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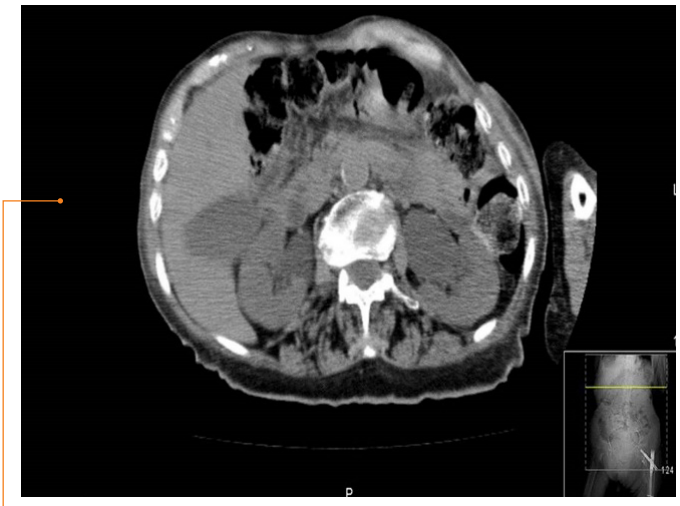

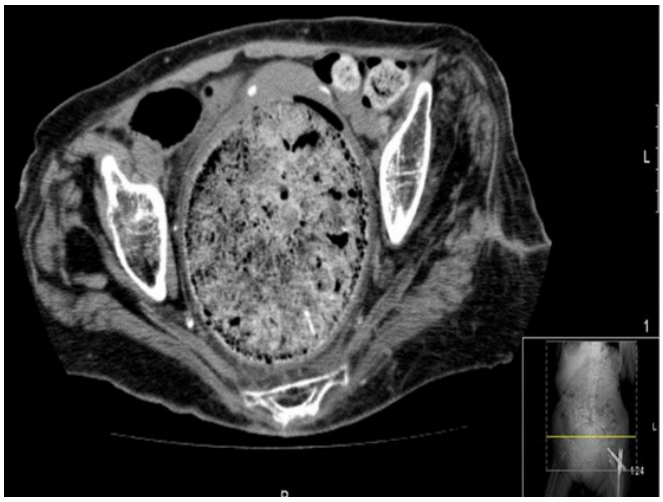

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RAMB - REVISTA DA ASSOCIAÇÃO MÉDICA BRASILEIRA (JOURNAL OF THE BRAZILIAN MEDICAL ASSOCIATION)

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Phytotherapeutic medicines: reality or myth?

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Over the past decades, there has been an increase in medical professionals' interest in the use and knowledge of plant-based medications — phytotherapeutic medicines. This increased interest can be due to the possible risks of the irrational use of allopathic medicines, in addition to their high costs. The World Health Organization, since the Declaration of Alma-Ata (1978), acknowledges that around 85% of the population of developing countries uses plants or plant-based products in healthcare. Therefore, it is necessary to respect and value this approach.

Phytotherapy is recognized as a therapeutic process encouraged by the national health bodies, but not as a medical specialty. There is no resolution on the subject by the Federal Council of Medicine. The Regional Council of Ceará recommends careful State supervision of its use as treatment by doctors. It is worth pointing out that, in its general concept, it is considered complementary treatment, like acupuncture and homeopathy. With this regard, the misinformation on the subject is clear, since the procedure to register phytotherapeutic medicines follows the same precepts and rules of any synthetic drug.

To avoid confusion on the subject, in 2012, legislation with a classification of three categories of plant-based products went into effect: plant drug (for teas); phytotherapic (finished product of living pharmacies) and phytotherapeutic medication. Considering the date of publication, we can observe that up to that point there was much confusion about the concept of phytotherapeutic medication, which was then defined as a product obtained exclusively from plant-based raw materials, with constant quality and reproducibility; as well as the knowledge of its risks and effectiveness, characterized by ethnopharmacological studies, technical-scientific documentation in publications or clinical trials. They also follow the same Anvisa rules for synthetic drugs It is necessary to emphasize the differences between countries regarding legislation on phytotherapics. In the United States, plant-based products are considered "nutritional supplements" and are not under the jurisdiction of its regulatory agency, the strict FDA. However, in Brazil, phytotherapics can be considered drugs and are supervised by strict legislation, under Anvisa, despite the fact that the definition of phytotherapic includes any product used as therapeutic and of exclusively plant-based origin. This includes, therefore, teas and artisanal or home-made preparations.

In research that aimed to evaluate the opinion of health professionals on the use of phytotherapics in public health care of Basic Health Units, 60% reported having information only from popular culture. Only 20% obtained knowledge from journals. This offers a glimpse of the huge risk of misinformation, or inappropriate or even erroneous conclusions. The interviewees added that the government program was

introduced without the professionals involved being consulted or trained.

When doctors in the Family Health Program of the city of Canoas (RS), a city that practically does not have a rural area, were approached, of the 31 doctors in exercise, 27 agreed to be interviewed. In their great majority, they were up to 30 years old, which indicates they have concluded their studies recently. Virtually all reported not only using plants in their personal care, but also recommending them to patients (avoiding the term "prescribe"), but also demonstrated that their knowledge on the subject came from their families, or even from the patients themselves. The practically unanimous report that "there is no knowledge based on evidence-based medicine" clearly evidences the lack of knowledge of the professionals on the content of the numerous studies published in national or international journals, as well as a lack of knowledge that could have been obtained in their academic programs. Thus, although there was recognition of the pharmacological action of plant-based products, it can be considered as resulting from the trust and faith of the patient, in many cases (although the word was not mentioned) a placebo effect.

This lack of knowledge does happen exclusively in Brazil. In fact, in a study conducted in Switzerland highlighted the growing interest in the use of alternative practices such as phytotherapy, homeopathy, and acupuncture, but these subjects were also not included in the academic curricula of the country. The study aimed to evaluate precisely the doctors' and medical students' interest in this inclusion. The conclusion was that the desire exists and that there should be informative programs so that students can know about the real efficacy, interactions, side effects and safety of these methods.

About the medical training in phytotherapy, a study presented in Congress highlights the need for a greater approach to the subject in medical schools, and that much of what the medical class knows comes from the propaganda of the pharmaceutical industry and not from university education, although there is a national program encouraging the use of complementary and integrative practices.

A doctorate study addressed the inclusion of phytotherapy in the curricula of Brazilian public universities. It analyzed the teaching of phytotherapy in 59 public universities: in faculties of medicine there were 42 references to the teaching of phytotherapy; in faculties of pharmacy, 36; and in nutrition programs, 40. However, when the mandatory status of the discipline was analyzed, in medical courses only one (Federal University of Mato Grosso do Sul) addressed the topic, and in 36 pharmacy programs, it was part of the curriculum. It is worth noting that students and professionals have shown interest in its use and knowledge, provided that it is well established in scientific evidence.

Another important point is the misconception that plant-based products do not bring risks and can be ingested indiscriminately. It should be noted that like any other medication, plant derivatives have adverse and toxic effects. Also, the long-term effect of many of them is unknown and, therefore, requires education, research, and appropriate use. It is recommended that this should be done under medical supervision. Also, many times there is the advantage of a much lower cost than that of other therapeutic agents.

In a national study, the author reports data on phytotherapy training initiatives in different locations. Among several, she mentions the program "Uso de Plantas Medicinais e Fitoterápicos" [Use of Medicinal Plants and Phytotherapics] for community health agents, of the Avasus unit of the Ministry of Health, active since 2010. We had the opportunity to access it and observed that its primary objective is to inform the public, get a better understanding of the use of plants in the communities, but not particularly of phytotherapics. The study also mentions the phytotherapy program for doctors that was promoted by the Ministry of Health but taught only in 2003, to 300 participants. And the course "objective of the study," sponsored by the municipal government of São Paulo for health professionals, with several classes, since 2010, including the classes of 2014 and 2015, with a total of 165 participants included. However, only 27 of these were physicians.

In the interviews conducted in this study, the participants reported difficulties in obtaining scientific knowledge about the theme and some confusion between scientific data and popular knowledge. The widespread idea that there is no scientific evidence in relation to plant-based products is proved to be false by running a simple check. For example, in the website of the American National Health Service, PubMed, internationally used as a reference, in a search for Glycinemax (soya), we found 25,127 references. Using the word "therapy" as a filter, 2,942

studies returned. After searching for, we found respectively 3,991or 2,017 publications. For (balmmint), 2,625 or 931 papers were found. For (mint), 931 or 143 studies. For (passionfruit), 645 or 171 works. There are obviously plant extracts much studied , and others not so often mentioned. These data show that there are numerous researchers around the world interested in the subject. Not to mention, of course, the possibility of access to other search sources available.

On the other hand, the idea of prescribing phytotherapeutic medicine does not mean replacing traditional medicines but widening therapeutic possibilities, since these are medicines and, as such, approved by Anvisa.

Brazil has a program sponsored by the Ministry of Health - the National Plan of Complementary and Integrative Therapies (PNPIC) - which covers the recommendation of use, by the Health System (SUS), of several therapeutic processes, such as acupuncture, homeopathy, and others, among them, phytotherapy. However, what we have observed is that, despite 12 plants having been approved for use in medicines, the SUS professionals of the do not often have in-depth information about the scientific knowledge of these plants, and its popular uses, including those common in the area in which the SUS unit is located, and "Living Pharmacies", i.e., the use of medicinal plants cultivated are consequentially favored and used many times . We cannot fail to recognize that in some cities, the programs were very well developed, with a very insightful staff, among which Vitória and Curitiba are particularly noteworthy, in addition to others .

Thus, we consider it is quite clear that phytotherapics can be very useful as a full or complementary therapy for many clinical conditions, but it is necessary to provide more information to the medical class (in particular the actual inclusion of the subject in academic courses) as well as to the public so that they can trust in such a powerful tool. Phytotherapeutic medicines need to be understood as a medicine available, an additional option, which is not a placebo!

REFERENCES

1. Rosa C, Câmara SG, Béria JU- Representações e intenção de uso da fitoterapia na atenção básica à saúde. Programa de pós-graduação em Saúde Coletiva, Universidade Luterana do Brasil, Av Farroupilha, 8001, prédio 14, sala 228, Bairro São José. 92425-900 Canoas RS (caroliner2007@gmail.com)
2. Parecer CREMEC No 33/2008ww.portalmedico,org.br/pareceres/crmce/2008/33_2008.htm
3. Relatório da reunião em Brasília Política Nacional de Plantas Medicinais e Fitoterápicos (PNPMF)http://farmacia.saude.pe.gov.br/sites/farmacia.saude.pe.gov.br/files/programa-nacional-plantas-medicinais-fitoterapicos-sem-marca.pdf
4. Bruning MCR, Mosegui GBG, Vianna CMM- A utilização da fitoterapia e de plantas medicinais em unidades básicas de saúde nos municípios de Cascavel e Foz de Iguaçu – Paraná: a visão dos profissionais de saúde. www,scielo.br/pdf/csc/v17n10/17.pdf
5. Nicolao M, Täuber MG, Heusser P. How should complementary and alternative medicine be taught to medical students in Switzerland? A survey of medical experts and students. Med Teach. 2010 Jan;32(1):50-5
6. Costa YMN, Lopes RP, Sousa JL, Frota MRX, Terceiro HBA- Fitpterapia e a formação médica: como avançar?AnCongBrasMedFam Comunidade. Belem, 2013 Maio; 12:1117
7. Barreto BB- Fitoterapia como conteúdo nos cursos de graduação da área da saúde: importância para a formação profissional. Tese de doutorado, Universidade de Brasilia, Faculdade de Ciências da Saúde, Programa de pós-graduação em ciências da Saúde, 2015
8. Marques LC- Entre o conhecimento popular e o científico. www.comciencia,br/dossies-1-72-/reportagens/fito/fito1.htm
9. Haraguchi LMM-A fitoterapia praticada por profissionais de saúde participantes do curso "Plantas Medicinais e Fitoterapia" realizado pela Secretaria Municipal do Verde e do Meio Ambiente de São Paulo (2014/2015). Dissertação apresentada à Universidade Federal de São Paulo – Escola Paulista de Medicina – Departamento de Medicina Preventiva. 2p18
10. Carbonel AAF, Simões RS, Girão JHC, Sasso GRDS, Bertoncini CRA, Sorpreso ICE, Soares Junior JM, Simões MJ, Baracat EC. Isoflavones in gynecology. Rev Assoc Med Bras (1992). 2018;64(6):560-564.
11. del Giorno C, Fonseca AM, Bagnoli VR, Assis JS, Soares JM Jr, Baracat EC. Effects of Trifolium pratense on the climacteric and sexual symptoms in postmenopause women. Rev Assoc Med Bras (1992). 2010;56(5):558-62.
12. Lima SMRR, Campaner AB, Auge APF. Isoflavones derived from Glycine max (L.) Merr. in the treatment of vaginal atrophy: A new frontier. Rev Assoc Med Bras (1992). 2017 ;63(9):727-728.
13. Kaari C, Haidar MA, Júnior JM, Nunes MG, Quadros LG, Kemp C, Stavale JN, Baracat EC. Randomized clinical trial comparing conjugated equine estrogens and isoflavones in postmenopausal women: a pilot study. Maturitas. 2006 Jan 10;53(1):49-58.
14. Han KK, Soares JM Jr, Haidar MA, de Lima GR, Baracat EC. Benefits of soy isoflavone therapeutic regimen on menopausal symptoms. Obstet Gynecol. 2002;99(3):389-94.
15. Accorsi-Neto A, Haidar M, Simões R, Simões M, Soares J Jr, Baracat E. Effects of isoflavones on the skin of postmenopausal women: a pilot study. Clinics (Sao Paulo). 2009;64(6):505-10.

Depression in the workplace: screening and treatment

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The Guidelines Project, an initiative of the Brazilian Medical Association, aims to combine information from the medical field in order to standardize producers to assist the reasoning and decision-making of doctors.
The information provided through this project must be assessed and criticized by the physician responsible for the conduct that will be adopted, depending on the conditions and the clinical status of each patient.

Screening depression in the occupational setting has the potential of diagnosing workers with depression symptoms in different levels of severity. Depression and its treatment have the potential of modifying occupational outcomes of functionality, productivity, absenteeism, presenteeism, return to work, work engagement, unemployment, among others. It was carried out from the systematic review of literature in the medline database, recovering 21,232 papers, 54 (figure 1 – annex I) being selected to answer the clinical questions: is it necessary to screen workers for depression? And is treatment effective and safe? The details of the methodology and the results of this guideline are exposed in annex I.

KEY POINTS

Screening depression in the occupational setting has the potential to diagnose, in impactful prevalence and with acceptable accuracy, workers with symptoms of depression at different levels of severity, knowledge about the disease and volition for adherence to treatment.

Depression and its treatment have the potential to modify occupational outcomes of functionality, productivity, absenteeism, presenteeism, return to work, work engagement, unemployment, among others.

Healthcare teams in the occupational setting have the potential to educate about depression and its treatment, promote adherence to treatment, coordinate and prescribe treatment for depression, just as it happens in studies published in the workplace or in primary care.

Periodic testing is one of the tools for implementing depression screening. Mapping and interventions by electronic means can promote adherence to screening and treatment, and optimize the distribution of educational content.

Interventions involving treatment with antide-

pressants, interventions based on cognitive behavioral therapy in person or by phone or via the web, multimodal interventions, among others, have potential of positive benefit for both depression symptoms and occupational outcomes, and are implementable in an occupational setting.

INTRODUCTION

Depression is characterized as a mood disorder that affects the way a person feels, thinks, or behaves, leading to impairment in social or occupational functioning.[1] A major depressive episode is defined by the presence of five or more of the nine major symptoms of depression over a period of two weeks[2]. The onset of depression may be triggered by biological, psychosocial or environmental factors, including risk factors present in the workplace. Those who have experienced an episode of depression before are at greater risk of having future episodes[3].

The prevalence of depression demonstrates its importance in public health in Brazil. Using data from 2013 to 2014 of the National Health Survey of IBGE, it is estimated that 9.7% of Brazilian adults present some degree of depression and 3.9% present major depression. Among adults with depression, only 27.6% were diagnosed at some point in their lives[4]. These data are consistent with a Brazilian systematic review of 2014 that estimated an annual prevalence of 8% of depression among adults and of 17% throughout life[5]. Data from the INSS[6] show the importance of mental health problems in disability, and in 2016, mental and behavioral disorders resulted in 10,376 of 212,209 urban accident-related disability pensions, 178,613 of 1,983,708 urban disability pensions and 6,423 of 207,100 rural disability pensions.

A 2017 systematic review gathered evidence of occupational risk factors with common psychiatric diseases, including depression, concluding that there is moderate-level scientific evidence from prospective studies associating high labor demand, low control, effort and reward unbalance, organizational (in)justice, low social support and violence at work with common psychiatric diseases[7]. These data are consistent with the factors perceived by workers interviewed in the National Health Survey of IBGE[8].

Because depression is potentially treatable, there has been an interest in screening patients presenting in primary care settings. The United States Preventive Services Task Force recommends universal screening where there is support to ensure adequate follow-up[9]. However, this study specifically analyzed clinical outcomes.

In view of the epidemiological importance, pertinence of occupational risk factors and the presence of impact on absenteeism and disability, this guideline seeks to evaluate the scientific evidences that demonstrate the consistency and the effectiveness of the screening of depression in a workplace focused on the modification of occupational outcomes.

RESULTS – EVIDENCE OF SUPPORT TO THE RECOMMENDATIONS

1. Screening and prevalence of depression in workers

Table 1 (Appendix I) summarizes the methodology and results of experiences related to the mapping of depression in the working population. The selected studies illustrate the feasibility and accuracy of mapping in the occupational setting and extend the concept of mapping as a mere cross-sectional diagnostic tool, and can be extended to a prospective analysis of the intensity of symptoms in a population exposed to occupational risk factors of interest, or to primary, secondary or tertiary prevention interventions, including active workers or workers on any sickness leave. The concomitant mapping of suspicious or interest occupational risk factors may guide the design of control measures. The occupational physician can take advantage of the methodology from the studies mentioned in this guideline to implement the mapping in practice. Most of the studies follow a methodology that will reflect in occupational health programs: population mapping of symptoms of depression through a questionnaire or structured interview, stratification of the population regarding the severity of depression symptoms, mapping of demographic indicators and occupational risk factors of interest, evaluation of the association between the depression symptoms and occupational or demographic risk factors of interest, followed, finally, by the implementation of preventive or curative measures.

As an example, the Ahlin[10]**(B)** study addressed Swedish workers aged 16-64. Study participants were followed up every two years since 2006 (n = 9,214) or 2008 (n = 9,703) or 2010 (n = 2,572) or 2014 (n = 19,388). In all, 28,672 individuals (70%) re-

sponded to at least one follow-up questionnaire by 2016, while 6,387 had responded up to six times. Basically, the participants were evaluated in relation to the presence of depressive symptoms and in relation to occupational components perceived by the workers as high demand, control, social support. In this study, the population was classified according to the severity of the depression symptoms: One group (n = 94, 1.1%) was classified as severe persistent, another group (n = 588, 9.4%) was classified as moderate persistent, and another one (n = 995, 12.6%) represented the group with subclinical to mild symptoms, thus totaling 23.1% of workers with significant depressive symptoms identified. This same study indicated that high demand, low control, and low social support in the workplace increase depressive symptoms over time. With this mapping modality, the study evaluated the association of occupational risk factors with the different severity levels of depression symptoms.

Nakamura-Taira[11](B) conducted a study with Japanese workers evaluating the relationship between the presence of depressive symptoms, perceived stress at work and beliefs about mental illness. The study sample involved 3,718 employees (2,660 men, 1,058 women). The presence of depressive symptoms totaled 10.2% of the workers analyzed. In addition, these individuals were more likely to expect depression to improve without treatment and also did not recognize useful sources of support (for example, talking to friends/family, seeing a psychiatrist, taking medication, seeing a counselor) compared with patients without depressive symptoms. In this sense, the underestimation of stress was related to the worst clarification regarding mental health.

Nieuwenhuijsen[12](B) carried out a study with Canadian workers, totalizing a sample of 2,219 employee participants analyzed, either by a questionnaire over the telephone (n = 2,145) or by a web-based survey (n = 74). Basically, the participants were evaluated in relation to the presence of depressive symptoms and presence of fatigue associated with the work activity characterized as recovery time required after work activity. In this study, 783 workers (38%) were identified with mild to severe depressive symptoms, based on the score used to screen depression. At the same time, this study identified an association between the presence of depressive symptoms and recovery time after work activity, identifying a risk of depressive symptoms eight times greater in the group of workers with a high need for recovery than workers with less need for recovery.

Wang[13](B) carried out a study with male workers. In total, there were 841 participants, including 511 men at high risk of severe depression and 330 with low risk of severe depression. This study identified that male workers classified as high risk for depression were more likely to endorse the importance of accessing health resources online than low-risk men (83.4% vs. 75.0%, respectively; P = 0.01). Of the 17 different characteristics evaluated, the three main ones most used by high-risk men were: "information on how to improve sleep hygiene" (61.3%), "exercise practice to help reduce stress and depression symptoms" (59.5%) and "having access to quality information and resources on occupational stress issues" (57.8%). Qualitative data analysis revealed that privacy issues, disease-related stigma, ease of web tool navigation and lack of personal interaction, time and knowledge were identified as barriers to the use of mental health programs by working men who were at high risk of depression. One of the main results of this study was that 62.7% of participants who were at high risk of depression used the internet to obtain health information in the 12 months prior to the survey. In addition, more than 75% of men at high risk of depression considered that health information online is useful to help them make healthcare decisions and more than 72% would use a mental health program to deal with work-related stress. Since men often delay the search for mental health problems due to stigma and gender norms, results suggest that the privacy inherent in mental health programs makes these programs a promising tool for improving men's mental health.

Volker[14](B) carried out a study in the workplace, whose sample consisted of 170 employees with work leave between 4 and 26 weeks. Basically, the purpose of this study was to validate the PHQ-9 questionnaire for depression within a population of employees on sick leave using the Mini International Neuropsychiatric Interview (Mini) as the gold standard. As a result, data from 170 employees were included in the reviews. Of the total of 170 Minis, 36 employees scored positively for depression (prevalence = 21.2%). Regarding the PHQ-9 questionnaire, a cutoff value of 10 resulted in an adequate balance between sensitivity and specificity, determining a sensitivity of 86.1%, specificity of 78.4%, positive predictive value (PPV) of

51.7%, negative predictive value (NPV) of 95.5% and accuracy of 80.0%.

Wada[15](B) carried out a study in the workplace where the CES-D (Center for Epidemiologic Studies Depression Scale) questionnaire was sent to all workers who performed the periodic health examination (2,409 individuals). Concomitantly, a version of the Mini International Neuropsychiatric Interview - Mini section that addressed the major depressive episode was administered to all workers. The percentage of participants with a CES-D score above 19 was 9.5%. Sensitivity and specificity were calculated for various CES-D scores. Sensitivity ranged from 95.1% to 85.3% and the specificity ranged from 82.2% to 93.1% in the central range of the curve. With a cutoff point of 16, which is the traditional score in the literature, the sensitivity was 95.1% and the specificity was 85.0%. The appropriate balance cutoff score for depression screening was calculated at 19. The study demonstrated the validity of the CES-D questionnaire for screening depression in working populations.

The analysis of the articles compiled in this subgroup shows relevant points about the screening and prevalence of depression among workers. The prevalence of significant depressive symptoms in workers is high from 9.5% to 38%. The main questionnaires used in the literature in the general population for screening depression may be validated for use in the working population. Screening in the working population allows contact with individuals suffering from symptoms of depression but that are not aware of the disease or do not have volition to seek medical treatment.

IMPACT OF DEPRESSION ON WORKERS

The studies mentioned in Table 2 in Annex I illustrate the high prevalence, the impact of depression on workers' health, on functionality and productivity, on presenteeism and absenteeism, being these indicators of interest in the practice of occupational medicine both for identification of the population of greater risk and evaluation of the effectiveness of interventions adopted. The instruments or questionnaires used in studies to measure the impact of depression on workers can be used in versions translated, validated and adapted, in whole or in part, in the practice of occupational medicine.

Asami[24](B) carried out an observational study with 17,820 workers in Japan. As a primary result, labor productivity was assessed using the validated questionnaire, a six-item instrument consisting of the following metrics: loss of general work productivity (general disability estimate, which is a combination of absenteeism and presenteeism), absenteeism (percentage of work time lost due to illness in the last seven days), presenteeism (percentage of compromise suffered during work in the last seven days due to illness) and impairment of activity (percentage of health-related disability in daily activities in the last seven days). The main independent variables for this project were those based on the self-reported diagnosis of depression and the outcome of the validated questionnaire response for depression screening (PHQ-9). Among workers (n = 17,820), 3.8% were diagnosed with depression within 12 months (n = 678). Among those with a diagnosis, 51.0% (n = 346) presented PHQ-9 scores of 10 and over, while among those without diagnosis, 7.8% (n = 1336) scored 10 and over. In other words, 7.5% among workers (1,336 out of 17,820) reported that they were not diagnosed as depressed, but had PHQ-9 scores of 10 and over. Absenteeism and general analyzes of deterioration of work included 16,906 out of 17,820 workers, while presenteeism included 17,428 workers. In all measures of productivity and work activity, the greatest losses were observed among those diagnosed compared to those undiagnosed. The interaction between PHQ-9 scores and the diagnosis of depression was significant for general work impairment, presenteeism and impairment of the activity ($P < 0.01$). The mean percentage disabilities adjusted by subgroups indicate that the effects of PHQ-9 on productivity were, in all cases, stronger in the undiagnosed than in the diagnosed group. This study demonstrated the impact of the underdiagnosed depression on the work performance of workers and, at the same time, the negative repercussion of the presence of significant depressive symptoms on presenteeism. On this item, the result of this article was corroborated by other studies selected in this guideline that analyzed the relationship between depression and presenteism[23,26-29,33,34](B).

Lamichhane[18](B) conducted a prospective study in a group of registered workers for health examination at the Department of Occupational and Environmental Medicine at a university hospital who work in 23 small and medium-sized manufacturing enterprises. Thus, the analysis was carried out using data from 2,349 individuals (1,807 men and 542 women).

Depressive symptoms were measured using the Center for Epidemiological Studies Depression Scale (CES-D). The dependent variable was whether or not a worker was absent from work due to an accident or illness in the previous year. Those who answered "yes" to (1) "were absent from work due to an accident at work last year?" or (2) "were absent from work due to illness last year?" were included in the absenteeism group. The percentages of workers who scored within the reference range of depressive symptoms (CES-D ≥ 16) and were absent from work due to illness were 16.9% for men and 27.5% for women. Men and women with depressive symptoms at the beginning of the study were more likely to be absent due to the disease at follow-up. Non-adjusted models showed a significant effect of depressive symptoms in absence of disease (OR = 3.67, 95% CI 2.17-6.21 for men and 2.14, 95% CI 1.29-3.56 for women). When gross odds ratios (OR) were calculated for absence due to accidents, men with depressive symptoms showed a statistically significant OR (OR = 2.95, 95% CI, 1.41-6.18). This study demonstrated the significant impact of depression symptoms on absenteeism due to illness or accident in affected workers. The result of this article was corroborated by other studies selected in this guideline that analyzed the relationship between depression and absenteeism[19,27,28,32,34](B).

Porru[17](B) conducted a prospective study on 5,263 European workers. The mean age was 55.0 years old. The primary outcome in this study was the status in self-reported work. Work status was measured after two and four years. Depressive symptoms were defined according to the validated Euro-D scale. In this study, it was observed that individuals with significant depressive symptoms were more likely to have an impact on their work capacity from long-term leave due to social security benefits with a significant hazard ratio (HR = 2.46, 95% CI 1.68 -3.60). In all, 19% of men and 20% of women who left work being paid through social security benefits had their leave attributed to significant depressive symptoms. This study demonstrated the impact on workers of the presence of significant depressive symptoms on long-term work leave. The result of this article was corroborated by other studies that analyzed the relationship between depression and prolonged leave selected in this guideline[21,22,25,26](B).

Weaver[16](B) conducted a prospective cohort study with healthcare professionals at four academic hospitals. The analysis included 416 participants who were monitored monthly for six months. In this study, the Patient Health Questionnaire for Anxiety-Depression (PHQ-4) was used to screen anxiety and depression. Adverse safety outcomes included motor vehicle collisions, "near misses", exposures to potentially infectious materials (occupational exposures), and adverse events to patients. Positive screening for anxiety or depressive symptoms was associated with a 63% increase in the incidence of adverse safety outcomes after multivariate adjustment. Workers with anxiety or depression had 124 adverse safety outcomes with RR = 1,63 (95% CI 1.58-1.69). This study demonstrated the impact on workers of the presence of significant depressive symptoms on accidents with motor vehicles, work accidents with biological material and adverse events for patients.

Newcomb[20](B) conducted a study with 205 employees selected for depression using the nine item Patient Health Questionnaire (PHQ-9). Screening for depression was associated with an increased diagnosis of depression compared to the control group (30% versus 4%, P <0.001). There was a significant difference in the need for activity restriction in the workplace, being reduced to 97 days for the screened employees compared to 159 days for the controls (P <0.001). Thus, depression screening was associated with a lower chance of receiving temporary work restrictions with OR = 0.55 (95% CI 0.38-0.78) or permanent restrictions with OR = 0.35 (95% CI 0.23-0.52). This study demonstrated the repercussion in workers of the presence of significant depressive symptoms on the need of activity adaptation in the workplace.

Lexis[31](B) carried out an observational study that examined the relationship between the presence of significant depressive symptoms and perceived health. The study was conducted among employees working in a large bank. The screening instrument contained four questions about the results of the health complaint experience and aid in the pursuit of health behavior. Of all the employees who responded to the screening instrument, 13.3% were identified as being at high risk of sick leave in the future and 8.3% were identified as having mild to severe depressive complaints. Of the employees identified as being at high risk of sick leave, 48% reported having complaints about their own health, compared to 20% of employees identified as being without risk. Complaints about their own health among employees identified with depressive com-

plaints were higher (57%), compared with 21% of employees without depressive complaints. This study and others selected[30] in this guideline demonstrated the repercussion in workers of the presence of significant depressive symptoms in relation to the perception of health at work.

In this subgroup of articles, the impact of depression on workplace was evaluated in 19 prognostic studies carried out on the working population. In eight studies, the impact of significant depressive symptoms on presenteeism and productivity was evaluated[23,24,26-29,33,34](B). In six studies, the impact of depression on absenteeism was analyzed[18,19,27,28,32,34](B).

In five studies, the impact of significant depressive symptoms on the long-term absence from work was evaluated[17,21,22,25,26](B). In four other studies, the impact of depression on other outcomes such as commute accidents, work accidents, work restriction, perception of health at work and adverse events in patients was analyzed[16,20,30,31](B).

The compilation of these studies demonstrates the extent of the repercussion of significant depressive symptoms on occupational outcomes in the workplace, determining the relevant impact on workers in presenteeism, absenteeism, prolonged leave, work accident, commute accident, activity restriction, perception of health at work and adverse events for patients.

SCREENING OF DEPRESSION IN WORKERS ASSOCIATED TO TREATMENT

Selected studies have involved clinical trials or systematic reviews[35,45,58](A) of interventions with potential to improve depressive symptoms and other occupational outcomes of interest. The selected studies involved pharmacological treatment with antidepressants[35,38,63](A), occupational therapy[48,61](A), psychoeducation[51,56,62](A), cognitive behavioral therapy or other modalities of psychotherapy with psychotherapist present[43,54,55](A) or by phone[38,40,47,52,53,57,60](A),[59](B), multifaceted interventions[49,50,52](A), use of automated online tools with heterogeneous content, generally based on principles of cognitive behavioral therapy, psychoeducation, self-help and other diverse contents such as relaxation or meditation techniques associated with a previous screening of depression symptoms[36,37,39,41,42,44,46](A).

Lee[35](A) conducted a systematic review published in 2018 of clinical trials that examined the impact of pharmacological treatment of depression on occupational outcomes such as work functionality and absenteeism. The selected papers met the selection criteria defined: adult population with major depression, submitted to pharmacological intervention with antidepressants, in randomized, double-blind, placebo or comparative intervention clinical trials with outcomes of work functionality or absenteeism assessed quantitatively with standardized instruments. The analysis of 13 comparative clinical trials with placebo and four comparative clinical trials with other interventions reported the efficacy of antidepressants on subjective measures of commitment in the workplace. Treatment with antidepressants has improved standardized measures of functioning in the workplace. The study suggests that pharmacological treatment with antidepressants has a positive effect on productivity in the workplace.

Tan[45](A) conducted a systematic review published in 2014 that selected randomized clinical trials on workplace interventions that reported outcomes on mental health for individuals with depression. The selected studies compare at least two different intervention groups randomly allocated with at least one being a control group or waiting list. Study participants should be active-age adults (18-65 years old) who belonged to a working group. It was observed that most of the included studies used cognitive behavioral therapy (CBT) techniques. The overall standardized mean difference (SMD) between the intervention and control groups was 0.16 (95% CI 0.07-0.24, P = 0.0002), indicating a positive effect. A separate analysis using only interventions based on cognitive behavioral therapy (CBT) generated a significant difference of 0.12 (95% CI 0.02-0.22, P = 0.01). The results indicate that a number of different intervention programs on depression have positive effects on the workplace. When analyzed separately, universally distributed CBT interventions significantly reduced levels of depressive symptoms among workers. These results demonstrate that appropriate interventions based on psychotherapy in the workplace should be part of efforts to intervene in depression. Other clinical trials selected in this guideline analyzed the application of in-person psychotherapy in the workplace, individually or in groups, alone or in conjunction with other interventions[43,48,51,54,55,60,61,62](A), by telephone[38,40.47,52.53,57,60](A)[59](B), online[36,37,39,41,42,44,46](A). These findings are in line with other recent systematic reviews recently published

on the subject[64-67](A). In a meta-analysis published in 2019 by Nigatu[67](A), the benefit of CBT-based and non-CBT-based interventions had small to medium effect size with significant standardized mean differences (SMD) and, respectively, in -0.44 (95% CI -0.61 to -0.26, I^2 = 62.1%) and in -0.32 (95% CI -0.59 to -0.06, I^2 = 58%).

Martin[58](A) conducted a systematic review published in 2009 that selected articles that focused on workplace interventions that reported outcomes in mental health for individuals with depression and anxiety. The primary outcome measures were the composite measure scores of depression, anxiety or mental health used as screening tools for these conditions. The multimodal intervention aimed at mental health, directly or indirectly, through a program to promote mental health in the workplace that acts on a known risk factor for depression or anxiety, such as smoking, chronic illness, substance abuse, obesity, physical inactivity and organizational climate. In total, 22 studies met the inclusion criteria, with a total sample of 3,409 post-intervention employees, 17 of which were included in the meta-analysis. The pooled results indicated small but positive overall effects of interventions in relation to depression symptoms with standardized mean differences (SMD) of SMD = 0.28 (95% CI 0.12-0.44) and anxiety with SMD = 0.29 (95% CI 0.06-0.51). This study suggests that multimodal intervention on work organization that is part of a workplace mental health promotion program improves clinical outcomes in individuals with depression. This finding is consistent with the results of a systematic review published in 2014 on the positive benefit in reducing depression symptoms of occupational interventions based on exercise[68](B).

Smith[63](A) conducted a clinical trial on the impact of the primary healthcare service on occupational outcomes. The intervention consisted of "optimized care," in which healthcare providers (physicians and nurses) directed the treatment in accordance with the recommendations of the Agency for Healthcare Research and Quality (AHRQ) guideline. Control consisted of usual care for study participants. Of the 262 patients in the baseline sample eligible for this analysis, 219 (83.6%) were followed up at one year. The occupational outcomes analyzed were employability/turnover and organizational climate. The intervention significantly increased employability with 10.1% (p = 0.04, CI 90% 2.8-17.4%) and reduced unemployment by 4.3% (CI 90% 1.2-7.4 %). In addition, among participants on optimized care, there was a significantly lower probability of reporting workplace conflicts (8.1% vs. 18.9%, p = 0.04). This study suggests that the incorporation of the primary care logic with the training of physicians and nurses improves occupational outcomes in individuals with depression, thus determining the repercussion of the intervention on employability, turnover and organizational climate.

In this sense, the compilation of these studies demonstrates that screening for depression through validated questionnaire can be followed by therapeutic interventions, either in person or at a distance, with benefit on clinical and occupational outcomes, regardless of whether the worker is active or retired, or if they had a previous depressive episode or not. The set of interventions reflects the multifactorial nature of depression and the need for equally comprehensive interventions.

DISCUSSION

In order to assess the importance of screening for depression, specifically of workers (in a workplace), as well as their consequences in effective and safe therapeutic measures, it will be necessary to overcome some barriers and concepts that hinder not only the understanding of the scenario, but also the generation and/or the interpretation of available scientific evidence.

The extrapolation, from data and scientific information, on the screening of depression from the general population to the population of workers, based on the proximity between the prevalence indexes of these two populations, is insufficient. This is because the mechanisms of generation, maintenance and recurrence of depression in workers, although similar, present aspects specific to the occupational sphere and, consequently, the intervention measures present specific nuances. In addition, because of this extrapolation, there is a distinct accommodation of the scientific community in the generation of randomized trials comparing whether or not to screen for depression in the occupational population, which naturally reduces the strength of available evidence, supporting screening among workers.

At the same time, there is an unfounded concern that screening is used as an admission selection measure or to guide cost-centered post-admission actions, producing obvious detriment or reduction of care focused on the worker.

In this sense, in the specific evidence on depression in the workplace there are still biases, for example, the difficult individualization of exclusive populations of depressive and non-depressive patients in the screening, prevention and treatment actions, as well as the presence of other treatments concomitant with the interventions of the studies.

However, it is possible to indirectly build an adequate evaluation of the importance of the screening of depression in workers supported by prevalence data, the impact of depression on occupational outcomes, and response (efficacy and safety) to pharmacological and non-pharmacological treatment modalities in the workplace.

Thus, despite the fact that there are no experimental cohorts (RCTs) comparing screening and not screening worker populations as there is in the non-occupational population, we can see that the available evidence on the management of depression among workers through active screening and treatment offer for those diagnosed can be considered of moderate strength of scientific evidence based on observational cohorts and randomized clinical trials.

Among the recognized interventions, mention is made of pharmacological treatment with antidepressants, psychotherapy (presential or distance), multimodal intervention on work organization and primary care service centered on the worker's care.

Thus, the population of depressive patients among workers is underdiagnosed in an environment in which the worker seeks, with or without psychological attention, to remain in activity without his/her problem being noticed. On the other hand, the supervision of these workers does not notice the indirect or even direct signs of the presence of depression, and if they do notice, since they do not know how to deal with the situation, they do not take intervention measures, thus failing to generate benefits in occupational outcomes such as presenteeism, absenteeism, prolonged leave, work accident, commute accident, activity restriction, health perception and adverse events for patients.

RECOMMENDATION

Screening for depression in workers is recommended because of its high prevalence and underdiagnoses in the workplace. There is evidence that depression in workers has a relevant impact on occupational indicators and on the generation of comorbidities. Therefore, its early diagnosis and identification is recommended, as well as specific interventions, including actions on risk factors for depression at work.

Thus, screening for depression needs to be followed by diagnostic confirmation and pharmacological and non-pharmacological therapeutic measures, with the benefit and safety being verified in occupational outcomes such as presenteeism, absenteeism, prolonged leaves, work accident, commute accident, activity restriction, perception of health and adverse events for patients.

On screening, several instruments for screening or diagnosis of depression already validated for use in the workplace are found. Among the recognized interventions, mention is made of pharmacological treatment with antidepressants, psychotherapy (presential or distance), multimodal intervention on work organization and primary care service centered on the worker's care.

The findings and conclusions of this guideline are in agreement with systematic reviews published on the subject[35,45,58,62,63,64].

ANNEX I

Clinical question

Is it necessary to screen workers for depression? Is the treatment effective and safe?

Structures question

The research question was organized according to the acronym PICO (P for population or problem, I for intervention or indicator, C for control or comparison, and O for outcome). From the PICO format, from the descriptors of health science (MeSH terms) and their synonyms, search strategies were defined for each database.

P	adult patient in a workplace (worker) with or without current symptoms of depression
I	screening and/or treatment of depression in the workplace
C	usual measures or no intervention
O	job, absence or leave from work, recurrence, return to work

ELIGIBILITY CRITERIA

Inclusion

1. Elements of PICO
2. Randomized clinical trials; observational cohort studies; cross-sectional studies; systematic review with or without meta-analysis (the most recent)
3. No restriction on language and period
4. Full text or abstract with data

Exclusion

1. Population outside the workplace or scope
2. Treatment or adherence to treatment of depression
3. Papers assessing risk factors for depression
4. Papers assessing stress, burnout, anxiety, insomnia, mental health
5. Validation of diagnostic instruments
6. Quality assessment of workplace
7. Cost-effective or effectiveness models

ARTICLES SEARCH

Databases

Medline, Embase, Central Cochrane. Manual and gray search.

Research strategy

#1 (Depression* OR Depressive OR Depressive disorder) AND (screening OR questionnaire* OR score* OR scale* OR tool* OR survey*) AND (Worker OR Workplace OR Workplaces OR Work OR Job OR Worksite OR Employment OR Occupation* OR Occupational OR Industry OR Occupational diseases) – 19,850 papers.

#2 (Depression* OR Depressive OR Depressive disorder) AND (Worker OR Workplace OR Workplaces OR Work OR Job OR Worksite OR Employment OR Occupation* OR Occupational OR Industry OR Occupational diseases)))) AND random* - 4,062 papers.

CRITICAL ASSESSMENT

The selected evidence was graded according to the Oxford Centre for Evidence-based medicine – Levels of Evidence (March 2009)[69], with recommendation grades A or B being added to the references.

RESULTS EXTRACTION AND ANALYSIS METHOD

From each evidence included, the necessary data were extracted to support, through a text, the recommendations to answer the clinical question taking into account the characteristics of the patients, the interventions and comparisons, and the outcomes defined in the eligibility criteria.

RESULTS

Thus, 54 papers were selected to support the guideline, of which 25 were randomized clinical trials, 3 systematic reviews, 12 cross-sectional studies and 14 observational cohort studies. The scientific evidence included to support the recommendations is exposed in three subgroups for didactic purposes of discussion, and the content of each article is not necessarily limited to a single category:

1. Importance of depression in workers based on screening and prevalence (6 papers) [10-15].
2. Importance of depression in workers based on its consequences (19 papers)[16-34].
3. Screening for depression aggregated to efficacy and safety of treatment proposals (29 papers)[35-63].

FIGURE 1. INFORMATION FLOW WITH DIFFERENT STAGES OF THE SYSTEMATIC REVIEW

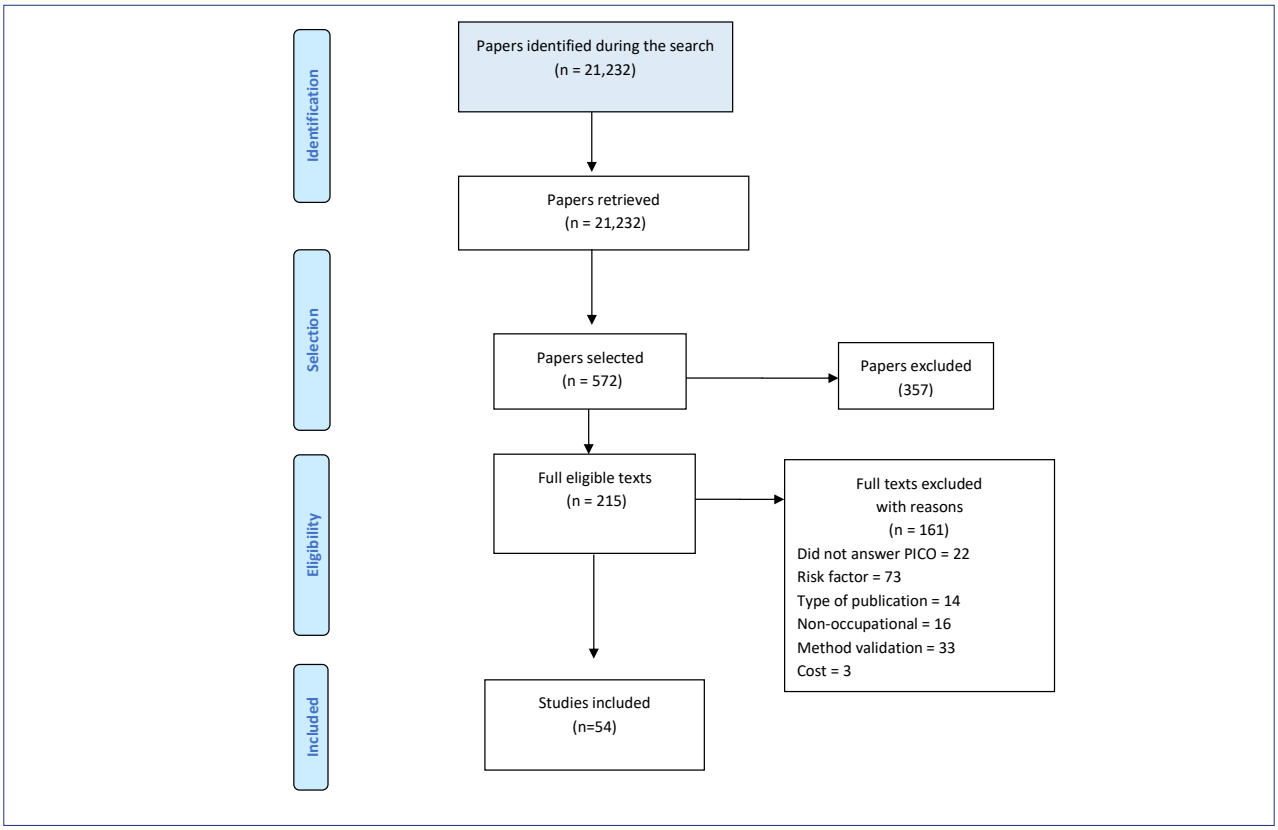

TABLE 1. STUDIES SELECTED RELATED TO MAPPING OF DEPRESSION IN WORKERS

Author	Population	Main results
Ahlin 2018[10]	Data from the Swedish Longitudinal Occupational Survey of Health (Slosh) study of 6,679 Swedish workers from 16 to 64 years of age who took the Symptom Checklist Core Depression (SCL-CD6), Hopkins Symptom Checklist (HSCL), Demand-Control-Support -Questionnaire (DCSQ) between 2006 and 2014 every two years.	1,336 workers maintained over the years very mild symptoms of depression (score 0-6). 3,299 workers maintained persistent mild symptoms (score 7-9). 995 evolved from borderline symptoms to mild depression (score 10-11). 588 had persistent moderate depression (score 12-15). 94 had persistent severe symptoms (score > 16). There was a significant association of high labor demand and low social support in both the population with mild persistent symptoms (RR = 1.26 CI 95% 1.06 - 1.51) and population with severe persistent symptoms (RR = 2.51 CI 95 % 1.43 - 4.41).
Nakamura-Taira 2018[11]	3,718 Japanese volunteer workers from various work activities completed a questionnaire about stress (SUB scale), Kessler Psychological Distress Scale (K6), and a knowledge assessment about depression by means of a vignette.	Underestimating stress has a positive association with failure to recognize symptoms of depression, knowledge about the disease, expectation of spontaneous resolution of depression, denial of the need for support from relatives, friends, drug treatment and psychotherapy.
Nieuwenhuijsen 2016[12]	2,219 Ontario volunteer workers completed questionnaires online or interviews. The Need for Recovery after Work (NFR), Patient Health Questionnaire (PHQ-8), Job Content Questionnaire were applied.	A high NFR score correlates with more severe symptoms of depression (OR = 8.3 95% CI 6.8 to 10.2). There was no significant association of depression with stress at work or job strain (association of high demand with low occupational control). Of 2,068 workers, 800 had a high NFR score with a mean of depression greater than 7.5 vs. 2.7 compared to workers with lower NFR scores.
Wang 2016[13]	841 Canadian workers without depression were evaluated by a predictor algorithm for major depression in 4 years.	511 considered high risk for depression and 330 low risk after application of the algorithm. Higher-risk workers were more likely to endorse the importance of accessing healthcare resources online than low-risk men (83.4% vs. 75.0%, respectively, P = 0.01). Of the 17 different characteristics evaluated, the three most commonly used by high risk men were: "information on how to improve sleep" (61.3%), "practice and exercise to help reduce symptoms of stress and depression" (59.5%) and "access to quality information and resources on work-related stress issues" (57.8%). Qualitative data analysis revealed that issues of privacy, perceived stigma, ease of navigation, personal relevance and lack of personal interaction, time and knowledge were identified as barriers to the use of mental health programs in working men who were at high risk of depression.

Author	Population	Main results
Volker 2016[14]	170 workers on work leave from 4 to 26 weeks were selected for validation of the PHQ-9 questionnaire for depression.	36 employees scored positively for depression. The optimal cutoff value in the questionnaire score is 10 points, resulting in sensitivity of 86.1%, specificity of 78.4%.
Wada 2007[15]	2,219 workers, during a periodic occupational examination, answered two questionnaires for depression: Center for Epidemiologic Studies Depression Scale (CES-D) and Mini International Neuropsychiatric Interview (Mini).	The optimal cutoff of the questionnaire in this population was 19 points for the screening of major depression with sensitivity of 92.7%, specificity of 91.8%. A proportion of 9.5% of this population presented CES-D above 19 points.

TABLE 2. STUDIES SELECTED RELATED TO THE IMPACT OF DEPRESSION ON WORKERS

Author	Methodology	Main results
Weaver 2018[16]	416 healthcare workers with a total of 1,367 person-months were assessed with questionnaires for sleep disorders, for anxiety and depression, for automobile accident events or with biological material, near misses and medical error.	Two out of five participants had at least one sleep disorder, with insomnia being the most common, followed by sleep apnea, restless leg syndrome, and shift work-related disorder. 23 reported an auto accident, there were 200 near-auto accident events by 94 participants, 66 accidents with biological material in 30 participants. 75% of reported medical errors were attributed to sleep deprivation or fatigue. There was a significant association between the sleep disturbance group with adverse safety events (p = 0.001), with an 83% increase in these events. Depression and anxiety independently added adverse events by 63%. There was no significant relationship between sleep disorders and symptoms of anxiety or depression. Individuals who present with sleep disorders and depression or anxiety present triple the risk for adverse safety events with RR = 2.97 (95% CI 2.12 to 4.16) and medical error with RR 2.24 (95% CI 1.42 to 3.55) and near misses with RR 2.47 (95% CI 1.62 to 3.47). 88% of the population with sleep disorders continued without diagnosis or treatment.
Porru 2018[17]	5,263 workers from 11 European countries aged between 50 years old and the specific retirement age of the local country were evaluated prospectively by interviews after two and four years. The study seeks to associate the questionnaire score for Euro-D depression with compensated work leave.	For both genders, high score in Euro-D has a significant association with disability pension with HR = 2.46 (95% CI 1.68 to 3.60). Among men, depressive symptoms have a non-significant association with unemployment with HR 1.55 (95% CI 0.94 to 2.57) and significant among women with HR = 2.10 (CI 95% CI 1.17 to 3.77). 19% of the study population had four or more symptoms of depression.
Lamichhane 2017[18]	2,349 plant workers were prospectively evaluated for one year. The study sought to associate depressive symptoms with absenteeism using the Center for Epidemiologic Studies Depression Scale (CES-D).	For both genders, the presence of depressive symptoms with a CES-D score greater than or equal to 16 is associated with higher chances of future absenteeism with OR = 4.06 (95% CI 2.32 to 7.11) in men and OR = 1.75 (95% CI 1.02-2.98) in women.
Vendrig 2018[19]	2,514 workers participated in the validation of the Work and Wellbeing Inventory (WBI).	The depression scale predicted absenteeism in future work (> 6 weeks) because of a common mental disorder in healthy workers. The scale of stress at work and the disease behavior scale predicted long-term absenteeism (> 3 months) in workers with short-term absenteeism.
Newcomb 2016[20]	205 workers were selected and completed the Patient Health Questionnaire (PHQ-9).	Screening for depression was associated with an increased diagnosis of depression compared to controls (30% versus 4%, P <0.001). There was no significant difference in length of absence at work or percentage of workplace restrictions. However, the restricted work duration was reduced to 97 days for the screened employees compared to 159 days for controls (P <0.001). The screening of depression was associated with a lower chance of receiving temporary work restrictions with odds ratio OR = 0.55 (95% CI 0.38 to 0.78) or permanent restrictions with OR = 0.35 (95% CI 0.23 to 0.52).
van Hoffen 2015[21]	2,782 postal workers were evaluated prospectively over a two-year period seeking an association between symptoms related to mental health and long-term absenteeism. Two questionnaires were applied: Four-Dimensional Symptom Questionnaire and Maslach's Burnout Inventory.	Scores of underlying mental health symptoms were significantly associated with prolonged absenteeism due to mental illness during the two-year follow-up. Mental health symptoms did not discriminate between postal workers with and without prolonged absenteeism due to all causes. Postal workers with prolonged absenteeism due to mental illness presented higher median scores of distress (40.5, interquartile range [IQR] 12.5 - 87.5), depressed mood (25.0, IQR 0.0 - 50.0) and fatigue (40.0, IQR 20.0 - 68.0) than postal workers without mental disorders. These workers who went on leave during the follow-up presented a score of 25.0 (IQR 9.4 - 53.1, P <0.01), 0.0 (IQR 0.0 - 25.0, P <0.01) and 28.0 (IQR 8.0 - 48.0, P <0.01) in suffering, depressed mood and fatigue, respectively.
Torske 2015[22]	The study sought to evaluate the association between rural activity and depression symptoms with disability pension. For 14 years (1985-2008), 29,016 Norwegian workers from different occupational activities participated with 3,495 farmers, all under 62 years of age and active. The Hospital Anxiety and Depression Scale (Hads) questionnaire was applied.	In comparison with the high educational level population, farmers presented a higher risk of pension due to occupational disability HR 2.07 (95% CI 1.80 to 2.38). Farmers with symptoms of depression have a higher risk of occupational disability pension HR 1.53 (95% CI 1.25 to 1.87) compared to farmers with no symptoms of depression. The same occurs in farmers with anxiety symptoms with HR 1.51 (95% CI 1.23 to 1.86).

Author	Methodology	Main results
Rose 2015[23]	The study assessed the association between symptoms of depression and functionality among 6,339 German active workers born in 1959 or 1965 in unspecified activities and workload. Civil servants and freelancers were not included in the study. The Beck Depression Inventory questionnaire (BDI-V) and two items of the Work Ability Index were applied.	The discussion of the study sought to define BDI-V scores associated with loss of functionality. For men, cutoff values between 20 and 24 points are associated with a sensitivity of 0.64 to 0.75 and a specificity of 0.64 to 0.75. In women, cutoff values between 23 and 28 points are associated with a sensitivity of 0.57 to 0.74 and a specificity of 0.60 to 0.74. The instrument does not define a cutoff score associated with disability but rather with loss of functionality.
Asami 2015[24]	The study sought to evaluate the association of severity of depression with absenteeism, presenteeism, loss of work functionality and in the daily activities of 17,820 Japanese workers. The Patient Health Questionnaire-9 (PHQ-9) and Work Productivity and Activity Impairment (WPAI) questionnaires were used.	1,336 of 17,820 or 7.5% of the workers had scores above 10 in PHQ-9 and were undiagnosed, 346 diagnosed with depression had PHQ-9> 10 points. There was a significant association between score in PHQ-9> 10 with presentism and loss of work functionality and in daily activities, but not significant with absenteeism. The effects on productivity and absenteeism were higher in the undiagnosed population compared to those diagnosed.
Ervasti 2015[25]	The study assessed the association between present or recent psychiatric illness and other comorbidities with return to work of 9,908 Finnish public officials with at least one episode of absenteeism due to depression between 2005 and 2011. In total there were 14,101 events of absenteeism due to depression in the period in this population.	12,486 or 89% of absenteeism episodes result in return to work, with a mean of 34 days. Comorbidities identified in the study were associated with a lower probability of return to work due to depression. Other psychiatric diseases with HR = 0.78 (95% CI = 0.74-0.83), cancer with HR = 0.66 (95% CI = 0.47-0.92), diabetes with HR = 0, 73 (95% CI = 0.62-0.86), cardiovascular disease with HR = 0.78 (95% CI = 0.62-0.99), hypertension with HR = 0.76 (95% CI = 0.67-0.85), osteomuscular disease HR = 0.82 (95% CI = 0.77-0.87) and asthma HR = 0.84 (95% CI 0.75-0.94).
Wang 2014[26]	A cross-sectional study of 1,000 Brazilian active workers currently or in the last 12 months, from 16 to 64 years of age, sought to assess the prevalence of symptoms of depression and loss of work functionality using a questionnaire adapted from the European Depression Association and another one for functionality.	One in five workers reported having received at some point in their lives a diagnosis of depression. 73.5% of the diagnosed workers remained active. 60% of these workers reported loss of work productivity associated with the presence of cognitive symptoms such as loss of interest (59%), bad mood or sadness (52%), sleep disorders (44%) and difficulty concentrating (36%). In individuals already defined as depressed, a total loss of 4,139 working days per year was reported by the 63 individuals who left work because of depression. The calculated absenteeism considering part-time workers (n = 11) was 3,795 days. The mean number of off-day days was 65.7 days. More than half of these workers have been away for 21 or more days.
Jain 2013[27]	1,051 adult and active workers with a diagnosis of depression were evaluated for severity of symptoms and their association with loss of productivity, applying the questionnaires Patient Health Questionnaire (PHQ-9), World Health Organization Health and Performance Questionnaire (HPQ) and Work Productivity and Activity Impairment Questionnaire (WPAI).	All levels of depressive severity were associated significantly and proportionally with worsening in work productivity, either by the measure of presenteeism through HPQ and WPAI and absenteeism through WPAI. Mean unadjusted scores in the HPQ (no depressive symptoms [81.0], mild [73.5], moderate [68.6], moderately severe [66,1] and severe [61,5] depression; P <0, (P <0.001) and mean WPAI scores (no depressive symptoms [10.7], mild [26.2], moderate [38.8], moderately severe [44.7] and severe [54.3] depression; P < 0.0001) presented progressive worsening of labor productivity (presenteeism) with the increase of depression severity.
Woo 2011[28]	102 South Korean workers aged 20-60 years of age who had untreated depression and who had no other major physical or mental comorbidities were compared and matched to 91 healthy workers. The intervention group received pharmacological treatment and psychotherapy for 20 to 30 minutes. Intensity of symptoms of depression and productivity were assessed after four and eight weeks of treatment. The questionnaires used were the World Health Organization's Health and Work Performance Questionnaire (HPQ) and the Hamilton Rating Scale for Depression (HAM-D).	After eight weeks of antidepressant treatment with supportive psychotherapy, HAM-D scores improved significantly (24.00 vs. 7.19, P <0.001). The number of workdays absent due to health problems and that due to any other reason decreased significantly after eight weeks of treatment (P = 0.041; P = 0.008). The number of days missed due to health problems during the last four weeks also decreased (P = 0.003). The performance of the self-assessed work increased significantly from 4.90 to 6.46 (P <0.001). Compared with the control group, there was a significant difference for a higher prevalence of absenteeism and presenteeism in the intervention group. The self-reported average productivity in workers in the intervention group is 32.3% lower compared to the control group and 22.2% lower compared to the past one to two years.
Harvey 2011[29]	An ecological study with adherence of 1,161 English telephone operators sought to evaluate the association of depressive symptoms with productivity. In the study were used the Patient Health Questionnaire depression scale (PHQ-9) and four telemarketing productivity indicators.	The implementation in the online environment of online questionnaires via the web is feasible. 623 or 54% of the workers presented PHQ-9 with scores above or equal to 5 points, demonstrating a high prevalence of population with at least subclinical symptoms. There was an association between severity of depressive symptoms and level of productivity at work.
Munir 2011[30]	Data from completed questionnaires of Danish workers from 2000 (n = 5,510) and 2005 (n = 8,393) were assessed seeking association of long-term absenteeism in workers with depression with positive occupational psychosocial factors. The questionnaires used were the 5-Item Mental Health Inventory and Copenhagen Psychosocial Questionnaire (COPSOQ).	Leadership quality was associated with a reduction in sick leave to a stronger degree for those with moderate depressive symptoms (hazard ratio = 0.88, 95% CI = 0.78-0.98) than for those without depressive symptoms, and high decision autonomy was a protective factor against depression (RR = 0.91, 95% CI = 0.85-0.97).

Author	Methodology	Main results
Lexis 2010[31]	The study aims to evaluate the association of depressive symptoms with future risk of absenteeism and the adherence profile to the search for treatment. Data of 8,893 bankers. Questionnaires used in the study: Hospital Anxiety and Depression (HAD) and The Balansmeter (BM).	2,311 workers presented health complaints and 1,848 of 2,311 workers previously sought help: 1,293 for primary care, 341 for the occupational physician, 951 for other professionals. Of 463 untreated workers, 35% have a future intention to seek help and 46% do not intend to seek help. Among workers with depression at any level and with a high risk profile for absenteeism and without treatment started, only 50% expressed intention to seek treatment, a scenario that may be a barrier to occupational prevention programs.
Lexis 2009[32]	Data from 3,339 Dutch workers were assessed for association between depressive symptoms as predictors of absenteeism during a 10-month period in 2001. The Hospital Anxiety and Depression (HAD-D) questionnaire and absenteeism data provided by companies were used.	In both men and women, a significant association was found between depressive complaints and the onset time of the first period of sick leave. In men, the hazard ratio (HR) was 1.03 (1.01 to 1.05) and 1.04 (1.01 to 1.07) in women. For both men and women, the HR for the relationship between depressive complaints and the time of the beginning of the first period of sick leave were all in the expected direction, where those with mild or moderate-severe complaints had a higher risk of taking leave of absence earlier than employees who scored within the reference range.
Wang 2004[33]	The study aims to evaluate the association of depressive symptoms with momentary work performance in 105 aviation reserve agents and 181 telephone operators. For seven days, a diary and data collection via pager were performed. For screening of depression, the Composite International Diagnostic Interview Short Form questionnaire was used.	Major depression was significantly associated with a decrease in task focus at decreases of approximately 12 points in task focus and approximately 5 points in productivity on their scales from 0 to 100. These effect sizes are equivalent to a reduction of standard deviation of 0.4 in the focus of the task and a reduction of 0.3 standard deviation in productivity. As these effects are based on random experience sampling method assessments during the workday, they can reasonably be considered to describe the average decreases in these results at all times in a typical workweek.
Druss 2001[34]	The study evaluated the association between depressive symptoms, level of health service satisfaction and occupational outcomes after two years (1993 to 1995) in a population of 6,239 workers. For depression, the 36-item and 12-item versions of the Short-Form Health Survey were applied.	In all, 1,200 enrolled in the longitudinal sample (19.2%) met the criteria for depressive symptoms. Multivariate models indicated that the odds of absence from work due to illness were 2.17 times higher for those interviewed with chronic depressive disease than for participants without depressive symptoms, and the odds of reporting decreased efficacy at the workplace were 7.20 times higher. Incident depressive symptoms were associated with an intermediate impact on workplace function. Participants whose symptoms were resolved showed some persistent difficulties in workplace function but these effects were substantially lower than those for groups with chronic and incident depressive symptoms.

TABLE 3. STUDIES OF SCREENING FOR DEPRESSION IN WORKERS ASSOCIATED TO TREATMENT

Author	Methodology	Results
Lee 2018[35]	Systematic review of clinical trials that evaluated the effects of antidepressants on functionality at work. We selected papers published until 07/28/2017 in databases or search engines: Medline, ClinicalTrials.gov, Google Scholar.	Thirteen comparative clinical trials with placebo and four active comparators reported the efficacy of antidepressants on subjective measures of workplace impairment. Overall, antidepressant treatment has improved standardized measures of workplace functioning (for example, Sheehan Disability Scale work item). Evidence available suggests that antidepressant treatment improves workplace outcomes in MDD. The increase with other interventions may, additionally, facilitate the return to pre-morbid levels of functioning and return of patients to work (for example, lifestyle changes, cognitive-behavioral therapy, work-directed interventions, sleep normalization).
Beiwinkel 2017[36]	Clinical trial using the online tool "HelpID" based on cognitive behavioral therapy, mindfulness and counseling containing interactive content, videos, audios, graphics and others to promote education for depression, self-knowledge and relaxation, among others. The intervention was weekly, from 30 to 45 minutes. The Patient Health Questionnaire (PHQ9) and Beck Depression Inventory (BDI II) questionnaires were applied before, at the end of the 12 week intervention and after 24 weeks.	180 participants were randomized into intervention group (n = 100, final adhesion of 31) and control (n = 80, final adhesion of 27). The control group received informative texts on depression. Both groups presented a significant reduction of absenteeism in -67.23%, P <.001 in the intervention group and -82.61%, P <.001 in the control group. There was no significant difference in absenteeism reduction between the two groups (P = 0.07). A significant difference between groups in favor of the intervention group was found for PHQ9 (P <0.001), which corresponds to a mean effect size. For those in BDI-II, a significant difference was found between the posttreatment groups (P = 0.004), which corresponds to a moderate effect size. In the intervention group, 63% (63/100) of the participants showed a reliable change of symptoms from baseline to post-intervention and were classified as responders. In the control group, 33% (27/80) were classified as responders. The difference in the reliable change of symptoms between the intervention and the control group was significant (p <0.001). This resulted in an NNT of 4.08.

Author	Methodology	Results
Hirsch 2017[37]	Clinical trial using myStrenght digital tool with content based on cognitive behavioral therapy, religion, mindfulness, mood monitoring and others. The intervention group accessed the content freely during the 26 weeks of the study. The control group received e-mails with educational content on depression. The Depression, Anxiety, and Stress Scale (Dass21) questionnaire was applied at the beginning of the intervention and after 14, 60 and 180 days.	96 participated in the intervention group and 69 in the control group. The primary outcome of interest is the change in depression score over time. Both participants in the experimental study and active control experienced a reduction of depressive symptoms over time. Depressive symptoms, especially mild ones, tend to decrease, even without intervention, on the account of time. On average, study participants in the intervention group accessed the myStrength platform 6.09 times during the 26-week study period. Receiving mental health treatment, such as outpatient therapy or taking antidepressant medication, was an independent predictor of depression score reduction over time (P = .026). The main effect of time was statistically significant, which means that, overall, a reduction of 0.83 in the depression score was achieved at each time point (P = 0.002). This main effect finding is qualified by the interaction group x time (P <.001). Taking into account potential confounding factors, the experimental group experienced an accelerated course of reducing symptoms of depression to a factor 1.35 times faster than the control group.
Sarfati 2017[38]	Clinical trial that seeks to compare differences in productivity and functionality at work between intervention groups of patients with depression treated with escitalopram and eight weekly sessions of cognitive behavioral therapy by telephone vs. control group treated with escitalopram. The Montgomery-Åsberg Depression Rating Scale (Madrs), Employment Absence and Productivity Scale (Leaps), Work Performance Questionnaire (HPQ), and Sheehan Disability Scale (SDS) questionnaires were applied.	Among the 99 randomized patients, 86 completed the 12-week study. Remission of symptoms, irrespective of treatment, was associated with significant improvement in work performance, by the Leaps productivity subscale, by the overall HPQ performance, and by the SDS work/school item; a non-significant trend (P = 0.08) was observed with the HPQ productivity subscale. Effect sizes indicate small to medium effects likely to be clinically significant.
Imamura 2015[39]	Clinical trial that investigated the preventive effect for depression in Japanese IT company workers from an online cognitive behavioral therapy program. Intervention group (n = 381) participated in six weekly sections of the online program, control group (n = 381) received e-mails with nonspecific monthly stress management tips over six months. To evaluate the incidence of depression after 6 and 12 months, the Beck Depression Inventory - I (BDI-II) and Kessler's Psychological Distress Scale (K6) questionnaires were used.	At the end of six months, 272 (71.4%) participants in the intervention group and 320 (84.0%) of the control group completed the follow-up study. After 12 months, 239 (62.7%) participants from the intervention group and 272 (71.4%) from the control group completed the follow-up research. Dropout rates were significantly higher in the intervention group at both the six-month follow-up (p <0.01) and at the 12-month follow-up (p = 0.01). Participants in the intervention group had a significantly lower incidence than the control group at the six-month follow-up (p = 0.07) and a significantly lower incidence at the 12-month follow-up (p <0.01). The hazard ratio (HR) of major depression for the intervention group compared to the control group was 0.22 (95% CI 0.06-0.75) during the 12-month follow-up. At six months of follow-up, the relative risk (RR) of having major depression (MD) in the intervention group was 0.30 (p> 0.05). In the 12-month follow-up, the relative risk of having major depression in the intervention group was 0.20 (p <0.01); the number needed to achieve the prevention of one case of MD onset was 32 (95% CI 19-100).
Lerner 2014[40]	The study aimed to evaluate the improvement of the functionality at work in workers over 45 years of age with depression and with limitations at work. The intervention group was exposed to eight telephonic sessions of cognitive behavioral therapy of 50 minutes every two weeks with experienced psychologists. The control group received standard care of referral to the physician. A total of 380 participants completed the study. Patient Health Questionnaire-9 (PHQ-9), Primary Care Screener for Affective Disorder and Work Limitations Questionnaire (WLQ) were applied.	At baseline, 39% of participants in the overall sample had persistent depressive disorder, 25% had major depression, and 36% had both. Among those with persistent depressive disorder and those with persistent depressive disorder and major depression, 73% (N = 192 of 264) had moderate symptoms and 27% (N = 72 of 264) had severe symptoms. The intervention group improved significantly in each outcome, and the improvements were significantly higher than those observed for the usual care group. The loss of labor productivity improved significantly by 44% (p <0.001) in the intervention group compared with 13% (p <0.001) in the usual care group. Improvements measured by the four WLQ work performance scales were significant in favor of the intervention group. Absences decreased 53% in the WFI group (p <0.001) versus 13% in the usual care group (p = 0.31) (p <0.001 for the difference in change). The mean severity scores for depression symptoms dropped by 51% (p <0.001) in the intervention group versus 26% (p <0.001) in the usual care group (p <0.001 for the difference in change).
Volker 2015[41]	A clinical trial that evaluated, in workers with depression symptoms on leave for any disease for 4 to 26 weeks, the effects of an online intervention with content in five modules: psychoeducation, cognitive perception of return to work, problem-solving skills, management of pain and fatigue, prevention of relapse with the purpose of promoting partial or total return to work. The control group received usual care. The tool notifies via e-mail the occupational doctor that monitors and advises the worker during the leave.	A total of 220 workers were allocated initially in the intervention group (n = 131) and control (n = 89). Return to work presented higher and earlier rates of return to work in the intervention group. Partial or total return of 84% (72/86) in the intervention group vs. 87.7% (114/130) in the control group and total early return to work with difference of 47 days in the intervention group compared to the control group; however, this difference was not statistically significant.

Author	Methodology	Results
Imamura 2015[42]	762 IT workers were randomized into a clinical trial, which sought to evaluate the improvement of work engagement between intervention group exposed to a weekly online behavioral cognitive behavioral therapy program of 30 minutes in six modules vs. control group exposed to emails with simple stress management tips. After three and six months, work engagement was measured. The Utrecht Work Engagement Scale (Uwes), WHO Health and Work Performance Questionnaire (HPQ) and Beck Depression Inventory II (BDI-II) questionnaires were applied.	At the end of three months, 270 (70.9%) participants from the intervention group and 336 (88.2%) from the control group completed the follow-up study. At the six-month follow-up, 272 (71.4%) participants from the intervention group and 320 (84.0%) from the control group completed the follow-up research. The program showed a significant effect on Uwes (P = 0.04), but at low intensity of effect: 0.11 (95% CI, -0.05 to 0.27) at three months of follow-up and 0.16 95% CI, 0.0007 to 0.32) at six months follow-up. The program showed a marginally significant effect on days of sick leave in the last three months (P = 0.07), with small effect sizes: -0.16 (95% CI -0.32 to 0.0003) at three-month follow-up and -0.14 (95% CI -0.30 to 0.02) at the six-month follow-up. The conclusion of the study was positive results in the work engagement through the program, effect partially explained with the improvement of depression symptoms.
Kröger 2015[43]	26 workers on leave due to depression were divided between usual behavioral cognitive therapy (control group, n = 13) and work-related cognitive behavioral therapy. Each group received around 24 sessions of psychotherapy. The aim of the study was to compare the improvement of depression symptoms and absenteeism between the groups. Beck Depression Inventory (BDI), Life Satisfaction Questionnaire (FLZ) and Symptom-Checklist 90-Revised (GSI) questionnaires were applied.	All participants reported a BDI score ≥ 15 points. The mean pre-treatment BDI score was 20.58 (SD ± 5.20), indicating a moderate level of depression. Absenteeism was significantly reduced in both types of treatment. There was no difference between groups in terms of early partial return to work (p = 0.722). Eight versus six employees in the intervention and control group, respectively, returned to the work place on a part-time basis. For both types of treatment, BDI and GSI scores decreased significantly over time, while FLZ scores increased significantly.
Geraedts 2014[44]	The clinical trial tested the effects on absenteeism and symptoms of depression in workers not on leave for depression from an online Happy @ Work self-help program compared to usual care. This tool is based on problem-solving treatment (PST) and cognitive therapy (CT) and a guideline to help employees prevent work-related stress. The Trimbos and iMTA Questionnaire on Costs Associated with Psychiatric Illness (TiCP), Short Form Health and Labor Questionnaire (SFHLQ), Center for Epidemiologic Studies Depression Scale (CESD), Maslach Burnout InventoryGeneral Scale (MBI), WHO Health and Work Performance Questionnaire (HPQ) were applies, among others.	A total of 231 participants were included in the study, of which 116 were in the intervention group and 115 in the control group. Of the 231 participants, 10 (4.3%) used medication without psychological treatment, 24 (10.4%) received psychological treatment, but no medication, and four participants (1.7%) used both medications and received psychological treatment in the beginning of the study. All participants had improved depressive symptoms and this improvement was sustained over time. However, the estimated overall mean difference between groups over time was not significant. There were improvements between baseline and posttreatment assessment in the secondary outcomes, and these improvements were sustained over time, but there were no significant differences between groups over time. The total between the effect sizes of the group for secondary outcomes was small. The result of absenteeism was expressed in the duration of absenteeism during the period between two evaluations. Therefore, it is not possible to study whether there was an increase or decrease in the duration of absenteeism over time, but only the differences between the groups regarding the duration of absenteeism can be examined. The estimated overall mean difference between groups over time was not significant.
Tan 2014[45]	Systematic review of clinical trials of any intervention in an occupational setting for the prevention of depression. Articles were searched in Medline, PsycInfo and Embase databases.	Most of the included studies used cognitive behavioral therapy (CBT) techniques. The overall standardized mean difference (SMD) between the intervention and control groups was 0.16 (95% CI 0.07, 0.24, P = 0.0002), indicating a small positive effect. A separate analysis using only CBT-based interventions generated a significant SMD of 0.12 (95% CI 0.02, 0.22, P = 0.01). The results indicate that a number of different depression prevention programs produce small positive effects in the workplace. When analyzed separately, CBT-based interventions significantly reduced levels of depressive symptoms among workers. These results demonstrate that appropriate evidence-based interventions in the workplace should be part of efforts to prevent the development of depression. In conclusion, there is good quality evidence that universal mental health interventions can reduce the overall level of depression symptoms in workers. Specifically, CBT-based interventions in the workplace are effective in reducing universal symptoms for depression. Further research is required to determine the extent to which such interventions can prevent new cases of depression and establish economic and practical strategies for large-scale implementation. Overall, the results of this review provide support for occupational mental health interventions and raise the imperative that depression should no longer be ignored in health promotion programs in the workplace.
Phillips 2014[46]	Clinical trial that evaluated the effects on depression symptoms in workers of an online self-help program based on cognitive behavioral therapy, lasting five weeks and evaluating the effects after 6 and 12 weeks. The program has five modules of one hour each and weekly. The Work and Social Adjustment Scale (WSAS) and Patient Health Questionnaire-9 (PHQ-9) questionnaires were used.	In the experimental and control groups, depression scores improved in six weeks, but the loss of participants was high. There was no evidence of a difference in the median effect of MoodGYM treatment on WSAS, nor for a difference in any of the secondary outcomes.

Author	Methodology	Results
Lam 2013[47]	The clinical trial sought to compare improvement in symptoms of depression and functionality at work among workers with major depression with participants in a group exposed to cognitive behavioral therapy (CBT) by phone + escitalopram and a group exposed only to escitalopram. The Montgomery-Asberg Depression Rating Scale (Madrs), Sheehan Disability Scale (SDS), Lam Employment Absence and Productivity Scale (Leaps), Health and Work Performance Questionnaire (HPQ) were applied.	After randomization, 48 were allocated to the telephone CBT group and 51 to the escitalopram group. There were 40 (83%) patients evaluated at the outcome of 12 weeks for the CBT + escitalopram group and 46 (90%) for the escitalopram group. There was significant improvement in Madrs change scores within each treatment condition, with large start point effect sizes at the end. However, there was no significant difference between groups. In the analysis, response rates were 63% in the CBT + escitalopram group and 61% in the escitalopram group, and remission rates were 56% and 53%, respectively (p = 0.74). The SDS item of work/function showed improvement within each treatment condition, but there were no significant differences between the treatment conditions. The other work scales showed significant differences between the treatment conditions favoring CBT + escitalopram.
Hees 2013[48]	Clinical trial evaluating the effects of sessions with occupational therapists on return to work of workers on leave for at least eight weeks due to major depression compared to workers who received usual care. A total of 18 sessions, nine individual, eight in group and one with the employer were applied in the intervention group. The outcomes measured were the rates and delay of partial or total return to work, absenteeism and variation of the depression symptoms. The Hamilton Rating Scale for Depression (HRSD) and Utrecht Coping List (UCL) questionnaires were applied, among others.	Both groups decreased significantly in their hours of absenteeism (p <0.001), with the largest decrease between 6 and 12 months (p <0.001). However, there were no significant differences between the groups. Over time, participants in the intervention group had greater improvement in depression symptoms than in the control group, both in terms of severity (p = 0.03) and long-term remission (HRSD ≤ 7, OR = 1, 8, 95% CI 1.0 to 3.3). In addition, the percentage of participants achieving sustainable remission - defined as remission for ≥ 6 months - was higher in the intervention group (92%) than in the control group (69%; p = 0.04). Both groups decreased significantly in their work limitations (all three WLQ scales p <0.001), with the greatest decrease between 6 and 12 months (Output: p = 0.01, Time Management: p = 0.02, Mental/ Interpersonal: p = 0.02). Likewise, both groups increased the work efficiency (p <0.001), with the highest increase between 6 and 12 months (p = 0.01). However, no significant differences were found between groups for these measures.
Vlasveld 2013[49]	The clinical trial sought to evaluate the effectiveness of a collaborative care program to promote return to work (RTW) in workers on leave for 4 to 12 weeks for major depression compared to usual care. The intervention involves 6 to 12 sessions of problem-solving therapy, use of antidepressants in selected cases, self-help manuals, and meetings between worker and employer. The Patient Health Questionnaire (PHQ-9) was used.	126 participants were included in the study and were randomized to either the usual care group (N = 61) or the collaborative care group (N = 65). Participants in collaborative care did not differ significantly from usual care participants in the odds of not achieving remission or response from depression (p> 0.05). For the participants who achieved remission, the mean time to first remission was 6.5 months in the collaborative care group (N = 27/65) and 7.9 months in the usual care group (N = 29/61). Within one year of follow-up, 64.6% of collaborative care participants and 59.0% of usual care participants had achieved long-lasting and complete RTW. The mean duration of complete and long-term RTW, calculated from day of randomization, was 190 days (DP of 120 days) in the collaborative care group and 210 days (DP of 124 days) in the usual care group.
Raiskila 2013[50]	The study evaluated the effectiveness of a multiprofessional program involving psychotherapist, psychiatrist, social worker, among others, in order to promote support for professional and personal stressors. The program offers courses and sessions based on cognitive behavioral therapy and principles of psychodynamics, workplace visits and assessments associated with employer support and occupational health service, family support with convening family members. The Beck Depression Inventory (BDI) and the Structured Clinical Interview for DSMIV (SCID I and II) were applied.	A total of 355 individuals were referred to the project, and 283 of them were randomized to the intervention (N = 142) and control (N = 141) groups. The selected workers had BDI scores higher than 9. According to SCID I interviews, 34.3% of participants in the intervention group (N = 134) presented mild depression, 59.0% moderate and 6.7% severe depression at the beginning of the study. In the control group (N = 100), the respective rates were 49.0%, 45.0% and 6.0%. The mean BDI score at the beginning of the study was 20.8 in the intervention group and 19.3 in the control group and, after one year of follow-up, 9.1 and 8.8, respectively. The mean reduction in BDI scores in the intervention group was 11.6 and 10.8 in the control group. The decrease was statistically significant in both groups (P <0.001). The only significant difference between the study groups was the decrease in BDI scores above 9 points during the one-year follow-up period, which occurred in two-thirds of the intervention group and half of the control group (P = 0.013).
Ahola 2012[51]	The study sought to promote the prevention of active workers through skills training sessions. The intervention consisted of four half-day sessions, which were held for one or two weeks. The skills involved principles of lifelong learning, organizational change management practice, social conflict resolution, career management, among others. The Beck Depression Inventory (BDI) questionnaire and the Job Content Questionnaire were applied.	A total of 43 medium and large organizations were contacted and had the opportunity to participate in a study about an intervention program. The final study population consisted of 566 people (79%), with 296 (80%) in the intervention group and 270 (77%) in the comparison group. In the follow-up, the odds of depression were lower in the intervention group (OR = 0.40 95% CI 0.19 to 0.85) than in the comparison group when adjusted for initial depressive symptoms, work stress, and demographic data. The odds of depression among those with stress at work (OR = 0.15, 95% CI 0.03 to 0.81) were lower after the intervention. There was no statistically significant effect among those with depressive symptoms at baseline.

Author	Methodology	Results
Lerner 2012[52]	The clinical trial sought to evaluate the efficacy of a multifaceted telephonic program for workers with depression and lost productivity at work. The program involves coaching, coordination of care with negotiation of a therapeutic plan, education about depression and its treatment, referral to medical treatment, use of cognitive behavioral therapy strategies. The Work Limitations Questionnaire (WLQ) and Patient Health Questionnaire 9 (PHQ-9) questionnaires were applied.	79 workers were randomized into the control group (27) and the intervention group (52). All results improved significantly (p <0.01) in the intervention group. The results from the control group were worse compared to baseline or did not improve significantly. The magnitude of the improvement in all outcomes was significantly higher in the intervention group (p <0.01). Within the intervention group, job performance improved an average of 18.1 points for time management, 10.9 points for physical tasks, 11.8 points for interpersonal mental work tasks and 14.2 points for exit tasks. These represent between 20% and 50% of the initial mean of the WLQ scale. The loss of work productivity for the intervention group declined from 10.3% (SD = 4.3) to 6.8% (SD = 4.3), for an average improvement of 3.5 percentage points (p < 0.01). Absence in the intervention group improved from 1.7 (SD = 1.7) to 1.0 day (SD = 1.2 p <0.01). Loss of productivity due to absences improved 7.1% (p <0.01).
Furukawa 2012[53]	Clinical trial that sought to evaluate the effectiveness of an eight-session cognitive behavioral therapy program by telephone in active workers with the purpose of reducing presenteeism and symptoms of depression. In both the control group and the intervention group, a worker assistance program for diagnosis and stress reduction was available. The K6, Health and Work Performance Questionnaire (HPQ) and Beck Depression Inventory II (BDI-II) were used.	The planned sample size was 108 per group, but the test was interrupted early because of the low participation rate. In total, 118 individuals were randomized, of which 58 were in the intervention group and 60 in the control group. The BDI-II scores decreased from the mean of 17.3 at the baseline to 11.0 in the intervention group and to 15.7 in the control group after four months (p <0.001), with effect size at 0.69 (95% CI 0.32 to 1.05). However, there was no statistically significant reduction in absolute or relative presenteeism.
Sandahl 2011[54]	A sample of 120 office workers on prolonged leave of over 90 days due to work-related depression was distributed to focal psychodynamic therapy (FGT), cognitive therapy (CGT) or a comparison group (CC). The study sought to evaluate the effectiveness of the intervention in promoting return to work.	In the six-month telephone interview, 74% of the patients in the therapy group reported that they were satisfied or very satisfied with the group therapy treatments, and 75% reported that they felt better or recovered. There was no difference in this respect between the two group treatments. In the 12-month follow-up, 69% reported that they were working part-time or longer. The pattern of return to work differed somewhat between groups, but this can be explained by different patterns of inclusion. Therefore, the conclusion was that even with regard to return to work, there was no difference between the three treatment conditions.
Lexis 2011[55]	Clinical trial sought to evaluate the effectiveness of 10-12 sessions of psychotherapy based on principles of problem-solving therapy and cognitive behavioral therapy in the prevention of long-term absenteeism in high-risk workers with symptoms of depression. The Balansmeter, Hospital Anxiety and Depression Scale (HAD Scale), Short Form Health Survey (SF-36), Job Content Questionnaire, among others, were applied.	A total of 139 employees were included in the study and randomized to the intervention group (n = 69) or the control group (n = 70). A significant difference in the total duration of absence due to illness was found between the intervention (27.5 days) and the control group (50.8 days) at the 12-month follow-up with a 46% reduction (p = 0.017). The intervention group showed a non-significantly lower proportion of long-term absenteeism episodes compared to the control group (p = 0.127) at the 12-month follow-up. Regarding the complaints of depression, significant differences were observed after 6 and 12 months of follow-up in favor of the intervention group, with a reduction of 19.2% and 19.8%, respectively, with a number needed to treat (NNT) in 5.2 (95% CI 2.7 to 55.5) and 5.0 (95% CI 2.7 to 32.1), respectively.
Farzanfar 2011[56]	Clinical trial comparing the efficacy of an automated screening program associated with psychoeducation, counseling for self-help measures, and specialized follow-up versus isolated, telephone-based screening for workers with stress and untreated symptoms. The questionnaires used were Work Limitation Questionnaire, Medical Outcomes Questionnaire Short Form-12, Patient Health Questionnaire 9, Perceived Stress Scale 4 and WHO-Five Well-being Index.	A total of 164 workers were randomized into the intervention (87) and control (77) groups. Those in the intervention group showed a significantly greater reduction in depression (p ≤ 0.05) at three months, a non-significant improvement in overall mental health at six months (p ≤ 0.10), as well as a significantly greater improvement in subscale of the work limitation questionnaire, at the mental-interpersonal scale (p ≤ 0.05) at three months and at the time and scheduling scale (p ≤ 0.05) at six months. Based on the analysis, participants in the intervention group found the system easy to use (84% reporting very easy or slightly easy to use), friendly (80% very or slightly friendly), appropriately rhythmic (67%) and informative (76% reporting very or slightly informative). In addition, 65% reported that the system was very or partially useful and 47% agreed that the system reduced the time spent at the doctor.
Bee 2010[57]	Clinical trial evaluated the effects of telephone sessions of cognitive behavioral therapy on symptoms of depression and productivity in workers with symptoms of depression and history of absenteeism in the last ten months. The questionnaires applied were the 34-item Clinical Outcomes in Routine Evaluation Outcome measure (Core-OM), Hospital Anxiety & Depression Scale (Hads), Work and Social Adjustment Scale (WSAS) and WHO Work Performance Questionnaire.	53 workers were randomized, of which 26 were in the intervention group. Although the clinical results were not statistically significant, the direction of the effect favored the intervention, which was associated with moderate effects sizes in clinical outcomes and in the labor productivity score.

Author	Methodology	Results
Martin 2009[58]	Systematic review and meta-analysis of published clinical trials between 1997 and 2007 to evaluate the effects of occupational health promotion programs on symptoms of depression or anxiety. The intervention aimed at mental health, directly or indirectly, through a known risk factor for depression or anxiety. The revised interventions were limited to those delivered at the workplace.	In all, 22 studies met the inclusion criteria, with a total sample of 3,409 post-intervention workers, and 17 of these studies were included in the meta-analysis, representing 20 intervention-control comparisons. The pooled results indicated small but positive overall effects of interventions for symptoms of depression [SMD 0.28, 95% confidence interval (CI 95%), 0.12-0.44] and anxiety [SMD 0.29, 95% CI 0.06-0.51), but no effect on composite mental health measures [SMD 0.05, 95% CI 0.03-0.13). Interventions that included a direct focus on mental health had benefit in the symptoms of depression and anxiety, as well as interventions with an indirect focus on risk factors.
Nakao 2007[59]	A cohort study of 283 Japanese workers exposed to a worker assistance program (EAP) aimed at improving the symptoms of depression. The program offered psychological counseling by email or telephone, which was free and anonymous, and referral to a psychiatric clinic affiliated with the institute. In addition, work-related mental health seminars were held for all employees five times a year. The Hamilton Depression Scale (HAM-D) and Job Content Questionnaire (JCQ) questionnaires were applied.	In the EAP group, scores on the full-scale HAM-D decreased significantly during the study. The following results were obtained for the total sample: HAM-D scores decreased by 149 people (53%), remained unaltered in 27 people (10%) and increased in 107 people (38%). In the EAP group, changes in the HAM-D scores for suicidal thoughts, agitation, psychomotor retardation, guilt, and depressed mood were significant. Specifically, 22 men in the EAP group responded positively to the suicidal thoughts item of HAM-D at baseline. The total scores of these 22 subjects decreased significantly over the two-year period, and 19 subjects (86%) reported no suicidal thoughts at the end of the study period. The total HAM-D at the beginning of the study did not differ significantly between the PAD group and the reference group, and there were no significant changes in the total scores and items in the reference group during the intervention period. The three JCQ scores at baseline were not significantly different between the EAP group and the reference group.
Wang 2007[60]	A clinical trial that sought to evaluate the effectiveness of a telephone depression screening and monitoring program for workers with depression. The structured telephone intervention program systematically evaluated treatment needs, facilitated entry into personal treatment (psychotherapy and antidepressant medication), monitored and supported adherence to treatment, and offered a structured psychotherapeutic intervention by telephone. The Quick Inventory of Depressive Symptomatology (QIDS-SR) and WHO Health and Productivity Questionnaire (HPQ) questionnaires were applied.	QIDS-SR scores were significantly lower at intervention than the usual care group at 6 and 12 months. The proportion whose symptoms improved substantially (50% improvement in QIDS-SR) was also significantly higher among interventions than usual care, but not until the 12-month evaluation (30.9% vs. 21.6%, OR = 1.7). The proportion of participants with recovery (QIDS-SR ≤ 5) was also significantly higher in the intervention than the usual care group, but not before 12 months (26.2% vs. 17.7%, OR = 1.7). The scores in the effectiveness measure of hours worked were significantly higher in the intervention than in the usual care group at 6 and 12 months. This overall effect was due to significant improvements in job retention (92.6% vs. 88.0% in 12 months, OR = 1.7) and hours worked among the participants. Participants in the intervention group were significantly more likely than those in usual care to receive any special mental health treatment (OR = 1.6), but somewhat less likely to receive any treatment of depression in primary or non-medical care (OR = 0.6-0.7). The average number of treatment contacts in all occupations (including care manager contacts) was almost twice as high in the intervention versus the usual care group (12.7 vs. 6.5, t = 5.7, p <0.001).
Schene 2007[61]	Clinical trial that sought to evaluate the effects of occupational therapy added to the usual care in workers with depression and absenteeism. The intervention consists of multiple phases seeking to clarify the activity of the job, promote preparation for reintegration to work, monitor the progress of return to work and advise on difficulties of adaptation. The Beck Depression Inventory (BDI) and the Questionnaire Organization Stress (QOS) were used.	The percentage of patients who met the DSM-IV criteria for Major Depressive Episode decreased from 100% to 29% in the control group and to 44% in the intervention group. This recovery was statistically significant (p = 0.000), particularly in the first six months (p = 0.007), but not in the following six months (p = 0.23). The total BDI score decreased from 23.6 to 14.0 for the control group and from 27.1 to 12.3 for the intervention group, with significant interaction (p = 0.015), which did not occur in the 12 months (p = 0.950), but emerged between months 13 and 42 (p = 0.032). The time to return to work was measured between the groups. In the intervention group, the mean time to return to work was 207 days. 299 days in the control group, with a relative risk of RR = 2.71 (95% CI, 1.16-6.29, p = 0.01).
Mino 2006[62]	The clinical trial sought to evaluate the effectiveness of a stress reduction program in preventing depression in workers who engaged in high-stress activities. The stress management program included lectures on stress perception, measures to address it, stress management records, and e-mail counseling. The General Health Questionnaire (GHQ), Center for Epidemiologic Study for Depression (CES-D), Questionnaire of Work-Related Stress and Effort-Reward Imbalance Questionnaire were used.	58 workers were randomized into intervention group (28) and control group (30). In the intervention group, a significant improvement in the depressive symptoms was observed, compared to the control group through the CES-D questionnaire. In the multiple regression analysis, the effect of stress control on depressive symptoms at follow-up was significant (p = 0.041).

Author	Methodology	Results
Smith 2002[63]	The clinical trial evaluated the effects of depression treatment offered by 12 primary care centers on reducing rates of unemployment and interpersonal conflict at work in workers with depression. Primary care physicians worked on providing treatment for depression and nurses worked on monitoring symptoms and adherence. Of the 262 patients in the baseline sample eligible for this analysis, 219 (83.6%) were followed up at one year.	The intervention significantly increased subsequent employment in one year by 10.1% (p = 0.04, CI 90% 2.8-17.4%), reduced unemployment by 5.8% (CI 90% 1.6 -10.0%) and underemployment at 4.3% (CI 90% 1.2-7.4%). Of the difference of 10.1% in employment between intensive and usual care patients in one year, 3.4 percentage points are explained by the intervention. The additional 6.7 percentage points represents the ability to intervene to reduce the length of the periods of unemployment/underemployment among those who reported unemployment/underemployment in six months. Among those employed in one year, intensive care patients were significantly less likely than usual patients to report workplace conflicts in the following year (8.1% vs. 18.9%, p = 0.04).

EVIDENCE APPLICATION – RECOMMENDATION

For the presentation of the recommendations, the suggestion of conduct was made with the preparation of the recommendations by the authors of the technical guideline, considering the characteristics of the evidence synthesis and being submitted for validation of all the authors participating in the work group. The degree of recommendation comes directly from the available strength of the studies included according to Oxford[69] and the use of the Grade[70] system.

Conflict of interest

There is no conflict of interest related to this review to be declared by any of the authors.

Final statement

The Guidelines Project, an initiative of the Brazilian Medical Association in conjunction with the Specialty Societies, aims to reconcile medical information in order to standardize behaviors that aid the physician's reasoning and decision making. The information contained in this project should be submitted to the evaluation and critique of the physician responsible for the conduct to be followed, in view of the reality and clinical condition of each patient.

REFERENCES

1. Public Health Agency of Canada Mood disorders. In: The human face of mental health and mental illness in Canada 2006. Ottawa (ON): The Agency; 2006. p. 57?70 Available: www.phac-aspc.gc.ca/publicat/human-humain06/index-eng.php.
2. American Psychiatric Association Diagnostic and statistical manual of mental disorders. Fourth edition Text revision. Washington (DC): The Association; 2000
3. Buckman JEJ, Underwood A, Clarke K, Saunders R, Hollon SD, Fearon P et al. Risk factors for relapse and recurrence of depression in adults and how they operate: A four-phase systematic review and meta-synthesis. Clinical psychology review; 2018
4. Barros MBA, Lima MG, Azevedo RCS., Medina LBP, Lopes CS, Menezes PR et al. Depression and health behaviors in Brazilian adults?PNS 2013. Revista de saude publica 2017, 51, 8s.
5. Silva MT, Galvao TF, Martins SS, Pereira MG. Prevalence of depression morbidity among Brazilian adults: a systematic review and meta-analysis. Rev Bras Psiquiatr. 2014 Sep;36(3):262-70.
6. Anuario Estatistico da Previdencia Social, Ministerio da Fazenda, Secretaria de Previdencia, Empresa de Tecnologia e Informacoes da Previdencia. Anuario Estatistico da Previdencia Social 2016. Brasilia: MF/DATAPREV.
7. Harvey SB, Modini M, Joyce S, Milligan-Saville JS, Tan L, Mykletun A et al. Can work make you mentally ill? A systematic meta-review of work-related risk factors for common mental health problems. Occup Environ Med. 2017 Mar;74(4):301-310.
8. Oenning NSX, Ziegelmann PK, Goulart BNG, Niedhammer I. Occupational factors associated with major depressive disorder: A Brazilian population-based study. J Affect Disord. 2018 Nov;240:48-56.
9. US Preventive Services Task Force recommendation statement Screening for depression in adults. Ann Intern Med 2009;151: 784?92
10. Åhlin JK, Rajaleid K, Jansson-Fröjmark M, Westerlund H, Magnusson Hanson LL. Job demands, control and social support as predictors of trajectories of depressive symptoms. J Affect Disord 2018; 235: 535-543. doi: 10.1016/j.jad.2018.04.067. PMID: 29689506.
11. Nakamura-Taira N, Izawa S, Yamada KC. Stress underestimation and mental health literacy of depression in Japanese workers: A cross-sectional study. Psychiatry Res 2018; 262: 221-228. doi: 10.1016/j.psychres.2017.12.090. PMID: 29471260.
12. Nieuwenhuijsen K, Sluiter JK, Dewa CS. Need for Recovery as an Early Sign of Depression Risk in a Working Population. J Occup Environ Med 2016; 58: e350-e354. PMID: 27820770.
13. Wang J, Lam RW, Ho K, Attridge M, Lashewicz BM, Patten SB, et al. Preferred Features of E-Mental Health Programs for Prevention of Major Depression in Male Workers: Results From a Canadian National Survey. J Med Internet Res 2016; 18: e132. doi: 10.2196/jmir.5685. PMID: 27267782.
14. Volker D, Zijlstra-Vlasveld MC, Brouwers EP, Homans WA, Emons WH, van der Feltz-Cornelis CM. Validation of the Patient Health Questionnaire-9 for Major Depressive Disorder in the Occupational Health Setting. J Occup Rehabil 2016; 26: 237-44. doi: 10.1007/s10926-015-9607-0. PMID: 26377480.
15. Wada K, Tanaka K, Theriault G, Satoh T, Mimura M, Miyaoka H, et al. Validity of the Center for Epidemiologic Studies Depression Scale as a screening instrument of major depressive disorder among Japanese workers. Am J Ind Med 2007; 50: 8-12. PMID: 17096372.
16. Weaver MD, Vetter C, Rajaratnam SMW, O'Brien CS, Qadri S, Benca RM, et al. Sleep disorders, depression and anxiety are associated with adverse safety outcomes in healthcare workers: A prospective cohort study. J Sleep Res 2018: e12722. doi: 10.1111/jsr.12722. [Epub ahead of print] PMID: 30069960.
17. Porru F, Burdorf A, Robroek SJW. The impact of depressive symptoms on exit from paid employment in Europe: a longitudinal study with 4 years follow-up. Eur J Public Health 2018. doi: 10.1093/eurpub/cky136. [Epub ahead of print]. PMID: 30052918.

18. Lamichhane DK, Heo YS, Kim HC. Depressive symptoms and risk of absence among workers in a manufacturing company: a 12-month follow-up study. Ind Health 2018; 56:187-197. doi: 10.2486/indhealth.2017-0065. PMID: 29225216.

