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Letter to the editor

Letter to the editor

Dr. Inácio de Barros Melo Neto[1]

With another issue of the journal Annals of the Faculty of Medicine of Olinda, our hope and faith are renewed after difficult moments during the COVID-19 pandemic. Although the pandemic persists, the number of cases and deaths has decreased due to the broad vaccination coverage of the Brazilian population. This control allowed us to return with even more determination to fulfill our mission.

In our institution, the first semester of 2022 was celebrated with actions, including the construction of the new Medical Skills Laboratory, the new school clinic, the creation of the Institutional Research and Extension Support Fund, and the Regulation of Actions of Extension. These initiatives, associated with our innovative medical training project, enabled the institution to obtain top marks in the re-accreditation process with the Instituto Nacional de Estudos e Pesquisas Educacionais Anísio Teixeira – Ministério da Educação consolidating our mission of offering excellent medical education.

For us, it is gratifying to consolidate, every day, our expectations in building an institution with values that we consider essential for the training process and a fair and sustainable society. The engagement of our students and teachers in the activities proposed within the scope of this journal is a source of great pride.

In this way, we reaffirm our commitment to the pillars of higher education, strengthening the inseparable foundations of teaching, research, and extension. Thus, the Faculty of Medicine of Olinda will become a training center for doctors with a critical and humanistic vision focused on working closer to society.

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Letter from editor

Prof. Paulo Sávio A. Goes, PhD.

We reached number 7 in the journal Annals of the Faculty of Medicine of Olinda. This issue brings even more advances in scientific dissemination and knowledge production in the medical field. Thanks to institutional investments, this journal is making great strides toward indexation.

All copies include the Digital Object Identifier, which allows our studies to be cited in any database. In addition, it enables greater accuracy of bibliographic citation and registration in the Creative Commons database, which is an important step that will take us faster towards internationalization.

From this issue onwards, the submission flows of the journal will be computerized, relying on blind evaluation by evaluators in the area. Also, a new submission layout will be available for manuscripts, following the standards of each type of study, namely original articles, systematic and narrative literature reviews, case reports, experience reports focusing on our scope of social responsibility, and reviews.

We have no doubt that the journal Annals of the Faculty of Medicine of Olinda has established itself as a reference in the medical field, focusing on humanistic medicine and being committed to the needed transformations in society.

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4D PRINTING OF EXTRACELLULAR MATRIX: A PILOT PROJECT FOR THE DEVELOPMENT OF COMPLEX TISSUES

Impressão 4D de matriz extracelular: projeto piloto para o desenvolvimento de tecidos complexos

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ABSTRACT

Introduction: The shortage of organs and tissues for transplant is a great challenge in the medical field. The 3D printing emerges as a feasible solution for this problem. This study presents preliminary results of the first models of a 4D extracellular matrix (ECM) using a biomaterial.

Methods: This experimental multicenter study was conducted at the laboratory of simulation and digital fabrication of the Federal Institute of Pernambuco. A solution of sodium alginate at 2% was used to print the ECM on a 3D printer model Zmorph type fused deposition modeling.

Results: The geometric figure used to print the ECM was a square with 1cm^2 of area initially planned in a computer aided design and developed using the Voxelizer software. The biomaterial in gel was successfully printed with extrusion speed of 1mm/s and printing speed of 3mm/s. The uniform deposit of the material during printing resulted in the geometric figure designed.

Discussion: This paper shows preliminary results of the printing of a 4D ECM. The sodium alginate might be a promising biopolymer to the future phases of this study.

Conclusion: The variation of the nozzle area and extrusion speed highly influences the analysis of the 3D printer final parameters.

Keywords: Bioprinting; extracellular matrix; regenerative medicine; bioengineering; scaffolds.

RESUMO

Introdução: A indisponibilidade de órgãos e tecidos para transplante com o objetivo de atender a demanda existente é um dos grandes desafios na área médica. Uma solução viável é a impressão tridimensional de órgãos e tecidos. O presente estudo apresentou resultados preliminares dos primeiros modelos da matriz extracelular (MEC) 4D utilizando um biomaterial.

Métodos: Estudo experimental multicêntrico realizado no laboratório de Simulação e Fabricação Digital do Instituto Federal de Pernambuco. Uma solução de alginato de sódio a 2% foi utilizada para a fabricação da MEC em uma impressora 3D Zmorph do tipo fused deposition modeling.

Resultados: A figura geométrica da MEC foi um quadrado com 1 cm^2 de área desenvolvida pelo software Voxelizer, projetado inicialmente no *computer aided design*. A seguir, o biomaterial em forma de gel foi impresso com uma velocidade de extrusão de 1 mm/s e uma velocidade de impressão de 3 mm/s. Obteve-se êxito na impressão. O depósito uniforme do material durante a impressão permitiu a obtenção da forma geométrica projetada.

Discussão: O presente estudo mostra resultados preliminares do processo de impressão de uma MEC 4D. O êxito da impressão qualifica o alginato como um dos principais biopolímeros a ser utilizado nas futuras fases desse estudo.

Conclusão: A variação da área da ponteira e a velocidade de extrusão têm grande influência na análise dos parâmetros finais na utilização da impressora tridimensional.

Palavras-chave: bioimpressão; matriz extracelular; medicina regenerativa; bioengenharia; tecidos suporte.

INTRODUCTION

The shortage of organs for transplant is a major challenge in the medical field. Approximately six million people worldwide are in the final stage of organ failure, [1] and in several countries, high mortality rates affect patients on the waiting list for transplants. In the United States, around 40,000 patients receive transplants each year. However, 120,000 remain on the waiting list, and 7,600 die while waiting. Furthermore, the increasing demand for transplants might be proportional to the increased life expectancy and incidence of chronic diseases in the next years.[2]

The COVID-19 pandemic worsened this scenario, revealing a strong temporal association between the increased coronavirus infections and the reduced transplants of solid organs. In France and United States, the rate of transplants of deceased donors decreased by 90.6% and 51.1%, respectively. In addition, the organ procurement and transplant rates were reduced even in regions less affected by the pandemic, suggesting a global effect.[3] In Brazil, population aging follows the global trend, which combines with the rising number of patients with organ failure and increases the demand for transplants.[4]

Technologies inspired by regenerative medicine challenge the current allograft transplant model. The 3D printing of organs and tissues might be a feasible solution for the shortage of organs for transplant. Combined with tissue bioengineering, regenerative medicine allows new approaches for replacing and regenerating tissues and organs, with the potential to correct congenital defects using laboratory-grown organs and tissues, known as bioartificial organs.[5]

A systematic review highlighted the revolutionary potential of 3D printing in patient counseling, preoperative and intraoperative surgical planning, and education in urology. Although time and costs of production remain as challenges, 3D printing might represent a step forward in meeting the needs of patients and surgeons.[6]

Despite experimental, regenerative medicine has already remarkable results. Atala et al.[7], a world reference in 3D printing of biological material, developed a biocompatible tissue capable of fully or partially replacing the bladder; however, without functional contraction. The full implant of the organ developed was successful and significantly improved the quality of life of patients. [7] Atala is responsible for several other studies on regenerative medicine using 3D printing, such as the production of a human endothelialized myocardium from stem cells in a microfluidic hydrogel ECM.[8]

The 3D printing enables the fabrication of complex shapes with high precision by adding layers of smart materials. These materials change shape, color, or generate electrical currents in response to external stimuli, being bioactive or fulfilling a desired function. The 3D printing allowed the development of dynamic 3D struc-

tures, known as 4D printing.[9]

Tissues *in vivo* are dynamic structures composed of various cell types, an ECM, and a range of signaling molecules. The ECM is a crucial component of the cellular microenvironment, forming a complex 3D network.[10] In 3D printing, cells added to the ECM may be organized into complex structures, creating tissues or organs.[11] Currently, regenerative medicine aims to develop a technique of 4D printing for biological material capable of implantation in the human body, replacing tissues or organs while reducing the risk of rejection due to biological compatibility.