19. Vendrig AA, Schaafsma FG. Reliability and Validity of the Work and Well-Being Inventory (WBI) for Employees. J Occup Rehabil 2018; 28: 377-390. doi: 10.1007/s10926-017-9729-7. PMID: 28887747.

20. Newcomb RD, Steffen MW, Breeher LE, Sturchio GM, Murad MH, Wang Z, et al. Screening for depression in the occupational health setting. Occup Med (Lond) 2016; 66: 390-3. doi: 10.1093/occmed/kqw043. PMID: 27154983.

21. van Hoffen MF, Joling CI, Heymans MW, Twisk JW, Roelen CA. Mental health symptoms identify workers at risk of long-term sickness absence due to mental disorders: prospective cohort study with 2-year follow-up. BMC Public Health 2015; 15: 1235. doi: 10.1186/s12889-015-2580-x. PMID: 26655203.

22. Torske MO, Hilt B, Bjørngaard JH, Glasscock D, Krokstad S. Disability pension and symptoms of anxiety and depression: a prospective comparison of farmers and other occupational groups. The HUNT Study, Norway. BMJ Open 2015; 5: e009114. doi: 10.1136/bmjopen-2015-009114. PMID: 26525724.

23. Rose U, March S, Ebener M, du Prel JB. Cut-off values for the applied version of the Beck Depression Inventory in a general working population. J Occup Med Toxicol 2015; 10: 24. doi: 10.1186/s12995-015-0067-4. PMID: 26191076.

24. Asami Y, Goren A, Okumura Y. Work productivity loss with depression, diagnosed and undiagnosed, among workers in an Internet-based survey conducted in Japan. J Occup Environ Med 2015; 57: 105-10. doi: 10.1097/JOM.0000000000000310. PMID: 25563547.

25. Ervasti J, Vahtera J, Pentti J, Oksanen T, Ahola K, Kivekäs T, et al. Return to work after depression-related absence by employees with and without other health conditions: a cohort study. Psychosom Med 2015; 77: 126-35. doi: 10.1097/PSY.0000000000000138. PMID: 25675157.

26. Wang YP, Gorenstein C. Attitude and impact of perceived depression in the workplace. Int J Environ Res Public Health 2014; 11: 6021-36. doi: 10.3390/ijerph110606021. PMID: 24914639.

27. Jain G, Roy A, Harikrishnan V, Yu S, Dabbous O, Lawrence C. Patient-reported depression severity measured by the PHQ-9 and impact on work productivity: results from a survey of full-time employees in the United States. J Occup Environ Med 2013; 55: 252-8. doi: 10.1097/JOM.0b013e31828349c9. PMID: 23439268.

28. Woo JM, Kim W, Hwang TY, Frick KD, Choi BH, Seo YJ, et al. Impact of depression on work productivity and its improvement after outpatient treatment with antidepressants. Value Health 2011; 14: 475-82. doi: 10.1016/j.jval.2010.11.006. PMID: 21669372.

29. Harvey SB, Glozier N, Henderson M, Allaway S, Litchfield P, Holland-Elliott K, et al. Depression and work performance: an ecological study using web-based screening. Occup Med (Lond) 2011; 61: 209-11. doi: 10.1093/occmed/kqr020. PMID: 21525074.

30. Munir F, Burr H, Hansen JV, Rugulies R, Nielsen K. Do positive psychosocial work factors protect against 2-year incidence of long-term sickness absence among employees with and those without depressive symptoms? A prospective study. J Psychosom Res 2011; 70: 3-9. doi: 10.1016/j.jpsychores.2010.09.014. PMID: 21193095.

31. Lexis MA, Jansen NW, Stevens FC, van Amelsvoort LG, Kant I. Experience of health complaints and help seeking behavior in employees screened for depressive complaints and risk of future sickness absence. J Occup Rehabil 2010; 20: 537-46. doi: 10.1007/s10926-010-9244-6. PMID: 20467796.

32. Lexis MA, Jansen NW, van Amelsvoort LG, van den Brandt PA, Kant I. Depressive complaints as a predictor of sickness absence among the working population. J Occup Environ Med 2009; 51: 887-95. doi: 10.1097/JOM.0b013e3181aa012a. PMID: 19625974.

33. Wang PS, Beck AL, Berglund P, McKenas DK, Pronk NP, Simon GE, et al. Effects of major depression on moment-in-time work performance. Am J Psychiatry 2004; 161: 1885-91. PMID: 15465987.

34. Druss BG, Schlesinger M, Allen HM Jr. Depressive symptoms, satisfaction with health care, and 2-year work outcomes in an employed population. Am J Psychiatry 2001; 158: 731-4. PMID: 11329394.

35. Lee Y, Rosenblat JD, Lee J, Carmona NE, Subramaniapillai M, Shekotikhina M, et al. Efficacy of antidepressants on measures of workplace functioning in major depressive disorder: A systematic review. J Affect Disord 2018; 227: 406-415. doi: 10.1016/j.jad.2017.11.003. PMID: 29154157.

36. Beiwinkel T, Eißing T, Telle NT, Siegmund-Schultze E, Rössler W. Effectiveness of a Web-Based Intervention in Reducing Depression and Sickness Absence: Randomized Controlled Trial. J Med Internet Res 2017; 19: e213. doi: 10.2196/jmir.6546. PMID: 28619701.

37. Hirsch A, Luellen J, Holder JM, Steinberg G, Dubiel T, Blazejowskyj A, et al. Managing Depressive Symptoms in the Workplace Using a Web-Based Self-Care Tool: A Pilot Randomized Controlled Trial. JMIR Res Protoc 2017; 6: e51. doi: 10.2196/resprot.7203. PMID: 28377368.

38. Sarfati D, Stewart K, Woo C, Parikh SV, Yatham LN, Lam RW. The effect of remission status on work functioning in employed patients treated for major depressive disorder. Ann Clin Psychiatry 2017; 29: 11-16. PMID: 27901522.

39. Imamura K, Kawakami N, Furukawa TA, Matsuyama Y, Shimazu A, Umanodan R, et al. Does Internet-based cognitive behavioral therapy (iCBT) prevent major depressive episode for workers? A 12-month follow-up of a randomized controlled trial. Psychol Med 2015; 45:1907-17. doi: 10.1017/S0033291714003006. PMID: 25562115.

40. Lerner D, Adler DA, Rogers WH, Chang H, Greenhill A, Cymerman E, et al. A randomized clinical trial of a telephone depression intervention to reduce employee presenteeism and absenteeism. Psychiatr Serv 2015; 66: 570-7. doi: 10.1176/appi.ps.201400350. PMID: 25726984.

41. Volker D, Zijlstra-Vlasveld MC, Anema JR, Beekman AT, Brouwers EP, Emons WH, et al. Effectiveness of a blended web-based intervention on return to work for sick-listed employees with common mental disorders: results of a cluster randomized controlled trial. J Med Internet Res 2015; 17: e116. doi: 10.2196/jmir.4097. PMID: 25972279.

42. Imamura K, Kawakami N, Furukawa TA, Matsuyama Y, Shimazu A, Umanodan R, et al. Effects of an internet-based cognitive behavioral therapy intervention on improving work engagement and other work-related outcomes: an analysis of secondary outcomes of a randomized controlled trial. J Occup Environ Med 2015; 57: 578-84. doi: 10.1097/JOM.0000000000000411. PMID: 25749132.

43. Kröger C, Bode K, Wunsch EM, Kliem S, Grocholewski A, Finger F. Work-related treatment for major depressive disorder and incapacity to work: preliminary findings of a controlled, matched study. J Occup Health Psychol 2015; 20: 248-58. doi: 10.1037/a0038341. PMID: 25402222.

44. Geraedts AS, Kleiboer AM, Twisk J, Wiezer NM, van Mechelen W, Cuijpers P. Long-term results of a web-based guided self-help intervention for employees with depressive symptoms: randomized controlled trial. J Med Internet Res 2014; 16: e168. doi: 10.2196/jmir.3539. PMID: 25008127.

45. Tan L, Wang MJ, Modini M, Joyce S, Mykletun A, Christensen H, Harvey SB. Preventing the development of depression at work: a systematic review and meta-analysis of universal interventions in the workplace. BMC Med 2014; 12: 74. doi: 10.1186/1741-7015-12-74. PMID: 24886246.

46. Phillips R, Schneider J, Molosankwe I, Leese M, Foroushani PS, Grime P, et al. Randomized controlled trial of computerized cognitive behavioural therapy for depressive symptoms: effectiveness and costs of a workplace intervention. Psychol Med 2014; 44: 741-52. doi: 10.1017/S0033291713001323. PMID: 23795621.

47. Lam RW, Parikh SV, Ramasubbu R, Michalak EE, Tam EM, Axler A, et al. Effects of combined pharmacotherapy and psychotherapy for improving work functioning in major depressive disorder. Br J Psychiatry 2013; 203: 358-65. doi: 10.1192/bjp.bp.112.125237. PMID: 24029535.

48. Hees HL, de Vries G, Koeter MW, Schene AH. Adjuvant occupational therapy improves long-term depression recovery and return-to-work in good health in sick-listed employees with major depression: results of a randomised controlled trial. Occup Environ Med 2013; 70: 252-60. doi: 10.1136/oemed-2012-100789. PMID: 23117218.

49. Vlasveld MC, van der Feltz-Cornelis CM, Adèr HJ, Anema JR, Hoedeman R, van Mechelen W, et al. Collaborative care for sick-listed workers with major depressive disorder: a randomised controlled trial from the Netherlands Depression Initiative aimed at return to work and depressive symptoms. Occup Environ Med 2013; 70: 223-30. doi: 10.1136/oemed-2012-100793. PMID: 23112266.

50. Raiskila T, Blanco Sequeiros S, Kiuttu J, Kauhanen ML, Läksy K, Vainiemi K, et al. The Impact of an Early Eclectic Rehabilitative Intervention on Symptoms in First Episode Depression among Employed People. Depress Res Treat 2013; 2013: 926562. doi: 10.1155/2013/926562. PMID: 24324883.

51. Ahola K, Vuori J, Toppinen-Tanner S, Mutanen P, Honkonen T. Resource-enhancing group intervention against depression at workplace: who benefits? A randomised controlled study with a 7-month follow-up. Occup Environ Med 2012; 69: 870-6. doi: 10.1136/oemed-2011-100450. PMID: 22718708.

52. Lerner D, Adler D, Hermann RC, Chang H, Ludman EJ, Greenhill A, et al.

Impact of a work-focused intervention on the productivity and symptoms of employees with depression. J Occup Environ Med 2012; 54: 128-35. doi: 10.1097/JOM.0b013e31824409d8. PMID: 22252528.

53. Furukawa TA, Horikoshi M, Kawakami N, Kadota M, Sasaki M, Sekiya Y, et al. Telephone cognitive-behavioral therapy for subthreshold depression and presenteeism in workplace: a randomized controlled trial. PLoS One 2012; 7: e35330. doi: 10.1371/journal.pone.0035330. PMID: 22532849.

54. Sandahl C, Lundberg U, Lindgren A, Rylander G, Herlofson J, Nygren A, et al. Two forms of group therapy and individual treatment of work-related depression: a one-year follow-up study. Int J Group Psychother 2011; 61: 539-55. doi: 10.1521/ijgp.2011.61.4.538. PMID: 21985258.

55. Lexis MA, Jansen NW, Huibers MJ, van Amelsvoort LG, Berkouwer A, Tjin A Ton G, et al. Prevention of long-term sickness absence and major depression in high-risk employees: a randomised controlled trial. Occup Environ Med 2011; 68: 400-7. doi: 10.1136/oem.2010.057877. PMID: 20924024.

56. Farzanfar R, Locke SE, Heeren TC, Stevens A, Vachon L, Thi Nguyen MK, et al. Workplace telecommunications technology to identify mental health disorders and facilitate self-help or professional referrals. Am J Health Promot 2011; 25: 207-16. doi: 10.4278/ajhp.100118-QUAN-14. PMID: 21192751.

57. Bee PE, Bower P, Gilbody S, Lovell K. Improving health and productivity of depressed workers: a pilot randomized controlled trial of telephone cognitive behavioral therapy delivery in workplace settings. Gen Hosp Psychiatry 2010; 32: 337-40. doi: 10.1016/j.genhosppsych.2010.01.006. PMID: 20430241.

58. Martin A, Sanderson K, Cocker F. Meta-analysis of the effects of health promotion intervention in the workplace on depression and anxiety symptoms. Scand J Work Environ Health 2009; 35: 7-18. PMID: 19065280.

59. Nakao M, Nishikitani M, Shima S, Yano E. A 2-year cohort study on the impact of an Employee Assistance Programme (EAP) on depression and suicidal thoughts in male Japanese workers. Int Arch Occup Environ Health 2007; 81: 151-7. PMID: 17492306.

60. Wang PS, Simon GE, Avorn J, Azocar F, Ludman EJ, McCulloch J, Petukhova MZ, et al. Telephone screening, outreach, and care management for depressed workers and impact on clinical and work productivity outcomes: a randomized controlled trial. JAMA 2007; 298: 1401-11. PMID: 17895456.

61. Schene AH, Koeter MW, Kikkert MJ, Swinkels JA, McCrone P. Adjuvant occupational therapy for work-related major depression works: randomized trial including economic evaluation. Psychol Med 2007; 37: 351-62. PMID: 17112401.

62. Mino Y, Babazono A, Tsuda T, Yasuda N. Can stress management at the workplace prevent depression? A randomized controlled trial. Psychother Psychosom 2006; 75: 177-82. PMID: 16636633.

63. Smith JL, Rost KM, Nutting PA, Libby AM, Elliott CE, Pyne JM. Impact of primary care depression intervention on employment and workplace conflict outcomes: is value added? J Ment Health Policy Econ 2002; 5: 43-9. PMID: 12529569.

64. Yunus WMAWM, Musiat P, Brown JS. (2018). Systematic review of universal and targeted workplace interventions for depression. Occup Environ Med, 75(1), 66-75.

65. Joyce S, Modini M, Christensen H, Mykletun A, Bryant R, Mitchell PB, et al. (2016). Workplace interventions for common mental disorders: a systematic meta-review. Psychological medicine, 46(4), 683-697.

66. Carolan S, Harris PR, Cavanagh K. (2017). Improving employee well-being and effectiveness: systematic review and meta-analysis of web-based psychological interventions delivered in the workplace. Journal of medical Internet research, 19(7).

67. Nigatu YT, Huang, J, Rao S, Gillis K, Merali Z, Wang J. (2019). Indicated Prevention Interventions in the Workplace for Depressive Symptoms: A Systematic Review and Meta-analysis. American journal of preventive medicine, 56(1), e23-e33.

68. Chu AHY, Koh D, Moy FM, Müller-Riemenschneider F. (2014). Do workplace physical activity interventions improve mental health outcomes? Occupational Medicine, 64(4), 235-245.

69. Oxford Centre for Evidence-based medicine – Levels of Evidence (March 2009). Disponível em: Levels of Evidence and Grades of Recommendations - Oxford Centre for Evidence Based Medicine. Disponível em URL: http://cebm.jr2.ox.ac.uk/docs/old_levels.htm.

70. Goldet G, Howick J. Understanding GRADE: an introduction. J Evid Based Med 2013; 6:50-4

Dual platelet antiaggregation therapy after myocardial revascularization surgery

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SUMMARY

Coronary artery bypass graft (CABG) is a consolidated treatment in patients with coronary artery disease (CAD) for both symptom control and improvement of prognosis. The patency of venous grafts is still the most vulnerable point of the surgical treatment since it presents a high prevalence of occlusion both in the immediate postoperative period and in the long-term follow-up. Aspirin plays a well-established role in this setting, and for a long time, clopidogrel use has been restricted to patients allergic to aspirin. Recently, subgroup analyses of studies with different anti-platelet therapies have shown reduced mortality and cardiovascular events in patients on dual anti-platelet antiplatelet therapy (DAPT) undergoing CABG, although such studies have not been designed to evaluate this patient profile. However, there is still an insufficient number of randomized studies using DAPT in this context, resulting in a disagreement between the European and American cardiology societies guidelines regarding their indication and generating doubts in clinical practice.

KEYWORDS: *Coronary artery bypass. Coronary artery disease. Aspirin. Platelet Aggregation Inhibitors.*

Myocardial revascularization surgery (CABG) has been performed since the 1960s. It is routine treatment in patients with coronary artery disease (CAD) to control symptoms or improve prognosis. The evolution of perioperative care and surgical techniques increased the percentage of complete revascularization and reduced morbidity and mortality related to the procedure over the years.

The patency of venous grafts is still the vulnerable point of the surgical treatment with a prevalence of 10%-15% of occlusion in the immediate postoperative period and 50% occlusion in ten years. One of the primary mechanisms of occlusion in the first year is graft thrombosis. The use of aspirin, from the immediate postoperative period, in patients who undergo CABG, is already well established, with a reduction of mortality and combined adverse cardiac and cerebrovascular events, without a significant increase in bleeding.[1-3]

The progression of the atherosclerotic disease even after CABG is not unusual, culminating in adverse cardiac events. After all, the surgical procedure consists only of correcting the coronary lesions existent at the time of the procedure. Aspirin has a well-established role in this scenario, and for a long time, the use of clopidogrel was restricted to patients allergic to aspirin. However, the association of antiplatelet therapy has been shown to be beneficial in

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curbing the progression of atherothrombosis and to the viability of the venous grafts, in view of the possible additional effect of clopidogrel in reducing intimal hyperplasia, one of the main mechanisms of chronic coronary occlusion of venous grafts, an action which is not evidenced with the use of aspirin.[4]

Thus, given the possible benefits of inhibitors of P2Y12 receptors, the study TiCAB[5], randomized and double-blind, assessed the use of aspirin compared with Ticagrelor, both in monotherapy in patients after surgical revascularization. After a one-year follow-up, the study, which was interrupted early due to lack of funding by the sponsor, did not show the superiority of the use of Ticagrelor in comparison to the use of aspirin for reducing cardiovascular outcomes and cerebrovascular diseases.

Since the monotherapy with the P2Y12 inhibitors did not show better results when compared with aspirin, it was suggested that Dual platelet antiaggregation therapy (Dapt) could be superior to the use of only one antiplatelet agent after CABG. In this sense, the eight-year follow-up of the Cascade[6] study was published, comparing the effectiveness of Dapt with clopidogrel *versus* ASA and placebo. In this study, there was no difference between mortality or more significant cardiovascular events between the groups. Although there is less progression of atherosclerosis in the native beds, the patency of the grafts remained equal between the groups, as assessed using coronary angiography.

On the other hand, it is estimated that 30% of the population has some degree of resistance to clopidogrel. In acute coronary syndrome, for example, the effectiveness and safety of other antiplatelets, prasugrel or ticagrelor, are well established. Thus, several studies are being conducted to extend the indication of these platelets in relation to clopidogrel.

A subgroup analysis of the Plato[7] study, which evaluated patients submitted to surgical revascularization, demonstrated a reduction of mortality and cardiovascular events in patients using dual antiplatelet dual with ticagrelor in comparison with Dapt with clopidogrel; however, the study was not designed to evaluate this profile of patients. Since the evidence remains scarce in the postoperative context, some randomized studies are being carried out to assess the real benefit of Dapt after CABG.

The Dacab trial[8], an open and randomized study, evaluated the patency of venous grafts one year after CABG as a primary outcome in 500 patients into three branches (ASA, ticagrelor, and Dapt). A total of 12.2% was observed in patency favorable to the Dapt group (88.7%) in relation to the ASA group (76.5%), with no significant difference between the ASA and ticagrelor groups. The rate of events was low, with no difference in mortality and cardiovascular events in the three branches. There was also an increase of major bleedings between groups.

The POPular trial[9], currently at the randomization stage and expected to wrap up this year (NCT02352402), has as its objective to compare the patency of venous graft after a year between Dapt (ticagrelor and ASA) and monotherapy (ASA and placebo). The results of this study can contribute to a better understanding of the role of dual antiplatelet in surgical revascularization.

Although the majority of studies evaluate the patency of venous grafts as the primary outcome, there is still a difficulty in correlating the reduction of cardiovascular events and the patency of the grafts. The Prevent IV[10] study demonstrated that the occlusion of venous grafts was not correlated with an increase of acute myocardial infarction in four years.

In summary, we found an insufficient quantity of randomized studies with the use of Dapt in patients undergoing CABG. New studies, preferably with clinical outcomes, therefore, are required to better consolidate specific recommendations after the procedure, since the lack of evidence results in an inconsistency between the recommendations of the American and European Societies of Cardiology.[11, 12]

In stable patients, for example, the first considers the possibility of Dapt after surgical revascularization, while the second considers that the data are insufficient for such recommendation. In our practice, for stable patients submitted to CABG who did not make use of cardiopulmonary bypass and/or patients considered at low risk of bleeding, we consider the use of Dapt for 12 months.

For acute patients, to whom CABG is the revascularization therapy applied in a smaller proportion of cases (approximately 10% of the patients in the Plato[7] study), our position is favorable to the use of Dapt for at least 12 months. This recommendation is consolidated and referenced by the most recent European and American guidelines, both with class I recommendation.

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RESUMO

A cirurgia de revascularização miocárdica (CRM) é tratamento fundamental em pacientes com doença arterial coronariana (DAC) tanto para controle de sintomas quanto para melhora do prognóstico. A patência dos enxertos venosos ainda hoje é o ponto mais vulnerável do tratamento cirúrgico, por apresentar alta prevalência de oclusão tanto no pós-operatório imediato como no seguimento em longo prazo. A aspirina tem papel bem estabelecido neste cenário e, por muito tempo, o uso do clopidogrel ficou restrito a pacientes alérgicos a aspirina. Recentemente, análises de subgrupos de estudos com diferentes terapias antiplaquetárias demonstraram redução de mortalidade e eventos cardiovasculares em pacientes em uso de dupla antiagregação plaquetária (Dapt) submetidos à CRM, ainda que tais estudos não tenham sido desenhados para avaliar este perfil de pacientes. Contudo, há ainda uma quantidade insuficiente de estudos randomizados com uso de Dapt nesse contexto, resultando em uma discordância entre as diretrizes europeia e americana de cardiologia quanto à sua indicação e gerando dúvidas na prática clínica.

PALAVRAS-CHAVE: *Ponte de artéria coronária. Doença da artéria coronariana. Aspirina. Inibidores da agregação de plaquetas.*

REFERENCES

1. Motwani JG, Topol EJ. Aortocoronary saphenous vein graft disease: pathogenesis, predisposition, and prevention. Circulation. 1998;97(9):916-31.
2. Goldman S, Copeland J, Moritz T, Henderson W, Zadina K, Ovitt T, et al. Saphenous vein graft patency 1 year after coronary artery bypass surgery and effects of antiplatelet therapy. Results of a Veterans Administration Cooperative Study. Circulation. 1989;80(5):1190-7.
3. Mangano DT; Multicenter Study of Perioperative Ischemia Research Group. Aspirin and mortality from coronary bypass surgery. N Engl J Med. 2002;347(17):1309-17.
4. Herbert JM, Dol F, Bernat A, Falotico R, Lalé A, Savi P. The antiaggregating and antithrombotic activity of clopidogrel is potentiated by aspirin in several experimental models in the rabbit. Thromb Haemost. 1998;80(3):512-8.
5. ClinicalTrials.gov [Internet]. Bethesda (MD): National Library of Medicine - Identificador NCT 01755520, Study comparing ticagrelor with aspirin for prevention of vascular events in patients undergoing CABG (TiCAB); 24 Dezembro 2012. Disponível em: https://clinicaltrials.gov/ct2/show/NCT01755520.
6. Hage A, Voisine P, Erthal F, Larose É, Glineur D, Chow B, et al. Eight-year follow-up of the Clopidogrel After Surgery for Coronary Artery Disease (CASCADE) trial. J Thorac Cardiovasc Surg. 2018;155(1):212-22.
7. Wallentin L, Becker RC, Budaj A, Cannon CP, Emanuelsson H, Held C, et al; PLATO Investigators. Ticagrelor versus clopidogrel in patients with acute coronary syndromes, N Engl J Med. 2009;361(11):1045-57.
8. Zhao Q, Zhu Y, Xu Z, Cheng Z, Mei J, Chen X, Wang X. Effect of Ticagrelor Plus Aspirin, Ticagrelor Alone, or Aspirin Alone on Saphenous Vein Graft Patency 1 Year After Coronary Artery Bypass Grafting: A Randomized Clinical trial. JAMA. 2018; 319 (16): 1677-1686.
9. ClinicalTrials.gov [Internet]. Bethesda (MD): National Library of Medicine - Identificador NCT02352402, The effect of ticagrelor on saphenous vein graft patency in patients undergoing coronary artery bypass grafting surgery (POPular CABG); 2 Fevereiro, 2015. Disponível em: https://clinicaltrials.gov/ct2/show/NCT02352402.
10. Alexander JH, Hafley G, Harrington RA, Peterson ED, Ferguson TB Jr, Lorenz TJ; PREVENT IV Investigators. Efficacy and safety of edifoligide, an E2F transcription factor decoy, for prevention of vein graft failure following coronary artery bypass graft surgery: PREVENT IV: a randomized controlled trial. JAMA. 2005;294(19):2446-54.
11. Levine GN, Bates ER, Bittl JA, Brindis RG, Fihn SD, Fleisher LA, et al. 2016 ACC/AHA guideline focused update on duration of dual antiplatelet therapy in patients with coronary artery disease: a report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines. An Update of the 2011 ACC/AHA/SCAI guideline for percutaneous coronary intervention, 2011 ACCF/AHA guideline for coronary artery bypass graft surgery, 2012 ACC/AHA/ACP/AATS/PCNA/SCAI/STS guideline for the diagnosis and management of patients with stable ischemic heart disease, 2013 ACC/AHA guideline for the management of ST-elevation myocardial infarction, 2014 AHA/ACC guideline for the management of patients with non-ST-elevation acute coronary syndromes, and 2014 ACC/AHA guideline on perioperative cardiovascular evaluation and management of patients undergoing noncardiac surgery. Circulation. 2016;134(10):e123-55.
12. Valgimigli M, Bueno H, Byrne RA, Collet JP, Costa F, Jeppsson A; ESC Scientific Document Group; ESC Committee for Practice Guidelines (CPG); ESC National Cardiac Societies. 2017 ESC focused update on dual antiplatelet therapy in coronary artery disease developed in collaboration with EACTS: The Task Force for dual antiplatelet therapy in coronary artery disease of the European Society of Cardiology (ESC) and of the European Association for Cardio-Thoracic Surgery (EACTS). Eur Heart J. 2018;39(3):213-60.

Critical analysis of the classic indications for myocardial revascularization

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SUMMARY

Treatment of stable coronary artery disease (CAD) relies on improved prognosis and relief of symptoms. National and international guidelines on CAD support the indication of revascularization in patients with limiting symptoms and refractory to optimal medical treatment, as well as in clinical situations where there is a prognostic benefit of interventional treatment. Most of the studies that support the guidelines for indication of revascularization date back to the 1980s and1990s of the last century. Recent studies have revisited the theme and brought a new breath. The present review provides a critical analysis of classic indications for revascularization, reviewing evidence from the studies of the 1970s to the recent controversial ORBITA study.

KEYWORDS: *Coronary artery disease. Myocardial Revascularization. Angina, Stable.*

INTRODUCTION

Once the diagnosis of stable coronary artery disease (CAD) is established and the optimal medical therapy (OMT) introduced, the clinical cardiologist is faced with the dilemma of whether or not coronary intervention (surgical or percutaneous) associated to the OMT is necessary. There is no doubt that the OMT is absolutely necessary and responsible for over 70% of the reduction in the relative risk of death or myocardial infarction in two years in the context of secondary prevention[1].

However, the dilemma is real. The evidence directs us to the lack of prognostic benefit of surgical or percutaneous revascularization when compared to exclusive OMT, when indicated routinely and generally[2,3]. This same evidence, however, identifies specific subpopulations, whose high risk of adverse outcomes could be reduced with myocardial revascularization[4]. The recognition of these subpopulations is an indispensable part of the therapeutic management of patients with stable CAD.

CLASSICAL INDICATIONS FOR REVASCULARIZATION

The identification of the spectra of stable CAD that benefit from revascularization procedures in addition to the OMT is based on the concept that the benefit of revascularization is greater the more severe is the CAD. The severeness is considered from a clinical (limiting symptoms), angiographic (lesions

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in the topography of poor prognosis), ischemic (extensive area at risk), or ventricular function (systolic dysfunction of the left ventricle) perspective.

Thus, the decisive elements for defining the indication of revascularization are focused on the patient: symptoms, anatomy, ischemia and left ventricular systolic function. Some of these variables, in addition to assisting in the indication of myocardial revascularization, also help in selecting the strategy of revascularization. However, it is necessary to differentiate the tools necessary for the best judgment of indication of those useful in the selection of the intervention strategy (Figure 1).

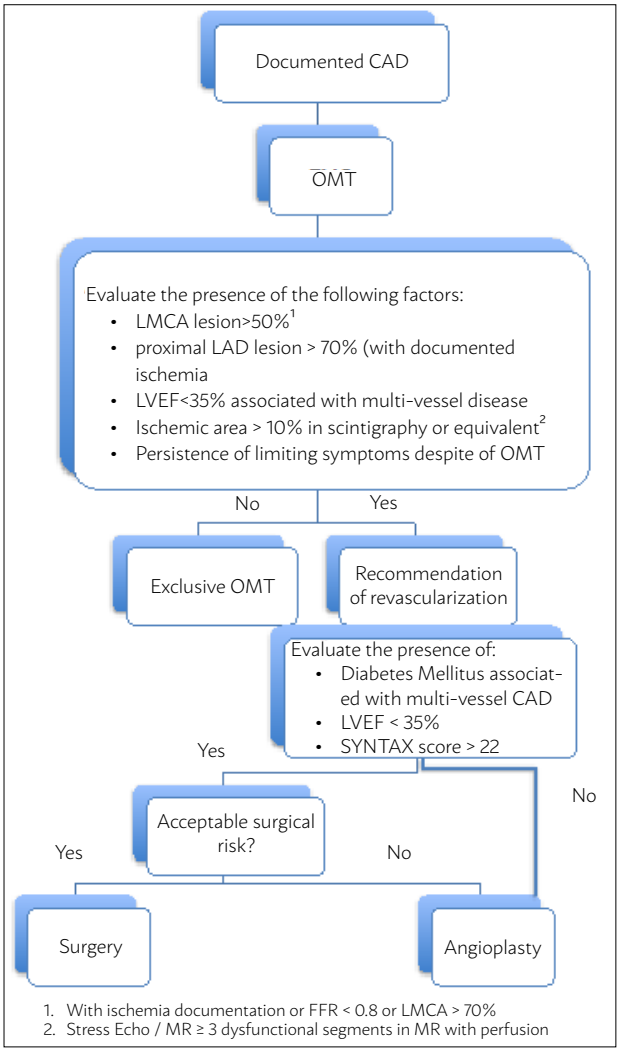
PRESENCE OF DOCUMENTED ISCHEMIA

Classically, the presence of moderate-to-severe ischemic burden constitutes as one of the indications for myocardial revascularization in chronic coronary artery disease (CAD). A retrospective study with observational data evaluated the relationship between the ischemic area detected by means of stress and resting scintigraphy and cardiovascular mortality[5]. Patients were randomly divided into two groups, clinical (9,956 individuals) or surgical (678 patients), in accordance with the established treatment after 60 days of the scintigraphy. In patients in which they identified an ischemic area of 1%-5%, there was a mortality rate of 1% in the clinical group and 1.8% in the surgical group. However, above 10%, there was a mortality rate of 4.8% in the group of drug therapy alone *versus* 3.3% in the group of surgical revascularization. Finally, for those with significant ischemia (> 20%), the mortality rate in the clinical group was more than three times higher than that of the intervention group (6.7% *versus* 2.0%). A careful analysis of these findings is necessary because this is a retrospective study in which the cause-and-effect association cannot be fully established.

A recent prospective and randomized study compared the angioplasty guided by FFR with drug-eluting stents, combined with pharmacological treatment, and the clinical treatment alone in patients with stable CAD[6]. The lesions with FFR < 0.8 were treated. There was a reduction of the composite outcome of mortality due to all causes, non-fatal myocardial infarction and unplanned hospitalization with new revascularization in two years. However, this reduction was due only to the decrease in additional revascularizations, without impact on myocardial infarction or mortality. On the other hand, in patients with lesions anatomically significant but without ischemia evidenced by FFR, there was an excellent prognosis with clinical treatment alone. There are also some limitations of this study that are noteworthy. The early interruption in the recruitment of patients after the analysis of a high rate of events in the control group may have reduced the power of the study. In addition, doctors and patients were not blinded regarding the group they belonged to. This fact may have influenced the decision-making process during the follow-up.

Conversely, it is worth mentioning the nuclear subanalysis of the Courage study[7], in which the treatment with percutaneous angioplasty (PCI) was superior to the clinical treatment in the reduction of ischemia assessed using myocardial scintigraphy (2.7% reduction *versus* 0.5%). However, this finding did not translate into improved survival after adjusted risk analysis.

FIGURE 1.



The moderate-to-severe ischemic burden seems to indicate the worst prognosis. However, the thresholds of both ischemic area and fractional flow reserve still need to be reviewed as indicators of intervention. In this perspective, the ischemia trial (ClinicalTrials.gov Identifier: NCT01471522), randomized, controlled, under development, will evaluate more consistently the prognostic impact of ischemia in stable CAD, as well as the role of myocardial revascularization in this context.

PRESENCE OF ANGINA

In addition to the prognostic improvement, symptomatic relief is one of the main objectives of the treatment of stable angina. Data from the literature demonstrate that the intervention treatments can be used in conjunction with a clinical treatment for this purpose. In this perspective, the quality of life analysis of the classic CASS study[2] showed that in patients with angina CCS I or II myocardial revascularization surgery, compared to clinical treatment alone, improved the quality of life, reduced angina more significantly, with a reduction of the daily need of beta-blockers and nitrates. A subanalysis of the STICH study[8] demonstrated that the presence of angina was not a predictor of a worse prognosis in patients with ventricular dysfunction and CAD and did not identify patients who could present better survival with surgical revascularization. Notwithstanding, surgery reduced angina more consistently than drug treatment alone. Percutaneous angioplasty with stent was also assessed for symptomatic control. Parisi et al.[9] demonstrated, in a prospective study of patients with angina and documented ischemia on cardiac stress testing and coronary lesions in main vessels above 70%, that drug therapy alone or its association with percutaneous angioplasty (PCI) were effective in reducing the incidence of symptoms, as well as increasing exercise time and improving the quality of life. However, these benefits were more significant in the group submitted to PCI.

The Orbita[10] study was the target of great controversy, since it demonstrated, randomly and blindly, the futility of the percutaneous treatment in the improvement of exercise tolerance in a population with univessel CAD by visual estimation. The selection of individuals (univessel, oligosymptomatic, approximately 25% without demonstrated ischemia), as well as the primary outcome selected (time of exercise tolerance, with objective ischemia or class of secondary angina outcomes) may explain the results of this study[11]. Thus, the Orbita study should not be generalized to the heterogeneous population with CAD.

Thus, it is recommended that clinical treatment be optimized as the initial strategy for angina control. If there is symptomatic resistance in its duration, both percutaneous and surgical treatment may be indicated, and the choice of each method depends on the anatomical complexity of comorbidities, functional status, and the preference of the patient.

ANATOMICAL COMPLEXITY OF CORONARY ARTERY DISEASE

Lesion in the proximal LAD

The most commonly used definition of the lesion in the proximal LAD consists in the stenosis between the end of the LMCA and the first great septal or first diagonal, whichever is nearest[12]. The prevalence of atherosclerotic involvement at this location can reach 41% in a population of patients in CAD assessment[13].

Classically, due to the large extension of myocardium at risk associated with stenosis of the proximal LAD, the interventionist approach is recommended in this scenario. In the European study, the subgroup of patients with stenosis above 50% in the proximal LAD showed higher mortality in the group submitted to drug treatment when compared to the surgical group, mainly in association with three-vessel disease[14]. Since then, the presence of a lesion in the proximal LAD has been associated with a worse prognosis in the long term, irrespective of the number of arteries affected. The two studies in question, however, were carried out in the period in which the clinical treatment for CAD consisted of symptomatic medications, with little or no impact on prognosis.

The Mass I[15] study, after comparing three therapeutic strategies (surgery, balloon angioplasty, or drug treatment alone) in a sample of 214 patients with isolated involvement of the proximal LAD and preserved left ventricular function, showed no difference in mortality or infarction among the three groups. In the surgical group, there was a reduction in the primary composite outcome at the expense of a lower rate of additional revascularization. In a subanalysis of the Courage study[16], the presence of a lesion in the proximal LAD, even in three-vessel patients, was not associated with the increase in the primary outcome composed of death, myocardi-

al infarction, and unstable angina. In the same way, when compared with the clinical treatment alone, revascularization was not proved to be beneficial in this subgroup of patients. Thus, in individuals who do not present angina that is limiting and resistant to optimal medical therapy, the initial therapeutic approach with regular clinical follow-up and optimization of medications is an acceptable strategy in this population in relation to survival. The guidelines that recommend revascularization in this scenario with prognosis benefit are based on reducing the need for additional revascularization during follow-up, with no change in mortality. On the other hand, when opting for myocardial revascularization in this context (indication of revascularization for improvement of symptoms, for example), the choice of the best interventionist strategy (surgical or percutaneous) is still a subject of debate in clinical practice.

In a meta-analysis including nine randomized studies that compared the strategies of revascularization in 1,210 patients with isolated involvement of the proximal LAD, Kapoor et al.[17] showed there was no difference in mortality between the groups, but there was less need for additional revascularization and greater symptomatic relief in the surgical group.

In the presence of stenosis of the proximal LAD, therefore, after an initial strategy of optimal clinical treatment, if there is a persistence of symptoms, the two options of interventionist approach may be considered. In the case of percutaneous treatment, however, there is a greater likelihood of a need for new revascularization procedures during outpatient follow-up, but without prognostic impact.

Lesion in the left main coronary artery disease

The lesion in the left main coronary artery (LMCA), among the many segments of the coronary tree, is the most feared because of its association with adverse events, given the magnitude of the territory irrigated by the left coronary system.

Conley et al.[18] followed-up patients who presented LCT lesion documented by cardiac catheterization and did not undergo myocardial revascularization surgery. The survival rate of 163 patients in clinical treatment with lesion ≥ 50% was 79% and 50% in 1 and 3 years, respectively. However, these data differ when comparing lesions between 50%-70% and lesions above 70%. Patients kept under clinical treatment with obstructions in the LCT above 70% presented lower survival in a three-year follow-up compared to those with lesions between 50%-70% (41% *vs.* 66%; $p < 0.05$). Thus, not only a significant lesion in the LMCA but also its severity, represent a prognostic value in this peculiar population of coronary heart disease patients.

The records of the CASS study[19] show 1,484 patients with LCT lesion ≥ 50% allocated to surgical and clinical treatment (MT). Coronary artery bypass grafting (CABG) was the initial treatment in 1,153 patients (78%), in accordance with the preferences of medical staff and patients. The population submitted to CABG had more angina, lesions of greater severity in the LCT, but better left ventricular function. In the greatest long-term follow-up ever published for this type of patients, 15 years, the median survival was of 13.3 years in the surgical group and 6.6 years in the clinical group ($p < 0.001$). However, it should be noted that this significant difference in survival rate between the groups disappears when analyzing only those with preserved ventricular function, even when in association with right coronary lesion greater than 70%. On the other hand, the surgical treatment proposed in the present study, as well as the clinical one, is far from the current standards. The left mammary artery graft, for example, was used in only 9.5% of the patients.

The Iris-Main[20] records are a contemporary study, non-randomized, multicenter, and observational of Asian hospitals, including consecutive patients with LCT lesion ≥ 50% submitted to clinical treatment, percutaneous or surgical. The results were interpreted according to the generation of *stents* used in pre-specified historic times. During all periods, the rates of combined events (death, myocardial infarction, and cerebrovascular accident) were greater in patients under clinical treatment alone in comparison with interventional therapy, regardless of the strategy used. However, they observed a gradual reduction in the rate of events in the population that was kept under the conservative treatment, possibly related to the growing use of statins and dual platelet antiaggregation therapy during the three periods. The rates of events in the group under surgical treatment remained stable, while a significant reduction of these outcomes was observed in patients undergoing percutaneous treatment, in the pre-specified historic times. When comparing the interventional therapies, higher rates of new revascularization always were identified in the percutaneous group (PCI). However, with the advent and development of

pharmacological stents associated with the use of functional revascularization and imaging tools (fractional flow reserve, intravascular ultrasound, and optical coherence tomography), we observed a gradual reduction in the difference for this outcome between the PCI and CABG groups[21].

The choice of clinical treatment alone in patients with a significant lesion in the LMCA is reserved for situations of fragility, low life expectancy, refusal to interventional therapy and anatomy unfavorable for revascularization. The current discussion focuses on the definition of the interventionist strategy more appropriate for this subgroup of patients.

Lesion in the single remaining patent coronary artery

It is defined as lesion in the single remaining patent coronary artery > 50% with the occlusion of all other coronary beds The indication for myocardial revascularization in patients with severe stenosis in the single remaining patent coronary artery consists of a class I recommendation in the major national and international guidelines[22,23]. However, this recommendation is based on physiopathology and not on clinical evidence. In fact, an event associated with this artery will most likely result in a fatal event. However, the selection of the best strategy of revascularization should be based on angiographic aspects, presence of comorbidities, and left ventricular function.

PRESENCE OF VENTRICULAR DYSFUNCTION

Coronary artery disease (CAD) is the most common etiology of heart failure with reduced ejection fraction. The presence of left ventricular dysfunction confers a worse prognosis for patients with chronic coronary disease. The objective of myocardial revascularization for this group of patients consists of contractile recovery after the coronary flow is reestablished, the reduction of cardiovascular events, and the improvement of symptoms and functional capacity[24].

The STICH study[25] (Surgical Treatment of Ischemic Heart Failure), in a sample of 1,212 patients with ventricular dysfunction (ejection fraction less than 35%) associated with multivessel coronary disease, compared the optimized clinical treatment with surgical myocardial revascularization. After a median follow-up of five years, there was no difference between the two therapeutic strategies with regards to the primary outcome of general mortality, but there was a trend of reduction of cardiovascular mortality in the surgical group. With the objective of better selecting individuals with ventricular dysfunction that could benefit from a surgical approach, Velazquez et al.[4], in a subanalysis of the STICH, study identified three predictors of prognostic benefit with revascularization surgery (three-vessel disease, ejection fraction lower than 27%, and indexed end-systolic volume greater than 79 ml/m2). In the presence of two or more of these factors, the patients had lower mortality when submitted to a surgical procedure in comparison with the sample with one or no predictor. Additionally, the results of the very long-term follow-up of the STICH study corroborate the indication for surgical revascularization for ischemic cardiomyopathy. In this analysis, the surgical group presented lower overall mortality rates and cardiovascular mortality when compared to patients undergoing clinical treatment alone.

In patients with left ventricular dysfunction secondary to multivessel CAD, therefore, current evidence suggests that the prognosis benefit of the surgical strategy should be prioritized in the therapeutic management, regardless of the documentation of myocardial viability. However, patients with high surgical risk, fragile and/or with less complex anatomy, can be eligible for percutaneous treatment, especially when undergoing complete revascularization[26].

PATIENTS WITH DIABETES MELLITUS

A patient with diabetes mellitus (DM) presents a higher risk of death and adverse events in the context of CAD. Due to angiographic (more extensive disease, affecting more coronary segments and/or more diffuse), plaque (a higher probability of plaques called “vulnerable”), and individual (greater association with other morbidities, kidney insufficiency, for example) peculiarities, the indication of an earlier intervention in the natural history of CAD is questioned in these individuals in an attempt to prevent more serious cardiovascular events. However, the evolution of the atherosclerotic disease in diabetic patients undergoing the various procedures of intervention is also marked by complications. Thus, we know that the diabetic population is more susceptible to thrombosis and restenosis of stents, as well as a higher rate of early (CVA, infections) and late (lower patency of grafts in the long term) surgical complica-

tions. Thus, the safety of keeping a diabetic patient with CAD in a drug treatment alone was researched in studies with long-term follow-up.

The BARI 2D[27] study, which included a population of 2,368 diabetic patients with multivessel CAD, showed no difference in the incidence of AMI, CVA, or death in the long-term follow-up, comparing the intervention (with surgery or conventional angioplasty with stent) and optimal medical therapy (OMT). However, there was a lower incidence of the combined primary outcome of AMI, CVA, and death in the subgroup submitted to surgery compared to the OMT group (22.4% *versus* 30.5%; p = 0.01).

In our environment, a subanalysis of the MASS study[28] compared three therapeutic modalities (surgery, angioplasty, and OMT) in 190 diabetic patients with stable multivessel CAD, out of the 611 included in the original study. After the first year of follow-up, a lower mortality rate among those submitted to interventionist procedures (surgery and angioplasty) was observed when compared to the OMT group (p = 0.039). These data were confirmed in a ten-year follow-up of the same population, with the benefit of the surgery over the clinical treatment alone among diabetics[29].

The indication of revascularization in diabetic patients with CAD without further formal indications of revascularization is controversial and lacks evidence. However, once the revascularization is indicated, there is the benefit of surgical treatment over the percutaneous treatment among patients with multivessel CAD[30].

CONCLUSION

The indications for myocardial revascularization are substantiated on more than 30 years of evidence and aspects that focus on symptoms, coronary anatomy, as well as the presence of extensive ischemia and left ventricular dysfunction. The variables that govern the indication of myocardial revascularization should be properly distinguished from those that assist in the selection of the best strategy of revascularization.

RESUMO

O tratamento da doença arterial coronariana estável (DAC) se baseia na melhora do prognóstico e alívio de sintomas. Diretrizes nacionais e internacionais sobre a DAC respaldam a indicação de revascularização em pacientes com sintomas limitantes e refratários ao tratamento medicamentoso, bem como em situações clínicas nas quais há benefício prognóstico do tratamento intervencionista. Grande parte dos estudos que norteiam as diretrizes de indicação de revascularização data das décadas de 1980 e 1990. Estudos recentes têm revisitado o tema e trazido novo fôlego. A presente revisão faz uma análise crítica das indicações clássicas de revascularização, revisando a evidência desde os estudos da década de 1970 ao recente e polêmico estudo Orbita.

PALAVRAS-CHAVE: *Doença da artéria coronariana. Revascularização miocárdica. Angina estável.*

REFERENCES

1. Yusuf S. Two decades of progress in preventing vascular disease. Lancet. 2002;360(9326):2-3.
2. Coronary artery surgery study (CASS): a randomized trial of coronary artery bypass surgery. Quality of life in patients randomly assigned to treatment groups. Circulation. 1983;68(5):951-60.
3. Boden WE, O'Rourke RA, Teo KK, Hartigan PM, Maron DJ, Kostuk WJ, et al.; COURAGE Trial Research Group. Optimal medical therapy with or without PCI for stable coronary disease. N Engl J Med. 2007;356(15):1503-16.
4. Velazquez EJ, Lee KL, Jones RH, Al-Khalidi HR, Hill JA, Panza JA, et al.; STICHES Investigators. Coronary-artery bypass surgery in patients with ischemic cardiomyopathy. N Engl J Med. 2016;374(16):1511-20.
5. Hachamovitch R, Hayes SW, Friedman JD, Cohen I, Berman DS. Comparison of the short-term survival benefit associated with revascularization compared with medical therapy in patients with no prior coronary artery disease undergoing stress myocardial perfusion single photon emission computed tomography. Circulation. 2003;107(23):2900-7.
6. De Bruyne B, Fearon WF, Pijls NH, Barbato E, Tonino P, Piroth Z, et al. Fractional flow reserve-guided PCI for stable coronary artery disease. N Engl J Med. 2014;371(13):1208-17.
7. Shaw LJ, Berman DS, Maron DJ, Mancini GB, Hayes SW, Hartigan PM, et al.; COURAGE Investigators. Optimal medical therapy with or without percutaneous coronary intervention to reduce ischemic burden: results from the Clinical Outcomes Utilizing Revascularization and Aggressive Drug Evaluation (COURAGE) trial nuclear substudy. Circulation. 2008;117(10):1283-91.
8. Jolicoeur EM, Dunning A, Castelvecchio S, Dabrowski R, Waclawiw MA, Petrie MC, et al. Importance of angina in patients with coronary disease, heart failure, and left ventricular systolic dysfunction: insights from STICH. J Am Coll Cardiol. 2015;66(19):2092-100.
9. Parisi AF, Folland ED, Hartigan P. A comparison of angioplasty with medical therapy in the treatment of single-vessel coronary artery disease. Veterans Affairs ACME Investigators. N Engl J Med. 1992;326(1):10-6.
10. Al-Lamee R, Thompson D, Dehbi HM, Sen S, Tang K, Davies J, et al.; ORBITA investigators. Percutaneous coronary intervention in stable angina (ORBITA): a double-blind, randomized controlled trial. Lancet. 2018;391(10115):31-40.
11. Oliveira VD, Giugni FR, Martins EB, Azevedo DFC, Lima EG, Serrano Júnior CV. The ORBITA trial: A point of view. Rev Assoc Med Bras (1992). 2018;64(2):100-3.
12. Myers WO, Davis K, Foster ED, Maynard C, Kaiser GC. Surgical survival in the Coronary Artery Surgery Study (CASS) registry. Ann Thorac Surg. 1985;40(3):245-60.
13. Min JK, Dunning A, Lin FY, Achenbach S, Al-Mallah M, Budoff MJ, et al.; CONFIRM Investigators. Age- and sex-related differences in all-cause mortality risk based on coronary computed tomography angiography

findings results from the International Multicenter CONFIRM (Coronary CT Angiography Evaluation for Clinical Outcomes: An International Multicenter Registry) of 23,854 patients without known coronary artery disease. J Am Coll Cardiol. 2011;58(8):849-60.

14. European Coronary Surgery Study Group. Long-term results of prospective randomised study of coronary artery bypass surgery in stable angina pectoris. Lancet. 1982;2(8309):1173-80.

15. Hueb WA, Bellotti G, Oliveira SA, Arie S, Albuquerque CP, Jatene AD, et al. The Medicine, Angioplasty or Surgery Study (MASS): a prospective, randomized trial of medical therapy, balloon angioplasty or bypass surgery for single proximal left anterior descending artery stenoses. J Am Coll Cardiol. 1995;26(7):1600-5.

16. Mancini GB, Hartigan PM, Bates ER, Chaitman BR, Sedlis SP, Maron DJ, et al. Prognostic importance of coronary anatomy and left ventricular ejection fraction despite optimal therapy: assessment of residual risk in the Clinical Outcomes Utilizing Revascularization and Aggressive DruG Evaluation Trial. Am Heart J. 2013;166(3):481-7.

17. Kapoor JR, Gienger AL, Ardehali R, Varghese R, Perez MV, Sundaram V, et al. Isolated disease of the proximal left anterior descending artery comparing the effectiveness of percutaneous coronary interventions and coronary artery bypass surgery. JACC Cardiovasc Interv. 2008;1(5):483-91.

18. Conley MJ, Ely RL, Kisslo J, Lee KL, McNeer JF, Rosati RA. The prognostic spectrum of left main stenosis. Circulation. 1978;57(5):947-52.

19. Caracciolo EA, Davis KB, Sopko G, Kaiser GC, Corley SD, Schaff H, et al. Comparison of surgical and medical group survival in patients with left main coronary artery disease. Long-term CASS experience. Circulation. 1995;91(9):2325-34.

20. Lee PH, Ahn JM, Chang M, Baek S, Yoon SH, Kang SJ, et al. Left main coronary artery disease: secular trends in patient characteristics, treatments, and outcomes. J Am Coll Cardiol. 2016;68(11):1233-46.

21. Harskamp RE, Park DW. Stenting versus surgery for significant left main disease. Curr Cardiol Rep. 2015;17(4):18.

22. Windecker S, Kolh P, Alfonso F, Collet JP, Cremer J, Falk V, et al.; Grupa Robocza Europejskiego Towarzystwa Kardiologicznego (ESC); Europejskie Stowarzyszenie Chirurgii Serca i Klatki Piersiowej (EACTS) do spraw rewaskularyzacji mięśnia sercowego; European Association for Percutaneous Cardiovascular Interventions (EAPCI). [2014 ESC/EACTS Guidelines on myocardial revascularization]. Kardiol Pol. 2014;72(12):1253-379.

23. Cesar LA, Ferreira JF, Armaganijan D, Gowdak LH, Mansur AP, Bodanese LC, et al.; Sociedade Brasileira de Cardiologia. Guideline for stable coronary artery disease. Arq Bras Cardiol. 2014;103(2 Suppl 2):1-56.

24. Lima EG, Carvalho FPC, Linhares Filho JPP, Pitta FG, Serrano CV Jr. Ischemic left ventricle systolic dysfunction: an evidence-based approach in diagnostic tools and therapeutics. Rev Assoc Med Bras (1992). 2017;63(9):793-800.

25. Velazquez EJ, Lee KL, Deja MA, Jain A, Sopko G, Marchenko A, et al.; STICH Investigators. Coronary-artery bypass surgery in patients with left ventricular dysfunction. N Engl J Med. 2011;364(17):1607-16.

26. Bangalore S, Guo Y, Samadashvili Z, Blecker S, Hannan EL. Revascularization in patients with multivessel coronary artery disease and severe left ventricular systolic dysfunction: everolimus-eluting stents versus coronary artery bypass graft surgery. Circulation. 2016;133(22):2132-40.

27. BARI 2D Study Group, Frye RL, August P, Brooks MM, Hardison RM, Kelsey SF, MacGregor JM, et al. A randomized trial of therapies for type 2 diabetes and coronary artery disease. N Engl J Med. 2009;360(24):2503-15.

28. Soares PR, Hueb WA, Lemos PA, Lopes N, Martinez EE, Cesar LA, et al. Coronary revascularization (surgical or percutaneous) decreases mortality after the first year in diabetic subjects but not in nondiabetic subjects with multivessel disease: an analysis from the Medicine, Angioplasty, or Surgery Study (MASS II). Circulation. 2006;114(1 Suppl):I420-4.

29. Lima EG, Hueb W, Garcia RM, Pereira AC, Soares PR, Favarato D, et al. Impact of diabetes on 10-year outcomes of patients with multivessel coronary artery disease in the Medicine, Angioplasty, or Surgery Study II (MASS II) trial. Am Heart J. 2013;166(2):250-7.

30. Farkouh ME, Domanski M, Sleeper LA, Siami FS, Dangas G, Mack M, et al. Strategies for multivessel revascularization in patients with diabetes. N Engl J Med. 2012;367(25):2375-84.

Eosinophilic fasciitis: an atypical presentation of a rare disease

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SUMMARY

Eosinophilic fasciitis, or Shulman's disease, is a rare disease of unknown etiology. It is characterized by peripheral eosinophilia, hypergammaglobulinemia, and high erythrocyte sedimentation rate. The diagnosis is confirmed by a deep biopsy of the skin. The first line of treatment is corticotherapy.

We present a rare case of eosinophilic fasciitis in a 27-year-old woman with an atypical presentation with symmetrical peripheral edema and a Groove sign. The patient responded well to treatment with corticosteroids at high doses and, in this context, was associated with hydroxychloroquine and azathioprine. After two and a half years, peripheral eosinophilia had increased, and more of her skin had hardened. At that time, the therapy was modified to include corticoids, methotrexate, and penicillamine. It is of great importance to publicize these cases that allow us to gather experience and better treat our patients.

KEYWORDS: *Fasciitis. Eosinophils. Eosinophilia. Edema/etiology.*

INTRODUCTION

Eosinophilic fasciitis is a rare disease characterized by skin alterations such as scleroderma, peripheral eosinophilia, hypergammaglobulinemia, and high erythrocyte sedimentation rate.[1] It more frequently involves the inferior limbs. The absence of Raynaud's phenomenon or sclerodactyly distinguishes Eosinophilic fasciitis from sclerodermia.[1] Literature has described fewer than 300 cases. Shulman syndrome, as it is also known, was first described in 1974. Its etiology remains unknown.[2] It affects both sexes equally, with a higher incidence among individuals of 40-50 years old and no associations of race, risk factors, or family history.[3]

The importance of this case is related to the rarity of the disease, its atypical presentation, and the difficulty of its treatment. The reduced number of reported cases makes it harder to approach these situations and emphasizes the importance of sharing any experience.

We present the case of a 27-year-old woman with Eosinophilic fasciitis characterized by symmetrical peripheral edema and maculopapular skin rash.

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CASE REPORT

We report the case of a 27-year-old caucasian woman who works as a secretary. She has diabetes mellitus type 1 and hypothyroidism, treated and supervised. She was sent to us because of the emergent and progressive deterioration of symmetrical peripheral edema (Figure 1), asthenia, and maculopapular pruritic rash with a 2-month evolution. She had been previously treated with insulin and levothyroxine and had no other complaints or relevant history. At the objective examination, she presented with a hardening of the subcutaneous tissue of the forearms and inferior limbs with symmetric edema, with no evidence of arthritis and a maculopapular rash on the right arm. Analytically, she presented haemoglobin of 13.1 g/dL (11.7-15.7), leucocytes 26,3x10^3 uL (4-10), eosinophils 8.7x10^3 u/L (33%), plaquettes 650,000 uL (150,000-450,000); sedimentation velocity of 58 mm/h (<20), LDH 497 U/L (240-480), reactive protein C 18,1 mg/L (<5), antinuclear antibody (ANA) 1/320, anti-dsDNA 121,9 UI/mL (<25), rheumatoid factor 52 uI/mL (<14) and electrophoresis of the serum proteins with a monoclonal peak of IgG de 2920 mg/dL (700-1600). The swab of peripheral blood showed eosinophilia and lymphocytes (with no morphologic alterations), thrombocytes, and some giant plaquettes. The myelogram and bone biopsy revealed granulocytic hyperplasia with mature eosinophils. We made a nuclear resonance imaging of the right hand, which revealed diffuse thickening of the subcutaneous cell tissue, associated with edema that reached the fascias and the intermuscular planes diffusely with no clear predominance; this made it reasonable to reconsider the clinical hypothesis of fasciitis with confirmation by cutaneous biopsy (Figure 2). The deep cutaneous biopsy between the subcutaneous cell tissue and the skeletal muscle showed the involvement of the fascia by numerous polymorphonuclear eosinophils. The remaining examinations, such as capillaroscopy, manometry, breathing function proofs, echocardiogram, and computerized axial tomography of thorax, abdomen, and pelvis did not show any alterations. The examinations led us to conclude that it was eosinophilic fasciitis. The patient started pulses of methylprednisolone (500 mg daily for 3 days), followed by 40 mg of oral prednisolone (slowly weaning, during four months, until reaching 10 mg/day) as well as hydroxychloroquine 400 mg/day and azathioprine (AZA) 150 mg/day for 2.5 years. After that period, she displayed an increase of peripheral eosinophils with worsening of cutaneous complaints (hardening of the skin). She repeated pulses of methylprednisolone 1gr monthly for 5 months as well as methotrexate 20 mg /week, penicillamine 150 mg/day, and hydroxychloroquine 400 mg/day. One year later, there was an improvement of the subcutaneous hardening maintaining hyperpigmentation areas (figure 3).

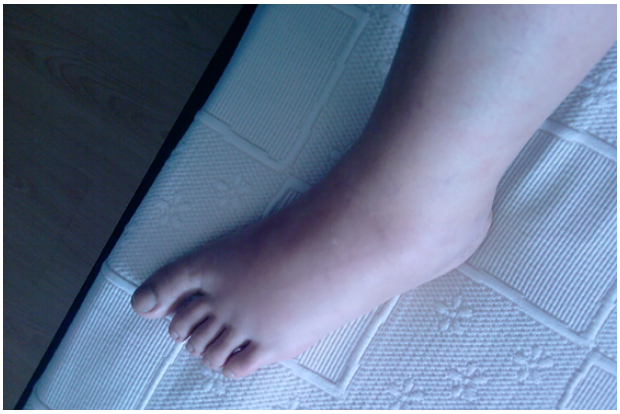

FIGURE 1: EDEMA OF THE INFERIOR MEMBER

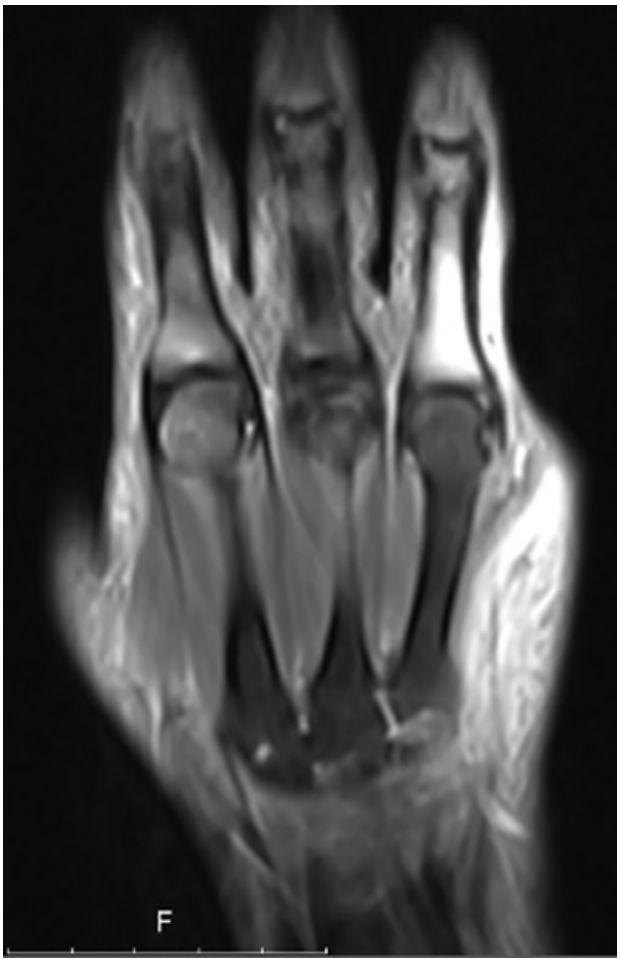

FIGURE 2: RESONANCE IMAGING OF THE RIGHT HAND

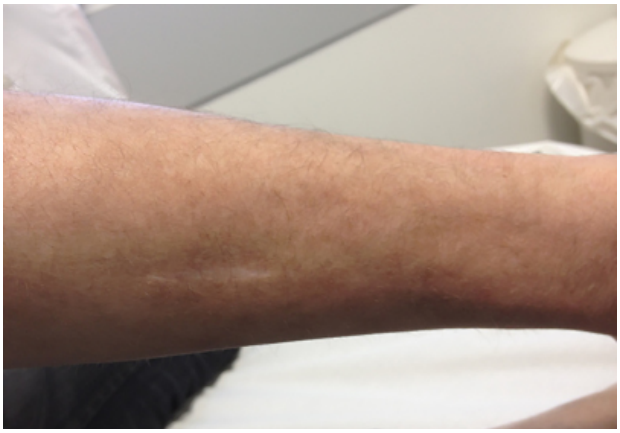

FIGURE 3: GROOVE SIGN ON THE RIGHT FOREARM

DISCUSSION

Fasciitis was first described by Shulman in 1974.[4] Its etiology remains unknown. In literature, the disease is sometimes said to have been preceded by intense physical activity.[1] Some cases are associated to other factors, such as the beginning of hemodialysis, infection by Borrelia burgdorferi, radiotherapy, hematological disturbances, graft versus host disease, or medications such as simvastatin, atorvastatin, and fenitoin.[1,5-8] In our case, no leading factor was detected. Serologies and cultural examinations were negative. About 15% of patients have a malign underlying disease like leukemia or lymphoma. There are also cases described as being associated with hematologic disturbances, such as aplastic anemia, hemolytic anemia, self-immune peripheric thrombocytopenia, and pernicious anemia.[1] Self-immune diseases like Hashimoto Thyroiditis, morphea, systemic sclerosis, Sjögren syndrome, and antiphospholipid antibody syndrome were also reported in some patients.[1] In our case, the patient has not presented any malign disease so far. She was already under treatment and supervision for type 1 diabetes mellitus and hypothyroidism.

Patients often present unspecific symptoms like weakness, general discomfort, fever, and weight loss[9]. At the objective examination, we noticed a hardening of the skin (looking like "peau d'orange") with venous nodes (Groove sign) and occasionally localized morphea.[10] The prayer sign is common, and the Raynaud phenomenon is not frequent.[10] In this case, the patient presented a Groove sign in the right forearm and symmetric edema in the inferior limbs, which led to the differential diagnosis conditions associated to low oncotic pressure or venous failure and congestive heart failure. It was necessary to consider other differential diagnoses with similar symptoms and peripheral eosinophilia such as scleroderma, systemic sclerosis, eosinophilia-myalgia syndrome, and localized fibrosing disorders.[5] Analytically, we found peripheral eosinophilia, hypergammaglobulinemia (generally IgG or IgM) and elevation of the velocity of erythrocyte sedimentation. Our patient presented peripheral eosinophilia and a peak in IgG with no specific antibody.[9] The diagnosis is established by the histopathological report samples through a deep skin biopsy, which must include the muscle. Magnetic resonance imaging may prove useful to define the area for the biopsy, and later, to monitor the response of the disease to the treatment.[1] The typical findings are thickening of the fascia (twice to fifteen times the normal size) associated with inflammatory cells formed by lymphocytes, plasmocytes, histiocytes and eosinophils (in 69-75% of the cases) in variable rates. There is no necrotic vascular injury. The presence of peripheral eosinophils in the fascia is not confirmed in the diagnosis.[9] Histologically, the differential diagnosis should be done with nodule erythema, Wells Syndrome and a reaction to insect bites or parasitic diseases.[10]

Eosinophilic fasciitis is a rare disease, which makes the treatment strategy more difficult. Corticotherapy is currently the first line of treatment at a dosage of 1mg/kg/day until analytical normalization and clinic remission. Many patients do not answer to corticotherapy, and in the case of those who do, there may be a relapse. In these situations, other immunomodulators are used, such as azathioprine, hydroxychloroquine, cyclosporine, dapsone, and methotrexate.[7,10,11]

The prognosis is generally good, except when an underlying malign disease is associated. Diagnosis at ages younger than 12 years, the involvement of the body, and cutaneous sclerosis such as morphea can be evidence of refractory disease.[1]

CONCLUSION

Eosinophilic fasciitis is a rare disease and its treatment can be a challenge. We have just presented a rare case of eosinophilic fasciitis with symmetric peripheral edema. Patients often present relapsing or do not respond to corticotherapy alone. In this case, other immunomodulator agents had to be added. It is important to spread these experiences, which allow us to gather information and knowledge to better treat and care for our patients.

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RESUMO

A fasciite eosinofílica ou doença de Shulman é uma doença rara de etiologia desconhecida. É caracterizada por eosinofilia periférica, hipergamaglobulinemia e velocidade de sedimentação eritrocitária elevada. O diagnóstico é confirmado por biópsia profunda da pele. O tratamento de primeira linha é a corticoterapia.

Apresentamos um caso raro de fasciite eosinofílica numa mulher de 27 anos com uma apresentação atípica com edema periférico simétrico e sinal de Groove. A paciente respondeu bem ao tratamento com corticoides, mas em doses elevadas, e, nesse contexto, associou-se hidroxicloroquina e azatioprina. Ao fim de dois anos e meio verificou-se aumento de eosinofilia e novamente pele mais endurecida. Nessa altura alterou-se a terapêutica para corticoides, metrotexato e penicilamina. É de grande importância a divulgação desses casos que nos permitem reunir experiência e assim melhor tratar os nossos doentes.

PALAVRAS-CHAVE: *Fasciite. Eosinófilos. Eosinofilia. Edema/etiologia.*

REFERENCES

1. Chun JH, Lee KH, Sung MS, Park CJ. Two cases of eosinophilic fasciitis. Ann Dermatol. 2011;23(1):81-4.
2. Massarente VL, Valadares FGC, Muzy GSC, Polônio KS. Fasciíte eosinofílica: relato de caso. Arq Med Hosp Fac Cienc Med Santa Casa São Paulo. 2017; 62(2):107-9.
3. Falcão S, Mourão AF, Ribeiro C, Pinto TL, Mateus M, Araújo P, et al. Fasceíte eosinofílica e aplasia medular. Acta Reumatol. Port. 2009;34(1):120-4.
4. Shulman LE. Diffuse fasciitis with eosinophilia: a new syndrome? Trans Assoc Am Physicians. 1975;88:70-86.
5. Chang CW, Lau MS. Atypical presentation of eosinophilic fasciitis with pitting edema. Hawaii J Med Public Health. 2015;74(9 Suppl 2):36-8.
6. Bischoff L, Derk CT. Eosinophilic fasciitis: demographics, disease pattern and response to treatment: report of 12 cases and review of the literature. Int J Dermatol. 2008;47(1):29-35.
7. Lebeaux D, Francès C, Barete S, Wechsler B, Dubourg O, Renoux J, et al. Eosinophilic fasciitis (Shulman disease): new insights into the therapeutic management from a series of 34 patients. Rheumatology (Oxford). 2012;51(3):557-61.
8. Moulton SJ, Kransdorf MJ, Ginsburg WW, Abril A, Persellin S. Eosinophilic fasciitis: spectrum of MRI findings. AJR Am J Roentgenol. 2005;184(3):975-8.
9. Lamback EB, Resende FS, Lenzi TC. Eosinophilic fasciitis. An Bras Dermatol. 2016;91(5 suppl 1):57-9.
10. Servy A, Clérici T, Malines C, Le Parc JM, Côté JF. Eosinophilic fasciitis: a rare skin sclerosis. Patholog Res Int. 2011;2011:716935.
11. Manzini CU, Sebastiani M, Giuggioli D, Manfredi A, Colaci M, Cesinaro AM, et al. D-penicillamine in the treatment of eosinophilic fasciitis: case reports and review of the literature. Clin Rheumatol. 2012;31(1):183-7.

Mucosal vitiligo in angles of the mouth: clinical and fluorescence aspects

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SUMMARY

Vitiligo is the most common depigmenting, chronic acquired disease of the skin and mucosa. However, vitiligo of an unclassified type and mucosal subtype affecting only one area of the mucosa is considered quite uncommon. The diagnosis of vitiligo, regardless of its type, is clinical. Nonetheless, a device that allows the visualization of the tissue fluorescence may be useful for confirming the diagnosis. We present the use of wide-field optical fluorescence device for complementary examination and diagnosis of unusual cases of mucosal vitiligo located only in angles of the mouth.

KEYWORDS: *Vitiligo. Diagnostic imaging. Diagnosis. Fluorescence. Pathology. Mouth.*

INTRODUCTION

Vitiligo is an acquired chronic disease of the skin or mucosa characterized by the progressive loss of melanocytes and resulting in the reduction or absence of melanin in the affected site[1-3]. The estimated prevalence of vitiligo worldwide is of 0.5 to 1%, and it is considered the most common depigmenting disorder[1,2]. However, vitiligo of an unclassified type and mucosal subtype affecting only one area of the mucosa represents only 2.3% of the total cases of vitiligo; therefore, a quite uncommon condition[4].

Clinically, the lesion presents itself as white patches that cannot be removed by scraping1. Thus, although vitiligo is not a fatal disease, the presence of facial lesions may cause aesthetic impairment affecting patients psychosocial aspects[1,2,4].

The treatment of vitiligo in the oral region may involve immunomodulators, micropigmentation, phototherapy, and invasive surgical treatments[1,4]. However, vitiligo in the oral region is resistant to treatment; therefore, the early diagnosis contributes to both treatment success and reduction of aesthetic damage[4].

The diagnosis of vitiligo, regardless of its type, is clinical[1,2,4]. However, it can also be confirmed by examination with devices that allow visualizing the tissue fluorescence[1-5]. In this sense, the diagnosis of vitiligo is confirmed by the visualization of an increase in fluorescence in the region of the lesions due to the lack of absorption of light by melanin caused by the reduction of melanocytes[1-4]. Otherwise, in adjacent

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healthy tissues with the presence of melanin, the fluorescence is reduced[5].

We present the use of a wide-field optical fluorescence device for complementary examination and diagnosis of an uncommon case of mucosal vitiligo located only in angles of the mouth.

METHODS

The patient was attended at the Oral Diagnosis Center of the Divinopolis Health Department through a partnership with the Federal University of São João del Rei (Divinópolis, Minas Gerais, Brazil). The protocol consisted of a clinical examination and, in the same consultation, the patient was submitted to the wide-field optical fluorescence complementary examination using the Evince® device (MMOptics, São Carlos, Brazil), which, emits light at a wavelength of 400 ±10nm.

This study has been approved by the Ethics Committee of the Federal University of São João del-Rei, Dona Lindu Center-West Campus (CAAE: 59621516.8.0000.5545; Approval number: 1.756.617).

CASE

A 55-year-old man with a history of white patches located bilaterally at the angles of the mouth was referred by a general dental practitioner for an oral diagnosis consultation. The diagnostic hypothesis by the general dental practitioner was angular cheilitis. In anamnesis, the patient reported that he noticed the whitish lesions about 3 years ago, which aesthetically bothered him. In addition, the patient reported that the lesions were always painless, without ulcerations or crusts and with a history of unsuccessful treatment with topical corticosteroids and antifungals. There was no report of any disease or family history of autoimmune diseases or vitiligo.

In the general physical examination, no other patches or disorders were observed. During the clinical examination, the presence of whitish patches, bilaterally, on the angles of mouth was observed (Figure 1). The lesions observed with an open mouth were 20 mm x 4 mm on the right side (Figure 2A) and 20 mm x 9 mm on the left side (Figure 2B). The white patches could not be scraped off with gauze and exhibited similar consistency to the adjacent healthy tissue.

In the same medical appointment, during the wide-field optical fluorescence examination, a bright green appearance was observed in the region of the lesions, bilaterally at the angles of the mouth, which is characteristic of increased local fluorescence (Figures 2C, 2D).

Based on the clinical examination and the presentation under fluorescence, the final diagnosis was es-

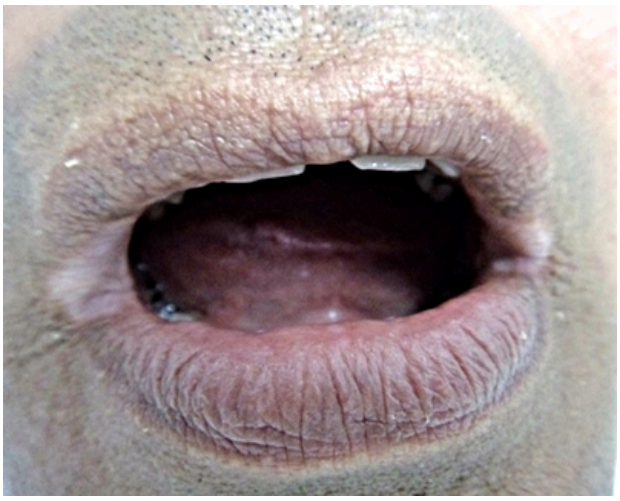

FIGURE 1 Clinical presentation of mucosal vitiligo with the presence of whitish patches bilaterally at the angles of the mouth.

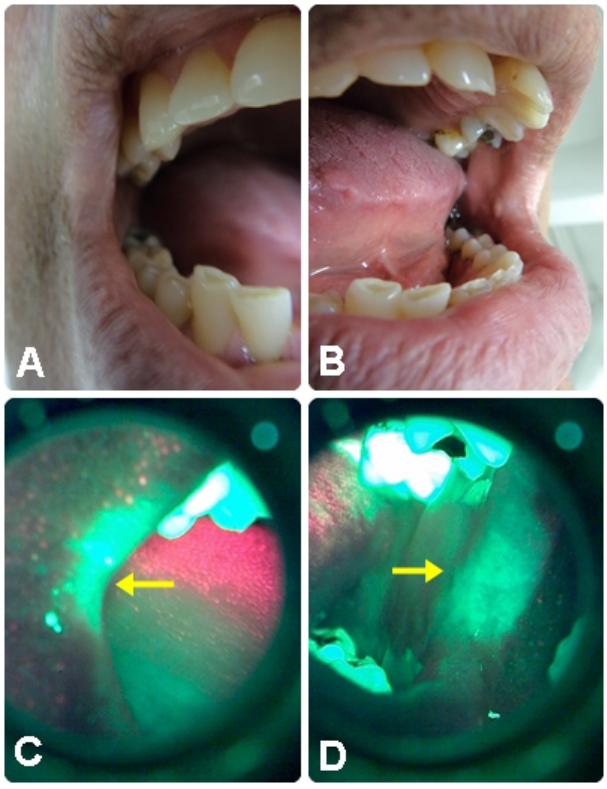

FIGURE 2 Clinical presentation of mucosal vitiligo with the presence of whitish patches at the angle of the mouth on the right side (A) and the left side (B). Wide-field optical fluorescence presentation of mucosal vitiligo; the yellow arrows indicate the increase of bright green fluorescence at the angles of the mouth on the right (C) and left(D).

tablished as vitiligo. The vitiligo was categorized as an unclassified type and mucosal subtype since the patient presented the lesions only in the region of the angle of the mouth without affecting other areas of the body, and the lesions had been persistent for 3 years. Thus, the patient was referred to a dermatologist for treatment.

DISCUSSION

This case of unclassified or undetermined type and mucosal subtype of vitiligo is considered quite unusual since, according to the Vitiligo Global Issues Consensus Conference, to receive such classification, the vitiligo lesion, must occur only in the oral mucosa area, for a time longer than 2 years and without the lesions anywhere else on the body[2,4]. Furthermore, according to Prasad[4], vitiligo lesions usually affect the lip vermilion without extending across the wet labial mucosa. This makes this case even more unusual because when observing the lesion on the left side of the mouth it is observed, both clinically and by fluorescence, that the lesion extends to the wet labial mucosa (Figures 2B,2D).

The general dental practitioner probably considered the hypothesis of angular cheilitis because it is one of the most prevalent oral lesions affecting the oral angle. Moreover, Park et al.[6] described that in its early stages, angular cheilitis presents as a greyish white thickening at the angles of the mouth. However, this hypothesis was immediately discarded due to the persistence of the lesion for 3 years with treatment failure, consistency of the lesion similar to the adjacent tissue, absence of pain, ulceration, and crusts, which are incompatible with angular cheilitis. It is noteworthy that, during clinical examination, many oral pathologies exhibit similar clinical features making diagnosis difficult[7-9].

Thus, to increase the perception of oral lesions, which, could be imperceptible upon clinical examination, the technology of wide-field fluorescence has been approved by the United States Food and Drug Administration (FDA) for use as a complementary examination[8-10]. In summary, we concluded that in this case report wide-field optical fluorescence was a useful complementary examination, as it confirmed the diagnosis of vitiligo through the visualization of an increase in fluorescence caused by the reduction of melanin in the vitiligo area, which, was imperceptible in the conventional clinical examination.

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RESUMO

O vitiligo é a doença crônica adquirida despigmentante mais comum da pele e/ou da mucosa. Entretanto, o vitiligo do tipo não classificado e subtipo de mucosa afetando apenas uma área da mucosa é considerado bastante incomum. O diagnóstico de vitiligo, independentemente do seu tipo, é clínico. No entanto, o uso de um dispositivo que permite a visualização da fluorescência tecidual pode ser útil para a confirmação do diagnóstico de vitiligo. Apresentamos o uso do dispositivo de exame complementar de fluorescência óptica de campo amplo para o diagnóstico de um caso incomum de vitiligo de mucosa localizado apenas em ângulos da boca.

PALAVRAS-CHAVE: *Vitiligo. Diagnóstico por imagem. Diagnóstico. Fluorescência. Patologia. Boca.*

REFERENCES

1. Picardo M, Dell'Anna ML, Ezzedine K, Hamzavi I, Harris JE, Parsad D, et al. Vitiligo. Nat Rev Dis Primers. 2015;1:15011.
2. Ezzedine K, Lim HW, Suzuki T, Katayama I, Hamzavi I, Lan CC, et al; Vitiligo Global Issue Consensus Conference Panelists. Revised classification/nomenclature of vitiligo and related issues: the Vitiligo Global Issues Consensus Conference. Pigment Cell Melanoma Res. 2012;25(3):E1-13.
3. Klatte JL, van der Beek N, Kemperman PM. 100 years of Wood's lamp revised. J Eur Acad Dermatol Venereol. 2015;29(5):842-7.
4. Parsad D. Mucosal vitiligo. In: Vitiligo [Internet]. Berlin, Heidelberg: Springer;2010. p.57-9. Available from: http://link.springer.com/10.1007/978-3-540-69361-1_7
5. Gupta LK, Singhi MK. Wood's lamp. Indian J Dermatol Venereol Leprol. 2004;70(2):131-5.
6. Park KK, Brodell RT, Helms SE. Angular cheilitis, part 1: local etiologies. Cutis. 2011;87(6):289-95.
7. Lane PM, Gilhuly T, Whitehead P, Zeng H, Poh CF, Ng S, et al. Simple device for the direct visualization of oral-cavity tissue fluorescence. J Biomed Opt. 2006;11(2):024006.
8. Andrade SA, Pratavieira S, Ribeiro MM, Bagnato VS, Pilla Varotti F. Oral cancer from the perspective of wide-field optical fluorescence: diagnosis, tumor evolution and post-treatment follow up. Photodiagnosis Photodyn Ther. 2017;19:239-42.
9. Shin D, Vigneswaran N, Gillenwater A, Richards-Kortum R. Advances in fluorescence imaging techniques to detect oral cancer and its precursors. Futur Oncol. 2010;6(7):1143-54.
10. Andrade SA, Pilla Varotti F, Bagnato VS, Pratavieira S. Firearm projectile in the maxillary tuberosity located by adjunctive examination of wide-field optical fluorescence. Photomed Laser Surg. 2018;36(2):112-5.

Easy resolution of severe obstructive kidney injury

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SUMMARY

Chronic constipation is a common diagnosis with a high prevalence in the elderly. Constipation affects the quality of life of sick individuals, bringing several clinical complications.

Keywords: Constipation. Elderly. Acute Kidney Injury.

CASE

We present the case of an 82-year-old caucasian woman living in a nursing home who went to the ER due to strong sputum and fever. She had a previous history of controlled high blood pressure, early-stage dementia, and chronic constipation due to laxatives. She presented leukocytosis and severe kidney injury with a creatinine of 3.4 mg/dL and urea of 231 mg/dL. To clarify the situation, a computed abdominal and pelvis axial tomography was made, which revealed a prominence of the excretory channels on the left side of both kidneys (figure 1) and prominent ureters in the proximal segment conditioned by the dilation of the sigmoid and colon with extensive coprostasis and exuberant fecaliths (figure 2). After eliminating the fecaliths with cleaning enemas, the patient presented normalized kidney function with a creatinine of 1.1 mg/dL and urea of 60 mg/dL.

DISCUSSION

Chronic constipation is a common diagnosis in the elderly with a prevalence of 30% to 40% in patients over 65 years old; it is also more common among women. For patients living in nursing homes, the prevalence increases to 70%. Constipation affects the quality of life of sick individuals, causing many clinical complications and increasing healthcare expenses.[1,2] In Portugal, for instance, over 10 million

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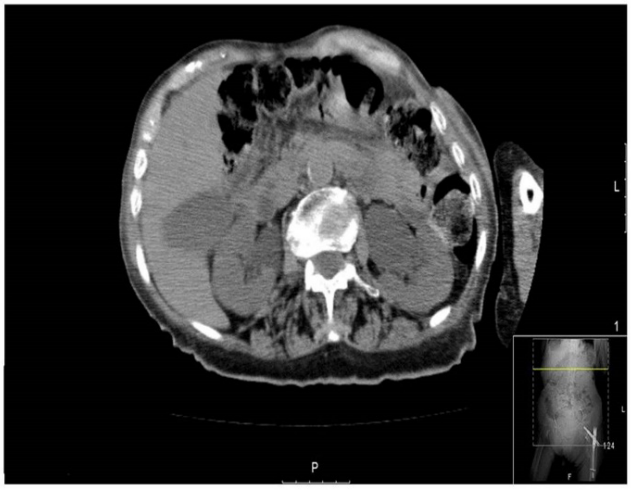

FIGURE 1: PROMINENCE OF THE EXCRETORY CHANNELS OF BOTH KIDNEYS AND PROMINENT URETERS.

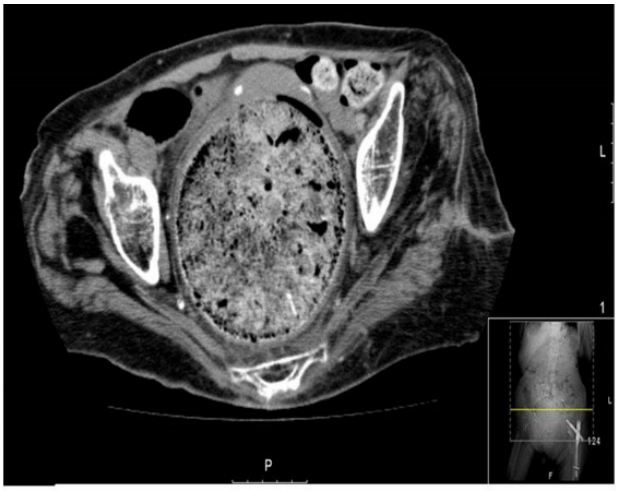

FIGURE 2: EXUBERANT FECALITHS

euros are spent annually on laxatives and more than 50,000 medical appointments.[3] In the USA, 820 million dollars are spent on laxatives, and in the UK, over 48 million pounds, thirty times the amount spent on high blood pressure treatment.[2]

Constipation is associated with complications such as symptoms of the inferior urinary tract and fecal impaction, which may lead to perforation of the colon. The complications are more frequent in hospitalized or bedridden patients with neurodegenerative diseases.[1,2]

One of the aspects of constipation that makes it difficult to deal with is the absence of an agreed-upon definition. The most common criterion is the limited number of three evacuations a week, although the main complaints are due to hard or irregular feces and an excessive effort in defecation.[1,2] The Rome III criteria are currently the most commonly used.[2,4]

It is important to clearly make a distinction between primary constipation – which may be divided into normal intestinal transit, slow intestinal transit, and dysfunction of the pelvic pavement – and secondary constipation due to medicines, neuropathic and myopathic alterations, hydro electrolyte alterations, and intestinal and endocrine-metabolic diseases. The therapeutic strategy varies according to the etiology.[1,2]

Currently, there are no guidelines for treating constipation. Nonpharmacological measures depend on a lifestyle change with increased ingestion of liquids and fiber, as well as physical exercise. However, these remedies are often ineffective in the elderly, so they require a multifactorial approach. The pharmacological measures include the use of laxatives, which should be prescribed individually bearing in mind the elderly's cardiovascular and renal conditions and their regular medication. In cases of constipation resistant to common laxatives, new agents such as lubiprostone, linaclotide, plecanatide, prucalopride, and elobixibat are available. These new agents have proven to be safe and efficient in adults but are only partially validated for the elderly.[1,2]

We insist on the importance of creating guidelines that define constipation and its treatment in the elderly so these patients' quality of life can be genuinely improved and complications avoided.

All the authors contributed to the manuscript and read the final version.

There is no conflits of interest.

RESUMO

A obstipação crônica é um diagnóstico com alta prevalência comum em idosos. A constipação afeta a qualidade de vida das pessoas doentes, carregando muitas complicações clínicas.

PALAVRAS-CHAVE: *Constipação intestinal. Idoso. Lesão renal aguda.*

REFERENCES

1. De Giorgio R, Ruggeri E, Stanghellini V, Eusebi LH, Bazzoli F, Chiarioni G. Chronic constipation in the elderly: a primer for the gastroenterologist. BMC Gastroenterol. 2015;15:130.
2. Couto JRS. Obstipação crónica no idoso: opções terapêuticas [Dissertação de mestrado]. Coimbra: Faculdade de Medicina da Universidade de Coimbra; 2014. 79p.
3. Villanueva T, Alves MM. Obstipação intestinal e impactação fecal. Programa Harvard Medical School Portugal. [Cited 2018, April 28]. Available from: https://hmsportugal.wordpress.com/2012/04/10/obstipacao-intestinal-e-impactacao-fecal/
4. Bharucha AE, Pemberton JH, Locke GR 3rd. American Gastroenterological Association technical review on constipation. Gastroenterology. 2013;144(1):218-38.

Downregulation of lncrna uca1 as a diagnostic and prognostic biomarker for ovarian endometriosis

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SUMMARY

OBJECTIVE: *Ovarian endometriosis seriously affects the quality of life of females, and long non-coding RNA lncRNA urothelial carcinoma-associated 1 (UCA1) plays pivotal roles in the pathogenesis of various ovarian diseases. However, the involvement of lncRNA UCA1 in ovarian endometriosis remains unknown to date. Therefore, the present study aims to study the role of UCA1 in ovarian endometriosis.*

METHODS: *A total of 98 patients with ovarian endometriosis and 28 healthy females were included. The expression of lncRNA UCA1 in ectopic and eutopic endometrium tissues of ovarian endometriosis patients and controls was detected using qRT-PCR. A ROC curve analysis was performed to evaluate the diagnostic values of serum lncRNA UCA1 for ovarian endometriosis. Patients were followed up for 2 years after discharge, and the recurrence of ovarian endometriosis was recorded.*

RESULTS: *The expression level of lncRNA UCA1 was significantly higher in ectopic endometrium tissues than in paired eutopic endometrium tissues for most of the patients. The serum lncRNA UCA1 level showed no significant correlations with either patients' age or living habits. After the treatment, the serum lncRNA UCA1 level increased, and serum levels of lncRNA UCA1 on the day of discharge were significantly lower in patients with recurrence than those in patients without recurrence. Conclusion: The downregulation of lncRNA UCA1 is involved in the pathogenesis of ovarian endometriosis and may serve as a promising diagnostic and prognostic biomarker for the disease.*

KEYWORDS: *Endometriosis. RNA, Long Noncoding. Biomarkers. Endometrium/pathology.*

INTRODUCTION

Endometriosis is the abnormal growth of endometrial tissue outside the uterine cavity. As a chronic gynecological disease, it can cause chronic pelvic pain and infertility[1]. Endometriosis may affect different organs and tissues. Based on the sites of endometrial-type mucosa, endometriosis can be divided into different subtypes such as ovarian and peritoneal endometriosis[2]. Ovarian endometriosis is one of the most common types and can reduce or even eliminate the fertility potential of females. The incidence of ovarian endometriosis is predicted to be increasing among females worldwide due to the extended ovulatory menstruation period[3]. At present, laparoscopic surgery is the most commonly used treatment for patients with ovarian endometriosis[4]. However, the recurrence rate of ovarian endometriosis within 2 years for patients without proper postoperative treatment can be as high as 40% [5]. Even for patients

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with appropriate postoperative treatment, the incidence is still around 10% [6]. Therefore, how to inhibit postoperative recurrence is the primary objective of the treatment for ovarian endometriosis.

Long non-coding RNA lncRNA urothelial carcinoma-associated 1 (UCA1) has been proved to participate in the pathogenesis of various human diseases including different types of ovarian diseases, such as ovarian cancer [7]. A recent microarray study has shown that lncRNA UCA1 expression was inhibited in patients with ovarian endometriosis, [8] indicating the potential involvement of lncRNA UCA1 in the disease. In this study, the expression of lncRNA UCA1 in ectopic endometrium tissues and paired eutopic endometrium tissues of ovarian endometriosis patients as well as serum from both patients and controls were detected. A ROC curve analysis was performed to evaluate the diagnostic value of serum lncRNA UCA1 for ovarian endometriosis. Patients were followed up for 2 years after discharge, and the recurrence of ovarian endometriosis was recorded. We found that the downregulation of lncRNA UCA1 is very likely to be involved in the pathogenesis of ovarian endometriosis and the serum lncRNA UCA1 may serve as a promising diagnostic and prognostic biomarker for the disease.

METHODS

Patients

A total of 98 patients with ovarian endometriosis (diameter between 6 and 7 cm under laparoscopy) were selected from January 2013 to January 2015 in the Tongren Hospital of the Wuhan University. The age of those patients ranged from 22 to 41 years old. All patients were diagnosed using pathological examinations and received laparoscopic surgeries so that ectopic endometrium tissues and paired eutopic endometrium tissues from the same person were obtained during surgical operations. Patients with abnormal menstrual cycles and treated with hormonal medicine for 3 months before surgery were excluded. The American Fertility Society (AFS) staging was performed per the revised American Society for Reproductive Medicine staging (1997). There were 19 patients in stage I, 21 patients in stage II, 33 patients in stage III and 25 patients in stage IV. No significant differences in age and other basic background information were found among patients in different AFS stages. At the same time, 28 normal healthy females with the same age distribution were also selected to serve as the control group, which was only subjected to blood extraction. The Tongren Hospital of the Wuhan University ethics committee approved this study, and all the patients signed informed consent. All patients were followed up for 2 years through telephone and outpatient service. The recurrence of ovarian endometriosis was diagnosed by pathological examinations.

Preparation of serum samples

Fasting blood (20 ml) was extracted from each patient one day before the surgery and on the day of discharge. Blood samples were kept at room temperature for 2 hours, followed by centrifugation at 1200g for 15 min to collect serum. The serum was kept at -80 ºC before use.

Real-time quantitative PCR (qRT-PCR)

Trizol reagent (Invitrogen, USA) was used to extract total RNA from serum samples, ectopic and paired eutopic endometrium tissues. All RNA samples were tested using NanoDrop™ 2000 Spectrophotometers (Thermo Fisher Scientific, USA), and only the ones that showed a ratio of A260/A280 between 1.8 and 2.0 were used in the reverse transcription for cDNA synthesis using iScript™ cDNA Synthesis Kit (Bio-Rad, CA, USA). The PCR reaction system was prepared using SYBR® Green Real-Time PCR Master Mixes (Thermo Fisher Scientific, USA). The PCR reactions were performed on the ABI 7500 System. The primers used in PCR reactions were: 5'-TTTATGCTTGAGCCTTGA-3' (forward) and 5'-CTTGCCTGAAATACTTGC-3' (reverse) for lncRNA UCA1; 5'-GACCTCTATGCCAACACAGT-3' (forward) and 5'-AGTACTTGCGCTCAGGAGGA-3' (reverse) for β-actin. The PCR reaction conditions were: 95 °C for 40 s, followed by 40 cycles of 95 °C for 20 s and 60 °C for 45 s. Ct (threshold cycle) values were processed using the 2-ΔΔCT method. The relative expression level of lncRNA UCA1 was normalized to endogenous control β-actin.

STATISTICAL ANALYSIS

SPSS19.0 (SPSS Inc., USA) was used for statistical analysis. Measurement data were recorded by (`x±s), and comparisons between two groups were performed using the t-test. While comparisons among

multiple groups were performed using one-way analysis of variance and LSD test, the correlation between serum levels of lncRNA UCA1 and clinical data of patients was analyzed using the chi-square test. A ROC curve analysis was used to evaluate the diagnostic value of serum lncRNA UCA1 for ovarian endometriosis patients in different AFS stages. $P < 0.05$ was considered to be statistically significant.

RESULTS

Comparison of expression levels of lncRNA UCA1 in ectopic and paired eutopic endometrium tissues from ovarian endometriosis patients

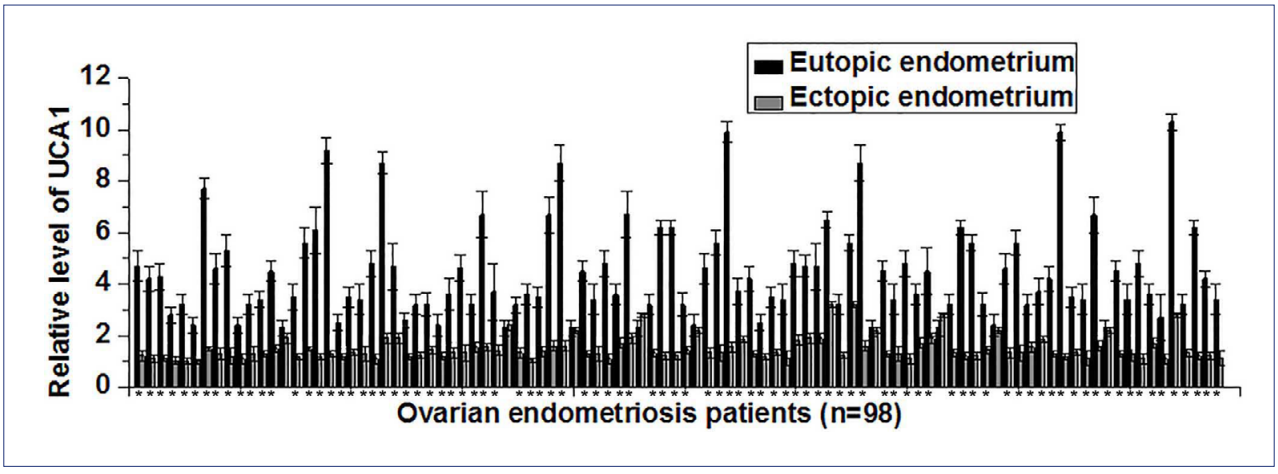
The expression of lncRNA UCA1 in ectopic and paired eutopic endometrium tissues in patients with ovarian endometriosis was detected by qRT-PCR. As shown in Fig. 1a, the expression level of lncRNA UCA1 was significantly lower in ectopic endometrium tissues than that in paired eutopic endometrium tissues in 89 out of 98 patients with ovarian endometriosis ($p < 0.05$).

Serum levels of lncRNA UCA1 on healthy controls and patients with different stages of ovarian endometriosis

Serum levels of lncRNA UCA1 (one day before surgery) on healthy controls and patients with different stages of ovarian endometriosis were measured using qRT-PCR. As shown in Fig. 1b, compared with healthy controls (9.8± 3.3), serum levels of lncRNA UCA1 were significantly reduced in patients with different stage I (7.7± 1.8), stage II (4.9± 1.1), stage III (4.1± 0.5) and stage IV (2.9 ± 0.3) ovarian endometriosis ($p < 0.05$). Also, serum levels of lncRNA UCA1 were significantly reduced with the increased AFS stages ($p < 0.05$).

Diagnostic values of serum lncRNA UCA1 on patients with different stages of ovarian endometriosis

A ROC curve analysis was performed to evaluate the diagnostic values of serum lncRNA UCA1 on patients with different stages of ovarian endometriosis. As shown in Fig. 2A, the area under the curve (AUC)



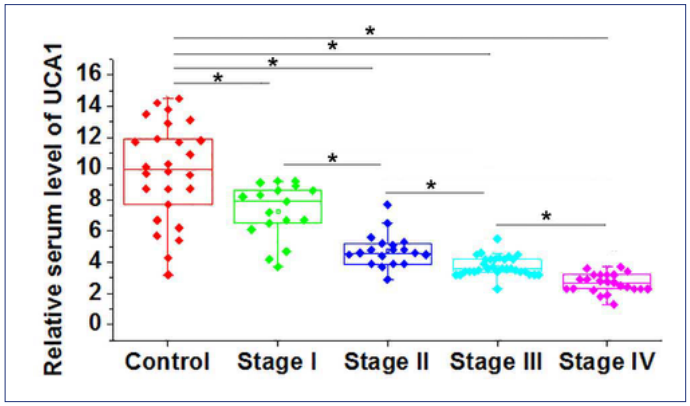


FIGURE 1. A. Expression of lncRNA UCA1 in ectopic endometrium tissues and paired eutopic endometrium tissues in patients with ovarian endometriosis. A. Serum levels of lncRNA UCA1 in healthy controls and patients with different stages of ovarian endometriosis. Data were analyzed using one-way analysis of variance, followed by LSD test.

Notes: *compared with paired eutopic endometrium tissues, $p < 0.05$

of serum lncRNA UCA1 in the diagnosis of ovarian endometriosis patients in Stage I was 0.7509 with 95 % confident interval of 0.6109 to 0.8910, specificity of 80.1% and sensitivity of 76.7% (p = 0.003820). For stage II, AUC was 0.9175 with 95 % confident interval of 0.8308 to 1.004, specificity of 85.6% and sensitivity of 81.1% (p < 0.0001; Fig 2, panel B). For stage III, AUC was 0.9605 with 95 % confident interval of 0.8982 to 1.023, specificity of 89.1% and sensitivity of 88.1% (p < 0.0001; Fig 2. panel C). For stage IV, AUC was 0.9921 with 95 % confident interval of 0.9747 to 1.010, specificity of 90.5 % and sensitivity of 89.0% (p < 0.0001; Fig 2, panel D).

Correlation between serum levels of lncRNA UCA1 and the clinical data of patients

According to the median serum level of lncRNA UCA1, the 98 patients with ovarian endometriosis were divided into a high expression and a low expression group. The serum level of lncRNA UCA1 was not affected by the patients' age and living habits, including vegetarian, smoking and drinking (p < 0.05).

Comparison of serum levels of lncRNA UCA1 before and after treatment and between patients with or without recurrence of ovarian endometriosis

Compared with the pretreatment level of serum lncRNA UCA1, the serum level of lncRNA UCA1 was significantly increased in those patients after treatment (p <0.05; Fig 3, panel A). A total of 28 cases of ovarian endometriosis recurrence were observed during a two-year follow-up. The serum levels of lncRNA UCA1 (on the day of discharge) were significantly lower in patients with recurrence than in patients without recurrence (p <0.05; Fig 3, panel B).

DISCUSSION

The abnormal expression of lncRNA UCA1 has been observed in the development of various types of human diseases. In the study of lung cancer, Wang et al.[9] found that lncRNA UCA1 expression in plasma was more significantly upregulated in lung cancer patients than in healthy controls, and the increased

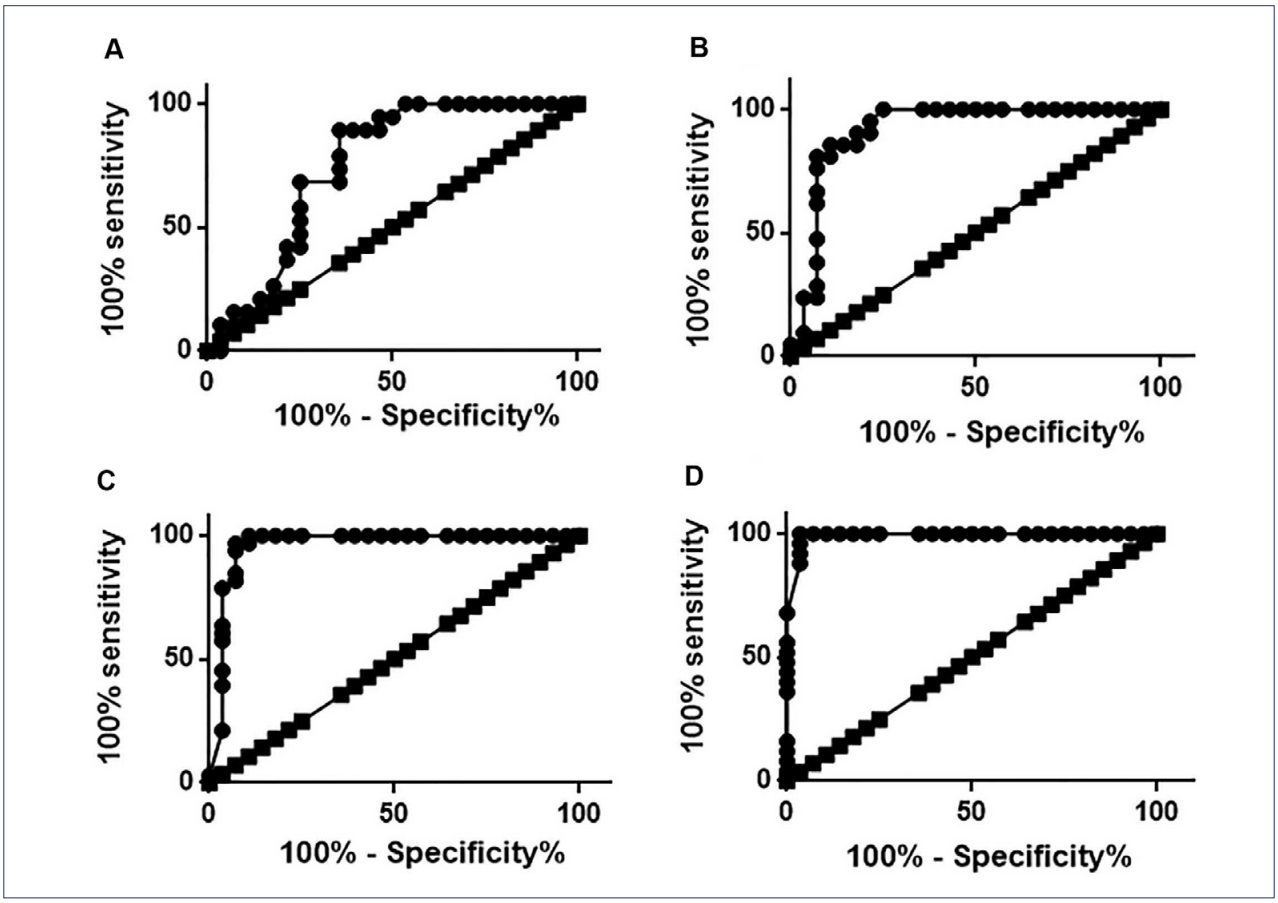


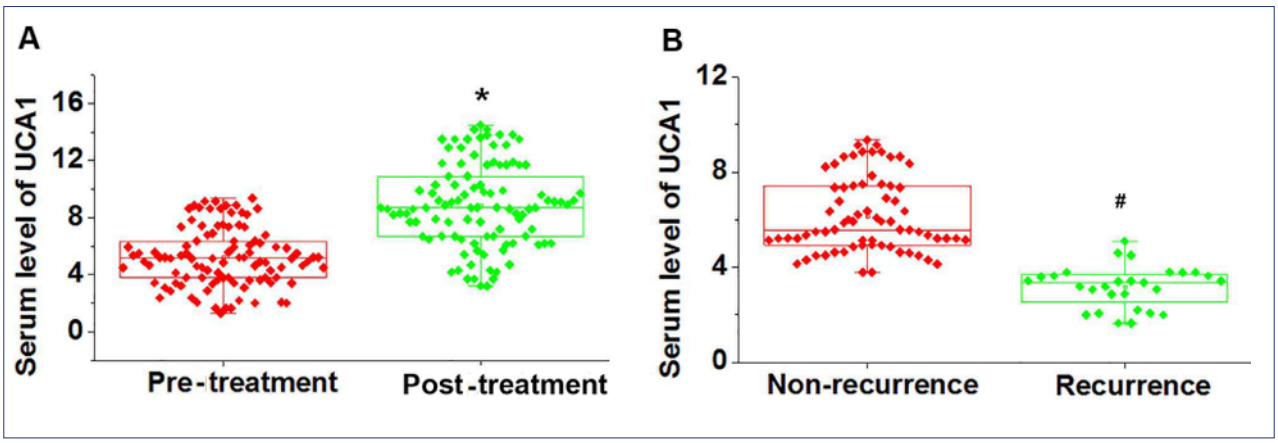
FIGURE 2. Diagnostic value of serum lncRNA UCA1 for patients with different stages of ovarian endometriosis. A-D indicate diagnostic values of serum lncRNA UCA1 for ovarian endometriosis patients in stage I, II, III and IV, respectively.

expression level of lncRNA UCA1 contributed to the progression of the disease. Upregulation of lncRNA UCA1 was also observed in breast cancer and was proved to be correlated with an accelerated epithelial-mesenchymal transition of the breast tumor. In contrast, the occurrence of acute myocardial infarction was accompanied by the reduced level of circulating lncRNA UCA1. LncRNA UCA1 showed different expression patterns in different ovarian diseases. LncRNA UCA1 overexpression not only related to the progression of ovarian cancer but was also responsible for poor treatment outcomes after adjuvant chemotherapy. However, lncRNA UCA1 expression was downregulated in patients with ovarian endometriosis [8]. Consistent with previous studies, in this study, the expression level of lncRNA UCA1 was found to be significantly lower in ectopic endometrium tissues than in paired eutopic endometrium tissues in 89 out of 98 patients with ovarian endometriosis. Also, serum levels of lncRNA UCA1 were significantly reduced in patients with different stages of ovarian endometriosis in comparison with healthy controls, and serum levels of lncRNA UCA1 were significantly reduced with increased AFS stages. These data suggest that the downregulation of lncRNA UCA1 is involved in the development and progression of ovarian endometriosis.

In this study, a ROC curve analysis was performed to evaluate the diagnostic values of serum lncRNA UCA1 on patients with different stages of ovarian endometriosis. We found that serum lncRNA UCA1 level could be referred to diagnose ovarian endometriosis patients accurately. Especially for those in AFS stage IV, AUC reached 0.9921 with 95 % confident interval of 0.9747 to 1.010. Therefore, serum lncRNA UCA1 may serve as a promising diagnostic marker for ovarian endometriosis. The expression of certain lncRNAs is significantly affected by an individual's age and living habits such as smoking, alcohol consumption, and diet. In this study, no significant correlations were found between serum levels of lncRNA UCA1 and patients' age and living habits including drinking, smoking, and vegetarianism. Therefore, these variables may be ignored in the diagnosis of ovarian endometriosis using lncRNA UCA1.

After the treatment, serum levels of lncRNA UCA1 were significantly increased compared with the pretreatment levels. At present, the treatment for ovarian endometriosis using laparoscopic surgery is still challenged by the high postoperative recurrence rate. The recurrence rate of ovarian endometriosis within 2 years after laparoscopic surgery for patients without proper postoperative treatment reaches 40% [5], and for patients with appropriate postoperative treatment, the incidence is still as high as 10% [6]. In the current study, all patients were followed up for two years after discharge, and none of them was missed. During follow-up, a total of 28 cases of recurrence were observed, which accounted for 28.6 % of the total. A comparison of serum levels of lncRNA UCA1 on the day of discharge showed that the serum levels of lncRNA UCA1 were significantly lower in patients with recurrence than in patients without recurrence. These results suggest that lncRNA UCA1 may serve

FIGURE 3. Comparison of serum levels of lncRNA UCA1 regarding treatment and recurrence of ovarian endometriosis
A. Comparison of serum levels of lncRNA UCA1 before and after treatment; B. Comparison of serum levels of lncRNA UCA1 between patients with and without recurrence of ovarian endometriosis.



Notes: *compared with pretreatment level, p < 0.05; #compared with patients with recurrence, p < 0.05

as a target for the treatment of ovarian endometriosis, and low serum levels of lncRNA UCA1 after laparoscopic surgery may predict recurrence of the disease. A recent study revealed that different types of endometriosis might have a different gene expression pattern. Our future study will focus on identifying the functionality of lncRNA UCA1 in different types of endometriosis, such as superficial and deep infiltrating endometriosis[10].

CONCLUSION

The expression level of lncRNA UCA1 was significantly higher in ectopic endometrium tissues than that in paired eutopic endometrium tissues in most patients. Compared with healthy controls, serum levels of lncRNA UCA1 were decreased in patients with ovarian endometriosis, and serum levels of UCA1 were further decreased with the progression of this disease. The serum lncRNA UCA1 level showed no significant correlations with either patients' age or living habits. After the treatment, the serum lncRNA UCA1 level was increased, and the serum levels of lncRNA UCA1 on the day of discharge were significantly lower in patients with recurrence than in those without recurrence. Based on these data, we can conclude that the downregulation of lncRNA UCA1 is involved in the pathogenesis of ovarian endometriosis and might be a promising diagnostic and prognostic biomarker for the disease. Our study is limited by the small sample size due to the limited resources of patients and healthy volunteers. Future studies with more significant sample size are needed to confirm these conclusions further.

RESUMO

OBJETIVO: *A endometriose ovariana afeta seriamente a qualidade de vida das mulheres, e o carcinoma urotelial 1 de urcélio de RNA não codificador longo 1 (UCA1) desempenha um papel crucial na patogênese de várias doenças ovarianas. No entanto, o envolvimento do lncRNA UCA1 na endometriose ovariana permanece desconhecido até o momento. Portanto, o presente estudo tem como objetivo estudar o papel do UCA1 na endometriose ovariana. Métodos: Um total de 98 pacientes com endometriose ovariana e de 28 mulheres saudáveis foi incluído. A expressão de lncRNA UCA1 em tecidos de endométrio ectópico e eutópico de pacientes com endometriose ovariana e controles foi detectada por qRT-PCR. A análise da curva ROC foi realizada para avaliar os valores diagnósticos do lncRNA UCA1 sérico para endometriose ovariana. Os pacientes foram acompanhados por dois anos após a alta, e a recorrência da endometriose ovariana foi registrada.*

RESULTADOS: *O nível de expressão do lncRNA O UCA1 foi significativamente maior nos tecidos do endométrio ectópico do que nos tecidos do endométrio eutópico pareados para a maioria dos pacientes. O nível sérico de UCA1 foi diminuído com a progressão da endometriose ovariana. O soro UCA1 pode ser usado para diagnosticar com precisão a endometriose ovariana. O nível sérico de UCA1 não apresentou correlações significativas com a idade ou com os hábitos de vida dos pacientes. Após o tratamento, o nível sérico do lncRNA UCA1 foi aumentado, e os níveis séricos de lncRNA UCA1 no dia da alta foram significativamente menores nos pacientes com recidiva do que naqueles sem recorrência.*

CONCLUSÃO: *A regulação negativa do lncRNA UCA1 está envolvida na patogênese da endometriose ovariana e pode servir como um promissor biomarcador diagnóstico e prognóstico para a doença.*

PALAVRAS-CHAVE: *Endometriose. RNA longo não codificante. Biomarcadores. Endométrio/patologia.*

REFERENCES

1. Vercellini P, Viganò P, Somigliana E, Fedele L. Endometriosis: pathogenesis and treatment. Nat Rev Endocrinol. 2014;10(5):261-75.
2. Nisolle M, Donnez J. Peritoneal endometriosis, ovarian endometriosis, and adenomyotic nodules of the rectovaginal septum are three different entities. Fertil Steril. 1997;68(4):585-96.
3. Bulun SE. Ovarian endometriosis: the nemesis of eggs. Fertil Steril. 2014;101(4):938-9.
4. Duffy JM, Arambage K, Correa FJ, Olive D, Farquhar C, Garry R, et al. Laparoscopic surgery for endometriosis. Cochrane Database Syst Rev. 2014;(4):CD011031.
5. Cucinella G, Granese R, Calagna G, Svelato A, Saitta S, Tonni G, et al. Oral contraceptives in the prevention of endometrioma recurrence: does the different progestins used make a difference? Arch Gynecol Obstet. 2013;288(4):821-7.
6. Somigliana E, Vercellini P, Vigano P, Benaglia L, Busnelli A, Fedele L. Postoperative medical therapy after surgical treatment of endometriosis: from adjuvant therapy to tertiary prevention. J Minim Invasive Gynecol. 2014;21(3):328-34.
7. Yang Y, Jiang Y, Wan Y, Zhang L, Qiu J, Zhou S. UCA1 functions as a competing endogenous RNA to suppress epithelial ovarian cancer metastasis. Tumor Biol. 2016;37(8):10633-41.
8. Sun PR, Jia SZ, Lin H, Leng JH, Lang JH. Genome-wide profiling of long noncoding ribonucleic acid expression patterns in ovarian endometriosis by microarray. Fertil Steril. 2014;101(4):1038-46.
9. Wang HM, Lu JH, Chen WY, Gu AQ. Upregulated lncRNA-UCA1 contributes to progression of lung cancer and is closely related to clinical diagnosis as a predictive biomarker in plasma. Int J Clin Exp Med. 2015;8(7):11824-30.
10. Fettback PB, Pereira RM, Rocha AM, Soares JM Jr, Smith GD, Baracat EC, et al. Expression of stem cell-related genes in the endometrium and endometriotic lesions: a pilot study. Gynecol Endocrinol. 2016;32(1):82-6.

Comparison of different types of endovascular mechanical embolectomy in acute ischemic stroke

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SUMMARY

Background: To compare the treatment efficacy of different types of endovascular mechanical embolectomy in acute ischemic stroke (AIS). Material and Methods: A total of 89 patients with AIS were selected in our hospital from January 2014 to January 2016 and divided into tPA group (n=27), tPA+Trevo group (n=30) and tPA+Solitaire FR group (n=32) for different treatments. Treatment effectiveness was evaluated using NIHSS and mRS system. The NIHSS score, vascular recanalization rate and postoperative complications were compared among groups. Results: The NIHSS score of the tPA group was significantly lower than that of other two groups at 1 d after the operation ($p < 0.05$), but it was significantly higher than that of other two groups at 3 d and 3 w after the operation ($p < 0.05$). After the treatment, no significant difference in NIHSS score was found between the tPA+Trevo and tPA Solitaire FR groups. The revascularization rate was significantly higher, but the mortality rate in 90 d was significantly lower in the tPA+Trevo and tPA+Solitaire FR groups than that in the tPA group ($p < 0.05$), and no significant difference was found between the tPA+Trevo and tPA+Solitaire FR groups. The incidence rate of symptomatic intracranial hemorrhage was significantly lower in the tPA+Solitaire FR group than that in tPA+Trevo group ($p < 0.05$) or tPA group ($p < 0.01$). Significantly more patients with mRS no higher than 2 points were found in the tPA+Trevo and tPA+Solitaire FR groups than those in tPA group ($p < 0.05$), and no significant difference was found between the tPA+Trevo and tPA+Solitaire FR groups. Conclusion: TPA+Solitaire FR is a type of thrombectomy that is superior to tPA and tPA+Trevo in the treatment of patients with AIS.

Keywords: Stroke. Embolectomy. Tissue plasminogen activator.

INTRODUCTION

A cerebrovascular accident (CVA), also called stroke, is a serious disease that ranks in second place among all death-causing diseases [1]. Besides death, stroke can also cause disability, which in turn seriously affects the patients' quality of life and brings heavy economic burden to both patients and their families [2-4]. According to the pathogenesis, stroke can be divided into subtypes including hemorrhagic stroke and ischemic stroke. As the dominant type of stroke, ischemic stroke accounts for about 80 % of all stroke cases [5-7]. An ischemic stroke is caused by insufficient oxygen and blood supply to the brain, which can be caused by a variety of reasons, and in turn leads to brain tissue necrosis. As a type of ischemic stroke, acute ischemic stroke (AIS) is characterized by the sudden loss of blood supply to brain tissues [8]. Clinical studies have proven that thrombotic or embolic occlusion is the main cause of most AIS [9].

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Therefore thrombolysis has become a central part of the AIS treatment.

Intravenous recombinant tissue plasminogen activator, or tPA, has been recognized as the standard treatment for AIS. But no more than half of the patients showed complete recovery after the treatments with tPA within 4.5 h of stroke onset, and the motility rate is high [10]. The application of tPA treatment is also challenged by the existence of contraindications, and only less than 10 % of AIS patients can be treated with it. In addition, tPA treatment usually fails to provide satisfactory outcomes for occlusion in main artery. The combined use of tPA and mechanical thrombectomy devices has been shown to be able to significantly improve the recanalization rate and clinical outcome. Solitaire FR stent and Trevo stent are two mostly used thrombectomy devices. However, the comparison of treatment efficacy and safety of those two systems still hasn't been reported.

In this study, a total of 89 patients with AIS were selected to be treated with different measures. The treatment outcomes and safety of those treatments were compared.

MATERIALS AND METHODS

Patients

A total of 89 patients with AIS enrolled from January 2014 to January 2016 in the Suqian People' hospital of Nanjing Durm Tower hospital Group were selected. Patients were divided into tPA group (n=27), tPA+Trevo group (n=30) and tPA+Solitaire FR group (n=32) for different treatments according to their willingness and the physician's recommendations. Inclusion criteria: (1) Patients met the diagnostic criteria of AIS in China; (2) Patients diagnosed by imaging and biochemical examinations. Exclusion criteria: (1) patients with severe heart, liver, and kidney dysfunction; (2) patients who were not willing to receive treatment.

Treatment

Patients in the tPA group were treated with emergent intravenous thrombolysis: patients were treated with recombinant tissue plasminogen activator (rt-PA) alteplase at a dose of 0.9mg/kg through intravenous injection (the first 10 %) and continuous intravenous pumping for 1 h. The treatment was stopped if patients showed severe headache, hypertension, and nausea or vomiting. Patients in the TAP+Solitaire FR group were treated with emergent intravenous thrombolysis combined with mechanical thrombectomy with solitaire FR stent: whole-brain CT digital subtraction angiography was performed to identify the cerebral arterial occlusion site and responsible vessels. A micro-guidewire was pushed through the thrombus occlusion site under the guidance of a path map. The stent delivery catheter was introduced to go through the micro-guidewire and pass the distal end of occlusion, and the Solitaire FR stent was introduced to go through the catheter. The Solitaire FR stent was opened and the opening state was maintained for 2-3 min after the recovery of local blood flow. This operation can be repeated no more than 3 times. Imaging examination was performed to evaluate the treatment efficacy. Patients in the tAP+Trevo group were treated with emergent intravenous thrombolysis that combined mechanical thrombectomy with the Trevo stent using the same method described above.

Observation indicators

Treatment effectiveness was evaluated using the NIHSS system at 1d, 3d, and 3w before and after the treatment. A lower score indicates a better outcome. The vascular recanalization rate and symptomatic intracranial hemorrhage rate were recorded. Treatment outcomes were also evaluated using modified Rankin Scale (mRS), and patients with mRS score ≤ 2 were recorded.

Statistical analysis

SPSS19.0 software was used for all of the statistical analyses. Comparisons between groups were performed by t-test or chi-square test and $p < 0.05$ was considered to be statistically significant.

RESULTS

Comparison of general information between groups

No significant differences in age, gender, weight, cultural level, disease types and the sites of intracranial occlusion were found among three groups of patients (Table 1).

Comparison of imaging results before and after mechanical embolectomy

Using a patient from the tAP+Trevo group as an example, whole cerebral angiography showed oc-

TABLE1 COMPARISON OF GENERAL INFORMATION AMONG GROUPS

	tPA n=27	tPA + Trevo n=30	tPA + Solitaire FR n=32	
Age	65.17(5.84)	68.87(7.69)	63.87(4.13)	p>0.05
Gender				p>0.05
	Male	15	17	17
	Female	12	13	15
Weight	54.81(7.85)	55.43(6.46)	56.43(5.87)	p>0.05
Cultural level				p>0.05
	Junior high school	7	9	11
	Senior high school	10	11	12
	College degree and above	10	10	9
Type of disease				p>0.05
	Hypertension	16	20	19
	Diabetes	6	6	8
	Atrial fibrillation	5	4	5
Site of intracranial occlusion				p>0.05
Carotid T	6	6	8	
Proximal M1	9	11	11	
Distal M1	5	4	5	
M2		3	5	3
Basilar artery	2	1	2	
Proximal ICA stenosis >70%	2	3	3	

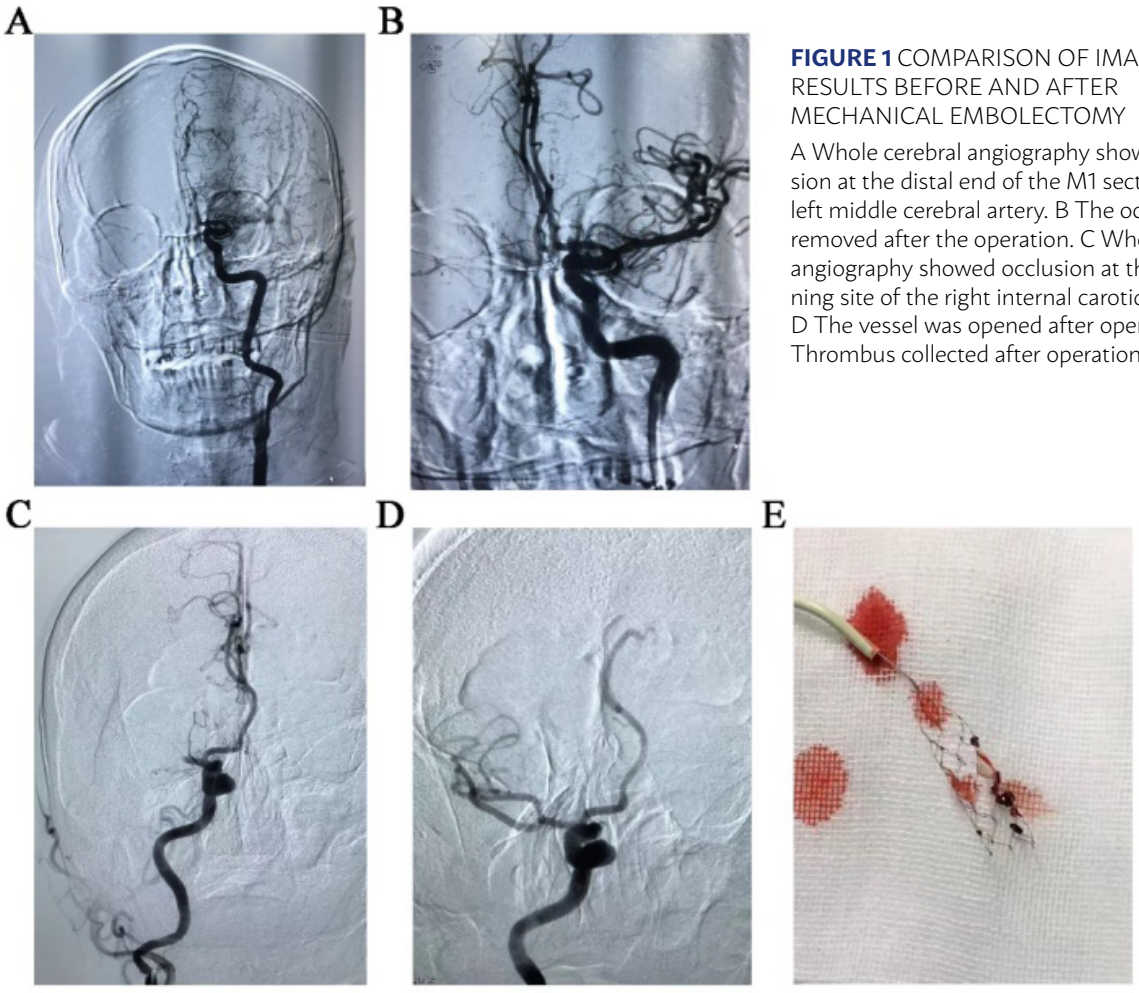

FIGURE 1 COMPARISON OF IMAGING RESULTS BEFORE AND AFTER MECHANICAL EMBOLECTOMY

A Whole cerebral angiography showed occlusion at the distal end of the M1 section of the left middle cerebral artery. B The occlusion was removed after the operation. C Whole cerebral angiography showed occlusion at the beginning site of the right internal carotid artery. D The vessel was opened after operation. E Thrombus collected after operation.

TABLE 2. COMPARISON OF TREATMENT OUTCOMES

	No.	TICI	SICH	Mortality 90 D (%)	mRS≤2
tPA	27	16(59.26)	3(10.69)	6(22.22)	9(33.33)
tPA + Trevo	30	22(73.30)	2(6.67)	5(16.67)	17(56.67)
tPA + Solitaire FR	32	26(81.25)	1(3.125)	4(12.50)	20(62.50)
p value[a]	<0.05	>0.05	>0.05	<0.05	
p value[b]	<0.05	<0.05	<0.05	<0.05	
p value[c]	>0.05	<0.01	>0.05	>0.05	

Notes: **a**, comparison between the tPA+Trevo and tPA groups; **b**, comparison between the tPA+ Solitaire FR and tPA groups; **c**, comparison between the tPA + Trevo and tPA+Solitaire FR groups; TICI, Thrombolysis In Cerebral Infarction; SICH, symptomatic intracranial hemorrhage.

clusion at the distal end of the M1 section of the left middle cerebral artery (Fig. 1A); the occlusion was removed after mechanical thrombectomy with Trevo stent (Fig. 1B). Using a patient from the tAP+Trevo group as another example, whole cerebral angiography showed occlusion at the beginning site of the right internal carotid artery (Fig. 1C), and the vessel was opened after mechanical thrombectomy with Solitaire FR stent (Fig. 1D).

Comparison of NIHSS scores among three groups before and after the treatment

There was no significant difference in NIHSS scores among three groups before the treatment. The NIHSS score of the tPA group was significantly lower than that of other two groups at 1 d after the operation ($p < 0.05$), but it was significantly higher than that of other two groups at 3 d and 3 w after the operation ($p < 0.05$). No significant difference in NIHSS scores was found between the tPA+Trevo and tPA Solitaire FR groups after the operation.

Comparison of treatment outcomes among three groups

The revascularization rate of the tPA+Trevo and tPA+Solitaire FR groups was significantly higher than that of the tPA group ($p < 0.05$), but no significant difference was found between the tPA+Trevo and tPA+Solitaire FR group. The incidence rate of symptomatic intracranial hemorrhage was significantly lower in the tPA+Solitaire FR group than that in the tPA+Trevo ($p < 0.05$) and tPA groups ($p < 0.01$), and no significant difference was found between the tPA+Trevo and tPA groups. The mortality rate at 90 d was lower in the tPA+Trevo and tPA+Solitaire FR groups than that in tPA groups, but significant difference was only found between the tPA+Solitaire FR and tPA groups ($p < 0.05$). The number of patients with mRS no higher than 2 points is significantly bigger in the tPA+Trevo and tPA+Solitaire FR groups than in the tPA group ($p < 0.05$), but no significant difference was found between the tPA+Trevo and tPA+Solitaire FR groups (Table 2).

DISCUSSION

AIS is a serious disease that causes unacceptably high morbidity and mortality rates worldwide. Treatment with tPA is the standard for AIS patients. However, the treatment with tPA alone usually fails to provide satisfactory outcomes due to the short treatment time window, which is only 3 to 4.5 hours. Therefore, how to extend this time window has become a focus of the AIS treatment. A previous study reported that the combined use of tPA and 3-methyl-1-phenyl-2-pyrazoline-5-one (a free radical scavenger) could significantly extend the ischemic therapeutic time, which in turn would increase survival rates and reduced the incidence of postoperative complications. In another study, Fisher and Albers found that the application of tPA under the guidance of advanced imaging can also extend the therapeutic time window of AIS. Although those newly developed techniques showed promising outcomes in the AIS treatment in comparison with tPA alone, all those treatments may fail to increase the revascularization rate, especially for occlusions in main artery due to the lack of endovascular treatment.

In recent years, endovascular mechanical embolectomy combined with tPA treatment has been widely used in the treatment of AIS, and it showed high efficiency in enhancing revascularization as well as expanding the treatment time window. A series of mechanical thrombectomy devices and stent retrievers have been developed. Since these are the two most commonly used mechanical thrombecto-

my devices, more and more patients have accepted the application of Solitaire FR stent and Trevo stent. As a kind of clot retrieval, Solitaire Flow stent contains a self-expanding stent retriever that can remove proximal vessel occlusion in patients with AIS to restore blood flow. A prospective study carried out by Pereira et al. found that the treatment with Solitaire Flow stent could significantly improve clinical outcomes and reduce postoperative complications. Another type of clot retrieval, Trevo works in a similar way to that of Solitaire FR stent. In an animal model, Trevo was proved to be effective in inducing reperfusion of blocked arteries without leading to significant disruption of the vascular integrity.

To date, the comparison of the treatment efficacy and safety of Solitaire FR as well as Trevo has not been well studied although there was one relevant study by Mendonça et al. In that study, they reported that the revascularization rate of patients who received Solitaire Stent treatment (60 %) is lower than that of patients in the Trevo group (77 %), but the difference was not significant. The incidence rate of postoperative symptomatic intracranial hemorrhage was lower in the Trevo group (0 %) than that in Solitaire Stent group (15 %). In addition, no significant difference in mortality rate after 3 months was found between the Trevo stent and Solitaire Stent groups. Consistent with this study, in our study, no significant difference in revascularization rate was found between the tPA+Trevo and tPA+Solitaire FR groups, but the revascularization rate of tPA+Trevo and tPA+Solitaire FR groups was significantly higher than that of tPA group. In addition, mortality rate at 90 d was lower in tPA+Trevo and tPA+Solitaire FR groups than in the tPA group. Those results suggest that, compared with the use of tPA alone, the combined use of Trevo stent and tPA+Solitaire stent can significantly increase the revascularization rate and reduce the mortality rate. Different results were also found in our study. In this study, the incidence rate of symptomatic intracranial hemorrhage was significantly lower in the tPA+Solitaire FR group than that in tPA+Trevo and tPA groups, but no significant difference was found between the tPA+Trevo and tPA groups. This result suggests that, compared with the use of Trevo stent, the Solitaire FR can significantly reduce the incidence of symptomatic intracranial hemorrhage.

In this study, NIHSS and mRS scoring systems were used to evaluate the treatment outcomes as well as the safety of different treatments. The NIHSS score of the tPA group was significantly lower than that of other two groups at 1 d after the operation, but it was significantly higher than that of other two groups at 3 d and 3 w after the operation, indicating that the use of Solitaire and Trevo could only improve the medium- and long-term treatment outcomes but not the short-term outcomes. In addition, a number of patients with mRS that no higher than 2 points is significantly bigger in the tPA+Trevo group and tPA+Solitaire FR groups than in the tPA group, which further confirmed that advantages of the combined treatment.

CONCLUSION

In conclusion, the effectiveness and safety of tPA+Trevo and tPA+Solitaire in the treatment of AIS patients were better than those of tPA alone. tPA+Solitaire is also superior to tPA+Trevo in some aspects. This study is limited by the small sample size. Further studies with bigger sample size are needed to confirm the conclusions of this study.

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RESUMO

OBJETIVO *Comparar a eficácia do tratamento de diferentes tipos de embolectomia mecânica endovascular em acidente vascular cerebral isquêmico agudo (AIS).*

MATERIAL E MÉTODOS *Um total de 89 pacientes com AIS foi selecionado em nosso hospital de janeiro de 2014 a janeiro de 2016, e os pacientes foram divididos em: grupo tPA (n = 27), tPA + grupo Trevo (n = 30) e grupo tPA + Solitaire FR (n = 32) para diferentes tratamentos. A eficácia do tratamento foi avaliada usando NIHSS e sistema mRS. Escore NIHSS, taxa de recanalização vascular e complicações pós-operatórias foram comparados entre os grupos.*

RESULTADOS *A pontuação NIHSS do grupo tPA foi significativamente menor do que a dos outros dois grupos em um dia após a operação (p < 0,05), mas foi significativamente maior do que nos outros dois grupos em três dias e três semanas após a operação (p < 0,05). Após o tratamento, não houve diferença significativa no escore NIHSS entre o grupo tPA + Trevo e o gru-*

po tPA Solitaire FR. A taxa de revascularização foi significativamente maior, mas a taxa de mortalidade em 90 dias foi significativamente menor nos grupos tPA + Trevo e tPA + Solitaire FR do que no grupo tPA (p < 0,05) e não houve diferença significativa entre os grupos tPA + Trevo e tPA + Solitaire FR. A taxa de incidência de hemorragia intracraniana sintomática foi significativamente menor no grupo tPA + Solitaire FR do que no grupo tPA + Trevo (p < 0,05) ou no grupo tPA (p < 0,01). Significativamente mais pacientes com mRS não maiores que 2 pontos foram encontrados no grupo tPA + Trevo e tPA + Solitaire FR do que no grupo tPA (p < 0,05), e nenhuma diferença significativa foi encontrada entre os grupos tPA + Trevo e tPA + Solitaire FR. Conclusão O tPA + Solitaire FR é um tipo de trombectomia superior ao tPA e tPA + Trevo no tratamento de pacientes com EIA.
PALAVRAS-CHAVE: *Acidente vascular cerebral. Embolectomia. Ativador de plasminogênio tecidual.*

REFERENCES

1. Yang J, Yin P, Zhou M, Ou CQ, Li M, Li J, et al. The burden of stroke mortality attributable to cold and hot ambient temperatures: epidemiological evidence from China. Environ Int. 2016;92-93:232-8.
2. Park JS, Hwang NK, Oh DH, Chang MY. Effect of head lift exercise on kinematic motion of the hyolaryngeal complex and aspiration in patients with dysphagic stroke. J Oral Rehabil. 2016;44(5):385-91.
3. Keppel Hesselink JM. NS1209/SPD 502, a novel selective AMPA antagonist for stroke, neuropathic pain or epilepsy? Drug development lessons learned. Drug Dev Res. 2017;78(2):75-80.
4. Seto SW, Chang D, Jenkins A, Bensoussan A, Kiat H. Angiogenesis in ischemic stroke and angiogenic effects of Chinese herbal medicine. J Clin Med. 2016;5(6). pii: E56.
5. Vasileva D, Lubenova D, Mihova M, Grigorova-Petrova K, Dimitrova A. Orthostatic reactivity in patients with ischemic stroke in the chronic period. Open Access Maced J Med Sci. 2015;3(3):397-402.
6. Boisserand LS, Kodama T, Papassin J, Auzely R, Moisan A, Rome C, et al. Biomaterial applications in cell-based therapy in experimental stroke. Stem Cells Int. 2016;2016:6810562.
7. Liang Y, Huang J, Tian J, Cao Y, Zhang G, Wang C, et al. The prevalence and risk factors of stroke in patients with chronic schizophrenia. Neuropsychiatr Dis Treat. 2016;12:1131-4.
8. Powers WJ, Derdeyn CP, Biller J, Coffey CS, Hoh BL, Jauch EC, et al; American Heart Association Stroke Council. 2015 American Heart Association/American Stroke Association Focused Update of the 2013 guidelines for the early management of patients with acute ischemic stroke regarding endovascular treatment: a guideline for healthcare professionals from the American Heart Association/American Stroke Association. Stroke. 2015;46(10):3020-35.
9. Chimowitz MI. Endovascular treatment for acute ischemic stroke: still unproven. N Engl J Med. 2013;368(10):952-5.
10. Wardlaw JM, Murray V, Berge E, del Zoppo G, Sandercock P, Lindley RL, et al. Recombinant tissue plasminogen activator for acute ischaemic stroke: an updated systematic review and meta-analysis. Lancet. 2012;379(9834):2364-72.

ORIGINAL ARTICLE

Attitudes and experiences during training and professional expectations in generation-y surgical residents

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SUMMARY

BACKGROUND: *Residency programs, especially in surgery, have been undergoing constant changes. The profile of residents in surgical fields is changing too since residents are now part of the Generation Y (Millenials). This change in profile mandates a re-evaluation to adapt surgical residency programs. Six years ago, we carried out a study evaluating attitudes and experiences during training, and the professional expectations of residents. This study aims to survey surgical residents to evaluate current attitudes, experiences, and expectations.*

METHODS: *We surveyed 50 residents to determine professional satisfaction, residency-program satisfaction, future expectations, financial expectations, and correct attitude towards patients.*

RESULTS: *Our results show that half of the residents are satisfied with the residency program. However, dissatisfaction reaches 40% on surgical volume and 80% on mentorship; 62% of the residents are not confident to perform operations after the residency, the majority believes a specialization is necessary; most residents believe financial compensation will decrease with time, but concerns with reimbursement are low; and most residents are worried about injuring patients, but only two thirds are satisfied working with patients.*

CONCLUSIONS: *Current residents present lower job satisfaction and more criticism of teaching techniques. These changes compared to previous results match the profile of Generation Y, who is more iconoclastic when compared to previous generations.*

KEYWORDS: *Internship and Residency. General Surgery/Education. Intergenerational Relationships.*

INTRODUCTION:

Residency programs, especially in surgery, have been undergoing constant changes. These changes must consider attitudes and experiences during training, and the professional expectations of residents. Six years ago, we carried out a study evaluating these domains[1]; however, the profile of residents in surgical fields has changed. According to McLeod, in her inaugural address to the Society for Surgery of the Alimentary Tract (SSAT)[2], we are living in times when the residents are part of Generation Y, also known as *Millennials*, which includes people born between 1982 and 2002. Studying the portrait of this generation helps to understand the attitudes and needs of current residents and, therefore, adapt surgical residency programs to their profile.

This study aims to assess attitudes and experiences during training and the professional expectations of surgical residents.

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METHODS

Surgical residents of a quaternary teaching hospital, from different years of training and specialties, were surveyed with a dedicated questionnaire. Participation was voluntary, and an independent investigator presented the questionnaires in order to avoid authority biases. Questionnaires were not identified and the responses were kept anonymous.

Population

Fifty surgical residents (64% males, mean age 26 years) were surveyed. 80% were junior, and 20% were senior residents.

Questionnaire

We employed an adapted questionnaire by Yeo et al.[3], previously translated and adapted[1].

Individuals were asked to agree, be neutral or disagree with forty-two affirmations, taking into account human, technical and professional domains, randomly oriented. Questions were grouped into professional satisfaction, residency program satisfaction, future expectations, financial expectations and correct attitude towards patients.

The questionnaire is presented in Table 1.

Ethics

The project was approved by the local ethics committee and all participants signed a consent form before entering the study. The questionnaire was reused with the authors' permission.

There are no conflicts of interest. The authors are responsible for the study, no professional or ghostwriter was hired.

RESULTS

All individuals agreed to participate.

The answers are presented in Table 1. The main results for the 5 domains are shown in figures 1 to 5.

DISCUSSION

Generation Y comprises individuals born between 1982 and 2002. It is the largest demographic group since Baby Boomers; thus their influence will have an impact on social norms, work, pleasure, and politics. It is estimated that they will account for 50% of the workforce in 2020. This population grew up used to having technology as part of their daily lives, and many have been raised with extensive parental involvement. They want to make a difference in the world and expect to have meaningful, fulfilling work. They may be impatient and think that they have much to teach older generations, especially when it comes to living a proper life, respecting their health and psychological limits and living a life that makes them happy, regardless of financial success[2,4-7].

Generation Y residents follow the same characteristics described above. One would expect a low professional satisfaction in these residents since surgery demands great dedication[8] and work may be a secondary priority, with life quality and the pursuit of happiness being primary. Indeed, life-work balance is currently an important factor in the decision against surgery as a career after medical school[9]. Our study shows that almost all residents enjoy operating, but less than half are happy at work. Future expectations follow the same ideals. The prospect of surgical specialty is not a big concern among our residents. Curiously, a newspaper article entitled "the generation that found success after being fired" told the stories of young graduates from prominent Universities that were fired or quit their jobs and radically changed their lifestyle to live a simple life[10]. Financial expectations are not a motivator to Millenials as well. In our survey, only a third of residents were influenced by good financial compensation when choosing their careers as surgeons and worry about making enough money when finishing residency. Interestingly, not only for this domain but especially on it, the number of answers as neutral is significant.

The education of Generation Y is under constant debate[11]. Personal beliefs and opinions, conduct in front of patients, dress codes, attitude towards pets, charity, etc. may be more important to create or destroy a mentor-mentee relation than medical knowledge and educational skills. Millenial residents must be taught that hierarchy is important in surgery. Our study shows some concerning issues. Although half of the residents are satisfied with the residency program, more than half have considered leaving it. Interestingly, they are different individuals. 54% of the residents satisfied with the program and 67% of the unsatisfied ones considered leaving the program. Lifestyle issues have been imputed as the main cause for the high number of attrition in surgical programs[12]. Our residents, however, did not answer that working hours or stress causes a strain on family life. They were very critical about the program but confident on their performance, as shown by other studies, including Y'ers as well[13].

TABLE 1. PROFESSIONAL SATISFACTION, RESIDENCY PROGRAM SATISFACTION, FUTURE EXPECTATIONS, FINANCIAL EXPECTATIONS AND CORRECT ATTITUDE TOWARDS PATIENTS FOR SURGICAL RESIDENTS (N=50)

QUESTIONS		AGREE	NEUTRAL	DISAGREE
1.	Overall, I am very satisfied with my program	26 (52%)	12 (24%)	12 (24%)
2.	As a surgical resident, my opinions are important	21 (42%)	16 (32%)	13 (26%)
3.	The program has support structures in place that provide me with someone to turn to when I am struggling	17 (34%)	22 (44%)	11 (22%)
4.	To be a good surgeon you must give up your sensitivity	0	2 (4%)	48 (96%)
5.	I feel I can turn to members of the faculty when I have difficulties in the program	24 (48%)	15 (30%)	11 (22%)
6.	I feel I can turn to members of the faculty when I am struggling with how to treat a patient	33 (66%)	11 (22%)	6 (12%)
7.	I look forward to coming to work every day	27 (54%)	18 (36%)	5 (10%)
8.	I am satisfied with the teaching in my program	11 (22%)	24 (48%)	15 (30%)
9.	I am satisfied with the operative experience in my program	11 (22%)	20 (40%)	19 (38%)
10.	I have considered leaving my program	28 (56%)	4 (8%)	18 (36%)
11.	I do not feel respected by my attendings	4 (8%)	21 (42%)	25 (50%)
12.	I am happy when I am at work	21 (42%)	23 (46%)	6 (12%)
13.	Surgery training is too long	18 (36%)	14 (28%)	18 (36%)
14.	I feel uncomfortable with some of the ethical decisions I see some attendings make	25 (51%)	15 (31%)	9 (18%)
15.	I am given so much to do that I am afraid I will hurt someone	5 (10%)	17 (34%)	28 (56%)
16.	I feel that my operating skill level is appropriate	27 (54%)	14 (28%)	9 (18%)
17.	I worry that I will not feel confident enough to performs procedures by myself before I finish training	31 (62%)	8 (16%)	11 (22%)
18.	I am not happy with the personality that I must have to become a good surgeon	6 (12%)	12 (24%)	32 (64%)
19.	The hours I am working are causing a strain on my family life	13 (26%)	13 (26%)	24 (48%)
20.	The stress of my work is causing a strain on my family life	11 (22%)	18 (36%)	21 (42%)
21.	My attendings will think worse of me if I ask for help when I do not know how to do a procedure	5 (10%)	8 (16%)	37 (74%)
22.	My attendings will think worse of me if I ask for help when I do not know how to manage a patient	10 (20%)	7 (14%)	33 (66%)
23.	I really care about my patients	43 (86%)	3 (6%)	4 (8%)
24.	I worry about performing poorly in front of my senior residents	36 (72%)	7 (14%)	7 (14%)
25.	I worry about performing poorly in front of my attendings	38 (76%)	7 (14%)	5 (10%)
26.	The personal cost of surgical training is not worth for me	3 (6%)	6 (12%)	41 (82%)
27.	I feel that I fit in well at my training program	43 (86%)	7 (14%)	
28.	I get a tremendous amount of satisfaction working with patients	33 (66%)	15 (30%)	2 (4%)
29.	I get along with my fellow residents	29 (58%)	18 (36%)	3 (6%)
30.	I am committed to completing my general surgery residency training	43 (86%)	6 (12%)	1 (2%)
31.	I enjoy operating	48 (96%)	2 (4%)	
32.	I worry about hurting patients	42 (84%)	7 (14%)	1 (2%)
33.	My operative experience so far has helped me to develop my skills well	35 (70%)	11 (22%)	4 (8%)
34.	If I have a problem I feel I can count on other residents to help me out	45 (90%)	5 (10%)	
35.	I worry that the field of general surgery is going to become obsolete	7 (14%)	5 (10%)	38 (76%)
36.	I worry that other medical professionals will take over some of the procedures that I do	13 (26%)	11 (22%)	26 (52%)
37.	The modern general surgeons must become specialty trained in order to be successful	39 (80%)	6 (12%)	4 (8%)
38.	Surgeons do not make as much money now as they used to	25 (51%)	21 (43%)	3 (6%)
39.	I worry about the high cost of malpractice insurance	29 (59%)	12 (24%)	8 (16%)
40.	One of the factors that influenced my decision to be a surgeon was the expectation of good financial compensation	17 (35%)	15 (31%)	17 (35%)
41.	Each year my expectations for the amount of money I am going to make when I finish training seem to go down	14 (29%)	16 (33%)	19 (39%)
42.	I worry about making enough money as a surgeon	28 (57%)	15 (31%)	6 (12%)

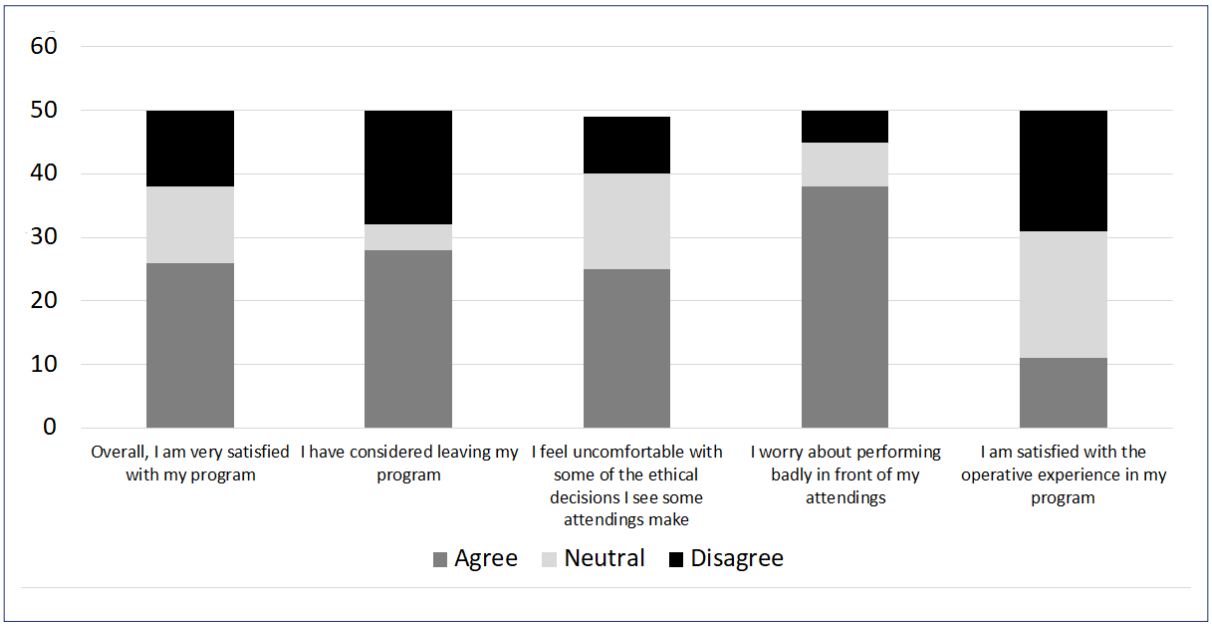

FIGURE 1. MAIN RESULTS FOR "RESIDENCY PROGRAM SATISFACTION" DOMAIN

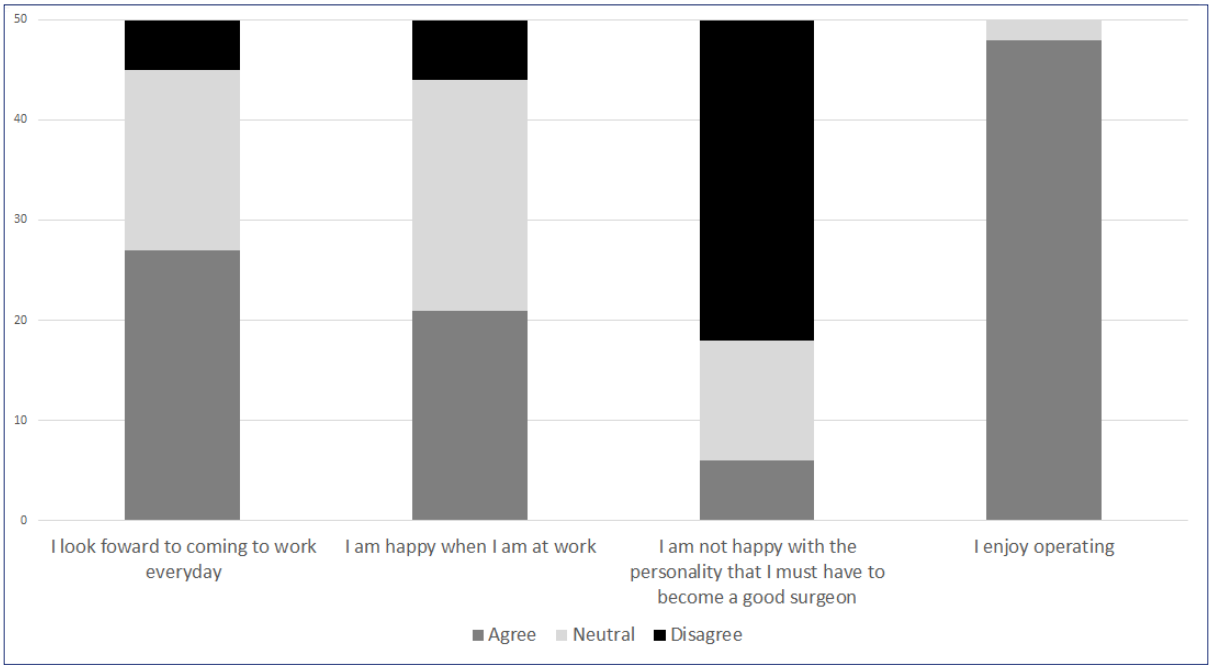

FIGURE 2. MAIN RESULTS FOR "PROFESSIONAL SATISFACTION" DOMAIN

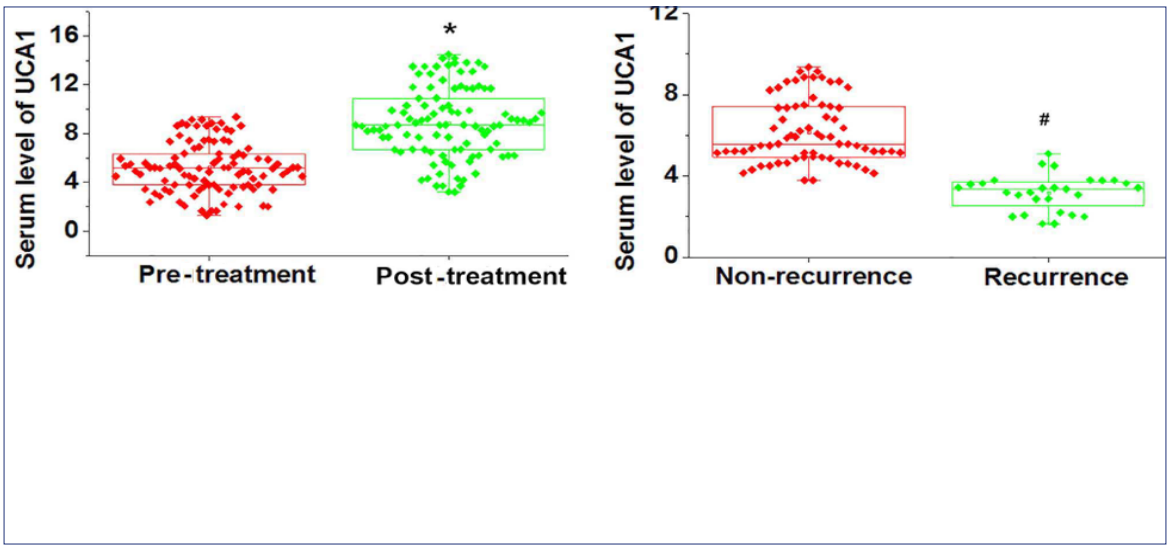

FIGURE 3. MAIN RESULTS FOR "ATTITUDE TOWARDS PATIENTS" DOMAIN

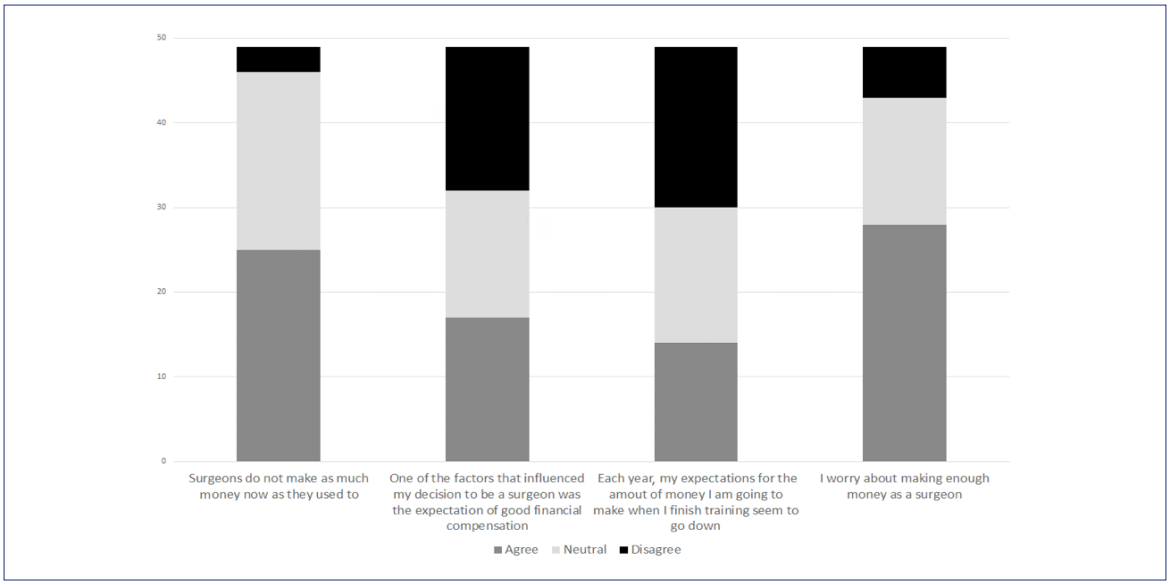

FIGURE 4. MAIN RESULTS FOR "FINANCIAL EXPECTATIONS" DOMAIN

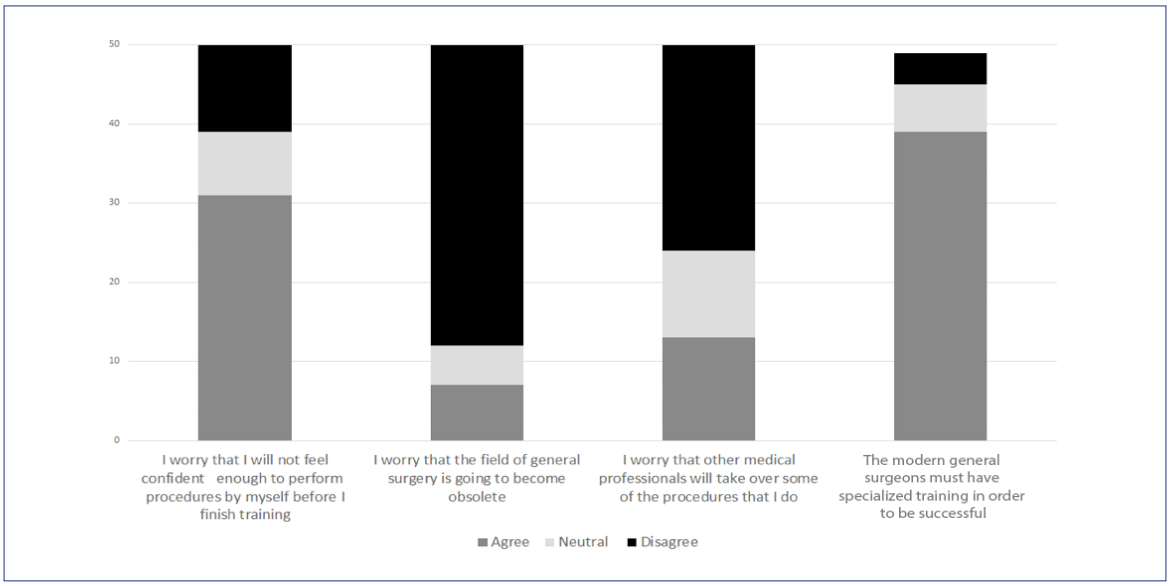

FIGURE 5. MAIN RESULTS FOR "FUTURE EXPECTATIONS" DOMAIN

We surveyed our residents 6 years ago[1]. Although most of the residents at that time were Millenials, the world is changing fast. We did notice a change in profile when repeating the same questionnaire. In 2011, residents had great satisfaction with a surgical career but high financial concerns and conflicting opinions about the future of this specialty. Only 20% of the respondents had thought about leaving the program. A USA national survey at that time[2] showed similar conclusions. Current residents present lower job satisfaction and more criticism of teaching techniques.

Different ideas on how to deal with generation Y residents have been proposed. Among them: (a) provide challenging, constant and variable tasks as they become bored quickly[4]; (b) technology as a teaching and working tool is very attractive to these individuals[4,6,7]; (c) millennials are independent of hierarchy but at the same time work environment, support and leadership must mimic the helicopter parenting style they are used too[4]; (d) they like to have their opinion heard with instantaneous feedback[5,6]; (e) teamwork should be encouraged as they are part of the "we" generation[4,7]; and (f) millennials do not fight for money, fame, fortune or status; do not offer these as advantages[5]. In summary, Y'ers must be understood.

Our study has some limitations. First, the results have a regional influence and may not be extrapolated elsewhere, although Generation Y is a worldwide phenomenon. Second, the small number of participants, that was defined by the current number of residents in the program, did not allow a subanalysis of results according to the level of training, different specialties, and gender. As a strength, the manuscript was written by 2 medical students and 1 fellow part of the generation Y, 1 senior surgeon part of the X generation, and 1 senior surgeon part of the Baby Boomer generation to include multigeneration opinions and critiques.

In conclusion, current residents present lower job satisfaction, more criticism of teaching techniques and greater concerns about preparedness for the future. These changes compared to previous results match the profile of Generation Y, who is more iconoclastic when compared to previous generations. Program directors, senior colleagues and all other individuals dealing with new surgical residents must be aware of these new characteristics.

RESUMO

INTRODUÇÃO: *Os programas de residência, especialmente em cirurgia, estão em constante mudança. O perfil dos residentes nos campos cirúrgicos também vem mudando, dado que atuais residentes fazem parte da Geração Y (Millennials). Essa mudança de perfil demanda uma reavaliação para adaptar os programas de residência. Este estudo tem como objetivo entrevistar os residentes de áreas cirúrgicas para avaliar suas atuais atitudes, experiência e expectativas.*

MÉTODOS: *Entrevistamos 50 residentes para determinar satisfação profissional, expectativas em relação ao futuro, expectativas financeiras e atitude correta em relação aos pacientes.*

RESULTADOS: *A insatisfação com o volume cirúrgico chega a 40% e a 80% com a preceptoria; 62% dos residentes não se sentem confiantes para realizar procedimentos sozinhos após o fim do programa e a maioria acredita que uma especialização cirúrgica é necessária; a maioria dos residentes acredita que os ganhos monetários diminuirão com o tempo.*

CONCLUSÃO: *Os atuais residentes apresentam menor satisfação com o trabalho quando comparados com os antigos, e são mais críticos quanto ao ambiente de ensino. Essas mudanças seguem as premissas da Geração Y, cujos participantes são mais iconoclastas quando comparados a gerações passadas.*

PALAVRAS-CHAVE: *Geração Y. Programas de residência. Educação em cirurgia.*

REFERENCES

1. Herbella FA, Fuziy RA, Takassi GF, Dubecz A, Del Grande JC. Evaluation of training and professional expectations of surgery residents. Rev Col Bras Cir. 2011 Jul-Aug;38(4):280-4.
2. McLeod RS. SSAT presidential address 2014: here comes Generation Y! J Gastrointest Surg. 2015 Jan;19(1):1-5. doi: 10.1007/s11605-014-2703-1.
3. Yeo H, Viola K, Berg D, Lin Z, Nunez-Smith M, Cammann C, Bell RH Jr, Sosa JA, Krumholz HM, Curry LA. Attitudes, training experiences, and professional expectations of US general surgery residents: a national survey. JAMA. 2009 Sep 23;302(12):1301-8. doi: 10.1001/jama.2009.1386.
4. Schlitzkus LL, Schenarts KD, Schenarts PJ. Is your residency program ready for Generation Y? J Surg Educ. 2010 Mar-Apr;67(2):108-11. doi: 10.1016/j.jsurg.2010.03.004.
5. Money SR, O'Donnell ME, Gray RJ. In the time of significant generational diversity - surgical leadership must step up! Surgeon. 2014 Feb;12(1):3-6. doi: 10.1016/j.surge.2013.09.007.
6. Desy JR, Reed DA, Wolanskyj AP. Milestones and Millennials: A Perfect Pairing-Competency-Based Medical Education and the Learning Preferences of Generation Y. Mayo Clin Proc. 2017 Feb;92(2):243-250. doi: 10.1016/j.mayocp.2016.10.026
7. Venuta F. ESTS Presidential Address. Education motivation... inspiration of Generation Y. The evolution of our species. Eur J Cardiothorac Surg. 2014 Nov;46(5):761-6. doi: 10.1093/ejcts/ezu295. Epub 2014 Aug 1.
8. Zubair MH, Hussain LR, Williams KN, Grannan KJ. Work-Related Quality of Life of US General Surgery Residents: Is It Really so Bad? J Surg Educ.

2017 Nov - Dec;74(6):e138-e146. doi: 10.1016/j.jsurg.2017.09.018. Epub 2017 Oct 4.

9. Kleinert R, Fuchs C, Romotzky V, Knepper L, Wasilewski ML, Schröder W, Bruns C, Woopen C, Leers J. Generation Y and surgical residency - Passing the baton or the end of the world as we know it? Results from a survey among medical students in Germany. PLoS One. 2017 Nov 27;12(11):e0188114. doi: 10.1371/journal.pone.0188114. eCollection 2017.

10. http://emais.estadao.com.br/blogs/ruth-manus/a- geracao-que- encontrou-o-sucesso-no- pedido-de- demissao/ Accessed 06/06/2017

11. Evans KH, Ozdalga E, Ahuja N. The Medical Education of Generation Y. Acad Psychiatry. 2016 Apr;40(2):382-5. doi: 10.1007/s40596-015-0399-5. Epub 2015 Aug 18.

12. Khoushhal Z, Hussain MA, Greco E, Mamdani M, Verma S, Rotstein O, Tricco AC, Al-Omran M. Prevalence and Causes of Attrition Among Surgical Residents: A Systematic Review and Meta-analysis. JAMA Surg. 2017 Mar 1;152(3):265-272. doi: 10.1001/jamasurg.2016.4086.

13. Mundschenk MB, Odom EB, Ghosh TD, Kleiber GM, Yee A, Patel KB, Mackinnon SE, Tenenbaum MM, Buck DW 2nd. Are Residents Prepared for Surgical Cases? Implications in Patient Safety and Education. J Surg Educ. 2017 Jul 18. pii: S1931-7204(16)30390-7. doi: 10.1016/j.jsurg.2017.07.001.

ORIGINAL ARTICLE

An anterior neurovascular interval approach to coronal shear fractures of the distal humerus: a prospective clinical study with short- to mid-term follow-up

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SUMMARY

OBJECT: *To explore the treatment effect of the anterior medial neurovascular interval approach to coronal shear fractures of the distal humerus.*

METHODS: *This prospective study included two female patients who were 30-64 years old, with a mean age of 47 years. Fractures were caused by falling from a bicycle. The time between the injury and operation was 1-2 days, with a mean time interval of 1.5 days. Two patients with coronal shear fracture of the distal humerus were treated with open reduction and internal fixation using anterior neurovascular interval approach.*

RESULTS: *There were no intraoperative and postoperative neurological and vascular complications or infections, and the fracture was united. At 12 months after the surgery, the patient returned to work without pain, and with a normal range of motion for elbow and forearm rotation. The X-rays revealed excellent fracture union, no signs of heterotopic ossification, and no traumatic arthritis. According to Mayo's evaluation standards for elbow function, a score of 100 is excellent.*

CONCLUSIONS: *The application of the anterior neurovascular interval approach of the elbow in the treatment of shear fracture of the articular surface of the distal humerus, particularly the trochlea of the humerus, can reduce the stripping of the soft tissue.*

KEYWORDS: *anatomy, anterior approach, elbow joint, neurovascular interval, coronal, shear, fracture, distal, humerus, reduction, and internal fixation.*

INTRODUCTION

Coronal shear fractures of the distal humerus are rare and complex and have become a challenge for orthopedists[1-3]. Insufficient soft tissue attachments at the fracture site often contribute to displacement[2], which determines the need for surgery. Determining a good surgical approach is crucial to ensure the success of the surgery and reduce complications such as joint stiffness, joint pain, osteoarthritis, heterotopic ossification, instability of the elbow and limited elbow function activity. Medial and lateral approach-

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es require excessive soft tissue dissection, making exposure and fixation more difficult; while the posterior approach is often combined with olecranon osteotomy. Thus, these three approaches have limitations[4,5]. The application of the anterior approach has also been reported[6] to have an advantage of performing fewer dissections, which is more applicable for capitellum fractures.

According to anatomical findings of the anterior neurovascular interval of the elbow, we applied a different anterior approach to treat shear fractures of the distal humeral articular surface, especially fractures at the trochlear region. This approach has various advantages, including a simple anatomical layer, it is easy to master, the tissues are moderately dissected during surgery, fewer side injuries, easy to expose, provides a clear operative field, reduces direct visualization and strong operability. These are reported as follows.

MATERIALS AND METHODS

General Information

This prospective study included two females aged between 30-64 years, with a mean age of 47 years. Their fractures were caused by a bicycle fall. The time between the injury and the operation was 1-2 days, with a mean interval of 1.5 days. Both patients with coronal shear fracture of the distal humerus were treated with open reduction and internal fixation by the anterior neurovascular interval approach. Reduction and fixation of the coronoid fracture were performed through the same approach on one patient with an ulna coronoid process fracture, while in another patient with a fracture of the lateral epicondyle of the humerus the fixation of the fracture was performed through another incision. Plaster splints were used for fixation in both patients, and function exercises were performed after the removal of the external fixation at 2-3 weeks postoperative.

Typical case: A 64-year-old female fell off the bicycle and got injured when her right hand and forearm hit the ground. Her right elbow was swollen and painful with limited mobility. She presented to the Emergency Department of our hospital 30 minutes after the injury. Upon examination, the right elbow was swollen with local ecchymosis and tenderness, as well as an elbow flexion of 30°. Elbow flexion/extension and forearm rotation were limited. No signs of neurovascular injury were detected, radial artery pulse was good, and the right wrist and fingers moved easily. After checking the vital signs of the patient admitted to the hospital, X-ray, CT scan, and three-dimensional imaging examinations revealed a coronal shear fracture of the distal humerus, which was comminuted and affected the lateral epicondyle of the humerus; the fracture surface was sagittal. Various routine examinations were performed before surgery for perioperative preparation.

The patient received an open reduction and internal fixation (ORIF) 48 hours after the injury. The patient was placed in supine position, and the operative elbow was extended on a see-through operating table, without a tourniquet. Based on the following surgical technique, the fracture of the distal humeral articular surface was exposed for anatomical reduction of fractures. At the trochlear region of the articular surface, three 2.0-mm bioabsorbable screws (provided by Inion, Finland) were fixed from front to back, perpendicular to the fracture surface. The lateral epicondyle connected to the capitellum was a whole bone. Another 2-cm small incision was made on the lateral side, and two 1.2-mm Kirschner wires were used for fracture fixation vertical to the fracture surface of the epicondyle. Due to the anatomical reduction of the fracture and reliable fixation, no elbow instability was detected after the surgery. Indomethacin was administered at the early stage after the injury to prevent heterotopic ossification. A functional plaster slab was provided for immobilization after surgery, as well as routine anti-inflammatory and anti-swelling treatment.

Surgical techniques

Under brachial plexus block anesthesia, the patient was placed in the supine position; and the operative elbow was extended on the operating table. A “S” type incision started 2cm proximal to the elbow flexion crease, curved cross the elbow crease, and extended distally along the radial border of the forearm at approximately 2cm distal to the flexion crease. During the skin incision, the main superficial veins were identified and protected to ensure non-ligation and ready extraction. Careful blunt subcutaneous dissection was performed to expose the medial antebrachial cutaneous nerve, which should be protected and retracted medially. The deep fascia, biceps, and bicipital aponeurosis were unroofed, as well as the distal brachioradialis and pronator teres. The trans-

verse split of the bicipital aponeurosis provided excellent exposure of the proximal brachial artery and vein, as well as the median nerve. The latter extended distally from the space between the humeral head of the pronator teres muscle and ulnar head. The brachioradialis and pronator teres muscle were separated for distal exposure. The clearance between the brachial artery and median nerve allowed insertion due to a loose structure and absence of neurovascular branches. The brachial artery, biceps tendon, and brachioradialis were retracted laterally, while the median nerve and pronator teres were retracted medially, providing the best exposure of the brachial muscle. The longitudinal muscle split and retraction accomplished good visualization of the anterior capsule and displaced fragment of the coronal fracture of the distal humerus. The anatomical reduction of the fracture was performed under direct visualization, followed by front-to-back fixation with headless compression screws or absorbable screws (Figure 1).

Postoperative management and follow-up

The suture was removed two weeks after surgery, followed by the removal of the plaster slab two weeks later. Functional exercises were initiated with a small ROM, combining active and passive activities. After removing the Kirschner wires four weeks later, ROM was gradually increased, and regular functional training was appointed. ROM was increased moderately every 3-5 days. After eight weeks, both elbow flexion/extension and forearm pronation/supination were restored to normal, followed by fracture healing three months after the surgery. The patient was followed-up regularly at 1, 3, 6 and 12 months (Figure 2).

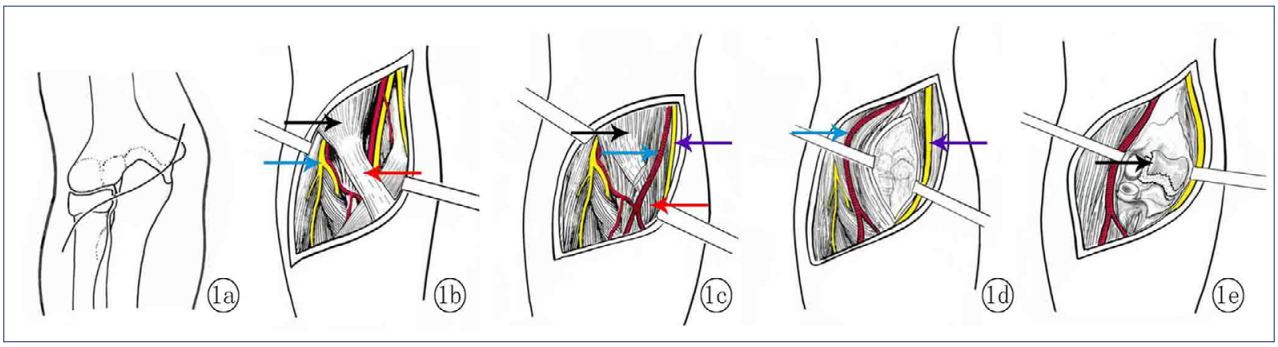

FIGURE 1. Illustration of the anterior neurovascular interval approach of the elbow. a. "S" shaped incision in the front of elbow joint; b. Exposure of biceps (black arrow) and bicipital aponeurosis (red arrow), lateral cutaneous nerve of the forearm (blue arrow) after subcutaneous dissection; c. Horizontal incision of bicipital aponeurosis, showing biceps (black arrow), part of pronator teres (red arrow), brachial artery (blue arrow), and median nerve (purple arrow); d. Entering through the neurovascular interval, biceps and brachial artery (blue arrow) to the lateral side, and pronator teres, median nerve (purple arrow) to the medial side, exposure and longitudinally splitting of brachial muscle and distraction; e. Exposure of trochlea fracture fragment (black arrow)

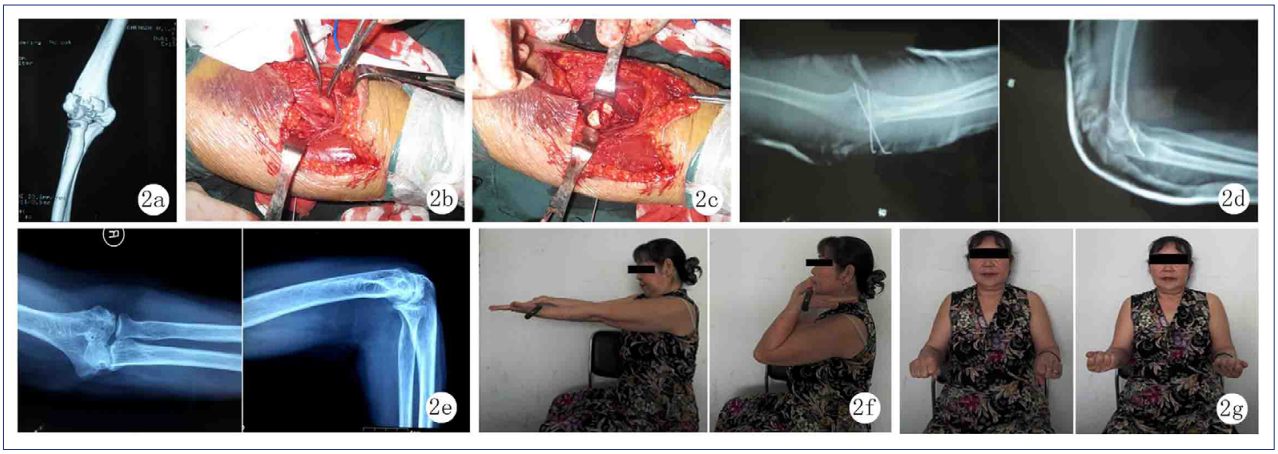

FIGURE 2. A 64-year-old female patient with coronal shear fractures of the right distal humerus a. Preoperative three-dimensional CT image; b. Exposure of brachial artery and median nerve in operation; c. Exposure of trochlea fracture fragment in operation; d. Anteroposterior and lateral X-rays 2 days after the operation; e. Anteroposterior and lateral X-rays 1 year after the operation; f. Function of the flexion and extension of the elbow 1 year after the operation; g. Function of the pronation and supination of the arm 1year after the operation

RESULTS

The patient revealed no intraoperative and postoperative neurovascular complications, and no deep infection, with a Class-A healing rate. Elbow function was restored to normal during the 12-month follow-up. Furthermore, the patient was able to resume her job and reported no complications such as pain, heterotopic ossification, traumatic arthritis, and poor joint mobility. The arc of the elbow flexion and extension was 135°, and the forearm pronation/supination was 155°. Based on the evaluation criteria of elbow performance by Mayo, the result was categorized as excellent.

DISCUSSION

Shear fractures of the distal humeral articular surface are a relatively rare intra-articular fracture that often leads to displacement. Conservative treatment provides poor outcomes and often contributes to complications such as chronic pain, instability, and mechanical symptoms of joints[1-3]. Its principles of treatment include the anatomical reduction of the fracture, restoration of the smooth articular surface, strong and reliable internal fixation, and early functional exercises, thereby reducing the non-osteoporosis complications of the elbow including arthritis, pain, joint stiffness and joint instability[2]. Basically, all patients require surgery, including ORIF. Arthroscopic-assisted surgery has also been reported. In terms of severely comminuted fractures that could not be fixed, total elbow replacement surgery is viable[7]. ORIF surgery can produce superior therapeutic effects in most patients with coronal shear fracture of the distal humerus. This effect derives from the anatomic reduction of fractures and stable fixation, as well as early and active function exercises on this basis[1].Coronal shear fractures of the distal humerus represent significant articular injuries and are usually more complex than those suggested by radiographic imaging. The fracture pattern and extent of articular involvement dictate the method of surgical exposure and the internal fixation technique used for treatment[8].The complexity of elbow injuries and pathology requires a surgeon to be comfortable with a range of approaches to the elbow joint. In trauma, the selection of the most helpful approach is determined by a combination of experience and familiarity with the anatomy interpreted to the fracture pattern. This enables the optimal exposure of relevant parts of the elbow joint[9]. Good surgical approaches are necessary for a successful operation. Multiple surgical approaches have been described for coronoid repair[10]. There are several treatment options similar to those that concern shear fracture of the distal humeral articular surface, which include the extensile lateral approach, the posterior approach with olecranon osteotomy, and the anterolateral approach. The extensile lateral approach has the disadvantages of excessive soft tissue dissection around the joint and great damage[11]. Furthermore, we believe that this approach is preferable for humeral head fractures. However, it is difficult to expose the ulnar side of the distal humerus. The posterior approach is often combined with olecranon osteotomy, therefore bringing about the extension of the fracture, the risk of nonunion olecranon, and complications caused by the internal fixator[12]. Therefore, we also assume that this approach is applicable to free-bone fractures of the anterior elbow joint. Furthermore, it has also been reported in the literature that for the anterolateral approach, an incision is made at the space between the lateral biceps tendon and brachioradialis, where the radial nerve passes and the nerve branches are given off. This would easily damage these branches. The function of nerve branches is difficult to restore after injury. In addition, the biceps tendon ends at the radial tuberosity. Hence, it is more difficult to expose the ulnar side of the distal humerus by medial retraction. Therefore, this approach is also more applicable for radial fractures of the distal humerus, such as fractures of the humeral head. Some scholars have applied two surgical approaches, called the combination of the lateral and anterior approaches, which can reduce soft tissue dissection. The anterior approach is similar to the approach for the fixation of ulna coronoid fractures. However, through this approach, the incision is made at the space between the biceps nerve and vascular bundles[13]. The ideal surgical approach for elbow fracture should be the one that requires as little surgical dissection as possible, does not affect the visualization of the fracture, and has minimal risk of heterotopic ossification and elbow stiffness[14]. In this study, despite the similar skin incision induced through the anterior approach and other anterior approaches that have been reported, the specific tissue space selected in this approach is different from other anterior approaches. As for the superficial layer of the deep fascia, other reports[15,16] revealed that the brachial artery and the median

nerve were retracted to one side; while in our study, we made an incision space between the brachial artery and median nerve. The separation was available due to the loose and wide space. The brachial artery, biceps tendon, and brachioradialis were retracted laterally, while the median nerve and pronator teres were retracted medially, providing the best exposure of the brachial muscle. The longitudinal muscle split and retraction accomplished a good visualization of the anterior capsule and anterior humeral fracture. Through the anatomical study of cadavers, we found that the brachial artery and vein were accompanied by the median nerve for a long distance, and there were no branches traveled across them. Furthermore, the brachial artery had no medial branches but lateral ones, which nourished muscles such as the brachioradialis and supinator muscle. In contrast, the median nerve has no lateral branches except for medial ones, which control the pronation teres, flexor carpi radialis, and musculus flexor digitorum sublimis. The gap between these two was very loose to be separated and retracted easily. However, anatomical studies also have variations. Based on local anatomical characteristics, we propose that an incision is made at the space between the brachial artery and median nerve. Due to elbow flexion/extension functions, nerves, and blood vessels, this site has great ROM for retraction to both sides, causing no neurovascular complications. This approach avoids medial retraction of the neurovascular bundle, therefore preventing the branches of the brachial artery from being cut off. Some scholars[17] have questioned the reports provided by Han SH et al. Some arterial branches constitute a "vascular tree"; therefore, the distance of the brachial artery retracted medially was constricted. What is the best option to manage arterial branches? An incision made at the space between the brachial artery and the median nerve was the optimal approach. This reduced injury to vascular branches and intraoperative bleeding, and saved operation time. Additionally, lateral retraction to expose the blood vessels and nerves would damage the medial branches of the median nerve, resulting in neurological symptoms, which have been reported by some scholars[18]. There are several ways to fix coronal shear fractures of the distal humerus, and the most commonly used ones include headless compression screws. Absorbable screws were applied to the patient reported in our study. We presume that both can be used for articular fractures producing good therapeutic effects. The biomechanical experiments conducted by Elkowitz et al.[19] revealed that front-to-back fixation with headless compression screws was superior to back-to-front fixation with cancellous bone screws. Front-to-back vertical fixation is the most stable one in coronal shear fractures, and this approach is convenient for the front-to-back placement of the fixator, especially for intra-articular fractures, free-anterior fracture blocks, and fractures with coronal planes. Moreover, with this approach, less soft tissue dissection contributes to anatomical reduction and stable fixation, reducing complications such as arthritis, nonunion, pain, stiffness, and heterotopic ossification. The limitation of this study is its small sample size. Therefore, studies with large sample sizes are needed to accumulate experience.

CONCLUSIONS

The anterior neurovascular interval approach of the elbow can be used for the treatment of shear fractures of the distal humeral articular surface, especially the trochlea. This approach reduces the dissection of soft tissue, ensures the most significant exposure, reduces fracture and fixation under direct visualization, and reduces complications. It can be applied either alone or in combination with other surgical approaches, producing optimal therapeutic effects.

RESUMO

OBJETIVO: *Explorar o efeito do tratamento com uma abordagem anterior do intervalo neurovascular médio para fraturas de cisalhamento coronal da porção distal do úmero.*

METODOLOGIA: *Este estudo prospectivo incluiu duas pacientes do sexo feminino de 30-64 anos de idade, com idade média de 47 anos. As fraturas foram causadas por quedas de bicicleta. O tempo entre a lesão e a operação foi de 1-2 dias, com um intervalo de tempo médio de 1,5 dias. Duas pacientes com cisalhamento coronal da porção distal do úmero foram tratadas com redução aberta e fixação interna utilizando a abordagem anterior do intervalo neurovascular.*

RESULTADOS: *Não houve complicações neurológicas e vasculares intra e pós-operatórias, nem complicações ou infecções, e a fratura foi unida. Após 12 meses da cirurgia, as pacientes retornaram ao trabalho sem dor e com uma amplitude normal de movimento de rotação do antebraço e cotovelo. Os raios-X revelaram excelente união das fraturas, sem sinais de ossificação heterotópica e sem artrite traumática. De acordo com as diretrizes da clínica Mayo para avaliação da função do cotovelo, uma pontuação de 100 é considerada excelente.*

CONCLUSÃO: *A aplicação da abordagem anterior do intervalo neurovascular do cotovelo no tratamento de uma fratura de cisalhamento da superfície articular da porção distal do úmero, especificamente da tróclea do úmero, pode reduzir o desgaste do tecido mole.*

PALAVRAS-CHAVE: *Anatomia; abordagem anterior; articulação do cotovelo; intervalo neurovascular; processo coronóide da ulna; redução; fixação interna.*

REFERENCES

1. Nauth A, McKee MD, Ristevski B, Hall J, Schemitsch EH. Distal humeral fractures in adults. J Bone Joint Surg Am 2011; 93: 686-700.
2. Lee JJ, Lawton JN. Coronal shear fractures of the distal humerus. J Hand Surg Am 2012; 37: 2412-2417.
3. Ruchelsman DE, Tejwani NC, Kwon YW, Egol KA. Coronal plane partial articular fractures of the distal humerus: current concepts in management. J Am Acad Orthop Surg 2008; 16: 716-728.
4. Watts AC, Morris A, Robinson CM. Fractures of the distal humeral articular surface. J Bone Joint Surg Br 2007; 89: 510-515.
5. Guitton TG, Doornberg JN, Raaymakers EL, Ring D, Kloen P. Fractures of the capitellum and trochlea. J Bone Joint Surg Am 2009; 91: 390-397.
6. Ruchelsman DE, Tejwani NC, Kwon YW, Egol KA. Open reduction and internal fixation of capitellar fractures with headless screws. Surgical technique. J Bone Joint Surg Am 2009; 91 Suppl 2 Pt 1: 38-49.
7. Kuriyama K, Kawanishi Y, Yamamoto K. Arthroscopic-assisted reduction and percutaneous fixation for coronal shear fractures of the distal humerus: report of two cases. J Hand Surg Am 2010; 35: 1506-1509.
8. Shahram S Y, Nathan L B, Miguel A C, Lee M R. Management of distal humeral coronal shear fractures. World J Clin Cases 2015; 3: 405-417.
9. Chris Peach, David Stanley. Surgical approaches to the elbow. Orthopaedics and Trauma 2012; 26: 297-302.
10. Shukla DR, Koehler SM, Guerra SM, Hausman MR. A novel approach for coronoid fractures. Tech Hand Surg 2014; 18: 189–193.
11. Bilsel K, Atalar AC, Erdil M, Elmadag M, Sen C, Demirhan M. Coronal plane fractures of the distal humerus involving the capitellum and trochlea treated with open reduction internal fixation. Arch Orthop Trauma Surg 2013; 133: 797-804.
12. Dubberley JH, Faber KJ, Macdermid JC, Patterson SD, King GJ.Outcome after open reduction and internal fixation of capitellar and trochlear fractures. J Bone Joint Surg Am 2006; 88: 46-54.
13. Reichel LM, Milam GS, Reitman CA. Anterior approach for operative fixation of coronoid fractures in complex elbow instability. Tech Hand Up Extrem Surg 2012; 16: 98-104.
14. Ring D, Jupiter JB. Surgical Exposure of Coronoid Fractures. Techn Shoulder Elbow Surg 2002; 3: 48–56.
15. O'Driscoll SW, Jupiter JB, Cohen MS, Ring D, McKee MD. Difficult elbow fractures: pearls and pitfalls. Instr Course Lect 2003; 52: 113-134.
16. Han SH, Yoon HK, Rhee SY, Lee JK. Anterior approach for fixation of isolated type III coronoid process fracture. Eur J Orthop Surg Traumatol 2013; 23: 395-405.
17. Zhu XZ, Wang X , Ma ZH. Comment on the anterior approach for coronoid process fracture. Eur J Orthop Surg Traumatol 2014; 24: 123–124.
18. Zuo YM ,Wang ZHQ , Wang YG, Zhao GZ, Ma SL. Treatment of ulna coronoid fractures. Chin J Orthop 2006; 26: 366-370.
19. Elkowitz SJ, Polatsch DB, Egol KA, Kummer FJ, Koval KJ. Capitellum fractures: a biomechanical evaluation of three fixation methods. J Orthop Trauma 2002; 16: 503-506.

ORIGINAL ARTICLE

Increased levels of plasma IL-1b and BDNF can predict resistant depression patients

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SUMMARY

BACKGROUND: *There is no strong evidence on the link between inflammatory profile and pattern of drug treatment response in depressive patients that could result in Coronary Artery Disease occurrence.*

OBJECTIVE: *This study aimed to compare the subclinical atherosclerosis markers, inflammatory profile, and BDNF production in Resistant Depression (RD) or Bipolar Affective Disorder (BAD) patients under conventional treatment.*

METHODS: *The population evaluated was comprised of 34 RD, 43 BAD, and 41 controls. Subclinical atherosclerosis markers were evaluated using ultrasonography, tomography, and exercise stress test. Plasma concentrations of TNFα, IL-1β, IL-6, and BDNF were measured using Luminex100™. The usCRP concentration was measured using turbidimetric immunoassay. IL1B, IL6, and TNFA expression were determined using TaqMan®. For the statistical analysis, the significance level was established at p<0.05.*

RESULTS: *Concerning subclinical atherosclerosis markers, only O2 consumption was reduced in the BAD group (p = 0.001). Although no differences were found in gene expression, BDNF and IL-1β plasma concentration was increased in the RD group (p = 0.002 and p = 0.005, respectively) even with an antidepressant treatment, which suggests that these drugs have no effect in IL-1β secretion and that the inflammasome may play a role in therapy response.*

CONCLUSION: *Taken together, both BDNF and IL-1β plasma concentrations could be used to the early identification of RD patients.*

KEYWORDS: *Bipolar Affective Disorder, Depression, Inflammation, Atherosclerosis, BDNF, IL-1β*

INTRODUCTION

Mood disorders have been recognized by the World Health Organization (WHO) as a major public health problem. Depression is a devastating disorder and one of the leading causes of disability worldwide, affecting up to 10% of the adult population [1,2]. Bipolar Affective Disorder (BAD) is a relatively rare affective disorder when compared with unipolar depression, with a lifetime prevalence estimated at 1% to 1.5% of the world population[3].

Clinical and experimental research supports

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a mutual relationship between inflammation and mood disorders. Patients with major depressive disorder and other neuropsychiatric diseases exhibit increased expression of pro-inflammatory cytokines, including interleukin-6 (IL-6), interleukin-1β (IL-1β), tumor necrosis factor-α (TNF-α), and their receptors, and elevated concentrations of C-reactive protein (CRP) [4]. IL-6, TNFα, and CRP are also implicated in BAD, and an increase in their plasma concentration can occur during both mania and depression episodes [5,6]. Also, studies have found reduced circulating BDNF protein concentration during acute phases of BAD, and this reduction is more significant than in a major depressive disorder. Conversely, BDNF expression is upregulated in response to prolonged treatment with anti-depressant drugs [7,8].

Although effective treatments are available, approximately one-third of all patients with mood disorder fail to respond to conventional antidepressant therapies [9]. The response to antidepressants or lithium may be affected due to inflammatory activity before treatment [10,11]. Moreover, the concentration of IL-6 and acute-phase proteins have been shown to be higher in non-responder patients compared to responder patients [10].

Depressive symptoms have received special attention by cardiologists because the high concentrations of inflammatory markers are risk factors to the development of atherosclerosis, an important Coronary Artery Disease (CAD). BDNF is also expressed in atherosclerotic coronary arteries suggesting its possible role in the pathogenesis of CAD. Thus, controlling inflammation in depression syndrome is an important strategy to prevent CAD in these patients [12-14].

This study aimed to compare subclinical atherosclerosis markers, inflammatory profile, and BDNF production between Resistant Depression (RD) and BAD patients under conventional treatment and euthymic controls, as a way of providing indicators of both the risk for cardiovascular diseases and drug treatment response in a Brazilian population.

MATERIALS AND METHODS

Subjects

Seventy-seven individuals were recruited from an Affective and Anxiety Disorders Program, including 34 Resistant Depression (RD) and 43 Bipolar Affective Disorder (BAD) patients diagnosed based on the Structured Clinical Interview for Diagnostic for DSM-IV Axis I Disorders (SCID-I), a standard research tool to identify bipolar disorder, using the validated questionnaire in Portuguese, applied to all patients by at least two experienced clinicians [15].

According to SCID-I, bipolar disorder is defined by several symptoms during the depressive episode such as anhedonia, sadness, hopelessness, insomnia or hypersomnia, loss of appetite, insecurity. That differs from the manic state, which is characterized by agitation, lack of sleep, hyperactivity, and impulsivity. The symptoms experienced in both episodes affect emotion and cognition and may compromise interpersonal relationship, work capacity and cause psychic distress. Resistant depression was defined as the failure of two or more standard antidepressant therapies at adequate dosing and duration to promote response and remission. The aim of antidepressant therapy is symptom remission or the reinstatement of euthymia—often defined as a score ≤7 on the total Hamilton Depression Rating Scale (HDRS) [16]. In our study, the resistant depression group had failed to respond to at least two previous drug treatments.

Forty-one subjects were controls (C). Control participants were euthymic with no previous mood disorder (based on SCID), chronic inflammatory condition or known coronary artery disease or any atherosclerotic vascular disease.

For all groups, individuals of any gender who were in the range of 21 to 70 years old were recruited. To reduce potential additional confounders, we also excluded subjects with other psychiatric diagnosis (depression with response or remission, schizophrenia, personality disorder), patients with known coronary artery disease, any atherosclerotic vascular disease, or any chronic inflammatory disorder or auto-immune disease such as Chrom's colitis, systemic lupus erythematosus, rheumatoid arthritis or other autoimmune or infectious diseases.

This study was approved by the Ethics Committee of Dante Pazzanese Institute of Cardiology, São Paulo and all subjects who agreed to participate in the study signed a written informed consent.

Subclinical atherosclerosis markers

Subclinical atherosclerosis evaluation was based on carotid intima-media thickness (CIMT) obtained with a high-resolution echo-colour-Doppler (Acusson, Model Aspen Advanced or similar) and 10 MHz linear transducer. The search for coronary atherosclerotic plaques was made with ultrafast comput-

ed tomography using the Imatron RC-150 (Imatron Corporation, San Francisco, California). Imaging procedures were characterized by axial heart with 3 mm slice thickness, at the end of diastole electrocardiography-triggered at 100 ms time interval during the inspiratory pause. Furthermore, the quantity or score of coronary calcium was estimated. Myocardial ischemia was diagnosed using the exercise test (ET) per Ellestad protocol [17]. The studied population was asymptomatic and had no previous history of coronary events.

RNA isolation and cDNA synthesis

The whole blood was collected into PAXgene® tubes (QIAGEN, Hilden, Germany) and PAXgene® Blood RNA Kit (QIAGEN, Hilden, Germany) was used to purify the total RNA per manufacturer's instructions. The RNA quantification and integrity were determined by Qubit® 2.0 Fluorometer (Life Technologies, Carlsbad, USA) and Agilent 2200 Tape Station® platform (Agilent Technologies, Santa Clara, USA), respectively. For the cDNA synthesis, 800ng of total RNA and High-Capacity cDNA Reverse Transcription kit (Life Technologies, Carlsbad, USA) were used per manufacturer's instructions.

Quantitative RT-PCR (qPCR)

The mRNA quantification of *IL1B, IL6,* and *TNFA* was performed by qPCR using TaqMan® Gene Expression Assay (Life Technologies, Carlsbad, USA) and RotorGene® platform (QIAGEN, Hilden, Germany). The GAPDH gene was chosen as an endogenous reference using GeNorm (qbase+, Biogazelle). The PCR was performed in a multiplex format using Quantifast® Multiplex PCR assay (QIAGEN, Hilden, Germany). Thus, target genes and GAPDH were labeled with fluorescent dye FAM and VIC, respectively. Data were analyzed using Rotor-Gene® Q - Pure detection software (QIAGEN, Hilden, Germany). The results were obtained by relative quantification method ($2^{-\Delta\Delta Ct}$) [18].

Protein measurements

TNFα, IL-1β, IL-6 and BDNF concentrations in EDTA plasma were measured by multiplex assay using Luminex 100™ system (Luminex Corporation Austin, EUA) and custom Milliplex Map Kits (EMD Millipore Corporation, Billerica, USA), following the manufacturer's instructions. The data were analyzed using Xponent 3.1 software. The usPCR concentration was measured by an automated turbidimetric immunoassay using Hitachi 912 Chemistry Analyzer (Myco Instrumentation, Renton, WA).

Statistical analysis

The results were analyzed using SPSS 16 (SPSS Inc., Chicago, IL, USA) and the significance level was established at $p<0.05$. Through the Kolmogorov-Smirnov test, it was verified that variables did not follow a normal distribution. For non-parametric data, the Kruskal-Wallis test was performed, and data are presented as median and interquartile. Multiple comparison analysis was performed to identify differences between groups. To compare the frequencies of qualitative variables, chi-square test ($\chi 2$) or Fisher's exact were performed. Spearman Correlation Coefficient was used to examine the relationship between gene expressions and protein measurements with time since diagnosis and subclinical atherosclerosis markers. Multiple logistic regression analysis was used with BMI as a confounder variable.

RESULTS

In table 1, the demographic characteristics from controls, RD, and BAD groups are shown. There were no differences in gender, ethnicity, Body Mass Index (BMI), and age. When subclinical atherosclerosis markers were evaluated, only the O2 consumption was reduced in the BAD group (table 1). Regarding drugs, the RD group was taking significantly more antidepressants and the BAD group antipsychotics. When we fragmented the class of antidepressants and compared patients under SSRIs treatment between all 3 groups, we observed a p-value of 0.000. This difference may be related to the controls that were not under drug treatment compared to both patient groups. However, when we compared only both patient groups (RD and BAD), this significance disappeared.