Therefore, this study aimed to fabricate 4D ECMs of sodium alginate using a 3D printer, with the purpose of adding cells hereafter originators of tissues and organs. This study seeks future application in the regenerative medicine with animals and humans.

ECMs were produced in the laboratory of simulation and digital fabrication at the research center of the Federal Institute of Pernambuco (Recife, Brazil). Thus, this study shows preliminary results of the printing, including training for printer operation and mechanical fabrication.

METHODS

This multicenter experimental study involved the Faculty of Medicine of Olinda, the Federal Institute of Pernambuco, and the Federal University of Pernambuco. An extensive literature review was conducted to define the biomaterial, printer, extrusion nozzle adapted for bio-inks, software, geometric shape, and printing technique for the fabrication of ECMs.

The biomaterial selected for the initial tests was sodium alginate (Figure 1), defined as a class of biopolymers initially derived from the cell wall of brown algae from various species (e.g., *Laminaria*, *Macrocystis* and *Sargassum*).[12, 13]

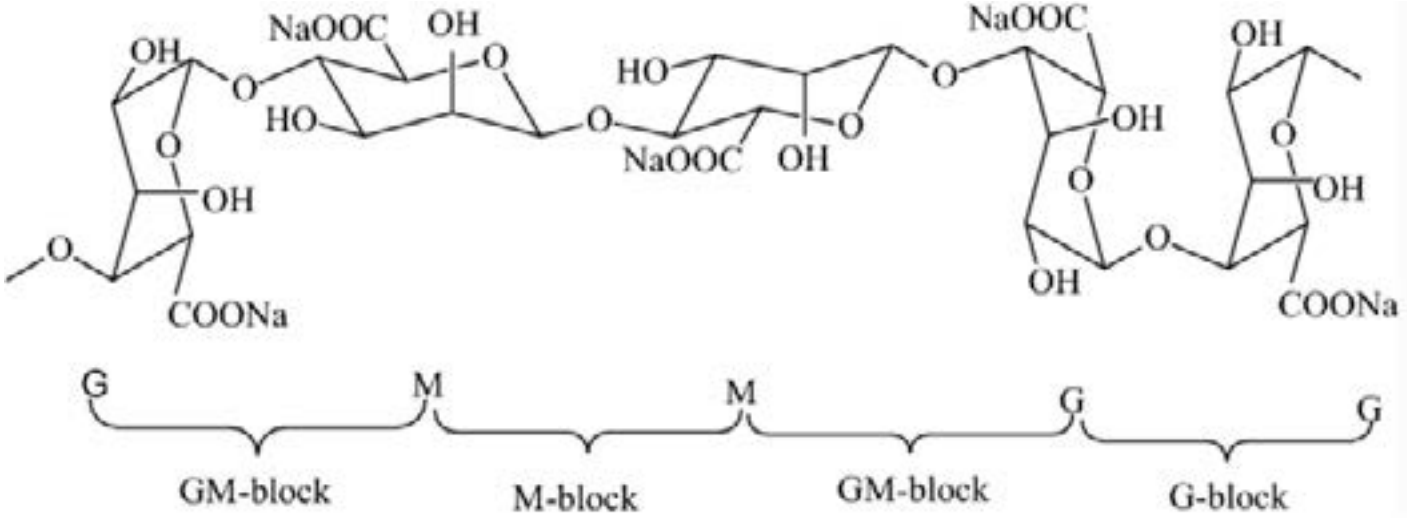

Figure 1. Different blocks of sodium alginate linked by 1→4 glycosidic bonds.

For ECM fabrication, a gel was produced consisting of a solution containing 2% sodium alginate and 0.25% sodium chloride diluted in water. Under mechanical stirring and heated to 80° C, calcium chloride was added at a concentration of 0.25% in a ratio of 1:5 mL, respectively.

The 3D printer model selected was the Zmorph type fused deposition modeling. For the initial tests, The Voxelizer software version 2.0(Zmorph S.A., Wrocław, Poland was used to convert the computer-aided design (CAD) files into GCODE format (i.e., the language used to describe how the 3D printer should execute a printing). The printer used a paste extrusion nozzle in aluminum of 4 mm, operating at a flow rate of 5 mL/s in a linear format to construct squares with 1 cm^2 of area. Initially, polylactic acid (PLA) and cellulose were used to define printing parameters.

RESULTS

The preliminary results included training in printer operation and mechanical fabrication. Initially, the structure of ECMs was designed in CAD (Figure 2), and the Voxelizer signaled the printer to execute the printing (Figure 3).

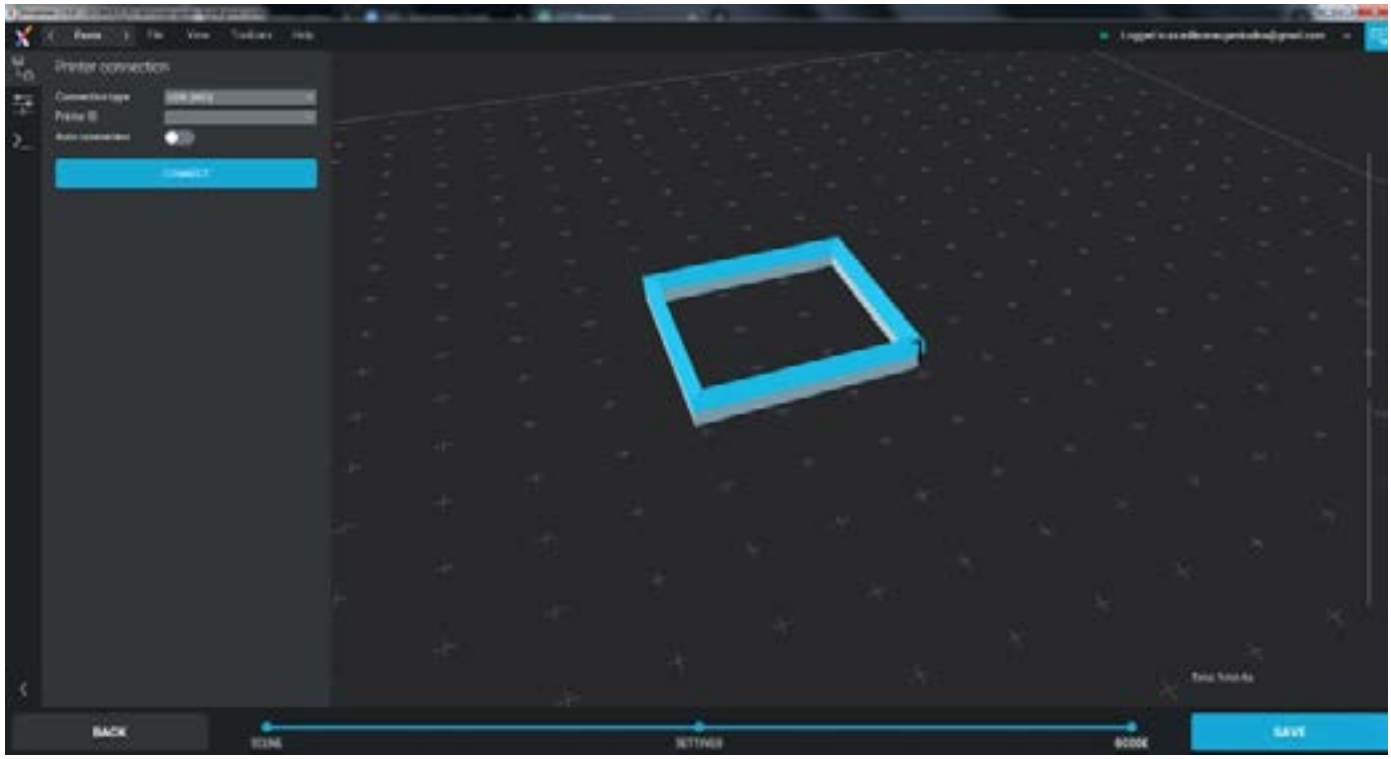

Figure 2. Design of the ECM structure.