Although molecular analysis showed that *IL1B, IL6* and *TNFA* expression were not different between the groups (data not shown), BDNF and IL-1β plasma concentrations showed a statistical difference (table 2). After multiple comparison analysis, BDNF plasma concentrations were increased in both the RD and BAD groups compared with control subjects, while IL-1β plasma concentration was higher in the RD group than in the controls (figure 1 A and B).

Since obesity commonly occurs with depression

and is associated with inflammatory processes, it may act as a confounder variable. So, after controlling for BMI, using regression analysis, BDNF concentrations of the RD and BAD groups were 88% and 42% more than the controls, respectively, but remained significant only in the RD group. IL-1β concentrations were different only between the RD and the controls (p = 0.007). When controlled for BMI, logistic regression showed that the RD and BAD groups were 1.79 and 0.84 times more likely to be IL-1β positive than controls, but were only significant in the RD group (figure 1 C and D). We found a positive correlation of time since diagnosis with BDNF (Spearman's rho = 0.336, p = 0.001) and IL-1β (Spearman's rho = 0.299, p = 0.003) (data not shown) and a negative correlation between BDNF with right CIMT (Spearman's rho = -0.371, p = 0.022) and left CIMT (Spearman's rho = -0.375, p = 0.020) in Control group (data not shown).

DISCUSSION

Heart diseases and depression are two of the major illnesses worldwide and seem to co-occur in a bi-directional way. However, there is still an unknown complex physiologic mechanism to explain both conditions. When we evaluated subclinical atherosclerosis markers, the BAD group showed less O2 consumption in the cardiac stress test. This could be due to the side effects of BAD drug therapies, that are fre-

TABLE 1. DEMOGRAPHIC CHARACTERISTICS AND SUBCLINICAL ATHEROSCLEROSIS MARKERS OF THE CONTROL, RD AND BAD GROUPS

	C	RD	BAD	p
Gender				
Female	33 (33)	29 (29)	38 (38)	0.600*
Male	8 (44.4)	5 (27.8)	5 (27.8)	
Ethnicity				
White	20 (31.7)	19 (30.2)	24 (38.1)	0.908*
Brown	14 (40.0)	9 (25.7)	12 (34.3)	
Black	6 (40.0)	5 (33.3)	4 (26.7)	
Asian	1 (25.0)	1 (25.0)	2 (50.0)	
Others	0 (0)	0 (0)	1 (100)	
Age (years)	50.4 ± 9.0	50.5 ± 12.0	46.5 ± 10.6	0.157**
BMI (kg/m^2)	28.7 ± 8.4	30 ± 6.9	29.4 ± 4.9	0.259**
WC (cm)	61.3 ± 43.5	99.3 ± 13.0	83.5 ± 34.0	0.005**
CIMT				
Right (mm)	0.71 ± 0.19	0.66 ± 0.18	0.62 ± 0.22	0.272**
Left (mm)	0.69 ± 0.16	0.66 ±0.17	0.6 ± 0.20	0.206**
Ankle Brachial Index				
Right	0.92 ± 0.09	0.93 ± 0.14	0.94 ± 0.07	0.195**
Left	0.89 ± 0.09	0.86 ± 0.19	0.91 ± 0.10	0.647**
Tomography				
Calcium score	17.9 ± 84.7	34.9 ± 148.8	9.36 ±46.5	0.309**
Cardiac Stress test				
Effective (%)	40.5	27.4	32.1	0.499*
Ineffective (%)	25	41.7	33.3	
O2 consumption (TEM)	11.0 ± 3.2	10.9 ±7.3	8.3 ±2.0	0.001**
Framingham score (Directive 2007)	1.88 ± 2.6	2.35 ± 3.7	1.48 ± 3.3	0.367**

Values are shown as mean (%); mean ± SD for age, BMI, WC, Right and Left CIMT, Ankle Brachial Index Right and Left, and Calcium score; and mean (%) for Cardiac Stress test, Effective and Ineffective; C - Controls; RD - Resistant Depression; BAD - Bipolar Affective Disorder; BMI - Body Mass Index; WC - Waist Circumference; CIMT - Carotid intima-media thickness. *Chi-square test; **Kruskal-Wallis Test.

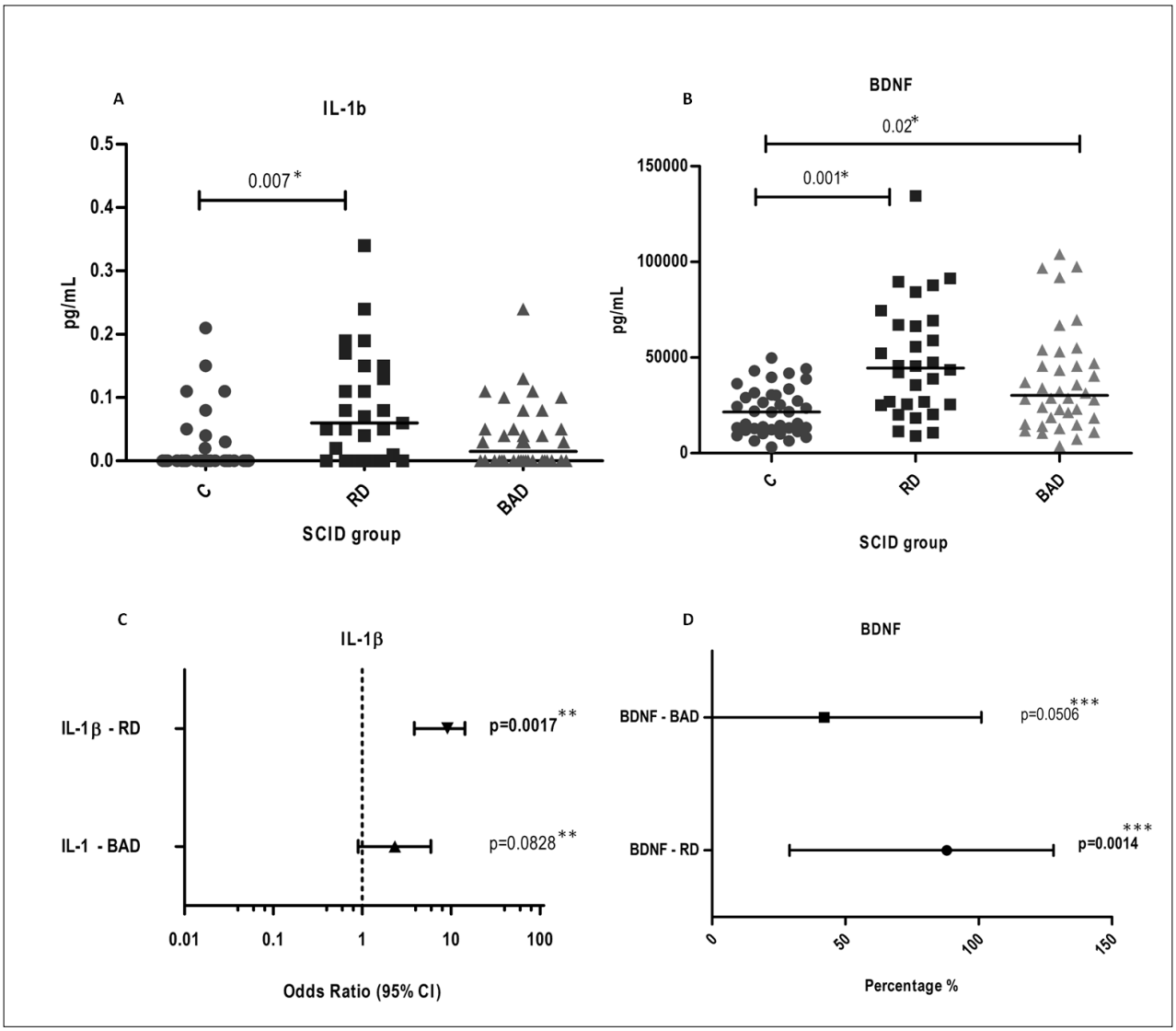

FIGURE 1. A AND B: MULTIPLE COMPARISON ANALYSIS OF IL-1B AND BDNF PLASMA CONCENTRATION BETWEEN CONTROLS (C), RESISTANT DEPRESSION (RD) AND BIPOLAR AFFECTIVE DISORDER (BAD) GROUPS; C AND D: MULTIPLE LOGISTIC REGRESSION FOR BMI VARIABLE ASSOCIATED WITH IL-1B AND BDNF PLASMA CONCENTRATION. * MULTIPLE COMPARISON ANALYSIS; ** LOGISTIC REGRESSION; ***GENERALIZED GAMMA MODEL.

quently associated with weight gain and metabolic disturbance [19]. However, the remaining parameters were not different between groups.

Since atherosclerosis is a slow and progressive disease, it was observed that 30.9% of the patients had the disease for less than five years when they participated in the study, the time between the diagnosis of mood disorder and laboratory testing was not sufficient to observe the association between depression symptoms and CAD. However, a study observed that patients with slow coronary flow have higher rates of depression, anxiety, and overall psychological distress. So, slow-coronary-flow patients should have special medical monitoring [20].

Besides that, our patients have been polymedicated so far, which might explain the lack of association of patients groups with atherosclerosis markers. One of the medications used by our patients was Selective Serotonin Reuptake Inhibitors (SSRI), a widely used antidepressant that is also associated with a lower risk of death or recurrence of myocardial infarction [21]. A similar result was found for coronary heart disease. The authors have confirmed that inflammation and metabolic factors are significantly increased in depression and suggest a higher risk of coronary heart disease. Using SSRI may reduce these factors and, consequently, the risk of coronary heart disease, as suggested in a previous study [22]. Inflammation underlies both heart diseases and depression, so inflammatory markers seem to be elevated in both conditions [23].

We found that BDNF is significantly higher in RD and BAD patients than in controls. It is known that BDNF blood concentrations are associated with clinical changes in depression [24]. However, regarding the cause and effect relationship, Molendijk and colleagues showed in a longitudinal study that the decrease in plasma BDNF concentration is a peripheral manifestation of depression[25].

Since our patients were under drug treatment, mainly antidepressants (data not shown), the high concentrations of BDNF found in both RD and BAD groups were expected, suggesting a positive pharmacotherapy response. However, there is no consensus about the effect of antidepressant treatment on BDNF changes. Bus and colleagues (2014) found that persistent and remitted major depressive disorder patients presented a decline in BDNF levels, which were not influenced by antidepressants [26]. On the other hand, low BDNF concentrations have been associated with major depressive disorders and found to normalize with antidepressant treatment [27]. The same results were observed by Castren in 2016 [28]. A recent meta-analysis shows that a period of treatment with conventional antidepressants leads to the increase of BDNF concentration[29], making this molecule an essential determinant of antidepressant efficacy. However, in these studies, the follow-up period was 2 years or less. In our study, RD and BAD patients had used antidepressants for more than 2 years, 74.3% and 73.8%, respectively. Also, 60% of RD patients and 50% of BAD patients had been using antidepressant drugs for more than five years.

Although some researchers suggest BDNF as a biomarker for major depressive and bipolar disorders [25,30], when controlled for the confounding variable of BMI, only the RD group remained significantly associated with BDNF.

When we analyzed BDNF with subclinical atherosclerosis markers between groups, we found a significant negative correlation between BDNF with right/left CIMT only in the control group. These results were not found in both disease groups (table 6). Hatch and colleagues showed that lower BDNF was associated with higher mean CIMT in adolescents with symptomatic Bipolar Disorder [31]. However, our patients were under medication treatment which may have led to an increase in BDNF concentrations and consequently prevented carotid intima-media thickening. This could explain the lack of association between subclinical atherosclerosis markers and the study groups (table 2).

In addition to BDNF measurement, we also evaluated gene expression and plasma concentration of IL-1β, IL-6 and TNF-α, which are the most important biomarkers of an inflammatory condition in depression, according to literature [4,32]. In this study, only IL-1β concentrations were significantly higher in refractory depression patients than controls. Even after controlling BMI as a confounding variable, the RD group remained significantly associated with IL-1β. In 2012, a meta-analysis showed that only in the European studies the IL-1β concentrations were significantly higher in patients with major depressive disorder [33].

In depression syndrome, some studies showed a role of IL-1β in mediating BDNF decrease [4,27] This means that when there is an increase of IL-1β concentrations, a reduction in BDNF concentrations occurs [27]. According to literature, in patients undergoing antidepressant treatment, there is a decrease of IL-1β and an increase of BDNF concentrations. In our study, the results did not show this profile. Although BDNF concentrations were increased in RD patients undergoing treatment, IL-1β concentrations remained high in this group.

A meta-analysis study including 6 publications showed that antidepressant treatments reduce IL-1β concentrations [34]. Our results, however, showed a different profile that is consistent with many clinical studies and *in vitro* assays. Hernandéz et al (2008) studied patients with major depressive disorder undergoing treatment with SSRI for 52 weeks and found an 86% increase in IL-1β concentrations between 0

TABLE 2. BDNF, IL-1B, IL-6 AND TNFA CONCENTRATIONS IN CONTROL, RD AND BAD GROUPS

Protein	C	RD	BAD	p*
BDNF (pg/mL)	21793 (12613; 32555.5)	43642 (25612; 66384)	32333 (19989; 53524)	0.002
IL-1β (pg/mL)	0.00 (0.00; 0.03)	0.05 (0.00; 0.14)	0.02 (0.00; 0.05)	0.005
IL-6 (pg/mL)	0.41 (0.10; 0.93)	0.26 (0.14; 0.89)	0.38 (0.12; 1.43)	0.731
TNFα (pg/mL)	6.15 (3.96; 10.20)	5.69 (3.93; 7.92)	6.87 (5.19; 9.26)	0.244
usCRP (mg/dL)	0.60 (0.22; 0.90)	0.60 (0.14; 1.00)	0.60 (0.26; 0.95)	0.813

Values are shown as median (25%; 75%); C - Controls; RD - Resistant Depression; BAD - Bipolar Affective Disorder; BDNF - Brain-derived neurotrophic factor; IL1-β - Interleukin-1beta; IL-6 - Interleukin-6; TNFα - Tumor necrosis factor-alpha; usCRP - ultrasensitive C-reactive protein. *Kruskal-Wallis Test.

and 52 weeks [35]. Also, an *in vitro* study that evaluated the effect of three antidepressant drugs on cytokine secretion showed that Citalopram and Mirtazapine increase IL-1β secretion [36]. This suggests that different types of antidepressants can lead to a variation in IL-1β secretion, probably due to the mechanism of action of the drug [37].

According to literature, IL-1β production may affect the neuronal plasticity through impairing the signaling triggered by the binding of BDNF to TrkB receptor [38,39]. Therefore, the results may suggest that in patients with resistant depression, despite the high production of BDNF in response to drug treatment, there is also an increased production of IL-1β that could interfere with BDNF effect. *IL1B* was not expressed differently between the groups; however, the elevated IL-1β secretion in the RD group leads us to expect that inflammasome complexes are an individual characteristic which may play an important role in the antidepressant drug resistance.

In 2013, Iwata proposed the "inflammasome hypothesis of depression" [40]. It was based on the pathway between NLRP3 to IL-1β. NLRP3 is a cytosolic protein complex, also known as inflammasome. Usually, this complex is formed in response to infection caused by pathogenic and non-pathogenic microorganisms. Inflammasome proteins recognize these microorganisms, activate caspase-1, which cleaves the pro- IL-1β in secreted cytokine [4,41].

Alcocer-Gómez et al. (2014) have shown that IL-1β concentrations, NLRP3, and caspase-1 gene expression were increased in non-treated major depressive disorder patients and were reduced in patients who receive amitriptyline [42]. This study is in agreement with the inflammasome hypothesis of depression, which states that psychological stress activates NLRP3 and leads to the development of depression [40].

In our study, the patients were diagnosed with refractory depression because they did not respond to previous antidepressant treatment. About 29% to 46% of patients with depression do not adequately respond to the first antidepressant treatment, and between 19% and 34% of these patients were considered as non-responders [43,44].

Our results showed that BDNF and IL-1β concentrations were increased in RD patients, but there was no difference in gene expression in this group compared to others. This evidence suggests that, in Refractory Depression, the antidepressant drugs acted on mRNA transcription but had no effect on the secretion mechanism of IL-1β. Given the role of the inflammasome in depression, our results lead us to suppose that refractory depression patients have a different cleavage mechanism to secret IL-1β. Also, a positive correlation was also found between IL-1β, BNDF and time of diagnosis that highlight our hypothesis. However, more studies are necessary to validate it.

Many studies have discussed the use of both anti-inflammatory and antidepressants drugs in RD treatment. However, there is still a concern about the adverse events of anti-inflammatory drugs and the interaction between these drugs and antidepressants [45].

CONCLUSION

In conclusion, our results suggest that when taken together, both BDNF and IL-1β plasma concentrations could be used in the early identification of RD patients, allowing a pharmacotherapy association with anti-inflammatory drugs, for example. However, further studies are necessary in order to validate these findings.

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RESUMO

FUNDAMENTAÇÃO: *Não há fortes evidências sobre a associação entre o perfil inflamatório e o padrão de resposta ao tratamento medicamentoso em pacientes depressivos que podem resultar em ocorrência de doença coronariana.*

OBJETIVO: *O objetivo deste estudo foi comparar os marcadores de aterosclerose subclínica, o perfil inflamatório e a produção de BDNF em pacientes com Depressão Resistente (DR) ou Transtorno Afetivo Bipolar (BAD) sob tratamento convencional.*

MÉTODOS: *A população avaliada incluiu 34 RD, 43 BAD e 41 controles. Os marcadores de aterosclerose subclínica foram avaliados por ultrassonografia, tomografia e teste de esforço. As concentrações plasmáticas de TNFα, IL-1β, IL-6 e BDNF foram medidas utilizando*

Luminex100TM. A concentração de usCRP foi medida por imunoensaio turbidimétrico. A expressão de IL1B, IL6 e TNFA foi determinada usando TaqMan®. Para as análises estatísticas, foi estabelecido o nível de significância de p < 0,05.

RESULTADOS: *Quanto aos marcadores de aterosclerose subclínica, apenas o consumo de O2 foi reduzido no grupo BAD (p = 0,001). Embora não tenham sido encontradas diferenças na expressão gênica, a concentração plasmática de BDNF e IL-1β foi aumentada no grupo RD (p = 0,002 e p = 0,005, respectivamente) mesmo sob tratamento antidepressivo, o que sugere que esses medicamentos não têm efeito na secreção de IL-1β e que o inflamassomo pode desempenhar um papel na resposta terapêutica.*

CONCLUSÃO: *Juntas, as concentrações BDNF e IL-1β poderiam ser usadas para a identificação precoce de pacientes com DR.*

PALAVRAS-CHAVE: *Transtorno afetivo bipolar. Depressão. Inflamação. Aterosclerose. BDNF, IL-1β.*

REFERENCES

1. Moussavi S, Chatterji S. Depression, chronic diseases, and decrements in health: results from the World Health Surveys. Lancet. 2007;370:851–8.
2. Global Burden of Disease Study 2013 Collaborators. Global, regional, and national incidence, prevalence, and years lived with disability for 301 acute and chronic diseases and injuries in 188 countries, 1990-2013: a systematic analysis for the Global Burden of Disease Study 2013. Lancet (London, England). 2015 Aug;386(9995):743–800.
3. S. de Sá A, Campos C, B.F. Rocha N, Yuan T-F, Paes F, Arias-Carrión O, et al. Neurobiology of Bipolar Disorder: Abnormalities on Cognitive and Cortical Functioning and Biomarker Levels. CNS Neurol Disord - Drug Targets [Internet]. 2016 Jul 12;15(6):713–22. Available from: http://www.eurekaselect.com/openurl/content.php?genre=article&issn=1871-5273&volume=15&issue=6&spage=713
4. Miller AH, Raison CL. The role of inflammation in depression: from evolutionary imperative to modern treatment target. Nat Rev Immunol. 2016;16(1):22–34.
5. Brietzke E, Stertz L, Fernandes BS, Kauer-Sant'Anna M, Mascarenhas M, Escosteguy Vargas A, et al. Comparison of cytokine levels in depressed, manic and euthymic patients with bipolar disorder. J Affect Disord [Internet]. 2009 Aug;116(3):214–7. Available from: http://linkinghub.elsevier.com/retrieve/pii/S0165032708004795
6. Jacoby AS, Munkholm K, Vinberg M, Pedersen BK, Kessing LV. Cytokines, brain-derived neurotrophic factor and C-reactive protein in bipolar I disorder – Results from a prospective study. J Affect Disord [Internet]. 2016 Jun;197:167–74. Available from: https://linkinghub.elsevier.com/retrieve/pii/S0165032715314439
7. Fernandes BS, Gama CS, Kauer-Sant'Anna M, Lobato MI, Belmonte-de-Abreu P, Kapczinski F. Serum brain-derived neurotrophic factor in bipolar and unipolar depression: A potential adjunctive tool for differential diagnosis. J Psychiatr Res [Internet]. 2009 Oct;43(15):1200–4. Available from: http://linkinghub.elsevier.com/retrieve/pii/S0022395609000910
8. de Sousa RT, van de Bilt MT, Diniz BS, Ladeira RB, Portela L V., Souza DO, et al. Lithium increases plasma brain-derived neurotrophic factor in acute bipolar mania: A preliminary 4-week study. Neurosci Lett [Internet]. 2011 Apr;494(1):54–6. Available from: http://linkinghub.elsevier.com/retrieve/pii/S030439401100245X
9. Corey-Lisle PK, Nash R, Stang P, Swindle R. Response, Partial Response, and Nonresponse in Primary Care Treatment of Depression. Arch Intern Med [Internet]. 2004 Jun 14;164(11):1197. Available from: http://archinte.jamanetwork.com/article.aspx?doi=10.1001/archinte.164.11.1197
10. Sluzewska A, Sobieska M, Rybakowski JK. Changes in Acute-Phase Proteins during Lithium Potentiation of Antidepressants in Refractory Depression. Neuropsychobiology [Internet]. 1997;35(3):123–7. Available from: https://www.karger.com/Article/FullText/119332
11. Lanquillon S, Krieg JC, Bening-Abu-Shach U, Vedder H. Cytokine production and treatment response in major depressive disorder. Neuropsychopharmacology. 2000 Apr;22(4):370–9.
12. Bozzini S, Falcone C. The Interface of Coronary Artery Disease and Depression : Pathophysiology and Diagnosis. J Cardiovasc Disord. 2015;2(2):1–6.
13. Rosenblat JD, Cha DS, Mansur RB, McIntyre RS. Inflamed moods: A review of the interactions between inflammation and mood disorders. Prog Neuro-Psychopharmacology Biol Psychiatry [Internet]. 2014 Aug;53:23–34. Available from: https://linkinghub.elsevier.com/retrieve/pii/S0278584614000141
14. Ejiri J. Possible Role of Brain-Derived Neurotrophic Factor in the Pathogenesis of Coronary Artery Disease. Circulation. 2005 Oct;112(14):2114–20.
15. Del-Ben CM, Vilela JAA, Crippa JA de S, Hallak JEC, Labate CM, Zuardi AW. Confiabilidade teste-reteste da entrevista clínica estruturada para o DSMIV - Versão clínica (SCID-CV) traduzida para o português. Rev Bras Psiquiatr. 2001 Sep;23(3):156-9.
16. Thase ME. Evaluating antidepressant therapies: remission as the optimal outcome. J Clin Psychiatry. 2003;64 Suppl 1:18–25.
17. Ellestad M. Prova de Esforço. Rio de Janeiro: Cultura Médica; 1984.
18. Livak KJ, Schmittgen TD. Analysis of Relative Gene Expression Data Using Real-Time Quantitative PCR and the 2−ΔΔCT Method. Methods. 2001 Dec;25(4):402–8.
19. Kemp DE. Managing the side effects associated with commonly used treatments for bipolar depression. J Affect Disord. 2014 Dec;169:S34–44.
20. Karatas MB, Sahan E, Özcan KS, Çanga Y, Güngör B, Onuk T, et al. Anxiety, Depression, and General Psychological Distress in Patients with Coronary Slow Flow. Arq Bras Cardiol. 2015;
21. Barr Taylor C, Youngblood ME, Catellier D, Veith RC, Carney RM, Burg MM, et al. Effects of Antidepressant Medication on Morbidity and Mortality in Depressed Patients After Myocardial Infarction. Arch Gen Psychiatry. 2005;62(7):792–8.
22. Mathews MJ, Mathews EH, Liebenberg L. The mechanisms by which antidepressants may reduce coronary heart disease risk. BMC Cardiovasc Disord. 2015 Dec;15(1):82.
23. Halaris A. Co-morbidity between cardiovascular pathology and depression: Role of inflammation. In: Inflammation in Psychiatry. 2013.
24. Brunoni AR, Lopes M, Fregni F. A systematic review and meta-analysis of clinical studies on major depression and BDNF levels : implications for the role of neuroplasticity in depression. Int J Neuropsychopharmacol. 2008;11(8):1169–80.
25. Molendijk ML, Spinhoven P, Polak M, Bus BAA, Penninx BWJH, Elzinga BM. Serum BDNF concentrations as peripheral manifestations of depression: evidence from a systematic review and meta-analyses on 179 associations (N=9484). Mol Psychiatry [Internet]. 2014 Jul 20;19(7):791–800. Available from: http://www.nature.com/articles/mp2013105
26. Bus BAA, Molendijk ML, Tendolkar I, Penninx BWJH, Prickaerts J, Elzinga BM, et al. Chronic depression is associated with a pronounced decrease in serum brain-derived neurotrophic factor over time. Mol Psychiatry [Internet]. 2015 May 26;20(5):602–8. Available from: http://www.nature.com/articles/mp201483
27. Lotrich FE, Albusaysi S, Ferrell RE. Brain-Derived Neurotrophic Factor Serum Levels and Genotype: Association with Depression during Interferon-a Treatment. Neuropsychopharmacology. 2013;38263(10):985–95.
28. Castrén E, Kojima M. Brain-derived neurotrophic factor in mood disorders and antidepressant treatments. Neurobiol Dis. 2016 Jul;
29. Zhou C, Zhong J, Zou B, Fang L, Chen J, Deng X, et al. Meta-analyses of comparative efficacy of antidepressant medications on peripheral BDNF concentration in patients with depression. Hashimoto K, editor. PLoS One [Internet]. 2017 Feb 27;12(2):e0172270. Available from: https://dx.plos.org/10.1371/journal.pone.0172270
30. Fernandes BS, Molendijk ML, Köhler CA, Soares JC, Manuel S Leite CG,

Machado-Vieira R, et al. Peripheral brain-derived neurotrophic factor (BDNF) as a biomarker in bipolar disorder: a meta-analysis of 52 studies. BMC. 2015;13(289):1-22.

31. Hatch JK, Scola G, Olowoyeye O, Collins JE, Andreazza AC, Moody A, et al. Inflammatory Markers and Brain-Derived Neurotrophic Factor as Potential Bridges Linking Bipolar Disorder and Cardiovascular Risk Among Adolescents. J Clin Psychiatry. 2017 Mar;78(3):e286–93.

32. Miller AH, Maletic V, Raison CL. Inflammation and Its Discontents: The Role of Cytokines in the Pathophysiology of Major Depression. Biol Psychiatry. 2009 May;65(9):732–41.

33. Liu Y, Ho RCM, Mak A. Interleukin (IL)-6, tumour necrosis factor alpha (TNF-α) and soluble interleukin-2 receptors (sIL-2R) are elevated in patients with major depressive disorder: A meta-analysis and meta-regression. Journal of Affective Disorders. 2012.

34. Hannestad J, Dellagioia N, Bloch M. The Effect of Antidepressant Medication Treatment on Serum Levels of Inflammatory Cytokines: A Meta-Analysis. Neuropsychopharmacology. 2011;36132(10):2452–9.

35. Hernández ME, Mendieta D, Martínez-Fong D, Loría F, Moreno J, Estrada I, et al. Variations in circulating cytokine levels during 52 week course of treatment with SSRI for major depressive disorder. Eur Neuropsychopharmacol. 2008;18:917-24.

36. Munzer A, Sack U, Mergl R, Schönherr J, Petersein C, Bartsch S, et al. Impact of Antidepressants on Cytokine Production of Depressed Patients in Vitro. Toxins (Basel). 2013;5:2227-40.

37. Ho PS, Yeh YW, Huang SY, Liang CS. A shift toward T helper 2 responses and an increase in modulators of innate immunity in depressed patients treated with escitalopram. Psychoneuroendocrinology. 2015;

38. Tong L, Balazs R, Soiampornkul R, Thangnipon W, Cotman CW. Interleukin-1β impairs brain derived neurotrophic factor-induced signal transduction HHS Public Access. Neurobiol Aging [Internet]. 2008 [cited 2018 Nov 2];29(9):1380–93. Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4052889/pdf/nihms62176.pdf

39. Carlos AJ, Tong L, Prieto GA, Cotman CW. IL-1β impairs retrograde flow of BDNF signaling by attenuating endosome trafficking. J Neuroinflammation [Internet]. 2017 Dec 2;14(1):29. Available from: http://jneuroinflammation.biomedcentral.com/articles/10.1186/s12974-017-0803-z

40. Iwata M, Ota KT, Duman RS. The inflammasome: Pathways linking psychological stress, depression, and systemic illnesses. Brain Behav Immun. 2013;

41. Strowig T, Henao-Mejia J, Elinav E, Flavell R. Inflammasomes in health and disease. Nature. 2012 Jan;481(7381):278–86.

42. Alcocer-Gómez E, de Miguel M, Casas-Barquero N, Núñez-Vasco J, Sánchez-Alcazar JA, Fernández-Rodríguez A, et al. NLRP3 inflammasome is activated in mononuclear blood cells from patients with major depressive disorder. Brain Behav Immun. 2014;

43. Fava M, Davidson KG. Definition and epidemiology of treatment-resistant depression. Psychiatr Clin North Am. 1996;19(2):79–200.

44. Keller MB. Issues in treatment-resistant depression. J Clin Psychiatry. 2005;66(SUPPL. 8):5–12.

45. Andrade C. Antidepressant augmentation with anti-inflammatory agents. J Clin Psychiatry. 2014;75(9):975–7.

ORIGINAL ARTICLE

Subtar arthroscopic debridment for the treatment of sinus tarsi syndrome: case series

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SUMMARY

OBJECTIVE: *The objective of this study is to report the results of arthroscopic debridement of the subtalar joint in eight patients with Sinus Tarsi Syndrome (STS) refractory to conservative treatment.*

METHODS: *This is a retrospective study of eight patients with STS who underwent subtalar arthroscopy for debridement of the sinus tarsi between January 2015 and January 2017 after six months of conservative treatment. All patients answered an epidemiological questionnaire and underwent functional evaluation with the Visual Analogue Pain Scale (VAS) and the American Orthopedic Foot and Ankle Society Score (AOFAS) in the preoperative and in the last evaluation (average of 12 months - 6-24 months).*

RESULTS: *All patients showed severe synovitis in the region. Seven patients had remnants of the talocalcaneal ligaments and six of the cervical ligament. AOFAS increased by 30 points on average (51.75 in the preoperative period to 82.62 in the last follow-up) and the VAS decreased on average by 5 points (7.37 preoperatively to 2.12 in the last follow-up). These results were statistically significant with p = 0.043 and p = 0.032 respectively. Six patients described the result as excellent and two as good. No complications were reported. All patients returned to sports after six months of follow-up.*

CONCLUSION: *The arthroscopic debridement of the subtalar joint is an effective and safe alternative in the treatment of STS refractory to conservative treatment. More studies, with a prospective methodology, are necessary to prove the results of this technique.*

KEYWORDS: *Arthroscopy/methods. Tarsal bones. Subtalar joint. Debridement.*

INTRODUCTION

The tarsal sinus syndrome (TSS) can be understood as a set of conditions that affect this topography and produce similar signs and symptoms. Even though its incidence is not reported to be high, it often causes pain and permanent disability, likely removing affected individuals from their activities for long periods. In this syndrome, the presence of pain within the tarsal sinus, which worsens with deep palpation, may be accompanied by local edema and sense of instability[1-3].

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The etiology of OSH is often undefined. However, the presence of previous trauma can be found in approximately 70% of the cases[1,4]. Inversion sprains, fractures, and repetitive traumas are pointed out as responsible for injuries of the talocalcaneal ligaments that, when poorly healed, could produce a subtalar instability, a finding usually associated with the syndrome[1,4-6].

Ligament remnants and the presence of joint lassitude cause synovitis and chronic local irritation that translates to the patient as local pain and discomfort[7,8]. The initial treatment is based on motor physical therapy and on the use of semi-rigid orthoses, which reduce the mobility of the area[3,4,6].

In the failure of a non-surgical approach, the arthroscopic debridement is a viable alternative with high rates of positive outcomes.[1,2,4,5,9] The technique is performed using two portals anterior to the lateral malleolus, seeking visualization and resection of the ligament debris, as well as of the hyperplastic synovial tissue.

The association of the subtalar instability with the TSS has led some authors to propose adjuvant stabilizing procedures for this region. These approaches include from a simple Brostrom-Gould ligamentoplasty to reconstructions with free grafts and talocalcaneal arthrodesis. Despite the few series of cases, the techniques have shown consistent and lasting results [6,10,11].

The objective of this study is to report the results of subtalar arthroscopic debridement in eight patients with TSS resistant to conservative treatment.

METHODS

This is a retrospective study with eight TSS patients who underwent subtalar arthroscopy for debridement of the tarsal sinus between January 2015 and January 2017. Four men and four women, with an average age of 43 years (33-57 years), were diagnosed with TSS for reporting pain on the tarsal sinus, exacerbated by deep palpation and mobilization of the subtalar. All patients were submitted to x-rays with foot and ankle load, which showed an absence of degenerative diseases, in addition to an evaluation by magnetic resonance imaging, which showed an increase of signal in the area and/or the presence of talocalcaneal ligament injuries. None of the patients presented sequels from fractures, osteoarthritis or tarsal coalition. Only after six months of conserva-

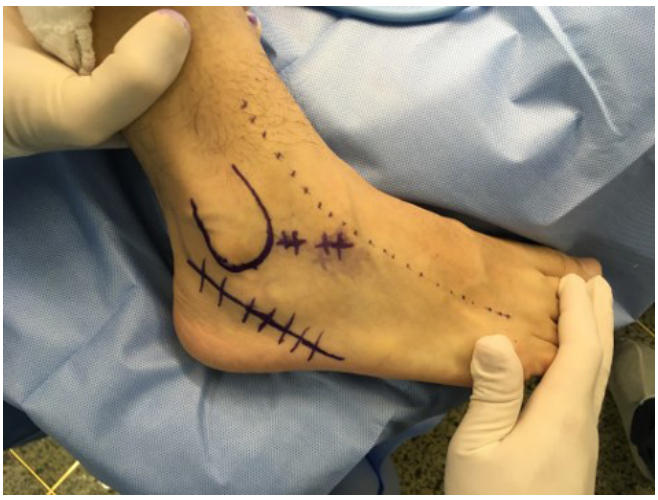

FIGURE 1. DEMARCATION OF THE ARTHROSCOPIC PORTALS.

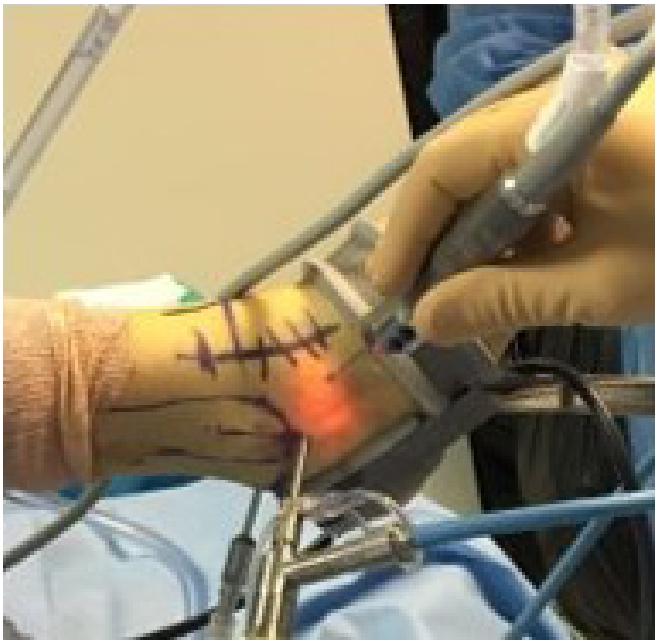

FIGURE 2. ARTHROSCOPIC TRIANGULATION.

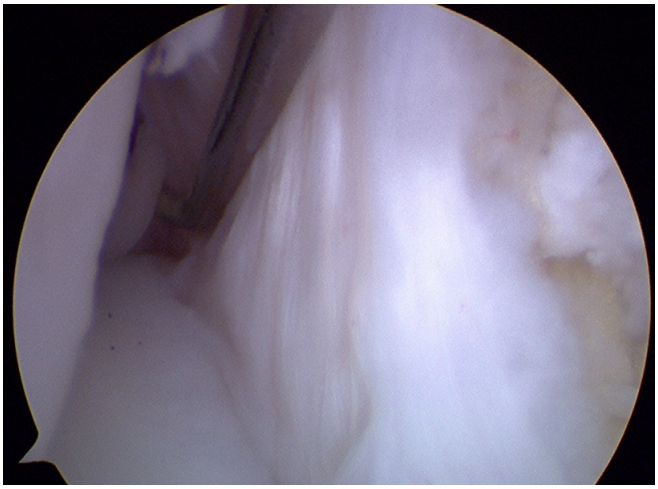

FIGURE 3. ARTHROSCOPIC IMAGE SHOWING REMNANTS OF INTEROSSEOUS LIGAMENTS AND LATERAL TALOCALCANEAL AND INTENSE LOCAL SYNOVITIS.

tive treatment, including rest, analgesic and anti-inflammatory medication, muscle strengthening, and use of semi-rigid orthosis, surgical treatment was indicated.

After approval by the ethics committee of the institution and the signature of the informed consent term, all patients filled out the epidemiological questionnaire and underwent functional assessment using the Visual Analog Scale for pain (VAS) and the American Orthopaedic Foot and Ankle Society Score (Aofas) in the preoperative consultation and during follow-up, with an average of 12 months (6-24 months).

The statistical analysis was performed with the Gretl (2017c) software. Student t-test was used to compare pre-operative and post-operative data. In this study, we adopted a 0.05 value for the alpha error and consequent rejection of the null hypothesis.

SURGICAL TECHNIQUE

The patients underwent general anesthesia associated with the blocking of the saphenous and femoral nerves. After positioning the patients on dorsal decubitus with a small cushion under the gluteus and pneumatic tourniquet on the thigh, they were all marked for the arthroscopic portals and the local anatomical references (Figure 1).

The intermediary portal was established at 1 cm from the anterior edge of the lateral malleolus, at the height of its apex, directly on the tarsal sinus. The anterolateral portal was performed 1 cm distal and 3 cm above the apex of the fibula[12]. Skin incisions, transverse, with an 11 blade, were made in the places previously established, followed by the introduction of a gripper for opening the capsule. The arthroscopy was performed with a 4.5 mm and 30º lens, instrumented by a shaver and 4.5 mm grippers (Figure 2). An electrocoagulation tip of the same size was also used. The resection of the synovitis and the hypertrophic ligament remains was performed, switching the portal and visualization used whenever necessary. The procedure was successful and finalized after verification of complete cleaning of the cavity and the absence of impact of soft parts to the movement of the subtalar (Figure 3).

After the suture, a sterile bandage was applied, followed by a splint. In the first postoperative follow up, after one week, the bandage was replaced, and the splint substituted by an immobilizing boot. Physical therapy started on the same week, stimulating the mobility of the ankle and subtalar in a controlled

TABLE 1. PATIENTS' DESCRIPTIVE DATA

Patient no.	Side	Age	Gender	Aofas Pre	Aofas Post	Eva Pre	Eva Post	Follow-up (Months)	Diagnosis	Procedure
1	E	55	M	58	69	9	4	8	TSS + Ankle lateral instability	Subtalar arthroscopy + Brostrom-Gould
2	E	52	F	68	90	5	1	6	TSS	Subtalar arthroscopy
3	E	57	F	32	69	7	4	12	TSS + Ankle lateral instability + Pes cavus	Subtalar arthroscopy + Brostrom-Gould + Dwyer
4	D	33	M	71	84	8	2	24	TSS + Ankle lateral instability + Pes cavus	Subtalar arthroscopy + Brostrom-Gould + Dwyer
5	E	35	F	32	69	9	3	10	TSS + Ankle lateral instability	Subtalar arthroscopy + Brostrom-Gould
6	D	37	M	76	90	7	1	12	TSS + Ankle lateral instability	Subtalar arthroscopy + Brostrom-Gould
7	D	37	F	33	90	6	1	7	TSS + Ankle lateral instability	Subtalar arthroscopy + Brostrom-Gould
8	D	40	M	44	100	8	1	18	TSS	Subtalar arthroscopy
Average		43.25		51.75	82.62	7.37	2.12	12.12		

way, avoiding the inversion and internal rotation up to the sixth week. The load was initiated during the first week progressively. The boot was replaced by a rigid anklet on the fourth week, and light physical activities were initiated. The patients were allowed to resume sporting activities at the 16th week.

RESULTS

All eight patients reported a previous history of trauma in the ankle region, seven of them by inversion sprains and one by nonspecific trauma. The descriptive data of the patients can be found in Table 1. It is worth noting that only two patients presented isolated tarsal sinus syndrome and received arthroscopic debridement of the subtalar as an isolated intervention. The other four had lateral instability of the ankle and two, in addition to the instability, also had associated rearfoot varus. In the patients who had instability, Brostrom-Gould ligamentoplasty was performed (which also allowed for the stabilization of the subtalar), and, in patients with rearfoot varus, Dwyer osteotomy of the calcaneus was performed.

During the subtalar arthroscopy, all patients showed exuberant synovitis in the region. Seven patients had remnants of talocalcaneal ligaments and six of the cervical ligament. No cartilage or tumoral lesion was found.

Regarding the scores measured, the Aofas increased by 30 points on average (51.75 at the preoperative to 82.62 at the last follow-up) and VAS decreased on average 5 points (7.37 at the preoperative to 2.12 at the last follow-up) during the evolution. These results were statistically significant, with $p = 0.043$ and $p = 0.032$, respectively. Six patients described the result as excellent and two as good. No complication was reported. All patients resumed sporting activities after six months of follow-up.

DISCUSSION

Nonspecific complaints of pain in the lateral region of the ankle motivate a good number of athletes and sedentary patients to seek medical care. Among the differential diagnoses possible, TSS stands out. It is a condition common in the region that causes pain and is often not diagnosed and poorly treated. This condition is characterized by pain on the tarsal sinus, commonly associated with a previous traumatic episode or instability of the ankle and subtalar[13,14].

Pathology anatomy studies prove the presence of intense inflammatory infiltration in the tarsal sinus, associated with the presence of chronic synovitis and disorganized ligament remnants[7,8]. These findings support the theory of previous trauma (acute or repetition) as the primary etiologic factor in the pathogenesis of this disease. All patients in our study also presented a history of previous trauma, corroborating the data in the literature.

Initially, the approach used in these individuals is non-operative, comprising of a resting phase, muscle strengthening, analgesia, and functional immobilization[3]. Some authors consider the infiltration of the tarsus with corticosteroids an alternative for resistant cases[1,2]. The failure of these measures, however, requires surgical resolution, which can be performed using an open approach or endoscopy.

Frey et al.[5] described the results of the arthroscopic treatment of TSS in 49 cases. With an average follow-up of seven years, they found a rate of satisfaction of 94%. The author reported only five minor complications; the most common of them was the transient neuropraxia of the superficial fibular nerve. Among the injuries observed during the arthroscopy, the most prevalent was the rupture of the interosseous ligament[5]. Oloff et al.[2], in turn, reported good results using the same technique, with an average Aofas of 85 at the last follow-up. Of the 29 patients in their series, only one was subjected to a secondary to arthrofibrosis arthrotomy. No complications were reported 2. Lee et al.[1] demonstrated the results of arthroscopy in 31 patients (33 feet) with a follow-up of 24 months. The VAS of these patients showed an average improvement of 7.3 to 2.7, and the average trend of the Aofas went from 43.1 to 86.2. Only one patient progressed with irritation of the superficial fibular nerve, showing complete resolution after a neurolysis procedure[1].

This study is in agreement with the positive results presented in the literature. Our patients reported a high rate of satisfaction, 75% considered them excellent, and two results were considered good. We also had high rates of pain reduction, with an average VAS reduction from 7.37 at the preoperative to 2.12 at the last follow-up. The Aofas, in turn, increased from 51.75 at the preoperative to 82.62 at the last follow-up. These results, with statistical significance, were similar to those pre-

viously observed[1,2,5] and proved the effectiveness of the subtalar arthroscopy for treating resistant TSS. In addition, no complications were observed in this study, which demonstrates the safety of the technique.

We believe that the arthroscopic debridement of the subtalar is a viable alternative and a reproducible method for the treatment of TSS irresponsive to the conservative treatment. This study is relevant because it deals with a topic of sparse description in the literature and details the results with objective parameters for evaluation. The disadvantages of this study are its retrospective methodology, low number of patients, and the presence of associated conditions and adjuvant procedures that carry biases to the interpretation of the data.

CONCLUSIONS

Arthroscopic debridement of the subtalar is a safe and effective alternative for the treatment of TSS resistant to conservative treatment. Further studies with a prospective methodology are needed to confirm the results of the technique.

Conflict of interest

The authors declare that there are no conflicts of interest.

RESUMO

OBJETIVO: *O objetivo desse estudo é relatar os resultados do desbridamento artroscópico da subtalar em oito pacientes portadores da Síndrome do Seio do Tarso (SST) refratária ao tratamento conservador.*

MÉTODOS: *Este é um estudo retrospectivo com oito pacientes com diagnóstico de STT que foram submetidos à artroscopia subtalar para desbridamento do seio do tarso entre janeiro de 2015 e janeiro de 2017, após seis meses de tratamento conservador. Todos os pacientes responderam questionário epidemiológico e foram submetidos à avaliação funcional com a Escala Visual Analógica de dor (EVA) e o American Orthopaedic Foot and Ankle Society Score (Aofas) no pré-operatório e na última avaliação, em uma média de 12 meses (6-24 meses).*

RESULTADOS: *Todos os pacientes exibiram intensa sinovite na região. Sete pacientes tinham resquícios de ligamentos talocalcaneanos e seis do ligamento cervical. O Aofas aumentou 30 pontos em média (51,75 no pré-operatório para 82,62 no último seguimento) e a EVA diminuiu em média 5 pontos (7,37 no pré-operatório para 2,12 no último seguimento). Esses resultados foram estatisticamente significativos com p = 0,043 e p = 0,032, respectivamente. Seis pacientes descreveram o resultado como excelente e dois como bom. Nenhuma complicação foi relatada. Todos os pacientes retornaram ao esporte após seis meses de acompanhamento.*

CONCLUSÃO: *O desbridamento artroscópico da subtalar é uma alternativa eficaz e segura no tratamento da SST refratária ao tratamento conservador. Mais estudos, com metodologia prospectiva, são necessários para comprovar os resultados da técnica.*

PALAVRAS-CHAVE: *Artroscopia/métodos. Ossos do tarso. Articulação talocalcânea. Desbridamento.*

REFERENCES

1. Lee KB, Bai LB, Song EK, Jung ST, Kong IK. Subtalar arthroscopy for sinus tarsi syndrome: arthroscopic findings and clinical outcomes of 33 consecutive cases. Arthroscopy. 2008;24(10):1130-4.
2. Oloff LM, Schulhofer SD, Bocko AP. Subtalar joint arthroscopy for sinus tarsi syndrome: a review of 29 cases. J Foot Ankle Surg. 2001;40(18):152-7.
3. Helgeson K. Examination and intervention for sinus tarsi syndrome. N Am J Sports Phys Ther. 2009;4(1):29-37.
4. Pisani G, Pisani PC, Parino E. Sinus tarsi syndrome and subtalar joint instability. Clin Podiatr Med Surg. 2005;22(1):63-77.
5. Frey C, Feder KS, DiGiovanni C. Arthroscopic evaluation of the subtalar joint: does sinus tarsi syndrome exist? Foot Ankle Int. 1999;20(3):185-91.
6. Mittlmeier T, Wichelhaus A. Subtalar joint instability. Eur J Trauma Emerg Surg. 2015;41(6):623-9.
7. Akiyama K, Takakura Y, Tomita Y, Sugimoto K, Tanaka Y, Tamai S. Neurohistology of the sinus tarsi and sinus tarsi syndrome. J Orthop Sci. 1999;4(4):299-303.
8. Rein S, Manthey S, Zwipp H, Witt A. Distribution of sensory nerve endings around the human sinus tarsi: a cadaver study. J Anat. 2014;224(4):499-508.
9. Muñoz G, Eckholt S. Subtalar arthroscopy: indications, technique and results. Foot Ankle Clin. 2015;20(1):93-108.
10. Aynardi M, Pedowitz DI, Raikin SM. Subtalar instability. Foot Ankle Clin. 2015;20(2):243-52.
11. Jung HG, Park JT, Shin MH, Lee SH, Eom JS, Lee DO. Outcome of subtalar instability reconstruction using the semitendinosus allograft tendon and biotenodesis screws. Knee Surg Sport Traumatol Arthrosc. 2015;23(8):2376-83.
12. Beimers L, Frey C, van Dijk CN. Arthroscopy of the posterior subtalar joint. Foot Ankle Clin. 2006;11(2):369-90.
13. Maceira E, Monteagudo M. Subtalar anatomy and mechanics. Foot Ankle Clin. 2015;20(2):195-221.
14. Choudhary S, McNally E. Review of common and unusual causes of lateral ankle pain. Skeletal Radiol. 2011;40(11):1399-413.

ORIGINAL ARTICLE

Sleep disorders in polycystic ovary syndrome: influence of obesity and hyperandrogenism

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SUMMARY

OBJECTIVE: *This study aims to evaluate the sleep of subjects with polycystic ovary syndrome (PCOS), with and without hyperandrogenism, in comparison with a healthy control group and examine the effects of hyperandrogenism and obesity on sleep parameters.*

METHODS: *A total of 44 volunteers were recruited to participate in the study. Clinical, biochemical and polysomnographic parameters were used to diagnose PCOS and hyperandrogenism. The evaluation of sleep quality was made using validated questionnaires and polysomnography test. The frequency of obstructive sleep apnea was also compared between the groups.*

RESULTS: *The study revealed that women with PCOS presented poorer subjective sleep quality, increased incidence of snoring and a higher risk of obstructive sleep apnea, based on the Berlin questionnaire. Also, after adjusting for body mass index, PCOS subjects had rapid eye movement (REM) time lower than those in the control group. PCOS women versus those without hyperandrogenism did not differ on any sleep measurement. Women with obstructive sleep apnea were only diagnosed in the PCOS group.*

CONCLUSIONS: *Our results indicate that PCOS impairs subjective sleep quality, as well as objective sleep quality, due to a reduction in REM sleep stage time in women diagnosed with the syndrome. Obesity affected sleep-related parameters but hyperandrogenism had no effect. Only the PCOS group had obstructive sleep apnea diagnosis.*

ABBREVIATIONS: *AHI = apnea-hypopnea index; BMI = body mass index; ESS = Epworth Sleepiness Scale; OSA = obstructive sleep apnea; PCOS = polycystic ovary syndrome; PSG = polysomnography; PSQI = the Pittsburgh Sleep Quality Index; REM = rapid eye movement;*

KEYWORDS: *hormonal; hyperandrogenism; polycystic ovary syndrome; sleep; women.*

INTRODUCTION

Obstructive sleep apnea (OSA) is a syndrome characterized by recurrent events of partial or total obstruction of the upper airway during sleep, leading to intermittent hypoxemia, which has obesity as the pillar of its physiopathology.[1,2] Studies suggest that androgens influence sleep architecture, favoring the development of OSA.[3-5] In general, symptoms of OSA include fatigue, tiredness, maintenance insomnia, with polysomnography findings showing an increase in the apnea-hypopnea index (AHI), hypoxia

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and some impact on sleep architecture[6,7] Sleep complaints in women vary according to hormonal fluctuations dictated by their menstrual cycles.

Although women have proportionally less severe OSA and it is considered to predominantly affect the male population, a recent study found the disease in 26.1% of women[8]. The problem is likely to be more common, as women with sleep-disordered breathing are 2 to 3 times less likely to report classic symptoms of the disease (snoring, gasping, snorting and sleep apnea), which may lead to reduced clinical recognition of OSA in women compared to men.[9] Women suffering from PCOS tend to have higher levels of respiratory sleep events. It has been suggested that this may be linked to the increased androgen levels often associated with the syndrome.[2] Sleep itself acts as an important modulator of several aspects of endocrine function, making the relationship between these factors difficult to elucidate.[4,10,11]

Polycystic ovary syndrome (PCOS) is the most common endocrine disease, affecting approximately 8% of women in the reproductive stage.[12,13] It is well known that OSA prevalence is increased in women with PCOS compared with women without the disease.[13-15] There also are a broad range of hormonal and metabolic abnormalities in PCOS, and it has been suggested that the hormonal profile of those with the condition is associated with OSA.[16] Previous studies have shown higher levels of testosterone in PCOS patients to be related to OSA.[12,17] However, neither the outcomes of OSA nor the relationship of the symptoms with sleep architecture have been fully explored in this population. It has been hypothesized that obesity and hormonal factors, caused by the disease, act synergistically to impair quality of sleep. Therefore, our hypothesis is that females with PCOS are at increased risk of OSA and other sleep disorders. Our aim is to clarify this, and examine whether hyperandrogenism has any effect on sleep parameters by evaluating PCOS subjects (with and without hyperandrogenism) using polysomnography and sleep questionnaires and compare these patients with a healthy control group.

METHODS

Population

A total of 55 subjects were selected to participate in the study. The volunteers, ranging in age from 16 to 45 years, were recruited from the Endocrinology Division of the Federal University of São Paulo, Brazil. The diagnosis of PCOS was based on the latest 2003 Rotterdam consensus,[18] requiring the presence of at least two of the following features: (*1*) oligomenorrhea or chronic anovulation, (*2*) clinical and/or biochemical hyperandrogenism, and (*3*) ultrasound appearance of polycystic ovaries. These women were distributed into two groups: SOP with hyperandrogenism and SOP without hyperandrogenism.

The control group was comprised of 17 women. Inclusion criteria: a regular menstrual cycle of 28-30 days, normal BMI and in the follicular phase of the menstrual cycle. Exclusion criteria: neurologic conditions and/or being under psychiatric treatment; use of medication for chronic diseases that might interfere with the study results; participation in another clinical study or having participated in a clinical study within a period of 3 months; being a carrier of a disease; having a history of stroke; use of hypnotic, psychotropic, psychostimulant, and/or analgesic drugs; use of hormonal contraceptives; and presence of dysmenorrhea or endometriosis that may interfere with sleep patterns.

All procedures performed in the studies involving human participants followed the ethical standards of the institutional and/or national research committee and the 1964 Helsinki declaration and its later amendments or comparable ethical standards. The study was approved by the Ethics Committee for Research of the Federal University of Sao Paulo (#0588/2010), and informed written consent was obtained from all subjects.

Subjects with other known causes of hyperandrogenism (such as congenital adrenal hyperplasia, androgen-secreting tumors and Cushing's syndrome), using oral contraceptives, corticosteroids, antidiabetic or lipid-lowering drugs in the previous 3 months, having a history of liver disease (such as viral hepatitis B and C, hemochromatosis and autoimmune hepatitis), diabetes mellitus, untreated hypothyroidism, renal, hepatic, cardiac or pulmonary disease, receiving treatment for sleep apnea using medications that alter liver enzymes, with a daily ingestion of more than 20 grams of ethanol, using drugs (sympathomimetics, sympatholytics, and β-blockers), with depression or with chronic diseases were excluded.[16]

Clinical, Anthropometric and Sonographic Measurement

Questionnaires were used to document clinical history, including regularity and length of menstrual cycles, and ovulation status. Signs of androgen excess (hirsutism, alo-

pecia, acne) were noted in the physical examination. Hirsutism with a Ferriman-Gallwey score of 8 or above was considered clinical evidence of androgen excess. Weight (in kilograms) and height (in meters) were measured. The body mass index (BMI) was calculated from the ratio of the weight to height squared.

All subjects underwent an ultrasound examination of the pelvis by the same radiologist. LOGIQ P5 (GE Healthcare®, Wauwatosa, WI) with an 8 MHz transvaginal transducer was used for the ultrasound of the pelvis.

Evaluation of Sleep and Polysomnography

Full-night polysomnography (PSG) was performed, using a digital system (EMBLA® S700®, Embla Systems Inc, Broomfield, CO) at the sleep laboratory for one night. Trained technicians visually scored all of the PSG data according to standardized criteria for investigating sleep.[19] Electroencephalogram arousals and sleep-related respiratory events were scored following the criteria outlined in the American Academy of Sleep Medicine Manual for Scoring Sleep and Associated Events.[20] OSA classification was defined according to the AHI.[21] Participants were diagnosed with OSA if they presented an AHI≥5 and sleep complaints. Participants with an AHI≥15 were diagnosed with OSA, regardless of whether they had any additional complaint.

For subjective evaluation of sleep, we used the Pittsburgh Sleep Quality Index (PSQI), which is an instrument for evaluating the subjective quality of sleep, as well as the number of sleep disturbances occurring during a period of 1 month.[22,23] The Berlin questionnaire, previously validated in a Brazilian Portuguese version, was used to assess the risk for sleep apnea.[24,25] Using this questionnaire's total score, it is possible to differentiate "good sleepers" (score≤5) and "poor sleepers" (score>5). Also, daytime somnolence was evaluated subjectively using the Epworth sleepiness scale (ESS),[26] with a score ≥10 considered excessive daytime sleepiness.

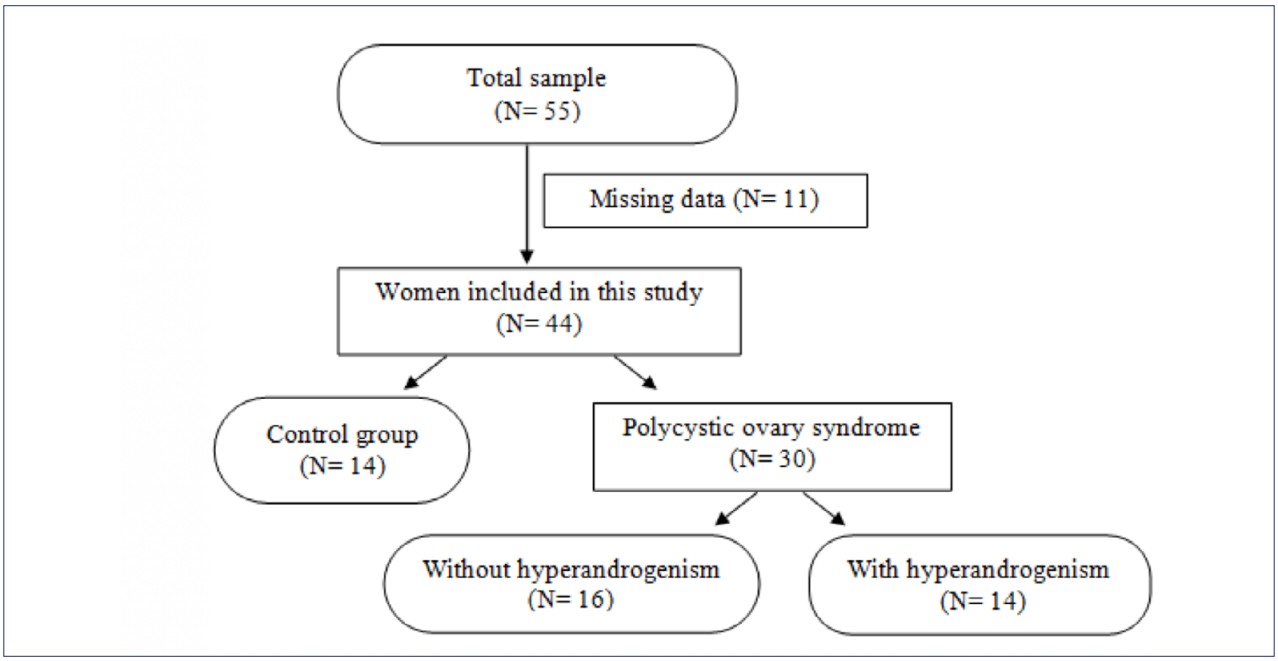
Laboratory Analysis

Total testosterone levels were measured using a UniCel Dxl 800 Immunoassay System (Beckman Coulter®, Brea, CA). The within-assay coefficient of variation for testosterone was 1.99%, and the between-assay coefficient was 4.22%. There are some limitations to measuring testosterone using a chemiluminescence immunoassay, but this was the only laboratory technique available. Serum-free testosterone and bioavailable testosterone were estimated using a previously validated formula.[27] All biochemical assays were performed at the Sleep Institute laboratory.

Statistical analysis

The variables were evaluated regarding normality (Shapiro-Wilk's test) and homogeneity (Levene's test). When the distribution was demonstrated to be nonparametric, the data were standardized through

FIGURE 1. FLOW CHART OF THE DESCRIPTION OF THE STUDY SAMPLE.



Z-score. Statistical analysis of the sample was carried out using the General Linear Model through one-way analysis of variance for continuous variables, and Pearson's chi-squared test was performed to determine the association between categorical variables. To evaluate the relationship between age and BMI with the sleep-related factors, Pearson's correlation test was performed. BMI and age were used as adjustment factors in evaluating the effect of PCOS and hyperandrogenism on sleep, respectively. The results were submitted to adjustment only when the groups had significant statistical differences in age or BMI. The significance level was set to $P<0.05$. Data are presented as mean±standard error of the mean or as frequency (percentage).

RESULTS

From a total of 55 women initially included in the study, 11 individuals were excluded because of missing data (8 related to the PSQI and 3 to BMI). Our final sample of 44 women comprised 14 healthy women and 30 women with PCOS, of whom 14 had a diagnosis of hyperandrogenism (Figure 1).

The descriptive data of the sample analyzed in the study were distributed as healthy women (control group) and women with PCOS. We observed a higher BMI in the PCOS group ($F1,42=36,404$; $P<0.001$) compared to the control group. Regarding the sleep evaluation questionnaires, a higher frequency of women with PCOS was categorized as: high risk (Berlin Questionnaire) ($\chi^2=12,156$; $df=1$; $P<0.001$), poor sleepers (PSQI) ($\chi^2=8,696$; $df=1$; $P<0.01$), snorers ($\chi^2=3,889$; $df=1$; $P<0.05$) and OSA ($\chi^2=5,280$; $df=1$; $P<0.05$). There were no differences in frequency of sleepiness as measured by the Epworth Sleepiness Scale between women with PCOS and the control group. Results presented in Table 1 indicated that age was not statistically different between the groups since the inclusion criteria of the study were strictly followed. Table 2 shows the polysomnography results from the PCOS group adjusted for BMI, which indicate that the percentage of REM sleep was lower in the PCOS group than in the control group ($F1,41=7,245$; $P<0.05$). The effect of BMI as confounding factor was a significant covariate for Pittsburgh sleep scale ($P=0.039$), N1 sleep stage ($P=0.010$), N3 sleep stage ($P=0.003$), REM sleep stage ($P=0.043$), number of Arousals ($P=0.002$), arousals index ($P<0.001$), PLM index ($P=0.021$) and mean oxygen saturation ($P=0.033$).

We also evaluated the possible effect of hyperandrogenism on the objective and subjective aspects of sleep in women with PCOS. A description of the sample indicated that women with hyperandrogenism were younger than the PCOS subjects without hyperandrogenism, 32.2±1.3 for women without hyperandrogenism and 25.6±1.4 3 for women with hyperandrogenism ($F1,28=15,674$; $P<0.001$). There were no observed differences in the following variables: BMI, the frequency of women with high-risk of apnea according to the Berlin Questionnaire, poor sleepers, sleepiness, snoring and those diagnosed with OSA. No differences were seen in the scores for the ESS and the PSQI questionnaires, and there were no differences in the polysomnographic parameters of women with hyperandrogenism in comparison with those without hyperandrogenism when adjusted for age.

The effect of age as a confounding factor was a significant covariate for the following factors: N3 sleep stage ($P=0.039$) and REM sleep stage ($P=0.026$). Sleep-related factors were correlated with age and BMI through Pearson's correlation test considering all participants. The REM sleep stage was the only significant parameter in the correlation between age and sleep-related factors ($r=0.329$, $P=0.029$). On the other hand, the correlation between BMI and sleep-related

TABLE 1. DESCRIPTIVE PARAMETERS OF ALL THE PATIENTS RECRUITED FOR THIS STUDY.

Variables	Control (N=14)	PCOS (N=30)	P-value
Age (years)	27.9±1.7	29.7±1.2	0.412
Body Mass Index (weight/height2)	22.4±1.6	34.3±1.1***	<0.001
High risk for OSA (Berlin questionnaire)	1 (7.1%)	19 (63.3%)***	<0.001
High daytime sleepiness (ESS, score≥10)	5 (35.7%)	15 (50%)	0.375
Poor sleep quality (PSQI, score>5)	7 (50%)	27 (90%)**	0.003
Reported snoring	10 (71.4%)	28 (93.3%)*	0.049
Meet criteria for OSA (from PSG)	0 (0%)	9 (30%)*	0.022

Legend: ESS, Epworth sleepiness scale; OSA, obstructive sleep apnea; PCOS, polycystic ovary syndrome; PSQI, Pittsburgh sleep quality index. The data were presented as mean±SEM or number (%).Note: *P<0.05, **P<0.01 and ***P<0.001 compared to the control group.

TABLE 2. SUBJECTIVE AND OBJECTIVE PARAMETERS OF SLEEP FOR EVALUATION OF POLYCYSTIC OVARY SYNDROME (PCOS) AND CONTROL GROUP, P-VALUE WAS ADJUSTED PER BODY MASS INDEX.

Variables	Control (N=14)	PCOS (N=30)	P-value
ESS score	8.4±1.2	8.5±0.8	0.575
PSQI score	4.6±0.8	8.5±0.5	0.105
Sleep latency (min)	15.9±5.2	26.3±3.5	0.432
REM latency (min)	120.7±16.0	126.6±10.9	0.742
Total sleep time (min)	366.8±16.4	339.4±11.2	0.396
Sleep efficiency (%)	88.0±2.9	80.0±2.0	0.358
N1 sleep stage (% TST)	3.3±1.6	11.5±1.1	0.144
N2 sleep stage (% TST)	47.7±2.3	45.9±1.6	0.585
N3 sleep stage (% TST)	27.9±2.6	25.2±1.8	0.152
REM sleep stage (% TST)	20.5±1.6	17.3±1.1*	0.010
WASO (min)	30.7±10.7	59.1±7.3	0.596
Arousal index (events/h)	12.6±3.2	18.9±2.2	0.198
PLM (events/h)	0.3±0.7	1.5±0.5	0.630
AHI (events/h)	1.6±3.8	9.2±2.6	0.765
Basal oxygen saturation	97.3±0.3	96.9±0.2	0.714
Mean oxygen saturation	96.6±0.3	95.9±0.2	0.873
Minimum oxygen saturation	92.0±1.3	89.5±0.9	0.890

Legend: AHI, apnea-hypopnea index; ESS, Epworth sleepiness scale; PCOS, polycystic ovary syndrome; PLM, periodic limb movements; PSQI, Pittsburgh Sleep Quality Index; REM, rapid eye movement; WASO, wake after sleep onset. The data were presented as mean±SEM. Note: *P<0.05 compared to the control group.

TABLE 3. SUBJECTIVE AND OBJECTIVE PARAMETERS OF SLEEP FOR EVALUATION OF HYPERANDROGENISM, P-VALUE WAS ADJUSTED PER AGE.

	Women with polycystic ovary syndrome		
Variables	Without hyperandrogenism (N=16)	With hyperandrogenism (N=14)	P-value
ESS score	8.8±1.2	8.2±1.3	0.380
PSQI score	8.2±1.0	8.8±1.1	0.739
Sleep latency (min)	22.9±5.4	30.2±5.8	0.677
REM latency (min)	116.6±16.1	138.1±17.2	0.480
Total sleep time (min)	337.6±15.3	341.4±16.4	0.883
Sleep efficiency (%)	80.4±3.1	79.6±3.3	0.469
N1 sleep stage(% TST)	10.7±1.8	12.5±1.9	0.276
N2 sleep stage (% TST)	45.0±2.1	46.8±2.3	0.599
N3 sleep stage (% TST)	26.1±2.3	24.3±2.5	0.095
REM sleep stage (% TST)	18.3±1.3	16.3±1.4	0.605
WASO (min)	61.6±11.9	56.3±12.7	0.437
Arousal index (events/h)	18.6±3.2	19.3±3.4	0.515
PLM (events/h)	1.4±0.8	1.6±0.8	0.320
AHI (events/h)	6.1±4.3	12.8±4.6	0.507
Basal oxygen saturation	96.9±0.3	96.9±0.3	0.573
Mean oxygen saturation	96.1±0.4	95.8±0.4	0.407
Minimum oxygen saturation	91.1±1.4	87.7±1.5	0.184

Legend: AHI, apnea-hypopnea index; ESS, Epworth sleepiness scale; PCOS, polycystic ovary syndrome; PLM, periodic limb movements; PSQI, Pittsburgh Sleep Quality Index; REM, rapid eyes movements; WASO, wake after sleep onset. The data were presented as mean±SEM.

factors showed statistical significance for: sleep efficiency (r=-0.338, *P*=0.025), N1 sleep stage (r=0.611, *P*<0.001), N3 sleep stage (r=-0.410, *P*=0.006), wakefulness after sleep onset (r=0.388, *P*=0.009), arousals index (r=0.533, *P*<0.001), periodic limb movements (r=0.403, *P*=0.007), apnea–hypopnea index (r=0.406, *P*=0.006), basal oxygen saturation (r=-0.333, *P*=0.027), mean oxygen saturation (r=-0.409, *P*=0.006) and minimum oxygen saturation (r=-0.370, *P*=0.013).

DISCUSSION

The findings of this study revealed that the PCOS group presented poorer sleep quality and reduced REM sleep time when compared to the control group. Also, there was a higher risk of apnea according to the Berlin Questionnaire, poorer sleep quality measured by the PSQI and a higher frequency of snorers in PCOS subjects compared to controls. In women with PCOS, no effect of hyperandrogenism was observed on sleep pattern, neither subjectively or objectively

Obesity is common among women with PCOS, but it is not part of the diagnostic criteria. This association has been previously demonstrated.[28] Moreover, the increased anthropometric measures arising from obesity also impact the following factors: increased obstruction of the upper airway events, hypoxia, sleep fragmentation, fatigue and perception of non-refreshing sleep.[7] Thus, the PSG data were adjusted for BMI in the analysis of the PCOS effect. The effect of PCOS on sleep architecture is controversial in the literature,[29,30] however several studies point to an increased risk of obstructive sleep apnea, decreased REM sleep and sleep efficiency.[12,31-33] These studies demonstrated that PCOS promotes a decrease in REM sleep, which is not seen in an obese control group without PCOS. The cause of decreased REM sleep in women with PCOS is still unknown. Our main hypothesis is that obesity and PCOS are strongly associated, resulting in a variety of consequences for the body, specifically neurophysiological impacts. It is possible that adjusting for BMI is not enough to account for all the repercussions promoted by the synergistic action of both factors. The sleep quality, snoring and diagnosis of OSA are factors clinically important due to the increased weight observed in women with PCOS.

In obese women with PCOS the incidence of OSA is increased at 41–58%[13] with the finding that their BMI does not correlate with their OSA severity.[2,34] In adolescent girls (15 years) with PCOS (n=31) compared with healthy obese girls without PCOS (n=19) neither group had significant OSA although total sleep time, percentage of REM sleep and sleep efficiency was lower in girls with PCOS.[33] Symptoms of PCOS usually begin in adolescence and perhaps OSA develops in a sub-group of females over time along with worsening insulin resistance. Thus, age might have been a protective factor for OSA in the group of women with PCOS and hyperandrogenism in the current study. A relationship between OSA severity with waist-to-hip ratio and elevated serum testosterone may over time contribute to the higher prevalence of OSA in women with PCOS.

The chronic reduction of REM sleep can lead to memory loss, failure to consolidate cognitive processes and metabolic disorders. Thus, untreated PCOS can have long term effects and cause other health problems, in addition to infertility. The findings show that the consequences of the disease were associated with damage to subjective sleep quality. Regarding objective aspects of sleep, the lower percentage of REM sleep in women with PCOS sleep could explain the perception of poor quality sleep in this group. Suppression of REM sleep can jeopardize women's health by damaging long term memory, increasing pain sensitivity and weight gain.[35-37] The reduction of REM sleep observed in women with PCOS (Table 2) does not seem to be related to PCOS, but due to increased BMI, snoring, high-risk group classification in the Berlin Questionnaire and frequency of women diagnosed with OSA. The current knowledge of the pathophysiology of PCOS has no evidence of the influence of the disease on sleep architecture.[38,39]

The negative correlation between sleep efficiency, N3 sleep stage and BMI demonstrated that increased body weight affects the distribution of sleep stages and hinders the deepening of sleep. Simultaneously, increased BMI showed a positive correlation with N1 stage sleep, wakefulness after sleep onset, arousals and apnea-hypopnea index. These results demonstrate a poor quality, fragmented superficial sleep. As a result, oxygen saturation levels were decreased significantly.

The statistical analysis of the sleep questionnaire shows that the PCOS group are at increased risk for presence of obstructive sleep apnea syndrome. Also, the self-perception of snoring during sleep can be a complementary signal in the clinical assessment of sleep. The frequency of women considered poor sleepers was significantly higher in the PCOS group. The increase in the prevalence of obstructive sleep apnea in PCOS subjects was associated with changes in sex hormones (increased androgens and/or decreased estrogens) and increased visceral adiposity.[13]

Considering only the women with PCOS, this study indicates that there were no differences regarding the risk for OSA in the analysis of the effect of hyperandrogenism. The results show that the

group with hyperandrogenism was younger than those in the group without hyperandrogenism. The adjustments for obesity and age were essential to exclude the influence of potential confounding factors. Also, the subjective and objective parameters of sleep did not differ between groups. This finding reveals that higher testosterone levels do not impair women's sleep quality. The initial hypothesis that testosterone could be responsible for snoring, sleep fragmentation, and respiratory disorder is not supported by our findings.[40] Other studies support the fact that women's sleep quality is not associated with increased testosterone, but that female hormones actually have a protective effect.[41] Progesterone increases respiratory drive and the action of the dilator muscles of the upper airway;[42,43] corroborating the suggestion that hormone therapy in postmenopausal women can act as a protective factor against to the obstructive sleep apnea syndrome.

Exogenous administration of testosterone has been shown to induce sleep apnea events in women.[44] The adverse effects of testosterone therapy on sleep cause a shortened sleep, worsened sleep apnea, and increased hypoxemia.[4] Testosterone increases baseline ventilation during wakefulness, altering the apneic threshold and increasing ventilator sensitivity to CO2 during sleep in healthy women.[3,5,45] However, a randomized, double-blind, placebo-controlled study demonstrated that impaired sleep quality as a consequence of sleep-disordered breathing was fleeting in the first weeks of daily administration of testosterone and that testosterone does not have a long-term effect.[46,47] A large epidemiological study, The Seattle Midlife Women's Health Study, found no significant association between disruption in sleep and testosterone, merely observing a negative trend.[48] Therefore all women with PCOS should undergo a sleep evaluation as there does seem to be a link between PCOS and sleep problems.[49]

In addition to hyperandrogenism and ovulatory dysfunction, PCOS may cause other common characteristics, such as abnormal gonadotrophin secretion, insulin resistance, and dyslipidemia. Insulin resistance and hyperinsulinemia are relevant pathophysiological consequences of the disease, affecting up to 75% of women with the syndrome.[50] Regarding the treatment of PCOS, the first choice is to adopt healthy lifestyle habits, such as dietary reeducation and physical exercise. As a result, weight loss potentially favors the fall of circulating androgens, improving lipid profile, reducing peripheral insulin resistance and regularization of ovulatory function. The prescription of low-dose oral hormonal contraceptives promotes control of menstrual irregularity and reduced risk of endometrial cancer.[51]

Some limitations of the study need to be considered. Biochemical analysis of testosterone was performed only in the sample PCOS women but not in the control group (healthy) because of the absence of clinical criteria for the disease. The analysis of both groups could provide a comparative assessment of the hormone levels in PCOS and hyperandrogenism. Despite these limitations, this study reveals that REM sleep time was reduced in women with PCOS.

CONCLUSIONS

Our results indicate that PCOS impairs subjective and objective sleep quality, due to reduced REM sleep time. Hyperandrogenism, characterized by higher free testosterone levels, did not have any effect on sleep-related parameters. Therefore, the findings confirm the hypothesis that women's sleep is mainly affected by obesity.

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The authors would like to thank all the women who took part in the study.

RESUMO

OBJETIVO: *Este estudo objetivou avaliar o sono de mulheres com síndrome do ovário policístico, com e sem hiperandrogenismo, em comparação com um grupo controle saudável, e estudar os efeitos do hiperandrogenismo e da obesidade nos parâmetros do sono.*

MÉTODOS: *Um total de 44 voluntárias foram recrutadas para participar do estudo. Os parâmetros clínicos, bioquímicos e polissonográficos e foram usados para diagnosticar SOP e hiperandrogenismo. A avaliação da qualidade de sono foi feita usando questionários validados e o exame polissonográfico. A frequência de síndrome da apneia obstrutiva também foi comparada entre os grupos.*

RESULTADOS: *O estudo revelou que mulheres com SOP apresentaram menor qualidade de sono subjetiva, incidência aumentada de ronco e maior risco para síndrome da apneia obstrutiva, baseada no questionário de Berlin. Ademais, após o ajuste para índice de*

massa corpórea, mulheres com SOP tiveram menor tempo de sono REM do que aquelas do grupo controle. Dentre as mulheres com SOP, aquelas com hiperandrogenismo não tiveram diferenças em nenhuma variável do sono. Mulheres com síndrome da apneia obstrutiva foram diagnosticadas no grupo SOP.

CONCLUSÕES: *Nossos resultados indicam que a SOP afeta a qualidade subjetiva de sono, bem como a qualidade objetiva e do sono, em razão da redução do tempo de sono REM em mulheres diagnosticadas com a síndrome. A obesidade afetou parâmetros relacionados ao sono, mas o hiperandrogenismo não teve efeito. A síndrome da apneia obstrutiva somente foi diagnosticada em mulheres com SOP.*

PALAVRAS-CHAVES: *hormonal; hiperandrogenismo; síndrome do ovário policístico; sono; mulheres.*

REFERENCES

1. Vgontzas AN, Papanicolaou DA, Bixler EO, et al. Sleep apnea and daytime sleepiness and fatigue: relation to visceral obesity, insulin resistance, and hypercytokinemia. *J Clin Endocrinol Metab* 2000;85:1151-8.
2. Fogel RB, Malhotra A, Pillar G, Pittman SD, Dunaif A, White DP. Increased prevalence of obstructive sleep apnea syndrome in obese women with polycystic ovary syndrome. *J Clin Endocrinol Metab* 2001;86:1175-80.
3. Behan M, Wenninger JM. Sex steroidal hormones and respiratory control. *Respir Physiol Neurobiol* 2008;164:213-21.
4. Andersen ML, Alvarenga TF, Mazaro-Costa R, Hachul HC, Tufik S. The association of testosterone, sleep, and sexual function in men and women. *Brain Res* 2011;1416:80-104.
5. Zhou XS, Rowley JA, Demirovic F, Diamond MP, Badr MS. Effect of testosterone on the apneic threshold in women during NREM sleep. *J Appl Physiol* 2003;94:101-7.
6. International classification of sleep disorders. *Diagnostic and coding manual (ICSD-3)*. Westchester, IL: American Academy of Sleep Medicine Task Force (AASM); 2014.
7. Polesel DN, Hirotsu C, Nozoe KT, et al. Waist circumference and postmenopause stages as the main associated factors for sleep apnea in women: a cross-sectional population-based study. *Menopause* 2015;22:835-44.
8. Tufik S, Santos-Silva R, Taddei JA, Bittencourt LR. Obstructive sleep apnea syndrome in the Sao Paulo Epidemiologic Sleep Study. *Sleep Med* 2010;11:441-6.
9. Ye L, Pien GW, Weaver TE. Gender differences in the clinical manifestation of obstructive sleep apnea. *Sleep Med* 2009;10:1075-84.
10. Spiegel K, Leproult R, Van Cauter E. Impact of sleep debt on metabolic and endocrine function. *Lancet* 1999;354:1435-9.
11. Leproult R, Van Cauter E. Role of sleep and sleep loss in hormonal release and metabolism. *Endocr Dev* 2010;17:11-21.
12. Mokhlesi B, Scoccia B, Mazzone T, Sam S. Risk of obstructive sleep apnea in obese and nonobese women with polycystic ovary syndrome and healthy reproductively normal women. *Fertil Steril* 2012;97:786-91.
13. Tasali E, Van Cauter E, Ehrmann DA. Polycystic Ovary Syndrome and Obstructive Sleep Apnea. *Sleep Med Clin* 2008;3:37-46.
14. Nandalike K, Strauss T, Agarwal C, et al. Screening for sleep-disordered breathing and excessive daytime sleepiness in adolescent girls with polycystic ovarian syndrome. *J Pediatr* 2011;159:591-6.
15. Nitsche K, Ehrmann DA. Obstructive sleep apnea and metabolic dysfunction in polycystic ovary syndrome. *Best Pract Res Clin Endocrinol Metab* 2010;24:717-30.
16. Tock L, Carneiro G, Togeiro SM, et al. Obstructive sleep apnea predisposes to nonalcoholic Fatty liver disease in patients with polycystic ovary syndrome. *Endocr Pract* 2014;20:244-51.
17. Yang HP, Kang JH, Su HY, Tzeng CR, Liu WM, Huang SY. Apnea-hypopnea index in nonobese women with polycystic ovary syndrome. *Int J Gynaecol Obstet* 2009;105:226-9.
18. Group REA-SPCW. Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome. *Fertil Steril* 2004;81:19-25.
19. Rechtschaffen A, Kales A. *A manual of standardized terminology, techniques and scoring system for sleep stages of human subjects*. Los Angeles, CA: Brain Information Service/Brain Research Institute, UCLA; 1968.
20. Iber C, Ancoli I, Cheeson A, Quan S. *The AASM manual for scoring of sleep associated events: rules, terminology and techical specifications*. Wetchester, IL: American Academy of Sleep Medicine; 2007.
21. *Sleep-related breathing disorders in adults: recommendations for syndrome definition and measurement techniques in clinical research*. The Report of an American Academy of Sleep Medicine Task Force. Sleep 1999;22:667-89.
22. Buysse DJ, Reynolds CF, Monk TH, Berman SR, Kupfer DJ. The Pittsburgh Sleep Quality Index: a new instrument for psychiatric practice and research. *Psychiatry Res* 1989;28:193-213.
23. Bertolazi AN, Fagondes SC, Hoff LS, et al. Validation of the Brazilian Portuguese version of the Pittsburgh Sleep Quality Index. *Sleep Med* 2011;12:70-5.
24. Netzer NC, Stoohs RA, Netzer CM, Clark K, Strohl KP. Using the Berlin Questionnaire to identify patients at risk for the sleep apnea syndrome. *Ann Intern Med* 1999;131:485-91.
25. Massierer D, Martinez D, Fuchs SC, et al. Obstructive sleep apnea, detected by the Berlin Questionnaire: an associated risk factor for coronary artery disease. *Cad Saude Publica* 2012;28:1530-8.
26. Johns MW. A new method for measuring daytime sleepiness: the Epworth sleepiness scale. *Sleep* 1991;14:540-5.
27. Vermeulen A, Verdonck L, Kaufman JM. A critical evaluation of simple methods for the estimation of free testosterone in serum. *J Clin Endocrinol Metab* 1999;84:3666-72.
28. Esmaeilzadeh S, Andarieh MG, Ghadimi R, Delavar MA. Body mass index and gonadotropin hormones (LH & FSH) associate with clinical symptoms among women with polycystic ovary syndrome. *Glob J Health Sci* 2015;7:101-6.
29. de Sousa G, Schlüter B, Buschatz D, et al. The impact of insulin resistance and hyperandrogenemia on polysomnographic variables in obese adolescents with polycystic ovarian syndrome. *Sleep Breath* 2012;16:169-75.
30. Moran LJ, March WA, Whitrow MJ, Giles LC, Davies MJ, Moore VM. Sleep disturbances in a community-based sample of women with polycystic ovary syndrome. *Hum Reprod* 2015;30:466-72.
31. Tasali E, Van Cauter E, Ehrmann DA. Relationships between sleep disordered breathing and glucose metabolism in polycystic ovary syndrome. *J Clin Endocrinol Metab* 2006;91:36-42.
32. de Sousa G, Schlüter B, Menke T, Trowitzsch E, Andler W, Reinehr T. A comparison of polysomnographic variables between adolescents with polycystic ovarian syndrome with and without the metabolic syndrome. *Metab Syndr Relat Disord* 2011;9:191-6.
33. de Sousa G, Schlüter B, Menke T, Trowitzsch E, Andler W, Reinehr T. Relationships between polysomnographic variables, parameters of glucose metabolism, and serum androgens in obese adolescents with polycystic ovarian syndrome. *J Sleep Res* 2011;20:472-8.
34. Gopal M, Duntley S, Uhles M, Attarian H. The role of obesity in the increased prevalence of obstructive sleep apnea syndrome in patients with polycystic ovarian syndrome. *Sleep Med* 2002;3:401-4.
35. Roehrs T, Hyde M, Blaisdell B, Greenwald M, Roth T. Sleep loss and REM sleep loss are hyperalgesic. *Sleep* 2006;29:145-51.
36. Liu X, Forbes EE, Ryan ND, Rofey D, Hannon TS, Dahl RE. Rapid eye movement sleep in relation to overweight in children and adolescents. *Arch Gen Psychiatry* 2008;65:924-32.
37. Meerlo P, Mistlberger RE, Jacobs BL, Heller HC, McGinty D. New neurons in the adult brain: the role of sleep and consequences of sleep loss. *Sleep Med Rev* 2009;13:187-94.
38. Suri J, Suri JC, Chatterjee B, Mittal P, Adhikari T. Obesity may be the common pathway for sleep-disordered breathing in women with polycystic ovary syndrome. *Sleep Med* 2016;24:32-9.
39. Franik G, Krysta K, Madej P, et al. Sleep disturbances in women with polycystic ovary syndrome. *Gynecol Endocrinol* 2016;32:1014-7.
40. Cistulli PA, Grunstein RR, Sullivan CE. Effect of testosterone administration on upper airway collapsibility during sleep. *Am J Respir Crit Care Med* 1994;149:530-2.
41. Hachul H, Andersen ML, Bittencourt L, Santos-Silva R, Tufik S. A popu-

lation-based survey on the influence of the menstrual cycle and the use of hormonal contraceptives on sleep patterns in São Paulo, Brazil. *Int J Gynaecol Obstet* 2013;120:137-40.

42. Martins AB, Tufik S, Moura SM. Physiopathology of obstructive sleep apnea-hypopnea syndrome. *J Bras Pneumol* 2007;33:93-100.

43. Marcouiller F, Boukari R, Laouafa S, Lavoie R, Joseph V. The nuclear progesterone receptor reduces post-sigh apneas during sleep and increases the ventilatory response to hypercapnia in adult female mice. *PLoS One* 2014;9:e100421.

44. Johnson MW, Anch AM, Remmers JE. Induction of the obstructive sleep apnea syndrome in a woman by exogenous androgen administration. *Am Rev Respir Dis* 1984;129:1023-5.

45. Ahuja D, Mateika JH, Diamond MP, Badr MS. Ventilatory sensitivity to carbon dioxide before and after episodic hypoxia in women treated with testosterone. *J Appl Physiol* 2007;102:1832-8.

46. Killick R, Wang D, Hoyos CM, Yee BJ, Grunstein RR, Liu PY. The effects of testosterone on ventilatory responses in men with obstructive sleep apnea: a randomised, placebo-controlled trial. *J Sleep Res* 2013;22:331-6.

47. Melehan KL, Hoyos CM, Yee BJ, et al. Increased sexual desire with exogenous testosterone administration in men with obstructive sleep apnea: a randomized placebo-controlled study. *Andrology* 2016;4:55-61.

48. Woods NF, Smith-Dijulio K, Percival DB, Tao EY, Taylor HJ, Mitchell ES. Symptoms during the menopausal transition and early postmenopause and their relation to endocrine levels over time: observations from the Seattle Midlife Women's Health Study. *J Womens Health (Larchmt)* 2007;16:667-77.

49. Chatterjee B, Suri J, Suri JC, Mittal P, Adhikari T. Impact of sleep-disordered breathing on metabolic dysfunctions in patients with polycystic ovary syndrome. *Sleep Med* 2014;15:1547-53.

50. Junqueira PAA, Fonseca AM, Aldrighi JJ. Síndrome dos Ovários Policísticos. *Rev Assoc Med Bras* 2003; 49:13-4.

51. Lopes IM, Baracat MC, Simões MdeJ, Simões RS, Baracat EC, Soares Junior JM. Endometrium in women with polycystic ovary syndrome during the window of implantation. *Rev Assoc Med Bras* 2011;57:702-9.

ORIGINAL ARTICLE

Effects of eccentric exercise in pressure pain threshold in subjects with functional ankle equinus condition

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SUMMARY

Stretching exercises are widely used by the population before sporting activities. One of the most common technique is eccentric exercise. Here, we made a clinical examination of 98 subjects with equinus condition before activity and after 30 min of running (49 participants with previous eccentric exercise and 49 with no previously eccentric exercise). The clinical assessment of the Achilles tendon was based on the pressure pain threshold (PPT). We identified significant PPT changes between the previous eccentric stretching and the non-previous eccentric stretching group in the Achilles tendon evaluations. Based on our findings, we propose that subjects with equinus condition could use eccentric stretching in order to improve the Achilles tendon status.

KEYWORDS: *Ankle. Foot. Sports. Equinus Deformity.*

INTRODUCTION

Equinus is a clinical ankle condition that limits the dorsiflexion range of movement. Equinus could be explained as the "limitation of the ankle for dorsiflexion with the knee extended and/or flexed (excluding osseous-restriction)" [1,2]. Although Equinus is a non-symptomatic pathologic pattern, it may promote a clinical alteration in the Achilles tendon (AT) and triceps surae muscle. Equinus condition is deeply related to lower limb injuries (as anterior cruciate ligament rupture), hip pain, ankle sprain, asymmetric load patterns, and changes in muscle activity for the triceps surae [3-6].

Equinus condition could promote a higher basal activation for triceps surae and hamstring muscles. This clinical condition has been investigated using force-pressure platforms [7-10] and its relationships be-

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tween muscle activity patterns have been studied by electromyography [11-13]. Equinus condition is deeply related to gait, posture [9,10] and injuries. The activation of the triceps surae musculature could modify the mechanosensitivity of the Achilles tendon and promote other alterations[14-17]. Human posture is actively supported by the contraction of muscles as gluteus, hamstrings and triceps surae.

We aimed to assess with an algometer the PPT values for the Achilles tendon. The main aim of the study was to check PPT values between eccentric and non-eccentric stretching in subjects with Equinus before and after running exercise.

METHODS

This was an observational study; 98 healthy male subjects were recruited. A consecutive sampling method was used to select participants. A total of 49 subjects with eccentric stretching and 49 with non-eccentric stretching were recruited. All participants completed the study. The exclusion criteria were the presence of sprains, infection, bone alteration, musculoskeletal disorders, tendon injuries, low back and pelvic pain, scoliosis, and use of drugs in the week preceding the test.

The studied variable was PPT. The pain pressure threshold was measured with an algometer. Twenty eccentric stretching exercises were carried out before the activity. Subjects then ran for 30 minutes on a treadmill at a speed of 10 km/h, and the PPT assessment was repeated.

Sample size calculation

The sample size calculation was carried out by the difference between two independent groups with the G*Power statistic tool and based on the right-Achilles tendon PPT of a case-control pilot study with 2-groups [mean (SD)], n = 10 subjects with Equinus condition [1.60 (0.57) kg/cm2] and n = 10 subjects without Equinus condition [1.98 (0.57) kg/cm]. Indeed, 1 tail hypothesis, an effect size of 0.67, an α-error of 0.05, and a power of 0.95 were used for the sample size calculation. Therefore, a total sample size of 98 subjects, 49 for each group, was obtained.

Ethical considerations

The Ethics Committee of the HULP (Madrid, Spain; record number: CE 2828A) approved the study. All participants provided signed informed consent before the beginning of the study. The ethical standards for human experimentation of the Declaration of Helsinki and the Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) guidelines were applied.

Clinical exploration

First, subjects lay in a supine position, and their Equinus condition was assessed with the knee extended/ flexed. The range of movement was checked using a goniometer to evaluate the angle between the plantar line of the foot and the tibia bone axis. The PPT was checked using an algometer before and after activity. The Equinus assessment and PPT was carried out by the same Podiatry Doctor (D.R.S) in order to ensure the reliability of measurements.

FIGURE 1

STATISTICAL ANALYSIS

Statistical analyses were carried out with SPSS (version 22.0 for Windows, IBM SPSS Statistics for Windows, Armonk, NY, IBM Corp) with an α error of 0.05 (95% confidence interval [CI]), with the desired power of 80% (β error of 0.2).

The Kolmogorov-Smirnov test was used to assess data normality. All data were normally distributed, and the parametric statistical tests were selected. The mean and standard deviation of the temperature data were obtained for the selected lower limb muscles Achilles tendon.

Unpaired sample Student's t-test were performed to test for statistically significant differences in height, weight, body mass index, and age between the two groups. Paired Student's t-tests were performed to determine differences between the groups (Eccentric and non-eccentric stretching) as well as between sessions (before and after running).

RESULTS

We found no statistically significant differences between the eccentric and non-eccentric groups for participant height, weight, age, or body mass index (Table 1).

TABLE 1. PARTICIPANT CHARACTERISTICS (N = 49 ECCENTRIC STRETCHING AND 49 NON-ECCENTRIC STRETCHING)

	Eccentric exercise group	Non-eccentric group
Age (years)*	34.56 ± 3.7	36.2 ± 2.4
Height (cm)*	174.5 ± 5.3	168.1 ± 4.9
Weight (kg)*	71.8 ± 2.1	70.2 ± 1.9
Body mass index*	21.1 ± 1.2	20.8 ± 1.4

* No statistically significant difference between groups ($p \geq 0.05$).

We found significant differences for pressure pain threshold in the Achilles tendon between both groups (eccentric and non-eccentric stretching exercises) after the activity (Table 2).

DISCUSSION

Here, we identified that the PPT values were also lower for the non-eccentric stretching group vs. the eccentric stretching participants.

The triceps surae requires stronger activation in subjects with Equinus condition, therefore, might be affected early by pain or fatigue, thus explaining why we observed an increase in PPT values for the eccentric stretching participants compared with the non-eccentric stretching participants. In the assessment before running, we did not find any significant differences in the PPT values. The running exercise may serve as a stimulus to increase PPT values in muscles and consequently differences between groups.

TABLE 2. PRESSURE PAIN THRESHOLD ASSESSMENT

	PPT Before Running	PPT After Running
	Mean (SD)	Mean (SD)
Eccentric stretching	2.7 (0.22)	3.5 (0.33)
Non-eccentric stretching	2.5 (0.14)	2.8 (0.13)
p-value	0.17*	0.01†

*No statistically significant difference between groups before running. † Statistically significant difference between groups before running

Eccentric stretching exercise may assist triceps surae extensibility during loading and running and promote venous return. Biomechanically, the maximum ankle dorsiflexion during the stance-phase of a normal gait occurs before heel lift with the knee completed extended.[1] The most deeply known range of movement values for ankle dorsiflexion in the reviewed literature for static evaluation is the minimum dorsiflexion movement of the ankle for normal gait, which is 10° of motion.[3, 18-23]

Further studies are necessary to improve our knowledge of muscle condition and establish the clinical relevance of the association between PPT in non-equinus subjects [25,26]. Based on our findings, we propose that eccentric stretching exercise be recommended for runners with GSE condition.

CONCLUSIONS

The eccentric stretching group showed a higher PPT-value for the Achilles tendon after exercise than the non-eccentric group. Therefore, an algometer that measures pain pressure threshold could be promoted as a screening tool for prevention or therapeutic actions. Based on our findings, we propose that eccentric exercise be recommended for subjects with GSE condition

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RESUMO

Exercícios de alongamento são amplamente utilizados pela população antes da atividade esportiva. Uma das técnicas mais comuns é o exercício excêntrico. Aqui, fizemos um exame clínico de 98 indivíduos com condição de pé equino antes da atividade e após 30 minutos de corrida (49 corredores com exercício excêntrico anterior e 49 sem exercício excêntrico anterior). A avaliação clínica do tendão de Aquiles foi baseada no limiar de dor à pressão (PPT). Identificamos modificações significativas no PPT entre alongamentos prévios excêntricos e nenhum exercício anterior excêntrico de alongamento para as avaliações do tendão de Aquiles. Com base em nossos achados, propomos que sujeitos com condição de pé equino poderiam fazer alongamentos com exercícios excêntricos para melhorar o status do tendão de Aquiles.

PALAVRAS-CHAVE: *Tornozelo. Pé. Esportes. Pé equino.*

REFERENCES

1. DiGiovanni CW, Kuo R, Tejwani N, Price R, Hansen ST Jr, Cziernecki J, et al. Isolated gastrocnemius tightness. J Bone Joint Surg Am. 2002;84-A(6):962-70.
2. Downey MS, Banks AS. Gastrocnemius recession in the treatment of nonspastic ankle equinus. A retrospective study. J Am Podiatr Med Assoc. 1989;79(4):159-74.
3. Lamm BM, Paley D, Herzenberg JE. Gastrocnemius soleus recession: a simpler, more limited approach. J Am Podiatr Med Assoc. 2005;95(1):18-25.
4. Silfverskiold N. Reduction of the uncrossed two-joints muscles of the leg to one-joint muscles in spastic conditions. Acta Chir Scand. 1924;56:315-30.
5. Downey MS. "Ankle equinus". In: McGlamry ED, Banks AS, Downey MS, eds. Comprehensive textbook of foot surgery. 2nd ed. vol. 1. Baltimore: Williams & Wilkins; 1992. p.687.
6. Root ML, Orien WP, Weed JH. Normal and abnormal function of the foot. Los Angeles: Clinical Biomechanics Corp; 1977.
7. Blustein SM, D'Amico JC. Limb length discrepancy. Identification, clinical significance, and management. J Am Podiatr Med Assoc. 1985;75(4):200-6.
8. Mahar RK, Kirby RL, MacLeod DA. Simulated leg-length discrepancy: its effect on mean center-of-pressure position and postural sway. Arch Phys Med Rehabil. 1985;66(12):822-4.
9. Bhave A, Paley D, Herzenberg JE. Improvement in gait parameters after lengthening for the treatment of limb-length discrepancy. J Bone Joint Surg Am. 1999;81(4):529-34.
10. Blake RL, Ferguson H. Limb length discrepancies. J Am Podiatr Med Assoc. 1992;82(1):33-8.
11. Vink P, Huson A. Lumbar back muscle activity during walking with a leg inequality Acta Morphol Neerl Scand. 1987;25(4):261-71.
12. Gurney B, Mermier C, Robergs R, Gibson A, Rivero D. Effects of limb-length discrepancy on gait economy and lower-extremity muscle activity in older adults. J Bone Joint Surg Am. 2001;83-A(6):907-15.
13. Balestra G, Frassinelli S, Knaflitz M, Molinari F. Time-frequency analysis of surface myoelectric signals during athletic movement. IEEE Eng Med Biol Mag. 2001;20(6):106-15.
14. Merla A, Iodice P, Tangherlini A, De Michele G, Di Romualdo S, Saggini R, et al. Monitoring skin temperature in trained and untrained subjects throughout thermal video. Conf Proc IEEE Eng Med Biol Soc. 2005;2:1684-6.
15. Zontak A, Sideman S, Verbitsky O, Beyar R. Dynamic thermography: analysis of hand temperature during exercise. Ann Biomed Eng. 1998;26(6):988-93.
16. Ammer K, Formenti D. Editorial: Does the type of skin temperature distribution matter? Thermol Internat. 2016;26(2):51-4.
17. Formenti D, Ludwig N, Rossi A, Trecoci A, Alberti G, Gargano M, et al. Skin temperature evaluation by infrared thermography: comparison of two image analysis methods during the nonsteady state induced by physical exercise. Infrared Physics and Technology. 2017;81:32-40.
18. McGlamry ED, Kitting RW. Aquinus foot: an analysis of the etiology, pathology and treatment techniques. J Am Podiatry Assoc. 1973;63(5):165-84.
19. Knutzen KM, Price A. Lower extremity static and dynamic relationships with rearfoot motion in gait. J Am Podiatr Med Assoc. 1994;84(4):171-80.
20. Nuber GW. Biomechanics of the foot and ankle during gait. Clin Sports Med. 1988;7(1):1.
21. Lavery LA, Armstrong DA, Boulton AJM; Diabetex Research Group. Ankle equines deformity and its relationship to high plantar pressure in a large population with diabetes mellitus. J Am Podiatr Med Assoc. 2002;92(9):479-82.
22. Wrobel JS, Connolly JE, Beach ML. Associations between static and functional measures of joint function in the foot and ankle. J Am Podiatr Med Assoc. 2004;94(6):535-41.
23. Winter DA. Kinematic and kinetic patterns in human gait: variability and compensating effects. Human Mov Sci. 1984;3:51-76.
24. Brodersen A, Pedersen B, Reimers J. Foot deformities and relation to the length of leg muscles in Danish children aged 3-17 years. Ugeskr Laeger. 1993;155(48):3914-6.
25. Abate M, Carlo LD, Romualdo SD, Ionta S, Ferretti A, Romani GL, et al. Postural adjustment in experimental leg length difference evaluated by means of thermal infrared imaging. Physiol Meas. 2010;31(1):35-43.
26. Bernardo WM, Meleiro SAS, Mendes VTA, Kaleka CC, Cury RPL. Update of treatment of isolated lesions of the posterior cruciate ligament. Rev Assoc Med Bras. 2015;61(2):100.

ORIGINAL ARTICLE

Positive outcomes of phosphodiesterase type 5 inhibitor on histopathologic and biochemical changes induced by ureteral obstruction

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SUMMARY

OBJECTIVES: *We examined the effects of tadalafil, one of the phosphodiesterase type 5 (PDE5) inhibitors, in a rat model of with partial and complete unilateral ureteral obstruction (UUO).*

METHODS: *The rats were divided into 5 groups: sham (n=6), partial unilateral ureteral obstruction (PUUO, n=6), PUUO with tadalafil treatment (PUUO+T; Cialis, 10 mg/72 h, intragastric; Lilly, Indianapolis, Indiana, USA), complete unilateral ureteral obstruction (CUUO, n=6), and CUUO with tadalafil treatment (CUUO+T).*

RESULTS: *Fifteen days after the UUO, the ureter presented changes in the layers of urothelium and significant infiltration of inflammatory cells in the PUUO and CUUO groups. Compared with the sham, PUUO and CUUO groups had severe increased inflammatory cell infiltration. The urothelial epithelium exhibited cell degeneration and loss because of the swollen, atrophic, and denuded epithelial cells in the PUUO and CUUO groups. In the PUUO+T and CUUO+T groups, the urothelium revealed less epithelial cell degeneration and loss.*

The expressions of α-smooth muscle actin (α-SMA) and **transforming growth factor**-β *(TGF–β) exhibited up-regulation in the PUUO and CUUO groups. The expression of TGF-β decreased positively correlated with that of α-SMA in the tadalafil therapy groups, PUUO+T and CUUO+T.*

CONCLUSION: *The phosphodiesterase type 5 inhibitor's tadalafil reduced expressions of α-SMA and TGF-β in the obstructed ureters, measured by biochemical examinations. In addition, tadalafil decreased urothelium degeneration due to the decreased epithelial cell loss and inflammatory cell infiltration. Our results show that tadalafil prevents or slows down the onset of ureter inflammation and urothelial degeneration in rats with UUO.*

KEYWORDS *Phosphodiesterase type 5 inhibitor, Tadalafil, obstruction, histopathologic and biochemical*

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INTRODUCTION

Ureteric obstruction, one of the most common diseases in the urinary tract, can lead to obstructive uropathy [1]. During ureteric obstruction, the dilated ureter presents hypertrophy of the ureteric smooth muscle and proliferation of connective tissues [2,3].

UUO, a commonly used experimental model of chronic kidney injury is characterized by tubular atrophy, inflammation, and interstitial fibrosis[4,5]. Unilateral ureteral obstruction causes significant tubulointerstitial fibrosis in the kidney. Ureteric obstruction, both complete and incomplete, can lead to severe ureteric injury[1]. Nevertheless, the degeneration, proliferation, and regeneration of the urethelial layer in the obstructed ureters have not been well documented and fully understood[6].

Renal fibrosis is a common feature of various kidney diseases leading to end-stage renal failure[7]. Epithelial-to-mesenchymal transition contributes to renal fibrosis in chronic kidney disease[8]. Transforming growth factor-β (TGF-β), one of the profibrotic growth factors produced by epithelial cells and macrophages, plays a crucial role in the pathogenesis of tissue fibrosis[9,10].

The myofibroblasts are major contributors to the increased extracellular matrix deposition seen in kidney fibrosis[11]. Fibrosis is characterized by the accumulation of myofibroblasts defined by the expression of α-smooth muscle actin (α-SMA). Fibroblast differentiation to the myofibroblast phenotype is associated with α-SMA expression and regulated by cytokines. The TGF-β can stimulate and inhibit myofibroblast differentiation [12]. Many studies demonstrate that renal tubular cells can be converted to myofibroblasts during epithelial-mesenchymal transition stimulated by TGF-β[13]. The TGF-β promoted α-SMA expression or myofibroblast differentiation[12].

Tadalafil is one of the most commonly used phosphodiesterase type 5 (PDE5) inhibitors 14. It has a markedly different molecular structure than sildenafil and vardenafil, and these structural differences have implications for the selectivity and pharmacokinetics of the three PDE5 inhibitors 15. In 2011, the first experimental study to use tadalafil in the kidney during ischemia was published, with a finding of a decrease in leukocyte infiltration in animals that used this medication 16. However, the effect of tadalafil on inflammation and the urothelial layer in the ureter has not been studied.

We examined the effects of tadalafil one from PDE5 inhibitors in a rat model of partial and complete unilateral ureteral obstruction in the ureter. The histopathologic and biochemical changes in obstructed ureters were determined using Hematoxyline and Eosin staining and tissue analysis of α-SMA and TGF-β in Sprague-Dawley rats.

METHODS

The study comprised male Sprague-Dawley rats (body weight 250-300 g). All experimental protocols followed the principles of the Declaration of Helsinki and received full approval from the Animal Ethical Committee of the Ordu University, Turkey.

The rats were divided into five groups, sham-operated (n=6); partial unilateral ureteral obstruction (PUUO, n=6); and PUUO with tadalafil treatment (PUUO+T; Cialis, 10 mg/72 h, intragastric; Lilly, Indianapolis, Indiana, USA); complete unilateral ureteral obstruction (CUUO, n=6); and CUUO with tadalafil treatment (CUUO+T).

The unilateral ligation of the left ureters was performed in the ureteral obstruction groups, as described previously [1]. Briefly, under general anesthesia (ketamine, 50 mg/kg, im), laparotomy was performed on the ureter. The PUUO and PUUO+T groups were then received a 24-gauge intravenous catheter into the proximal ureter, and the ureter and catheter around the ureter were connected using a 4-0 silk suture. The remainder ureters were subsequently narrowed by placing a ligature over the catheter. After partial ureteral obstruction, the abdominal incision was closed. In the CUUO and CUUO+T groups, the left ureter was ligated with silk at two points and cut between the ligatures. After ureteric ligation, the surgical incision was closed.

The sham rats underwent the same procedure, except that the left ureters were not ligated. The rats were killed for examination 15 days after the ligation. The ureters were removed and divided; some portions were stored at -80 °C for tissue analysis, while others were fixed in 4% paraformaldehyde. Following fixation, the tissues were embedded in paraffin, and about 5 µm sections were cut for histopathologic examinations.

HISTOPATHOLOGIC EXAMINATION

The proximal portion of the ureter was prepared for light microscopy by staining with hematoxylin

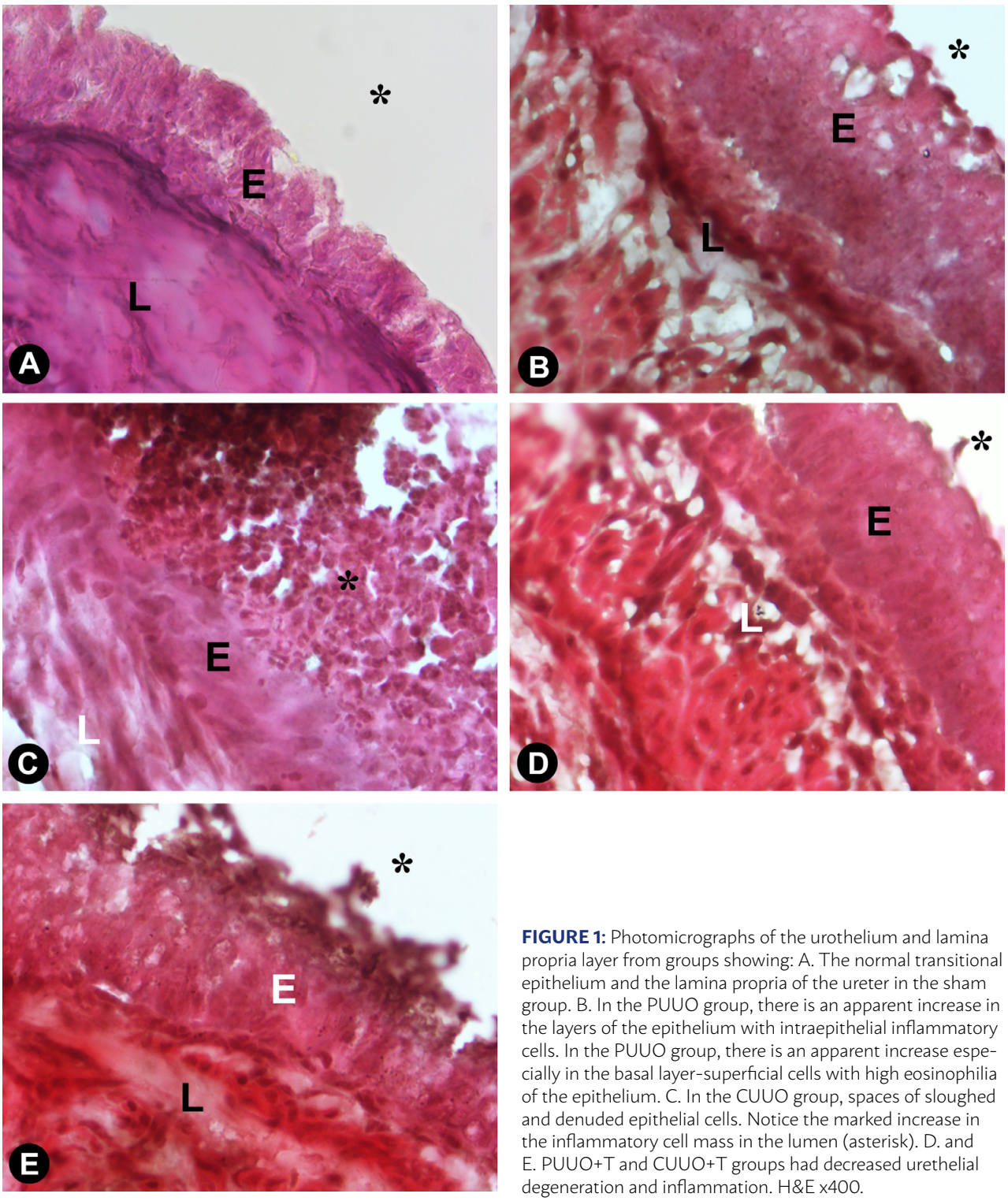


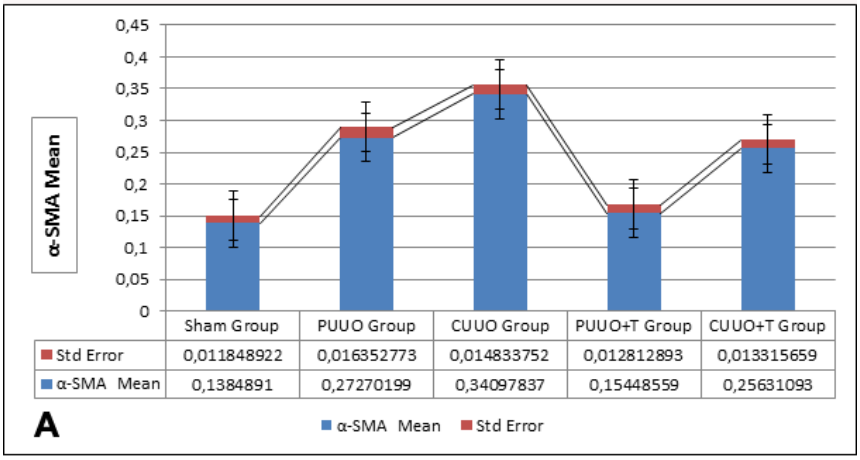
FIGURE 1: Photomicrographs of the urothelium and lamina propria layer from groups showing: A. The normal transitional epithelium and the lamina propria of the ureter in the sham group. B. In the PUUO group, there is an apparent increase in the layers of the epithelium with intraepithelial inflammatory cells. In the PUUO group, there is an apparent increase especially in the basal layer-superficial cells with high eosinophilia of the epithelium. C. In the CUUO group, spaces of sloughed and denuded epithelial cells. Notice the marked increase in the inflammatory cell mass in the lumen (asterisk). D. and E. PUUO+T and CUUO+T groups had decreased urethelial degeneration and inflammation. H&E x400.

and eosin (H&E) for morphological evaluations.

The loss of epithelial cells was assessed by the total number of urothelial nuclei in a selected field (magnification x400). The mean value was calculated from randomly 2 selected fields per ureter in the 6 animals for each group [17].

The ureter sections were acquired systematically and sampled randomly. They were scored depending on the quantity of the inflammatory cell infiltration as follows: score 0: no cells; score 1: a few cells; score 2: many cells; and score 3: many cells in the in the ureter [18].

Finally, images of the stained sections were captured with a Leica DFC295 HD color digital camera mounted on a Leica DM2500 microscope and stored as Tagged Image File Format images.



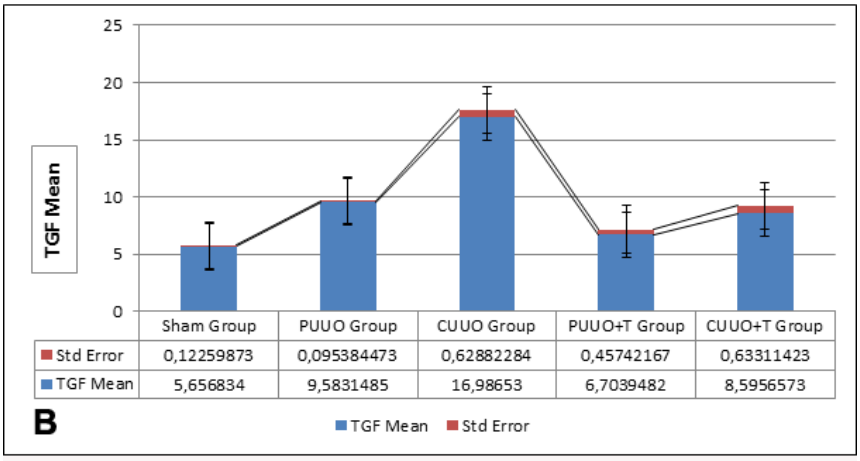


FIGURE 2: A. Tissue analysis of α –smooth muscle (α-SMA) and B. Transforming growth factor-β (TGF-β) expressions in ureters using ELISA. The α-SMA and TGF-β expression levels were up-regulated in the PUUO and CUUO groups; however, they were down-regulated in the PUUO+T and CUUO+T groups (* P < 0.0001 versus control).

BIOCHEMICAL EXAMINATIONS

Tissue analyzes of α-SMA and TGF-β were performed in the ureter tissue homogenates (Witeg Labortechnik GmgH, WiseTis HG-15D Homogenizer). After two freeze-thaw cycles, the homogenates were centrifuged for 5 minutes at 5000 g (2-8 °C). In the supernates, α-SMA and TGF-β levels were assayed using appropriate enzyme-linked immunosorbent assay (ELISA) kits (Cusabio CSB-E14027r and Boster EK0514, respectively). The analyses were performed according to the product instruction and read using an absorbance microplate reader (BioTek, ELx800). The concentration of total proteins in the homogenates was determined by the Bradford method using Coomassie reagent (Thermo scientific; 23200) and bovine serum albumin as standard.

Statistics

Data were expressed as mean ± standard error of the mean (SEM) and analyzed using analysis of variance (ANOVA) for comparison among multiple groups using the Tukey post-test analysis for comparison and KaleidaGraph 4.0 software. All p-values lower than 0.05 were considered to be statistically significant.

RESULTS

The histopathologic examination using H&E staining showed normal urothelium and lamina propria layer in the ureters of the sham group. The urothelium of the ureter was histologically normal in the sham group. The normal ureter was lined by three to four cells deep transitional epithelium and no inflammatory cell infiltration (score 0, Figure 1A).

The PUUO group exhibited inflammatory cell infiltration (score 2), severe epithelial atrophy, and swelling of the epithelial cells. In the PUUO group, the epithelium was thickened and lined by three to eight-layered cells (P < 0.0001). In the PUUO group, vacuoles appeared in the squamous epithelial cells (Figure 1B).

In the CUUO group, the urothelium was partially or completely denuded with a loss of epithelial cells. The epithelial cells had irregularly shaped and darkly stained pyknotic nuclei. In the CUUO group, the epithelium became thinner, down to 2-4 layers (P <

0.0001), and the cells were exposed to the ureteral lumen. The CUUO group exhibited epithelial hemorrhage as well as infiltration of inflammatory cells (score 3) into the urothelium and lamina propria layer. The urothelium, interstitial spaces, and ureteral lumen were filled with large macrophages (Figure 1C).

The PUUO+T group exhibited attenuated inflammatory cell infiltration (score 1) and less swelling of the epithelial cells compared with the PUUO group ($P < 0.0001$, Figure 1D). The inflammation was higher in the CUUO+T group than in the PUUO+T group. In the CUUO+T group, the inflammation was less significant than the CUUO group ((score 2, Figure 1D).

The PUUO and CUUO groups exhibited up-regulation of α-SMA and TGF-β ($P < 0.0001$). The PUUO+T and CUUO+T groups exhibited down-regulation of α-SMA and TGF-β ($P < 0.0001$). There was no significant difference between the expressions of the α-SMA and TGF-β in the control, PUUO+T and CUUO+T groups (Figure 2A and B).

DISCUSSION

Ureteric obstruction is one of the most common problems faced by urologists [19,20]. Ureteric obstruction, both complete and partial, can lead to severe ureteric injury [1]. Ureteric obstruction causes ureteric injury as well as renal hemodynamic and metabolic changes, leading to tubular injury and renal inflammation, characterized by macrophage infiltration. Infiltration of macrophages, causing a release of cytokines to induce tubular cell apoptosis, activate and increase proliferation of fibroblasts [5,21]. Upon recruitment and activation, macrophages produce various pro-inflammatory cytokines, such as TGF-β, which in turn promote expression of adhesion molecules and contribute to further recruitment of circulating inflammatory cells [21,22]. The present study demonstrated that there was an increase in inflammatory cell infiltration and degenerative changes of epithelial cells in the obstructed ureters. These results agree with the clinical changes during hydroureter [23] and those of other animal studies [2,3].

Several studies have shown that TGF-β stimulates α-SMA expression and tissue fibrosis in both experimental and human kidney disease. The blocking of TGF-β protects mice with diabetes from renal dysfunction, especially glomerular hypertrophy and fibrosis [9,24]. In the present study, the tadalafil therapy groups exhibited decreased inflammatory cell infiltration in the ureters. Our results also indicate that partial and complete unilateral ureteral obstruction led to a marked the up-regulation of α-SMA and TGF-β, whereas the tadalafil treatment moderately improved these effects in the ureters.

The first experimental study with tadalafil in ischemia was performed in cardiomyocytes [25], in which the use of tadalafil prior to coronary occlusion in rats led to improved endothelial function and decreased ischemic area in myocardial infarction. Several clinical studies have shown favorable safety and efficacy profiles of tadalafil for men with erectile dysfunction and diabetes [26], hypertension, or heart disease [27,28]. The present study showed that tadalafil therapy decreases atrophy, vacuoles, swelling, and pyknotic nuclei in epithelial cells.

CONCLUSIONS

Our results suggest that tadalafil merits further exploration as a therapeutic agent in the prevention and treatment of ureteral obstruction disease. T ameliorate by the tadalafil, one of the PDE5 inhibitors, can have positive outcomes on ureteral obstruction. More investigation needs to be carried out, but the early evidence is promising for different doses and combination therapy.

Funding

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Conflict of Interest

The authors declared no potential conflicts of interest concerning the research, authorship, and/or publication of this article.

RESUMO

OBJETIVOS: *Examinamos os efeitos do tadalafil em um dos inibidores da fosfodiesterase tipo 5 (PDE5) em um modelo de rato com obstrução ureteral unilateral parcial e completa (UUO).*

MÉTODOS: *Os ratos foram divididos em cinco grupos: sham (n = 6), obstrução ureteral unilateral parcial (PUUO, n = 6), PUUO com tadalafil (PUUO T; Cialis, 10 mg/72 h, intragástrica; Lilly, Indianapolis, Indiana, EUA), completa obstrução ureteral unilateral (CUUO, n = 6) e CUUO com tratamento com tadalafil (CUUO T).*

RESULTADOS: *Quinze dias após a UUO, o ureter apresentou alterações nas camadas de urotélio e infiltração significativa de células inflamatórias nos grupos PUUO e CUUO. Em comparação com os grupos sham, PUUO e CUUO, houve um aumento grave da infiltração de células inflamatórias. O epitélio urotelial exibiu degeneração e perda celular devido às células epiteliais inchadas, atróficas e desnudas nos grupos PUUO e CUUO. Nos grupos PUUO T e CUUO T, o urotélio revelou menor degeneração e perda de células epiteliais. Nós mostramos que a expressão da actina do músculo liso-α (α-SMA) e do fator de crescimento transformador-β (TGF-β) foram exibidas como sub-regulação nos grupos PUUO e CUUO. A expressão do TGF-β foi diminuída positivamente correlacionada com a da α-SMA nos grupos de terapia com tadalafil, PUUO T e CUUO T.*

CONCLUSÃO: *O tadalafil do inibidor da fosfodiesterase tipo 5 reduziu as expressões α-SMA e TGF-β nos ureteres obstruídos, medidos por exames bioquímicos. Além disso, o tadalafil diminuiu a degeneração do urotélio devido à diminuição da perda de células epiteliais e da infiltração de células inflamatórias. Nossos resultados mostram que o tadalafil previne ou retarda o início da inflamação do ureter e degeneração urotelial em ratos com UUO.*

PALAVRAS-CHAVE: *Inibidor da fosfodiesterase tipo 5. Tadalafil. Obstrução. Histopatologia e Bioquímica .*

REFERENCES

1. Chuang YH, Chuang WL, Liu KM, Chen SS, Huang CH. Tissue damage and regeneration of ureteric smooth muscle in rats with obstructive uropathy. *Br J Urol*. 1998; 82: 261-6.
2. Cheng EY, Maizels M, Chou P, Hartanto V, Shapiro E. Response of the newborn ureteropelvic junction complex to induced and later reversed partial ureteral obstruction in the rabbit model. *J Urol*. 1993; 150: 782-9.
3. Harada T, Issa MM, Kigure T, Tsuchida S. Ureteral compliance and histology in partial obstruction in a canine model. *J Urol*. 1992; 148: 1274-8.
4. Stefanska A, Eng D, Kaverina N et al. Cells of renin lineage express hypoxia inducible factor 2alpha following experimental ureteral obstruction. *BMC Nephrol*. 2016; 17: 5.
5. Ucero AC, Benito-Martin A, Izquierdo MC et al. Unilateral ureteral obstruction: beyond obstruction. *Int Urol Nephrol*. 2014; 46: 765-76.
6. Bianchi D, Vespasiani G, Bove P. Acute kidney injury due to bilateral ureteral obstruction in children. *World J Nephrol*. 2014; 3: 182-92.
7. Okada H, Ban S, Nagao S, Takahashi H, Suzuki H, Neilson EG. Progressive renal fibrosis in murine polycystic kidney disease: an immunohistochemical observation. *Kidney Int*. 2000; 58: 587-97.
8. Carlisle RE, Heffernan A, Brimble E et al. TDAG51 mediates epithelial-to-mesenchymal transition in human proximal tubular epithelium. *Am J Physiol Renal Physiol*. 2012; 303: F467-81.
9. Liu X, Hong Q, Wang Z, Yu Y, Zou X, Xu L. Transforming growth factor-beta-sphingosine kinase 1/S1P signaling upregulates microRNA-21 to promote fibrosis in renal tubular epithelial cells. *Exp Biol Med (Maywood)*. 2016; 241: 265-72.
10. Leask A, Abraham DJ. TGF-beta signaling and the fibrotic response. *Faseb j*. 2004; 18: 816-27.
11. Yang J, Liu Y. Dissection of key events in tubular epithelial to myofibroblast transition and its implications in renal interstitial fibrosis. *Am J Pathol*. 2001; 159: 1465-75.
12. Zhang HY, Phan SH. Inhibition of myofibroblast apoptosis by transforming growth factor beta(1). *Am J Respir Cell Mol Biol*. 1999; 21: 658-65.
13. Liu Y. New insights into epithelial-mesenchymal transition in kidney fibrosis. *J Am Soc Nephrol*. 2010; 21: 212-22.
14. Sarhan NR, Omar NM. An immunohistochemical and ultrastructural analysis of the retina in tadalafil (Cialis) treated rats. *Acta Histochem*. 2018.
15. Socala K, Nieoczym D, Pierog M et al. Effect of Tadalafil on Seizure Threshold and Activity of Antiepileptic Drugs in Three Acute Seizure Tests in Mice. *Neurotox Res*. 2018.
16. Guzeloglu M, Yalcinkaya F, Atmaca S et al. The beneficial effects of tadalafil on renal ischemia-reperfusion injury in rats. *Urol Int*. 2011; 86: 197-203.
17. Girshovich A, Vinsonneau C, Perez J et al. Ureteral obstruction promotes proliferation and differentiation of the renal urothelium into a bladder-like phenotype. *Kidney Int*. 2012; 82: 428-35.
18. Benli E, Ayyildiz SN, Cirrik S et al. The effect of tadalafil therapy on kidney damage caused by sepsis in a polymicrobial septic model induced in rats: a biochemical and histopathological study. *Int Braz J Urol*. 2017; 43: 345-55.
19. Juan YS, Chuang SM, Long CY et al. Protein kinase C inhibitor prevents renal apoptotic and fibrotic changes in response to partial ureteric obstruction. *BJU Int*. 2012; 110: 283-92.
20. Zhang QF. Ulinastatin inhibits renal tubular epithelial apoptosis and interstitial fibrosis in rats with unilateral ureteral obstruction. *Mol Med Rep*. 2017; 16: 8916-22.
21. Zhao J, Wang L, Cao A, Jiang M, Chen X, Peng W. Renal Tubulointerstitial Fibrosis: A Review in Animal Models. *Journal of Integrative Nephrology and Andrology*. 2015; 2: 75-80.
22. Chevalier RL, Thornhill BA, Forbes MS, Kiley SC. Mechanisms of renal injury and progression of renal disease in congenital obstructive nephropathy. *Pediatr Nephrol*. 2010; 25: 687-97.
23. Hanna MK. Ureteral Structure and Ultrastructure. Part V. The Dysplastic Ureter. *The Journal of Urology*. 1979; 122: 796-8.
24. Dey N, Ghosh-Choudhury N, Kasinath BS, Choudhury GG. TGFβ-Stimulated MicroRNA-21 Utilizes PTEN to Orchestrate AKT/mTORC1 Signaling for Mesangial Cell Hypertrophy and Matrix Expansion. *PLOS ONE*. 2012; 7: e42316.
25. Sesti C, Florio V, Johnson EG, Kloner RA. The phosphodiesterase-5 inhibitor tadalafil reduces myocardial infarct size. *Int J Impot Res*. 2007; 19: 55-61.
26. Saenz de Tejada I, Anglin G, Knight JR, Emmick JT. Effects of tadalafil on erectile dysfunction in men with diabetes. *Diabetes Care*. 2002; 25: 2159-64.
27. Emmick JT, Stuewe SR, Mitchell M. Overview of the cardiovascular effects of tadalafil. *European Heart Journal Supplements*. 2002; 4: H32-H47.
28. Kloner RA, Mitchell M, Emmick JT. Cardiovascular effects of tadalafil in patients on common antihypertensive therapies. *Am J Cardiol*. 2003; 92: 47m-57m.

ORIGINAL ARTICLE

Non-pharmacological motor-cognitive treatment to improve the mental health of elderly adults

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SUMMARY

OBJECTIVE: *To propose a program of physical-cognitive dual task and to measure its impact in Chilean institutionalized elderly adults.*

METHOD: *Experimental design study with pre and post-intervention evaluations, measuring the cognitive and depressive levels by means of the Pfeiffer test and the Yesavage scale, respectively. The program was applied for 12 weeks to adults between 68 and 90 years old. The statistical analysis was based on the nonparametric Wilcoxon test for paired samples and was contrasted with its parametric version. The statistical software R was used.*

RESULTS: *Statistically significant differences were obtained in the cognitive level (p-value < 0.05) and highly significant (p-value < 0.001) in the level of depression with both tests (parametric and nonparametric).*

CONCLUSION: *Due to the almost null evidence of scientific interventions of programs that integrate physical activity and cognitive tasks together in Chilean elderly adults, a program of physical-cognitive dual task was proposed as a non-pharmacological treatment, easy to apply and of low cost to benefit their integral health, which improves significantly the cognitive and depressive levels of institutionalized elderly adults.*

KEYWORDS: *Cognitive Impairment. Depression. Dual Task. Elderly People. Physical Activity*

INTRODUCTION

The aging process is characterized by the gradual accumulation of cellular and molecular damage during the entire human life[1]. This accumulation produces widespread and progressive deterioration of the human body, besides of structural and functional changes that alter the psychomotor capacity[2]. It also causes a greater vulnerability to environmental factors and increased risk of diseases and death[1]. In addition to the biological and physiological problems, aging causes a decrease of the functions of the central nervous system[3], related to an increase in mental disorders [4] in elderly adults, referred to hereafter as EA." Among these age-related diseases are cognitive disorders such as mild cognitive impairment, de-

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mentia, and Alzheimer's[5], as well as emotion disorders such as anxiety and depression. These mental pathologies have economic, social, and public health impacts in the communities[6].

Cognitive dysfunction manifests progressively in people, starting with a mild cognitive impairment, a change in memory caused by aging, until it becomes dementia[5,7,8]. Dementia is a deterioration of mental faculties that produces serious conduct disorders and prevents the EA from engaging in daily life activities[5]. Dementia affects more than 47 million people in the world, and it is estimated that, by 2030, more than 75 million people will have this pathology, a number expected to double by the year 2050[7]. Whereas mild cognitive impairment is a modification of the memory that is higher than that expected for a specific age and degree of formal education, but with no signs of dementia, allowing the EA to engage properly in their daily activities[9].

One of the recurring emotional disorders in EA is depression[5], a disease that reduces the quality of life of people considerably[1]. Depression is affected by several factors, such as those of biological (neurochemical variations in the level of neurotransmitters or neuronal connections), psychological (history of mental illness, in the family or the individual), situational (stressful events), environmental (maltreatment, little exposure to sunlight), and sociocultural (loss of social functions) nature[10,11].

Regarding the strategies used to cope with the problems of aging, physical activity is perhaps the most successful[10-12]. When practiced on a regular and systematic manner, it is a powerful tool that promotes the well-being of people, since it maintains and improves the skeletal-muscle[13], cardio-respiratory[14], endocrine-metabolic[15], and psycho-neurological[16] functions. The effects produced by physical activity in the nervous system are the least known; two examples are the neuronal plasticity[17,18] and the increase in the activity of the hippocampus and of the neurotrophic factor, among others[19]. Thus, exercising not only improves physical health, but also mental health[20], leading to a lower incidence of cognitive deterioration[21] and depression[16].

The simultaneous implementation of physical exercise and cognitive tasks has been referred to as physical-cognitive dual task, abbreviated as "PCDT" from here on, which is defined as the interaction between physical activity and the cognitive processing of a task. For example, doing a balance exercise while reading the newspaper. Among the advantages of PCDT programs are an improvement ambulation of EA, both in its rhythm as its speed[22]. Good results were also obtained when there are changes of tasks, for example, alternating attention between one task and another[23], or prioritizing a task focusing on one of two[24]. Using PCDT programs, it is possible to see changes in physical performance and cognitive abilities of EA, due to the "bottleneck theory." In our context, this theory indicates that, due to the decrease in the processing of information, it is only possible to process one task at a time since the processing of a second task begins when the first ends. This "bottleneck" usually generates a higher response time for one of the two tasks when it uses a PCDT program, reducing the physical performance and cognitive capacity, which could be considered as a disadvantage. Several studies have demonstrated the positive effects of PCDT programs in EA, particularly its improvement in cognitive function[25-27], emotional state[10] and physical function[28]. The implementation of PCDT programs also improves memory[29], selective attention[18], reaction time[28], body awareness and spatial structure[30]. Thus, the combination of physical and cognitive tasks benefits the cognition and emotional state of EA. To the best of our knowledge, no PCDT programs targeted at EA has been applied in Chile.

The objectives of this study are: (i) to propose a PCDT program for EA to reduce their levels of cognitive impairment and depression; and (ii) to measure the impact of this program in institutionalized Chilean EAs. The PCDT program is proposed as a non-pharmacological, easy implementation, and low-cost strategy to benefit the health of EA.

The article is organized as follows: the second section describes the method used, detailing the PCDT program proposed in this study, the subjects of study, the ethical aspects, the measuring instruments, the variables analyzed, and the experimental design used for the statistical analysis that will allow us to obtain information to validate this program. The third section introduces the case study and the statistical analysis of the experimental data that leads to numerical results that validate the PCDT program. The last sections provide a discussion of the research performed and the final conclusions. A complete and updated list of references accompanies this study.

METHODS

The PCDT program

The double task program for EA proposed in this study is divided into three units of work, detailed in Table 1, following the didactic principle from the simplest to the most complex. It is proposed that each unit lasts 12 sessions, with a frequency of three times per week (on non-consecutive days), repeating the same routine of physical exercises and cognitive tasks during each week to observe the advances on the PCDT Program, enhancing the learning ability and motivation of the EA.

We propose to conduct the sessions at a moderate intensity, according to the rating of perceived exertion (RPE), which goes from 6 to 20 points[31,32]. The RPE of "moderate" intensity corresponds to the class between 40% and 59% of the heart rate reserve[33]. This moderate intensity corresponds to 12 or 13 points of RPE, and from 6 to 20 points in the Borg scale[32].

It is recommended that sessions last approximately 45 minutes, with each divided into three stages: warm-up, main stage and cool-down, detailed as follows:

Warm-up routine (8-10 minutes): the initial stage which prepares the body for the workout routine and is composed of articular mobility and of Tai Chi sequences (three to five sequences per session). This stage contains a system of psychophysical exercises that combine martial arts movements, breathing and stretching[19]. Tai Chi brings tranquility and relaxation to its practitioners and has a high healing power of physical and mental aspects, improving longevity[19].

Main stage (25 -30 minutes): main stage focused on developing the objectives planned in each unit as described in Table 1. Physical activity should be multicomponent, combining the physical qualities of strength, stretching, balance, coordination and metrics, the latter which corresponds to the coordinated movement of the limbs. In addition, it is necessary to have three levels of exercise (basic, medium and advanced), depending on the capabilities of the EA:

- Basic level: execution on sitting position/with or without implements.
- Medium level: execution on standing position/with or without implements.
- Advanced level: execution using displacement/with or without implements.

To reeducate, develop, and improve motricity, we propose three series per type of exercise, each series with 10 to 15 repetitions, according to the recommendations of the American College of Sports Medicine[34]. Regarding the cognitive tasks, the following must be exercised (according to the order of the units of the PCDT program detailed in Table 1): body awareness, memory, and attention, as well as space-time orientation and reasoning. The physical and cognitive tasks must be done simultaneously or one immediately after the other.

Cool-down (5-8 minutes): the final stage whose goal is to bring the body back to the initial resting condition. With that purpose, a stretching session (which will be repeated every session to improve memorization) and conscious breathing (guided inhaling and exhaling) is conducted.

Subjects and ethical aspects

The subjects of study are EA, and each one of them, as well as their representative, must follow the study accordingly and sign an informed consent form indicating that they agree to participate in the research and that they have knowledge of the PCDT program.

Measuring instruments

The instruments used will be questionnaires drawn following the guidelines of the Ministry of Health of Chile (www.minsal.cl) to measure the levels of cognition and depression of EA[35]. These questionnaires are the Pfeiffer[36,37] test of mental state and the Yesavage[38] scale of geriatric depression, which are detailed below:

TABLE 1: UNITS OF PCDT PROGRAM AND THEIR OBJECTIVE

	Unit 1 "Movement and awareness"	Unit 2 "Movement and memory"	Unit 3 "Movement and orientation"
Objective	Reeducate motricity and body awareness through exercises for strength and elongation.	Develop motricity, attention, and visual memory through exercises for strengthening the body scheme and balance.	Improve motricity, reasoning, quality of movement and sense of direction through exercises of coordination.
No. of sessions	12	12	12

- Pfeiffer Test: also known as "short portable mental status questionnaire" and abbreviated as "SPMSQ"[36]. This questionnaire is an instrument to assess the organic brain deficit in EA, easy to use, reliable, valid and brief, since it consists of 10 questions (see questionnaire in Annex 1). The test Pfeiffer assigns a score from 0 to 10, depending on the amount of incorrect questions answered, assigning one point for each incorrect question. The test corrects for schooling by subtracting from the total a point for EAs with basic or no schooling and adding a point to EAs with higher education. Then, on the basis of the errors made, these 10 questions measure the cognitive deterioration and determine its degree through four levels as follows: (0-2 points) normal, (3-4) light, (5-7 points), moderate and (8-10 points) severe.
- Yesavage Scale: also known as the "geriatric depression scale" and abbreviated as "GDS"[38]. This questionnaire is an instrument to detect depression in EAs who are 65 years old or older of also easy application, reliable, valid and consists of 15 questions with dichotomous answers such as "yes/no" (see the questionnaire in Annex 2). Then, the Yesavage scale assigns a score from 0 to 15 following the sum indicated in the questionnaire in Annex 2 and classifying the level of depression of the EA as follows: (0-5 points) normal, (6-9 points) light, and (10-15 points) established.

The study variables and experimental design

An experimental design of the "pre and post-test" type is used by applying the aforementioned instruments to measure the cognitive level with the Pfeiffer test and depressive level with the Yesavage scale. These instruments are applied before and after the intervention with the PCDT program, for twelve weeks, with each session of 45 minutes and a frequency of three times a week. The variables studied are defined in Table 2.

CASE STUDY RESULTS

Study subjects

This case analysis considers as study subjects (units of analysis) EAs institutionalized who lived in the retirement home "Silver House" (long-term stay private establishment) located in the community of Villa Alemana, V Region, Chile. The sample comprised 15 EAs institutionalized; however, after applying the inclusion criteria and due to the death of one of them, the sample included 10 EAs institutionalized (n =10), 6 women and 4 men, with ages between 68 and 90 years. This sample is considered random, since "Silver House" can be considered a retirement home representative of long-term stay establishments in Chile. The criteria for inclusion in the sample were the following:

- EAs institutionalized at the retirement home "Silver House".
- Age equal or over 65 years.
- Absence of a declared psychiatric pathology.
- Motor functionality compatible with the PCDT program proposed.

Data

The PCDT program proposed was applied to EAs institutionalized in the sample mentioned by two physical education teachers for twelve consecutive weeks. The data collection, using the instruments mentioned, was performed by the kinesiologist of the "Silver House", who interviewed the EAs three days before the program was implemented and three days after its last application. The kinesiologist re-

TABLE 2: VARIABLES STUDIED, THEIR RATING AND MEASURING INSTRUMENT

Rating	Variable	Measuring instrument
X1	Cognitive level of EA before the intervention	Pfeiffer Test (from zero to ten points: 0-10)
X2	Level of depression of the EA before the intervention	Yesavage Scale (from zero to fifteen points: 0-15)
Y1	Cognitive level of the EA after the intervention	Pfeiffer Test (from zero to ten points: 0-10)
Y2	Depression level of the EA after the intervention	Yesavage Scale (from zero to fifteen points: 0-15)
Z	Age of EA	(in years)

corded the EAs' answers in the questionnaires, and the data were digitally recorded using Office Excel. The data are shown in Table 3 and were analyzed with R (www.R-project.org), a free statistical software to make graphs and analysis of data widely used by the international scientific community. As mentioned, the unit of analysis is the institutionalized EA of the sample mentioned and the variables studied are defined in Table 2. According to the type of variable denoted in Table 2, such as X1, X2, Y1, Y2, and Z, the data analysis was based as follows:

(i) Descriptive statistical methods uni- and bivariate (correlation analysis).

(ii) Parametric and non-parametric statistical hypothesis tests for paired samples.

Statistical analysis

Table 4 and Figures 1 and 2 present a descriptive summary and uni- and bivariate graphs of the variables studied for n = 10 EA. This aspect is very important to decide what type of inferential statistical method should be used to assess whether the PCDT program significantly improves or not cognitive levels and depression of EAs.

Based on the results obtained in the exploratory analysis of the data to assess whether the PCDT program significantly improved or not the EAs' cognitive and depression levels, the Wilcoxon non-parametric test for paired samples was applied. As comparison and validation, the p-value of the Wilcoxon test is accompanied by the p-value of the statistical test corresponding to its parametric counterpart, that is, the Student-t test for paired samples. The results of both tests for the cognitive and depression levels, in accordance with the computational output of the R software, are shown in Table 5.

TABLE 4: DESCRIPTIVE SUMMARY OF THE VARIABLES AND UNITS OF MEASUREMENT LISTED IN TABLE 2 FOR EA DATA

Variable	Mean	Standard deviation	Coefficient of variation	Coefficient of asymmetry	Coefficient of kurtosis
X1	4.0	2.981	74.5%	0.611	2.152
X2	2.3	1.767	76.8%	0.461	1.551
Y1	6.3	3.467	5.0%	0.263	1.258
Y2	4.2	3.676	87.5%	0.799	2.414
Z	81	7.846	9.7%	-0.324	1.609

Based on the analysis of data it is possible to obtain the following information relevant to assessing the hypothesis of this research:

The cognitive levels of EA measured before and after the intervention showed an asymmetric distribution (non-normal or Gaussian), a situation that is repeated with the remaining variables under study with different degrees of asymmetry and kurtosis. It is important to remember that the normal distribution has coefficient of asymmetry equal to zero and a coefficient of kurtosis equal to three. In addition, even if the sample obtained can be regarded as representative of the population of Chilean EAs institutionalized, the information of the sample presented in Table 5 should be understood as correspondent to a small sample and could be different in a larger sample. Nevertheless, due to the characteristics of this sample, the results are statistically valid.

The lack of normality of the data, for the different variables studied, requires the use of non-parametric inferential methods. In the present study,

TABLE 3: DATA OBTAINED FOR THE VARIABLES AND UNITS OF MEASUREMENT LISTED IN TABLE 2.

Patient	Z	X1	Prior level of deterioration (Pfeiffer test)	X2	Post level of deterioration (Pfeiffer test)	Y1	Prior level of depression (Yesavage scale)	Y2	Post level of depression (Yesavage scale)
1	89	7	Moderate	2	Normal	10	Established	8	Light
2	77	6	Moderate	4	Light	3	Normal	0	Normal
3	80	5	Moderate	5	Moderate	12	Established	12	Established
4	90	10	Severe	5	Moderate	3	Normal	4	Normal
5	79	3	Light	1	Normal	8	Light	6	Light
6	90	3	Light	2	Normal	4	Normal	1	Normal
7	85	2	Normal	2	Normal	9	Light	5	Normal
8	82	2	Normal	1	Normal	3	Normal	2	Normal
9	70	2	Normal	1	Normal	3	Normal	2	Normal
10	68	0	Normal	0	Normal	8	Light	2	Normal

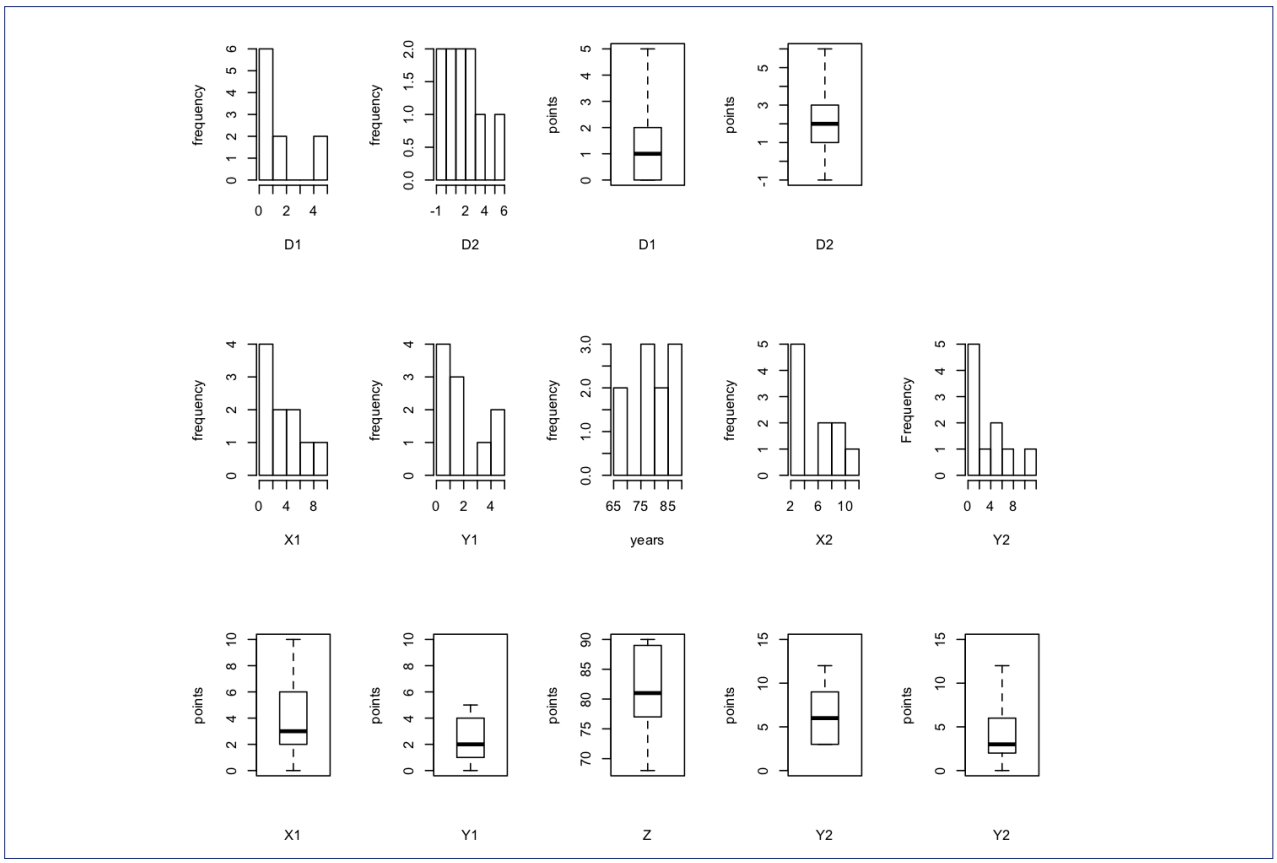

FIGURE 1: HISTOGRAMS AND BOXPLOT OF VARIABLE INDICATED FOR EA DATA.

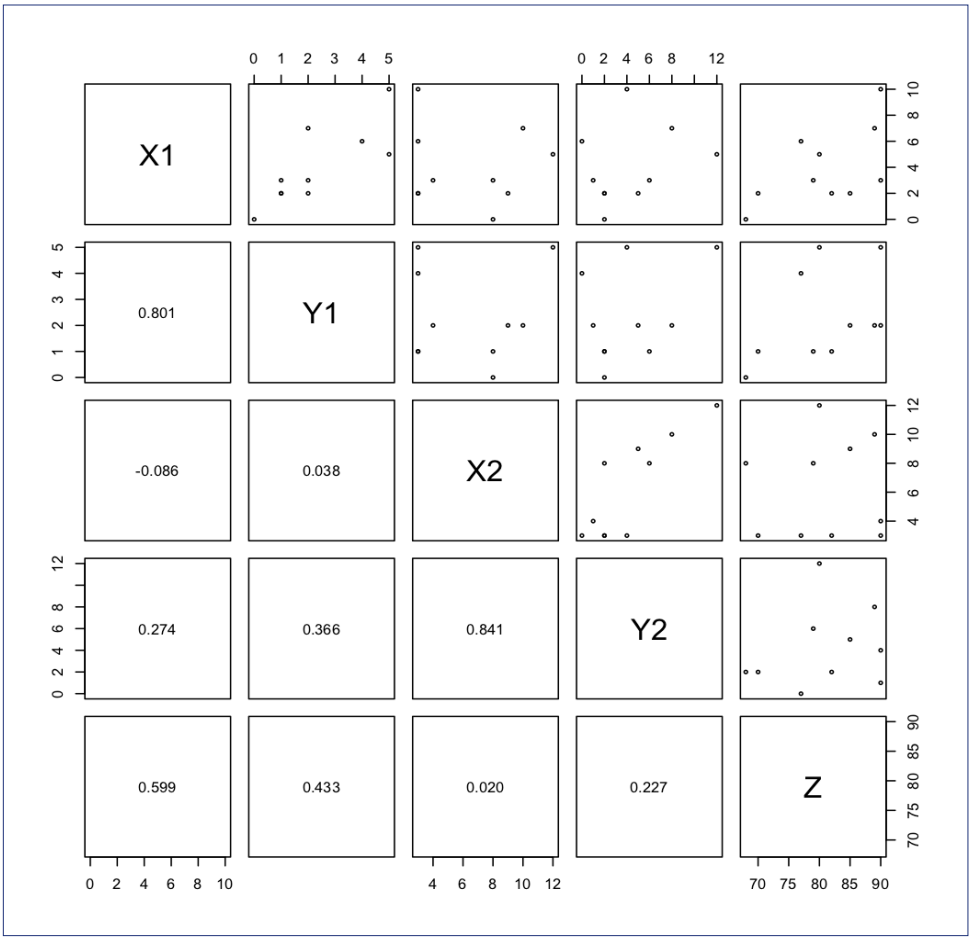

FIGURE 2: SCATTER DIAGRAMS AND THEIR PEARSON CORRELATION OF THE VARIABLES LISTED FOR EA DATA

TABLE 5: P-VALUE OF THE TEST AND VARIABLE INDICATED WITH EA DATA

Variable	Wilcoxon test	Student-t-test
D1 = X1 - Y1	W = 28; p-value = 0.0211 Alternative hypothesis: Median different from zero	T = 2.8465; degrees of freedom = 9; p-value = 0.0192 Alternative hypothesis: Mean different from zero Confidence interval of 95%: [0.3490; 3.0510] Sample mean of differences: 1.7
D2 = X2 - Y2	W = 43; p-value = 0.0172 Alternative hypothesis: Median different from zero	T = 3.2796; degrees of freedom = 9; p-value = 0.0095 Alternative hypothesis: Mean different from zero Confidence interval of 95%: [0.6515; 3.5485] Sample mean of differences: 2.1

this method corresponds to the Wilcoxon signed-rank test. In any case, as mentioned, to compare and validate the parametric statistical test equivalent to the Wilcoxon test, that is, the Student-t test, is also applied and reported.

From the exploratory analysis (see Table 5 and Figure 1), we can observe that the variability of cognitive levels of the EAs decreases after the intervention. This is relevant because the PCDT program transformed the EA sample into a more homogeneous group in cognitive level. However, this aspect reverses when the level of depression is assessed. This means that the PCDT program transformed the EA sample into a less homogeneous group in terms of depression.

The bivariate exploratory analysis provided in Figure 2 shows there is a high correlation between the cognitive levels before and after the intervention, a situation that is repeated with the level of depression. This implies that the hypothesis of independence necessary to use a usual parametric statistical test for comparing the means or medians is not valid. This invalidation is resolved by applying a test for paired samples, which simply means to analyze data from the differences in the cognitive levels (or depression) instead of the original data.

In Figure 1, it is possible to note that the differences between the cognitive levels of EAs before and after the intervention still do not have a non-normal distribution, requiring once again the use of the Wilcoxon non-parametric test. Once more it is accompanied by the Student-t test for comparison and validation, despite its incompatibility with the data from the differences in levels. Nevertheless, this non-normally detected in the distribution of cognitive levels does not seem to be violated in the depression levels, for which the normality it is observed as a reasonable hypothesis, based on the evidences available from the sample. This implies that, for data of differences in levels of depression, the Student-t test for paired samples is a valid method for assessing whether the PCDT program significantly improved or not the levels of EA depression. In any case, and again as a comparison and validation, the results of the Student-t test are accompanied by those obtained from the Wilcoxon test.

From Table 5, we concluded that the PCDT program proposed in this research allows to improve significantly (Wilcoxon test p-value = 0.0211; Student-t p-value = 0.0192) the EA cognitive level in 5%, increasing this level by 1.7 points in the Pfeffer test, with a confidence interval of 95% of [0.3490; 3.0510] points in this test.

Also based in Table 5, we concluded that the PCDT program proposed in this research has significantly improved (Student-t p-value = 0.0095; Wilcoxon p-value = 0.0172) the level of EA depression in 1%, improving this level by 2.1 points on the Yesavage scale, with a confidence interval of 95% of [0.6515; 3.5485] points on this scale.

Another interesting result obtained from the statistical analysis of the data that leaves one open problem that should continue to be studied is the relationship between levels of cognition and depression and the ED age. In Figure 2, it is possible to observe that there are very reasonable correlations between age and the mentioned levels. In addition, it shows that the PCDT program proposed produces an interesting effect on the EDs, since that age seems not to have an impact on these levels after the intervention, producing an equilibrium at such levels regardless of age.

DISCUSSION

The main objective of this study was to propose a PCDT program to improve the levels of EA cognition and depression. After the implementation of the PCDT program and analyzing the statistical results, it is possible to declare that the EA who participated

in the program showed significant cognitive improvement, as well as a highly significant improvement in their depressive state from a statistical point of view.

The results of previous studies show that physical activity alone improves the EA cognitive level[18,27,36]. Nevertheless, it has been demonstrated that there is a greater improvement in cognition when the physical training is combined with cognitive tasks[10,21,29,39-41] in comparison with physical training or cognitive tasks alone[25]. The present study employed a different methodology, without groups with physical activity or only cognitive tasks alone; therefore, it is not possible to confirm this statement. However, other studies[10,25,40,41] confirm the line of action taken in our research, allowing us to conclude that the PCDT program is a good intervention to improve EA cognition. It has been demonstrated that this type of intervention brings benefits to the independence and well-being of EA[21.26], mainly by significant improvements in the speed of information processing[25,28-30], the cognitive component used in most of the activities of daily life. However, the present study cannot prove such conclusions, since it did not employ an evaluation instrument for the functionality of the EAs; it only measured the general cognitive level through the Pfeiffer test. The results of the PCDT program of this research coincide with the improvements in EA cognitive levels[21,29,30,41] and depression[10,16,25] after participating in a program with physical and cognitive components.

The improvement of the cognitive level is given by the specific improvement of its components, such as memory[19,25,29], attention[25.30], body awareness, and spatial orientation[34]. Although this study did not include a specific evaluation of each cognitive component, the results coincide with previous research, showing an improvement in the post-test cognitive level results.

The institutionalized EA suffer from a greater number of pathologies, and neuropsychological diseases are more severe than in non-institutionalized EA who remain with their families[10]. This contributes to a high level of fragility and a drop in the functionality[25] of the institutionalized EA. With this same argument, some authors[10] have indicated that there are other factors that could be added to the sources of stress experienced by institutionalized EA in developing countries, such as an inadequate infrastructure, low professionalism of staff responsible for their care, and lack of comprehensive treatments, which also generate a negative impact on the emotional and cognitive aspects of the EA.

The results obtained in previous studies[10] have demonstrated the effectiveness of a combined training program on the improvement of emotional, cognitive, and independence abilities. After one year of the intervention with institutionalized EA, the authors[10] observed a positive trend of the program in reducing the levels of anxiety and depression in male EA. In addition, the cognitive deterioration of the EA did not increase throughout the sample. A recent study[28] confirmed these results with an eight-week intervention on institutionalized EA. It found significant improvements in physical condition, visual attention, executive functioning and speed of information processing. This proved the impact of such a type of programs in EA functionality, even of institutionalized ones. These results are related to the results obtained in the present study, since after the implementation of the PCDT program there was a statistically significant improvement in both cognitive and depression levels of the institutionalized EA.

Based on the results of this study, we concluded that the implementation of a PCDT program improves significantly the cognition and depression of institutionalized EA. This is reason enough for us to consider this type of intervention as a new form complementary non-pharmacological treatment thatis low cost and easy to implement to EA.

ANNEX 1: PFEIFFER TEST

Instructions: Ask from number 1 to 10 and complete the answer. These should be answered by the elderly individuals without seeing a calendar, newspaper or another element that facilitates the correct answer.

1.	What day is today?	(+)	(-)
2.	What day of the week is today?		
3.	What is this place or building called?		
4.	What is your phone number? What is your address? (only if there is no telephone)		
5.	How old are you?		
6.	What is the date of your birth? (Month/ Day/Year)		
7.	Who is the current president of Chile?		
8.	Who was the previous president?		
9.	What is your mother's last name?		
10.	Subtract 3 from 20 and continue to subtract 3 from each result until it is no longer possible to continue (20-17-14-11-8-5-2).		
TOTAL OF MISTAKES			

Formal education: Basic or no education ( ) Medium education ( ) Higher education ( )

Note: Subtract one in case of basic or no education. Add one in case of higher education.

Instructions for correction:	
Questions 1 and 6	It will be considered correct only the exact day, month and year.
Question 2	Self-explained.
Question 3	It will be marked correct if there is some description of the place.
Question 4	It will be correct by confirming the phone number or full address
Question 5	It will be correct if it matches with the date of birth.
Question 7 and 8	It will be correct if they say the last name of the President.
Question 9	It will be correct if they say what is in the clinical record.
Question 10	It will be correct only if the whole series is correct (20-17-14-11-8-5-2).

Evaluation of the results	
Number of mistakes	State
0-2	Normal (no cognitive deterioration)
3-4	Light cognitive deterioration
5-7	Moderate cognitive deterioration
8-10	Severe cognitive deterioration

ANNEX 2: YESAVAGE SCALE

Instructions: Answer each of the questions about how you felt during the last week.

Are you satisfied with your life?	yes	NO
Have you noticed a decrease or lack of interest in many of previous interests or activities?	YES	no
Do you feel your life is empty?	YES	no
Do you often feel bored?	YES	no
Do you feel excited most of the time?	yes	NO
Do you worry or fear that something might happen?	YES	no
Do you feel happy most of the time?	yes	NO
Do you often feel helpless?	YES	no
Do you rather stay at home than go out and do new things?	YES	no
Do you feel you have more memory problems than other people your age?	YES	no
Do you believe it is wonderful to be alive?	yes	NO
Do you feel useless or despicable in your current state?	YES	no
Do you feel energized?	yes	NO
Do you find yourself hopeless about your current situation?	YES	no
Do you believe other people are, in general, better off than you are?	YES	no

Add up all the YES answers (in capital letters) or NO (in capital letters),

NO (in capital letters), this is, YES = 1; yes = 0; NO = 1; no = 0, Total = __

Evaluation of the results	
Points	State
0-5	Normal (without depression)
6-10	Moderate depression
11-15	Established depression

RESUMO

OBJETIVO: *Propor um programa físico-cognitivo e medir seu impacto em idosos institucionalizados chilenos.*

MÉTODO: *Estudo de planejamento experimental com avaliações pré e pós-intervenção, medindo os níveis cognitivo e depressivo por meio do teste de Pfeiffer e da escala de Yesavage, respectivamente. O programa foi aplicado por 12 semanas a idosos entre 68 e 90 anos de idade. A análise estatística foi baseada no teste não paramétrico de Wilcoxon para amostras pareadas e foi contrastada com sua versão paramétrica. O software estatístico R foi utilizado.*

RESULTADOS: *Diferenças estatisticamente significantes foram obtidas no nível cognitivo ($p < 0,05$) e altamente significante ($p < 0,001$) no nível de depressão com ambos os testes (paramétrico e não paramétrico).*

CONCLUSÃO: *Porque quase não existe evidência científica de programas de intervenções que integram a atividade física e tarefas cognitivas em chilenos idosos, um programa físico-cognitivo foi proposto como tratamento não farmacológico, fácil de implementar e de baixo custo, para beneficiar a sua saúde integral, melhorando significativamente os níveis cognitivos e depressivos de idosos institucionalizados.*

PALAVRAS-CHAVE: *Atividade Física. Depressão. Deterioração Cognitiva. Dupla Tarefa. Idosos.*

REFERÊNCIAS

1. Organización Mundial de la Salud. Informe mundial sobre el envejecimiento y la salud. Ginebra: Organización Mundial de la Salud; 2015.
2. Almeida ST, Soldera CLC, Carli GA, Gomes I, Resende TL. Análise de fatores extrínsecos e intrínsecos que predispõem a quedas em idosos. Rev Assoc Med Bras. 2012; 58(4):427-33.
3. Tseng CN, Gau BS, Lou MF. The effectiveness of exercise on improvising cognitive function in older people: a systematic review. J Nurs Res. 2011;19(2):119-31.
4. Szalewska D, Radkowski M, Demkow U, Winklewski PJ. Exercise strategies to counteract brain aging effects. Adv Exp Med Biol. 2017;1020:69-79.

5. Dias FLC, Silva RMFL, Moraes EN, Caramelli P. Perfil clínico e autonômico de pacientes com doença de Alzheimer e demência mista. Rev Assoc Med Bras. 2013;59(5):435-41.
6. Zivin, K, Wharton T, Rostant O. The economic, public health, and caregiver burden of late-life depression. Psychiatric Clin North Am. 2013;36(4):631-49.
7. Organización Mundial de la Salud. La Salud mental y los adultos mayores. Nota descriptiva. Ginebra: Organización Mundial de la Salud; 2017. [cited 2018 Jul 5]. Available from: http://www.who.int/es/news-room/fact-sheets/detail/la-salud-mental-y-los-adultos-mayores.
8. Falck RS, Landry GJ, Best JR, Davis JC, Chiu BK, Liu-Ambrose T. Examining relationships of physical activity and sedentary behaviour with cognitive function among older adults with mild cognitive impairment: a cross-sectional study. Alzheimers Dement. 2017;13:P865-6.
9. Petersen RC, Parisi JE, Dickson DW, Johnson KA, Knopman DS, Boeve BF, et al. Neuropathologic features of amnestic mild cognitive impairment. Arch Neurol. 2006;63(5):665-72.
10. Garín MBG, Aranda AB, Jiménez SB. Programa combinado para mejorar el estado emocional y prevenir el deterioro cognitivo de adultos mayores institucionalizados. Informació Psicológica. 2014;106:41-53.
11. Tribess S, Virtuoso Júnior JS, Oliveira RJ. Atividade física como preditor da ausência de fragilidade em idosos. Rev Assoc Med Bras. 2012;58(3):341-7.
12. Bauman A, Merom D, Bull FC, Buchner DM, Fiatarone Singh MA. Updating the evidence for physical activity: summative reviews of the epidemiological evidence, prevalence, and interventions to promote "active aging". Gerontologist. 2016;56(Suppl 2):S268-80.
13. Valenzuela T. Efficacy of progressive resistance training interventions in older adults in nursing homes: a systematic review. J Am Med Dir Assoc. 2012;13(5):418-28.
14. Koster A, Stenholm S, Schrack JA. The benefits of physical activity for older people. In: Nyman SR, Barker A, Haines T, Horton K, Musselwhite C, Peeters G, et al., eds. The Palgrave handbook of ageing and physical activity promotion. Basingstoke: Palgrave Macmillan; 2018. p.43-60.
15. Bouaziz W, Vogel T, Schmitt E, Kaltenbach G, Geny B, Lang PO. Health benefits of aerobic training programs in adults aged 70 and over: a systematic review. Arch Gerontol Geriatr. 2017;69:110-27.
16. Olson RL, Brush CJ, Ehmann PJ, Alderman BL. Randomized trial of aerobic exercise on cognitive control in major depression. Clin Neurophysiol. 2017;128(6):903-13.
17. Erickson KI, Gildengers AG, Butters MA. Physical activity and brain plasticity in late adulthood. Dialogues Clin Neurosci. 2013;15(1):99-108.
18. Bherer L. Cognitive plasticity in older adults: effects of cognitive training and physical exercise. Ann N Y Acad Sci. 2015;1337:1-6.
19. Sungkarat S, Boripuntakul S, Kumfu S, Lord SR, Chattipakorn N. Tai Chi improves cognition and plasma BDNF in older adults with mild cognitive impairment: a randomized controlled trial. Neurorehabil Neural Repair. 2018;32(2):142-9.
20. Blondell SJ, Hammersley-Mather R, Veerman JL. Does physical activity prevent cognitive decline and dementia? A systematic review and meta-analysis of longitudinal studies. BMC Public Health. 2014;14:510.
21. Karssemeijer EGA, Aaronson JJ, Bossers WW, Smits T, Olde Rikkert MGM, Kessels RPC. Positive effects of combined cognitive and physical exercise training on cognitive function in older adults with mild cognitive impairment or dementia: A meta-analysis. Ageing Res Rev. 2017;40:75-83.
22. Wollesen B, Voelcker-Rehage C. Training effects on motor-cognitive dual-task performance in older adults: a systematic review. Eur Rev Aging Phys Act. 2013;11(1):5-24.
23. Silsupadol P, Shumway-Cook A, Lugade V, van Donkelaar P, Chou LS, Mayr U, et al. Effects of single-task versus dual-task training on balance performance in older adults: a double-blind, randomized controlled trial. Arch Phys Med Rehabil. 2009;90(3):381-7.
24. Bherer L, Kramer AF, Peterson MS, Colcombe S, Erickson K, Becic E. Transfer effects in task-set cost and dual-task training in older and younger adults: further evidence for cognitive plasticity in attentional control in late adulthood. Exp Aging Res. 2008;34(3):188-219.
25. Oswald WD, Gunzelmann T, Rupprecht R, Hagen B. Differential effects of single versus combined cognitive and physical training with older adults: the SimA study in a 5-year perspective. Eur J Ageing. 2006;3(4):179.
26. Theill N, Schumacher V, Adelsberger R, Martin M, Jäncke L. Effects of simultaneously performed cognitive and physical training in older adults. BMC Neurosc. 2013;14:103.
27. Sáez de Asteasu ML, Martínez-Velilla N, Zambom-Ferraresi F, Casas-Herrero Á, Izquierdo M. Role of physical exercise on cognitive function in healthy older adults: a systematic review of randomized clinical trials. Ageing Res Rev. 2017;37:117-34.
28. Marmeleira J, Galhardas L, Raimundo A. Exercise merging physical and cognitive stimulation improves physical fitness and cognitive functioning in older nursing home residents: a pilot study. Geriatr Nurs. 2018;39(3):303-9.
29. Lai L, Bruce H, Bherer L, Lussier M, Li KZH. Comparing the transfer effects of simultaneously and sequentially combined aerobic exercise and cognitive training in older adults. J Cognitive Enhancement. 2017;1(4):478-90.
30. Valencia C, López-Alzate E, Tirado V, Zea-Herrera MD, Lopera F, Rupprecht R, et al. Cognitive effects of combined memory and psychomotor training in elderly adults. Rev Neurol. 2008;46(8):465-71.
31. American College of Sports Medicine Position Stand. The recommended quantity and quality of exercise for developing and maintaining cardiorespiratory and muscular fitness, and flexibility in healthy adults. Med Sci Sports Exerc. 1998;30(6):975-91.
32. Borg GA. Psychophysical bases of perceived exertion. Med Sci Sports Exerc. 1982;14(5):377-81.
33. Kaufman C, Berg K, Noble J, Thomas J. Ratings of perceived exertion of ACSM exercise guidelines in individuals varying in aerobic fitness. Res Q Exerc Sport. 2006;77(1):122-30.
34. American College of Sports Medicine. ACSM's guidelines for exercise testing and prescription. Philadelphia: Lippincott Williams & Wilkins; 2013.
35. Chile. Ministerio de Salud. Manual de aplicación del examen de medicina preventiva del adulto mayor. Santiago: Ministerio de Salud; 2018. [cited 2018 Jul 5]. Available from: http://www.minsal.cl/portal/url/item/ab1f81f43ef0c2a6e04001011e011907.pdf
36. Pfeiffer E. A short portable mental status questionnaire for the assessment of organic brain deficit in elderly patients. J Am Geriatr Soc.1975;23(10):433-41.
37. Martínez de la Iglesia J, Dueñas Herrero R, Onís Vilches MC, Aguado Taberné C, Albert Colomer C, Luque Luque R. Spanish language adaptation and validation of the Pfeiffer's questionnaire (SPMSQ) to detect cognitive deterioration in people over 65 years of age. Med Cli. 2001;117(4):129-34.
38. Sheikh JI, Yesavage JA. Geriatric Depression Scale (GDS): recent evidence and development of a shorter version. Clin Gerontol. 1986;5(1-2):165-73.
39. Kelly ME, Loughrey D, Lawlor BA, Robertson IH, Walsh C, Brennan S. The impact of exercise on the cognitive functioning of healthy older adults: a systematic review and meta-analysis. Ageing Res Rev. 2014;16:12-31.
40. Rey Cao A, Canales Lacruz I. Cognitive and perceptual-motor improvements in elderly participating in a cognitive-motor integrated stimulation program. Rev Bras Geriatr Gerontol. 2012;15(1):27-39.
41. Middleton LE, Ventura MI, Santos-Modesitt W, Poelke G, Yaffe K, Barnes DE. The Mental Activity and eXercise (MAX) trial: Effects on physical function and quality of life among older adults with cognitive complaints. Contemp Clin Trials. 2018;64:161-6.

ORIGINAL ARTICLE

Id-1 expression in colorectal adenocarcinoma tissues and its clinical significance

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SUMMARY

BACKGROUND: *This study aims to investigate the expression of Id-1 in human colorectal adenocarcinoma tissues and explore its correlation with the clinical pathological parameters of colorectal cancer.*

METHODS: *The Id-1 mRNA and protein expression levels of 50 specimens of normal colorectal tissues and 50 specimens of colorectal adenocarcinoma tissues were detected using reverse-transcription polymerase chain reaction and western blot. Furthermore, Id-1 protein was detected using immunohistochemistry. The correlation between the expression of Id-1 and clinicopathologic features was analyzed.*

RESULTS: *The mRNA expression level of Id-1 in colorectal adenocarcinoma tissues and normal colorectal tissues was 0.96 ± 0.03 vs. 0.20 ± 0.04, respectively; and the difference was statistically significant (P=0.011). Furthermore, Id-1 protein expression was higher in colorectal adenocarcinoma tissues than in normal colorectal tissues (0.82 ± 0.04 vs. 0.31 ± 0.02, P=0.020). In addition, the positive protein expression rate of Id-1 was higher in colorectal adenocarcinoma tissues than in normal colorectal tissues (72.00% vs. 24.00%, X2=23.431, P=0.000). The expression of Id-1 was correlated with the depth of tumor invasion, TNM stage, lymph node metastasis, vessel invasion, and liver metastasis (P<0.01). However, this expression was not correlated with tumor size and differentiation degrees (P>0.05).*

CONCLUSIONS: *The high Id-1 expression in colorectal adenocarcinoma tissues play an important role in the process of cancer, and is expected to become a new tumor monitoring indicator for clinical diagnosis, treatment, and prognosis judgment.*

KEYWORDS: *Colorectal neoplasms. Immunohistochemistry. Blotting, Western. Adenocarcinoma.*

INTRODUTION

Inhibitors of DNA binding-1 (Id-1) have been an enthusiastically studied oncogene that can promote cell proliferation, induce tumor angiogenesis, enhance tumor growth invasion, and accelerate tumor growth and metastasis through animal experiments [1,2]. To date, it has been proven that Id-1 takes on a high expression state in multiple malignant tumors such as breast cancer, cervical cancer, and

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esophagus cancer [3-5]. However, studies in the field of colorectal cancer have been relatively few. This study aimed at investigating the expression of Id-1 in colorectal cancer and its relationship with related clinicopathological factors through immunohistochemistry, RT-PCR, and western blot analysis.

METHODS

Cases data

Fifty cases of colorectal cancer tissues excised during general surgery and 50 cases of normal tissues were collected between May 2012 and May 2014 from the First Affiliated Hospital of Hebei North University. Cancerous tissues were selected from the Cancer Lesions Center. In addition, incisal edges of specimen stub ends were selected, which were confirmed to be normal colorectal tissues by pathology. All cases had primary tumors, and targeted treatments such as neoadjuvant chemoradiotherapy were not performed prior to the operation. The male-to-female proportion of these subjects was 29:21, and their age ranged was 33-71 years, with an average age of 51.2 ± 2.9 years. Furthermore, among cases with normal colorectal tissues, the male-to-female proportion was 28:22, and their age ranged from 33-69 years, with an average age of 52.0 ± 2.1 years. The general data of cases in these two groups were comparable (P>0.05).

Main Reagents and Experimental Methods

Main Reagents

Rabbit anti-human monoclonal concentrated Id-1 antibody (ab50932 R2-1G6) was purchased from Abcam (Hong Kong); rabbit anti-human GAPDH polyclonal primary antibody (ab54621R2-3) and goat anti-rabbit secondary antibody (ab5214R2-2) were purchased from Santa Cruz Biotechnology (USA); Total RNA extraction reagent, reverse transcription kit, SDS-PAGE standard indicator, and BCA protein quantification kit were all purchased from Fermentas (Thermo Scientific, USA); RIPA reagents were purchased from Solarbio (Beijing, China); DAB color developing agent was purchased from Tiangen Biotech (Beijing, China). The primer was synthesized by Takara Bio .

Id-1 mRNA expression was detected by RT-PCR assay.

The total RNA concentration was extracted and determined. Id-1 upstream primer was 5'-CTGAG-GCACTGGCGAGGAGA-3' and Id-1 downstream primer was 5'-GAGAAGCACCAAACGTGACCAT-3'; and the length was amplified by 140 bp. GAPDH (β-actin): upstream primer was 5'-ACCCACTCCTC-CACCTTGA-3' and downstream primer was 5'-ACCACCCTGTT-GCTGTAGCC-3'; and the length was amplified by 107 bp. Reaction system: 2.5 ml of 1×PCR Buffer, 0.5 ml of dNTP, 2 ml of cDNA template, 1.25 U of Tag enzyme, 1 ml of each upstream and downstream primer, 1 ml of RNA enzyme inhibitor, and 25 ml of total reaction volume. The amplification was operated according to kit specifications: 1 ml of DNA sample was taken as the template, and deionized water was taken as the negative control template. The amplification was carried out under the following conditions: 95°C for two minutes pre-degeneration; at 95°C, for 10 seconds, at annealing temperature, for 15 seconds; at 72°C, extended to 45 seconds, with a total of 35 circulations; finally, at 72°C, extended again for 10 minutes. After adding 2% of sepharose gel, the product was electrophoretically separated; the result was observed under an ultraviolet radiator.

Id-1 protein expression was detected by western blot assay.

Total protein was extracted by RIPA lysate and quantified. Then, 50 mg of protein was taken, placed into a 5× buffer solution (4:1), water baths were performed three times for five minutes at 100°C, followed by electrophoresis with 8% polyacrylamide gel (SDS-PAGE), and the specimen was shifted onto PVDF membranes. Next, the membrane was removed, 5% skim milk powder was added for confining; the confining liquid was discarded. The specific primary antibody was added dropwise (Id-1: 1:1 500; GAPDH: 1:150) and incubated overnight at 4°C. TBST washing liquid was added three times for 10 minutes. The specific secondary antibody was added (1:1,500) and incubated for one hour at 37°C. TBST washing liquid was added three times again for 10 minutes. The stripes were detected by the ECL method, and quantitative analysis on density was conducted.

Protein expression was detected by immunohistochemical staining

Some specimens were fixed by 10% formaldehyde solution and embedded by paraffin wax, with a slice thickness of 4 mm. Then, hematoxylin and eosin (H&E) staining method were used for pathological diagnosis. The slices were roasted in an oven for 30

minutes at 80°C, then dehydrated using ethanol; endogenous peroxidase inactivated, hydrated repaired using citric acid buffer solution at high temperature and under high pressure, washed for three times by PBS. The primary antibody was added and cultured overnight at 4°C, then washed three times with PBS. The secondary antibody was added. At room temperature, the specimens were color developed by DAB, counterstained with hematoxylin, and differentiated with 1% hcl-ethanol for five minutes. Then, the specimens were rinsed with running water for five minutes, dehydrated with gradient ethanol, transparentized with turpentine for five minutes, and eventually sealed with gum. In the negative control group, the primary antibody was replaced by PBS.

Assessment of the positive expression of Id-1

In the cytoplasm, particles stained in claybank and brown were positive cells. Positive cell percentage was calculated and scored as follows: ≤5% was scored 0; 6%-25% was scored 1; 26%-50% was scored 2; 51%-75% was scored 3; and >75% was scored 4. No color was scored 0; yellow was scored 1; and deep yellow was scored 3. These two were multiplied, with 0 being (-), 1-4 being (+), 5-8 being (++), and 9-12 being (+++); and (+)-(+++) were all regarded as positive.

Statistical method

SPSS 17.0 statistical software was used for the analysis. Measurement data were indicated as x ± SD. The t-test was used for comparison between the two groups. Measurement data was presented in percentage. *The* X^2–test was used to test and analyze the expression of Id-1 and its relationship with clinicopathological parameters. For the inspection level, $\alpha = 0.05$ was assumed.

RESULTS

RT-PCR test results for Id-1

By taking the ratio of Id-1/GAPDH for the RT-PCR quantitative analysis, the mRNA expression level of Id-1 was significantly higher in cancerous tissues than in colorectal normal tissues; and the difference was statistically significant (0.96 ± 0.03 *vs.* 0.20 ± 0.04, $P = 0.011$; as shown in Figure 1).

Id-1 test results by western blot

The ratio of Id-1/GAPDH was taken for the protein quantitative test. In addition, the protein expression level of Id-1 was higher in colorectal cancer tissues than in colorectal normal tissues; the difference was statistically significant (0.82 ± 0. 04 *vs.* 0. 31 ± 0. 02, $P = 0.020$; as shown in Figure 2).

Immunohistochemical test results for Id-1

The positive expression rate in colorectal cancer tissues (72.00%, 36/50) was significantly higher than in normal mucous membranes (24.00%, 12/50), and the difference was statistically significant (X^2= 23.431, P=0.000; Figure 3).

The relationship between Id-1 protein and clinicopathological parameters

The protein expression level was not associated with the tumor size of patients and the differentiation degree but was closely related to tumor serosa infiltration, hepatic metastasis, lymphatic metastasis, tumor TNM stages, and vessel infiltration (P<0.01).

DISCUSSION

Id-1 is an important member of the helix-loop-helix (HLH) transcription factor family and a negative regulatory factor of cell differentiation. It has been widely expressed in the embryos of mammals, reproduction glands, and some immaturely differentiated histocytes. In normal tissues, it has a state of micro-expression or no expression. However, it has high expression in many malignant tumors and in vitro cultivated tumor cells. It is also related to the malignant potential and prognosis of tumors [6]. Most HLH structures have one closed adjacent basic HLH (bHLH) factor. These two are mutually combined to form a heterodimer, then are combined with targeted DNA to induce the E-box-like-structure target gene in the promoter to transcribe and integrate the so-called "E-box" DNA sequence in order to regulate and control cell proliferation and differentiation [7].

Id-1 enhances cell proliferation and invasion in multiple ways. Studies have confirmed that Id-1 forms a specific complex to inhibit E2A, activate P21, activate CDK2, induce Rb protein phosphorylation, inhibit G1/S stagnation during the cell division cycle and initiates the cell division cycle by combining with E2A [8]. The upregulation of Id-1 can enhance TGF-β1 expression, reduce E-calcium mucoprotein expression, and induce materialization between epithelial cells, which leads to loss of polarity between cells, as well as the reduction of adhesion. This would induce

cancerous cells to break through the basilar membrane barrier, metastasize, and infiltrate to tissues in the vicinity [9,10].

CONCLUSIONS

The results of this experiment revealed that the mRNA and protein expression of Id-1 was significantly higher in glandular cancer tissues than in normal tissues, which confirms that the specific high expression of Id-1 may play an important role in the process of occurrence and development of colorectal cancer.[11] It also revealed that the expression of Id-1 in non-small cell lung cancer gradually increased with the deepening of tumor infiltration. Furthermore, this was enhanced with lymph node and distant metastasis, which had a higher expression in lung cancer tissues at TNM III and IV stages. [12] Immunohistochemical assays were performed on 60 cases of esophageal squamous carcinoma tissues and revealed that the positive expression of Id-1 was significantly higher in esophageal squamous carcinoma than in normal esophagus mucous membranes. [13] The positive expression of Id-1 in the normal gastric

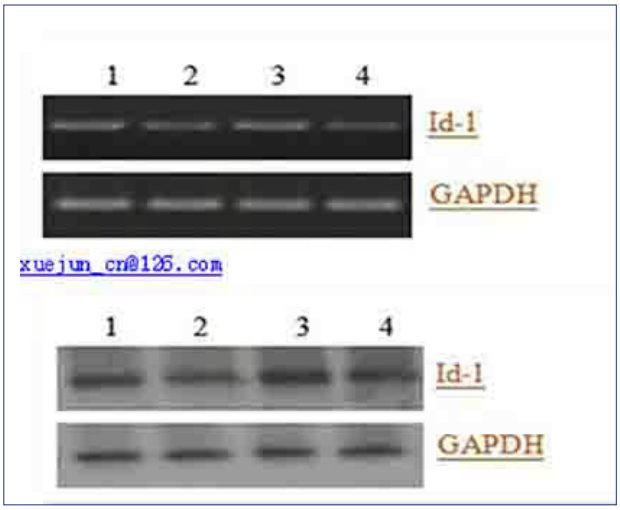

FIGURE 1. Expression level of Id-1 mRNA in colorectal adenocarcinoma and normal colorectal tissues detected by reverse-transcription polymerase chain reaction.
1–3 colorectal adenocarcinoma group 2–4: normal colorectalgroup

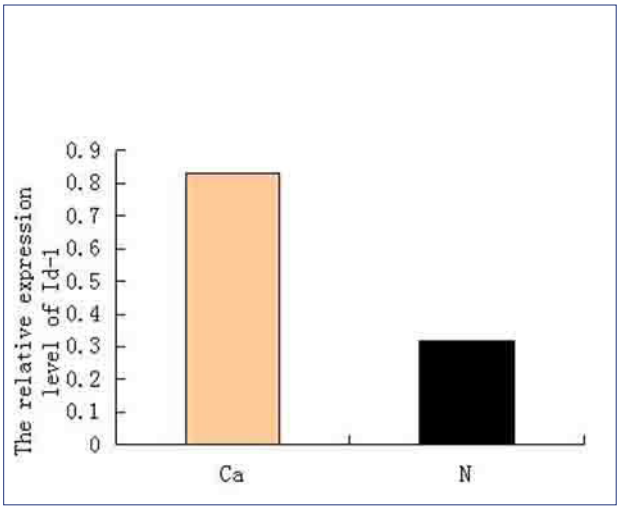

FIGURE 2. Expression level of Id-1 protein in colorectal adenocarcinoma and normal colorectal tissues detected by Western blot.

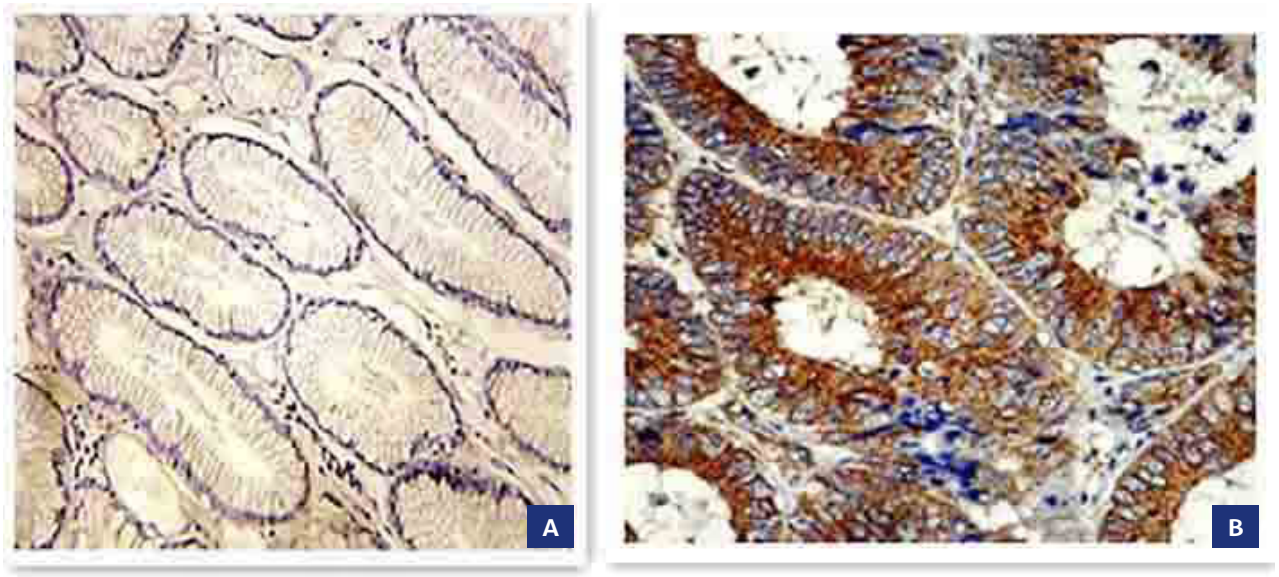

FIGURE 3. Expression level of Id-1 protein in colorectal tissues detected by Immunohistochemistry (SP×200).
A: Colorectal adenocarcinoma tissues B: Normal colorectal tissues

mucous membrane and atypical hyperplastic gastric mucous membrane had a gradually increasing trend, and the difference was statistically significant. Furthermore, this was significantly correlated to tumor differentiation degree, infiltration depth, TNM stages, lymphatic metastasis, and prognosis. The lifetime of Id-1 positive patients was significantly shorter than that of negative ones. In-depth studies have found that Id-1 protein expression in patients with colorectal cancer accompanied by serosa infiltration significantly increased when compared with non-infiltration. The higher the TNM stage was, the higher the Id-1 protein expression became. This shows that the protein expression level of Id-1 was closely correlated to the malignant biological behavior of colorectal tumors, which plays an accelerated role in the invasion and metastasis of colorectal cancer. [14] Immunohistochemical assays were performed on 62 cases of colorectal cancer tissues and found that Id-1 had 100% positive staining in cancerous tissues. This was closely related to tumor infiltration depth and lymphatic metastasis and was positively correlated to Ki-67 and VEGF, considering that Id-1, Ki-67, and VEGF had a synergistic effect. Furthermore, the study of Hu et al.[15] revealed that Id-1 in colorectal cancer could upregulate the expression level of MT1-MMP, inducing gene metal enzymes such as MMP-2 and MMP-9 to be overexpressed, thus enhancing the degradation of the basilar membrane and accelerating the infiltration and metastasis of cancer cells. In addition, studies have confirmed that Id-1 could also induce the formation of tumor angiogenesis and provide a nutrition basis for the growth, infiltration, and metastasis of tumors through the two aspects of the combined action of promoting VEGF expression and inhibiting TSP-1 expression[10].

In conclusion, the high expression of Id-1 is highly correlated to the occurrence, development, and malignant biological behavior of colorectal cancer. As a result, Id-1 is expected to become a new tumor monitoring index, providing an important reference for clinical treatment and prognosis monitoring. This experiment has only studied the expression of Id-1 in colorectal cancer and its relationship with clinicopathological features. However, the corresponding carcinogenic mechanism and role access were not studied. Furthermore, no related experiments have confirmed whether the application of Id-1 inhibitors can inhibit cell proliferation. This would be the future study direction for this subject.

List of abbreviations

- Inhibitors of DNA binding-1 Id-1
- hematoxylin and eosin H&E
- helix-loop-helix HLH
- membrane type-1 matrix metalloproteinase MT1-MMP
- sodium dodecyl sulfate polyacrylamide gel electrophoresis SDS-PAGE
- reverse transcription-polymerase chain reaction RT-PCR
- Tumor Node Metastasis TNM

Declaration

Ethics approval and consent to participate:

This study was conducted in accordance with the declaration of Helsinki.

Consent for publication

We agree that the article is published.

Availability of data and material

Not applicable.

Conflicts of interest statement:

We declare that we do not have any commercial or associative interest that represents a conflict of interest in connection with the work submitted.

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Authors' contributions

Xue-Liang Wu carried out the molecular genetic studies, participated in the sequence alignment and drafted the manuscript. Li-Kun Wang and Rui- Min Yang carried out the immunoassays. Bo-Liu participated in the sequence alignment. Jun Xue and Fei-Guo participated in the design of the study and performed the statistical analysis. Dong-Dong Yang and Ming Qu conceived of the study, and Jun Xue participated in its design and coordination and helped to draft the manuscript. All authors read and approved the final manuscript

RESUMO

OBJETIVO: *O objetivo deste estudo é investigar a expressão de Id-1 em tecidos de adenocarcinoma colorretal em humanos e investigar sua correlação com os parâmetros patológicos clínicos de câncer colorretal.*

MÉTODOS: *Os níveis de expressão de proteína e mRNA Id-1 em 50 amostras de tecido colorretal normal e 50 amostras de tecido de adenocarcinoma colorretal foram detectados através de reação em cadeia de polimerase precedida de transcrição reversa e western blot. Além disso, a proteína Id-1 foi detectada através de imuno-histoquímica. A correlação entre a expressão de Id-1 e características clínico-patológicas foi analisada.*

RESULTADOS: *O nível de expressão de mRNA Id-1 em tecidos de adenocarcinoma colorretal e tecidos colorretais normais foi de 0,96 ± 0,03 versus 0,20 ± 0,04, respectivamente; a diferença foi estatisticamente significativa (P= 0,011). Além disso, a expressão da proteína Id-1 foi maior em tecidos de adenocarcinoma colorretal do que em tecidos colorretais normais (0,82 ± 0,04 versus 0,31 ± 0,02, P= 0,020). Além disso, a taxa de expressão positiva de proteínas Id-1 foi maior em tecidos de adenocarcinoma colorretal do que em tecidos colorretais normais (72,00% vs. 24,00%, X2=23,431, p=0,000). A expressão de Id-1 foi correlacionada com a profundidade da invasão tumoral, estágio TNM, metástases linfonodais, invasão vascular e metástase hepática (P<0,01). Todavia, essa expressão não se correlacionou com o tamanho do tumor e graus de diferenciação (P>0,05).*

CONCLUSÃO: *A alta expressão de Id-1 em tecidos de adenocarcinoma colorretal desempenham um importante papel no processo do câncer, e é esperado que se torne um novo indicador de monitoramento de tumores para o diagnóstico clínico, tratamento e estimativa de prognóstico.*

PALAVRAS-CHAVE: *Neoplasias Colorretais. Imuno-histoquímica. Blotting, Western. Adenocarcinoma.*

REFERENCES

1. Yang G, Zhang Y, Xiong J, Wu J, Yang C, Huang H, et al. Downregulation of Id1 by small interfering RNA in gastric cancer inhibits cell growth via the Akt pathway. Mol Med Rep. 2012;5(4):1075-9.
2. Bhattacharya R, Kowalski J, Larson AR, Brock M, Alani RM. Id1 promotes tumor cell migration in nonsmall cell lung cancers. J Oncol. 2010;2010:856105.
3. Chen Z, Liu S, Sumida T, Sun S, Wei Y, Liu M, et al. Silencing Id-1 with RNA interference inhibits adenoid cystic carcinoma in mice. J Surg Res. 2011;169(1):57-66.
4. Ma LJ, He JC, Cai ZJ. Expression of inhibitor of differentiation -1 in cervical cancer tissues and its correlation with Ki-67. Chinese J Clin Pharmacol. 2015;31:627-30.
5. Li JF, Hu YH, Dong YL. Expression and significance of differentiation inhibitor 1,3 and matrix metalloproteinase-9 in brain metastatic tumors. Chinese J Exp Surg. 2013;30:300-2.
6. Rothschild SI, Kappeler A, Ratschiller D, Betticher DC, Tschan MP, Gugger M, et al. The stem cell gene "inhibitor of differentiation 1" (IDI) is frequently expressed in non-small cell lung cancer. Lung Cancer. 2011;71(3):306-11.
7. Sun R, Chen W, Zhao X, Li T, Song Q. Acheron regulates vascular endothelial proliferation and angiogenesis together with Id1 during wound healing. Cell Biochem Funct. 2011;29(8):636-40.
8. Schmidt M, Asirvatham AJ, Chaudhary J. Inhibitor of differentiation 1 (IDI) promotes cell survival and proliferation of prostate epithelial cells. Cell Mol Biol Lett. 2010;15(2):272-95
9. Li W, Zhang CH, Hong YL, Li J, Hu YM, Zhao CF. Inhibitor of DNA-binding-1/inhibitor of differentiation-I (ID-1) is implicated in various aspects of gastric cancer cell biology. Mol Biol Rep. 2012;39(3):3009-15.
10. Sharma P, Patel D, Chaudhary J. Idl and Id3 expression is associated with increasing grade of prostate cancer: Id3 preferentially regulates CDKNIB. Cancer Med. 2012;l(2):187-97
11. Pillai S, Rizwani W, Li X, Rawal B, Nair S, Schell MJ, et al. ID1 facilitates the growth and metastasis of non-small cell lung cancer in response to nicotinic acetylcholine receptor and epidermal growth factor receptor signaling. Mol Cell Biol. 2011;31(14):3052-67.
12. Li XM, Pang Y, Hou G. Expression and significance of Id-1 and HIF-1in esophageal carcinoma. Chinese J Clin. (Electronic Edition) 2013;7:1035-8.
13. Zhu B, Song C, Wang P. ID-I expression and clinical significance of ID1 in gastric cancer. J Shanghai Jiaotong University. 2013;33:298-302.
14. Guo Y. Expression and clinical significance of ID1 in gastric cancer. Chinese J Gen Surg. 2012;21:81-4.
15. Hu B, Chen ZH, Chen ZK, Wu SB, Ge J, Yuan WJ. Expression and correlation of Id-1 and MMP-9 in human colorectal carcinoma. China J Mod Med. 2010;20:2630-3.

ORIGINAL ARTICLE

Gravity of the non-authorized use of substances not intended for clinical use in invasive aesthetic procedures: the portuguese case

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13. Study conducted in Lisbon, Portugal

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SUMMARY

INTRODUCTION: *There is a worldwide increase in the number of invasive aesthetic procedures, and there is a general apprehension in medical societies towards the assurance of patient safety, that is dependent on the quality and certification of providers, of the materials and substances used, and where they take place.*

It is the main objective of this study to determine the perception of the gravity of non-authorized substances for clinical use in invasive aesthetic procedures among Portuguese plastic surgeons and its variation by the clinical sector of practice.

METHODS: *We proceeded to an inquiry by using a questionnaire, measured in a Linkert scale, and the collected data were statistically treated with a non-parametric Kruskal-Wallis test.*

RESULTS: *We obtained a 41,4% answer rate and a global perception that this is a serious problem – a median of 8,00 and mean of 7,45 points on a 1 to 10 scale. 70% of the plastic surgeons that answered the questionnaire work both in the private and public sector, 19% exclusively in the public sector and 11% only in private practice. The perception of the problem was most serious among those that work exclusively in the private sector (statistically significant difference).*

CONCLUSION: *The causes of the observed difference may reside in various reasons: the higher number of patients submitted to invasive aesthetic procedures exclusively in private practice; the higher perception of regulatory deficits in the private sector; scarce specific health politics for procedures outside the traditional boundaries of medicine; the difficulty for independent regulatory agencies to adopt effective measures.*

KEYWORDS: *government regulation; medical device legislation; esthetics; certification; plastic surgery.*

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INTRODUCTION

All around the world, worrisome reports of invasive procedures performed outside of health institutions — in business centers, hair salons, beauty salons, private houses, social gatherings, parties, etc. — have been surfacing. The risks of invasive procedures outside the clinical context are multiple: inadequate selection of patients, peer pressure to consent to treatment, professionals who are not qualified to perform the procedures or unable to control/treat possible complications, ineffective quality control, storage and dosage of substances administered, weak follow-up of patients in the period after the treatment, the possibility of alcohol and other substances being mixed with analgesic medication, among others[1].

To our knowledge, there are no published studies on the regulation of invasive aesthetic procedures in Portugal, on the number of treatments that are performed outside the clinical context, or about the impact they have on the health of patients. The real dimension of the problem of aesthetic procedures performed outside the clinical context and its direct impact on the health of patients is very difficult to determine with accuracy, since they are not, in their majority, reported to authorities or to the medical and scientific communities. You can infer, however, the seriousness of the matter through the perception of professionals who legally carry out these procedures and treat the complications derived from bad practices by others, and the regulatory mechanisms in relation to this specific issue.

METHODS

We used a survey by questionnaire as the empirical instrument of data collection for this study. The total universe of plastic surgeons who are active members of the Portuguese Society of Reconstructive and Aesthetic Plastic Surgery that were surveyed (128), resulted in 53 responses considered valid for the characterizing variable of "area of exercise of the professional activity," determining a response rate of 41.4%. For the characterization of this variable, we used the following categories: "public sector alone"; "private sector alone"; "public and private sectors."

The latent variable used was the perception that the use of devices or substances not authorized for clinical use, in aesthetic procedures, is a serious problem; this was measured using the Likert rating scale with 10 points, limited at the extremities — lower extremity: "completely disagree"; upper extremity: "agree completely."

The absence of normality in the distribution of levels obtained for the latent variable did not allow for the use of the Shapiro-Wilk test to determine the statistical relationships existing in the sample data. Therefore, it was necessary to use the Kruskal-Wallis non-parametric test[2,3]which allowed for the statistical analysis of the data. The statistical tests used 0.05 (5.00%) as a reference significance level.