Figure 3. Zmorph 3D printer, type fused deposition modeling initiating the printing.

For the experimental analysis of the printing, five tests were conducted using PLA, cellulose, and sodium alginate to configure parameters, which included extrusion and printing speeds, described as follows:

1) PLA: utilization of a nozzle with an area of 0.4 mm2, extrusion speed of 40 mm/s, and printing speed of 40 mm/s. Successful printing of the geometric figure (Figure 4).
2) Cellulose: utilization of a nozzle adapted for bio-inks with an area of 13.2 mm², extrusion speed of 7 mm/s, and printing speed of 10 mm/s. During the test, the extrusion value associated with the deposition speed applied to the material was too high, hindering the printing of the geometric figure.
3) Sodium alginate: utilization of a syringe with tips of 35.7 mm and 4.0 mm in diameter, extrusion speed of 1.5 mm/s, and a printing speed of 1 mm/s. A significant material deposition occurred at one pole, hindering the printing of the geometric figure.
4) Sodium alginate: utilization of a syringe with the same characteristics as the previous, extrusion speed of 1 mm/s, and printing speed of 0.5 mm/s. Similarly to the previous test, a high material deposition occurred at one pole, hindering the printing of the geometric figure.
5) Sodium alginate: utilization of a syringe with the same characteristics as test 3, extrusion speed of 1 mm/s, and printing speed of 3 mm/s. The material was uniformly deposited and the geometric figure was successfully printed (Figure 5).

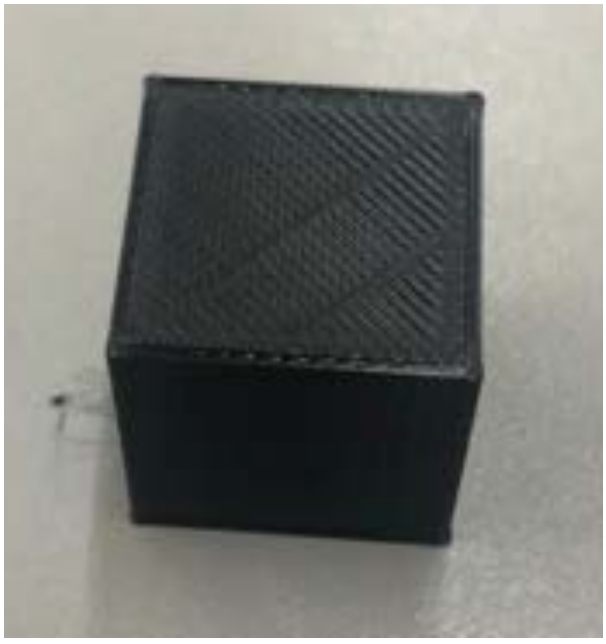

Figure 4. Model printed with polylactic acid.

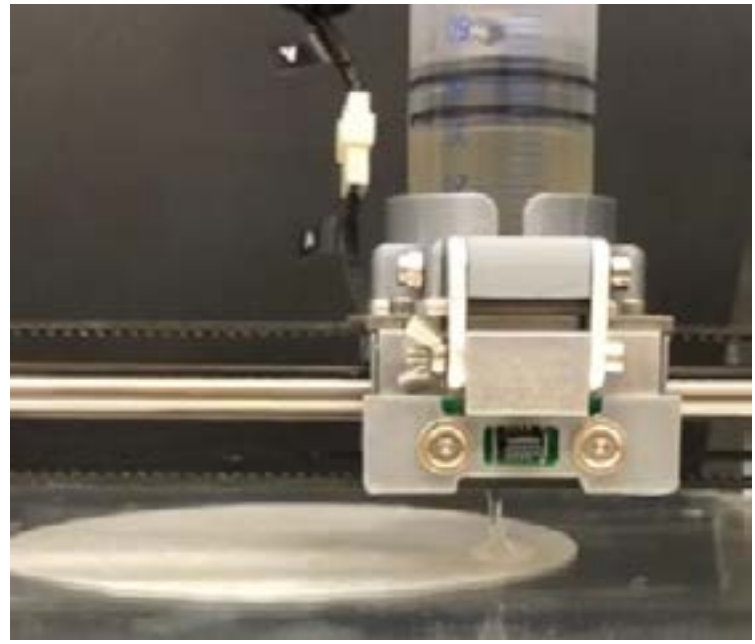

Figure 5. Model printed with sodium alginate.

DISCUSSION

The shortage of organs and tissues for transplant is one of the major challenges in the medical field. Furthermore, the COVID-19 pandemic negatively impacted worldwide transplant rates, decreasing procedures and organ donors, and increasing mortality among patients on the waiting list.[3, 14]

This scenario requires innovative therapies, such as regenerative medicine, which is crucial for replacing or restoring human cells, tissues, and organs. However, the fabrication of ECM (a pillar of regenerative medicine) represents one of the technological obstacles to tissue production. ECMs are structures made from synthetic or natural materials on which new tissues may grow to replace damaged tissues.[15] Creating a similar environment to the human ECM in which cells may attach and maintain their proper metabolism is complex.

This study shows preliminary results of the 4D printing of ECMs, including training for printer operation and mechanical fabrication. The equipment choice was defined by optimizing the existing resources since ECMs fabrication using 3D printers incurs high costs.[16]

The sodium alginate was selected as biopolymer for being one of the most used biomaterials in bioengineering due to its molecular similarity to polysaccharides. Sodium alginate enables the production of gels that incorporate stem cells and mimic an ECM microenvironment, facilitating tissue bioengineering and basic research.[17]

The first test was conducted with PLA, which presents superior thermomechanical characteristics compared to the gel of sodium alginate. The PLA has greater mechanical strength and a lower coefficient of thermal expansion, enhancing its printability and reducing biases during the production process.[18] In the second test, extrusion and deposition speeds were excessive, hindering the printing. Consequently, parameters were defined using sodium alginate.

Successfully printing the ECM was not possible using parameters previously obtained with the different materials, as showed in tests 3 and 4. However, these tests indicated that reducing the extrusion speed to 1 mm/s and increasing the printing speed to 3 mm/s would enable the intended printing. After applying the parameters, a uniform material deposition was achieved, resulting in the successful printing of the geometric figure.

This study had limitations. Statistical analyses were hindered by the absence of physicochemical analyses for a 4D ECM and the challenges associated with stabilizing the extruded filament according to the tested material. However, our findings are preliminary and foresee advancement in regenerative medicine and tissue bioengineering.

CONCLUSION

The variation in the nozzle area and the extrusion speed influenced the outcome of the 4D ECM printing. The ease of handling and the accessibility of sodium alginate were observed during the printing. Sodium alginate proved to be highly useful in tissue bioengineering and should be considered in future studies aimed at stabilizing the extruded filament.

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COMPARISON OF VASCULAR CALCIUM CHANNEL BLOCK PROMOTED BY VERAPAMIL AND NIFEDIPINE BY MOLECULAR MODELING

Comparação do bloqueio de canal de cálcio vascular promovido por verapamil e nifedipina por modelagem molecular

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ABSTRACT

Cardiovascular diseases are extensively studied due to their risks and global prevalence. Thus, searching for new treatments is an ongoing process, and plant-derived compounds are a promising source of drugs. Nowadays, molecular modeling analyses are conducted to understand the structure-function relationship of a pharmacological target and its protein-ligand interactions (i.e., the exact mechanism of pharmacological action). This study aimed to compare the voltage-dependent calcium channel (CaV) blockade promoted by verapamil and nifedipine using molecular modeling analyses. Tests and analyses were performed using the Dockthor and Chimera programs, respectively. The t-test was used to compare the affinity energies. Data analysis occurred in the GraphPad Prism software; statistical significance was set at $p < 0.05$. Verapamil had a higher affinity than nifedipine ($p < 0.0001$). Although the drugs presented different binding sites, they successfully blocked the CaV and prevented the calcium influx into the cell. These findings could be useful for the prospect of new drugs that block CaV.