RESULTS

Of the 53 observations, the majority (70%) corresponds to mixed work regimen (accumulation of functions), 19% are from the public sector exclusively and 11% from the private sector exclusively (Table 1).

The average of total value is 7.45 and the median, 8.00 in a Likert scale (1 to 10), reflecting a widespread perception that the problem of unauthorized substances for clinical use in aesthetic procedures is a serious one. The values obtained for the medians (and averages) of the seriousness of the problem, per sector of activity, are presented in Chart 1. This perception of the severity stands out on private sector category, with a median of 10.00 points (average of 9.33), i.e., 2.00 points above the value of the total median (1.88 point above the global average), while the categories that involve the public sector have lower scores — a median of 8.50 points (1.88 point above the global median) an average of 8.20 points (0.75 point above the global average) in the category of public sector alone; and median of 7.00 points (1.00 point below the global median) and an average of 6.95 points (0.50 point below the global average) in the mixed category with roles in the public and private sectors.

TABLE 1. NUMBER OF OBSERVATIONS PER SECTOR OF ACTIVITY AND ITS PERCENTAGE WEIGHT

Your clinical practice belongs to which sector of activity?	Number of observations	Percentage of total
Private sector alone	6	11.3
Public sector alone	10	18.9
Private and public sectors	37	69.8
Total	53	100

CHART 1. PERCEPTION VALUES OF THE GRAVITY OF THE USE OF SUBSTANCES NOT AUTHORIZED FOR CLINICAL USE IN AESTHETIC PROCEDURES PER SECTOR OF ACTIVITY

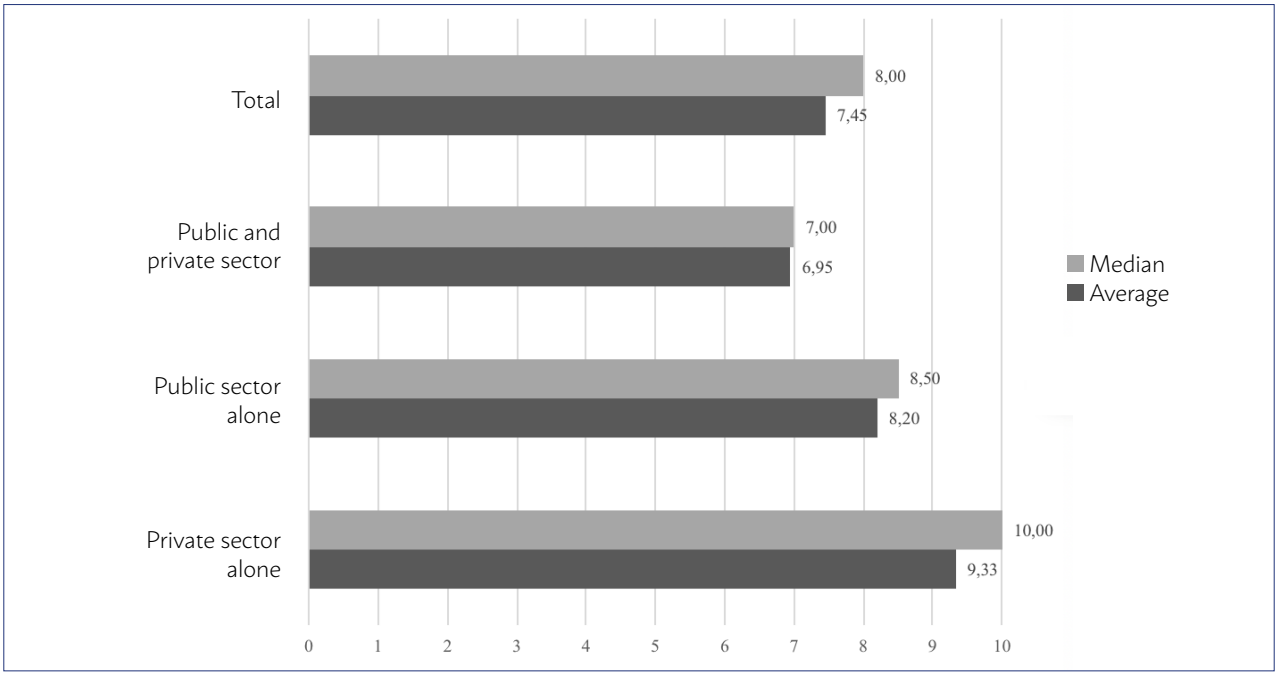

TABLE 2. SHAPIRO-WILK TEST APPLIED TO THE MEDIANS OF THE SCORES OF EACH SECTOR OF ACTIVITY (P-VALUE 0.05)

Your clinical practice belongs to which sector of activity? Statistics		Shapiro-Wilk		
		df	Sig.	
I believe that, in Portugal, the use of devices or substances not authorized for clinical use in aesthetic procedures is a serious problem.	Private sector alone	0.640	6	0.001
	Public sector alone	0.832	10	0.035
	Private and public sectors	0.898	37	0.003

TABLE 3. KRUSKAL-WALLIS TEST, WITH VARIABLE GROUPED PER "SECTOR OF ACTIVITY"

	I believe that, in Portugal, the use of devices or substances not authorized for clinical use in aesthetic procedures is a serious problem.
Chi-Square	6.112
df	2
Asymp. Sig.	0.047

It is necessary to determine whether these differences are statistically significant or may be due to random fluctuations in the data. Once it was determined that the distribution of scores was not normal using the Shapiro-Wilk test (p-value < 0.05), as described in Table 2, it was necessary to use the Kruskal-Wallis non-parametric test.

Examples of the use of this test can be found in other articles published in the scientific literature[3-6].

In Table 3 we can see that the p-value of 0.047 obtained from this test is below the level of significance (0.05).

From the results obtained by the comparison between the results of the Kruskal-Wallis test, through the stepwise method applied to the middle ranking of each sector of activity, it is possible to assume the existence of two subgroups of plastic surgeons: one that works exclusively in the private sector (with a higher value for the severity perception of the use of unauthorized substances in aesthetic procedures), and another which encompasses the public sector exclusively and the combination of public and private sectors (with lower perception values).

These facts lead to rejection of the null hypothesis of the test and the acceptance of the alternative hypothesis, which attests to the existence of statistically significant differences in the median of the perception of plastic surgeons on the seriousness of the use of substances not authorized for clinical use in invasive aesthetic procedures varying according to the sector of activity. Therefore, this perception is not uniform, and the relationship between it and the sector of activity is statistically confirmed by these data: the medians of scores of the private sector alone are higher than those that involve the public sector.

DISCUSSION

Iatrogenic lesions in plastic surgery caused by substances not suitable for clinical use are well

known, and there are cases described in which industrial silicone, liquid paraffin, soy oil, among others were used[7]. After the Second World War, this type of experimentation was frequent[8], but progressively its use was restricted and regulated. In recent years, there has been a growing number of aesthetic procedures at a global level: in 2015 more than 21 million surgical and non-surgical procedures were performed (an increase of 1 million since 2014); the application of botulinum toxin (the most frequently used) and the infiltration of hyaluronic acid had an increase of 4% and 7% compared to the previous year, respectively[9]. This boom in demand for aesthetic procedures seems to be followed by a growth in irregular situations. The widespread perception among the plastic surgeons who responded to the survey is that the use of substances or devices not approved for clinical use in aesthetic procedures is a serious problem, with a median of 8 points (average of 7.45) on a scale of 1 to 10. However, there is a statistically significant difference between the perception of plastic surgeons who work exclusively in the private sector and those who work in the public sector (exclusively or in a combination of roles). This difference may be due to:

- a greater number of patients who undergo this type of procedures conducted by plastic surgeons who devote their entire activity to the private sector, since these patients attend, almost exclusively, private institutions (except for major complications that require hospitalization). Consequently, they see a higher number of complications caused by the use of unauthorized substances used by others, which causes greater apprehension.

The work in private institutions alone has a different administrative paradigm from that of the public sector, which is more exposed to possible failures in the regulatory mechanisms of the professional activity and of the substances administered.

According to a survey conducted by the SPCPRE with its members and presented at the III Iberian Congress of Plastic Surgery (2016), half of the plastic surgeons recently observed patients that underwent implantation of devices or substances not authorized for clinical use[10]. This observation reflects a worrying reality for citizens' health and may be related to several factors. There are procedures that are not performed by qualified professionals, which can facilitate circumvention of the regulatory mechanisms and stimulate the unscrupulous use of appropriate substances; patients may not be adequately informed about the type of procedures, the substances used, and the qualifications of those who will practice invasive aesthetic procedures[11]; and the regulatory mechanisms may not be keeping up with this phenomenon, having difficulties with the effectiveness of their actions, because of:

The complexity inherent to certain specific issues, and in particular aesthetics, which are common to other countries, irrespective of their political and administrative reality.

The existence of different ethical foundations of Anglo-Saxon and European countries, with practical implications determinants of different administrative processes and organizational models — the Anglo-Saxon management and pragmatics, based on the implementation of effective, preventive, and formative mechanisms, in opposition to the European legalistic and conceptual model, with is repairing and far from the "practice"[12].

Cultural differences — beliefs and values of public administration, which are in conflict with this increasingly mixed model.

The relative lack of experience in this type of independence of regulatory agencies under the unifying State. There are similarities between the public and private administration in the Anglo-Saxon model, and the administrative process is seen as a means for achieving goals, with obvious gains in effectiveness and efficiency (instrumentalist); this model contrasts with the Continental European model, which sees the public and private administration as completely different spheres, with their own instruments — restraining even the public administration in a separate code of rules and legislative system, with few points of contact with the general law. Comparatively, the Anglo-Saxon approach there are similarities in the administrative functions of the public and private sector, differing mainly on strategic issues, while in the Continental European approach the similarities are avoided and the public administration is based on principles necessarily different from those adopted in private administration, very dependent on legal/normative regulations[12].

Aesthetic procedures introduce bodily changes with the aim of improving physical appearance and, consequently, increasing self-esteem. Some are not considered healthcare in Portugal (manicure, mechanical epilation, tattoos, for example) and are

performed in beauty and hair salons, for example, without the need for regulation of the provider. In the United Kingdom, on the other hand, the majority of these activities is seen as "health" activities and are regulated by the Ministry of Health. The already historically fluid boundaries between hygiene, medicine, and beauty are undefined when it comes to aesthetics.

The role of the State is also different in how it sees plastic surgery, depending on the type of procedure. Reconstructive plastic surgery procedures, as are included in the list of procedures offered by the National Health Service (SNS), inhabit the Continental European administrative paradigm with a strong presence of the State as provider, funder, and regulator and with a normative influence inherited from the French influence of the 18th and 19th centuries by Bonnin[13], based mainly on administrative law. Plastic surgery procedures, in its aesthetic aspect, are inserted in an open market, of private services and free competition, and fall more into the Anglo-Saxon paradigm typical of private providers of health care, in which the State retains only the role of regulator. This role of attentive spectator-intervener, in an open but regulated market, has long historical practice in common law countries, with a tradition and well-established mechanisms that have been refined over time[14]. In Portugal, there has been a gradual importation of management measures in public administration since the years 1980-1990 — New Public Management — with a greater increment in the period of financial rescues in the 1980s (mainly by the influence of the International Monetary Fund) and, more recently, in the troika intervention. Despite political and historical-institutional differences[15], as part of this package of measures independent regulatory institutions were created in various sectors. The need to regulate a market arises when one of the following assumptions is present: monopolies, externalities, asymmetric information, the need for continuity of service, anticompetitive behavior, the existence of a public good, when the bargaining power is unequal, if there is a shortage of resources and/or need of rationing, or due to a need for planning[16]. The public good identified, in the case of aesthetic procedures, is not only the direct result of the actions (improvement of the physical appearance or delay of the effects of aging) but the preservation of the security of these actions, i.e., mitigating or minimizing the health risk of these procedures. We can infer, then, that health or, more specifically, the maintenance of the health state in aesthetic procedures is also regarded as a public good.

The Law framework of independent administrative entities with regulation functions stipulates that "the regulation entities are collective persons of public law, with the nature of independent administrative entities with attributions in the field of regulation of economic activity, protection of services of general interest, protection of the rights and interests of consumers, and promotion defense of competition in private, public, social and cooperative sectors"[17]. In this way, the transition from the separation of State and market economy establishes a social value to the public regulation ensuring equality of opportunity for competition and guarantying the obligations of public service of the various sectors of society. There is, therefore, an understanding of regulation as a new form of relationship between the State and society, creating additional social value, by means of norms that ensure the balanced functioning of the open system (market) in agreement with public objectives.

The tendency for the formation of independent administrative authorities have in common at least three characteristics: the recognition that regulation has a specific logic and should, whenever possible, be separated from the political logic; the recognition that the market has flaws and not always gives guarantees of finding the balance of the system for the fulfillment of their public obligations; the recognition that the State is also flawed due to its administrative burden, its causal guidelines, corporatism, among others, which restrict the public interest[18].

In the health sector, these management modifications are mirrored in the plan of Reduction and Improvement of the Central Administration (Premac), considered by the tutelage as "[...] the beginning of a new phase of the reform of Public Administration, in order to make rational and efficient use of public resources. [...] Indeed, more than ever, the simultaneous achievement of the objectives of rationalization of state structures and better use of human resources is crucial in the process of modernization and optimization of the functioning of Public Administration".[19] This focus on the pursuit of efficiency and rationalization of resources was subsequently achieved by the creation of new independent administrative bodies with powers of supervision and regulation, such as the General Inspection of Health

Activities (Igas), or the independence of already existent autonomous structures, such as the Health Regulatory Body (ERS). This was created in 2005, in the process of autonomizing the regulatory agencies — which was already a management measure part of the New Public Management.

From the normative point of view, until recently the blurring of what is a health and a medical procedure persisted. We found that the Portuguese legal system does not include a isolated definition of the medical procedure, but there is a reservation of exercise on procedures of medical diagnosis, therapeutic prescription, and autonomous management of patients[20]. In addition to the difficulty in clarifying exactly what is and what is not a medical procedure, or a procedure restricted to health professionals, there is a normative vacuum in the classification of what is an invasive procedure". If one considers, on the one hand, that a tattoo, a body piercing, laser/pulsed light hair removal are invasive because they penetrate completely or partially the protective barrier of the skin, and because they can cause serious damage, temporary or permanent (transmission of diseases, hemorrhages, skin burns, eye injuries, among others), in practice, they are often used by people without any basic health training, performed solely based on "common sense". The project for a legal definition of Health Procedure is restricted to eight professions, trying to refer to a specific procedure of each professional activity, but described in very general terms[21]. Within these professions, it is understood from the remaining national regulatory framework that "acts of medical diagnosis (unlike the nursing diagnosis), prescribing treatment (unlike the non-therapeutic prescription) and autonomous management of patients constitute acts of absolute exclusivity of the medical profession. [...] The other acts, functionally considered as acts of the medical profession, appear to be susceptible to being delegable to other health professionals, including nurses, upon authorization of the Order of Doctors and within limits defined by a initial medical prescription"[20].

The legislation itself that defines the competencies of each entity with the power to regulate in the area of health has an overlapping of competences and their limits of action, which can induce the existence of "gray areas" that are the legal responsibility of several entities, but which, in practice, are not supervised by none. Perhaps to overcome some of these shortcomings, a protocol of cooperation between the ERS and the professional orders of the health sector was recently signed to "cooperate in the sharing of resources, both human and technical, and of knowledge with the purpose of improving the exercise of their respective attributions" and provides for the participation of professional orders in the planning and execution of supervisory actions, surveys, monitoring, and periodic evaluations"[22].

The Botulinum Toxin, for example, authorized by Infarmed for administration by "physicians ex-

TABLE 4. ENTITIES WITH RESPONSIBILITY IN THE REGULATION IN THE HEALTH AREA AND THEIR POWERS

Health Regulatory Entity	Regulates and inspects only the institutions registered as providers of health care — clinics, private clinics, hospitals — and has no powers of supervision of certification of the provider and of the quality of most substances and devices used in aesthetic procedures. However, it is stipulated that its influence extends to "any other places where it is materially identified the practice of activities that integrate the concept of health care provision, as defined by the ERS"[35].
General supervision of health activities	Its mission is to audit, inspect, supervise and develop disciplinary action in the health sector in order to ensure compliance with the law and high technical levels of performance in all areas of activity and the provision of health care through services, establishments and bodies of the Ministry of Health [...], or protected by it, either by private entities, collective persons, profit or non-profit". This agency has the power to "supervise the compliance with legal provisions and regulations and the applicable guidelines, as well as the quality of services rendered by any entity or professional in the field of health activities, through the implementation of actions of audit, i, spection and supervision"; and to carry out regular services of inspection at the level of safety and quality, in conjunction with the General Directorate of Health (DGS)"[19].
Infarmed	Has the mission to "contribute to the formulation of national health policies", "regulate, evaluate, authorize, punish, supervise, check analytically, and ensure the monitoring and control of research, production, distribution, marketing and use of medicinal products for human use and health products, which includes medical devices and cosmetics and personal hygiene products," as well as "ensure the regulation and supervision of the activities of research, production, distribution, marketing and use" of these products and devices[36].
Order of Doctors	Has the powers to regulate the access and the exercise of the profession of physician and exercise disciplinary power over them: its code of ethics provides disciplinary sto doctors doctors who go beyond the limits of their powers, taking into account the specialties and subspecialties — usually after complaint[37]. Should collaborate with other entities of the Public Administration in matters of public interest related to the medical profession and contribute to the protection of the health of citizens and the rights of patients, as well as participate in the drafting of legislation which relates to the access to and pursuit of the medical profession[38].

perienced in the application of botulinum toxin" (informative leaflet), is sold only with a prescription. However, this type of medication in pharmacies is easily obtained for application by non-doctors, and there is not a mandatory registration of the prescriber per package. In Spain, for example, the government has made some progress in regulating this type of procedures through the control of the circuit of the active substances and the certification of the provider. The Botulinum Toxin used with aesthetic indication has the same regulation of medicines for hospital use and can only be administered by "physicians with the proper qualification, experience in the treatment, and with the appropriate equipment and instrumentation. As a result, and to ensure that the proper administration of these treatments can only be performed in hospitals or health care institutions that are duly authorized in accordance with the legislation in force"[23]. The transport, storage, and recording of packages follow the same requirements of medicines for hospital use only. There is also a requirement that the doctor fills out a standard sheet with the reference of the packaging, the date, and the dose of administration, for reporting to the health authority, as well as an informational letter for patients who will receive the medication for the first time, containing the goals of the treatment, an explanation of the procedure and the risks involved.

Subcutaneous fillers to gain volume are also considered medical devices for exclusive medical use, but there is little control of their use. There are also reported cases of fraud and counterfeiting of facial filler products based on hyaluronic acid[24]. With globalization and the expansion of the digital commerce, nowadays it is easier to bypass regulating agencies and order substances and devices, over the internet, that are released for "clinical use" and "without control by national and international medication agencies."

However, this problem is not confined to the national reality; it is an international phenomenon. Rufai and Davis believe that cosmetic surgery in the United Kingdom is also "ubiquitous, unregulated, and seductive," there is no great adherence to national guidelines[25]. The lack of control over the qualification of suppliers may be a predisposing factor to the use of substances not authorized for clinical use. Then, the question that arises is: who has the qualifications to provide this type of care?[26] The use of training programs, the regulation of sectors, professional certification and the constant and systematic evaluation along the professional career are contemporary manifestations of the logic of scientific management. The desire of societies for certification, accuracy, organization and appropriate categorization of the world to find a sense in it is, without a doubt, as strong today as it was at the foundation of scientific management[27], and is compatible with the need for control and security of citizens themselves.

In the USA, the most important factor for patients in choosing a provider of aesthetic procedures is the certification in plastic surgery[28,29], and the greatest fear is to obtain "bad results"[30]. Even so, many of the providers of aesthetic procedures in the country are not certified by the American Society of Plastic Surgeons (ASPS)[31] or by other medical societies. A growing phenomenon is also the emergence of inaccurate qualifications that do not correspond to any type of degree or certification — "aesthetic surgeon", "aesthetic doctor", "facial plastic surgeon", "cosmetic surgeon", as a way to disguise the absence of a certification recognized by the national medical associations. Usually, the provider with this type of (self) qualification is a general medical practitioner or has another specialization less "seductive" for aesthetic procedures, or is a non-medical healthcare professional, or even someone without any qualification in health care provision. This type of marketing maneuver is frequent and is also observed abroad, with several calls for government intervention.[26,32].

In Portugal, the regulation of advertising of health procedures has recently been legislated as an attribution of the ERS and, in this context, it is deemed to be unlawful if it induces the patient to procedures that are unnecessary, harmful or without prior diagnosis or evaluation by a "qualified professional"; and "every time the professional favored by the practice of health advertising is also the health care provider, without actually being one, or if they are a health care provider but do not comply with the requirements of activity and operation, are not duly registered with the regulatory body of health and do not hold a license of operation, when applicable" .[33]

Recently, there has been a controversial attempt to regulate the activity of beauty salons by the European Committee for Standardization (CEN). About two years ago, the organization gathered a group of experts for the development of the CEN-409 normative for beauty salons[34]. This project was withdrawn in 2016 due to numerous complaints from local com-

mittees and national entities related to health, due to the possible ethical commitment to the safety of patients if the law were approved. On the basis of these allegations was the type of procedures that could be performed in "beauty salons" or equivalent, such as laser treatments, IPL, mesotherapy, ultrasounds, radiofrequency, chemical peels, without restrictive criteria of power, penetration, and depth, for example. According to the entities that complained about the normative proposal, these invasive aesthetic procedures should be performed by physicians or under the supervision of one, based on a clinical diagnosis, and in institutions properly regulated and supervised.

In practice, there seems to be a lack of supervision of economic activities not registered as health establishments based on clinical criteria — including "aesthetic centers", hair salons and beauty salons. There seems to be only, in practice, control of the activity in terms of quality of infrastructures, security, and tax transparency by means of the Economic and Food Safety Authority (ASAE).

CONCLUSION

Although the entities with the power to regulate the health sector have responsibilities defined by law to act on the various market segments of aesthetics — with an overlapping of functions — and have the duty to cooperate among themselves, there is a widespread perception among plastic surgeons who are accredited providers that the use of substances not authorized for clinical use in invasive aesthetic procedures is a serious problem. This perception is more pronounced in surgeons who exercise their activity exclusively in the private sector.

The regulation of the various segments that constitute the circuit of these substances — production, marketing, distribution, utilization, monitoring, inspection, evaluation — by agencies that are still incorporating instruments of independent or autonomous action, imported from countries with different administrative histories and cultures (management), may not prove to be effective in a real level, with the aggravating circumstance that this market also finds difficulties of effective regulation at an international level. The fact that this is a recent problem at a global scale has not yet allowed for the creation of specific health policies and effective administrative solutions.

Greater supervision and transparency are necessary in order to increase the safety of patients and the reliability of results[26].

Acknowledgments

Portuguese Society of Reconstructive and Aesthetic Plastic Surgery

RESUMO

INTRODUÇÃO: *Os procedimentos estéticos invasivos estão a aumentar globalmente, e são acompanhados por uma apreensão das sociedades médicas sobre a segurança desses procedimentos, dependentes da qualidade e certificação dos prestadores, dos dispositivos e substâncias utilizados e do local onde são efetuados. O presente estudo procura aferir a percepção dos cirurgiões plásticos portugueses sobre a gravidade da utilização de substâncias não autorizadas para uso clínico em procedimentos estéticos, e a sua variação consoante o setor em que exercem a atividade clínica.*

MÉTODOS: *Foi utilizado um inquérito sob a forma de questionário, medido numa escala de Likert, e os dados foram tratados estatisticamente pelo teste não paramétrico de Kruskal-Wallis.*

RESULTADOS: *Obteve-se uma taxa de resposta de 41,4% e a perceção global é a de que o problema é grave — mediana de 8,00 e média de 7,45 numa escala de 1 a 10. Setenta por cento dos cirurgiões plásticos que responderam ao inquérito trabalham num regime misto, 19% exclusivamente no setor público e 11% apenas no setor privado. A percepção do problema como mais grave (diferença estatisticamente significativa) foi observada na atividade exclusiva no setor privado.*

CONCLUSÕES: *A diferença observada pode dever-se a vários fatores: à maior observação de pacientes submetidos a esses procedimentos exclusivamente no setor privado; à maior percepção de déficits de regulação no setor privado; ao déficit de políticas de saúde específicas a técnicas utilizadas fora do contexto tradicional da medicina; à dificuldade de as agências administrativas reguladoras independentes adotarem práticas efetivas no setor privado da saúde.*

PALAVRAS-CHAVE: *Regulamentação governamental. Legislação de dispositivos médicos. Estética. Certificação. Cirurgia plástica.*

REFERENCES

1. ASPS, ASAPS. Injectables and Fillers: Legal and Regulatory Risk Management Issues. *Plast Reconstr Surg*. 2006;118(Suppl):129S-132S. doi:10.1097/01.prs.0000214727.21086.0c.
2. Kruskal WH, Wallis WA. Use of Ranks in One-Criterion Variance Analysis. *J Am Stat Assoc*. 1952;47(260):583. doi:10.2307/2280779.
3. Correia P, Carrapato P, Bilhim J. Administração hospitalar em Portugal: relação entre antiguidade e envolvimento laboral, e implicações para o risco de saída. *J Bras Econ da Saúde*. 2016;8(2):73-79. doi:10.21115/JBES.v8.n2.p73-79.
4. Catarino J, Correia P. Receitas fiscais e tributação geral sobre o consumo em Portugal : um estudo sobre eventuais assimetrias do comportamento dos sujeitos passivos do imposto no final da primeira década do século XXI [Tax Revenue and General Taxation on Consumption in Portuga. *Rev da FAE*. 2016;19(1):6-17.
5. Correia P, Catarino J. Ingreso Bruto Tributable del IVA: Evidencia de Diferenciación de los Municipios de la Costa Portuguesa [Gross Revenue for VAT Tax: Evidence of Differentiation at the Portuguese Littoral Municipalities]. *Rev del CLAD Reforma y Democr*. 2016;64:225-246.
6. Correia P, Videira SA. Troika's Portuguese Ministry of Justice Experiment, Part II: Continued positive results for civil enforcement actions in Troika's aftermath. *Int J Court Adm*. 2016;8(1):20. doi:10.18352/ijca.215.
7. Grotting JC, Neligan PC. *Plastic Surgery: Volume 5: Breast*. Elsevier Health Sciences; 2012. https://books.google.pt/books?id=DT7JhnV1_B4C.
8. Goulian D. Current status of liquid injectable silicone. *Aesthetic Plast Surg*. 1978;2(1):247-250. doi:10.1007/BF01577957.
9. International Society of Aesthetic Plastic Surgery. *ISAPS International Survey on Aesthetic/Cosmetic Procedures Performed in 2015*.; 2016. https://www.isaps.org/Media/Default/global-statistics/2016 ISAPS Results.pdf.
10. Sociedade Portuguesa de Cirurgia Plástica Reconstrutiva e Estética, Sociedad Española de Cirurgía Plastica Reparadora y Estetica. Segurança e Saúde Pública em Cirurgia Plástica. 2016. cprelisboa.org.
11. Manstein C. Preferences regarding board certification and organization relating to aesthetic surgery. *Plast Reconstr Surg*. 1998;(110):863–864.
12. Bilhim J, Ramos R, Pereira LM. Paradigmas administrativos, ética e intervenção do Estado na economia: o caso de Portugal. *Rev Digit Derecho Adm*. 2015;0(14):91. doi:10.18601/21452946.n14.07.
13. Bonnin CJ. *Les Principes D'administration Publique*. 3rd ed. Paris: Renaudiere, Imprimeur-libraire; 1812.
14. Bilhim J. *Ciência Da Administração*. Lisboa: Instituto Superior de Ciências Sociais e Políticas; 2013.
15. Moreira V. *Autoridades Reguladores E Independentes – Estudo E Projeto de Lei-Quadro*. Coimbra: Coimbra Editora; 2003.
16. Day P, Klein R, Redmayne S. *Why Regulate?: Regulating Residential Care for Elderly People*. Bristol: The Policy Press; 1996.
17. Assembleia da República. *Lei N.º 67/2013 de 28 de Agosto: Lei-Quadro Das Entidades Administrativas Independentes Com Funções de Regulação Da Atividade Económica Dos Setores Privado, Público E Cooperativo*. Portugal: Diário da República; 2015:7566-7584.
18. Pereira PT. Governabilidade, grupos de pressão e papel do Estado. In: *Actas*. Lisboa: APCP; 2001.
19. Ministério da Saúde. *Decreto-Lei Nº 33/2012 de 13 de Fevereiro*. Diário da República; 2012:3626-3633.
20. Temido M, Dussault G. Papéis profissionais de médicos e enfermeiros em Portugal : limites normativos à mudança. *Rev Port Saúde Pública*. 2014;2(1):45-54.
21. Assembleia da República. *Proposta de Lei Nº 34/XIII*.; 2016.
22. Entidade Reguladora da Saúde, Ordem dos Médicos, Ordem dos Enfermeiros, Ordem dos Farmacêuticos. *Protocolo de Colaboração, de 18 de Novembro de 2016*. Portugal; 2016. https://www.ordemdosmedicos.pt/send_file.php?tid=ZmljaGVpcm9z&did=4a1590df1d5968d41b855005bb8b67bf.
23. Agencia Española de Medicamentos y Productos Sanitarios, Ministerio de Sanidad y Política Social. *Circular 2/2010*. Espanha: CAIB; 2010.
24. INFARMED I.P. *Circular Informativa 044/CD de 2010*.; 2010.
25. Rufai SR, Davis CR. Aesthetic surgery and Google: ubiquitous, unregulated and enticing websites for patients considering cosmetic surgery. *J Plast Reconstr Aesthetic Surg*. 2014;67(5):640-643. doi:10.1016/j.bjps.2014.01.009.
26. Hruza GJ. Clinicians Performing Cosmetic Surgery in the Community: A Nationwide Analysis of Physician Certification. *Plast Reconstr Surg*. 2015;136(2):274e-275e. doi:10.1097/PRS.0000000000001420.
27. Frederickson HG, Smith KB, Larimer CW, Licari MJ. *The Public Administration Theory Primer*.; 2012. doi:10.5860/CHOICE.41-2423.
28. Waltzman JT, Scholz T, Evans GRD. What Patients Look for When Choosing a Plastic Surgeon. *Ann Plast Surg*. 2011;66(6):643-647. doi:10.1097/SAP.0b013e3181e19eeb.
29. Nowak LI, Washburn JH. Patient Sources of Information and Decision Factors in Selecting Cosmetic Surgeons. *Health Mark Q*. 1998;15(4):45-54. doi:10.1300/J026v15n04_03.
30. Galanis C, Sanchez IS, Roostaeian J, Crisera C. Factors Influencing Patient Interest in Plastic Surgery and the Process of Selecting a Surgeon. *Aesthetic Surg J*. 2013;33(4):585-590. doi:10.1177/1090820X13481228.
31. Housman TS, Hancox JG, Mir MR, et al. What Specialties Perform the Most Common Outpatient Cosmetic Procedures in the United States? *Dermatologic Surg*. 2007;34(1):1-8. doi:10.1111/j.1524-4725.2007.34000.x.
32. Berry MG. Commentary to "Aesthetic surgery and Google: Ubiquitous, unregulated and enticing websites for patients considering cosmetic surgery." *J Plast Reconstr Aesthetic Surg*. 2014;67(5):644-645. doi:10.1016/j.bjps.2014.02.005.
33. Ministério da Saúde. *Decreto-Lei Nº 238/2015 de 14 de Outubro*. Portugal: Diário da República; 2015:1-26.
34. European Comittee for Standardization. *CEN/TC 409: Beauty Salon Services - Requirements and Recommendations for the Provision of Service*. DIN; 2016.
35. Ministério da Saúde. *Decreto-Lei N.º 126/2014 de 22 de Agosto*. Portugal: Diário da República; 2014:4400-4416.
36. Ministério da Saúde. *Decreto-Lei Nº 46/2012, de 24 de Fevereiro*.; 2012:884-890.
37. Ordem dos Médicos. *Regulamento nº14/2009 - Código*. Portugal; 2009:1355-1369.
38. Assembleia da República. *Lei Nº 117/2015 de 31 de Agosto. Estatuto Da Ordem Dos Médicos*. Diário da República; 2015:7566-7584.

ORIGINAL ARTICLE

Reality of premature ovarian failure in Argentina

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SUMMARY

Premature Ovarian Insufficiency is defined as a decline in ovarian function that is accompanied by two biochemical determinations of Follicle Stimulating Hormone in hypergonadotropic values, in addition to low levels of circulating estrogens in women under 40 years old. Although some of its possible etiologies are recognized and diagnosed, most of the time, its cause remains unknown. It is a pathology with medical, psychological, and reproductive implications. Patients may experience climacteric symptoms, infertility, and emotional distress. In the medium and long term, cardiovascular and bone health can be affected, and some degree of cognitive deterioration can be evidenced. The therapeutic approach needs to be comprehensive for the patient and multidisciplinary. SAEGRE created in Argentina an interhospital network dedicated to gathering relevant statistical information regarding this and other pathologies in order to provide better assistance for these patients.

KEYWORDS: *Premature ovarian failure, premature ovarian insufficiency, Infertility, Ovarian reserve, hypoestrogenism, Hypergonadotrophic amenorrhea, Occult ovarian failure, Ovarian biochemical failure.*

INTRODUCTION

Premature ovarian insufficiency (POI), formerly known as "primary ovarian failure," is characterized by an accelerated depletion of ovarian reserve with a decrease in the number of residual follicles and deficiency in sex hormones, which is manifested in women as subfertility and hypoestrogenism years and even decades before the normal age of menopause. The subtleties in the clinical presentation and the relative lack of knowledge of the population with respect to this condition may lead to a delay in diagnosis and subsequent treatment. The condition is defined as a hypergonadrophic amenorrhea that occurs before the age of 40 years. It is detected in 5% to 10% of women who are evaluated with amenorrhea. Its prevalence in the general population is estimated with oscillation between 0.3% and 0.9% of women[1].

There are several causes described that can cause follicular atresia or dysfunction [1]. Table 1.

The most common symptoms are changes in the cycle, amenorrhea, and consequent infertility. There

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TABLE 1. CAUSES OF PREMATURE OVARIAN INSUFFICIENCY

SPONTANEOUS	INDUCED
Genetic	Radiotherapy
Autoimmune	Chemotherapy
Infectious	Surgical
Idiopathic	
Environmental toxins	

may also be symptoms of hypoestrogenism as hot flushes, vaginal dryness, sleep disorders, and irritability.

Women with premature ovarian insufficiency left untreated have a higher risk of osteoporosis and cardiovascular disease and all the effects of menopause, reducing their quality of life and leading to early mortality [2].

The diagnosis of these patients requires at least two determinants of FSH in the early follicular phase (between the 2nd and 5th day of the cycle). The presence of hypogonadotropic hypogonadism in a woman younger than 40 years old is consistent with a diagnosis of POI.

In Argentina, there are no studies on the prevalence in the population. For this reason, the Argentine Society of Gynecological and Reproductive Endocrinology (SAEGRE) has created the Interhospital Network of Endocrinology to build databases that allow us to improve the statistics on these matters.

METHODS

The SAEGRE Hospital Network of Gynecological and Reproductive Endocrinology proposed an epidemiological study with the purpose of learning the relevant aspects of POI in Argentinian women. A descriptive, observational, multicenter study collaborative in nature.

Public hospitals of the Autonomous City of Buenos Aires and the first sector of the province of Buenos Aires participated. We analyzed clinical histories of women who attended external offices of Gynecological Endocrinology from January 1st, 2011 to December 31st, 2016 (5 years analysis).

The inclusion criteria were women younger than 40 years with two determinants of FSH > or = 40 mUI/ml; other causes of primary and secondary of non-hypergonadrophic amenorrhea were excluded. The information of POI patients stored in the database was: sociodemographic factors (age, identification data, nationality, place of residence, occupation), family history (maternal age of menopause, POI in the family), personal history (pregnancies, autoimmune diseases, chromosomal alterations, gynecologic surgery, chemotherapy and radiotherapy, infections, smoking, alcoholism, drugs), reason for consultation.

RESULTS

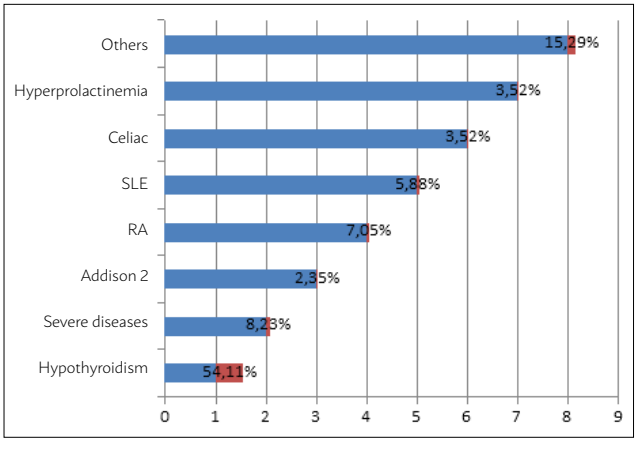
We studied 302 patients treated at the Endocrinology clinics of hospitals members of the SAEGRE Interhospital Network who met the inclusion criteria. The average age of our population was 33 years, with two cases of 19 years old as the younger age with a POI diagnosis. A total of 11.9% of the patients had a history of POI in their family.

As to his personal history, 50.3% did not have any previous pregnancy, were nulliparous, and of the remainder 49.7%, 6.9% had had previous abortions, with no pregnancies to term.

Of the total patients, 28.1% had an associated medical pathology, of which 27.5% of the total (83 patients), presented associated autoimmune diseases (among which autoimmune thyroid diseases predominated). A total of 54.11% were patients with hypothyroidism, followed by patients with Graves-Basedow diseases at 8.23%, and rheumatoid arthritis in 7.05%. There were other pathologies in smaller percentages, such as Systemic Lupus Erythematosus, hyperprolactinemia, Celiac, Addison. See Figure 1

A total of 7.6% of the 302 patients had received chemotherapy/radiotherapy due to prior oncologic diseases. A percentage of 8.20% had had gynecolog-

FIGURE 1. PATHOLOGIES ASSOCIATED WITH POI IN OUR POPULATION.



ical surgeries, of which 52% had been subjected to bilateral oophorectomy, 24% to total anexohysterectomy, 12% to bilateral adnexectomy, and 8% to unilateral oophorectomy.

Of the total number of patients, 2.3% had had some type of infection, most of them (57.1%,) had had tuberculosis, followed by HIV infection (28.5%).

Among the changes in the cycle that were the main reason for consultation, 55.6% had amenorrhea, followed by 40.3% who presented oligomenorrhea; only 0.33% had regular normal cycles.

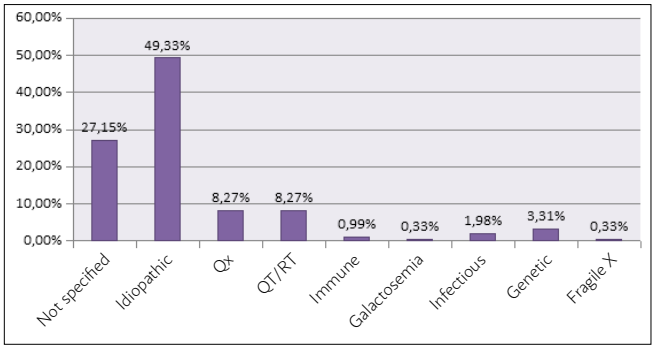
Regarding the causal pathology of POI, 49.33% had idiopathic causes, followed by 27.1% in which the cause was not specified (due to an incomplete study of the causes), with equal percentages of 8.2% were causes due to gynecological surgery and previous chemotherapy/radiotherapy. A total of 3.3% of the POI causes were genetic; in smaller percentages, there was infectious pathology, galactosemia, fragile X syndrome. See Figure 2.

DISCUSSION

As mentioned earlier, the evidence of hypogonadotropic hypogonadism in the context of a change in the cycle of women younger than 40 years old is consistent with a diagnosis of POI [2]. It is important to note that unlike women with menopause, which marks an irreversible state of ovarian senescence, women with POI continue with some degree of ovarian function. Therefore, fluctuations in the ovarian hormones, as well as occasional spontaneous ovulations may be found in a small proportion of women with POI, which adds complexity to the clinical picture.

A comprehensive review of medical and family history can provide certain information and guide doctors in the process of reaching a timely and correct diagnosis. Special attention should be given to the chronology of events, individual exposures, and personal and family history. In our population, 12% presented a history of first-degree ovarian failure. A detailed menstrual history containing the age of menarche, as well as the frequency and the menstrual pattern is useful to determine any change in the menstrual period at the beginning of the course of events. It is noteworthy that about 50% of our patients had a history of oligomenorrhea cycles during the years prior to diagnosis. Medical conditions, medications prescribed, or previous gonado-toxic exposures should be considered since they can directly affect the ovarian function and reserve. In these cases, the patient must be advised of different techniques to preserve their future fertility, that is why different medical specialties should know the impact of certain drugs and perform a multidisciplinary work on fertile women.

FIGURE 2. CAUSAL PATHOLOGIES ASSOCIATED WITH POI IN OUR POPULATION 2011-2016



It is important to consider the documentation of any endocrinopathy (such as type 1 diabetes mellitus and hypothyroidism associated with Hashimoto's thyroiditis) since it is common for these conditions to occur in patients with POI[3].

To determine the causes, it is useful to perform certain studies such as karyotype; genotyping for FMR (if there is a family history of POI); thyroid and adrenal glands antibodies. In most cases, the cause may not be evidenced; therefore, it is considered idiopathic. However, it is vital that these women are studied to demonstrate a possible cause, which can result in a specific treatment and assistance that is personal, familiar and includes possible future offspring.

Women with POI are subject to the entire range of symptoms experienced by women in menopause due to the presence of hypoestrogenism. Among these are the classic symptoms of menopause such as hot flushes, night sweats, sleep disorders, unstable state of mind and sexuality problems derived from dyspareunia, vaginal dryness, and decreased libido. These symptoms may be particularly intense in cases of iatrogenic POI, and the severity of the disease is often so great that it significantly affects the woman's quality of life, psychological well-being, and the intimate relations.

These women have a reduced life expectancy, in large part due to cardiovascular mortality and cerebrovascular accidents, changes in cognition, and low bone mass with an increased risk of fracture [4,5].

The adequate systemic replacement of hormones is critical to the control of symptoms, while local estrogens may be required to treat local symptoms such as dyspareunia or other genitourinary symptoms [6-11]. One thing that should be considered is that hormone therapy should be administered until the age of physiological menopause, i.e., around 51 years. This therapy is recommended by International Medical Societies, and its use does not present the same risks as the hormone therapy used during the climacteric.

Regarding the fertility, there are markers of ovarian reserve such as the FSH, FSH-estradiol binomial, HAM and recount of antral follicles, which can predict a low response to controlled ovarian stimulation and, therefore, a greater number of cancellations and lower response to treatments of IVF [12-14]. Nonetheless, none of them can predict the rate of pregnancy. The age of the patient is the most important marker to predict pregnancy rate since it usually is an indicator of oocitary quality [15].

There is a certain hormonal transition that is identified until the definitive establishment of the premature ovarian insufficiency diagnosis or primary ovarian failure, both biochemically as clinically.

It is the general gynecologist's job to detect this transition and timely forward the patient to a reproductologist and offer alternatives for her reproductive present or future.

There are two phases between the normal class and the establishment of a definitive diagnosis of POI:

- Occult ovarian failure, in patients who have values of FSH <10 UI/ml in the early follicular phase (FFT), but Estradiol > 60pg/ml in FFT, regular menstrual cycles, and normal or decreased fertility according to age. The younger the age, the higher the possibility of pregnancy.
- Biochemical ovarian failure, in patients who have values of FSH > 10 UI/ml, but < 25 IU/ml in FFT, regular or irregular menstrual cycles, or frequent polymenorrhea. The fecundity in these patients may be reduced compared with women of the same age and preserved ovarian reserve, but not necessarily reduced in comparison with older patients [16].

The reproductologist must identify the moment in which the patient is to offer therapeutic alternatives available, according to the desire of the patient.

If the patient still has regular cycles, she may or may not express a desire to get pregnant:

- If pregnancy is not desired at the time of diagnosis, it is of the utmost importance to inform her of the possibility of postponing maternity through the cryopreservation of oocytes and/or embryos, always when the patient is younger than 35 years, and genetic causes such as fragile X syndrome or Turner have been discarded. It is essential to provide clear and accurate information about the minimum number of oocytes required for cryopreservation to achieve a reasonable rate of pregnancy. Once the diagnosis of POI is established, it is not possible to use cryopreservation in oocytes. Once this treatment is finished, the patient should be advised about a contraceptive method.
- If pregnancy is desired, ovulation monitoring can be offered for a few months, if the patient has not had yet time for a search or IVF/ICSI, for patients older than 35 years, or searching for more than 6 months. |f the patient "qualifies" for IVF, different schemes of stimulation can be used.

If the patient already has no regularity in their menstrual cycles or amenorrhea:

- If there is no desire for fertility, advise on a contraceptive method.
- If there is a desire for fertility, without a doubt, the treatment of choice in these patients is oocyte donation, with pregnancy rates of 45-60% [17,18].

It is essential to mention the importance of preserving fertility in cases in which, for reasons such as cancer or other non-malignant systemic diseases, the patient must be exposed to treatments such as chemotherapy, radiotherapy and/or surgery, which can affect the ovarian reserve.

The methods currently available are:

- *Ovarian transposition:* for cases of Radiotherapy, oophoropexy or external shielding of the ovaries can be performed [19].
- *Use of analogs of GnRH:* There are two reviews and a meta-analysis that demonstrate a significant benefit in reducing the risk of ovarian failure [20,21].
- *Vitrification of oocytes and/or embryos:* by means of controlled ovarian stimulation and with cur-

rent techniques that facilitate the recovery of such cells and/or embryos.

- *Freezing and implantation of ovarian tissue:* proposed for children or adolescents even if it is still an experimental procedure.
- *Obtaining immature oocytes and in vitro maturation:* does not require stimulation but has a lower yield of fecundity.

It is of utmost importance to know that this entity has a high emotional impact on women, therefore, in addition to the recommendation of hormone therapy and advice on fertility, psychological support must be provided to promote acceptance and the best response during follow-up.

CONCLUSIONS

Premature ovarian insufficiency is a common condition that can have far-reaching consequences for the physical and mental health of women. Early diagnosis and treatment are crucial. We are Increasingly seeing improved access to new diagnostic tools and more effective options to preserve the ovarian function in young women who are treated for cancer as well as for fertility gain in other causes of POI. We need to understand the safety of hormone therapy for these women and achieve an effective treatment for improving the results in their health in the short, medium and long term. We believe that understanding our population will improve our knowledge and the treatment of this condition.

RESUMO

Insuficiência ovariana primária é definida como um declínio da função ovariana acompanhado por dois determinantes bioquímicos do Hormônio Folículo Estimulante em valores hipergonadotróficos, além de baixos níveis de estrogênios circulantes em mulheres com menos de 40 anos de idade. Embora algumas das suas possíveis etiologias serem reconhecidas e diagnosticadas, na maioria das vezes sua causa permanece desconhecida. Trata-se de patologia com a implicações médicas, psicológicas e reprodutivas. Os pacientes podem vivenciar sintomas climatéricos, infertilidade e problemas emocionais. A médio e longo prazo, a saúde cardiovascular e óssea pode ser afetada, e algum grau de deterioração cognitiva pode ser observado. A abordagem terapêutica precisa ser abrangente para o paciente e multidisciplinar. A SAEGRE criou na Argentina uma rede interospitalar dedicada a reunir informações estatísticas relevantes sobre esta e outras patologias, a fim de proporcionar uma melhor assistência para esses pacientes.

PALAVRAS-CHAVE: *Insuficiência ovariana primária, falência ovariana precoce, infertilidade, reserva ovariana, hipoestrogenismo, amenorréia hipergonadotrófica, insuficiência ovariana oculta, insuficiência ovariana bioquímica.*

REFERENCES

1. Rafique S, Sterling EW, Nelson LM. A new approach to primary ovarian insufficiency. Obstet Gynecol Clin North Am. 2012;39(4):567-86.
2. Rebar RW. Premature ovarian failure. Obstet Gynecol. 2009;113(6):1355-63.
3. Ayesha, Jha V, Goswami D. Premature ovarian failure: an association with autoimmune diseases. J Clin Diagn Res. 2016;10(10):QC10-2.
4. Rocca WA, Grossardt BR, Miller VM, Shuster LT, Brown RD Jr. Premature menopause or early menopause and risk of ischemic stroke. Menopause. 2012;19(3):272-7.
5. Archer DF. Premature menopause increases cardiovascular risk. Climacteric. 2009;12(Suppl 1):26-31.
6. Sarrel PM, Sullivan SD, Nelson LM. Hormone replacement therapy in young women with surgical primary ovarian insufficiency. Fertil Steril. 2016;106(7):1580-7.
7. Nelson LM. Clinical practice. Primary ovarian insufficiency. N Engl J Med. 2009;360(6):606-14.
8. Sullivan SD, Sarrel PM, Nelson LM. Hormone replacement therapy in young women with primary ovarian insufficiency and early menopause. Fertil Steril. 2016;106(7):1588-99.
9. Lobo RA. Menopause and care of mature woman: endocrinology, consequences of estrogen deficiency, effects of hormone therapy, and other treatment options. In: Lobo RA, Gershenson DM, Lentz GM, Valea FA, eds. Comprehensive gynecology. 7th ed. Philadelphia: Elsevier; 2017. p.258-93.
10. Huhtaniemi I, Hovatta O, La Marca A, Livera G, Monniaux D, Persani L, et al. Advances in the molecular pathophysiology, genetics, and treatment of primary ovarian insufficiency. Trends Endocrinol Metab. 2018;29(6):400-19.
11. Podfigurna A, Lukaszuk K, Czyzyk A, Kunicki M, Maciejewska-Jeske M, Jakiel G. Testing ovarian reserve in pre-menopausal women: why, whom and how? Maturitas. 2018;109:112-7.
12. Kwee J, Schats R, McDonnell J, Lambalk CB, Schoemaker J. Intercycle variability of ovarian reserve test: results of a prospective randomized study. Hum Reprod. 2004;19(3):590-5.
13. Hendriks DJ, Mol BW, Bancsi LF, Te Velde ER, Broekmans FJ. Antral follicle count in the prediction of poor ovarian response and pregnancy after in vitro fertilization: a meta-analysis and comparison with basal follicle-stimulating hormone level. Fertil Steril. 2005;83(2):291-301.
14. Tal R, Tal O, Seifer BJ, Seifer DB. Antimüllerian hormone as predictor of implantation and clinical pregnancy after assisted conception: a systematic review and meta-analysis. Fertil Steril. 2015;103(1):119-30.
15. Van Loendersloot LL, van Wely M, Limpens J, Bossuyt PM, Repping S, van der Veen F. Predictive factors in in vitro fertilization (IVF): a systematic review and meta-analysis. Hum Reprod Update. 2010;16(6):577-89.
16. Nelson LM, Covington SN, Rebar RW. An update: spontaneous premature ovarian failure is not an early menopause. Fertil Steril. 2005;83(5):1327-32.
17. European Society of Human Reproduction and Embryology. Management of women with premature ovarian insufficiency. Guideline of the European Society of Human Reproduction and Embryology; 2015.
18. Cox L, Liu JH. Primary ovarian insufficiency: an update. Int J Womens Health. 2014;6:235-43.
19. Quintana R. Preservación de la fetilidad en medicina. Ed Ascune Enero 2012. Pag 52-6;129-33.
20. Blumendfeld Z, von Wolff M. GnRH-analogues and oral contraceptives for fertility preservation in women during chemotherapy. Hum Reprod Update. 2008;14(6):543-52.
21. Clowse ME, Behera MA, Anders CK, Copland S, Coffman CJ, Leppert PC, et al. Ovarian preservation by GnRH agonists during chemotherapy: a meta-analysis. J Womens Health (Larchmt). 2009;18(3):311-9.

ORIGINAL ARTICLE

Metastasis from glioblastoma multiforme: a meta-analysis

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SUMMARY

OBJECTIVE: *Extracranial metastases of glioblastoma multiforme (GBM) are rare due to the short survival experienced by the patients. Therefore, the natural history of GBM metastases remains elusive. The identification of clinical factors promoting GBM metastases may help elucidate the mechanisms of tumor cell invasion in the brain. The aims of this study were to perform a meta-analysis evaluating the survival, characteristics, prognostic factors, and predictors of treatment outcome in patients with metastatic GBM and describe a case of metastatic extracranial GBM.*

METHODS: *We report the case of a patient diagnosed with GBM metastatic to the lungs and the results of a meta-analysis of 114 other cases of metastatic GBM identified through a MEDLINE and BIREME search.*

RESULTS: *The mean age of the patients was 38.2±16.1 years and 70.4% were male. The time elapsed between the identification of the metastasis and death was significantly increased in patients undergoing surgery (p=0.019), whereas the time from the diagnosis of the primary tumor to death was significantly increased in patients receiving radiation therapy (p=0.050). The time elapsed from metastasis to death and diagnosis to death was significantly longer in patients receiving chemotherapy (p<0.001 and p=0.027, respectively). The liver was the metastatic site associated with the shortest time elapsed from diagnosis to death (p=0.024).*

CONCLUSIONS: *In GBM, surgical resection is important in reducing the risk of metastasis, and chemotherapy and radiation therapy help to prolong survival in metastatic GBM. Metastases to the liver are associated with shorter survival compared with metastases to other sites.*

KEYWORDS: *Glioblastoma. Neoplasm metastasis. Neurosurgery. Meta-analysis.*

INTRODUCTION

Glioblastoma multiforme (GBM) is the most common primary malignant tumor of the central nervous system in adults, with an estimated incidence of 2–3 cases per 100,000 individuals in Europe and North America. Extracranial metastases of GBM are extremely rare, affecting 0.4–0.5% of all patients with GBM[1]. The rarity of this phenomenon is attributed to the very short survival experienced by the patients, in which not enough time is available for neoplastic cells to metastasize to extracranial organs.

Since the first case of GBM was reported in 1928 by Davis[2], the treatment of these extremely aggressive central nervous system (CNS) tumors has progressively improved. With the emergence of more

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aggressive neurosurgical therapies and ventricular peritoneal shunts, GBM metastases have been reported in association with hematogenous spreading of tumor cells or structural changes following surgery in the meningeal layers and skull. In a recent review[3], around 10% of the reported cases of extracranial GBM spread occurred without surgical intervention.

The natural history of extracranial GBM is unknown. The identification of clinical factors promoting extracranial metastases may help elucidate the mechanisms of invasion of tumor cells in the brain tissue. Based on these considerations, we report herein a case of GBM metastatic to the lungs, along with a meta-analysis evaluating the survival, characteristics, prognostic factors, and predictors of treatment outcome in patients with metastatic GBM.

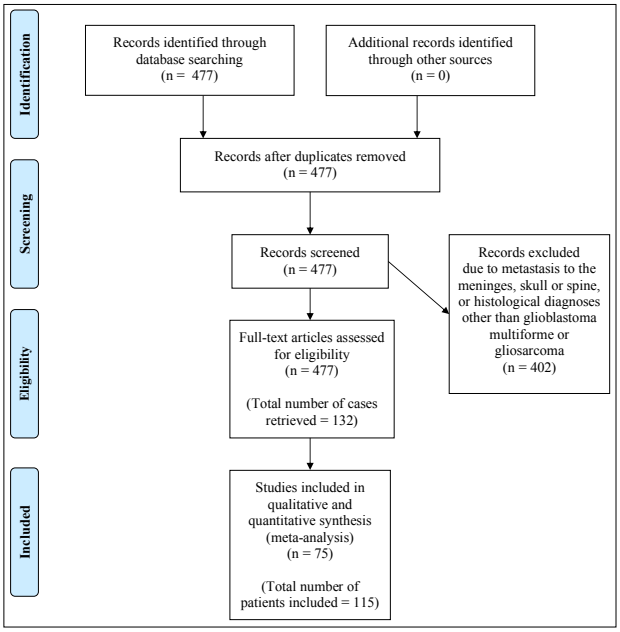
METHODS

The present article was written in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA; www.prisma-statement.org) (see the supplemental Figure for the PRISMA checklist).

Selection of patients

We performed an electronic survey of the MEDLINE and BIREME database in search of cases of GBM with extracranial metastases published between January 1928 and September 2015. We identified 457 articles retrieved from MEDLINE using the keywords "Neoplasm Metastasis" [MeSH] AND "Glioblastoma" [MeSH]. In the BIREME platform, 20 other publications not identified in the MEDLINE search were retrieved with the search terms "Neoplasm Metastasis" AND "Glioblastoma." Publications in languages other than English were included if abstracts were available in this language. Cases with suspected radiological diagnosis of GBM metastases but without corroborative histology were excluded. The selected cases include primary and secondary GBM and gliosarcoma, which subsequently progressed with extracranial metastases confirmed by surgery or autopsy. Cases with metastasis to the meninges, skull, or spine were included. All histological diagnoses other than GBM or gliosarcoma were excluded. After an individual analysis of 477 abstracts, 75 publications comprising cases or described series of metastatic GBM were selected (Figure 1).

FIGURE 1. PRISMA 2009 FLOW DIAGRAM OF SEARCH STRATEGY



Case report

The present case report is described in accordance with the ethical standards of our institution.

In addition to the cases selected from the literature, we added the case of a 43-year-old female patient who had undergone surgery at another institution for resection of a left frontal glial mass lesion with dimensions of 3.8 x 2.7 x 3.2 cm on magnetic resonance imaging (MRI). The lesion was diagnosed after the patient complained of headache that had progressively worsened over 3 months. No images are available from that period. After partial resection of the lesion (motor area), a diagnosis of GBM was established, and the patient underwent conformal radiotherapy (total 60 Gy) and chemotherapy, according to the protocol recommended by Stupp et al.[4]. At 23 months after the initial surgery, the patient was admitted to our hospital due to seizures and right hemiparesis that had developed 10 days earlier. A new brain MRI showed a solid-cystic lesion in the left frontoparietal region measuring 5.8 x 4. 7 x 5.4 cm in the largest diameter (Figure 2).

The patient underwent subtotal resection of the tumor due to the eloquence of the location. A pathological diagnosis was consistent with GBM, and the patient received chemotherapy with irinotecan. At 36 months after the diagnosis of the tumor, the patient was admitted to the emergency department for dyspnea lasting over 3 days. A chest computed tomography (CT) showed expansive lesions in the lung parenchyma and bilateral pleural effusion (Figure 3).

FIGURE 2. CEREBRAL AXIAL (LEFT) AND SAGITTAL (RIGHT) T1 MAGNETIC-RESONANCE IMAGING WITH CONTRAST SHOWING A SOLID-CYSTIC LEFT FRONTOPARIETAL LESION

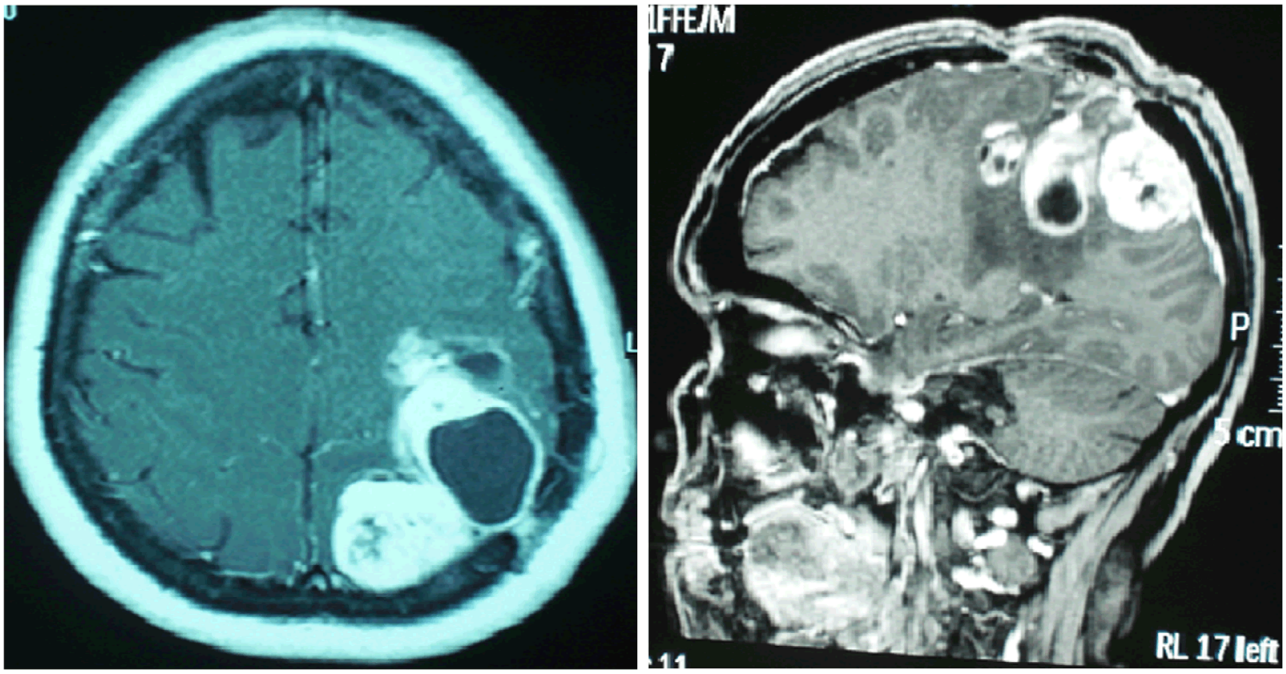
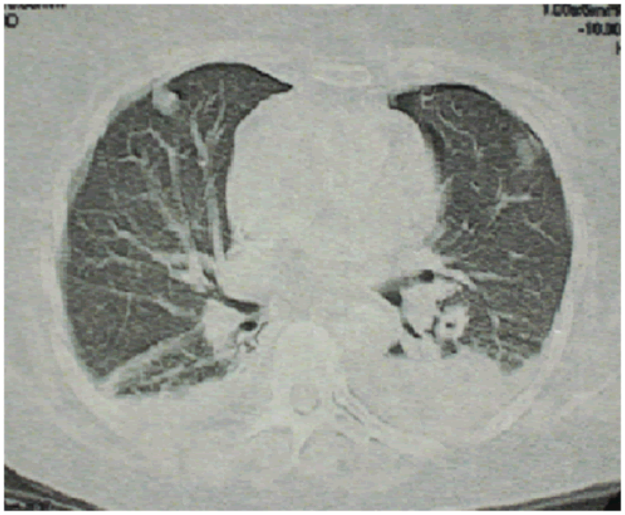


FIGURE 3. CONTRAST COMPUTED TOMOGRAPHY OF THE CHEST SHOWING BILATERAL PLEURAL EFFUSION, MORE PRONOUNCED ON THE LEFT, AND NODULES WITH WELL-DELINEATED MARGINS, SUGGESTIVE OF SECONDARY IMPLANTATION



A pathological diagnosis of pulmonary metastatic GBM was established by fiberoptic bronchoscopy. The patient died 2 months after the lesion was histologically confirmed to be metastatic GBM.

Data Collection

Our survey retrieved 132 cases of GBM with extracranial metastasis, of which 18 were excluded for not meeting the inclusion criteria. With the addition of our case, a total of 115 cases were included in the meta-analysis.

The data collected were organized in a spreadsheet and included the following:

- Survival time, divided into four stages: (A) from symptom onset to diagnosis of GBM; (B) from diagnosis of GBM to detection of extracranial metastases; (C) from the detection of extracranial metastasis to death, and (D) from the diagnosis of GBM to death;
- Year of publication, categorized from the 1950s until 2010 (cases published before 1950 were categorized in the same group);
- Patients' age, divided into decades: below 20 years, 20–29 years, 30–39 years, 40–49 years, 50–59 years, and above 60 years;
- Patients' described gender;
- Neuroimaging method used to diagnose the metastases, categorized as CT, MRI, both, or neither;
- GBM anatomical site, categorized as frontal, parietal, temporal, occipital, parietal + temporal, parietal + occipital, frontal + parietal, temporal + occipital , frontal + temporal, cerebellum, and brainstem;
- Location of the metastasis, categorized as systemic, bone, liver, lung, lymph nodes, and neck;
- Treatment performed, categorized as no therapy or a combination of tumor resection, radiation, chemotherapy, and/or ventriculoperitoneal shunt (VPS).

We excluded from the analysis the patients' neurological performance and extension of the surgical resection since these data were scarce in the reviewed literature.

When the GBM was located in three or more lobes or parts of the brain in a given patient, we considered the tumor to affect the entire hemisphere. We used the same criteria in regards to the metastatic site, grouping under the category "systemic" those metastases affecting organs not included in the five most prevalent groups described above. We considered to be the "metastatic site" the area other than the brain where the initial tumor cell growth was recorded and confirmed by biopsy.

Statistical analysis

The interval of time elapsed between variables of interest were evaluated with summary measures (mean, standard deviation, median, minimum, and maximum values). Comparisons of the time elapsed between the categories of characteristics were evaluated using the Kruskal-Wallis test followed by Dunn's multiple comparison or Mann-Whitney tests, as appropriate.

The tests were performed with a significance level of 5% and the software used for the analysis was SPSS 20.0 [5,6].

RESULTS

A total of 115 cases of GBM with extracranial metastases (110 cited as GBM and 5 as gliosarcoma) published between 1928 and 2015 were selected for the analysis. Table 1 shows the general characteristics of the study population and the time elapsed between events. The individual characteristics of each patient included in the meta-analysis are shown in the Supplemental Table.

We observed that the time elapsed between the emergence of symptoms and the diagnosis of the pri-

TABLE 1. GENERAL CHARACTERISTICS OF THE STUDY POPULATION

Variable	Values
Age (years)	
Mean (SD)	38.2 (16.1)
Median (min.– max.)	39.5 (6–68)
Sex, n (%)	
Female	34 (29.6)
Male	81 (70.4)
GBM site, n (%)	
Frontal	21 (18.6)
Temporal	25 (22.1)
Parietal	6 (5.3)
Occipital	9 (8)
Hemispheric	12 (8.8)
Parietal + Temporal	6 (5.3)
Parietal + Occipital	11 (9.7)
Frontal +Parietal	15 (13.3)
Temporal + Occipital	5 (4.4)
Frontal + Temporal	3 (2.7)
Cerebellum	1 (0.9)
Brain stem	1 (0.9)
CT/MRI, n (%)	
None	54 (47)
CT	16 (13.9)
MRI	11 (9.6)
Both	34 (29.6)
Surgery, n (%)	
No	12 (10.5)
Yes	102 (89.5)
Radiation, n (%)	
No	18 (16.8)
Yes	89 (83.2)
Chemotherapy, n (%)	
No	75 (66.4)
Yes	38 (33.6)
Shunt, n (%)	
No	102 (88.7)
Yes	13 (11.3)
Extracranial GBM site, n (%)	
Bone	28 (24.3)
Liver	10 (8.7)
Lung	25 (21.7)
Lymph nodes	14 (12.2)
Neck	10 (8.7)
Systemic	28 (24.3)
Time from symptoms to diagnosis (months)	
Mean (SD)	7.5 (14.2)
Median (min. – max.)	2.5 (0–75)
Time from diagnosis to metastasis (months)	
Mean (SD)	11.7 (11.9)
Median (min.– max.)	7.5 (0–62)
Time from metastasis to death (months)	
Mean (SD)	4.6 (9.3)
Median (min.– max.)	2.3 (0–78)
Time from diagnosis to death (months)	
Mean (SD)	15 (14.4)
Median (min.– max.)	10 (1–90)

Abbreviations: min. – minimum; max. – maximal; n – number; CT – computed tomography; MR – magnetic resonance.

mary tumor and between the detection of the metastasis and death differed significantly across decades, as shown in Table 2. The time elapsed between the diagnosis of the primary GBM tumor and the patients' death increased by 1.68 months per decade (p = 0.034), and the time elapsed between the detection of the metastasis until death increased by 0.87 months per decade (p = 0.024).

Given that CT and MRI can provide an earlier detection of the tumor, we analyzed whether the use of these imaging modalities affected the survival of patients with extracranial GBM. As shown in Table 3, the time elapsed between the detection of the metastases and death increased significantly with the use of CT, MRI, or both when compared with the absence of use of these imaging methods.

We also analyzed the effect of the delivered treatment on the patients' survival. As shown in Table 3, the time elapsed between the detection of the metastases and the patients' death was significantly longer in patients who had undergone surgical resection of the primary tumor. Similarly, the time between the diagnosis of the primary tumor and death was significantly longer in patients who had received radiation therapy, while the time between the detection of the metastasis and death and between diagnosis of the primary tumor and death were significantly longer in patients who received chemotherapy. The use of VPS had no influence on survival in the overall cohort.

As also shown in Table 3, the time elapsed between the diagnosis of the primary tumor and death was affected by the location of the metastases. On multiple comparison analysis, the time elapsed between the diagnosis of the primary tumor and death was longer in patients with neck metastases and lung metastases, and shortest in patients with liver metastases.

The GBM location in the brain or the patients' age and gender were not significantly associated with the development of metastases.

DISCUSSION

The small number of reported cases of patients with extracranial metastases of GBM prevents the development of prospective studies focused on this issue. Thus, the understanding of the impact of GBM metastases relies on meta-analyses of existing cases in the literature, such as the one reported in this

TABLE 2. TIME ELAPSED BETWEEN IMPORTANT EVENTS ACROSS DECADES IN PATIENTS WITH GLIOBLASTOMA MULTIFORME

Year	Time S-Dx (months)	Time Dx-M (months)	Time M-D (months)	Time Dx-D (months)
Up to 1950 Mean (SD) Median (min. – max.)	4.5 (3) 3 (2.5 – 8)	6 (6) (4 – 8)	2 (2) 2 (0 – 4)	8 (3.5) 6 (6 – 12)
1950 to 1959 Mean (SD) Median (min. – max.)	24.4 (26.6) 10.5 (2 – 75)	8.1 (5.1) 7.8 (0 – 17)	1.3 (1. 9) 0.3 (0 – 5)	8.5 (5) 9 (1 – 18)
1960 to 1969 Mean (SD) Median (min. – max.)	5.4 (3.6) 5 (1 – 11)	12.7 (10.3) 8 (3 – 32)	3.1 (3.2) 1.5 (1 – 9)	15.6 (13.3) 10.5 (1 – 60)
1970 to 1979 Mean(SD) Median (min. – max.)	10.8 (21) 1 (0.3 – 60)	6.1 (4.4) 6.5 (0 – 13)	0.9 (1.3) 0 (0 – 4)	6.6 (3.9) 7.5 (1 – 13)
1980 to 1989 Mean (SD) Median (min. – max.)	4.1 (4) 2 (0.5 – 12)	9.6 (9.3) 7.5 (1 – 31)	1.8 (2.1) 1 (0 – 7)	10.5 (9.7) 7.8 (1 – 32)
1990 to 1999 Mean (SD) Median (min. – max.)	4.8 (4.5) 3 (1 – 13.5)	12.8 (12) 10 (0 – 35)	6.2 (5.6) 3 (1 – 15)	18 (14.4) 15 (1 – 46)
2000 to 2009 Mean (SD) Median (min. – max.)	1.3 (1.2) 1 (0 – 4)	12 (9.6) 7.8 (3 – 34)	4.3 (3.6) 3 (1 – 13)	14.4 (9.3) 10 (3 – 36)
After 2010 Mean (SD) Median (min. – max.)	3.5 (3.7) 3 (0 – 12)	16.1 (17.7) 9 (1 – 62)	8.9 (16.7) 3 (1 – 78)	22.3 (22.3) 18 (1 – 90)
P	0.011	0.719	0.002	0.122

Kruskal-Wallis test. Abbreviations: S-Dx – symptoms to diagnosis; Dx-M – diagnosis to metastasis; M-D – metastasis to death; Dx-D – diagnosis to death; SD – standard deviation; min.: minimum; max.: maximum.

study. The current need for clinical elements to contribute to improving survival and the understanding of prognostic factors in patients with GBM was the rationale for this analysis.

Tumor dissemination generally occurs via lymphatic, vascular, or direct spreading. The occurrence of lymph node metastases from GBM suggests the participation of meningeal lymphatics in the process. This hypothesis is likely in patients undergoing surgery, but difficult to explain in patients with lymph node metastases who had not undergone surgery[7]. The brain lacks a true lymphatic system, thus limiting the lymphatic spread of tumor cells. The unique and characteristic vascular intracerebral glioblastoma network is notorious; the neoplastic vessels have thick walls with several layers of endothelial cells

TABLE 3. IMPACT OF (A) THE APPLICATION OF COMPUTED TOMOGRAPHY AND MAGNETIC RESONANCE IMAGING COMPARED WITH THE ABSENCE OF USE OF BOTH METHODS, (B) THE DELIVERED TREATMENT, AND (C) THE LOCATION OF THE EXTRACRANIAL METASTASIS ON TIME ELAPSED BETWEEN IMPORTANT EVENTS IN PATIENTS WITH GLIOBLASTOMA MULTIFORME

(A) CT/MR	Time S-Dx (months)	Time Dx-M (months)	Time M-D (months)	Time Dx-D (months)
None Mean (SD) Median (min. – max.)	13.2 (19.7) 5 (0.3 – 75)	9.3 (7.3) 7 (0 – 32)	1.7 (2.2) 1 (0 – 9)	12.8 (11.3) 9 (1 – 60)
CT Mean (SD) Median (min. – max.)	3.3 (3.6) 2 (0 – 12)	9.2 (10.5) 6 (0 – 35)	3.5 (4.4) 1.8 (0 – 15)	12.1 (12.5) 8 (1 – 46)
MR Mean (SD) Median (min. – max.)	1.2 (0.8) 1 (0.5 – 2)	14.1 (8.8) 15 (2 – 30)	4.5 (3.8) 3 (1 – 13)	18 (10.6) 18.8 (2 – 36)
Both Mean (SD) Median (min. – max.)	3.2 (3.2) 1.5 (0 – 12)	14.1 (15.9) 7.8 (0 – 62)	7.7 (14.4) 3 (1 – 78)	19.1 (19.7) 11.5 (1 – 90)
P	0.040	0.362	0.002	0.215

(B) Surgery	Time S-Dx (months)	Time Dx-M (months)	Time M-D (months)	Time Dx-D (months)
No Mean (SD) Median (min. – max.)	8.8 (15.8) 2 (0.3 – 50)	6.6 (6.3) 6 (0 – 17)	1.3 (1.7) 0.9 (0 – 5)	8.4 (4.8) 9 (2 – 18)
Yes Mean (SD) Median (min. – max.)	7.2 (14) 2.8 (0 – 75)	12.4 (12.3) 8 (0 – 62)	5.1 (9.9) 2.5 (0 – 78)	15.9 (15) 10 (1 – 90)
p	0.919	0.125	0.019	0.122
Chemoterapy				
No Mean (SD) Median (min. – max.)	9.8 (16.7) 3 (0 – 75)	10.3 (9.3) 7 (0 – 36)	2.2 (2.6) 1 (0 – 10)	13.1 (12) 9 (1 – 60)
Yes Mean (SD) Median (min. – max.)	2.8 (3) 2 (0 – 12)	14.2 (14.6) 8.5 (0 – 62)	8.1 (13.7) 3.3 (0 – 78)	19.5 (18) 13 (3 – 90)
p	0.093	0.203	<0.001	0.027
Radiation				
No Mean (SD) Median (min. – max.)	7.7 (9.7) 6 (1 – 36)	8.6 (11.2) 5 (1 – 36)	2.2 (2.7) 1.3 (0 – 10)	10.5 (12) 7 (1 – 46)
Yes Mean (SD) Median (min. – max.)	7.6 (15.2) 2 (0 – 75)	12.4 (12) 8 (0 – 62)	5.1 (10.2) 2.5 (0 – 78)	15.5 (14.5) 10 (1 – 90)
p	0.083	0.065	0.121	0.050
Shunt				
No Mean (SD) Median (min. – max.)	7.8(14.7) 2.5 (0 – 75)	11.8 (12) 7.5 (0 – 62)	4.7 (9.7) 2 (0 – 78)	15.5 (14.8) 10 (1 – 90)
Yes Mean (SD) Median (min. – max.)	3.5 (3.2) 4 (0 – 7)	10.4 (11.5) 8 (0 – 34)	3.1 (2.2) 3 (0 – 7)	9.6 (7.5) 8 (3 – 25)
p	0.683	0.605	0.694	0.188

(C) Local extracranial metastasis	Time S-Dx (months)	Time Dx-M (months)	Time M-D (months)	Time Dx-D (months)
Bone Mean (SD) Median (min. – max.)	3 (3) 2 (0.3 – 12)	10.8 (8.3) 8 (0 – 31)	8.5 (17) 3.3 (0 – 78)	18.3 (18.1) 12.5 (1 – 90)
Liver Mean (SD) Median (min. – max.)	11.1 (16.9) 4 (0.3 – 36)	6 (4.2) 4 (3 – 13)	1.5 (1.7) 1 (0 – 4)	7.1 (2.9) 6.5 (3 – 13)
Lung Mean (SD) Median (min. – max.)	3.5 (3.1) 2.8 (0 – 10)	12.1 (10.8) 8 (1 – 36)	2.2 (2.7) 2 (0 – 10)	16.9 (16) 10.5 (1 – 60)
Lymph node Mean (SD) Median (min. – max.)	19.4 (30.4) 2.5 (0.3 – 75)	9.9 (10) 9 (0 – 32)	3.5 (4.2) 1.5 (0 – 10)	13.9 (10) 11.8 (2 – 33)
Neck Mean (SD) Median (min. – max.)	5 (5.5) 3 (0.5 – 13.5)	17 (18.4) 8 (2 – 60)	5.5 (5.3) 2.8 (2 – 18)	22.4 (18.5) 17.3 (2 – 63)
Systemic Mean (SD) Median (min. – max.)	10.1 (15.9) 6 (0 – 60)	11.9 (14) 7 (0 – 62)	3.3 (4) 2 (0 – 15)	10.9 (9.4) 8 (1 – 35)
p	0.887	0.695	0.079	0.024

Kruskal-Wallis test. Abbreviations: S-Dx – symptoms to diagnosis; Dx-M – diagnosis to metastasis; M-D – metastasis to death; Dx-D – diagnosis to death; SD – standard deviation; min. – minimum; max. – maximum.

forming irregular interconnected glomerular structures arranged chaotically. Circulatory disorders, local hypoxia at low pH, and increased permeability of the tumor blood vessel wall occur as a result. Vascular proliferation is stimulated by the tumor itself, which produces growth factors and cytokines. Vascular endothelial growth factor (VEGF) is the most common among them, and its concentration is up to 50 times greater in the CNS of patients with GBM compared with healthy ones[8]. Unlike other malignant systemic diseases, glial cells do not spread through blood vessels, making hematogenous spread highly unusual.

Despite the notable progress in the quality of surgery, chemotherapy, and radiotherapy that has occurred over the decades evaluated in this study, survival has failed to increase in the same proportion. Overall, 70% of the patients with GBM die within 2 years from the diagnosis[4]. Molecular genetics has an intrinsic relationship with survival in individuals with GBM. Patients with epigenetic silencing of the O[6]-methylguanine-DNA methyltransferase (*MGMT*) gene have an estimated average survival of 48.9% at 2 years and 13.8% at 5 years when compared with those with unmethylated *MGMT*, whose corresponding average survival rates were reported to be around 14.8% and 8.3%, respectively[8]. The present study did not evaluate the methylation status of the *MGMT* gene due to the scarcity of this information in the retrieved cases, even in recently published ones. Our work also showed an increased survival of the patients over the decades, probably associated with improvement and agility in diagnostic imaging, as well as the benefit of combination therapies including surgery, radiotherapy, and chemotherapy. Currently, the molecular determinants that may predispose to the occurrence of GBM metastasis are unknown, and no molecular characteristics have been correlated with increased survival.

The paradoxical observation of a low (0.4%) incidence of metastases[1] in GBM combined with its highly invasive nature led Sullivan et al.[9] to search for circulating tumor cells (CTCs) in peripheral blood. The authors analyzed with immunofluorescence markers (especially EGFR) peripheral blood of 33 patients diagnosed with GBM. CTCs were present in 13 cases (39%), which is a much higher rate than the recorded rate of GBM metastases. Interestingly, the authors found that these GBM CTCs predominantly expressed genes associated with mesenchymal differentiation. This finding may be related to their greater phenotypic aggression, which may justify their ability to invade the cerebral blood flow and spread. Following the same line of reasoning, Müller et al.[10] used the glial fibrillary acidic protein (GFAP; mainly expressed in astrocytes and glial tumors) as a marker to isolate GBM cells in peripheral blood. Of the 141 patients included in the study, CTCs were identified in 29 cases (20.6%). Importantly, the analysis in both studies was performed preoperatively, indicating that the presence of CTCs was not the result of disruption of the blood brain barrier due to surgery. Another issue to be discussed is the fact that the studies used different methodologies to isolate CTCs based on different markers, raising questions regarding the

best method to detect CTCs. The molecular diversity of GBM must also be considered, and some tumor cells may not express GFAP markers and vice versa. In addition, CTCs expressing different markers may coexist and fluctuate over time. Thus, new technologies and more accurate identification of CTCs of GBM need to be developed to clarify these questions.

The patients in our study group had a mean age of 38.2 years, which is lower than that of typical adults with GBM, and a median survival of 15 months. This finding was also shown in a review by Lun et al.[11], which reported a mean age of 38 years and median survival of 10.5 months in 88 cases of GBM. In contrast, a review of 94 cases conducted by Anghileri et al.[12] found a mean age of 41.1 years and median survival of 12 months. Both studies found a mean time from the initial diagnosis of GBM to the emergence of metastases of 8.5 months, which is lower than that found in our analysis (11.7 months). It is possible that the occurrence of metastases from low-grade gliomas occur before these tumors undergo malignant transformation, enabling the ability of tumor cells to settle in metastatic sites. However, this hypothesis lacks proof [12].

In our analysis, the metastatic site influenced survival, considering the time from the diagnosis of the primary tumor to death. This fact was also evidenced in a previously published series, which showed a better prognosis of GBM metastatic to the neck and a worse prognosis for GBM metastatic to the lung[10]. Our study demonstrated that patients with metastases to the neck also showed a longer time from diagnosis of the primary tumor to death (22.4 ± 18.5 months). Patients with metastasis to the liver – and not to the lungs – had the lowest survival time from diagnosis to death (7.1 ± 2.9 months). This difference was statistically significant and may be explained by cellular and molecular heterogeneity, which was clearly demonstrated in a study by Fidler and Kripke[13] using B16 melanoma cells. Certain tumor clones had a predilection for specific organs such as the lung or liver. Thus, it is possible that GBM clones migrating to the neck are less aggressive than those migrating to the liver.

Despite GBM being the most common CNS tumor, the number of patients with metastatic GBM is very low. The explanation for this discrepancy is unclear but may involve the short survival experienced by the patients and a possible absence of a suitable environment for multiplication of metastatic cells. Rubenstein et al.[14] have reported that GBM cells may have preferential adhesion to the neural stroma. With disease progression, tumor cells can violate the existing cerebral vasculature, achieving a greater capacity to spread to the CNS via a hematogenous route. With prolonged survival of young patients with GBM, the possibility of GBM cells spreading to the bloodstream may increase, which will also increase the odds of distant metastasis[15]. There have been reports of the occurrence of extracranial GBM in recipients of liver and kidney grafts from donors with GBM. Additionally, immunosuppression for prevention of organ rejection may have a potentiating action on the development of tumors in recipients[16].

The development of extracranial GBM indicates that systemic spread occurs early in the disease[10]. The immune system appears to be a link in the understanding of this kind of behavior towards CTCs of GBM. As suggested by Seoane & de Mattos-Arruda[17], existing CTCs would require time to learn how to escape immune surveillance, which is not feasible considering the short survival experienced by these patients. GBM cells in the circulation may provide an opportunity for gene detection and analysis of the incipient intracranial GBM disease. These "liquid biopsy" are potentially powerful tools for characterization of patients with GBM. CTCs can represent materials derived from tumor sources and be regarded as tissue barcodes of brain tumor, with the advantage of being a minimally invasive procedure. Thus, these biomarkers can be molecularly categorized to reveal a repertoire of momentaneous somatic genomic aberrations and yield a longitudinal overview of the molecular characteristics of the tumor.

The "liquid biopsy" still needs further studies for validation. Some of its advantages include the discrimination of tumoral pseudoprogression, selecting specific therapies and monitoring mechanisms of resistance to cytotoxicity and therapeutic targets. In our view, the genomic changes "written" in the blood should be one of the most valuable tools to guide treatment and assist in the understanding of GBM and its molecular variants, given the scarcity of reported cases of extracranial GBM with the paradoxically higher incidence of GBM and amount of CTCs investigated in previous studies.

A potential limitation of our study was the small number of patients included in the analysis, retrieved from a period spanning almost a century. As mentioned earlier, this limitation is due to the

scarcity of cases of metastatic GBM described in the literature.

A meta-analysis of cases with extracranial metastatic GBM showed that over the past seven decades, patients affected with this disease experienced a decrease in the time elapsed between the onset of symptoms and the diagnosis of the primary tumor in addition to an increase in the time elapsed from the detection of metastasis to death. The increasing use of neuroimaging methods (CT and MRI) over time decreased the time elapsed between the development of symptoms and the diagnosis of GBM and prolonged the survival from the diagnosis of the metastasis until death. Surgery for treatment of the primary GBM tumor was associated with an increased time elapsed from the detection of the metastasis until death, while radiation increased the time elapsed between the diagnosis of the primary tumor and death. The time elapsed between the metastasis and death and between the diagnosis and death was significantly longer in patients who received chemotherapy. Hepatic metastasis of GBM was associated with the worst prognosis when compared with other metastatic sites.

The limited number of patients with GBMs presenting extracerebral metastases prevents the development of prospective studies. Improvements in molecular testing and immune response evaluation may help elucidate the occurrence of low metastatic rates in GBM. Perhaps this reversal path (investigation of the metastatic site) may shed new perspectives for the understanding of the adherence of CTCs into metastatic sites in patients with GBM.

CONCLUSIONS

The results of our study, spanning almost a decade and including patients with metastatic GBM, confirm the benefits of surgical resection as an important step in reducing the risk of metastasis, as well as those of chemotherapy and radiation therapy in prolonging survival in patients with metastatic GBM. Compared with other metastatic sites, the liver was the site associated with the shortest survival.

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RESUMO

OBJETIVO: *Metástases extracranianas do glioblastoma multiforme (GBM) são raras devido à baixa sobrevida dos pacientes. Portanto, a história natural das metástases do GBM permanece incerta. A identificação de fatores clínicos que promovem metástases no GBM pode ajudar a elucidar os mecanismos de invasão das células tumorais no cérebro. O objetivo deste estudo foi realizar uma meta-análise avaliando a sobrevida, características, fatores prognósticos e preditores de desfechos do tratamento em pacientes com GBM metastático e descrever um caso de GBM extracraniano metastático.*

MÉTODOS: *Relatamos o caso de uma paciente diagnosticada com GBM metastático para os pulmões e os resultados de uma meta-análise de 114 outros casos de GBM metastático identificados por meio de uma pesquisa no Medline e Bireme.*

RESULTADOS: *A média de idade dos pacientes foi de 38,2±16,1 anos e 70,4% eram do sexo masculino. O tempo decorrido entre a identificação da metástase e o óbito foi significativamente maior em pacientes submetidos à cirurgia (p = 0,019), enquanto que o tempo do diagnóstico do tumor primário até o óbito aumentou significativamente em pacientes submetidos à radioterapia (p = 0,050). O tempo decorrido da metástase até o óbito e do diagnóstico até o óbito foi significativamente maior nos pacientes que receberam quimioterapia (p < 0,001 e p = 0,027, respectivamente). O fígado foi o local metastático associado ao menor tempo decorrido do diagnóstico até a morte (p = 0,024).*

CONCLUSÕES: *No GBM, a ressecção cirúrgica é importante para redução do risco de metástase, e a quimioterapia e a radioterapia ajudam a prolongar a sobrevida no GBM metastático. Metástases para o fígado estão associadas a uma sobrevida mais curta quando comparadas a metástases para outros locais.*

PALAVRAS-CHAVE: *Glioblastoma. Metástase neoplásica. Neurocirurgia. Meta-análise.*

REFERENCES

1. Pasquier B, Pasquier D, N'Golet A, Panh MH, Couderc P. Extraneural metastases of astrocytomas and glioblastomas: clinicopathological study of two cases and review of literature. Cancer. 1980;45(1):112-25.
2. Davis L. Spongioblastoma multiforme of the brain. Ann Surg. 1928;87(1):8-14.
3. Anzil AP. Glioblastoma multiforme with extracranial metastases in the absence of previous craniotomy. Case report. J Neurosurg. 1970;33(1):88-94.
4. Stupp R, Hegi ME, Mason WP, van den Bent MJ, Taphoorn MJ, Janzer RC, et al; European Organisation for Research and Treatment of Cancer Brain Tumour and Radiation Oncology Groups; National Cancer Institute of Canada Clinical Trials Group. Effects of radiotherapy with concomitant and adjuvant temozolomide versus radiotherapy alone on survival in glioblastoma in a randomised phase III study: 5-year analysis of the EORTC-NCIC trial. Lancet Oncol. 2009;10(5):459-66.
5. Kirkwood BR, Sterne JAC. Essential medical statistics. Malden: Blackwell Science; 2006.
6. Kleinbaum DG, Klein M. Survival analysis: a self-learning text. New York: Springer; 1996.
7. Witoonpanich P, Bamrungrak K, Jinawath A, Wongwaisayawan S, Phudhichareonrat S, Witoonpanich R. Glioblastoma multiforme at the corpus callosum with spinal leptomeningeal metastasis. Clin Neurol Neurosurg. 2011;113(5):407-10.
8. Jain RK, di Tomaso E, Duda DG, Loeffler JS, Sorensen AG, Batchelor TT. Angiogenesis in brain tumours. Nat Rev Neurosci. 2007;8(8):610-22.
9. Sullivan JP, Nahed BV, Madden MW, Oliveira SM, Springer S, Bhere D, et al. Brain tumor cells in circulation are enriched for mesenchymal gene expression. Cancer Discov. 2014;4(11):1299-309.
10. Müller C, Holtschmidt J, Auer M, Heitzer E, Lamszus K, Schulte A, et al. Hematogenous dissemination of glioblastoma multiforme. Sci Transl Med. 2014;6(247):247ra101.
11. Lun M, Lok E, Gautam S, Wu E, Wong ET. The natural history of extracranial metastasis from glioblastoma multiforme. J Neurooncol. 2011;105(2):261-73.
12. Anghileri E, Castiglione M, Nunziata R, Boffano C, Nazzi V, Acerbi F, et al. Extraneural metastases in glioblastoma patients: two cases with YKL-40-positive glioblastomas and a meta-analysis of the literature. Neurosurg Rev. 2016;39(1):37-45.
13. Fidler IJ, Kripke ML. Metastasis results from preexisting variant cells within a malignant tumor. Science. 1977;197(4306):893-5.
14. Rubenstein JL, Kim J, Ozawa T, Zhang M, Westphal M, Deen DF, et al. Anti-VEGF antibody treatment of glioblastoma prolongs survival but results in increased vascular cooption. Neoplasia. 2000;2(4):306-14.
15. Awan M, Liu S, Sahgal A, Das S, Chao ST, Chang EL, et al. Extra-CNS metastasis from glioblastoma: a rare clinical entity. Expert Rev Anticancer Ther. 2015;15(5):545-52.
16. Jonas S, Bechstein WO, Lemmens HP, Neuhaus R, Thalmann U, Neuhaus P. Liver graft-transmitted glioblastoma multiforme. A case report and experience with 13 multiorgan donors suffering from primary cerebral neoplasia. Transpl Int. 1996;9(4):426-9.
17. Seoane J, De Mattos-Arruda L. Escaping out of the brain. Cancer Discov. 2014;4(11):1259-61.

ORIGINAL ARTICLE

The positive impact of physical activity on the reduction of anxiety scores: a pilot study

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SUMMARY

OBJECTIVES: *To compare anxiety scores between physical activity practitioners and sedentary, men and women and to relate them to physical activity frequency and age.*

METHODS: *In this cross-sectional study, a sample of 256 regular aerobic physical activity practitioners was compared to a sample of 256 sedentary individuals (control group). Anxiety scores were quantified by Anxiety Inventory Spielberger State-Trait (STAI). The scores of the groups were compared using the Student t-test and chi-square test for parametric and non-parametric data, respectively. The correlation between scores of different variables was performed using the Pearson test.*

RESULTS: *There was a significant difference between the average anxiety scores ($p < 0.001$) and the chi-square test proved there is a higher prevalence of severe anxiety ($p < 0.001$) in the sedentary group. Age did not correlate with worse anxiety scores ($p < 0.05$). Comparing by gender, women had a higher prevalence of intense anxiety.*

CONCLUSIONS: *Individuals who engage in regular physical activity have lower levels of anxiety, and both sexes are benefited with the anxiolytic potential of physical activity. Therefore, this study proved that the Roman poet Juvenal was right, and his expression "Mens sana in corpore sano," could also be interpreted in the opposite direction, i.e., a healthy body correlates with a healthy mind.*

KEYWORDS: *Anxiety. Exercise. Sedentary lifestyle. Psychiatry.*

INTRODUCTION

According to Plato (4th century BCE), "In order for man to succeed in life, God gave him two means, education and physical activity, one for the soul, the other for the body. It is only with just these two means together that man can reach perfection"[1]. In the 1st century A. D., the Roman poet Juvenal coined the famous quote "*Mens sana in corpore sano*'. To him, physical activity was considered the key to keeping our body and our mind in a state of healthy balance.[2]

The World Health Organization[3] estimated that physical inactivity is the fourth risk factor for overall mortality, ahead of overweight and obesity. Recent studies suggest that the practice of a minimum of daily physical activity is able to reduce mortality from all causes in up to 19%.[4]

The association between sedentary behavior and mental disorders, such as depression and low self-esteem, has been reported in some population groups, such as women and adolescents[5]. According to Almeida[6], anxiety is a warning sign that allows indi-

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viduals to be aware and take the necessary measures to deal with this threat. In its pathological form, it is an inadequate response to certain stimuli, due to its intensity or duration. The diagnosis of pathological anxiety can be achieved by means of well-established criteria, such as those of the American Psychiatric Association, through the 5th edition of the Diagnostic and Statistical Manual of Mental Disorders (DSM-V), or the World Health Organization, the 10th edition of the International Statistical Classification of Diseases and Related Health Problems (ICD-10). Moreover, the quantification of anxiety can be done using validated questionnaires, such as the Spielberger and Hamilton, which are the most used, including their versions in Portuguese.

The scientific evidence of the benefits of physical activity on mental health has been increasing significantly. Meta-analyses and systematic reviews have shown the benefits of different forms of physical activity in adults with anxiety disorders, with results as effective as pharmacotherapy and/or cognitive-behavioral therapy.[7]

Therefore, it is increasingly clear the association between the regular practice of physical activity and the improvement of psychiatric symptoms related to humor. The objective of this study was to quantify anxiety scores in a sample of individuals who practice physical activity regularly, comparing it to a sample of sedentary individuals. In addition, we verified whether there are inter-relationships between the variables (age, sex, and frequency of physical activity) in the results found. Therefore, we sought evidence that can prove that there is an association between "Healthy Body, Healthy Mind."

METHODS

The research was approved by the Research Ethics Committee of the University Center of East Minas Gerais - Unileste under decision No. 1,165.392, on 07/27/2015.

This is an observational study with a cross-sectional design conducted in the city of Ipatinga, MG. Since there are no data in the literature about anxiety scores for this municipality, to estimate the sample size in the present study, we employed the technique of sampling for convenience described by Portney and Watkins[8]. It uses a test sample, in which individuals are selected according to availability, by means of continuous recruitment of subjects until the minimum number that allows for the statistical analysis proposed is reached. According to Motta, in quantitative studies, samples that have a minimum of 30 subjects can represent with good accuracy the population average.[9]

The sample was composed of 512 individuals, 256 who practiced physical activity regularly (active group) and 256 sedentary (control group).

The criteria for inclusion in the active group were adults (18-60 years), of both sexes, healthy, who practiced physical activity regularly, of the aerobic type, following the criteria of the American Heart Association (AHA) for defining physically active: minimum of 25 minutes of intense activity for three or more times per week or a minimum of 30 minutes of moderate activity for five or more times per week.[10]

In the control group, we included adults (18-60 years) of both sexes, healthy, who did not practice physical activity on a regular basis.

Among the exclusion criteria for both groups were individuals with diagnosis and/or treatment for physical and/or mental conditions.

The recruitment occurred in recreational clubs and gyms of Ipatinga, as well as during sporting events in the municipality.

The selection of participants was made via a direct approach by the researchers. After clarification about the research, each individual who agreed to participate in the project was approached by the researchers, who then evaluated the inclusion and exclusion criteria and forwarded the volunteers to a place to carry out the assessment individually. At this place, they were given all the necessary information and were asked to sign the Informed Consent Form (ICF).

The anxiety symptoms were assessed and quantified using the State-Trait Anxiety Inventory, proposed by Spielberger in 1970, translated and standardized for Portuguese.[11] It is a self-assessment questionnaire in the form of two scales that allow the level of anxiety symptoms to be graded: state and trait. The term state-anxiety means anxiety concerning the present moment (the application of the questionnaire), while trait-anxiety refers to a personality tendency that does not vary with time. The trait scale was used in this study.

This scale consists of 20 statements that individuals use to describe themselves. Each affirmation receives a score of one to four and the values of all items are then added up. Those in which the high

score indicates low anxiety, the value of the score is reversed. For both scales, the following results apply: mild anxiety (less than or equal to 33 points); moderate anxiety (from 34 to 48 points); severe anxiety (greater than or equal to 49 points).

Since this is a self-assessment questionnaire, each individual first received instructions about its correct filling out and then received the questionnaire to read and answer. The interviewer was available for clarification of possible doubts.

For the statistical analysis, initially, the demographic data for each sample (age and sex) was described. Then the profiles of the sample of assets regarding the type of physical activity, as well as its weekly frequency, were described.

For anxiety scores, an analysis of the difference between the averages of the groups was performed using the Student's t-test for independent samples.

Considering the sample size of the present study, for 510 of freedom[(n1 – 1) + (n2 – 1)], the statistical difference should be considered significant when t >1.96 for significance (α) of 0.05 in samples of two-tailed distribution.[9]

To estimate the difference of qualitative data, we used the Pearson chi-square test (X^2), a nonparametric test of significance that deals essentially with the discrepancies between the observed and expected frequencies.[9]

The correlation between age and anxiety scores was assessed using the Pearson Correlation Coefficient, considered to be relevant when greater than 0.3.[8] This coefficient was also used to estimate the correlation between the frequency of physical activity and anxiety scores.

To estimate the associations between sedentary lifestyle/anxiety and sex/anxiety, we used the Odds-Ratio (OR).

The results were obtained by calculations performed in the Minitab® software, version 15.1.1.0. The minimum level of significance required to reject the null hypothesis (H0) was set at $p < 0.05$.

RESULTS

The sample was composed of 144 men (56.2%) and 112 women (43.8%) who practiced physical activity regularly. The sedentary group had 109 men (42.6%) and 147 women (57.4%). The sedentary group had an average age of 26.2, which was considered to be significantly lower in relation to the active group, with an average age of 29 (t = 3.18; p = 0.002). For the active group, 44.1% of the sample mentioned the practice of only one type of physical activity, while 47.3% reported practicing two and 8.6%, three or more types. It is worth mentioning that the practice of bodybuilding, as a complement to physical activity, was reported by 23.4% of the sample of active individuals. Running was the type of activity most reported by practitioners of physical activity, with 42.7%.