Keywords: Antihypertensive agents; calcium channel blocker; docking molecular.

RESUMO

As doenças cardiovasculares são extensamente estudadas devido aos seus riscos e prevalência mundial. Sendo assim, a pesquisa por novos tratamentos é um processo contínuo e as substâncias isoladas de plantas são uma fonte promissora de fármacos. Atualmente, análises de modelagem molecular são realizadas para compreender a relação entre estrutura-função de um alvo farmacológico e suas interações proteína-ligante, e assim o mecanismo exato da ação farmacológica. Diante disso, este trabalho visou comparar o bloqueio do canal de cálcio dependente de voltagem promovido por verapamil e nifedipina com análises de modelagem molecular. Os experimentos de *docking* molecular foram realizados usando o portal DockThor e análises com o programa Chimera. As energias de afinidade foram comparadas utilizando o programa GraphPad Prism, com o teste "t", em que valores de $p < 0,05$ foram considerados significantes. Os resultados mostram que o verapamil teve uma afinidade maior do que a de nifedipina ($p < 0,0001$). As simulações de configuração no canal foram diferentes, cada uma bloqueando o seu poro, o que impede o influxo de cálcio na célula. Estes dados serão úteis para comparar à prospecção de novas drogas que bloqueiam o canal de cálcio.

Palavras-chave: Agentes anti-hipertensivos; bloqueador de canal de cálcio; modelagem molecular.

INTRODUCTION

Cardiovascular diseases are extensively studied due to their risks and global prevalence. Although their incidence is decreasing in Brazil, they are still the leading cause of death.[1] Thus, searching for new treatments is an ongoing process, and plant-derived compounds are a promising source of drugs.[2]

Molecular modeling analyses are conducted to understand the structure-function relationship of a pharmacological target and its protein-ligand interactions.[3-6] These analyses provide information about the structural dynamics of the molecules and also the energy required to bind them in a target protein, which is essential for characterizing the mechanism of drug action.[7]

In blood pressure regulation, the α-1C subunit of L-type Ca^{2+} channels (CaV1.2) is the main pathway for Ca^{2+} influx into cells. Upon activation, these channels mediate Ca^{2+} into the cytoplasm, triggering vasoconstriction.[8-10] Voltage-gated calcium channel (CaV) blockers are commonly used for treating hypertension and are classified as dihydropyridines or non-dihydropyridines.[11-13] Dihydropyridines (e.g., amlodipine, nifedipine, and felodipine) cause a predominant vasodilatory effect with minimal interference in heart rate and systolic function, which justifies their use as antihypertensive drugs. Non-dihydropyridines (e.g., phenylalkylamines [verapamil] and benzothiazepines [diltiazem]) have a less pronounced vasodilatory effect and act on the cardiac muscle and the cardiac conduction system. Therefore, they reduce heart rate, exert antiarrhythmic effects, and may depress systolic function.[14] Thus, this study aimed to compare the CaV blockade promoted by verapamil and nifedipine using molecular modeling analyses.

MATERIAL AND METHODS

This study was quantitative and experimental. Verapamil (CID 2520, Figure 1A) and nifedipine (CID 4485, Figure 1B) were obtained from the PubChem database.[15] The homologous structure of CaV (ID 4MS2) was obtained from the Protein Data Bank (Figure 1C).

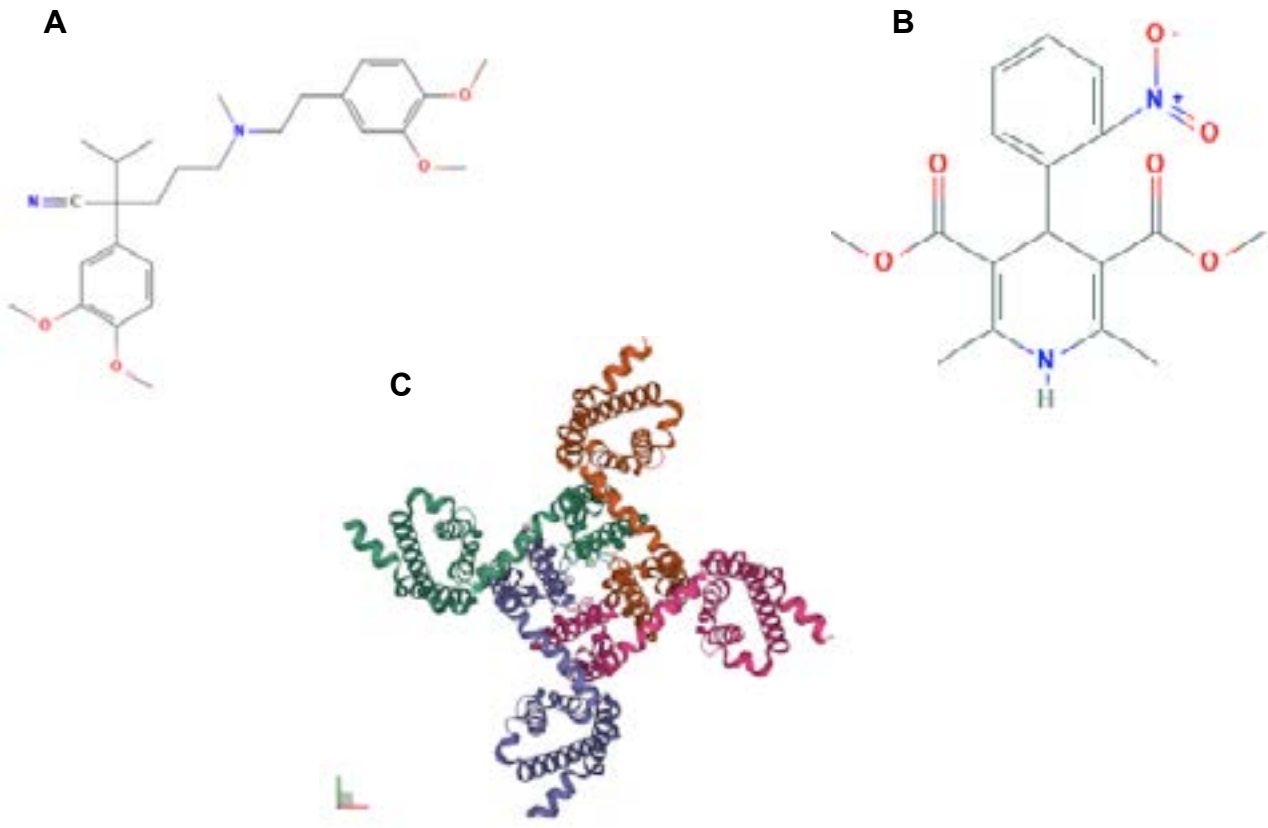

Figure 1. Molecular structure of verapamil (**A**, C27H38N2O4), nifedipine (**B**, C17H18N2O6) and the 3D structure of CaV (**C**).

Molecular modeling experiments were conducted using the online portal DockThor[16]; results were ranked by their highest affinity with the CaV protein. The three best docking configurations with the highest affinity for the target were selected for further analyses, and the positions were presented using the Chimera software (version 1.14).

Affinity energies were analyzed using the GraphPad Prism software, employing the t-test to compare the results of verapamil and nifedipine. Statistical significance was set at $p < 0.05$.

RESULTS

One million docking positions were generated for the drugs (verapamil and nifedipine) against the CaV protein (ID 4MS2). The three best positions based on binding energy values are presented in Table 1. Lower binding energy indicates a more stable interaction as a result of the hydrophobic and electrostatic bonds between the molecules and the CaV protein. Notably, verapamil demonstrated a significantly higher affinity for CaV than nifedipine.

Table 1. Binding affinities of drugs in the CaV channel.