As for the weekly frequency of physical activity, most of the active individuals reported practicing physical activity three times per week (45.7%), followed by 30.5% who practiced physical activity five times per week and 23.8% who practiced four times weekly.

Both in the active and sedentary groups the mean scores of anxiety fall in moderate. Although both the average values (35.9 for active individuals and 42.1 for sedentary) are classified as moderate anxiety by the Spielberger questionnaire, the Student t-test showed a significant difference between the average scores of the groups, i.e., in a confidence interval of 95% of chance for the difference between the averages, the t value was considered significant (Table 1).

Although there was a difference between the averages of the anxiety scores, they do not represent a clinically relevant difference, since both groups had a mean score compatible with moderate anxiety. To test the relevance, we used the chi-square test (X^2). For 2 degrees of freedom (number of groups - 1) and a significance level of 5%, the critical value of X^2 must be greater than or equal to 5.99 to be considered relevant (9). The results showed a significant difference between the groups (X^2 = 105.0; $p < 0.001$). Chart 1 summarizes the distribution of mild, moderate and severe anxiety prevalence in the groups.

The OR results to estimate the association between sedentary lifestyle and anxiety showed a positive correlation of 3.29 between a sedentary lifestyle and intense anxiety scores, i.e., a sedentary individual has approximately three times more chances of having intense anxiety scores when compared to a physically active individual.

TABLE 1. MEAN SCORES OF ANXIETY (IDATE)

	Group			
	Active	Sedentary	CI 95% for difference	T-test (p-value)
Average Score (SD)	35.9 (9.01)	42.1 (10.10)	(4.46 – 7.78)	7.24 (< 0.001)

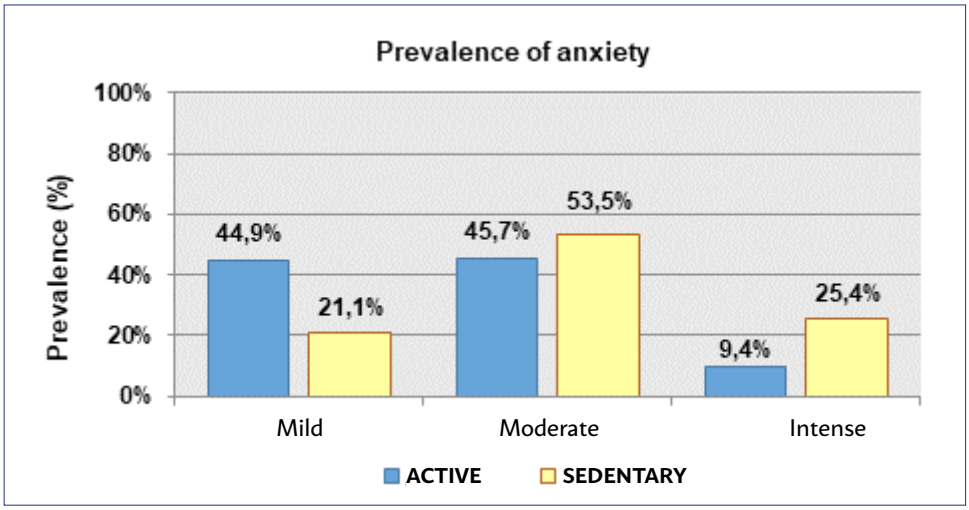

CHART 1. PREVALENCE OF ANXIETY

Since the samples were mixed regarding the distribution per sex, the distribution of clinical scores of anxiety per sex was evaluated in both groups. In both groups, there was a higher prevalence in females (OR = 2.83 for the active group and OR = 44.42 for the sedentary group). Both in the active and sedentary groups, women had higher scores of anxiety compared to men. In addition, the prevalence of severe anxiety in sedentary women is much higher if compared with the active ones.

To test the impact of age and the frequency of physical activity practice in the scores, we used the Pearson correlation (*r*). The results are summarized in Table 2. To be considered significant, the r-value must be greater than 0.3 (positive correlation) or lower than -0.3 (negative correlation). Thus, according to table 2, there was no correlation between age/anxiety scores and frequency of activity/anxiety scores.

DISCUSSION

Mens sana in corpore sano. In the 1st century A.D., Juvenal coined the famous phrase, probably from personal observation that physically active individuals had better emotional balance. It took almost 2 thousand years for scientific evidence to prove that he was right.

TABLE 2. CORRELATIONS BETWEEN AGE/FREQUENCY OF PHYSICAL ACTIVITY AND ANXIETY

	Active group r	Sedentary group r
Age x anxiety	- 0.223	- 0.04
Frequency of physical activity x anxiety	0.013	---

The present study quantified anxiety scores using the State-Trait Anxiety Inventory (STAI) in a sample of practitioners of aerobic physical activity, comparing it to a sample of sedentary, healthy individuals, using unpaired samples.

In the region of Vale do Aço, MG, there are no studies that evaluate the benefits of physical activity practice in combating anxiety, which makes this a pioneer study on the region. Furthermore, the majority of studies published to this day evaluate the benefits of physical activity in individuals with psychiatric disorders or other comorbidities. Thus, this study seeks to provide an innovative focus, evaluating only healthy individuals.

It is worth mentioning a conceptual issue. The focus of this study is the practice of physical activity for health benefits, unlike the practice of high-performance sports, which aims at improved results for competition purposes. It is also important to emphasize that the Spielberger questionnaire was not used with the purpose of defining any diagnosis, but rather to assess the propensity toward anxiety. To define a diagnosis effectively, it is necessary to consider the context in which signs and symptoms occur, the history of the individual and the evaluation of the psychiatrist.

Regarding the distribution of the groups in relation to sex and age, it is believed that there could be significant differences for these variables, which characterize a selection bias. However, when analyzing the data, we did not find such a difference using the chi-square test. Thus, although the samples were not paired per age and sex, the final result was not compromised.

The Student t-test showed a significant difference between the average anxiety scores of the groups. However, from a clinical point of view, both averages represent a clinical score compatible with moderate anxiety. For clinical relevance, we used the chi-square test to prove the difference between the two groups. The active group had significantly lower scores.

In the present study, we found a higher proportion of sedentary individuals who presented scores of intense anxiety (25.4%) when compared to those who practiced physical activity (9.4%). Mild anxiety scores, which can be considered physiological, were proportionally more significant in the active group (44.9% versus 21.1% of the sedentary group). These results are compatible with those of studies addressing similar themes, such as the Minghelli et al.[12], who also pointed out positive results of the practice of physical exercises in the reduction of anxiety symptoms.

Favorable results were also found by Stonerock et al.[13], who conducted a meta-analysis only on randomized clinical trials. In their study, on the effect of physical activity as a treatment for patients with anxiety disorder, they concluded that it is as effective as other modalities of treatment, despite the methodological limitations of some studies.

In the present study, the specific effects of the impact of the intensity of physical activity on anxiety scores were not analyzed. For this variable, there is no consensus in the literature about which intensity is ideal for mental health. While Jayakody et al.[14] found no relevant differences in their study, Conn[15], using only healthy individuals, found a greater benefit of exercises of moderate and high intensity in reducing anxiety symptoms.

This study was limited to evaluating practitioners of aerobic physical activity, comparing them with individuals with a sedentary lifestyle. A discussion on the beneficial potential of anaerobic activity, such as the bodybuilding activities, could be interesting. Jayakody et al.[14] showed that both aerobic and anaerobic physical activity have the capacity to reduce anxiety symptoms, with no difference between the modalities. Therefore, this is a topic to be explored in further investigation.

The data found in this study (higher prevalence of severe and moderate anxiety scores in the sedentary group and higher mild anxiety scores in the active group) can be justified by the fact that mild anxiety propels the individual to action, i.e., physiological, being the most significant clinically severe degree.

In the evaluation of the reasons for prevalence, it was noted that the chance of a practitioner of physical activity having high anxiety scores is three times lower than that of a sedentary individual. Although this is a significant result, we did not find in the literature data that could be compared to these results.

In relation to sex, it was evidenced that women are more prone to higher scores of anxiety. Comparing the sexes, there was a greater prevalence of intense anxiety in females, both for the active group (OR = 2.83) as well as for the sedentary group (OR = 44.42). Therefore, women present a reduction in anxiety scores with the practice of physical activity. Corroborating this study, Griffiths et al.[16] also demonstrated the benefits of physical activity on the mental health of women in all age groups studied, including older women. Whereas Herring et al.[17] reported a decrease in the levels of anxiety in young women who practice physical activity, and this reduction is related to the improvement of self-esteem.

Therefore, among the several current studies, there is sufficient evidence to prove the effectiveness of physical activity in the prevention, control and even in the treatment of anxiety disorders. This study also showed such benefits. What are the physiological mechanisms behind such significant findings?

According to Lissek[18], and Moylan et al.[19], the anxiolytic mechanisms of physical activity (PA) are not yet fully understood. However, several hypotheses have been used to explain the observed effects. Abnormalities in the fear conditioning processing are central in the physiopathology of anxiety. The excessive activation of the fear neurocircuit generates a state of chronic stress that can produce a variety of effects, which, in turn, leads to an anxiety disorder *ad eternum*. These effects include: hyperactivation of the hypothalamus-pituitary-adrenal axis (HPA) and the consequent production of cortisol; increased levels of pro-inflammatory cytokines (interleukins 1 and 6, tumor necrosis factor-alpha, interferon-gamma, among others); reactions of oxidation and free radical production, such as nitric oxide. This chronic proinflammatory state causes a reduction in the levels of neurotrophins, like the brain-derived neurotrophic factor (BDNF), which negatively affects neurogenesis in the brain and its neuroplasticity.

To Asmundson et al.[20], the PA can regulate the

HPA axis, reducing the hyperactivity of the sympathetic nervous system (seen in patients with anxiety disorders) and increase the role of the parasympathetic nervous system.

According to Viana and Andrade[21], PA is able to activate the anti-inflammatory mechanisms, with increased levels of anti-inflammatory cytokines, such as interleukin 1 and 10 receptor antagonist. In addition, regular and moderate PA reduce oxidative stress by increasing the activation of antioxidant enzymes (glutathione peroxidase, superoxide dismutase, and catalase) and increasing the synthesis of mitochondrial uncoupling protein 2 (UCP2), which increases the production of adenosine triphosphate and decreases the production of superoxide.

Moylan et al.[19] pointed out that the protective effects of PA on neurogenesis have been demonstrated by the increase in the synthesis of neurotrophic factors, including the insulin-like growth factor 1 (IGF-1) and the brain-derived neurotrophic factor (BDNF).

Other hypotheses highlight the anxiolytic effects of PA by modulating anandamide receptors and activating the endocannabinoid system, increasing the levels of circulating endocannabinoids, such as anandamide.[22,23]

The main limitation of this study was that it was not able to get a statistic per group of physical activity, trying to prove which method offers the best benefit.

This study excluded individuals who practiced activity at a frequency of fewer than three times per week, following the criteria of the AHA for defining physically active.

Therefore, the present study was able to prove the initial hypotheses, showing that the regular practice of physical activity is related to lower anxiety scores, which makes Juvenal's saying a two-way street - the body promotes a healthy mind and vice versa.

CONCLUSIONS

For the population studied, we concluded that practitioners of physical activity have lower anxiety scores.

Both man and woman who are physically active have lower anxiety scores compared by sex in relation to sedentary individuals.

No correlation was observed between the frequencies of activity (three to five times per week) and anxiety scores.

Therefore, we can affirm that the research has proven the initial hypothesis and its results can be used as raw material for future studies. In this sense, the need for defining strategies for encouraging the practice of physical activity becomes evident.

RESUMO

OBJETIVOS: *Comparar escores de ansiedade entre praticantes de atividade física e sedentários, entre homens e mulheres e relacioná-los com frequência de atividade física e idade.*

MÉTODOS: *Amostra de 256 praticantes de atividade física aeróbica regular foi comparada à amostra de 256 sedentários (grupo controle). Escores de ansiedade foram quantificados por meio do Inventário de Ansiedade Estado-Traço de Spielberger (Idate). Os escores dos grupos foram comparados por meio dos testes t de Student e qui-quadrado para dados paramétricos e não paramétricos, respectivamente. A correlação entre escores de diferentes variáveis foi realizada pelo teste de Pearson.*

RESULTADOS: *Houve diferença significativa entre os escores médios de ansiedade (p < 0,001) e o teste qui-quadrado comprovou haver maior prevalência de escores de ansiedade intensa (p < 0,001) no grupo de sedentários. Idade não se correlacionou com piores escores de ansiedade (p < 0,05). Em relação ao sexo, mulheres apresentaram maior prevalência de escores de ansiedade intensa.*

CONCLUSÕES: *Praticantes de atividade física possuem menores escores de ansiedade e ambos os sexos se beneficiam com o potencial ansiolítico da prática de atividade física. Portanto, foi comprovado que a máxima de Juvenal, Mens sana in corpore sano, também pode ser interpretada em sentido inverso, em que um corpo são se correlaciona com uma mente sã.*

PALAVRAS-CHAVE: *Ansiedade. Exercício. Estilo de vida sedentário. Psiquiatria.*

REFERENCES

1. Ströhle A. Physical activity, exercise, depression and anxiety disorders. J Neural Transm (Vienna). 2009;116(6):777-84.
2. Martínez de Morentin PB, López M. "Mens sana in corpore sano": exercise and hypothalamic ER stress. PLoS Biol. 2010;8(8). pii: e1000464.
3. World Health Organization. Global recommendations on physical activity for health. Geneva: World Health Organization; 2010.
4. Woodcock J, Franco OH, Orsini N, Roberts I. Non-vigorous physical activity and all-cause mortality: systematic review and meta-analysis of cohort studies. Int J Epidemiol. 2011;40(1):121-38.
5. Teychenne M, Costigan SA, Parker K. The association between sedentary behaviour and risk of anxiety: a systematic review. BMC Public Health. 2015;15:513.

6. Almeida JMD. Prevalência de sintomas de ansiedade e depressão em docentes do curso de Medicina da Universidade Federal da Bahia. [Monografia de Conclusão de Curso de Medicina]. Salvador: Universidade Federal da Bahia; 2013.

7. Mochcovitch MD, Deslandes AC, Freire RC, Garcia RF, Nardi AE. The effects of regular physical activity on anxiety symptoms in healthy older adults: a systematic review. Braz J Psychiatr. 2016;38(3):255-61.

8. Portney LG, Watkins MP. Foundations of clinical research: applications to practice. 3rd ed. New Jersey: Pearson Prentice Hall; 2009. p.912.

9. Motta VT. Bioestatística. 2ª ed. Caxias do Sul: EDUCS; 2006.

10. American Heart Association. American Heart Association recommendations for physical activity in adults. 2016. [cited 2018 May 5]. Available from: https://www.heart.org/en/healthy-living/fitness/fitness-basics/aha-recs-for-physical-activity-in-adults

11. Andrade L, Gorenstein C, Vieira Filho AH, Tung TC, Artes R. Psychometric properties of the Portuguese version of the State-Trait Anxiety Inventory applied to college students: factor analysis and relation to the Beck Depression Inventory. Braz J Med Biol Res. 2001;34(3):367-74.

12. Minghelli B, Tomé B, Nunes C, Neves A, Simões C. Comparação dos níveis de ansiedade e depressão entre idosos ativos e sedentários. Rev Psiq Clín. 2013;40(2):71-6.

13. Stonerock GL, Hoffman BM, Smith PJ, Blumenthal JA. Exercise as treatment for anxiety: systematic review and analysis. Ann Behav Med. 2015;49(4):542-56.

14. Jayakody K, Gunadasa S, Hosker C. Exercise for anxiety disorders: systematic review. Br J Sports Med. 2014;48(3):187-96.

15. Conn VS. Anxiety outcomes after physical activity interventions: meta-analysis findings. Nurs Res. 2010;59(3):224-31.

16. Griffiths A, Kouvonen A, Pentti J, Oksanen T, Virtanen M, Salo P, et al. Association of physical activity with future mental health in older, mid-life and younger women. Eur J Public Health. 2014;24(5):813-8.

17. Herring MP, O'Connor PJ, Dishman RK. The effect of exercise training on anxiety symptoms among patients: a systematic review. Arch Intern Med. 2010;170(4):321-31.

18. Lissek S. Toward an account of clinical anxiety predicated on basic, neurally mapped mechanisms of Pavlovian fear-learning: the case for conditioned overgeneralization. Depress Anxiety. 2012;29(4):257-63.

19. Moylan S, Eyre HA, Maes M, Baune BT, Jacka FN, Berk M. Exercising the worry away: how inflammation, oxidative and nitrogen stress mediates the beneficial effect of physical activity on anxiety disorder symptoms and behaviours. Neurosci Biobehav Rev. 2013;37(4):573-84.

20. Asmundson GJ, Fetzner MG, Deboer LB, Powers MB, Otto MW, Smits JA. Let's get physical: a contemporary review of the anxiolytic effects of exercise for anxiety and its disorders. Depress Anxiety. 2013;30(4):362-73.

21. Viana MC, Andrade LH. Lifetime prevalence, age, and gender distribution and age-of-onset of psychiatric disorders in the São Paulo Metropolitan Area, Brazil: results from the São Paulo. Megacity Mental Health Survey. Rev Bras Psychiatr. 2012;34(3):249-60.

22. Dietrich A, McDaniel WF. Endocannabinoids and exercise. Br J Sports Med. 2004;38(5):536-41.

23. Dworak M, Diel P, Voss S, Hollmann W, Strüder HK. Intense exercise increases adenosine concentrations in rat brain: implications for a homeostatic sleep drive. Neuroscience. 2007;150(4):789-95.

ORIGINAL ARTICLE

Most common histopathological patterns of the Minas Gerais Association of the Centers of Nephrology

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SUMMARY

INTRODUCTION: *We analyzed the distribution and frequency of glomerular diseases in patients biopsied between 1992 and 2016 in centers that make up the AMICEN (Minas Gerais Association of Nephrology Centers).*

METHODS: *We analyzed the biopsy reports of patients from 9 AMICEN nephrology centers. We took note of their age, gender, ultrasound use, post-biopsy resting time, whether the kidney was native or a graft, number of glomeruli and indication for the biopsy. The kidney biopsy findings were broken down into four categories: glomerular and non-glomerular diseases, normal kidneys and insufficient material for analysis. Those patients diagnosed with glomerular diseases were further divided into having primary or secondary glomerular diseases.*

RESULTS: *We obtained 582 biopsy reports. The median age was 38 years (1 to 85). The number of glomeruli varied between 0 and 70 (median = 13.0). In total, 97.8% of the biopsies were ultrasound guided. The main indication was nephrotic syndrome (36.9%), followed by hematuria-proteinuria association (16.2%). Primary glomerular diseases proved to be the most frequent (75.3%), followed by secondary diseases (24.7%). Among the primary glomerular diseases, FSGS was found at a higher frequency (28.8%), while among the secondary diseases, SLE was the most prevalent (42.4%). Regarding prevalence findings, those for both primary and secondary diseases were similar to those found in the large Brazilian registries published thus far.*

CONCLUSION: *Glomerular disease registries are an important tool to identify the prevalence of such disease in regions of interest and can serve as an instrument to guide public policy decisions concerning the prevention of terminal kidney diseases.*

KEYWORDS: *Epidemiology. Biopsy. Glomerulonephritis. Kidney Diseases.*

INTRODUCTION

Renal biopsy has been used in clinical practice for approximately 50 years.[1] Its initial description as a diagnostic method dates back to 1951 after Iversen and Brun developed the percutaneous technique.[2,3] This diagnostic modality has proven to be a valuable tool in establishing not only a diagnosis of certainty but also in evaluating disease severity and prognosis, as well as guiding the treatment of renal diseases.[4]

There has been growing interest in the prevalence of histopathological patterns of kidney diseases in

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Brazil and in the world. Some countries have a national registry of kidney histopathology. In Brazil, several attempts, although fragmented, have been made to gather such data.

Thus, we decided to contribute by trying to establish the epidemiology of the histopathological patterns found in kidney biopsies coming from "Zona da Mata" and "Campos das Vertentes" in the state of Minas Gerais, Brazil. We analyzed the distribution and frequency of glomerulopathies of patients biopsied between 1992 and 2016 in AMICEN member centers (Figure 1) and compared them with publications from national and international registries.[1-18]

METHODS

This is a cross-sectional study in which we analyzed biopsy reports of patients from 9 centers with nephrology units members of AMICEN. The kidney biopsies come from adults and children outpatients from "Zona da Mata" and "Campos das Vertentes" in the state of Minas Gerais, Brazil. The area covered by the study was mainly the Zona da Mata, with an estimated area of 35748 km², a population of 2.175.254 inhabitants, with a density of 60 in/km, a gross domestic product (GDP) per capita of US $ 2,575 and a Human Development Index (HDI) of 0.76, and the "Campos das Vertentes", with an area of 12 564 km², a population of 546,007 inhabitants, Gross domestic products (GDP) per capita of U $ 1470 and an Human development Index (HDI) of 0.77, both in the State of Minas Gerais.

This study was approved by the Human Research Ethical Board at the São João de Deus Hospital/ Fundação Geraldo Corrêa (number: 2.208.247).

The data were plotted onto a table, and supplementary information was obtained directly from the doctors of the respective clinics. After collection, the data were computed using Microsoft Office Excel 2016 software, and the results were obtained after the data were transferred to Statistical Package for Social Sciences(SPSS) 19.0 (SPSS, IBM Company, Chicago, IL). By these means, we carried out a descriptive analysis of the information collected, using data values of central tendency, variability, and frequency. Within the data gathered, we identified age, gender, ultrasound use, resting time after the biopsy, number of glomeruli and type of indication. Regarding kidney biopsy findings, the data were broken down into four categories: glomerular and non-glomerular diseases, normal kidneys, and insufficient material for analysis. The patients diagnosed with glomerulopathies were split into primary and secondary glomerulopathies. After splitting them, we analyzed the most frequent pathologies in each of these two groups.

RESULTS

In the period from 1992 to 2016, we carried out a renal biopsy study involving 672 patients from 9 nephrology centers that are associated with AMICEN. Of these, 49 were excluded because they had transplanted kidneys, as were another 41 lacking confirmed glomerular disease, thus leaving 582 biopsies remaining.

The median age was 38 years; the youngest patient was 1 year old and the eldest 85of age. Regarding the number of glomeruli, the lowest was zero, and the highest was 70, with a median of 13.0. In total, 97.8% of the biopsies were ultrasound guided. As far as patient resting time after the biopsy was concerned, we obtained data from 487 cases, of which 63.1% remained under observation for a period between 6 and 12 hours, and 20.6% for a period between 12 and 24 hours. Additionally, 49.1% of the biopsies were performed in females and 50.9% in males. In most cases (99.1%), the material was sufficient for analysis.

As noted in Table 1, the main reason for biopsy indication was nephrotic syndrome, which corresponded to 215 cases (36.9%), followed by proteinuria/ hematuria association in 94 (16.2%), hematuria alone in 34 (5.8%), acute kidney injury (AKI) in 20 (3.4%), chronic glomerulonephritis in 11 (1.9%), hematuria associated with hypertension in 2 (0.3%), and other causes in 206 (35.4%). The primary glomerulopathy was present in 438 cases (75.3%), and secondary glomerulopathy was present in 144 cases (24.7%), yield-

TABLE 1 – INDICATION OF RENAL BIOPSY

	Frequency	%
Nephrotic syndrome	215	36.9
Hematuria+ Proteinuria	94	16.2
Isolated Hematuria	34	5.8
AKI*	20	3.4
Chronic glomerulonephritis	11	1.9
Hematuria+ hypertension	2	0.3
Others	206	35.4
Total	582	100.0

*Acute Kidney Injury

ing a total of 582 cases of glomerular involvement.

Table 2 shows that 75.3% of the primary glomerulopathies could be classified as FSGS in 126 cases (28.8%), minimal change disease (MCD) in 84 cases (19.2%), IgA nephropathy in 80 cases (18.3%), membranous GN in 74 cases (16.9%), rapidly progressive GN in 27 cases (6.2%), diffuse glomerular sclerosis in 24 cases (5.5%), membranoproliferative GN (MPGN) in 15 cases (3.4%), and anti-GBM GN in 2 cases (0.5%). Other causes were found in 6 cases (1.4%).

Of the 144 cases with secondary glomerulopathy (Table 3), which represented 24.7% of the total, 61 cases (42.4%) had lupus nephritis. The second most common cause was diabetic nephropathy, which was found in 18 cases (12.5%), followed by thrombotic microangiopathy in 9 cases (6.3%), post-infectious GN in 5.6% and amyloidosis in 8 cases (5.6%). Diffuse HIV-related proliferative GN was found in 1 patient (0.7%). Other causes were found in 39 patients (27.1%).

DISCUSSION

In this study, we evaluated the different histopathological patterns in 582 patients from 9 centers of nephrology that make up AMICEN.

AKI was a reason for renal biopsy in a small percentage of our sample (3.4%). This is because most of the patients came from outpatient clinics adults and children.

As in other studies, primary glomerulopathies corresponded to the most frequent lesions, with FSGS being the most prevalent and found in 28.8% of cases.[17,18] These data are in line with those reported in the Paulista Registry of Glomerulopathy[10] and a review of 9617 histopathological findings from kidney biopsies performed in Brazil.[11] However, this is in disagreement with the Italian[7] and Spanish[9] registries, in which the most reported histologic type of primary glomerulopathy was IgAN.

IgAN was the third most frequent cause of primary glomerulopathy found in this study, corresponding to 18.3%. These data are similar to those reported in the Paulista Glomerulopathy Registry[10] and in a survey on the frequency of kidney biopsies performed in Brazil,[11] in which IgA nephropathy was also the third most frequently found histopathological pattern.[14,17] However, we must admit that several Brazilian nephrology centers belonging to AMICEN do not biopsy patients with hematuria alone, but only if they also present proteinuria, hypertension, or kidney function impairment. If these centers would biopsy their patients, it is very likely that the most common form of primary glomerulopathy found would be IgAN.

In this study, we found that MGN represented 16.9% of primary glomerulopathy cases, ranking fourth among the subgroups of primary glomerulopathies. Such data are in disagreement with a study carried out in the southern region of Brazil,[14] the Paulista Glomerulopathy Registry,[10] and the review of diagnoses by renal biopsy in Brazil[11].

Worldwide, especially in developed countries, there has been a reduction in the frequency of cases of infection-related glomerulopathies, such as membranoproliferative GN and post-infectious GN. In this study, we found that membranoproliferative GN was the seventh most common histopathological pattern among primary glomerulopathies, corresponding to

TABLE 2 – PRIMARY GLOMERULOPATHIES

	Frequency	%
FSGS	126	28.8
MCD	84	19.2
IgAN	80	18.3
MGN	74	16.9
RPGN	27	6.2
Diffuse Glomerular Sclerosis	24	5.5
MPGN	15	3.4
Anti-GBM GN	2	0.5
Other primary GN	6	1.4
Total	438	100.0

FSGS: Focal Segmental Glomerulosclerosis; MCD: Minimal Change Disease; IgAN: IgA Nephropathy; MGN: Membranous Glomerulonephritis; RPGN: Rapidly Progressive Glomerulonephritis; MPGN: Membranoproliferative Glomerulonephritis; Anti-GBM GN: Anti- Glomerular Basement Membrane Glomerulonephritis; Other primary GN: Other primary Glomerulonephritis

TABLE 3- SECONDARY GLOMERULOPATHIES

	Frequency	%
SLE	61	42.4
DM	18	12.5
Thrombotic microangiopathy	9	6.3
Post-infectious GN	8	5.6
Amyloidosis	8	5.6
HIV-related GN	1	0.7
Other secondary GN	39	27.1
Total	144	100.0

SLE: Systemic Lupus Erythematosus; DM: Diabetes Mellitus; GN: Glomerulonephritis

only 3.4% of cases, while post-infectious GN corresponded to 5.6% of secondary glomerulopathy cases. Such data are in disagreement with the Pernambuco Registry of Glomerulopathies (REPEG).[17]

Lupus nephropathy was the most common type of secondary glomerulopathy found, corresponding to 42.4% of cases. The low prevalence of diabetic nephropathy in our sample stems from the fact that we only biopsied diabetic nephropathy patients when they presented with a high degree of atypia. On the contrary, diabetic nephropathy would probably be regarded as the main cause of secondary GN in our population.

CONCLUSION

This study provides data regarding the frequency of histopathological findings from renal biopsies of 9 nephrology centers in the state of Minas Gerais. We found that FSGS was the most frequent primary glomerulopathy while lupus nephropathy was the main type of secondary glomerulopathy and that a low incidence of glomerulonephritis was associated with infectious conditions.

Creating and expanding these registries to identify the epidemiology of kidney diseases in our country will help create policies that can fight these diseases early in life in order to prevent an increase in the number of patients with chronic kidney failure, which is an important public health problem.

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RESUMO

INTRODUÇÃO: *Analisamos a distribuição e frequência de doenças glomerulares de pacientes biopsiados entre 1992 e 2016 em centros que compõem a Amicen (Associação de Minas Gerais de Nefrologia).*

MÉTODOS: *Analisamos os relatórios de biópsia de pacientes de nove centros de nefrologia da Amicen. Observamos idade, gênero, uso de ultrassom, tempo de descanso pós-biópsia, se o rim era nativo ou um enxerto, número de glomérulos e indicação para a biópsia. Os achados da biópsia do rim foram divididos em quatro categorias: doenças glomerulares e não glomerulares, rins normais e material insuficiente para análise. Os pacientes diagnosticados com doenças glomerulares foram ainda divididos em doenças glomerulares primárias ou secundárias.*

RESULTADOS: *Obtivemos 582 relatórios de biópsia. A idade mediana foi de 38 anos (1 a 85). O número de glomérulos variou entre zero e 70 (mediana = 13,0). No total, 97,8% das biópsias foram guiadas por ultrassom. A principal indicação foi síndrome nefrótica (36,9%), seguida de associação hematúria-proteinúria (16,2%). As doenças glomerulares primárias revelaram-se as mais frequentes (75,3%), seguidas de doenças secundárias (24,7%). Entre as doenças glomerulares primárias, o FSGS foi encontrado em maior frequência (28,8%), enquanto nas doenças secundárias, o lúpus eritematoso sistêmico foi o mais prevalente (42,4%). Quanto aos achados de prevalência, aqueles para doenças primárias e secundárias foram semelhantes aos encontrados nos grandes registros brasileiros publicados até o momento.*

CONCLUSÃO: *Os registros de doenças glomerulares são uma ferramenta importante para identificar a prevalência dessas doenças em regiões de interesse e pode servir como um instrumento para orientar decisões de políticas públicas relativas à prevenção de doenças renais terminais.*

Palavras-chave: Epidemiologia. Biópsia. Glomerulonefrite. Nefropatias.

REFERENCES

1. Cardoso ACD, Kirsztajn GM. Padrões histopatológicos das doenças glomerulares no Amazonas. J Bras Nefrol. 2006;28(1):39-43.
2. Cohen AH, Nast CC, Adler SG, Kopple JD. Clinical utility of kidney biopsies in the diagnosis and management of renal disease. Am J Nephrol. 1989;9(4):309-15.
3. Iversen P, Brun C. Aspiration biopsy of the kidney 1951. Am J Med. 1951;11(3):324-30.
4. Oliveira VS, Vieira Junior AE, Barreto JCS, Ramos Filho R. Biópsia renal: experiência do Hospital Geral de Goiânia. J Bras Nefrol. 2004;3(Suppl. 2):51.
5. Richards NT, Darby S, Howie AJ, Adu D, Michael J. Knowledge of renal histology alters patients management in over 40% of cases. Nephrol Dial Transplant. 1994;9(9):1255-9.
6. Barros RT, Alves MAVER, Dantas M, Kirsztajn GM, Sens YAS. Biópsia renal. In: Lima EQ, Barros RT, eds. Glomerulopatias: patogenia, clínica e tratamento. 2ª ed. São Paulo: Servier; 2006. p.100-5.
7. Gesualdo L, Di Palma AM, Morrone LF, Strippoli GF, Schena FP; Italian Immunopathology Group, Italian Society of Nephrology. The Italian experience of the national registry of renal biopsies. Kidney Int. 2004;66(3):890-4.

8. Mazzuchi Frantchez N, Di Martino Compte LA. Epidemiología de las glomerulopatías primarias en el Uruguay. Arch Med Interna. 1997;19(1):21-6.

9. Rivera F, López-Gómez JM, Pérez-García R; Spsnish Registry of Glomerulonephritis. Frequency of renal pathology in Spain 1994-1999. Nephrol Dial Transplant. 2002;17(9):1594-602.

10. Andrade LCF; Vieira PA, Reis MF, Pernambuco JM, Franco MN, Bastos MG. Análise de 121 biópsias renais (BXR) comparadas com o Registro Paulista de Glomerulopatias (RPG). J Bras Nefrol. Proceedings of the XXII Congresso Brasileiro de Nefrologia. 2004. Sep 18-22. Salvador, Bahia:Brasil;2004.p 9.

11. Polito MG, Moura LA, Kirsztajn GM. An overview on frequency of renal biopsy diagnosis in Brazil: clinical and pathological patterns based on 9,617 native kidney biopsies. Nephrol Dial Transplant. 2010;25(2):490-6.

12. Ferraz FHRP, Martins CGB, Cavalcanti JC, Oliveira FL, Quirino RM, Chicon R, et al. Perfil das doenças glomerulares em um hospital público do Distrito Federal. J Bras Nefrol. 2010;32(3):249-56.

13. Malafronte P, Mastroianni-Kirsztajn G, Betônico GN, Romão JE Jr, Alves MA, Carvalho MF, et al. Paulista Registry of glomerulonephritis: 5-year data report. Nephrol Dial Transplant. 2006;21(11):3098-105.

14. Crensiglova C, Rehme BB, Kinasz LR, Chula DC, Nascimento MM, Soares MF. Frequency and clinical histological analysis of glomerular diseases in a tertiary hospital in southern Brazil. J Bras Nefrol. 2016;38(1):42-8.

15. Sugiyama H, Yokoyama H, Sato H, Saito T, Kohda Y, Nishi S, et al.; Committee for Standardization of Renal Pathological Diagnosis; Committee for Kidney Disease Registry; Japanese Society of Nephrology. Japan Renal Biopsy Registry and Japan Kidney Disease Registry: Committee Report for 2009 and 2010. Clin Exp Nephrol. 2013;17(2):155-73.

16. Kasamatsu E, Nunes V MC, Morán M, Centurión M, Campos de Alvarenga S. Glomerulopathies in Paraguay. Report of Registry of Renal Biopsies in 1072 cases. Mem Inst Investig Cienc Salud. 2005;3:51-7.

17. Costa, DM, Valente LM, Gouveia PA, Sarinho FW, Fernandes GV, Cavalcante MA, et al. Comparative analysis of primary and secondary glomerulopathies in the northeast of Brazil: data from the Pernambuco Registry of Glomerulopathies - REPEG. J Bras Nefrol. 2017;39(1):29-35.

18. Rocha LP, Carminati CR, Machado JR, Laterza VL, Reis MA, Corrêa RR. Prevalence of nephropathies in children and adolescents and alterations in renal biopsies in Minas Gerais, Brazil, from 1996 to 2010. Ann Diagn Pathol. 2013;17(1):22-7.

ORIGINAL ARTICLE

Investigation of the effect of the virtual reality application on experimental pain severity in healthy

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SUMMARY

OBJECTIVE: *This study aimed to investigate the effect of virtual reality application on experimental ischemic pain created with a blood pressure instrument in healthy volunteers.*

METHODS: *The research sample consisted of 172 volunteer adult students who conformed to the inclusion criteria. These individuals were assigned into an experimental (n=86) and a control group (n=86) by a simple randomization method. All individuals in the experimental and control groups wereexperimentally subjected to pain for two minutes by applying 260 mmHg of pressure 3-4 cm above the antecubital region of the left arm with an aneroid adult-type blood pressure instrument. During the procedure, the volunteers in the experimental group watched virtual reality images, while those in the control group received no intervention. Immediately after the procedure, the pain levels of the individuals in both groups were assessed with a Visual Analog Scale (VAS).*

RESULTS: *We found that the mean pain score of the individuals in the experimental group was 2.62±1.82, and that of individuals in the control group was 5.75±1.65. Results of the statistical analysis showed a statistically significant difference between the mean pain scores of the individuals in the experimental and control groups (p<0.000).*

CONCLUSION: *This study found that the use of virtual reality was effective in reducing the level of pain in healthy individuals. This method used a smartphone with widespread availability and ease of transportation, which can be used by health professionals as a non-pharmacological method in the management of pain.*

KEYWORDS: *Virtual Reality. Pain. Distraction.*

INTRODUCTION

Distraction is a cognitive behavioral technique used to reduce the sensation of pain[1]. This technique has been developed following theories that attempt to explain the mechanism of pain, which was initially seen as a reflex event. In opposition to this idea, Melzack and Wall[2,3] proposed that pain signals do not reach the brain as soon as damage is caused. According to the Gate Control Theory that they developed, gate mechanisms exist at the level of the spinal cord and control the passage of pain. According to the neurocognitive model of attention to pain, which supports this theory, when more than

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one sensory signal arrives at the brain at the same time, the strongest is chosen and reaches the level of consciousness[4]. Thus, it is maintained that drawing a person's attention away from pain will reduce the level of pain felt[1].

The use of virtual reality (VR) based on the gate control theory is a method which uses attention processes to achieve a distancing from pain[5]. It is a simulation model which gives participants a feeling of reality, created by computer, and allows interaction. Today, three dimensional VR is possible on a smartphone by dividing the screen into two (for right and left eye images). A smart device can create a multi-dimensional and realistic image using a headset[6-8]. In this way, VR functions as a non-pharmacological method in the management of pain using cognitive and attention processes on the complex pain modulation system of the body[9].

Recently, the use of VR in pain management in health care has become more widespread. Studies in the literature show the effectiveness of VR in pain management in operations such as the debridement of burns, episiotomy, wound care, dental treatment, endoscopy, insertion of catheters, and chemotherapy[10-15]. Contrary to the results of these studies, others have reported that VR was not effective in pain management[16-17]. There are differences in the efficacy of its application in studies conducted on patients. Therefore, determining the effectiveness of the application in healthy individuals can provide a chance to evaluate it by eliminating factors which could affect pain levels such as the hospital environment, the psychology of being sick, the effects of analgesics, and the clinical situation[13,16]. This study aimed to determine the effect of virtual reality on the level of pain inflicted experimentally to healthy individuals by eliminating factors which could affect pain in the clinical environment.

METHOD

This was a randomized, single-blind, quasi-experimental study.

Setting and sample

The research was conducted between July and September 2017 at a university in the north of Turkey. The research sample consisted of 172 students, all healthy males and females. After a pilot study, a certain number of samples was collected, and a power analysis was performed using descriptive statistics. The sample size of each experimental group was calculated as 86 for 0.90 power. The relevant calculation was made using PASS 13.0 (PASS, Kaysville, Utah, USA).

The criteria for inclusion in the study were as follows: not having problems such as fracture, dislocation, crushing of the soft tissue or any circulation disorder of the upper limbs, no diseases which could

FIGURE 1. THE VR EQUIPMENT USED IN THIS STUDY WITH "ROLLER COASTERS" MEDIA WATCHED USING A SMARTPHONE LOCATED IN A VRBOX HEADSET.

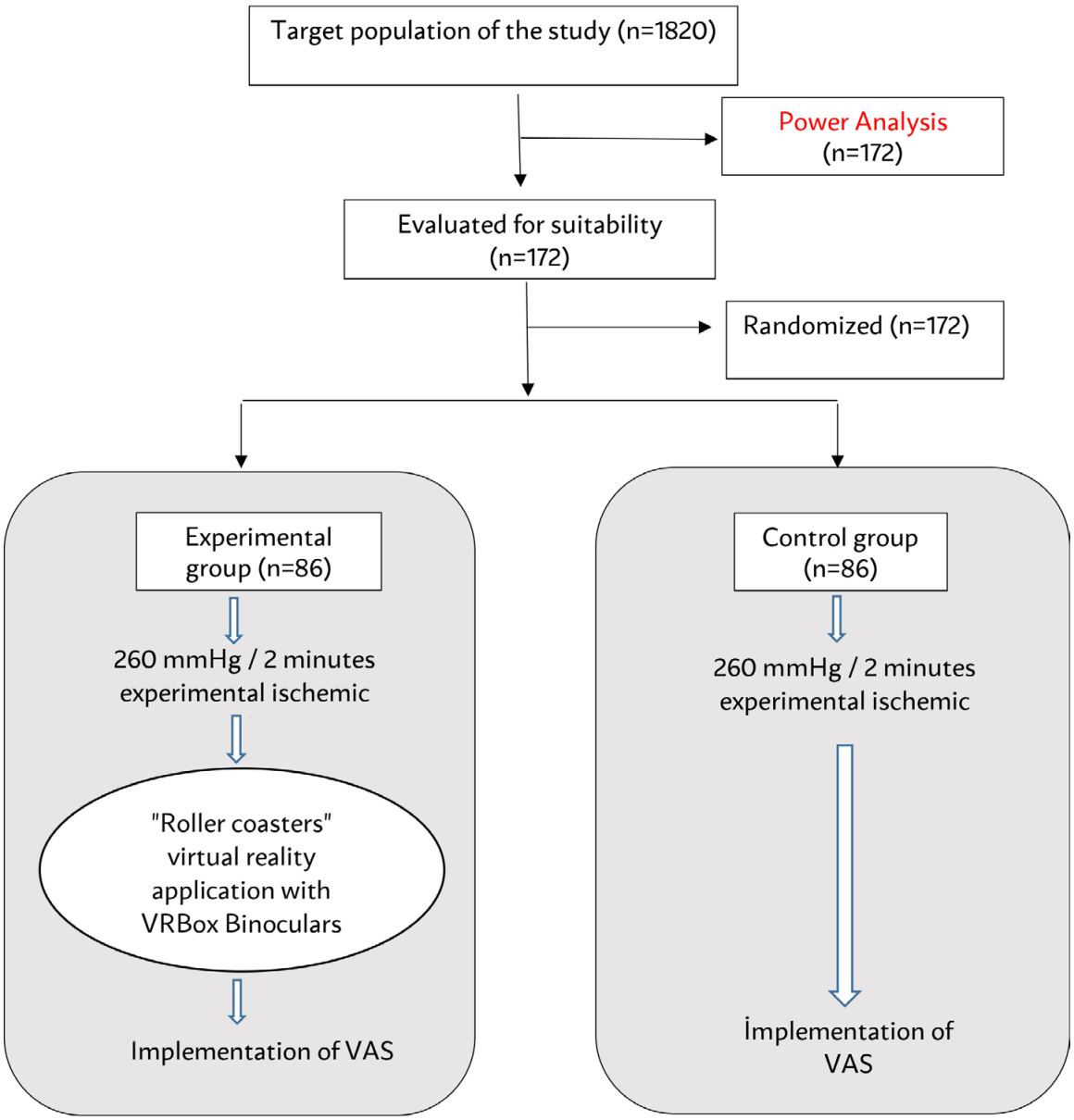

FIGURE 2. THE RESEARCH DESIGN APPLIED IN THIS STUDY

affect pain, no sight or hearing impairments. The students selected were not students of any of the researchers conducting the study and had no relationship of mutual interest with the teachers.

The students were randomized according to age and gender and randomly divided into an experimental and a control group using the coin method. Listed students were allocated into the experimental group (n=43 female and n=43 male) and the control group (n=43 female and n=43 male), taking into account the gender factor by an independent researcher.

Ethical considerations

The necessary legal permission to carry out the research was obtained from the Local Ethics Committee (Decision No. 2017-246). Also, the individuals were given information concerning the research before the study started. After information was given, they signed an informed consent form concerning their voluntary participation.

Measurements

An individual identification form was used to collect information on individuals' age and gender. A

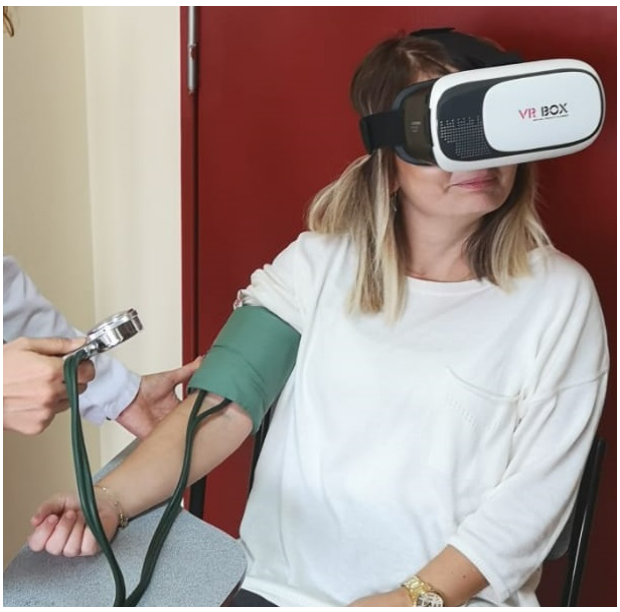

FIGURE 3. USING VR EQUIPMENT DURING PRACTICE

10 cm vertical Visual Analog Scale (VAS) was used to evaluate the intensity of pain felt by the individuals during the procedure. One end indicated no pain, and the other end the worst possible pain[18]. Pain intensity scores were assessed in cm.

"Roller coasters" was watched with a smartphone located in a VRBox headset. The images and realistic sounds created the feeling of being on a roller coaster in a virtual environment (Figure 1). Roller coaster is an application that gives the impression that people are moving in the speed of a train.

Data Collection

At the preparation stage of the study, an investigation was made with 10 people who were the same as the sample group, with the same equipment, and the same body part, to determine the mean mmHg and mean number of minutes needed to produce a sensation of pain. The minutes and pressure values at which these individuals felt the highest level of pain were determined and recorded. The averages of the determined pressure and minute values were calculated. As a result, the pain threshold for healthy individuals was set at 260 mmHg for two minutes.

After the participants had been assured that their participation was voluntary, their identification data was collected on the form, and then they were given information on the use of the Visual Analog Scale. After that, in order to achieve standardization, the left arm of all individuals was supported at the level of the heart and 260 mmHg pressure was applied 3-4 cm above the antecubital region for two minutes using an aneroid adult-type blood pressure instrument in order to induce pain.

While experimental ischemic pain was being applied to individuals in the experimental group, they were given "Roller Coasters" to watch using a virtual reality app on an android mobile phone placed in a VRBox headset (Figure 3). "Roller Coasters" was watched by the individuals in the experiment group, starting from one minute before the start of the procedure. After this procedure, the individuals' pain levels were assessed by an independent researcher using the VAS.

While experimental ischemic pain was being applied to individuals in the control group, no procedure was carried out to distract their attention. After the Experimental Ischemic Pain was applied, the individuals' pain levels were assessed by an independent researcher using the VAS. The research design of this study is shown in Figure 2.

Data analysis

Statistical analysis was performed using the Statistical Package for Social Sciences version 17.0 (SPSS Inc., Chicago, IL, USA). Shapiro-Wilk was used to examine normal distribution in numerical data. Distributions of identification data were given as means and standard deviation. The Mann-Whitney U test was used to compare both groups. A p-value < 0.05 was considered significant.

RESULTS

There was a total of 172 participants in the study. The mean age of all participants was 19.69 ± 0.77 years. The mean age of individuals in the experimental groups was 20.0 ± 1.00 years, and in the control group, it was 19.38 ± 0.94 years. The differences between both groups in terms of gender and age were found not to be statistically significant ($p > 0.05$), and the groups were found to have homogeneous distribution.

When the mean pain scores of the groups were examined, we found that the mean pain score of individuals in the experimental group was 2.62 ± 1.82, while that of the individuals in the control group was 5.75 ± 1.65. The mean pain score of the individuals in the experimental group was found to be statistically significantly lower than that of the individuals in the control group ($p < 0.001$) (Table 1).

TABLE 1. DISTRIBUTION OF PAIN SCORE OF EXPERIMENTAL AND CONTROL GROUPS

	Experimental group	Control group
	Mean ± SD	Mean ± SD
Pain score	2.62 ± 1.82	5.75 ± 1.65
Z= -8.92 p<.001		

SD = standart deviasyon; Z= Mann-Whitney U test

DISCUSSION

Pain is an unpleasant sensory and emotional experience in connection with actual or potential tissue damage or explained by this damage. Individuals can feel pain by focusing their limited attention capacity on the stimulus. For this reason, if a person's attention is directed away from the painful stimulus, the intensity of the pain experienced will be reduced[9]. Distraction is a cognitive behavioral technique used to reduce pain. Currently, this technique can be applied by means of the spatial illusion of virtual reality[1]. Virtual reality technology enables people to become immersed in a computer-simulated, three-dimensional environment[13].

There are differences in the results of clinical research evaluating the efficacy of VR on pain. JahaniShoorab et al[11] reported that patients who used virtual reality had lower pain scores during episiotomy repair. Similarly, in the study conducted by Gou et al[10], VR was effective in pain management on patients who had hand injury undergoing dressing change. However, Walker et al[17] reported that the effect of VR on pain did not differ between the two groups studied during cystoscopy using lidocaine jelly.

In nonclinical research, VR has a positive effect on experimental pain. When pain was induced in healthy individuals by means of thermal stimuli, their brain functions were assessed using the functional magnetic resonance imaging technique, and it was found that the activity of the areas concerned with pain was significantly reduced in individuals using VR[19]. Supporting this result, virtual reality has been found to be effective in reducing pain created experimentally by cold pressure[20]. Sulea et al[21] induced pain in healthy individuals using cold stimulus and mild electric shock methods and reported significantly lower pain scores in a group watching VR than in a group not watching VR. Another study comparing different methods concluded that for finger pressure pain, VR was a more effective method than hypnosis[22]. The results of our study are similar to that of other studies that investigated the effect of VR on pain. We too found that the use of virtual reality in healthy individuals subjected to pain by means of blood pressure was effective in reducing the levels of pain felt by diverting the attention away from the pain. Differently from other similar study results, our study used smartphones instead of computer-based software. VR can be used as a easy, cheap, and non-invasive method of pain management thanks to smartphones that have widespread availability and ease of transportation.

CONCLUSION

We found that the use of virtual reality to divert the attention elsewhere when healthy adults in the sample group were subjected to pain was an effective method in reducing the pain felt. Virtual reality created by smartphones can be effective for pain control and easily applied by health professionals for pain management aside from analgesic treatment.

Limitations

The study had some limitations. The results of the study are limited by the self-reporting of the individuals in the sample group. The method used in the study to inflict pain was manual. Also, although the pressure which created pain in the sample group of the study was determined as a standard in a pilot study, the true pain thresholds of individuals may be different. Thus, a further limitation in the study was that the pain thresholds of participants were not measured.

RESUMO

OBJETIVO: *El objetivo de este estudio fue investigar el efecto de la aplicación de realidad virtual en el dolor isquémico experimental creado con un instrumento de presión arterial en voluntarios sanos.*

MÉTODO: *La muestra de investigación consistió en 172 estudiantes adultos voluntarios que cumplieron con los criterios de inclusión. A estos individuos se les asignó mediante un método de aleatorización simple en un grupo experimental (n = 86) y uno de control (n = 86). Todos los individuos en los grupos experimentales y de control fueron sometidos experimentalmente a dolor durante dos minutos aplicando 260 mmHg de presión 3-4 cm por encima de la región antecubital del brazo izquierdo con un instrumento de presión arterial*

aneroide tipo adulto. Durante el procedimiento, los voluntarios en el grupo experimental observaron imágenes de realidad virtual, mientras que los del grupo de control no recibieron ninguna intervención. Inmediatamente después del procedimiento, los niveles de dolor de los individuos en ambos grupos se evaluaron con una Escala Analógica Visual (EAV).

RESULTADOS: *Se encontró que el puntaje promedio de dolor de los individuos en el grupo experimental fue 2.62 ± 1.82, y el de los individuos en el grupo control fue de 5.75 ± 1.65. Los resultados del análisis estadístico mostraron una diferencia estadísticamente significativa entre las puntuaciones medias de dolor de los individuos en los grupos experimental y control (p<0,000).*

CONCLUSÃO: *Se encontró en este estudio que el uso de la realidad virtual fue efectivo para reducir el nivel de dolor en individuos sanos. Este método, que se lleva a cabo mediante el uso del teléfono inteligente y que ofrece una amplia disponibilidad y facilidad de transporte, puede ser utilizado por profesionales de la salud como un método no farmacológico en el tratamiento del dolor.*

PALAVRAS-CHAVE: *Realidad virtual. Dolor. Distracción.*

REFERENCES

1. Sil S, Dahlquist LM, Thompson C, Hahn A, Herbert L, Wohlheiter K, et al. The effects of coping style on virtual reality enhanced videogame distraction in children undergoing cold pressor pain. J Behav Medi. 2014;37(1):156–65.
2. Dissanayake DWN, Dissanayake DMD. The physiology of pain: an update and review of clinical relevance. J Ceylon Coll Phys. 2016;46(1-2):19–23.
3. Melzack R. Gate control theory: On the evolution of pain concepts. Pain Forum. 1996;5(2):128-38.
4. Legrain V, Damme SV, Eccleston C, Davis KD, Seminowicz DA, Crombez G. A neurocognitive model of attention to pain: behavioral and neuroimaging evidence. Pain. 2009;144(3):230–2.
5. Wiederhold BK, Gao K, Kong L, Wiederhold MD. Mobile devices as adjunctive pain management tools. Cyberpsychol Behav Soc Netw. 2014;17(6):385–9.
6. Amer A, Peralez, P. Affordable altered perspectives: making augmented and virtual reality technology accessible. [Internet]. Paper presented at the Global Humanitarian Technology Conference, San Jose, CA, USA; 2014 [cited 2018 March 1]. Available from: http://ieeexplore.ieee.org/document/6970345/
7. Kim KH, Kabir E, Jahan SA. The use of personal hair dye and its implications for human health. Envir Int. 2016;89–90:222–7.
8. Sun F, Zhang Z, Liao D, Chen T, Zhou J. A lightweight and cross-platform Web3D system for casting process based on virtual reality technology using WebGL. Int J Adv Manufacturing Tech. 2015;80(5–8):801–16.
9. Li A, Montaño Z, Chen VJ, Gold JI. Virtual reality and pain management: current trends and future directions. Pain. 2011;1(2):147–57.
10. Guo C, Deng H, Yang J. Effect of virtual reality distraction on pain among patients with hand injury undergoing dressing change. J Clin Nurs. 2015;24(1–2):115–20.
11. Jahanishoorab N, Zagami SE, Nahvi A, Mazluom SR, Golmakani N, Talebi M, et al. The effect of virtual reality on pain in primiparity women during episiotomy repair: A randomize clinical trial. Iranian J Med Sci. 2015;40(3):219–24.
12. Maani CV, Hoffman HG, Morrow M, Maiers A, Gaylord K, McGhee LL, et al. Virtual reality pain control during burn wound debridement of combat-related burn injuries using robot-like arm mounted VR goggles. J Trauma. 2011;71(1):125–30.
13. Malloy KM, Milling LS. The effectiveness of virtual reality distraction for pain reduction: a systematic review. Clin Psychol Rev. 2010;30(8):1011–8.
14. Triberti S, Repetto C, Riva G. Psychological factors influencing the effectiveness of virtual reality–based analgesia: a systematic review. Cyberpsychol Behav Soc Netw. 2014;17(6):335–45.
15. Pandya PG, Kim TE, Howard SK, Stary E, Leng JC, Hunter OO, et al. Virtual reality distraction decreases routine intravenous sedation and procedure-related pain during preoperative adductor canal catheter insertion: a retrospective study. Korean J Anesthesiol. 2017;70(4):439-45.
16. Chan EA, Chung JW, Wong TK, Lien AS, Yang JY. Application of a virtual reality prototype for pain relief of pediatric burn in Taiwan. J Clin Nurs. 2007;16(4):786-93.
17. Walker MR, Kallingal GJ, Musser JE, Folen R, Stetz MC, Clark JY. Treatment efficacy of virtual reality distraction in the reduction of pain and anxiety during cystoscopy. Mil Med. 2014;179(8):891-6.
18. Kahl C, Cleland JA. Visual analogue scale, numeric pain rating scale and the McGill pain Questionnaire: an overview of psychometric properties. Physical Ther Revi. 2005;10(2):123–8.
19. Hoffman HG., Richards TL, Coda B, Bills AR., Blough D, Richards AL, et al. Modulation of thermal pain-related brain activity with virtual reality: evidence from fMRI. Neuroreport. 2004;15(8):1245–8.
20. Loreto-Quijada D, Gutiérrez-Maldonado J, Nieto R, Gutiérrez-Martínez O, Ferrer-García M, Saldaña C, et al. Differential effects of two virtual reality interventions: distraction versus pain control. Cyberpsychol Behav Soc Netw. 2014;17(6):353–8.
21. Sulea C, Soomro A, Wiederhlod BK, Wiederhlod MD. Quantifying the effectiveness of virtual reality pain management: a pilot study. Stud Health Tecnol Inform. 2014;199:94-7.
21. Enea V, Dafinoiu I, Opriş D, David D. Effects of hypnotic analgesia and virtual reality on the reduction of experimental pain among high and low hypnotizables. Int J Clin Exp Hypn. 2014;62(3):360–77.

REVIEW ARTICLE

Rational use of diagnostic tests for clinical decision making

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SUMMARY

OBJECTIVE: *To assist clinicians to make adequate interpretation of scientific evidence from studies that evaluate diagnostic tests in order to allow their rational use in clinical practice.*

METHODS: *This is a narrative review focused on the main concepts, study designs, the adequate interpretation of the diagnostic accuracy data, and making inferences about the impact of diagnostic testing in clinical practice.*

RESULTS: *Most of the literature that evaluates the performance of diagnostic tests uses cross-sectional design. Randomized clinical trials, in which diagnostic strategies are compared, are scarce. Cross-sectional studies measure diagnostic accuracy outcomes that are considered indirect and insufficient to define the real benefit for patients. Among the accuracy outcomes, the positive and negative likelihood ratios are the most useful for clinical management. Variations in the study's cross-sectional design, which may add bias to the results, as well as other domains that contribute to decreasing the reliability of the findings, are discussed, as well as how to extrapolate such accuracy findings on impact and consequences considered important for the patient. Aspects of costs, time to obtain results, patients' preferences and values should preferably be considered in decision making.*

CONCLUSION: *Knowing the methodology of diagnostic accuracy studies is fundamental, but not sufficient, for the rational use of diagnostic tests. There is a need to balance the desirable and undesirable consequences of tests results for the patients in order to favor a rational decision-making approach about which tests should be recommended in clinical practice.*

KEYWORDS: *Clinical Decision-Making.* Diagnostic Tests, Routine. *Evidence-Based Practice. Sensitivity and specificity. Predictive value of tests. Diagnostic equipment.*

INTRODUCTION

Diagnostic tests are used in clinical practice to identify the presence, absence or characteristics of a condition of interest in a patient, aiming to develop a plan to the appropriate treatment[1]. Additionally, they can be used to identify physiological changes, establish a prognosis, monitor the progress of a disease or response to treatment, and assist in clinical management.

A new test can play one of three fundamental roles: 1) act as a screening test (to minimize the use of invasive or expensive tests, as, for example, the use of magnetic resonance imaging before a biopsy in patients with prostate cancer); 2) replace a current test (by presenting a higher precision, being less invasive, having a shorter execution time or lower cost - for example, patients with pneumonia associated with mechanical ventilation who received a quick test of antimicrobial susceptibility had their results, on av-

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erage, 2.8 days before the standard tests for susceptibility and were able to receive early treatment, with better prognosis[2]; or 3) can be added to an existing test (improve the diagnostic accuracy of an existing test - for example, myocardial perfusion study and electrocardiogram in coronary artery disease)[3].

To recommend a diagnostic test in clinical practice, you must consider its impact on the improvement of outcomes that are important to the patient, such as those related to the disease (for example, reduction of mortality), reduction of the psychological consequences of tests to patients, reduction of risks associated to the test and also evaluate the outcomes related to the use of resources[4]. However, in clinical practice, decision making is often based only on parameters of test accuracy (sensitivity and specificity, among others), which is not the most appropriate approach, considering that the parameters of accuracy are considered surrogate endpoints capable of providing indirect evidence about the effectiveness of the diagnostic strategy adopted. It is necessary to bear in mind that merely establishing a diagnosis does not provide information about whether a patient or group of patients will benefit from the diagnosis[5].

New diagnostic tests are constantly developed, driven by demands for improvements in diagnostic speed, cost, ease of application, patient safety, and accuracy[6]. However, the efforts to detect diseases early can always be accompanied by unintentional damage. These include false-positive results and indeterminate discoveries that may worry patients, lead to more tests, increase the load of clinical work and distract doctors from the most important work. Also, excessive diagnoses may lead to unnecessary treatments[6]. Therefore, in clinical practice, it is common to have more than one option of test available for diagnosing a certain condition.

Thus, this review aims to discuss topics that help the decision/recommendation of a diagnostic test in clinical practice, reviewing the key concepts, study design, the proper interpretation of data accuracy, and inferring the clinical impact of the test in the diagnosis and the implications of choosing it.

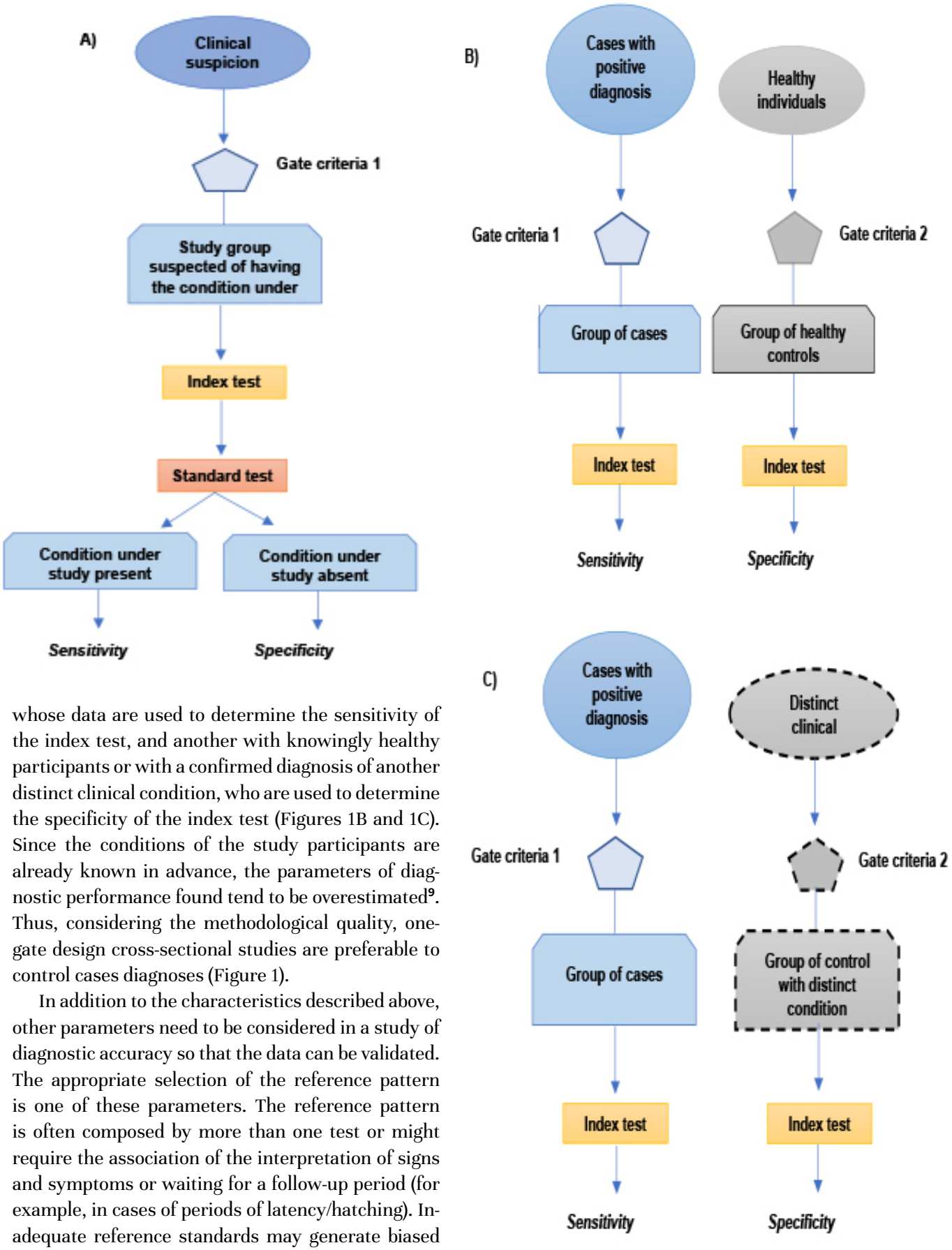
IDENTIFYING AND ASSESSING THE RISK OF BIAS IN STUDIES OF DIAGNOSTIC ACCURACY

Considering results in clinical practice, the best way to evaluate any diagnostic strategy, especially new ones with supposedly superior accuracy, is through a randomized clinical trial in which patients are grouped randomly into experimental diagnostic approaches and control, and the clinically relevant outcomes are measured[7,8]. For example, a randomized study on the diagnosis of tuberculosis compared the applicability and diagnostic accuracy between the Xpert MTB/RIF and the sputum smear microscopy, the latter being the most commonly used[8]. The cell culture was used as the gold standard. The TBscore for positive cultures was similar for both groups, and both showed high values of sensitivity and specificity. However, the Xpert test had more patients with a positive culture who started treatment on the same day and required a shorter treatment time[8].

As previously mentioned, studies on diagnostic tests rarely focus on this type of outcome, but on those that reflect the diagnostic performance of the test. Most of the literature available is based on specific randomized studies of diagnostic accuracy, which, by definition, is a cross-sectional study. The new test is referred to as the index test in the study, while the test that is already used in clinical practice, formerly the gold standard, is referred to as the reference standard. Since this study design allows variations and may be subject to bias in various stages, the first major challenge encountered by clinicians is the critical evaluation of the quality of the body of scientific evidence. Like any scientific literature, this is vast and needs to be interpreted using criteria as a basis for the decision on estimates that can be considered reliable.

Two main variations of studies of diagnostic accuracy are the most commonly used and they differ, essentially, on the source of inclusion of study participants: one-gate design cross-sectional studies of diagnostic accuracy (Figure 1A) and case-control diagnosis (*two-gate design*) (Figures 1B and 1C). One-gate design studies have a single source of study participants, all suspected of having the disease, but it is not known beforehand if the diagnosis is positive or negative. Thus, all participants in the study are submitted to both tests (index test and reference standard), and the accuracy outcomes of the index test are determined based on the diagnostic findings obtained from the standard reference. Whereas in case-control diagnosis studies (which differ from the concept of epidemiological case-control), two distinct sources of study participants are used: one that includes participants with a positive diagnosis of the disease, obtained using the reference standard,

FIGURE 1. TYPES OF STUDIES OF DIAGNOSTIC ACCURACY. A) CLASSIC DESIGN OF A CROSS-SECTIONAL DIAGNOSIS STUDY; B) CASE-CONTROL DIAGNOSIS USING HEALTHY CONTROLS; C) CASE-CONTROL DIAGNOSIS USING CONTROLS WITH A DISTINCT CLINICAL CONDITION. ADAPTED FROM RUTJES ET AL.[10]



whose data are used to determine the sensitivity of the index test, and another with knowingly healthy participants or with a confirmed diagnosis of another distinct clinical condition, who are used to determine the specificity of the index test (Figures 1B and 1C). Since the conditions of the study participants are already known in advance, the parameters of diagnostic performance found tend to be overestimated[9]. Thus, considering the methodological quality, one-gate design cross-sectional studies are preferable to control cases diagnoses (Figure 1).

In addition to the characteristics described above, other parameters need to be considered in a study of diagnostic accuracy so that the data can be validated. The appropriate selection of the reference pattern is one of these parameters. The reference pattern is often composed by more than one test or might require the association of the interpretation of signs and symptoms or waiting for a follow-up period (for example, in cases of periods of latency/hatching). Inadequate reference standards may generate biased

estimates of index test accuracy. In addition, it is extremely important that the results of the tests, both the index test and the reference standard, are interpreted without the prior knowledge of the results of the test previously applied (index or reference), so as not to influence the results, as in more subjective tests, such as imaging. The order in which the tests are conducted can be randomized or known, provided that researchers do not know, in advance, the results of the index and standard tests[11]. The time between conducting both tests must be sufficiently short so that the severity of the condition does not change. If the period between the reference standard test and index test allows a change in the spectrum of the disease, the performance of the index test may be overestimated.

One way to check the adequacy of the items mentioned above is to critically assess the risk of bias in studies of diagnostic accuracy. The Quadas-2 tool can be used to analyze the risk of bias through four domains: selection of patients, index test, reference standard, and flow and time[12]. This tool gives a more transparent analysis of bias, including guiding questions per domain, adding reliability to the findings of the study so that it can be used with more security in clinical practice[12].

ESTIMATING AND INTERPRETING OUTCOMES IN STUDIES OF DIAGNOSTIC ACCURACY

In primary studies of diagnostic accuracy, the experimental test (index test) is compared to the existing diagnostic test (reference standard), known as the best test currently available to identify, with precision, the presence or absence of the condition of interest. The results of both tests are then compared to assess the performance of the index test concerning the reference standard based on its sensitivity, specificity, positive and negative predictive values, and positive and negative likelihood ratios (Figure 2).

The sensitivity of a clinical trial is the proportion of individuals with the condition of diagnostic interest that is correctly identified by the index test concerning the proportion of individuals with a positive diagnosis of the disease obtained using the reference standard (true positives + false negatives by the test index)[13,14]. Specificity is the proportion of individuals without the condition that is correctly identified by the test index, concerning the proportion of individuals with a negative diagnosis of the disease obtained using the reference standard (true negatives + false positive by the test index)[13,14].

FIGURE 2. CALCULATION OF SENSITIVITY, SPECIFICITY, POSITIVE PREDICTIVE VALUE AND NEGATIVE PREDICTIVE VALUE, POSITIVE LIKELIHOOD RATIO AND NEGATIVE LIKELIHOOD RATIO OF A CLINICAL EXAMINATION FROM A CONTINGENCY TABLE 2 × 2.

		Condition	
		Present	Absent
Test	Positive	True positive (a)	False positives (b)
	Negative	False negatives (c)	True negatives (d)

Sensitivity = a/ (a+c)
Specificity = d/ (b+c)
Positive predictive value = a/ (a+b)
Negative predictive value = d/ c+d)
Positive likelihood ratio: a/ (a+c) / b (b+d) or (Sensitivity/ 1- Specificity)
Negative likelihood ratio: c/ (a+c) / d (b+d) or (1 - Sensitivity/Specificity)

The positive predictive value (PPV) of a test is defined as the probability of an individual having the disease, given that they had a positive result in the index test (true positives + false positive). The negative predictive value (NPV) of a test is defined as the probability of an individual not having the disease, given that they had a negative result in index test (true negative + false negative)[13].

Although the predictive values are more valuable to clinicians, since they provide a direct assessment of the practical usefulness of the test, they are influenced by the prevalence of the condition in the population to whom the test is applied[13,14]. Sensitivity and specificity, on the other hand, are not affected by the prevalence of the condition, but need to be interpreted in light of the population studied in the study, particularly regarding the spectrum of the disease, presenting limitations to the applicability of the test. For example, when assessing a particular condition in a general asymptomatic population (screening) or in a symptomatic one: a study that evaluated the sensitivity and specificity of the clinical test of breasts in a population of 752,081 women[15] identified a sensitivity of 85.2% and a specificity of 72.9% in symptomatic women *versus* 36.1% and 96.2%, respectively, in asymptomatic women.

The positive and negative likelihood ratios are the best indicators of the usefulness of a clinical trial. These proportions compare the probability of obtaining a positive or negative test result if the individual really has or does not have the condition, respectively. So, the greater the likelihood ratio of a positive test, the more certain it is that a positive test result indicates that the individual has the condi-

tion. A value of 10 or more is considered an indicator that a positive test result is very good at identifying a condition. In the same way, the lower the likelihood ratio of a negative test, the more certain it is that a negative test result indicates that the individual does not have the condition. A value of 0.1 or lower is considered an indicator that a negative test result is very good to dismiss the condition. If the likelihood ratio is close to 1.0, the test result is not a good indicator that the subject has (for a positive test result) or does not have (for a negative test result) the condition of interest. Since the calculation of these ratios uses the four parameters previously described and is not influenced by the prevalence of the condition, they are considered more valuable for clinical decision making[14].

MAKING INFERENCES ABOUT THE IMPACT AND CONSEQUENCES OF A DIAGNOSTIC TEST

Knowing how to interpret studies of diagnostic accuracy is an essential step, but that is not enough for recommending a test for clinical practice. As previously discussed, the preference should be for randomized clinical trials that evaluate the diagnostic strategies in outcomes that are relevant for the patient.

In the absence of those in the literature, the search must be for studies of diagnostic accuracy, preferably through systematic reviews and meta-analyses of performance estimates. In this sense, the synthesis of evidence in diagnostic tests is particularly challenging because the statistical methods used to aggregate data of diagnostic accuracy are conceptually complex (binomial and hierarchical models), often leading to difficulties in the interpretation of the results[16].

Additionally, evidence from studies of diagnostic accuracy is indirect. The clinician will need to make inferences about the probable impact and consequences of the test in outcomes that are important to the patient[17]. This situation also becomes a challenge, since many times there is no direct link between the results of diagnostic accuracy and a relevant outcome, especially when the test is part of an algorithm.

Generally, the inference requires the availability of an effective treatment[7]. However, even without an effective treatment, accurate testing can be beneficial to improve the well-being of patients with prognostic information, to reduce their anxiety, or even prevent them from being subjected to more unnecessary testing.

There are clinical situations in which it is more important to exclude a certain disease that has serious consequences if not identified. In these cases, the use of screening tests with high sensitivity and high negative predictive value may be of more value than the actual diagnosis of the disease, which becomes less important. This is the case, for example, with natriuretic peptides in congestive heart failure, given the consequences of the disease and the challenge of confirming it in patients with suspicion of failure, which occurs in only 40%-50% of cases[18].

When the treatment for a disease has high associated risks, in conditions of a difficult diagnosis, or when higher diagnostic precision is needed, multiple tests are often used. Thus, clinicians may be interested in additional benefits of a new adding-on test. An example of that is the use of computed tomography angiography tests additional to computed tomography without contrast in acute ischemic strokes[19].

Therefore, it is necessary to have a clear understanding of where this new test will be inserted along the diagnostic process, to achieve a balance between the desirable and undesirable consequences of the test. The inferences about the probable impact and consequences of the test in outcomes that are important to the patient may be carried out by simulating data in a hypothetical scenario of 1,000 patients, for example, using the prevalence data of the condition of interest or the pre-test probability of the patient having the disease based on signs and symptoms[20]. It is a simplified approach that classifies the test results to produce true positives and true negatives, false positives and false negatives cases, as in the following example.

Consider the consequences of replacing invasive angiography by multi-faceted spiral computed tomography (MCT) for the diagnosis of coronary artery disease (CAD). Suppose that the clinician wants to consider replacing it in patients in their clinical practice, which has 30% of patients with CAD.

The estimates of diagnostic accuracy of the computed tomography compared to angiography for CAD were obtained by identifying the meta-analysis of Hamon et al.[21], which included data from 27 studies (2,024 patients) corresponding to a cumulative number of 22,798 segments evaluated. This study

evidenced the summary estimates of 0.96 sensitivity (CI 95% 0.94-0.98), 0.74 specificity (0.65 to 0.84), 5.36 positive likelihood ratio (3.45 to 8.33), and 0.05 negative likelihood ratio (0.03 to 0.09).

Considering the context of service, it would be expected that the conventional coronary angiography would diagnosis 300 cases of CAD for every1,000 patients treated and 700 patients would be diagnosed with the absence of CAD using angiography. Based on the estimates from the meta-analysis, a MCT sensitivity of 96% represents the correct diagnosis of 288 out of 300 patients (true positives). In the same way, a specificity of 0.74 would represent the identification of 518 out of 700 non-cases of CAD (true negatives). Consequently, we would have 12 false negative and 182 false positive results (Figure 3).

FIGURE 3. DISTRIBUTION OF CASES OF TRUE POSITIVES, TRUE NEGATIVES, FALSE POSITIVES, AND FALSE NEGATIVES IN THE HYPOTHETICAL SCENARIO OF 1,000 PATIENTS AND SERVICE POPULATION WITH CAD PREVALENCE OF 30%. CAD: CORONARY ARTERY DISEASE; MCT: MULTIFACETED COMPUTED TOMOGRAPHY

		CAD (angiography)		
		Present	Absent	Total
MCT	Present	288	182	470
	Absent	12	518	530
	TotaL	300	700	1000

Table 1, below, shows the approach of the GRADE group (Gradings of Recommendations, Assessment, Development and Evaluation) to assess the consequences of the diagnostic strategies adopted in the above example. In this table, each possible outcome of the tests assumes a clinical/practical consequence. Thus, for the true positive cases, the patients would receive the treatments of known effectiveness (drugs, angioplasty, and stents, revascularization surgery) as clinical conduct, and the true negative cases would be spared the treatments and unnecessary use of resources. On the other hand, the false positive cases (26% of patients who would have a negative diagnosis using angiography) would be subjected to the unnecessary risks of the adverse events associated with the treatments (use of medication, segmental angioplasty, etc.), in addition to more anxiety and unnecessary loss of quality of life, while the false-negative cases would not receive adequate treatment that could help reduce the risk of subsequent coronary events, representing a loss of diagnosis in 4.0% of the patients with CAD.

Thus, it is relatively certain that minimizing false positives and false negatives will benefit patients. However, the complications of invasive angiography - cardiac catheterization, myocardial infarction, death -, although rare, are undoubtedly important.

TABLE 1. EXAMPLE OF THE CONSEQUENCES FOR THE PATIENT OF BEING CLASSIFIED ACCORDING TO THE PERFORMANCE OF THE TEST. SOURCE: HSU ET AL.[4]

Research question 1: What are the clinical implications of replacing invasive angiography with multifaceted computed tomography in the diagnosis of coronary artery disease?	
Population	Patients with suspected coronary artery disease (CAD)
Intervention	Multifaceted computed tomography (MCT)
Compared to	Conventional coronary angiography
Outcomes	
True positive	The patient will be submitted to the MCT, and the result will be positive; the patient will receive the treatments of known effectiveness (drugs, angioplasty, and stents, revascularization surgery) as the clinical conduct; there will be savings in expenses with the test
True negative	The patient will be submitted to the MCT, and the result will be negative; they will not have to go through other tests or unnecessary treatments, and there will be savings in expenses with the test
False positive	Anxiety, time and unnecessary expenses for the patient and family, unnecessary confirmatory tests; patients may be subjected to unnecessary risks of adverse events associated with treatments and loss of quality of life
False negative	The patient may have serious consequences from the disease, such as acute myocardial infarction, which may result in death; the true cause of the symptoms (i.e., CAD) will not be detected, leading to investigations and unnecessary treatments; an impediment to receiving the treatment
Inconclusive results	Time and resources spent; patients subjected to unnecessary risks, likewise the consequence of false negative and false positive
Tests complications	Both tests require the use of contrast in the patient, which can cause allergies, for example. Angioplasty is an invasive procedure, with the risk for cardiac catheterization
Use of resources (costs)	Angiography costs are higher than those of MCT, and it demands great resource utilization

CAD: coronary artery disease; MCT: multifaceted computed tomography.

Therefore, experts need to consider the desirable and undesirable consequences of diagnostic tests for patients, for making rational decisions about which test to recommend in clinical practice. The link between accuracy of results and clinical outcomes depends on this weighting, as well as the cognitive or emotional impact resulting from the knowledge of the diagnosis[22].

Other aspects should be part of the rational use of diagnostic tests, some of which have been already explored throughout the article, such as: the quality of the scientific evidence of studies available, the impact of the imprecision of the findings between studies, if the populations included in the studies correspond to the population that is the focus of the clinical decision making, the costs associated with the strategies and values and preferences of the patient[5]. Based on the example used, MCT could reduce the chance of adverse events of invasive angiography, which includes serious events, although rare, in addition to being less costly than angiography. On the other hand, the large number of false positives and the risk of losing patients with coronary artery disease treated effectively could lead to the patient's preference for the angiographic approach, even though it is more invasive.

CONCLUSIONS

Diagnostic tests are used to identify a condition of interest with the intention of providing adequate treatment and can be used as a diagnostic screening, replacing a diagnostic test, or be added to an existing test.

The appropriate decision regarding the adoption or recommendation of a diagnostic test should be based on evidence from randomized clinical trials, in which the diagnostic strategies are compared using measurements of clinical outcomes that are relevant to the patients.

However, such literature is scarce, and most of the commonly found evidence refers to estimates of the diagnostic performance of tests, measured using studies of diagnostic accuracy. Cross-sectional studies involving patients suspected of having the condition, without previous knowledge of the diagnosis status, provide the evidence of higher methodological quality when compared to diagnosis case-control designs.

Considering the accuracy parameters that are measured by this type of study, the positive and negative likelihood ratios are the best indicators of the usefulness of a clinical trial, because they can estimate a post-test probability of disease.

Clinical decision-making must, therefore, use indirect evidence from studies of diagnostic accuracy to make inferences regarding the impact and consequences of important results for the patient. The connection between accuracy outcomes, such true positives and false positives, and clinical results depends on the balance between benefits and damages of the tests available, as well as on the cognitive or emotional outcomes resulting from the knowledge of the diagnosis.

In addition, it is crucial that the clinician has knowledge in epidemiology and evidence-based health care, to properly evaluate the quality of evidence available, identify similar characteristics of the populations included in the studies in comparison with the population of the clinical practice and apply the estimates for the rational use of diagnostic tests.

Finally, considerations on resources should also be explored, as well as the preferences and values of the patient, who should participate in clinical decision making whenever possible.

RESUMO

OBJETIVO: *Auxiliar os clínicos na interpretação adequada das evidências científicas de estudos que avaliam testes diagnósticos, de modo a permitir seu uso racional na prática clínica.*

MÉTODOS: *Revisão narrativa da literatura dos principais conceitos, desenhos de estudo, interpretação adequada dos dados de acurácia diagnóstica e realização de inferências sobre o impacto do teste diagnóstico na prática clínica.*

RESULTADOS: *A maioria da literatura que avalia o desempenho de testes diagnósticos utiliza como delineamento os estudos transversais. Ensaios clínicos randomizados, avaliando desfechos clínicos, que seriam considerados ideais, são escassos. Os estudos transversais mensuram desfechos de acurácia diagnóstica que são considerados indiretos e insuficientes para definir o real benefício para os pacientes. Dentre os desfechos, as razões de verossimilhança positiva e negativa são as mais úteis para a decisão da conduta clínica. Variações no delineamento transversal do estudo, que podem acrescentar vieses aos resultados, bem como outros domínios que contribuem para diminuir a confiabilidade dos achados, são discutidos, além de como extrapolar tais achados de acurácia em impacto e*

consequências consideradas importantes para o paciente. Aspectos sobre custos, tempo para a obtenção do resultado, preferências e valores dos pacientes devem, preferencialmente, participar da tomada de decisão.

CONCLUSÃO: *Conhecer a metodologia dos estudos de acurácia diagnóstica é fundamental, porém não suficiente, para o uso racional de testes diagnósticos. Há a necessidade de se ponderarem as consequências desejáveis e indesejáveis dos resultados dos testes para os pacientes, de modo a favorecer a tomada de decisão racional acerca de qual teste recomendar na prática clínica.*

PALAVRAS-CHAVE: *Tomada de decisão clínica. Testes diagnósticos de rotina. Prática clínica baseada em evidências. Sensibilidade e especificidade. Valor preditivo dos testes. Equipamentos para diagnóstico.*

REFERENCES

1. White S, Schultz T, Enuameh YAK. Synthesizing evidence of diagnostic accuracy. Philadelphia: Lippincott Williams and Williams; 2011.
2. Bouza E, Torres MV, Radice C, Cercenado E, Diego R, Carrillo-Sánchez C, et al. Direct E-test (AB Biodisk) of respiratory samples improves antimicrobial use in ventilator-associated pneumonia. Clin Infect Dis. 2007;44(3):382-7.
3. Bossuyt PM, Irwig L, Craig J, Glasziou P. Comparative accuracy: assessing new tests against existing diagnostic pathways. BMJ. 2006;332(7549):1089-92.
4. Hsu J, Brozek JL, Terracciano L, Kreis J, Compalati E, Stein AT, et al. Application of GRADE: making evidence-based recommendations about diagnostic tests in clinical practice guidelines. Implement Sci. 2011;6:62.
5. Ferrante di Ruffano L, Hyde CJ, McCaffery KJ, Bossuyt PM, Deeks JJ. Assessing the value of diagnostic tests: a framework for designing and evaluating trials. BMJ. 2012;344:e686.
6. Hofmann B, Welch HG. New diagnostic tests: more harm than good. BMJ. 2017;358:j3314.
7. Bossuyt PM, Lijmer JG, Mol BW. Randomized comparisons of medical tests: sometimes invalid, not always efficient. Lancet. 2000;356(9244):1844-7.
8. Theron G, Zijenah L, Chanda D, Clowes P, Rachow A, Lesosky M, et al. Feasibility, accuracy, and clinical effect of point-of-care Xpert MTB/RIF testing for tuberculosis in primary-care settings in Africa: a multicentre, randomised, controlled trial. Lancet. 2014;383(9915):424-35.
9. Rutjes AW, Reitsma JB, Di Nisio M, Smidt N, van Rijn JC, Bossuyt PM. Evidence of bias and variation in diagnostic accuracy studies. CMAJ. 2006;174(4):469-76.
10. Rutjes AW, Reitsma JB, Vandenbroucke JP, Glas AS, Bossuyt PM. Case-control and two-gate designs in diagnostic accuracy studies. Clin Chem. 2005;51(8):1335-41.
11. Whiting P, Rutjes AW, Reitsma JB, Bossuyt PM, Kleijnen J. The development of QUADAS: a tool for the quality assessment of studies of diagnostic accuracy included in systematic reviews. BMC Med Res Methodol. 2003;3:25.
12. Whiting PF, Rutjes AW, Westwood ME, Mallett S, Deeks JJ, Reitsma JB, et al; QUADAS-2 Group. QUADAS-2: a revised tool for the quality assessment of diagnostic accuracy studies. Ann Intern Med. 2011;155(8):529-36.
13. Altman DG. Practical statistics for medical research. London: Chapman & Hall; 1991.
14. Davidson M. The interpretation of diagnostic test: a primer for physiotherapists. Aust J Physiother. 2002;48(3):227-32.
15. Bobo JK, Lee NC, Thames SF. Findings from 752,081 clinical breast examinations reported to a national screening program from 1995 through 1998. J Natl Cancer Inst. 2000;92(12):971-6.
16. Leeflang MM, Deeks JJ, Gatsonis C, Bossuyt PM; Cochrane Diagnostic Test Accuracy Working Group. Systematic reviews of diagnostic test accuracy. Ann Intern Med. 2008;149(12):889-97.
17. Lord SJ, Irwig L, Simes RJ. When is measuring sensitivity and specificity sufficient to evaluate a diagnostic test, and when do we need randomized trials? Ann Intern Med. 2006;144(11):850-5.
18. Roberts E, Ludman AJ, Dworzynski K, Al-Mohammad A, Cowie MR, McMurray JJ, et al. The diagnostic accuracy of the natriuretic peptides in heart failure: systematic review and diagnostic meta-analysis in the acute care setting. BMJ. 2015;350:h910.
19. Sabarudin A, Subramaniam C, Sun Z. Cerebral CT angiography and CT perfusion in acute stroke detection: a systematic review of diagnostic value. Quant Imaging Med Surg. 2014;4(4):282-90.
20. Fletcher RH, Fletcher SW, Fletcher GS. Diagnostics. In: Clinical epidemiology: the essentials. 5th ed. Baltimore: Wolters Kluwer; 2012.
21. Hamon M, Biondi-Zoccai GG, Malagutti P, Agostoni P, Morello R, Valgimigli M, et al. Diagnostic performance of multislice spiral computed tomography of coronary arteries as compared with conventional invasive coronary angiography: a meta-analysis. J Am Coll Cardiol. 2006;48(9):1896-910.
22. Segal JB. Choosing the important outcomes for a systematic review of a medical test. J Gen Intern Med. 2012;27(Suppl 1):S20-7.

REVIEW ARTICLE

Systemic dissemination of glioblastoma: literature review

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SUMMARY

INTRODUCTION: *Glioblastoma (GBM) is the most frequent primary malignant tumor from the central nervous system in adults. However, the presence of systemic metastasis is an extremely rare event. The objective of this study was to review the literature, evaluating the possible biological mechanisms related to the occurrence of systemic metastasis in patients diagnosed with GBM.*

RESULTS: *The mechanisms that may be related to GBM systemic dissemination are the blood-brain barrier breach, often seen in GBM cases, by the tumor itself or by surgical procedures, gaining access to blood and lymphatic vessels, associated with the acquisition of mesenchymal features of invasiveness, resistance to the immune mechanisms of defense and hostile environment through quiescence.*

CONCLUSIONS: *Tumor cells must overcome many obstacles until the development of systemic metastasis. The physiologic mechanisms are not completely clear. Although not fully understood, the pathophysiological understanding of the mechanisms that may be associated with the systemic spread is salutary for a global understanding of the disease. In addition, this knowledge may be used as a basis for a therapy to be performed in patients diagnosed with GBM distant metastasis.*

KEYWORDS: *glioblastoma, systemic metastasis, distant metastasis, extracranial, extraneural.*

INTRODUCTION

Glioblastoma multiforme (GBM) is the most common malignant tumor of the central nervous system (CNS) in adults, corresponding to 14.9% of primary tumors and 46.6% of primary malignant tumors of the central nervous system[1]. Data from The Central Brain Tumor Registry of the United States (CBTrus) have proven it to be more prevalent in Caucasian men and its incidence increases with age, being more common between the sixth and seventh decades of life. There is an estimate of 12,390 new cases in the year 2017; of these, 11,740 occur in people over 40 years old[1].

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In Brazil, in 2016, there was an estimate of 5,440 new cases of CNS cancer in men and 4,830 women. Disregarding non-melanoma skin tumors, CNS cancer in men is the 11th most frequent in the Southeast region. For women, it is the sixth most frequent in the Southern region. There are no specific histology data of the CNS tumor subtype in Brazil available[2].

The GBM is a tumor of high local aggressiveness, typically infiltrative and of rapid evolution, characteristically relapsing, resulting in limited prognosis. The survival of these patients progressed with the development of therapeutic protocols, especially the Stupp protocol in 2005, which added temozolomide (TMZ) to radiotherapy (RT) in the postoperative context and subsequent TMZ for six cycles, raising the overall survival (OS) in 2 years to 26.5%, compared with only 10.4% of exclusive postoperative RT[3]. It was also evidenced the benefit of survival even for the age range of 60-70 years, and the methylation of the promoter of O-6-methylguanine-DNA methyltransferase (MGMT) was established as the main predictor of the benefit of the TMZ association[4]. Recently, the association of tumor-treating fields (TTF) and TMZ maintenance, after the surgical protocol, RT + adjuvant TMZ, has been suggested as a new standard of treatment after an update of the phase III study, showing a gain of OS for this group[5]. Tumor-treating fields are the administration of alternating electric fields of low and medium intensity (200 kHz) via four electrodes placed on the hairless scalp and connected by cables to a portable generator. It must be used for 18 hours a day or more, and the electrodes must be changed twice per week[6]. The electrical fields generated hinder the formation of microtubules from the mitotic spindle on the metaphase, preventing the formation of the equatorial band, inducing cell vacuolization and failure of the mitosis in anaphase. In the telophase, they trigger dielectrophoretic movements of polar molecules and organelles during cytokinesis. Thus, it works as a mitotic inhibitor by selectively disrupting cell division, paralyzing mitosis and inducing apoptosis[6]. In view of all these recent therapeutic advances, still the overall survival (OS) in 2 years is 27%, and in 5 years, from 5% to 10%[6].

The systemic dissemination of GBM is considered a rare event, occurring in only 0.2 to 0.4% of cases[7], and its most common sites are bones, lymph node chains, liver, and lungs[8-10]. The first description of an event was made by Davis, in 1928, who named the disease Spongioblastoma[11].

An increase in reports of this type of case has been observed, particularly starting at the year 1994[12], perhaps due to the evolution of diagnostic methods with computed tomography and high-resolution magnetic resonance imaging, as well as PET-CT. Perhaps also due to the gain of survival using combined treatments, which may have increased the likelihood of dissemination and clinical manifestation of previously occult metastases.

Pietschmann et al., in their systematic review, assessed articles published from 1928 to 2013 and found 109 eligible studies, with a total of 150 patients diagnosed with GBM systemic metastasis[12]. From the data of the survey carried out for this study, other 14 cases reports were found from 2014 to 2017, totaling 17 patients[8,13-24]. The authors demonstrated that the average time from the initial diagnosis to the metastasis was nine months; the average survival after the metastasis was six months[12].

The present study aimed to perform a descriptive and analytical review of the literature to assess the possible biological mechanisms related to the occurrence of distant metastasis in patients with a diagnosis of GBM.

RESULTS

The exact reasons for the low occurrence of distant metastasis in patients with a GBM diagnosis are unknown.

Some authors speculate that patients with GBM, due to their low survival, do not have enough time for dissemination, neither to the neuraxis nor systemic. Furthermore, in some of the cases described in the literature, the systemic metastasis was not clinically detectable prior to death. Currently, autopsy is an uncommon procedure in cancer deaths, so the post-mortem diagnosis is unlikely[25].

THE GLYMPHATIC SYSTEM

Until recently, it was believed that one of the factors hindering the exit of GBM cells from the CNS was the absence of a local lymphatic system. This concept was disproved after two independent laboratories, first the Kipnis lab[26] and then the Alitalo lab[27], demonstrated the existence of lymphatic vessels in the meninges, baptized of glymphatic system. Such vessels were described as a network of narrow vessels that run along the blood vessels of the superior

and transverse, sagittal sinus. This lymphatic network becomes denser at the base of the brain and exits the skull along with the cranial nerves. These lymph vessels likely drain into deep cervical lymph nodes (dCLN), since the injection of dye into the cerebrospinal fluid was detected in the light of the lymphatic vessels marked by receivers Lyve-1 and VEGFR3, and in the dCLN, and not in the blood vessels. The ligation of lymphatic afferent to the dCLN abolished the drainage of dye for these nodules and made the meningeal vessels engorged[28]. In fact, there have been reports of cases with cervical nodal metastases in the absence of recurrence in the surgical scar or even without any pre-existing surgical procedure[29-31]. GBM rarely invades the meninges, and it is possible that surgical procedures facilitate the access of tumor cells to the meningeal vessels[32].

The presence of the blood-brain barrier (BBB)

The blood capillaries of the SNC differ from the capillaries of the rest of the body because they are not just a layer of endothelial cells loosely connected to each other, they are strongly connected among themselves by tight junctions, surrounded by a dense layer of basal membrane, pericytes and extensions of astrocytes, forming a highly selective microfilter, which only allows the passage of very small molecules. This barrier would make it impossible for cells to pass through due to their size. However, one of the main characteristics of high-grade gliomas as GBM is the breaking of the BBB, routinely described in imaging examinations of these patients. Thus, perhaps this is not the main obstacle to the formation of metastases[33].

It is understood that the tumor cells have access to blood circulation through the breaking of the BBB, but, as theorized by Liu et al.[34] in the theory of circulating stem cell (SCS), the tumor cells need specific skills to spread. As occurs in other malignant tumors, there is a cellular subtype in the tumor mass representative of less than 5% of the tumor population, which has preserved its potential for renewal,

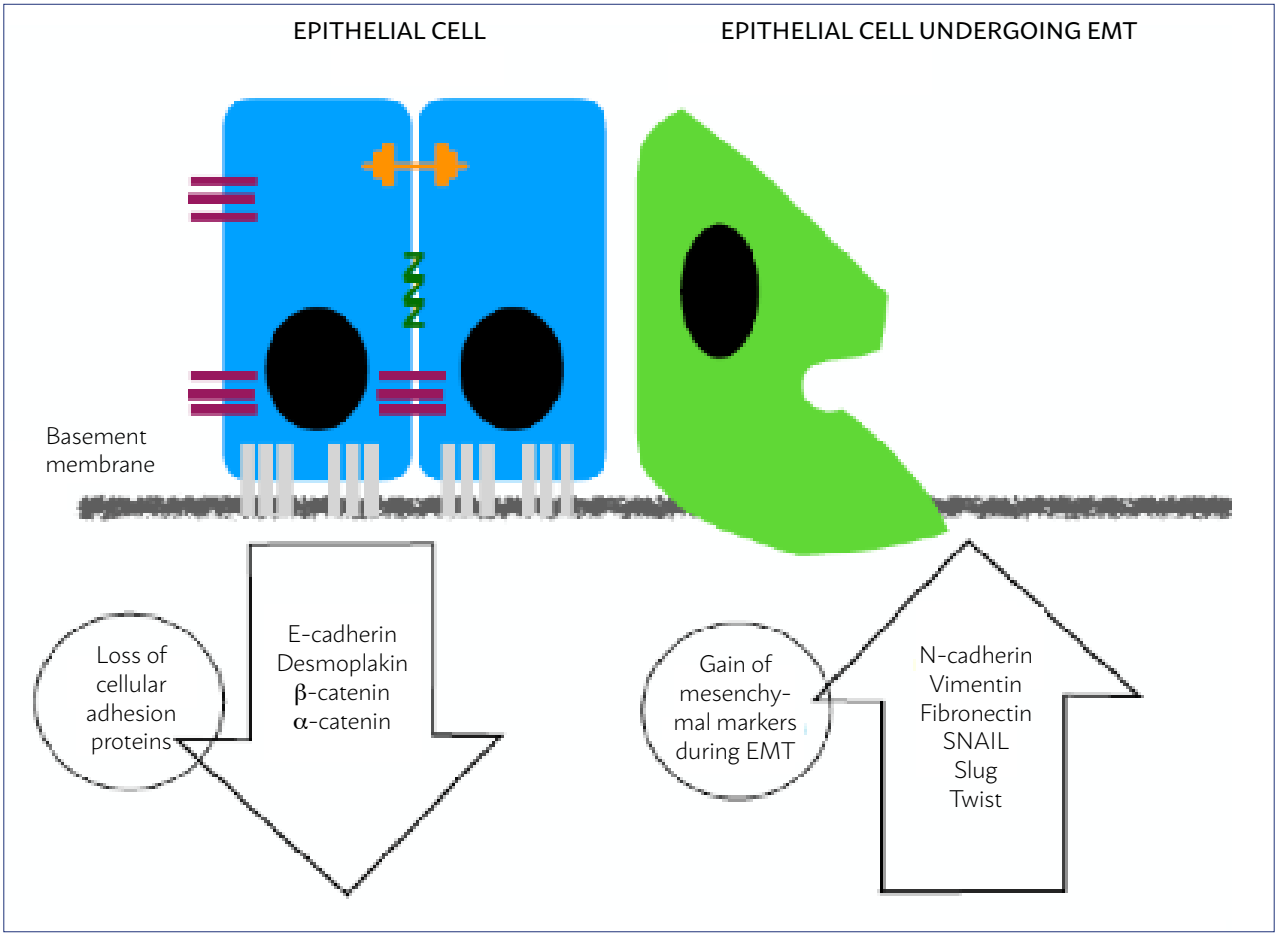

FIGURE 1: Changes in molecular markers during EMT. Loss of contact proteins: E-cadherin, Desmoplakin, β and α Catenins. Gain of mesenchymal markers: N-cadherin, Vimentin, Fibronectin, SNAIL, Slug, and Twist. Adapted from Serrano-Gomez et al. Molecular Cancer (2018) 15:18 (39).