Drug	**Affinity** (kcal/mol) Mean ± SEM
Verapamil	-9.696 ± 0.08*
Nifedipina	- 8.031 ± 0.009

Kcal/mol: kilocalorie per mole; SEM: standard error of the mean; *p < 0.0001 (verapamil vs. nifedipine), t-test

Configuration analysis of the drugs in the 3D structure of the CaV revealed that they occupy similar positions in the pore region. However, verapamil and nifedipine bind to different amino acid residues (Figure 2). Both drugs blocked the channel pore, hampering the influx of Ca^{2+} into the cells.

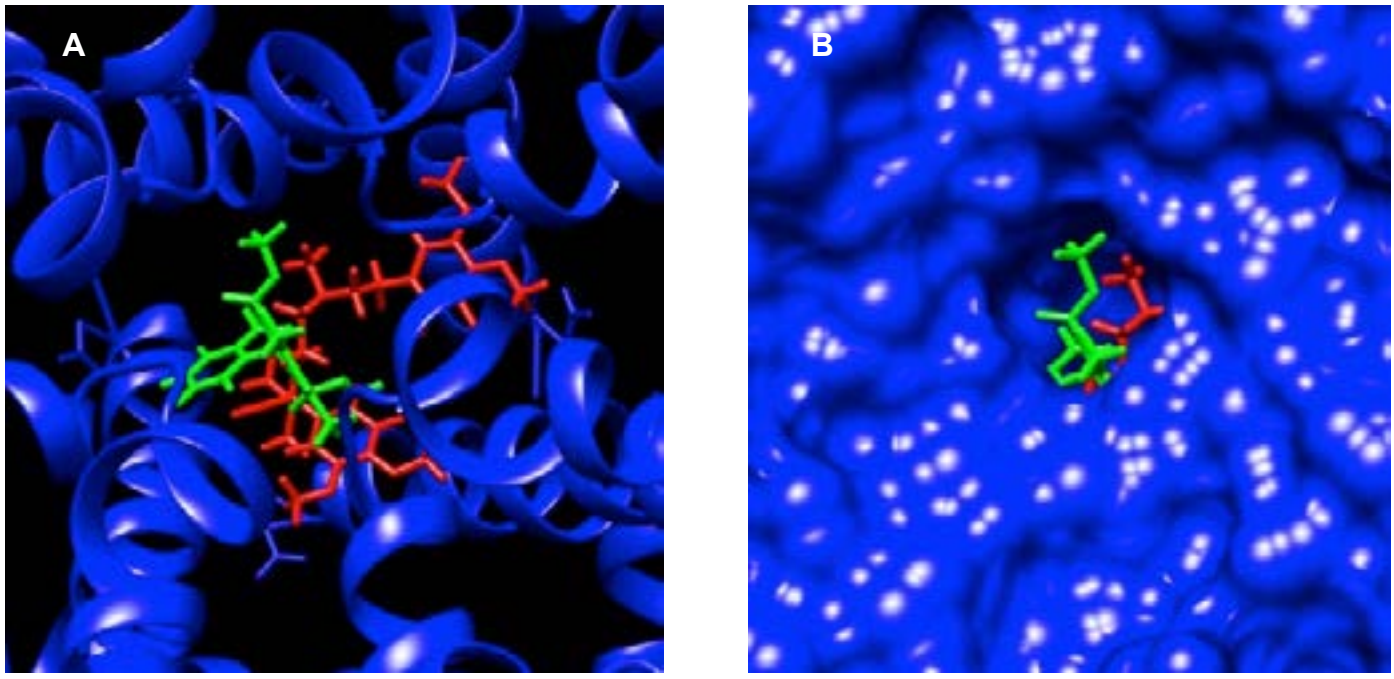

Figure 2. Positioning of verapamil (red) and nifedipine (green) in the 3D structure of the CaV channel (blue). **A:** quaternary protein structure; **B:** hydrophobic protein structure.

DISCUSSION

The CaV contains a large subunit (α1) that forms the pore using four homologous domains, each with six transmembrane segments and a loop that forms the pore[17](Figure 1C).

Phenylalkylamine-type CaV blockers (verapamil; Figure 1A) and benzothiazepines (diltiazem) are used for treating atrial arrhythmias.[18] Different affinities were observed for verapamil and nifedipine (Table 1); the former showed higher affinity. Verapamil presents a longer carbon chain than nifedipine, which allows more interactions with the amino acid residues of the CaV pore; this fact may justify the higher affinity. Phenylalkylamines have two aromatic rings linked by a central aliphatic chain, containing a tertiary amino group that can enter the external opening of the pore via the open activation gate and bind to a specific receptor site in a positively charged protonated form.[18,19]

Conversely, dihydropyridines (nifedipine; Figure 1B) interact with the S6 segments in domains III and IV, leading to a proposed interface-domain model for drug binding and action.[17,20] Another molecular modeling study suggests that these amino acid residues are located on the luminal side of the pore and collaborate to form the drug-binding site.[21]

Other dihydropyridine CaV blockers (nimodipine, benidipine, and amlodipine) have been evaluated for their orientation in blocking L-type calcium channels (CaV1.2).[22] Results similar to those of nifedipine (Figure 2) were obtained with uncarilan A, an alkaloid isolated from *Uncaria rhynchophylla*, a plant used in traditional Chinese medicine. The molecular simulation showed that uncarilan A inhibited the CaV, preventing calcium influx.[23]

This study used molecular modeling analyses to determine the CaV blockade promoted by verapamil and nifedipine as standard blockers used in clinical practice. Findings will serve to compare with new drugs that have potential CaV-blocking effects.

CONCLUSION

Nifedipine and verapamil blocked the CaV using different binding sites in the channel pore; the latter had a higher affinity. These findings will aid in the evaluation of new drugs that may also target and block the CaV.

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FUROSEMIDE BLOCKS THE ANION CHANNEL FORMED BY STAPHYLOCOCCUS AUREUS α-HEMOLYSIN

A furosemida bloqueia o canal aniônico causado por α-hemolisina de Staphylococcus aureus

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ABSTRACT

This study aimed to investigate the effect of furosemide on the *Staphylococcus aureus* α-hemolysin (α-HL) channel in planar lipid bilayer membranes using electrophysiological characterization and molecular docking analyses. Planar lipid bilayer membranes were prepared, and α-HL (0.07 mg/mL) was added to the standard solution in the *cis* compartment of the experimental chamber. All experiments were conducted at room temperature using the Axopatch 200A amplifier in voltage--clamp mode. The α-HL channels typically exhibited high conductance (~4 nS) and rarely switched to low conductance states at pH 7.5. Furosemide was added to the solution in the *cis* compartment after the ion channel was incorporated into the planar lipid bilayer membrane. For docking analyses, the Protein Data Bank (ID: 7AHL) was consulted for the atomic coordinates of the α-HL heptameric channel, and the furosemide structure was obtained from PubChem. The coordinates were built and minimized using the Avogadro software, and molecular docking experiments were conducted through the DockThor online platform. Furosemide inhibited the conductance of the α-HL channel in a voltage-dependent manner ($p < 0.05$). The two best docking solutions for the α-HL channel showed that the highest interaction affinity involved multiple hydrogen bonds, and residues 113 and 147 formed part of the constriction region of the α-HL channel. In conclusion, furosemide blocked ion currents at the constriction region of the *Staphylococcus aureus* α-HL channel.

Keywords: furosemide; *Staphylococcus aureus*; ion channel; virulence factors; anti-bacterial agent.

RESUMO

Investigar o efeito de furosemida no canal α-hemolisina (α-HL) de *Staphylococcus aureus* em bicamadas lipídicas planares (BLP) por caracterização eletrofisiológica e estudos de *docking* molecular. As BLP foram preparadas e α-HL (0,07 mg/mL) foi adicionada à solução padrão no compartimento *cis* da câmara experimental. Todos os experimentos foram realizados em temperatura ambiente usando um amplificador Axopatch 200A no modo *voltage clamp*. Em pH 7,5, os canais α-HL estavam geralmente em uma alta condutância (~ 4 nS) e raramente mudavam para estados de baixa condutância. Após a incorporação do canal iônico na BLP, a furosemida também foi adicionada à solução padrão no compartimento cis. Para os estudos de *docking*, as coordenadas atômicas para o canal heptamérico α-HL foram recuperadas *do Protein Data Bank* (ID: 7AHL), a estrutura de furosemida foi obtida do PubChem e suas coordenadas foram elaboradas e minimizadas com o software Avogadro. Os experimentos de *docking* molecular foram realizados usando o Dockthor online. A furosemida inibiu ($p < 0,05$) a condutância do canal α-HL de maneira voltagem-dependente. Foram avaliadas as duas melhores soluções de *docking* e o canal α-HL, observando-se que o modo de conexão com maior afinidade de interação possuía maior número de ligações de hidrogênio. Os resíduos de ligação foram o 113 e o 147, que formam os remanescentes de constrição do canal α-HL. Em conclusão, a furosemida bloqueia as correntes iônicas na

constrição do canal causado pela α-HL de *Staphylococcus aureus.*

Palavras-chave: furosemida; *Staphylococcus aureus;* canal iônico; fator de virulência; agente antimicrobiano.

INTRODUCTION

In the last decade, the rational design of drugs by using known chemical compounds for new indications (i.e., new targets) has been the most effective innovation approach, recognized as a model to advance drug research and development.[1] The *Staphylococcus aureus* α-hemolysin (α-HL) channel has an important pathogenic role and is a virulence factor involved in infection and multidrug-resistant strains.[2] Thus, the interest in repurposing known chemical compounds as antibiotics has emerged to target different bacterial mechanisms.[2]

Previous studies have assessed the ability of several compounds to inhibit α-HL by hindering its assembly in the membrane or directly blocking the channel.[3-6] In this context, furosemide is a loop diuretic with a wide range of pharmacological properties that compete with chloride to bind to the sodium-potassium-chloride cotransporter, inhibiting the reabsorption of sodium and chloride.[7] The α-HL is an exotoxin that forms lytic pores in the host cell membrane, playing a role in invasive infections.[8] During the exponential growth of *Staphylococcus aureus*, α-HL is secreted as a monomer, which oligomerizes into a heptameric transmembrane pore, causing osmotic cytolysis.[9]

Molecular docking studies have provided atomic details on protein-ligand interactions, which may enhance the knowledge of how compounds interact with the heptameric pore. Considering that furosemide is suggested as a potential therapeutic adjuvant for treating infected patients, the present study assessed its antimicrobial effect through the inhibition of the α-HL channel, which is a major virulence factor from *Staphylococcus aureus*.

MATERIALS AND METHODS

α-HL protein and other chemicals

The α-HL from wild-type *Staphylococcus aureus* was obtained from List Biological Laboratories. Solvent-free planar bilayer lipid membranes (PLM) with a capacitance of 40 pF were formed using the lipid monolayer apposition technique with diphytanoylphosphatidylcholine (DPhPC) in hexane at 25 ± 1 °C. Furosemide (4-chloro-2-[furan-2-ylmethylamino]-5-sulfamoyl benzoic acid) was purchased from Sigma, and DPhPC was purchased from Avanti Polar Lipids.

Single channel reconstitution in PLM

PLM were formed as previously described[10] by using two DPhPC monolayers. After membrane stabilization, a stock solution (0.1 to 0.4 μL) containing *α*-HL (0.07 mg/ml) was added to the *cis* compartment of the experimental chamber with the standard solution (4 M potassium chloride [KCl], 5 mM tris(hydroxymethyl)aminomethane, pH 7.5), resulting in a final concentration of ~2 ng/mL. All experiments were performed at room temperature (25 ± 2 °C) using the Axopatch 200A amplifier in voltage-clamp mode. The α-HL channels typically exhibit high conductance (~4 nS) and rarely switch to low conductance states at pH 7.5. After the ion channel was incorporated into PLM, anion inhibitors were added to the standard solution. All steps (from current measurement to data analysis) were performed using the equipment and software described in previous studies.[5,11,12]

Multiple channel reconstitution in PLM

PLM for multiple channel experiments were formed using the painting technique with a phosphatidylcholine/cholesterol mixture (1:1, w/w) across a ~0.3 mm diameter hole in a 25 mm-thick Teflon partition in a Teflon cell. The current was converted to voltage, filtered through an eight-pole Butterworth low-pass filter, and digitized at a sampling frequency of 0.5 kHz (for multiple channel experiments) using the Whole Cell Electrophysiology Program (V1.7b) or the Electrophysiology software.

Channels were formed by adding several microfilters of the α-HL stock solution (5 to 50 mg/mL) to the *cis* compartment of the chamber. The mean single-channel insertion current was 0.3 pA in the presence of calcium chloride (50 mM) and 40 mV potential. The potential was defined as positive when it was higher on the side of

protein addition. The α-HL channels typically remain in a high conductance and rarely switch to low conductance states at pH 7.5. The *t*-test was used to compare conductance in the GraphPad Prism software, and the statistical significance of mean differences was set at $p < 0.05$.

Molecular docking analyses

Docking experiments were conducted to predict furosemide position and orientation on the surface of α-HL channels and understand its atomic-level action mechanisms. Atomics coordinates for the α-HL heptameric channel were retrieved from Protein Data Bank (ID 7AHL).[13] The furosemide structure was obtained from PubChem, and its coordinates were built and minimized using the Avogadro software.[14] Molecular docking experiments were performed using the DockThor online platform,[15] focusing on the constriction region of the α-HL channel, which is probably the main region interacting with small ligands.[16] The output conformers from the software were ranked based on their increasing affinity for the channel, and the two conformers with the highest affinity were selected for further analysis.

RESULTS AND DISCUSSION

The ability of furosemide to block ion conductance was assessed using pores in artificial membranes due to the large single-channel conductance of the α-HL (~4 nS in 4 mM KCl) (Figure 1).

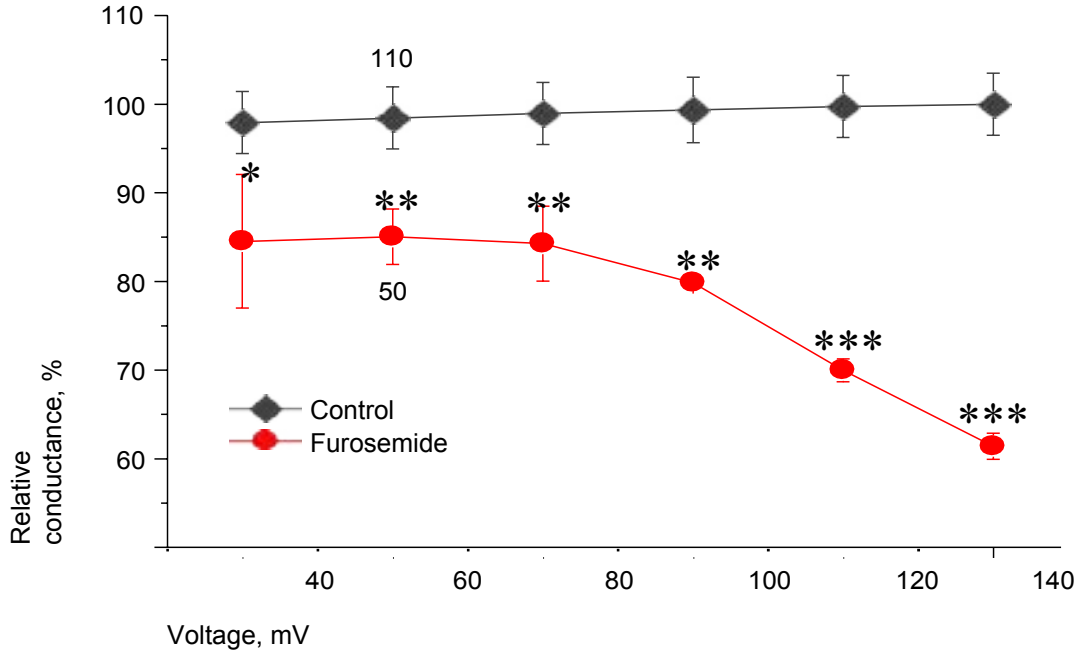

Figure 1. Voltage-dependent effect of inhibition from furosemide. The α-hemolysin channel conductance in the absence of anion inhibitors was considered 100% (control). The α-hemolysin channel conductance in the presence of 100 μM furosemide in the cis compartment was relativized to the control condition. Data presented as mean ± standard deviation for at least four experiments. *p < 0.05, **p < 0.01, ***p < 0.0001, t-test (control versus furosemide)

The relative conductance of the α-HL channel in the presence of furosemide was lower than in the standard solution and decreased with increasing transmembrane potential, indicating that its effect is voltage-dependent. The addition of 100 μM furosemide to the *cis* compartment of the membrane switched the channel to a closed state, similar to the "voltage-gated closed state" commonly observed for α-HL channels at ≥ 100 mV.