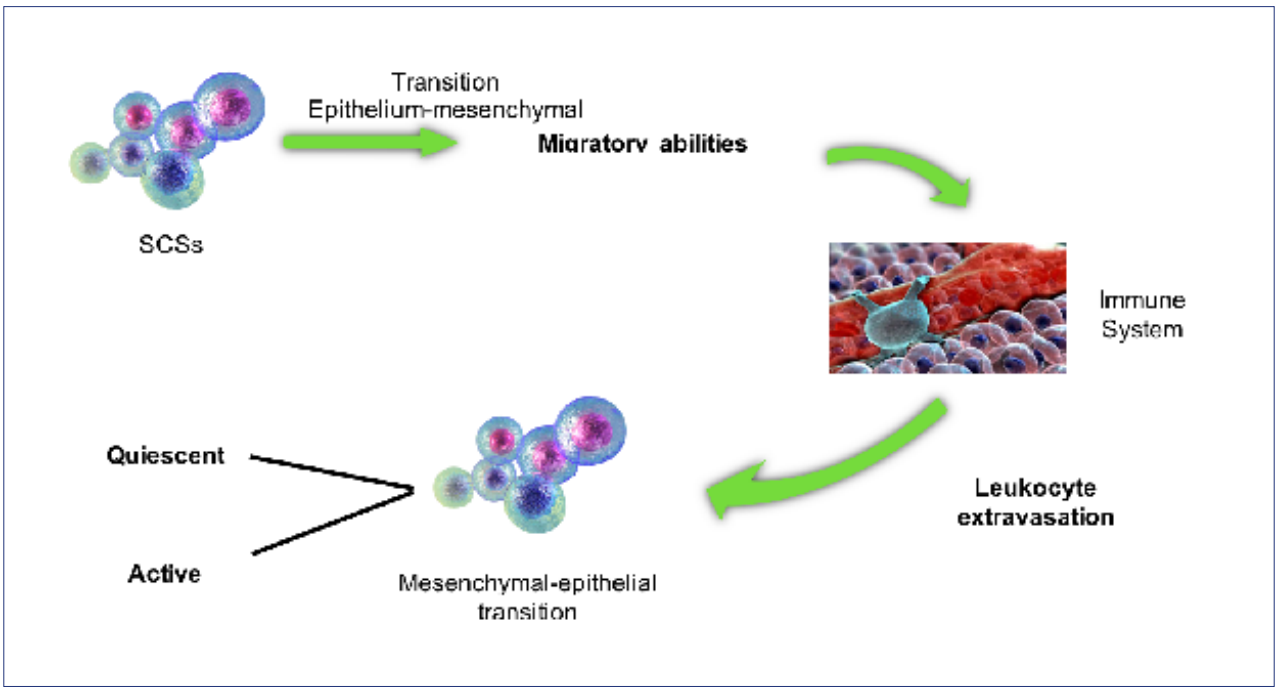

FIGURE 2. Scheme of the mesenchymal-epithelial transition.

is highly tolerant to hostile environments, to the defenses of the host and to the suppression of growth factors. They are the only cells capable of generating new tumors, the so-called cancer stem cells. Liu et al. hypothesized that these cells suffer a process of epithelial-mesenchymal transformation (EMT), acquiring skills for migration, such as the production of metalloproteinases and proteases enzymes capable of degrading the basal capillary membrane, allowing the breaking of the BBB[34]. Some of the EMT markers are: loss of epithelial markers and acquisition of mesenchymal cells, rupture of the intercellular adhesion, change of the apicobasal polarity and remodeling of the cytoskeleton. These changes are associated with the increase of cell migration and resistance to anoikis[33] (Figure 1).

Once in the bloodstream, the SCSs are subject to selection by shear stress and the immune system. It is argued that the SCSs can add to blood components such as platelets, "hiding" from the immune system and avoiding anoikis (programmed cell death by detachment from the extracellular matrix). The SCS exits the bloodstream through the capillaries of host tissue by the process of leukocyte extravasation. In the new environment, they go through a process reverse to the initial one, dependent on the absence of EMT inducer signs, the mesenchymal-epithelial transition (EMT), reassuming the initial epithelial phenotype, which may remain quiescent or multiply actively[35] (Figure 2).

Thus, EMT and SCSs are closely related in the process of tumor progression, especially in the initial stages of metastasis, which involves the invasion of surrounding stroma and the hematogenous dissemination to organs such as the liver, lungs, and bones[36,37].

The emergence of new technologies for detecting SCSs has led to the concept of liquid biopsy, which is the study of SCSs or even pieces of DNA from tumor cells in peripheral blood. The liquid biopsy can be used to detect cancer at an early stage, outline a treatment plan and assess its success or detect molecular changes and, consequently, resistance to tumor therapy[38].

An example of the EMT/EMT process is the sarcomatous metaplasia. It is the acquisition of a sarcomatous phenotype by neoplastic glial cells, allowing the acquisition of extracellular matrix of proteins necessary for vascular invasion. Gliosarcomas are more likely to invade the connective tissue, including the meninges. This transformation can be an undesired consequence of the GBM treatment, particularly RT and chemotherapy with alkylating agents, due to its potential to induce mutations on glial cells[39].

Another way of breaking the BBB is through surgery. According to Houston et al., about 96% of extraneural metastases occur after a neurosurgical procedure[31]. A craniotomy gives access to vessels of the cerebral parenchyma, meninges, and scalp. Ventriculoperitoneal shunts were also associated with

dissemination, not only for the reason mentioned above but also by allowing the migration of cells through its lumen[24]. In a case report with a review of the literature published by Lewis et al., approximately 20 cases were found of GBM with recurrence in the surgical scar or adjacent soft tissues until 2015[8]. On this publication and so far it is not possible to say what is the exact mechanism for such an occurrence, or if the surgical technique plays some role in it, but it is possible that tumor cells with molecular skills to survive outside the CNS are sown.

Suppression of the extracranial growth by the immune system

The prerequisite for the formation of metastases is that tumor cells reach the bloodstream so that they can be "distributed" to other organs. This also applies to GBM. Muller et al. published a study that demonstrated the presence of circulating cancer cells originating from the GBM in the peripheral blood of patients. The cells were detected using an immunocytochemical method, through the analysis of glial fibrillary acidic protein (GFAP), and its origin was confirmed by genomic tests (CGC and FISH). In treatment-naive patients, peripheral blood samples were collected before, during and after the surgical procedure. As a control, 23 healthy patients and five patients with cerebral metastases were also tested. In total, 141 patients with GBM were tested; 29 were positive for GFAP (20.6%). Only one patient in the control group tested positive for GFAP, demonstrating the high specificity of the test. Thirteen patients tested positive before the surgical procedure. This shows that the GBM cell can spread regardless of surgery. The mean follow-up time was 13.1 months, and no metastasis was detected[40].

If the GBM cells have proved capable of overcoming resistance mechanisms to exit the CNS and, as mentioned in the previous paragraph, reaching the bloodstream, why are more metastasis not detected? Perhaps due to the absence of GBM chemoattractives in peripheral tissues? Or, more likely, due to the action of the immune system (IS)?[25].

To discuss the relationship between cancer and the IS refers to the concept of immunosurveillance. Immunosurveillance was first hypothesized by Macfarlane Burnet and Lewis Thomas at the end of the 1950s, by stating that the immune system protects against the development of tumors by means of a patrolling of the body, eradicating cancer cells as soon as they appear[41].

Evidence points to the existence of immunosurveillance: 1) Immunosuppressed individual, such as transplant patients on prolonged use of immunosuppressants, have a risk one hundred times greater of developing neoplasms[42] and the more intense the suppression, the greater the risk[43]. There are case reports of GBM metastasis that developed in transplanted organs, but not in the donor[44-51]. 2) Cancer patients developing an immune response to the tumor that expresses molecularly identified antigens[52]. 3) Presence of tumor infiltrating lymphocytes (TIL) correlates with better survival in patients with melanoma, ovarian and colon cancer[53-56]. The paraneoplastic syndrome of neurological degeneration also demonstrates a spontaneous antitumor immunity[57].

If the IS is able to destroy early tumor cells, why immunocompetent patients develop cancer? To answer this question, a second concept is necessary: immunoediting.

These are acquired phenotypic characteristics common to almost all of the lineages of cancer cells, which allow them to circumvent barriers to growth and development: 1) autonomous growth; 2) angiogenic capacity; 3) unlimited replication; 4) resistance to apoptosis; 5) irresponsiveness to the growth inhibiting signs and 6) ability to metastasize[58].

However, none of these skills allows for escaping immunosurveillance. It was proposed then that tumors that arise in immunocompetent patients would have reduced immunogenicity if compared to those of immunodeficient patients. This was tested in a study using an animal model, and the results suggest that tumors that arise in the presence of normal lymphocytes are selected (edited) for their ability to withstand the antitumor lymphocyte response[59]. The IS can eradicate early tumors, but also promotes the development of immunoinvasive clones that support an immunologically intact medium, and this is the seventh phenotypic prerequisite for tumor development.

In gliomas, immunoediting is divided into three stages: 1) immunosurveillance or elimination, previously described; 2) balance, and 3) escape. The balance stage is a period of latency in the development of cancer, in which cells that were not eliminated during immunosurveillance remain in dynamic interaction with the IS in the tumor microenvironment. The evidence for the existence of this stage was demonstrat-

ed in a study published in 2007[60]. The CNS is considered immunologically distinct from the rest of the body due to the presence of the BBB. Many studies have attempted to clarify the mechanisms of anti-glioma immunity. Friese et al. described the transforming growth factor (TGF-β as a key molecule implicated in the reduced immune function of patients with gliomas responsible for the reduced expression of NKG2D in CD8+ T lymphocytes and NK cells through the serum and CSF *in vitro*[61].

As a result of this stage, the tumor can be eradicated or persist in this state of balance and non-progression, or even surpass the immune pressure and become clinically manifest, moving on to the escape stage. The escape stage represents the failure of the antitumor immunity, which is no longer able to restrict the growth of the tumor. The main escape mechanisms are intrinsic tumor changes, in which the tumor cell is more difficult to recognized or dead; and extrinsic changes, which diminish the effective capacity of the IS[41].

Inability of invading the extracranial connective tissue

As previously discussed, the SCSs disseminated to other anatomical sites have two options: to multiply actively forming a metastasis or become quiescent. The non-proliferative reversible state in which the cell remains "trapped" in their cell cycle in G0/G1 is called cellular quiescence. Intrinsic and extrinsic conditions of the microenvironment influence this process in a permissive or restrictive way[62]. Quiescent cancer cells are usually resistant to chemotherapy[63].

Confirmation of the existence of tumor quiescence was evidenced recently for gliomas. Endaya et al. failed to identify a fraction of quiescent cancer cells in a rat model with GBM. Ten years later, Naumov Ng et al. described that some cell lineages, including GBM, could not induce tumors *in vivo* for a long period[64,65].

Almog et al. hypothesized that quiescent tumors suffer a stable genetic reprogramming when they pass to the rapid growth phenotype and performed a wide transcriptional genomic analysis in search of the underlying molecular mechanism of these alternating states for various tumor histologies, including GBM. The comparative analysis between the quiescent tumor and rapid growth phenotypes proved that some specific genes are hyper-expressed in all the lineages of sleeper cells studied (breast carcinoma, GBM, osteosarcoma, and liposarcoma). They are: thrombospondin (inhibitor of angiogenesis), angiomotin (mediator of angiogenic activity of endogenous angiogenic inhibitor angiostatin), tropomyosin, transforming growth factor h2 (TGF-h2) and insulin-like growth factor binding protein 5 (IGFBP-5). The EphA5 expression, in particular, is significantly increased in quiescent cells of GBM. Therefore, the functional category most affected in the genetic signature of quiescent cells is the angiogenesis[66].

The ephrin tyrosine kinase family of receptors and its ligands are known for their participation in the embryonic and adult neurogenesis and angiogenesis. All the lineages of quiescent cells showed increased EphA5 RNA. In gliomas, the EphA5 expression revealed a correlation with the pathological degree. Their levels also lowered the more advanced the tumor stage was, having perhaps a suppressing role[66].

Treatment of glioblastoma with systemic metastasis

Due to the scarcity of cases, it is not possible to pinpoint what is the standard treatment for patients diagnosed with metastatic GBM. The drugs most often used are temozolomide and nitrosoureas. Bevacizumab (a recombinant humanized monoclonal antibody that blocks the action of vascular endothelial growth factor A-VEGF) has also been used as a monotherapy or in combination with chemotherapeutic agents[39].

In their meta-analysis of 88 cases of systemic metastatic GBM, Lun et al. found that patients who received multimodal treatment with local surgery and radiotherapy, systemic treatment with chemotherapy and *cerebrospinal shunt*, when necessary, were the ones that showed higher mean survival compared to less intense treatments[67].

Pietschmann et al. showed that only 40% of the cases evaluated had complete information about the treatment. The therapeutic approaches vary greatly, from exclusive palliative care to exclusive surgery, exclusive radiotherapy or chemotherapy alone, and various combinations of these methods. The authors also noted a greater predisposition for extra-CNS metastasis in young patients (average age of 42 years), often in good clinical conditions, suggesting a more aggressive treatment whenever possible, but remembering that there is no proven benefit of survival[12].

FINAL CONSIDERATIONS

There are several mechanisms to be overcome by tumor cells until a distant metastasis is formed. The first of these is that the tumor cells need to acquire the mesenchymal characteristics necessary to detach from their environment and erode the vascular basement membrane, without undergoing *anoikis*, overcoming the barriers of the CNS, among them the BBB. High-grade gliomas, such as GBM, characteristically break this barrier, and that is easily observed in imaging studies such as a MRI. Neurosurgical procedures also disrupt the BBB, which may facilitate this process. The cancer cells can then reach the circulation, either by hematologic or lymphatic pathways, becoming SCSs. However, once in the bloodstream, there is the action of a second filter, the immune system.

In immunocompetent patients, a large part of the SCSs ends up being detected and killed by Natural Killer cells (NK), while other few escape hiding from defense cells by binding to blood components such as platelets. To reach the connective tissue of the host organ, however, they can remain quiescent until the environmental conditions become favorable to its activation and proliferation. Immunocompromised patients, such as those who received organs or are in post-radiochemotherapy and develop systemic metastasis, indicate a clear relationship between the degree of competence of the immune system and such this type of event.

Although still not completely clear, the physiopathological understanding of the mechanisms associated with the systemic dissemination of GBM is important to the overall understanding of the disease. In addition, this knowledge can serve as a basis for selecting the therapy for patients with a diagnosis of GBM with distant metastasis.

RESUMO

INTRODUÇÃO: *Glioblastoma (GBM) é o tumor maligno mais comum do sistema nervoso central em adultos. Entretanto, metástase a distância de GBM é um evento extremamente raro. O presente estudo teve o objetivo de realizar uma revisão da literatura para avaliar os possíveis mecanismos biológicos relacionados com a ocorrência de metástase a distância de pacientes com diagnóstico de GBM.*

RESULTADOS: *Os mecanismos que podem estar relacionados com a capacidade de disseminação sistêmica do GBM são a quebra de barreira hematoencefálica (BHE) frequentemente vista em GBM, seja pela doença, seja por procedimentos cirúrgicos, dando acesso aos vasos sanguíneos e linfáticos, associada à aquisição de características mesenquimais de invasividade, resistência aos mecanismos de defesa do sistema imunológico e adaptação a hostilidades dos meios distantes por meio de quiescência.*

CONCLUSÕES: *As células tumorais necessitam vencer diversos obstáculos até a formação de uma metástase distante. Apesar de não totalmente esclarecido, o entendimento fisiopatológico dos mecanismos pelos quais podem estar associados à disseminação sistêmica do GBM é salutar para a compreensão global da doença. Além disso, esse conhecimento pode servir de base para a terapia a ser empregada diante do paciente com diagnóstico de GBM com metástase a distância.*

PALAVRAS-CHAVE: *Glioblastoma. Metástase sistêmica. Metástase a distância. Extraneural. Extracranial.*

REFERENCES

1. Ostrom QT, Gittleman H, Xu J, et al. CBTRUS Statistical Report: Primary Brain and Other Central Nervous System Tumors Diagnosed in the United States in 2009-2013. Neuro Oncol. 2016;18(suppl_5):v1-v75.
2. Estimativa 2018: Incidência de câncer no Brasil. [cited 2018 March 10]. Available from: http://www.inca.gov.br/estimativa/2018/estimativa-2018.pdf.
3. Stupp R, Mason WP, van den Bent MJ, et al. Radiotherapy plus concomitant and adjuvant temozolomide for glioblastoma. N Engl J Med. 2005;352(10):987-996.
4. Stupp R, Hegi ME, Mason WP, et al. Effects of radiotherapy with concomitant and adjuvant temozolomide versus radiotherapy alone on survival in glioblastoma in a randomized phase III study: 5-year analysis of the EORTC-NCIC trial. Lancet Oncol. 2009;10(5):459-466.
5. Stupp R, Taillibert S, Kanner AA, et al. Maintenance Therapy With Tumor-Treating Fields Plus Temozolomide vs. Temozolomide Alone for Glioblastoma: A Randomized Clinical Trial. JAMA. 2015;314(23):2535-2543.
6. Stupp R, Taillibert S, Kanner AA, et al. Effect of Tumor-Treating Fields Plus Maintenance Temozolomide vs. Maintenance Temozolomide Alone on Survival in Patients With Glioblastoma: A Randomized Clinical Trial. JAMA. 2017;318(23):2306-2316.
7. Beauchesne P. Extra-neural metastases of malignant gliomas: myth or reality? Cancers (Basel). 2011;3(1):461-477.
8. Lewis GD, Rivera AL, Tremont-Lukats IW, Ballester-Fuentes LY, Zhang YJ, Teh BS. GBM skin metastasis: a case report and review of the literature. CNS Oncol. 2017.
9. Senetta R, Cassoni P. Skin metastases of glioblastoma. In: Tumors of the Central Nervous System, Volume 2: Gliomas: Glioblastoma (Part 2). Hayat MA (Ed.). Springer, Dordrecht, The Netherlands, 143–149 (2011).
10. Laraqui L, Amarti A, Zouaidia F, Maher M, Kettani F, Saidi A. [Pulmonary metastasis from a glioblastoma. A case report]. Rev Pneumol Clin. 2001;57(3):225-228.

11. Davis L. Spongioblastoma Multiforme of the Brain. Ann Surg. 1928;87(1):8-14.

12. Pietschmann S, von Bueren AO, Kerber MJ, Baumert BG, Kortmann RD, Muller K. An individual patient data meta-analysis on characteristics, treatments and outcomes of glioblastoma/ gliosarcoma patients with metastases outside of the central nervous system. PLoS One. 2015;10(4):e0121592.

13. Kumar V, Singh D, Singh H, Saran RK. A case report of pontine glioblastoma presenting as subcutaneous metastasis in nape of neck in a child. J Pediatr Neurosci. 2015;10(4):386-388.

14. Johansen MD, Rochat P, Law I, Scheie D, Poulsen HS, Muhic A. Presentation of Two Cases with Early Extracranial Metastases from Glioblastoma and Review of the Literature. Case Rep Oncol Med. 2016;2016:8190950.

15. Xu M, Wang Y, Xu J, Yao Y, Yu WX, Zhong P. Extensive Therapies for Extraneural Metastases from Glioblastoma, as Confirmed with the OncoScan Assay. World Neurosurg. 2016;90:698 e697-698 e611.

16. Vandenbussche CJ, Ho CY, Nugent SL, Ali SZ. Extraneural metastases of primary central nervous system tumors identified by fine needle aspiration: a retrospective analysis. Acta Cytol. 2014;58(2):117-124.

17. Starnoni D, Yamgoue Y, Hottinger A, Bartanusz V. Multilevel severe radiculopathy from an extraneural glioblastoma cervical metastasis. Surg Neurol Int. 2016;7(Suppl 40):S1028-S1029.

18. Garcia JR, Corbella C, Baquero M, Bassa P, Soler M. Extracranial metastasis of multiforme glioblastoma detected by (11)C-methionine brain PET/CT. Rev Esp Med Nucl Imagen Mol. 2017;36(4):271-272.

19. Kup PG, Nieder C, Winnekendonk G, Adamietz IA, Fakhrian K. Extracranial oral cavity metastasis from glioblastoma multiforme: A case report. Mol Clin Oncol. 2016;5(4):437-439.

20. Anghileri E, Castiglione M, Nunziata R, et al. Extraneural metastases in glioblastoma patients: two cases with YKL-40-positive glioblastomas and a meta-analysis of the literature. Neurosurg Rev. 2016;39(1):37-45; discussion 45-36.

21. Khattab MH, Marciscano AE, Lo SS, et al. Antiangiogenic Therapies and Extracranial Metastasis in Glioblastoma: A Case Report and Review of the Literature. Case Rep Oncol Med. 2015;2015:431819.

22. Undabeitia J, Castle M, Arrazola M, Pendleton C, Ruiz I, Urculo E. [Multiple extraneural metastasis of glioblastoma multiforme]. An Sist Sanit Navar. 2015;38(1):157-161.

23. Forsyth TM, Bi WL, Abedalthagafi M, Dunn IF, Chiocca EA. Extracranial growth of glioblastoma multiforme. J Clin Neurosci. 2015;22(9):1521-1523.

24. Narayan A, Jallo G, Huisman TA. Extracranial, peritoneal seeding of primary malignant brain tumors through ventriculo-peritoneal shunts in children: Case report and review of the literature. Neuroradiol J. 2015;28(5):536-539.

25. Mourad PD, Farrell L, Stamps LD, Chicoine MR, Silbergeld DL. Why are systemic glioblastoma metastases rare? Systemic and cerebral growth of mouse glioblastoma. Surg Neurol. 2005;63(6):511-519; discussion 519.

26. Louveau A, Smirnov I, Keyes TJ, et al. Structural and functional features of central nervous system lymphatic vessels. Nature. 2015;523(7560):337-341.

27. Aspelund A, Antila S, Proulx ST, et al. A dural lymphatic vascular system that drains brain interstitial fluid and macromolecules. J Exp Med. 2015;212(7):991-999.

28. Dissing-Olesen L, Hong S, Stevens B. New Brain Lymphatic Vessels Drain Old Concepts. EBioMedicine. 2015;2(8):776-777.

29. Zhen L, Yufeng C, Zhenyu S, Lei X. Multiple extracranial metastases from secondary glioblastoma multiforme: a case report and review of the literature. J Neurooncol. 2010;97(3):451-457.

30. Hata N, Katsuta T, Inoue T, et al. [Extracranial metastasis of glioblastoma to the lung and heart with a histological resemblance to small cell carcinoma of the lung: an autopsy case]. No Shinkei Geka. 2001;29(5):433-438.

31. Houston SC, Crocker IR, Brat DJ, Olson JJ. Extraneural metastatic glioblastoma after interstitial brachytherapy. Int J Radiat Oncol Biol Phys. 2000;48(3):831-836.

32. Mondin V, Ferlito A, Devaney KO, Woolgar JA, Rinaldo A. A survey of metastatic central nervous system tumors to cervical lymph nodes. Eur Arch Otorhinolaryngol. 2010;267(11):1657-1666.

33. Savagner P. Epithelial-mesenchymal transitions: from cell plasticity to concept elasticity. Curr Top Dev Biol. 2015;112:273-300.

34. Liu J, Lin PC, Zhou BP. Inflammation fuels tumor progress and metastasis. Curr Pharm Des. 2015;21(21):3032-3040.

35. Lombard A, Goffart N, Rogister B. Glioblastoma Circulating Cells: Reality, Trap or Illusion? Stem Cells Int. 2015;2015:182985.

36. Nieto MA, Huang RY, Jackson RA, Thiery JP. Emt: 2016. Cell. 2016;166(1):21-45.

37. Thiery JP, Acloque H, Huang RY, Nieto MA. Epithelial-mesenchymal transitions in development and disease. Cell. 2009;139(5):871-890.

38. Lianidou ES. Gene expression profiling and DNA methylation analyses of CTCs. Mol Oncol. 2016;10(3):431-442.

39. Ray A, Manjila S, Hdeib AM, et al. Extracranial metastasis of gliobastoma: Three illustrative cases and current review of the molecular pathology and management strategies. Mol Clin Oncol. 2015;3(3):479-486.

40. Muller C, Holtschmidt J, Auer M, et al. Hematogenous dissemination of glioblastoma multiforme. Sci Transl Med. 2014;6(247):247ra101.

41. Dunn GP, Fecci PE, Curry WT. Cancer immunoediting in malignant glioma. Neurosurgery. 2012;71(2):201-222; discussion 222-203.

42. Penn I. Why do immunosuppressed patients develop cancer? Crit Rev Oncog. 1989;1(1):27-52.

43. Penn I. The changing pattern of posttransplant malignancies. Transplant Proc. 1991;23(1 Pt 2):1101-1103.

44. Nauen DW, Li QK. Cytological diagnosis of metastatic glioblastoma in the pleural effusion of a lung transplant patient. Diagn Cytopathol. 2014;42(7):619-623.

45. Armanios MY, Grossman SA, Yang SC, et al. Transmission of glioblastoma multiforme following bilateral lung transplantation from an affected donor: case study and review of the literature. Neuro Oncol. 2004;6(3):259-263.

46. Colquhoun SD, Robert ME, Shaked A, et al. Transmission of CNS malignancy by organ transplantation. Transplantation. 1994;57(6):970-974.

47. Frank S, Muller J, Bonk C, Haroske G, Schackert HK, Schackert G. Transmission of glioblastoma multiforme through liver transplantation. Lancet. 1998;352(9121):31.

48. Jonas S, Bechstein WO, Lemmens HP, Neuhaus R, Thalmann U, Neuhaus P. Liver graft-transmitted glioblastoma multiforme. A case report and experience with 13 multiorgan donors suffering from primary cerebral neoplasia. Transpl Int. 1996;9(4):426-429.

49. Morse JH, Turcotte JG, Merion RM, Campbell DA, Jr., Burtch GD, Lucey MR. Development of a malignant tumor in a liver transplant graft procured from a donor with a cerebral neoplasm. Transplantation. 1990;50(5):875-877.

50. Ruiz JC, Cotorruelo JG, Tudela V, et al. Transmission of glioblastoma multiforme to two kidney transplant recipients from the same donor in the absence of ventricular shunt. Transplantation. 1993;55(3):682-683.

51. Val-Bernal F, Ruiz JC, Cotorruelo JG, Arias M. Glioblastoma multiforme of donor origin after renal transplantation: report of a case. Hum Pathol. 1993;24(11):1256-1259.

52. Jager D, Stockert E, Gure AO, et al. Identification of a tissue-specific putative transcription factor in breast tissue by serological screening of a breast cancer library. Cancer Res. 2001;61(5):2055-2061.

53. Mihm MC, Jr., Clemente CG, Cascinelli N. Tumor infiltrating lymphocytes in lymph node melanoma metastases: a histopathologic prognostic indicator and an expression of local immune response. Lab Invest. 1996;74(1):43-47.

54. Clark WH, Jr., Elder DE, Guerry Dt, et al. Model predicting survival in stage I melanoma based on tumor progression. J Natl Cancer Inst. 1989;81(24):1893-1904.

55. Sato E, Olson SH, Ahn J, et al. Intraepithelial CD8+ tumor-infiltrating lymphocytes and a high CD8+/regulatory T cell ratio are associated with favorable prognosis in ovarian cancer. Proc Natl Acad Sci U S A. 2005;102(51):18538-18543.

56. Ohtani H. Focus on TILs: prognostic significance of tumor infiltrating lymphocytes in human colorectal cancer. Cancer Immun. 2007;7:4.

57. Dalmau J, Rosenfeld MR. Paraneoplastic syndromes of the CNS. Lancet Neurol. 2008;7(4):327-340.

58. Hanahan D, Weinberg RA. The hallmarks of cancer. Cell. 2000;100(1):57-70.

59. Mombaerts P, Iacomini J, Johnson RS, Herrup K, Tonegawa S, Papaioannou VE. RAG-1-deficient mice have no mature B and T lymphocytes. Cell. 1992;68(5):869-877.

60. Koebel CM, Vermi W, Swann JB, et al. Adaptive immunity maintains occult cancer in an equilibrium state. Nature. 2007;450(7171):903-907.

61. Friese MA, Wischhusen J, Wick W, et al. RNA interference targeting transforming growth factor-beta enhances NKG2D-mediated antiglioma im-

mune response, inhibits glioma cell migration and invasiveness, and abrogates tumorigenicity in vivo. Cancer Res. 2004;64(20):7596-7603.

62. Senft D, Ronai ZA. Immunogenic, cellular, and angiogenic drivers of tumor dormancy--a melanoma view. Pigment Cell Melanoma Res. 2016;29(1):27-42.

63. Adamski V, Hempelmann A, Fluh C, et al. Dormant glioblastoma cells acquire stem cell characteristics and are differentially affected by Temozolomide and AT101 treatment. Oncotarget. 2017;8(64):108064-108078.

64. Endaya BB, Lam PY, Meedeniya AC, Neuzil J. Transcriptional profiling of dividing tumor cells detects intratumor heterogeneity linked to cell proliferation in a brain tumor model. Mol Oncol. 2016;10(1):126-137.

65. Naumov GN, Bender E, Zurakowski D, et al. A model of human tumor dormancy: an angiogenic switch from the nonangiogenic phenotype. J Natl Cancer Inst. 2006;98(5):316-325.

66. Almog N, Ma L, Raychowdhury R, et al. Transcriptional switch of dormant tumors to fast-growing angiogenic phenotype. Cancer Res. 2009;69(3):836-844.

67. Lun M, Lok E, Gautam S, Wu E, Wong ET. The natural history of extracranial metastasis from glioblastoma multiforme. J Neurooncol. 2011;105(2):261-273.

REVIEW ARTICLE

Uremic neuropathy: an overview of the current literature

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SUMMARY

INTRODUCTION: *Peripheral neuropathy is a disorder that affects the cell body, axon or myelin of motor or peripheral sensory neurons and occurs in 60-100% of patients who are submitted to dialysis due to chronic kidney disease. Uremic neuropathy (UN) is attributed to the accumulation of organic waste, evident in patients with reduced glomerular filtration rate.*

OBJECTIVES: *This review aims to make clinical characteristics of uremic neuropathy evident enabling early diagnosis and treatment.*

METHODS: *This is a literature review of articles published on PubMed over the last 10 years using "Uremic Neuropathy" as "Title/Abstract".*

RESULTS: *A total of nine articles that met the inclusion criteria were included. UN is a distal symmetric sensorimotor polyneuropathy that occurs due to the accumulation of uremic toxins associated with an oxidative stress-related free radical activity. Hyperkalemia is thought to play an important role in its pathophysiology. Diagnosis depends on nerve conduction studies, and treatment includes dialysis or renal transplant.*

CONCLUSION: *Clinical presentations of UN are broad and non-specific; nonetheless, it is important to detect early changes in order to avoid its progression. The earlier UN is diagnosed and treated, the more successful are the clinical outcomes.*

KEYWORDS: *Neural conduction. Dialysis. Kidney Transplantation. Peripheral Nervous System Diseases. Uremia/complications.*

INTRODUCTION

Peripheral neuropathy (PN) is a disorder that affects the cell body, axon or myelin of motor or peripheral sensory neurons and can respectively be classified as neuropathological, axonal or demyelinating. This condition is either hereditary or acquired and may be further subdivided into sensory, motor or autonomic [1]. PN has a large spectrum of causes (such as nutritional deficiencies and toxic neuropathies [2] as well as clinical presentations [3]; however, constant and recurring pain occurs in almost all types of this disorder [4].

The overall prevalence of peripheral neuropathy is 2.4%. However, this number increases exponentially in certain age groups, and it may even be an under-

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estimate since traumatic causes are not included in this percentage [3].

Peripheral neuropathy occurs in 60-100% of patients who are submitted to dialysis due to chronic kidney disease (CKD) [5]. Uremic neuropathy (UN) occurs when renal dysfunction impairs filtration, leading to the accumulation of organic waste. This is evident in patients with reduced glomerular filtration rate (GFR) usually attributed to end-stage renal disease (ESRD) [6].

This review aims to make the clinical characteristics of uremic neuropathy evident enabling early diagnosis and treatment in order to prevent the effects of the advanced stages of this condition. The secondary purpose is to discuss the prognosis of uremic neuropathy based on data hinted in literature.

METHODS

This is a literature review of articles published on PubMed over the last 10 years using "Uremic Neuropathy" as "Title/Abstract". A total of 15 articles were found and 11 of them were available. Then, 9 articles were included as they met the inclusion criteria (Figure 1) – they were clinical studies and discussed uremic neuropathy.

RESULTS

Prevalence

In 1961, Martin and Tyler published the first report on uremic neuropathy in patients with hereditary intestinal nephritis with distal sensory-motor polyneuropathy [7,8]. Asbury et al, in 1963 used the term uremic polyneuropathy to describe distal sensorimotor changes due to uremic toxins. Uremic neuropathy is more frequent in males than in females [8] and is a common condition: studies have shown that it's prevalence varies from 50-100% in patients with chronic kidney disease [7,9-11]. This large range of values is due to the application of different criteria for the diagnosis of UN. The prevalence of UN in the pediatric population is unknown [10].

Pathology and pathophysiology of Uremic Neuropathy

UN is a distal symmetric sensorimotor polyneuropathy that typically affects lower limbs [7,12] and is due to length-dependent axonal degradation and secondary focal loss of myelin sheaths [7,8]. This is considered a demyelinating condition which leads to axonal degeneration and loss [8-10].

The accumulation of uremic toxins (the "middle toxins": guanidine compounds, parathyroid hormone, and myoinositol) [10,11] associated to oxidative stress-related free radical activity causes motor, sensory and autonomic nerve damage which leads to UN [8,13,14]. Although this exact mechanism remains unknown [10], there are hypotheses supporting the role of electrolytes in this process [8,9,12]. Hyperkalemia and hyperphosphatemia cause chronic uremic depolarization of nerves, contributing to the development of UN [8,12]. This occurs because potassium disrupts the normal ionic gradient and therefore activates calcium-mediated processes leading to axonal death [9].

UN is usually asymptomatic until renal function is under 15%, and glomerular filtration is lower than 10–12 ml/min, which usually happens 10-15 years after the onset of the underlying disease, such as diabetic neuropathy [7,8,10,11]. This type of neuropathy is one of the most frequent neurological manifestations of end-stage renal disease (ESRD) [11].

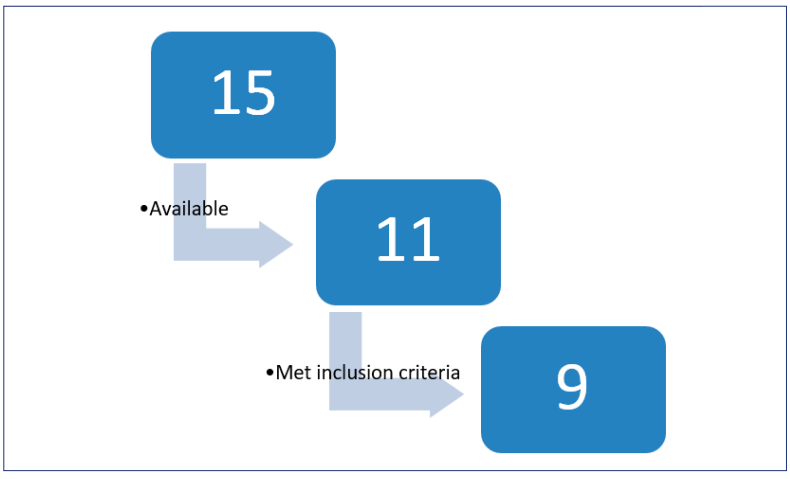


FIGURE 1. SELECTION PROCESS OF ARTICLES INCLUDED IN THIS REVIEW.

Both slowly and rapidly progressing sensorimotor axonal neuropathies are relatively common [10]. However, there have also been reports of fulminant motor neuropathies, which occurred under specific clinical conditions such as sepsis and severe CRF [8].

Hyperkalemia and hyperphosphatemia increase the risk of developing UN [9]. Other risk factors for UN are diabetes, advanced age and low creatinine and clearance [10].

A schematic representation of uremic neuropathy causes can be seen in Figure 2.

Symptoms of Uremic Neuropathy: Sensory and motor changes due to Uremic Neuropathy

Symptoms of UN vary, but it typically presents as a slowly progressing sensorimotor axonal neuropathy [9] which advances proximally, starting from the lower limbs and may spread to upper extremities [7]. Early symptoms are paresthesia, paradoxical heat sensation, restless leg syndrome, increased pain sensation, and cramps [7,9]. Long-term symptoms include weakness, impaired deep tendon reflexes, imbalance, numbness, and atrophy of the lower limbs [7-11].

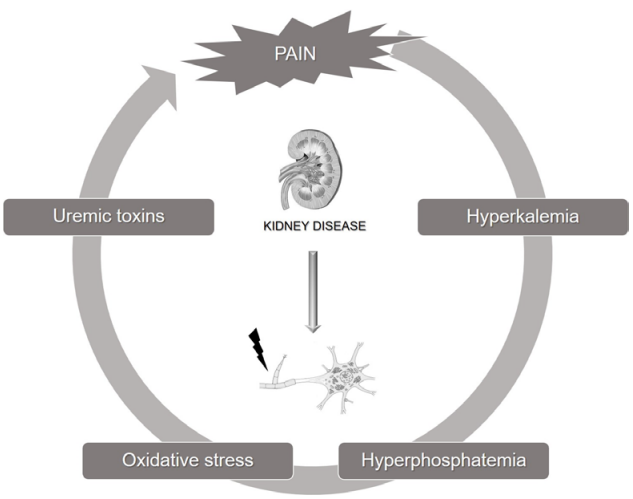
Nerve conduction and quantitation of sensory loss

Quantitation of sensory loss and nerve conduction is one of the main tools used to evaluate UN, as well as electromyography [11]. In patients suffering from this condition, nerve conduction velocity usually falls to 50-60% of normal values [8]; however, light touch and vibratory perception thresholds are more sensitive to evaluate either progression or recovery of UN than conduction velocity [8].

Some patients suffer thermal sensitivity impairment before they have sensory and motor damage. The number of functional axons in a nerve is evaluated according to changes in the amplitude of muscle response and sensory nerve action potentials [8]. The myelination of nerve fibers and their density is tested by velocity conduction. The most common morphological change in UN is the loss of large myelinated fibers, and positive neuropathic symptoms tend to correlate with quantitative results in conduction and sensory tests [8].

After nerve transplantation, there is an increase in nerve conduction due to remyelination [8]. Studies have shown that motor nerve conduction (MCV) is a significant predictor of mortality [13] in hemodialysis patients and achieves statistically significant values: (HR= 0.92; CI (0.86–0.99); $p < 0.05$) [15]. MCV correlates significantly with dialysis dose; however, further investigation is needed in order to confirm this hypothesis [13].

FIGURE 2. REPRESENTATIVE SCHEME OF UREMIC NEUROPATHY POSSIBLE CAUSES



Uremic Neuropathy in Children and Teenagers

The prevalence of UN in children is unknown [10], and this population usually does not present clinical evidence of UN; however, nerve conduction is altered, likewise in adults [8]. Authors showed that mean peroneal motor nerve conduction velocity (MNCV) was significantly decreased in children with mild renal failure (serum creatinine concentration, 1.5 to 2.9 mg/dL, normal range: 0.8–1.2 mg/dL), while ulnar MNCV was significantly decreased only when the serum creatinine value was at least 9 mg/dL. Within a year of renal transplantation, ulnar MNCV tends to return to normal values, and it takes 3 years for peroneal MNCV to go back to baseline values.

These parameters could potentially be used to evaluate the development of UN. However, they are only meaningful when renal function is very low or after a long period of time. Therefore, the periodic measurement of nerve conduction velocity is not useful to follow UN in children undergoing chronic hemodialysis [8].

Uremic Optic Neuropathy

Uremic optic neuropathy (UON) is a possible manifestation of UN that causes sudden vision deterioration and involves focal edema of the optic nerve

head [14]. Other related ophthalmic disorders include swelling of optic nerve heads, blurred margins of the optic disks seen using an ophthalmoscope. This disease should be taken into consideration as a possible diagnosis when patients with advanced chronic kidney failure present vision deterioration.

Seo et al. [14] described a patient suffering from UON who presented all the manifestations mentioned above, and the visual-evoked potential tests revealed reduced amplitude and increased latency in one of the eyes. This patient was treated with hemodialysis and corticosteroids, and his visual acuity and visual field improved, and the optic disk swelling was resolved.

The pathogenesis of UON is not well known. However, it has been shown to be related to the accumulation ofthat dialyzable toxin metabolites. Hemodialysis in combination to corticoids is the standard treatment for UON [14].

Diagnosis

The gold standard method to diagnose UN is a nerve conduction study [7,9]. Complementary methods include neurological assessment and biopsy of distal axons [12]. Since some UN symptoms are subjective and cannot be quantified using clinical tests, it might also be interesting to include psychological evaluation in order to investigate UN [15]. UN, however, may remain asymptomatic for a long time and only cause symptoms when severe damage has already been done. Therefore, an investigation should take place even in asymptomatic patients who present risk factors for UN [7].

Treatment of Uremic Neuropathy

Treatments that may reverse the effect of UN and improve nerve function are dialysis and renal transplantation [7,9,10]. Studies have shown a more significant reduction in the progression of UN due to an increase in dialysis dose, be it peritoneal or conventional dialysis [9]. It is controversial whether patients submitted to peritoneal dialysis have inferior results in the treatment of UN compared to those submitted to conventional hemodialysis [11]. Hemodiafiltration is another therapeutic option that also benefits motor nerve excitability.

Renal transplantation is the only definite treatment that interrupts the progression of UN and reverts symptoms. It is important to note that results are inversely proportional to the disease duration prior to the transplant: a shorter disease time leads to better post-transplant clinical outcomes [10]. Nonetheless, in patients who may not undergo this procedure, either due to clinical restrictions or personal denial, effective dialysis is definitely a good therapeutic option [11] as it also normalizes most nerve excitability parameters [8].

Comparison of uremic neuropathy with other types of neuropathies:

There are many different types of neuropathies other than Uremic, including Diabetic, Alcoholic, Chronic Inflammatory, and Infectious. Below is a comparative table with the symptoms and treatments of these main types of neuropathies.

TABLE 1. ASSOCIATION OF DIFFERENT TYPES OF NEUROPATHY AND ITS TREATMENTS

NEUROPATHY	MAIN SYMPTOMS	TREATMENT
Uremic Neuropathy	-Pain, numbness, and tingling in feet and legs; -Cramps, muscle twitches, or increased pain sensation in the feet and legs; -Muscle weakness [16]	-Dialysis -Kidney Transplant [16]
Diabetic Neuropathy	-Slowly and progressive primarily sensory deficit following a "stocking-glove distribution" [17]	-Treatment can range from lifestyle modification and strict glucose control to the use of immunosuppressant medications [17,18]
Alcoholic Neuropathy	-Painful paresthesia; -Muscle weakness; -Sensory and motor symptoms extend proximately into the arms and legs; -Gait impairment may be present [19,20]	- Thiamine treatment has been shown not to be effective; - Therapy ought to include cessation of alcohol ingestion, aiming at the toxic target(s) of alcohol [22]
Chronic Inflammatory Neuropathy	-Weakness is typically symmetric and characteristically involves proximal and distal muscles; -Sensory symptoms include numbness, tingling, gait imbalance, and, at times, painful paresthesias [21]	-Steroids, plasma exchange, and intravenous immunoglobulin may be used as 1st line treatment options [21]
Infectious neuropathy (HIV, HCV, CJ, LD[23])	-HIV, HCV, LD: pain, weakness, paresthesia and absent ankle reflexes -CJ: acute, ascending, motor neuropathy associated with Guillan-Barré syndrome (GBS) [23,24]	-HIV: analgesics, anticonvulsants [24] -HCV: steroids, antiviral therapy, rituximab, interferon alfa [25,26] -LD: doxycycline, amoxicillin, cefuroxime,ceftriaxone [27-29] -CJ: azithromycin, erythromycin, fluoroquinolones; treatment for GBS such as plasmapheresis, plasma exchange and intravenous immunoglobulin [30,31]

CONCLUSION

UN is a prevalent condition, affecting 60 to 100% of patients who suffer from chronic kidney disease, depending on the classification criteria used. The exact mechanism of the demyelinating process leading to axonal degeneration and loss is still uncertain. However, electrolytes, such as potassium, have shown to play an important role in the pathophysiology of UN. Clinical presentations of UN are broad and may be non-specific; nonetheless, it is essential to detect early changes in order to diagnose UN and avoid its progression. The earlier the signs and symptoms of UN are detected, the earlier UN is diagnosed using nerve conduction studies and treated with dialysis or renal transplant, leading to greater clinical success.

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RESUMO

INTRODUÇÃO: *A neuropatia periférica (NU) é um distúrbio que afeta o corpo celular, o axônio ou a mielina do motor ou neurônios sensoriais periféricos e ocorre em 60%-100% dos pacientes que são submetidos à diálise por doença renal crônica. A neuropatia urêmica é atribuída à acumulação de resíduos orgânicos, evidente em pacientes com taxa de filtração glomerular reduzida.*

OBJETIVO: *O objetivo desta revisão é fazer com que as características clínicas da neuropatia urêmica sejam evidenciadas, permitindo o diagnóstico e tratamento precoce.*

MÉTODO: *Esta é uma revisão da literatura de artigos publicados no PubMed nos últimos dez anos usando "Neuropatia Urêmica" como "Título/Resumo".*

RESULTADOS: *No total, foram incluídos nove artigos que atendem aos critérios de inclusão. A NU é uma polineuropatia sensório-motora simétrica distal que ocorre devido ao acúmulo de toxinas urêmicas associadas à atividade de radicais livres relacionados ao estresse oxidativo. A hipercalemia tem um papel importante na sua fisiopatologia. O diagnóstico depende de estudos de condução nervosa e o tratamento inclui diálise ou transplante renal.*

CONCLUSÃO: *As apresentações clínicas das NU são amplas e não específicas; no entanto, é importante detectar mudanças iniciais para evitar sua progressão. Quanto mais precoce for a detecção e tratamento da NU, melhor será o resultado clínico.*

PALAVRAS-CHAVE: *Condução nervosa. Diálise. Transplante de rim. Doenças do sistema nervoso periférico. Uremia/complicações.*

REFERENCES

1. Marchettini P, Lacerenza M, Mauri E, Marangoni C. Painful peripheral neuropathies. Curr Neuropharmacol. 2006;4(3):175-81.
2. Staff NP, Windebank AJ. Peripheral neuropathy due to vitamin deficiency, toxins, and medications. Continuum (Minneap Minn). 2014;20(5 Peripheral Nervous System Disorders):1293-306.
3. Misra UK, Kalita J, Nair PP. Diagnostic approach to peripheral neuropathy. Ann Indian Acad Neurol. 2008;11(2):89-97.
4. International Modulation Society. 2007. Neuropathy, neuropathic pain, and painful peripheral neuropathy: many kinds, causes, and treatments [cited 2017 March 12). Available from: http://www.neuromodulation.com/fact_sheet_painful_peripheral_neuropathy
5. Mambelli E, Barrella M, Facchini MG, Mancini E, Sicuso C, Bainotti S, et al. The prevalence of peripheral neuropathy in hemodialysis patients. Clin Nephrol. 2012;77(6):468-75.
6. Cooper JD, Lazarowitz VC, Arieff AI. Neurodiagnostic abnormalities in patients with acute renal failure. J Clin Invest. 1978;61(6):1448-55.
7. Ghazan-Shahi S, Koh TJ, Chan CT. Impact of nocturnal hemodialysis on peripheral uremic neuropathy. BMC Nephrol. 2015;16:134.
8. Said G. Uremic neuropathy. Handb Clin Neurol. 2013;115:607-12.
9. Witzel II, Jelinek HF, Khalaf K, Lee S, Khandoker AH, Alsafar H. Identifying common genetic risk factors of diabetic neuropathies. Front Endocrinol (Lausanne). 2015;6:88.
10. Kandil MR, Darwish ES, Khedr EM, Sabry MM, Abdulah MA. A community-based epidemiological study of peripheral neuropathies in Assiut, Egypt. Neurol Res. 2012;34(10):960-6.
11. Deger SM, Reis KA, Guz G, Bali M, Erten Y. A case of an accelerated uremic neuropathy. Renal Fail. 2011;33(3):371-2.
12. Sinha AD, Agarwal R. Chronic renal disease progression: treatment strategies and potassium intake. Semin Nephrol. 2013;33(3):290-9.
13. Stosovic M, Nikolic A, Stanojevic M, Simic-Ogrizovic S, Radovic M, Jovanovic D, et al. Nerve conduction studies and prediction of mortality in hemodialysis patients. Ren Fail. 2008;30(7):695-9.
14. Seo JW, Jeon DH, Kang Y, Lee DW, Lee HJ, Yoo WS, et al. A case of end-stage renal disease initially manifested with visual loss caused by uremic optic neuropathy. Hemodial Int. 2011;15(3):395-8.
15. Nowicki M, Zwiech R, Dryja P, Sobański W. Autonomic neuropathy in hemodialysis patients: questionnaires versus clinical tests. Clin Exp Nephrol. 2009;13(2):152-5.
16. University of Chicago. Types of peripheral neuropathy - systemic / metabolic. 2017. [cited 2017 April 20]. Available from: http://peripheralneuropathycenter.uchicago.edu/learnaboutpn/typesofpn/systemic/kidney.shtml.
17. Tracy JA, Dyck PJ. The spectrum of diabetic neuropathies. Phys Med Rehabil Clin N Am. 2008;19(1):1-26.
18. Schreiber AK, Nones CF, Reis RC, Chichorro JG, Cunha JM. Diabetic neuropathic pain: physiopathology and treatment. World J Diabetes. 2015;6(3):432-44.
19. Zeng L, Alongkronrusmee D, van Rijn RM. An integrated perspective on diabetic, alcoholic, and drug-induced neuropathy, etiology, and treatment in the US. J Pain Res. 2017;10:219-28.

20. Chopra K, Tiwari V. Alcoholic neuropathy: possible mechanisms and future treatment possibilities. Br J Clin Pharmacol. 2012;73(3):348-62.

21. Dimachkie MM, Barohn RJ. Chronic inflammatory demyelinating polyneuropathy. Curr Treat Options Neurol. 2013;15(3):350-66.

22. Mellion M, Gilchrist JM, de la Monte S. Alcohol-related peripheral neuropathy: nutritional, toxic, or both? Muscle Nerve. 2011;43(3):309-16.

23. Cashman CR, Höke A. Mechanisms of distal axonal degeneration in peripheral neuropathies. Neurosci Lett. 2015;596:33-50.

24. Smith HS. Treatment considerations in painful HIV-related neuropathy. Pain Physician. 2011;14(6):E505-24.

25. Prud'homme S, Nevens F, Casteels I. Bilateral simultaneous anterior ischemic optic neuropathy, an extrahepatic manifestation of hepatitis C cured with direct acting antivirals. GMS Ophthalmol Cases. 2016;6:Doc05.

26. Benstead TJ, Chalk CH, Parks NE. Treatment for peripheral neuropathy associated with hepatitis C virus infection. 2017. [cited 2017 April 20]. Available from: http://www.cochrane.org/CD010404/NEUROMUSC_treatment-for-peripheral-neuropathy-associated-with-hepatitis-c-virus-infection, 2014

27. Foundation for Peripheral Neuropathy. Lyme disease neuropathy. [cited 2017 April 20]. Available from: https://www.foundationforpn.org/what-is-peripheral-neuropathy/causes/inflammatory-infectious/lyme-disease/

28. Sigal LH, Williams S. A monoclonal antibody to *Borrelia burgdorferi flagellin* modifies neuroblastoma cell neuritogenesis in vitro: a possible role for autoimmunity in the neuropathy of Lyme disease. Infect Immun. 1997;65(5):1722-8.

29. Radzišauskienė D, Ambrozaitis A, Marciuškienė E. Delayed diagnosis of lyme neuroborreliosis presenting with abducens neuropathy without intrathecal synthesis of Borrelia antibodies. Medicina (Kaunas). 2013;49(2):89-94.

30. Nyati KK, Nyati R. Role of *Campylobacter jejuni* infection in the pathogenesis of Guillain-Barré syndrome: an update. Biomed Res Int. 2013;2013:852195.

31. Miljković-Selimović B, Lavrnić D, Morić O, Ng LK, Price L, Suturkova L, et al. Enteritis caused by *Campylobacter jejuni* followed by acute motor axonal neuropathy: a case report. J Med Case Rep. 2010;4:101.

REVIEW ARTICLE

Violence and sexually transmitted infections in pregnancy

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SUMMARY

OBJECTIVE: *To synthesize the knowledge produced in studies about the association between violence and STI during pregnancy.*

METHODS: *In this systematic review, we conducted basic activities of identification, compilation, and registration of the trials. The instruments of data collection were studies that investigated, explicitly, relationships between violence, gestation, and STI, from July 2012 to July 2017, using PubMed, Cochrane Library, SciELO, and LILACS.*

RESULTS: *In all, 26 articles were chosen to form the basis of the analysis of this study. The relationship between violence and STI was observed in 22 of the 26 studies, and in eight of them, the violence was practiced during the gestation period. In two studies, there was no evidence of this relationship. In one study, the lack of care for STI was attributed to the unpreparedness of health professionals. Mental disorders were cited as resulting from STI in three articles and in another as a result of violence. One study found more frequent violence against adolescents, while two others cited gestation as a protective factor.*

CONCLUSIONS: *IPV combines characteristics that have a different expression when the woman is in the gestational period. The literature points to a relationship between IPV against women and the presence of STI. The monitoring of pregnancy, whether in the prenatal or postpartum period, offers unique opportunities for the health professional to identify situations of violence and thus provide assistance.*

KEYWORDS: *Violence Against Women; Sexually Transmitted Infections; Pregnancy; Abortion; Stillbirth; Sex Offenses.*

INTRODUCTION

Studies have pointed to a relationship between intimate partner violence (IPV) and sexually transmitted infection (STI) in women[1,2], which makes it essential to detect this as part of the integrated healthcare, especially in the pregnancy-puerperal period, to protect the mother-child binomial. Identifying the presence of these diseases in the gestational period through clinical and serological diagnoses and correlating such findings with epidemiological data is fundamental so that, from this knowledge in our environment, we can establish early and timely preventive and/or therapeutic strategies or behaviors to avoid these infections which can lead to vertical transmission (VT).

The literature indicates not only the high prevalence of violence, as well as overlaps of several of its types in the gestational period[3]. Among them, there is sexual, psychological, and physical violence. Conjugal violence, practiced by an intimate partner, can combine characteristics of any of the above. Usually, it is used as a resource for interpersonal disputes, often complex, ambiguous, and with strong affective influence, in which local solutions go hand in hand with impunity[4].

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The preparation of professionals to deal with cases of violence against women presents gaps in the supply of these demands[5], and this contributes to the fact that the real magnitude of this social malaise remains unknown due to the high rate of underreporting[6]. However, in the course of prenatal care, delivery, and puerperium, there is a great opportunity to identify situations of violence and propose protective actions[7]. Therefore, studies that go deeper into the subject and bring information about violence, in its various forms, can contribute to training programs, not only to physicians but to all health professionals who work in the pregnant-puerperal period. During the months of follow-up, a trained professional will have the chance to establish an important bond of trust, which will help in the diagnosis of violent situations and open the way to act in the most appropriate form.

The high prevalence of congenital syphilis points to deficiencies in basic healthcare, especially prenatal care since the diagnostic test is part of those recommended by the Ministry of Health. Its prevention depends on measures that eliminate or reduce the risk of fetal infection, such as early diagnosis and appropriate treatment of the pregnant woman and her partner[8].

Also very prevalent, the *Human papillomavirus* (HPV), currently the most responsible for cervical cancer, appears as the most frequent viral STI. In a study of female adolescents between 10 and 14 years old, all of them sexually active, of the 22% who had some STI, 45% were caused by HPV [9]

HTLV infections are associated with several diseases such as adult T-cell leukemia/lymphoma (ATLL), myelopathy, tropical spastic paraparesis, uveitis, infectious dermatitis, rheumatic diseases, hairy cell leukemia, erythrodermatitis, bacterial infections coinfected with HIV, neurological diseases, and mycosis fungoides[10,11]. Transmission occurs through sexual intercourse, blood transfusions, injecting drugs, transplants, percutaneous exposure in health professionals, and through VT, especially through breastfeeding. The knowledge of the prevalence of infection in pregnant women and the factors related to a greater risk of VT in our country are of fundamental importance for prevention since there is no treatment and its consequences can be severe. There is no active or passive immunization, as well as specific antiviral therapy available against HTLV.

Therefore, the study of associations of situations of violence with STI in gestation provides subsidies for adopting preventive and protective measures, both for pregnant women and their newborns.

OBJECTIVE

To synthesize the knowledge produced by empirical studies in the health area on the association between violence and STI in gestation and subsidize new research and programs of prevention of IPV and vertical transmission of STI during pregnancy.

METHODS

In this systematic review, we chose four large databases namely: PubMed, SciELO, Cochrane, and LILACS.

We defined as inclusion criteria: articles that studied violence against pregnant women; articles that included the presence of STI during pregnancy; published in the last 5 years.

The search strategy used in PubMed, on 2018/01/12, was: ((“Violence” [Mesh]) AND “Pregnancy” [Mesh]) AND “Sexually Transmitted Diseases” [Mesh] Filters: 5 years. The following databases were searched: SciELO, Cochrane, and LILACS. In the SciELO, the descriptors Violence [All indexes] and Pregnancy [All indexes] and Sexually Transmitted Diseases [All indexes] were used. In Cochrane, it was used as search criteria: Violence and Pregnancy and Sexually Transmitted Diseases, Publication Year from 2012 to 2017. In LILACS we used the descriptors Violence [Words] and Pregnancy [Words] and Sexually Transmitted Diseases [Words].

RESULTS

Sixty-two studies were found in PubMed. In SciELO, no published works on these subjects were detected. In the LILACS database, of the four studies found, one was discarded by duplicity. Finally, in the Cochrane database, the survey found 28 bibliographic productions.

Ninety-three articles were selected in the screening of titles and abstracts, but six were excluded because they were protocols and consensuses.

After careful analysis of the 87 full texts, 10 were dismissed because they were literature reviews, 19 because they studied only one of the three themes chosen, and 32 because they focused on subjects that

were not related with the objectives present here.

Thus, 26 articles were chosen to form the basis for the analysis of this study, according to the schematic flowchart described in Figure 1 of Annex I.

The relationship between violence and STIs was observed in 22 of the 26 pieces of research, and in eight of them, violence was practiced during gestation. In two trials, there was no evidence of this relationship. In one study, the lack of care for STI was attributed to the unpreparedness of health professionals. Mental disorders were cited as resulting from STI in three articles and in another as a result of violence. One study found more frequent violence against adolescents, while two others cited gestation as a protective factor.

The description of the design of each study, its authors, sample, objective, and results are detailed in Annex II. Another 21 articles were added to enrich the introduction and discussion. With this, the article used 47 bibliographical references.

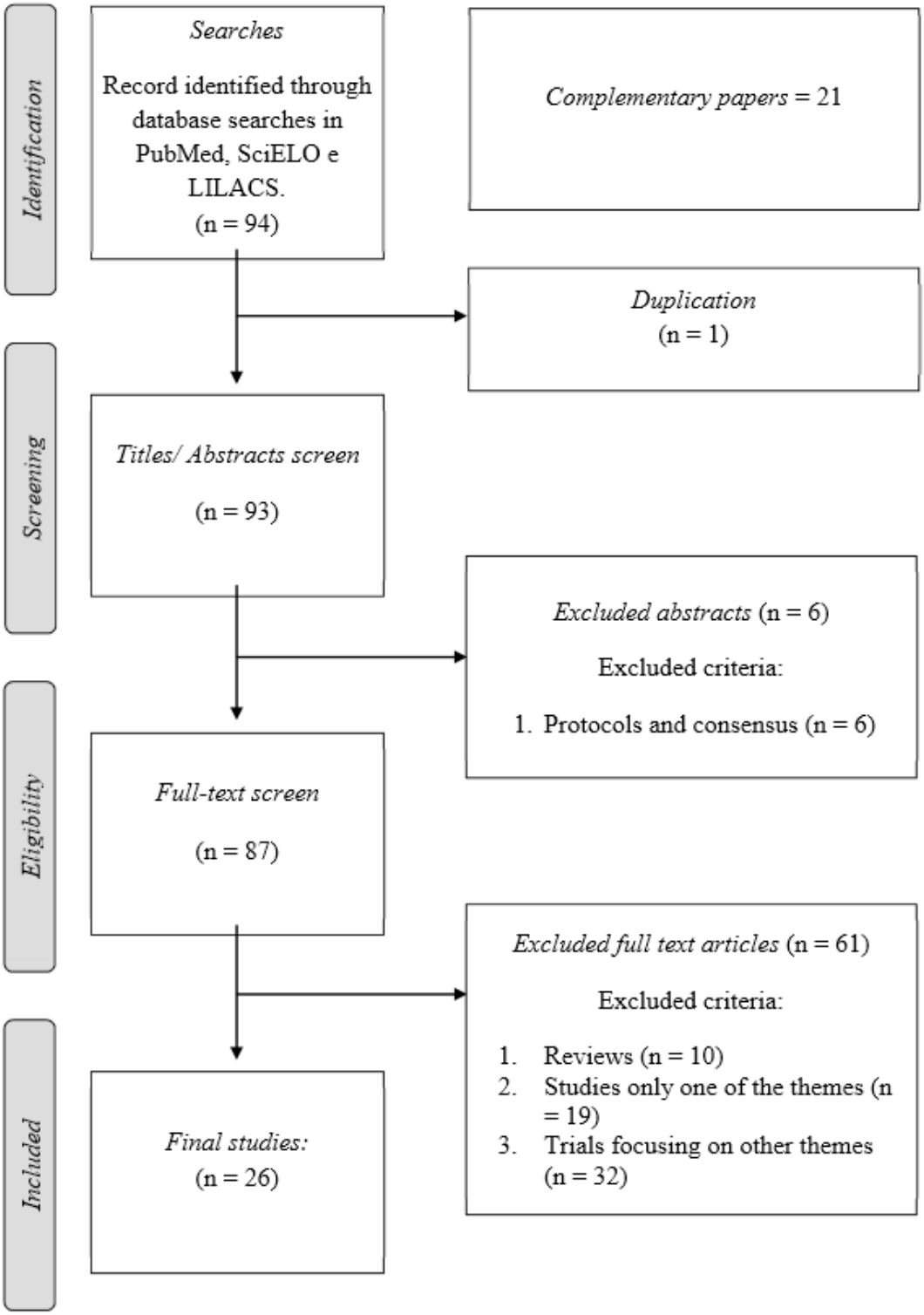
DISCUSSION

Violence in pregnancy may can the most varied responses. IPV, in addition to having physical consequences, indirectly worsens maternal health because pregnant women, in fear of violent reprisals from partners, may avoid prenatal care. Some even hide HIV prenatal testing from their partners because it can be perceived as a sign of infidelity. Equally regrettable is how some communities banalize IPV as a common and intractable issue[12]. Some people voluntarily abandon marital life as a strategy to escape violence. Others are expelled from their homes when they dare to exercise their autonomy, possibly due to their HIV status[13]. In some places, IPV during pregnancy can reach alarming levels, as in research conducted in Zimbabwe where the reports reached 63.1% of the interviewed, which is among the highest rates in the world[14]. Other factors may arise in this context, as in the interventional research of Rotheram-Borus et al., where they found that, over time, there is a significant relationship between alcohol, marital violence, and depression. However, simple measures such as home visits demonstrated efficacy in improving the mothers' emotional health, even when depression was not initially the objective [15]. Even so, Jahanfar et al. failed to establish the efficacy of interventions for domestic violence on outcomes of maternal and neonatal morbidity and mortality[16], although McDougal et al. have associated a history of miscarriage or stillbirth, not to physical violence but to sexual violence[17]. Therefore, high-quality work needs to be expanded to determine if intervention programs can prevent or reduce the deleterious effects of domestic violence on pregnant women and in pregnancy outcomes[16].

Although violent situations are present in all age groups, the greatest vulnerability among adolescents must be observed. In research about the factors that influenced reproductive and sexual health among women entering the adolescent sex industry, four themes were highlighted: early sexual abuse, early use of illicit drugs, continuous violence, and difficulties in accessing reproductive and sexual healthcare[18]. Recto and Champion highlighted the high risk of psychological distress in these adolescents involved in risk behaviors, victims of interpersonal violence, using illicit substances, or pregnant[19]. In a higher education institution in Colombia, students with a mean age of 20 years answered a self-administered questionnaire based on the Reproductive Health survey of the Pan American Health Organization. It was observed that had a history of pregnancy (12.3%), physical violence (21.6%), and sexual violence (4.6%), with a predominant silence of victims of sexual abuse (61.8%)[20]. Still among students, now in a high school located in a high-poverty region, about 20% of the girls surveyed between the ages of 14 and 17 reported a higher number of reproductive coercion than consensual intercourse. The most frequent reports were: "they said they did not use birth control," "took the condom out during sex to get pregnant," and "they said they would leave them if they were not pregnant." These adolescents had almost three times more chance to get chlamydia infection, had greater difficulty in recognizing abusive behavior and had worse communication with their sexual partners [21].

The association between violence, mainly sexual, unwanted pregnancy, and STI after forced and unprotected sex is well known[22]. However, the low representation of female adolescents in HIV/AIDS clinical trials may inhibit their access to future prevention techniques. In a formative in a community in Tanzania, situations of violence and recruitment difficulties for HIV testing among adolescents and young women were studied. Domestic violence related to the partner was present in the report of all

ANNEX I


Identification
Searches
Record identified through database searches in PubMed, SciELO e LILACS.
(n = 94)
Complementary papers = 21
Duplication
(n = 1)
Screening
Titles/ Abstracts screen
(n = 93)
Excluded abstracts (n = 6)
Excluded criteria:
1. Protocols and consensus (n = 6)
Eligibility
Full-text screen
(n = 87)
Excluded full text articles (n = 61)
Excluded criteria:
1. Reviews (n = 10)
2. Studies only one of the themes (n = 19)
3. Trials focusing on other themes (n = 32)
Included
Final studies:
(n = 26)

married participants. Many believed that beatings were normal. A third of the single women reported sexual abuse by relatives. Adolescents were the most frequent victims of domestic violence, often perpetrated by partners and relatives. It is, therefore, a public health priority to include adolescent abuse in HIV prevention [23].

Sexual violence can also be associated with humiliation, torture, and beatings during violation; and rape can be practiced by several men. Although Perry et al. have not established a relationship between chlamydia, gonorrhea, and miscarriage after rape [24], other studies point to the high prevalence and incidence of STI in women who suffer sexual crimes. They can often cause physical trauma, sexual disorders, unwanted pregnancy, psychological consequences, and even death. Thus, many of these women experience great uncertainty about their future and their children[25,26].

Already described in the literature, the relationships established between alcohol consumption and unprotected sexual intercourses indicate the incidence of HIV-positive women[27]. Some groups of pregnant women have the perception that IPV, an alcoholic partner, and lack of communication with the partner are forms of vulnerability that put them at risk of HIV[28]. A study conducted in Mombasa[27] with 400 HIV-negative women showed associations between alcohol-abuse disorders, the incidence of one year of unprotected sexual intercourse, HIV, and pregnancy. Abusive alcohol users had more unprotected sex and more partners than abstainers. Sexual intercourse under the influence of alcohol was frequent and associated with less frequent use of a condom. Compared to women who were sober, those who consumed excess alcohol experienced 4.1 times more sexual violence and 8.4 times more risk to have physical violence. It is, thus, perceived that unprotected sex, partner violence, and HIV incidence were higher in women with alcohol abuse disorders, a fact corroborated by Russell et al.[29].

In a study conducted in a hospital in Nigeria[30], the prevalence and patterns of intimate partner violence among HIV-positive and HIV-negative pregnant women receiving prenatal care were compared. In a total of 220 pregnant women, it was found that the cases did not differ significantly from the controls in relation to age, parity, tribe, religion, marital status, and monthly family income. Those interviewed with HIV positive experienced physical violence during pregnancy with rates six times higher than in controls, sexual violence about four times higher than in controls, and were 12 times more likely to be denied sex by their partner in comparison with the controls. The threat of being harmed, deprivation of financial support, and denial of communication were the most common forms of violence between sexual partners and HIV-positive pregnant women. In this study, HIV-positive status predisposes pregnant women to the increase in intimate partner violence but underlies the great impact of social rejection suffered as a result of HIV infection. It is worth remembering that, in addition to physical and psychological trauma in women, IPV also increases the chance of vertical transmission [31]. In another region of Africa, Charlotte Watts and Janet Seeley[32] address gender inequality and intimate partner violence as obstacles to a confrontation with an effective response to HIV. In this place, women and girls represent 57% of people living with HIV, and gender violence and inequality are determinants of vulnerability. Research shows qualitative data from women attending prenatal clinics in Johannesburg, which shows how the diagnosis of HIV during pregnancy and disclosure to the partner are relevant aspects of the occurrence of violence in the relationship. Low adherence to medications and the use of relief services are also described. Likewise, how the use of these services may be difficult since women feared that their partners would be alerted about their HIV status. Pregnant women also described that they could not refuse sex or negotiate condom use, prioritizing the physical safety of an unborn baby during pregnancy to the detriment of potential secondary transmission of HIV. Still in Africa, even in the absence of violence, several risk behaviors are pointed out during pregnancy, resulting from local customs and lack of information, such as the belief that antiretroviral drugs can cure HIV[33,34]. It is worth noting the efforts in South Africa in the fight against HIV, prevention of injuries and violence, in addition to initiatives for the promotion of maternal, neonatal, and child health[35]. Thus, there is still a long way to go.

National HIV prevention programs in Mexico focus on high-risk groups that do not include women in general. Although the epidemic is growing among them, their testing is restricted to prenatal screening. Even so, in a survey conducted in Mexico City, only 6% of the controls were tested for HIV

ANNEX II

TABLE 1 DETAILED DESCRIPTION OF THE ARTICLES USED IN THIS REVIEW.

Autor/ Year	Design	Sample	Objective	Results
Oza et al., 2015	cohort	25 ♀	Explore experiences during childhood and adolescence that influenced the sexual and reproductive health among women who entered in the sex industry.	Identified four main themes: early sexual abuse, early use of illicit drugs, ongoing violence and limited access to health care. The participants who reported these experiences face the risk of unintended pregnancy, miscarriage or stillbirth, and untreated STIs.ace
Recto & Champion, 2016	transversal	461 ♀	To study psychological disturbance and associated factors among adolescents.	Adolescents in risk behaviors, interpersonal violence, use of illicit substances and pregnant women are at risk of psychological distress.
Gómez-Camargo et al., 2014	transversal	934 ♀	To investigate the state of sexual and reproductive health among university students in Colombia.	12.3% had a history of pregnancy; physical violence (21.6%), sexual violence (4.6%, predominant silence of victims of sexual abuse (61.8%), history of STI (1.9%) and history of symptoms suggestive of STI (42%).
Northridge et al., 2017	transversal	149 ♀	To determine the prevalence of reproductive coercion and IPV, and risks to health.	Reproductive coercion is experienced by 1 in 5 girls of school age in a poor community and is associated with chlamydia and IPV infection.
Baumgartner et al., 2015	Clinical trial	138 ♀	Measure IPV and examine the associations between risk factors, sociodemographic factors, and HIV risk factors.	Married women reported IPV. Many believed that beatings were normal. One-third of the single women reported abuse by relatives. Teenagers have experienced high rates of domestic violence, and young people were more likely to report aggression.
Secor-Turner et al., 2013	cohort	241 ♀	Understand the interaction of multiple contexts of sexual risk behaviors of adolescents.	The latent construction of individual risk encompasses the use of illicit substances, the perpetration of violence, having suffered violence and witnessing situations of violence; the family breakdown included family disconnection, low-income family communication and perception of lack of safety at home.
Perry et al., 2015	transversal	19465 pregnant women	To estimate the prevalence of rape-related pregnancy as an indication for abortion and to describe the demographic and clinical correlations of these women.	Chlamydia and gonorrhea infection were no more prevalent among women who completed rape-related pregnancies than among those who ended up with other indications.
Zihindula & Maharaj, 2015	cohort	19 ♀	Explore the perceptions and experiences of the risk of sexual violence in the Democratic Republic of Congo.	Rape used to have extreme brutality against women, with long-term consequences, including unwanted pregnancies and/or HIV/AIDS. Many have lived with uncertainty about their future and that of their children.
Ceccon & Meneghel, 2015	transversal	161 ♀ HIV +	To determine the prevalence and forms of violence.	Psychological violence (72.7%), physical (54.6%) and sexual violence (25.4%).
Turan et al.,2016	cohort	614 pregnant women	Explore the forced migration of pregnant women in an area of high HIV prevalence as a strategy to escape from a violent partner.	Violent situations in pregnancy, such as voluntary leaving home to escape violence or being kicked out for exercising autonomy. In the quantitative analysis, pregnant women who migrated were more educated, less likely to live with their partners and with fewer children.
Shamu et al., 2013	transversal	2042 puerperal women	To describe the occurrence, dynamics, and predictors of IPV during pregnancy, including HIV links, in urban Zimbabwe.	63.1% of the interviewees reported physical, emotional and/or sexual IPV during pregnancy: 46.2% reported physical and/or sexual violence, 38.9% sexual violence, 15.9% physical violence and 10% reported severe violence during pregnancy. Physical violence was less common during pregnancy than in the last 12 months before pregnancy.
Hatcher et al., 2013	transversal	29 pregnant women	Explore the context and triggers of IPV in general and for pregnant women in rural Kenya.	Physical consequences and IPV induce pregnant women to avoid prenatal care and HIV testing because they fear violent reprisals from partners. The ♀ suffered physical, sexual and economic IPV, such as being kicked out of home or losing material support. They hide the HIV test from their partners because the test is seen as a sign of infidelity. The community considers IPV a common and untreatable issue, which seemed to normalize its use.
Teixeira et al., 2017	transversal	681 ♀	To analyze the factors associated with the occurrence of pregnancy after the diagnosis of HIV infection.	Violence related to the diagnosis of HIV/AIDS. Pregnancy after a diagnosis of HIV infection = 35.2%. Pregnancy after an HIV diagnosis is associated with a lower level of schooling.
Aguilar-Zapata et al., 2017	case-control	152 pregnant women	To study sociodemographic differences between recent pregnant women HIV positive and HIV negative.	Strong evidence of an association between HIV infection, STI history and history of violence. Only 6% of controls were tested for HIV during prenatal follow-up.

Autor/ Year	Design	Sample	Objective	Results
McDougal et al., 2013	cohort	582 ♀	To examine the prevalence of gestational losses among female professionals who inject drugs and measure their associations with physical and sexual violence.	30% of participants had at least one miscarriage/ stillbirth, 51% suffered sexual violence, and 49% had suffered physical violence. The history of miscarriage/ stillbirth was associated with sexual violence, but not with physical violence.
Narasimhan et al.,2016	transversal	945 ♀ HIV-positive	Determine the sexual and reproductive health priorities of women with HIV and apply their values and preferences to the construction of new guidelines.	89.0% feared or suffered gender violence; 56.7% had an unplanned pregnancy; 72.3% received advice on contraception; 58.8% had mental problems after they discovered their HIV status.
Ashaba et al., 2017	cohort	20 ♀	Explore the psychosocial challenges experienced by women living with HIV during pregnancy and postpartum.	Discrimination of health professionals and personal shame associated with pregnancy with HIV has led to the difficulty of care of HIV, especially when associated with structural barriers such as the lack of transport to the clinic. IPV and lack of support from partners and family members.
Maman et al.,2016	transversal	1092 ♀	Examine whether HIV increases the risk of IPV in the postpartum and the interaction in its disclosure.	HIV was not associated with physical IPV in the postpartum period. Statistically significant positive interaction between HIV and its disclosure.
Shamu et al., 2014	transversal	1951 puerperal women	Study the IPV after disclosure of HIV test results by pregnant women.	Factors associated with IPV were gender inequality, past of IPV, risky sexual behaviors and living with relatives. The IPV after the disclosure of HIV in pregnancy although lower is still high and strongly related to IPV before pregnancy.
Malaju & Alene, 2013	transversal	400 pregnant women	To study the reaction of the partners regarding the positive result for HIV in pregnant women.	78.50% expected a negative reaction from their partners over the positive HIV test. A positive reaction was associated to pregnant women with their own income, residing in urban areas, with a higher educational level, without a stigmatizing attitude with people living with HIV and having access to health care facilities.
Hampanda, 2016	transversal	320 ♀ HIV+ in the postpartum	To determine if there is a relationship between IPV and non-adherence to VT prevention.	Intimate partner violence is associated with non-adherence to VT prevention during and after pregnancy.
Peltzer et al., 2013	cohort	239 couples with pregnant women (n = 478 individuals)	Describe the high-risk sexual behavior among 239 couples during pregnancy, examine the relationship of sexual risk behavior to HIV serostatus and intimate partner violence.	31.8% of the pregnant women and 20.9% of the ♂ were HIV positive, with ± 2/3 knowing their serological status. 46.9% of the ♀ did not know the HIV status of their partner. 47.7% ♀ reported at least one unprotected sexual relation in the previous week. 17.6% of the ♂ and 10.0% of the ♀ indicated sexual intercourses with another person in the previous month. IPVs were associated with unprotected intercourse in the previous week.
Peltzer & Mlambo, 2013	transversal	1502 pregnant	To assess the sexual behavior of HIV risk and its associated factors among pregnant women in Mpumalanga, South Africa.	63% never used a condom with their primary sexual partner in the last 3 months, 60% did not know the partner's HIV status. Behaviors found: being HIV positive, experiencing partner violence and psychological distress due to the diagnosis of an STI other than HIV, unplanned pregnancy, belief that the medications cure HIV.
Chersich et al., 2014	cohort	400 ♀	Investigate alleged links between alcohol consumption, unprotected sex and HIV infection in sub-Saharan Africa.	Excessive alcohol led to more unprotected sex and a higher number of partners. Sex, when the woman was intoxicated, was frequent and associated with less frequent condom use. The occurrence of accidents with condoms has increased steadily with each increase in the AUDIT category. Associated violence and HIV incidence are higher in ♀ with alcohol use disorders.
Darak et al., 2014	cohort	460 ♀	Investigate women who, within the low-risk population, are potentially at higher risk of HIV infection.	19.4% of women perceived themselves to be at risk of HIV, and most of them reported some vulnerability in their relationship as couples, such as IPV, alcoholic partner and lack of communication with partners.
Rotheram-Borus et al., 2015	transversal	1238 pregnant women	To compare the standard care condition with the condition of home intervention in the use of alcohol, IPV and depression.	Alcohol, conjugal violence, and depression are significantly related over time. A home visit intervention improved the emotional health of low-income mothers, even when depression was not the initial target.
STI: Sexually Transmitted Infection; ♀: women; ♂: men; IPV: Intimate Partner Violence; AUDIT: Alcohol Use Disorders Identification Test.				

during prenatal care, which indicates a great fragility of the system[36].

In a survey with HIV-positive women, the majority (89.0%) feared or suffered gender-based violence, with more than half having an unplanned pregnancy and 58.8% experiencing psychiatric problems after discovering they had HIV[37]. Maman found a higher frequency of IPV among those who chose not to disclose to their partners their positive HIV status, perhaps because they had other problems in their relationships[38]. On the other hand, Shamu et al. found that IPV after disclosure of HIV positivity in pregnancy is also high, but lower and strongly related to IPV before pregnancy[39]. An exception was found by Malaju and Alene in an Ethiopian city called Gondar, they found positive reactions in partners who received results of an HIV positive test from their partners when they were financially independent and had good schooling[40].

Violence against women also has a high occurrence in Brazil and, in general, is the result of unequal relationships where the man expresses his will with greater power. HIV infection is a major concern for most women in situations of sexual violence, a fact that is justified by its higher incidence, in addition to other types, in the seropositive group[41,42]. Research carried out in the South of Brazil on the occurrence of pregnancies after a diagnosis of HIV infection has shown that this fact indicates the lack of a guarantee of the reproductive rights of women living with HIV/AIDS, since these pregnancies usually occur in contexts of great vulnerability[43].

"Though preventable, globally, each year about two million pregnant women become infected with syphilis." This statement by Shahrook et al. in their study about screening strategies during prenatal care portrays the situation of this malady that has been sweeping the world for decades. In this research, the proactive strategy of local screening for syphilis in prenatal care demonstrated better results on adequate treatment, of both the partner and the mother, which reflected in a lower incidence of congenital syphilis[44]. Thus, the importance of basic care during the opportunity of prenatal care is emphasized to track, address, and manage problems related to violence and STIs. However, the identification of the of violence is not always welcomed by the victim or even by health professionals, who have demonstrated unpreparedness for these care[45]. In addition, according to Ashaba, women can feel a stigma caused by the discrimination of health professionals themselves and the personal shame associated with pregnancy as HIV-positive women. This can make it difficult to engage in HIV care, especially in the presence of structural barriers, such as the difficulty of transport to the clinic. Added to this, the participants in their study also experienced intimate partner violence and lack of support from their families[46]. Therefore, in order to better meet the health needs of pregnant women, health systems must incorporate coordinated and interdisciplinary services that perform contextual analysis, in which relevant family and social aspects of pregnant women are considered[47].

CONCLUSIONS

Marital violence, perpetrated by an intimate partner, has characteristics that are perpetuating within the relationship, and manifest differently when the woman is in the gestational period. The replacement of physical violence by psychological is one of the ways to mask the harmful effects practiced by the partner since its identification is usually more difficult. These women may present, to a greater degree, symptoms of anxiety and post-traumatic stress.

Several studies show the high magnitude of violence practiced by partners, with severe and recurrent episodes. Cases of sexual violence, accompanied by physical aggression demonstrate the victim's difficulty in responding and leave them in a state of vulnerability.

The literature points to a relationship between intimate partner violence against women and the presence of sexually transmitted infections, which would make it essential to detection it as part of women's basic healthcare. HIV infection is a major concern for most women in situations of sexual violence. Studies have shown a higher prevalence of partner violence in HIV-positive pregnant women associated with unprotected sex and abusive use of alcohol.

Based on the results of the present study, we emphasize that violence against women is a significant public health problem. The follow-up periods in the gestational phase, whether in the prenatal or postpartum period, are unique opportunities for health professionals to identify situations of violence and thus provide assistance.

RESUMO

OBJETIVO: *Sintetizar o conhecimento produzido em estudos sobre a associação entre violência e IST na gestação.*

MÉTODOS: *Nesta revisão sistemática, envolvemos as atividades básicas de identificação, compilação e registro dos ensaios. Os instrumentos de coleta de dados foram os estudos que investigaram, explicitamente, as relações entre violência e gestação e IST, no período de julho de 2012 a julho de 2017, utilizando PubMed, Biblioteca Cochrane, SciELO e Lilacs.*

RESULTADOS: *Ao todo, 26 artigos foram escolhidos para formar a base da análise deste estudo. A relação entre violência e IST foi observada em 22 dos 26 estudos, sendo que em oito deles a violência foi praticada durante o período de gestação. Em dois estudos, não houve evidências dessa relação. Em um estudo, a falta de cuidados com a IST foi atribuída ao despreparo dos profissionais de saúde. Transtornos mentais foram citados como resultantes de IST em três artigos e em outro como resultado de violência. Um estudo encontrou violência mais frequente contra adolescentes, enquanto outros dois citaram a gestação como um fator de proteção.*

CONCLUSÕES: *A VPI combina características que possuem uma expressão diferenciada quando a mulher está no período gestacional. A literatura aponta para uma relação entre a VPI contra as mulheres e a presença de IST. O acompanhamento da gravidez, seja no pré-parto, seja no pós-parto, oferece oportunidades únicas para o profissional de saúde identificar situações de violência e, assim, prestar assistência.*

PALAVRAS-CHAVE: *Violência contra as mulheres. Infecções sexualmente transmissíveis. Gravidez. Aborto. Natimorto. Ofensas sexuais.*

REFERENCES

1. Barros C, Schraiber LB, França-Junior I. Associação entre violência por parceiro íntimo contra a mulher e infecção por HIV. Rev Saude Publica [Internet]. 2011 Apr [cited 2012 Nov 7];45(2):365–72. Available from: http://www.scielo.br/scielo.php?script=sci_arttext&pid=S0034-89102011000200015&lng=pt&nrm=iso&tlng=en
2. Walsh JL, Senn TE, Carey MP. Exposure to different types of violence and subsequent sexual risk behavior among female sexually transmitted disease clinic patients: A latent class analysis. Psychol Violence [Internet]. 2012;2(4):339–54. Available from: http://doi.apa.org/getdoi.cfm?doi=10.1037/a0027716
3. Schraiber LB, D'Oliveira AFPL, França-Junior I, Diniz S, Portella AP, Ludermir AB, et al. Prevalência da violência contra a mulher por parceiro íntimo em regiões do Brasil. Rev Saude Publica [Internet]. 2007 Oct;41(5):797–807. Available from: http://www.scielo.br/scielo.php?script=sci_arttext&pid=S0034-89102007000500014&lng=pt&nrm=iso&tlng=pt
4. Rifiotis T. As delegacias especiais de proteção à mulher no Brasil e a «judiciarização» dos conflitos conjugais. Soc e Estado. 2004;19(1):85–119.
5. Pedrosa CM, Spink MJP. A violência contra mulher no cotidiano dos serviços de saúde: desafios para a formação médica. Saúde e Soc [Internet]. 2011 Mar;20(1):124–35. Available from: http://www.scielo.br/scielo.php?script=sci_arttext&pid=S0104-12902011000100015&lng=pt&nrm=iso&tlng=pt
6. Okabe I, Fonseca RMGS da. Violência contra a mulher: contribuições e limitações do sistema de informação. Rev da Esc Enferm da USP [Internet]. 2009 Jun;43(2):453–8. Available from: http://www.scielo.br/scielo.php?script=sci_arttext&pid=S0080-62342009000200027&lng=pt&nrm=iso&tlng=pt
7. Silva EP, Ludermir AB, Araújo TVB de, Valongueiro SA. Freqüência e padrão da violência por parceiro íntimo antes, durante e depois da gravidez. Rev Saude Publica [Internet]. 2011 Dec;45(6):1044–53. Available from: http://www.scielo.br/scielo.php?script=sci_arttext&pid=S0034-89102011000600006&lng=pt&nrm=iso&tlng=en
8. Domingues RMSM, Saracen V, Hartz ZM de A, Leal M do C. Sífilis congênita: evento sentinela da qualidade da assistência pré-natal. Rev Saude Publica [Internet]. 2013;47(1):147–57. Available from: http://www.scielosp.org/scielo.php?script=sci_arttext&pid=S0034-89102013000100019
9. Teixeira SAM, Taquette SR. Violência e atividade sexual desprotegida em adolescentes menores de 15 anos. Rev Assoc Med Bras. 2010;56(4):440–6.
10. Goncalves DU, Proietti FA, Ribas JGR, Araujo MG, Pinheiro SR, Guedes AC, et al. Epidemiology, Treatment, and Prevention of Human T-Cell Leukemia Virus Type 1-Associated Diseases. Clin Microbiol Rev [Internet]. 2010 Jul 1;23(3):577–89. Available from: http://cmr.asm.org/cgi/doi/10.1128/CMR.00063-09
11. Bittencourt AL, Primo J, Oliveira M de FP de. Manifestations of the human T-cell lymphotropic virus type I infection in childhood and adolescence. J Pediatr (Rio J) [Internet]. 2006 Dec 13;82(6):411–20. Available from: http://www.jped.com.br/conteudo/Ing_resumo.asp?varArtigo=1573&cod=&idSecao=3
12. Hatcher AM, Romito P, Odero M, Bukusi EA, Onono M, Turan JM. Social context and drivers of intimate partner violence in rural Kenya: implications for the health of pregnant women. Cult Health Sex [Internet]. 2013 Apr;15(4):404–19. Available from: http://www.tandfonline.com/doi/abs/10.1080/13691058.2012.760205
13. Turan JM, Hatcher AM, Romito P, Mangone E, Durojaiye M, Odero M, et al. Intimate partner violence and forced migration during pregnancy: Structural constraints to women's agency. Glob Public Health [Internet]. 2016 Feb 7;11(1–2):153–68. Available from: http://dx.plos.org/10.1371/journal.pone.0176256
14. Shamu S, Abrahams N, Zarowsky C, Shefer T, Temmerman M. Intimate partner violence during pregnancy in Zimbabwe: a cross-sectional study of prevalence, predictors and associations with HIV. Trop Med Int Heal [Internet]. 2013 Jun;18(6):696–711. Available from: http://doi.wiley.com/10.1111/tmi.12078
15. Rotheram-Borus MJ, Tomlinson M, Roux I Le, Stein JA. Alcohol Use, Partner Violence, and Depression. Am J Prev Med [Internet]. 2015 Nov;49(5):715–25. Available from: http://linkinghub.elsevier.com/retrieve/pii/S0749379715002251
16. Jahanfar S, Janssen PA, Howard LM, Dowswell T. Interventions for preventing or reducing domestic violence against pregnant women. Cochrane database Syst Rev [Internet]. 2013 Feb 28;(2):CD009414. Available from: http://www.ncbi.nlm.nih.gov/pubmed/23450603
17. McDougal L, Strathdee SA, Rangel G, Martinez G, Vera A, Sirotin N, et al. Adverse pregnancy outcomes and sexual violence among female sex workers who inject drugs on the United States-Mexico border. Violence Vict [Internet]. 2013 Jun;28(3):496–512. Available from: http://linkinghub.elsevier.com/retrieve/pii/S0048357511000411
18. Oza KK, Silverman JG, Bojorquez I, Strathdee SA, Goldenberg SM. Examining negative effects of early life experiences on reproductive and sexual health among female sex workers in Tijuana, Mexico. Int J Gynecol Obstet [Internet]. 2015 Feb;128(2):169–73. Available from: http://doi.wiley.com/10.1016/j.ijgo.2014.08.014
19. Recto P, Champion JD. Psychological Distress and Associated Factors Among Mexican American Adolescent Females. Hisp Heal Care Int [Internet]. 2016 Dec 16;14(4):170–6. Available from: http://journals.sagepub.com/doi/10.1177/1540415316676224
20. Gómez Camargo DE, Ochoa Diaz MM, Canchila Barrios CA, Ramos Clason EC, Salguedo Madrid GI, Malambo García DI. Salud sexual y reproductiva en estudiantes universitarios de una institución de educación superior en Colombia. Rev Salud Pública [Internet]. 2015;16(5):660–72. Available from: http://www.revistas.unal.edu.co/index.php/revsaludpublica/article/view/39998
21. Northridge JL, Silver EJ, Talib HJ, Coupey SM. Reproductive Coercion in High School-Aged Girls: Associations with Reproductive Health Risk and Intimate Partner Violence. J Pediatr Adolesc Gynecol [Internet]. 2017;30(6):603–8. Available from: https://doi.org/10.1016/j.jpag.2017.06.007
22. Blake MDT, Drezett J, Machi GS, Pereira VX, Raimundo RD, Oliveira FR,

et al. Factors associated to late-term abortion after rape: literature review. Reprodução Clim [Internet]. 2014 May;29(2):60–5. Available from: http://dx.doi.org/10.1016/j.recli.2014.08.003

23. Baumgartner JN, Kaaya S, Karungula H, Kaale A, Headley J, Tolley E. Domestic violence among adolescents in HIV prevention research in Tanzania: participant experiences and measurement issues. Matern Child Health J [Internet]. 2015 Jan;19(1):33–9. Available from: http://link.springer.com/article/10.1007/s10995?014?1492?1

24. Perry R, Zimmerman L, Al-Saden I, Fatima A, Cowett A, Patel A. Prevalence of rape-related pregnancy as an indication for abortion at two urban family planning clinics. Contraception [Internet]. 2015;91(5):393–7. Available from: http://dx.doi.org/10.1016/j.contraception.2015.01.012

25. Drezett J, Blake MDT, Lira KSF de, Pimentel RM, Adami F, Bessa MMM, et al. Doenças sexualmente transmissíveis em mulheres que sofrem crimes sexuais. Reprodução Clim [Internet]. 2012 Sep;27(3):109–16. Available from: http://dx.doi.org/10.1016/j.recli.2013.03.004

26. Zihindula G, Maharaj P. Risk of sexual violence: Perspectives and experiences of women in a hospital in the Democratic Republic of Congo. J Community Health [Internet]. 2015;40(4):736–43. Available from: http://dx.doi.org/10.1007/s10900-015-9992-5

27. Chersich MF, Bosire W, King N, Temmerman M, Luchters S. Effects of hazardous and harmful alcohol use on HIV incidence and sexual behaviour : a cohort study of Kenyan female sex workers. Global Health [Internet]. 2014;10(1):1–11. Available from: Globalization and Health

28. Darak S, Gadgil M, Balestre E, Kulkarni M, Kulkarni V, Kulkarni S, et al. HIV risk perception among pregnant women in western India: Need for reducing vulnerabilities rather than improving knowledge! AIDS Care [Internet]. 2014 Jun 3;26(6):709–15. Available from: http://www.tandfonline.com/doi/abs/10.1080/09540121.2013.855303

29. Russell BS, Eaton LA, Petersen-Williams P. Intersecting epidemics among pregnant women: alcohol use, interpersonal violence, and HIV infection in South Africa. Curr HIV/AIDS Rep [Internet]. 2013 Mar 12;10(1):103–10. Available from: http://link.springer.com/10.1007/s11904-012-0145-5

30. Hyginus E, Chukwuemeka I, Lawrence I, Sunday M. HIV-related intimate partner violence among pregnant women in Nigeria. Vol. 9, East Afr J Public Health. 2012. p. 29–32.

31. Hampanda KM. Intimate partner violence and HIV-positive women's non-adherence to antiretroviral medication for the purpose of prevention of mother-to-child transmission in Lusaka, Zambia. Soc Sci Med [Internet]. 2016 Mar;153:123–30. Available from: http://linkinghub.elsevier.com/retrieve/pii/S0277953616300661

32. Watts C, Seeley J. Addressing gender inequality and intimate partner violence as critical barriers to an effective HIV response in sub-Saharan Africa. J Int AIDS Soc. 2014;17(1):5–7.

33. Peltzer K, Jones D, Weiss SM, Villar-Loubet O, Shikwane E. Sexual risk, serostatus and intimate partner violence among couples during pregnancy in rural South Africa. AIDS Behav [Internet]. 2013 Feb;17(2):508–16. Available from: http://www.ncbi.nlm.nih.gov/pubmed/22488126

34. Peltzer K, Mlambo G. Sexual HIV risk behaviour and associated factors among pregnant women in Mpumalanga, South Africa. BMC Pregnancy Childbirth [Internet]. 2013 Dec 4;13(1):57. Available from: BMC Pregnancy and Childbirth

35. Mayosi BM, Lawn JE, van Niekerk A, Bradshaw D, Abdool Karim SS, Coovadia HM, et al. Health in South Africa: changes and challenges since 2009. Lancet (London, England) [Internet]. 2012 Dec 8;380(9858):2029–43. Available from: http://dx.doi.org/10.1016/S0140-6736(12)61814-5

36. Aguilar-Zapata D, Piñeiruá-Menéndez A, Volkow-Fernández P, Rodríguez-Zulueta P, Ramos-Alamillo U, Cabrera-López T, et al. Sociodemographic differences among HIV-positive and HIV-negative recently pregnant women in Mexico City. Med (United States). 2017;96(27).

37. Narasimhan M, Orza L, Welbourn A, Bewley S, Crone T, Vazquez M. Sexual and reproductive health and human rights of women living with HIV: a global community survey. Bull World Health Organ [Internet]. 2016;94(4):243–9. Available from: http://www.who.int/entity/bulletin/volumes/94/4/14-150912.pdf

38. Maman S, Groves AK, McNaughton Reyes HL, Moodley D. Diagnosis and disclosure of HIV status: Implications for Women's Risk of Physical Partner Violence in the Postpartum Period. JAIDS J Acquir Immune Defic Syndr [Internet]. 2016 Mar;72(5):1. Available from: http://content.wkhealth.com/linkback/openurl?sid=WKPTLP:landingpage&an=00126334-900000000-97264

39. Shamu S, Zarowsky C, Shefer T, Temmerman M, Abrahams N. Intimate partner violence after disclosure of HIV test results among pregnant women in Harare, Zimbabwe. PLoS One. 2014;9(10).

40. Malaju MT, Alene GD. Women's expectation of partner's violence on HIV disclosure for prevention of mother to child transmission of HIV in North West Ethiopia. BMC Res Notes [Internet]. 2013;6(1):96. Available from: BMC Research Notes

41. Flores Ceccon R, Nazareth Meneghel S. HIV e violência contra mulheres: estudo em município com alta prevalência de Aids no Sul do Brasil. Rev Panam Salud Publica [Internet]. 2015;37(4/5):287--292 6p. Available from: http://ezp-prod1.hul.harvard.edu/login?url=http://search.ebscohost.com/login.aspx?direct=true%7B&%7Ddb=c8h%7B&%7DAN=109821053%7B&%7Dsite=ehost-live%7B&%7Dscope=site

42. Ghosh M, Rodriguez-Garcia M, Wira CR. Immunobiology of Genital Tract Trauma: Endocrine Regulation of HIV Acquisition in Women Following Sexual Assault or Genital Tract Mutilation. Am J Reprod Immunol [Internet]. 2013 Feb;69(SUPPL.1):51–60. Available from: http://onlinelibrary.wiley.com/doi/10.1111/aji.12027/full

43. Teixeira LB, Pilecco FB, Vigo Á, Drachler M de L, Leite JC de C, Knauth DR. Factors associated with post-diagnosis pregnancies in women living with HIV in the south of Brazil. PLoS One [Internet]. 2017;12(2):e0172514. Available from: http://dx.plos.org/10.1371/journal.pone.0172514

44. Shahrook S, Mori R, Ochirbat T, Gomi H. Strategies of testing for syphilis during pregnancy. Cochrane database Syst Rev [Internet]. 2014 Oct 29;10(10):CD010385. Available from: http://www.ncbi.nlm.nih.gov/pubmed/25352226

45. Cybulska B. Immediate medical care after sexual assault. Best Pract Res Clin Obstet Gynaecol [Internet]. 2013 Feb;27(1):141–9. Available from: http://dx.doi.org/10.1016/j.bpobgyn.2012.08.013

46. Ashaba S, Kaida A, Coleman JN, Burns BF, Dunkley E, O'Neil K, et al. Psychosocial challenges facing women living with HIV during the perinatal period in rural Uganda. Mazza M, editor. PLoS One [Internet]. 2017 May 1;12(5):e0176256. Available from: http://dx.plos.org/10.1371/journal.pone.0176256

47. Secor-Turner M, McMorris B, Sieving R, Bearinger LH. Life experiences of instability and sexual risk behaviors among high-risk adolescent females. Perspect Sex Reprod Health [Internet]. 2013;45(2):101–7. Available from: http://www.ncbi.nlm.nih.gov/pubmed/23750624