Figure 2 illustrates typical recordings of an ion current through a single α-HL channel. Even before the addition of compound 1, the conductance level of the single α-HL channels showed significant noise (Figure 2A, top track), which is characteristic of pores channels, exhibiting rapid flickering between open and fully closed conformations. The addition of furosemide to the *cis* compartment of the membrane (i.e., the toxin side) caused additional stepwise closures with an average duration of 6 ms (Figure 2B, middle).

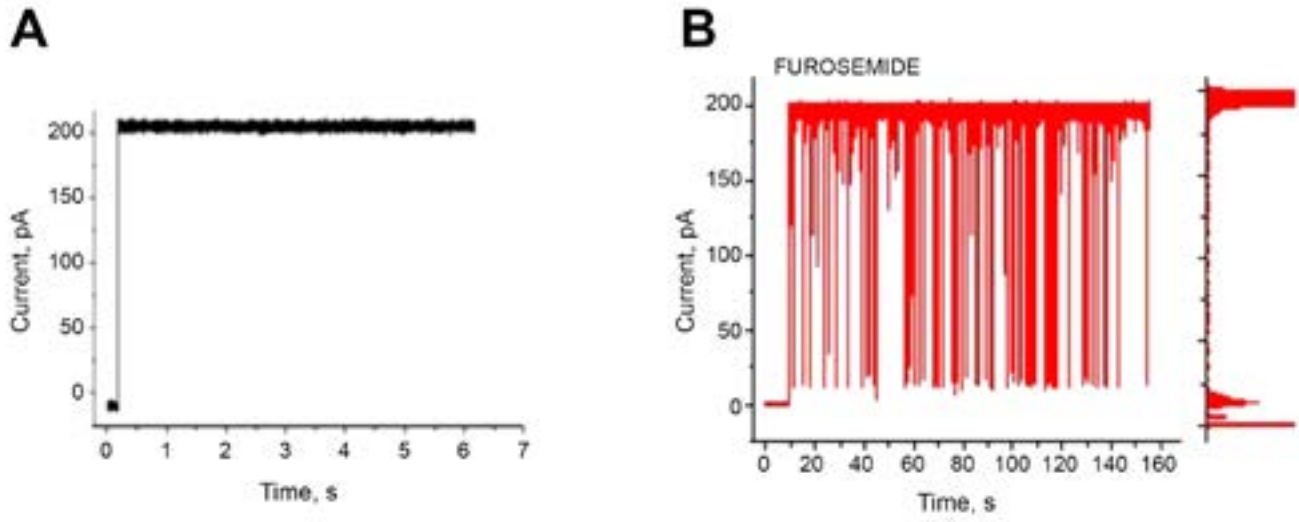

Figure 2. Effect of anion channel inhibitors on the α-hemolysin channel in planar lipid bilayer membrane. Ionic current through a single α-hemolysin channel in the absence of furosemide (A) and representative current recordings and all point current amplitude histograms illustrating the behavior of the α-hemolysin channel in the presence of 100 μM furosemide (B, red records) in the cis compartment. Voltage, +50 (upper traces) mV; filtering, 0.5 KHz; digitizing, 2.5 KHz.

These fluctuations were rapid transitions between the fully open and non-conducting states of the channel, and the blockade became frequent with 100 μM furosemide, similar to the voltage-gated closed state observed for α-HL channels at ≥ 50 mV (Figure 2B, bottom). The residual conductance in this closed state ranged between 1% and 15% of the total channel conductance. The current through a single α-HL channel remained stable, with no gating events observed at ≥ 100 mV.

Interestingly, the introduction of positive charges in the furosemide molecule enhanced its ability to block the α -HL channel on the *cis* compartment of the membrane. Unmodified drugs weakly bind to the heptameric α-HL channel when added to the *trans* (i.e., intracellular) compartment of the membrane. Also, furosemide presented similar effects to tamoxifen.[6]

The two furosemide conformers with the highest binding affinity to the α-HL channel presented binding energies of -6.766 and -6.634 kcal/mol, respectively. This binding showed the conformers were positioned in the same constriction region, occupying a small volume (Figure 3A). Furosemide binds to residues 113 and 147 of the α-HL channel, and four hydrogen bonds were observed with residue 147 near the constriction region of the channel (Figure 3B and 3C). Although these bonds stabilize furosemide within the nanopore lumen,[17] its polarity helps the solubilization within the solvent, justifying the rapid binding events observed in the experiments (Figure 2B).

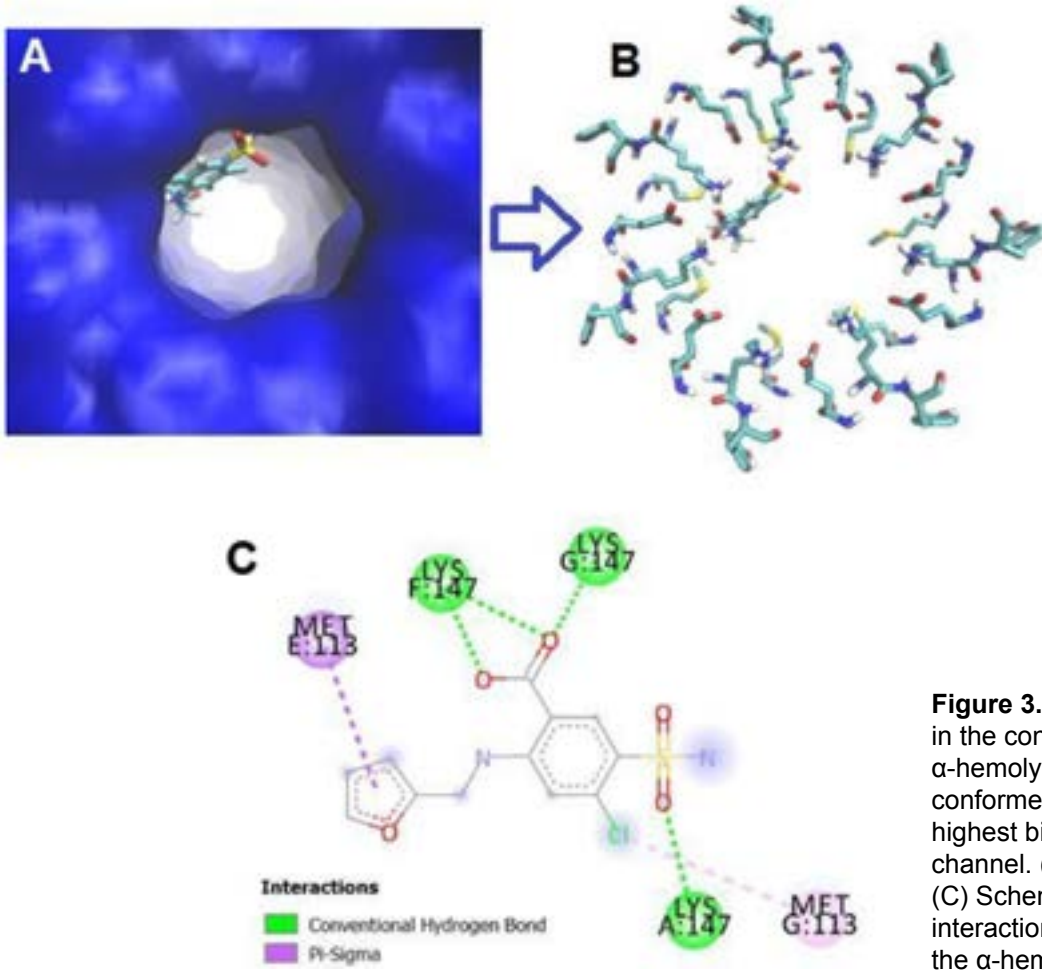

Figure 3. Illustration of furosemide in the constriction region of the α-hemolysin channel. (A) Docked conformer of furosemide with the highest binding energy coupled to the channel. (B) Licorice representation. (C) Schematic representation of interactions involving furosemide and the α-hemolysin channel.

The results of the present study suggested that furosemide blocked ion currents in the constriction region of the α-HL channel formed by *Staphylococcus aureus*. Thus, furosemide may be proposed as a potential therapeutic adjuvant for treating infected patients.

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DISTRIBUTION PATTERN OF ANASTOMOTIC VEINS OF LABBÉ AND TROLARD CONSIDERING THE LATERALITY AND SEX: AN ANGIOGRAPHIC STUDY

PADRÃO DE DISTRIBUIÇÃO DAS VEIAS ANASTOMÓTICAS DE LABBÉ E TROLARD EM FUNÇÃO DA LATERALIDADE E SEXO: UM ESTUDO ANGIOGRÁFICO

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ABSTRACT

INTRODUCTION: The superior (of Trolard; SAV) and inferior (of Labbé; IAV) anastomotic veins connect the middle cerebral vein to the superior sagittal and transverse sinuses, respectively. They are important for neurosurgery due to the high risk of injuries and the location in topographies of constant access. Consequences of injuries in SAV and IAV include cerebral edema, venous infarction, and hemorrhage. **AIM:** To investigate the distribution pattern of SAV and IAV in cerebral arteriographies considering the laterality and sex in cerebral arteriographies. **METHODS:** This cross-sectional and retrospective study was conducted at the Faculty of Medicine of Olinda. Twenty angiographies were analyzed as a pilot study for greater samples. Angiographies with lateral, oblique, and anteroposterior incidence angles and with a clear view of the cerebral venous anatomy were included. Data was analyzed using descriptive statistics. **RESULTS:** The IAV was prevalent on the right in females and the left in males. Meanwhile, the SAV was prevalent on the right in males and on the left in females. **CONCLUSION:** The present study described the distribution pattern of IAV and SAV considering the laterality and sex, relating to potential complications secondary to iatrogenic venous injury, which demonstrates the importance of knowing vascular structures and their neurosurgical implications.

Keywords: Cerebral veins; vein of Trolard; vein of Labbe; anatomical variations; neurosurgical anatomy.

RESUMO

INTRODUÇÃO: As veias anastomóticas superior (de Trolard) e inferior (de Labbé) são responsáveis por comunicar a veia cerebral média aos seios sagital superior e transverso, respectivamente. Elas são relevantes na área da neurocirurgia, pois são veias de alto risco para lesões em cirurgias devido à localização em topografias de acesso constante. Na literatura, são mostradas as consequências do acometimento dessas veias durante cirurgias, o que inclui edema cerebral, infarto venoso e hemorragia, dentre outras intercorrências. **OBJETIVO: I**nvestigar o padrão de distribuição da veia anastomótica de Labbé (VAL) e da veia anastomótica de Trolard (VAT) em exames de arteriografia cerebral em função da lateralidade e do sexo. **MÉTODOS:** Este projeto foi desenvolvido na Faculdade de Medicina de Olinda. Tratou-se de um estudo do tipo transversal, observacional e retrospectivo. Foram analisados 20 exames de angiografia como quantitativo inicial, tornando este trabalho um estudo piloto para análises posteriores com amostras mais significativas. Foram incluídos exames de arteriografias com avaliação da drenagem venosa do cérebro e excluídos os exames de arteriografia cerebral que não possuíam as três incidências utilizadas no exame. Para a análise dos dados, foi utilizada estatística descritiva de acordo com cada objetivo proposto. **RESULTADOS:** A VAL foi mais prevalente à direita no sexo feminino, enquanto no sexo masculino foi mais prevalente à esquerda. A VAT foi mais prevalente à direita no sexo masculino, enquanto

no sexo feminino foi mais prevalente à esquerda. **CONCLUSÃO:** O presente estudo descreveu o padrão de distribuição das VAL e VAT em função da lateralidade e do sexo, relacionando os achados com potenciais complicações secundárias à lesão venosa iatrogênica, demonstrando a importância do conhecimento da anatomia destas estruturas vasculares e suas implicações cirúrgicas.

Palavras-chaves: Veia cerebral; veia de Trolard; veia de Labbé; variações anatômicas; anatomia neurocirúrgica.

INTRODUCTION

The superior anastomotic vein (of Trolard; SAV) and the inferior anastomotic vein (of Labbé; IAV) connect the middle cerebral vein to the superior sagittal and transverse sinuses, respectively. These anastomotic veins are at high risk of injuries due to their location in constantly accessed topographies in neurosurgery[1].

The consequences of injuries in SAV and IAV include cerebral edema, venous infarction, hemorrhage, and ischemic complications in the central region of the cerebral hemisphere and temporal lobe. Additionally, thrombosis after prolonged or forced retraction is demonstrated, highlighting the importance of anatomical understanding [1,2].

A study using computed tomography observed distinct patterns of venous drainage due to insufficient venous flow regarding SAV and IAV, calling neurosurgeons to assess patterns of venous drainage[3,4].

Considering the high prevalence of anatomical variations in blood vessels, knowledge is crucial to the execution of surgical procedures, which provide information for radiological evaluations. Therefore, the present study aimed to investigate distribution patterns of SAV and IAV in cerebral arteriographies considering the laterality and sex.

METHODS

This study was conducted at the Faculdade de Medicina de Olinda and comprised the analysis of 20 cerebral digital angiography. The inclusion criteria were angiographies with lateral, oblique, and anteroposterior incidence angles and a clear view of the cerebral venous anatomy.

This cross-sectional and retrospective study was conducted between April 2021 and April 2022 with a convenience sample. Angiographies were selected, and the angioarchitecture of the deep cerebral venous system was analyzed using the Radiant DICOM software (Medixant, Greater Poland/Poland). Numbers of SAV and IAV were assessed considering the laterality and sex.

Data were analyzed using descriptive statistics by the Statistical Package for the Social Sciences (SPSS), version 17.0 (IBM Corp., NY/USA). This study was approved by the research ethics committee of the Medicine School of Olinda (no. 43998421.0.0000.8033).

RESULTS

Cerebral arteriographies presented 92.86% of the IAV and 50.00% of the SAV on the right. On the left, the IAV was observed in 92.86% and SAV in 64.29% of participants. Thus, the IAV did not show statistical difference between sides, whereas the SAV was significantly more prevalent on the left.

The prevalence of IAV and SAV in female participants on the right was 100% and 37.50%, respectively. On the left, the prevalence was 87.50% for the IAV and 75.00% for SAV. Data indicated an increased prevalence of the IAV on the right and the SAV on the left.

Regarding male participants, the prevalence of IAV and SAV on the right was 83.33% and 66.67%, respectively. On the left, the prevalence was 100% for IAV and 50.00% for SAV. Data indicated a greater prevalence of the IAV on the left and the SAV on the right.

The pattern of anastomotic veins was the opposite regarding sexe. While the IAV was prevalent on the right in females and the left in males, the SAV prevailed on the left in females and on the right in males.

In summary, the IAV was more prevalent (100.00%) on the right in females and the left

in males. Conversely, the SAV was more prevalent on the left in females (75.00%) and on the right in males (66.67%) (Figure 1).

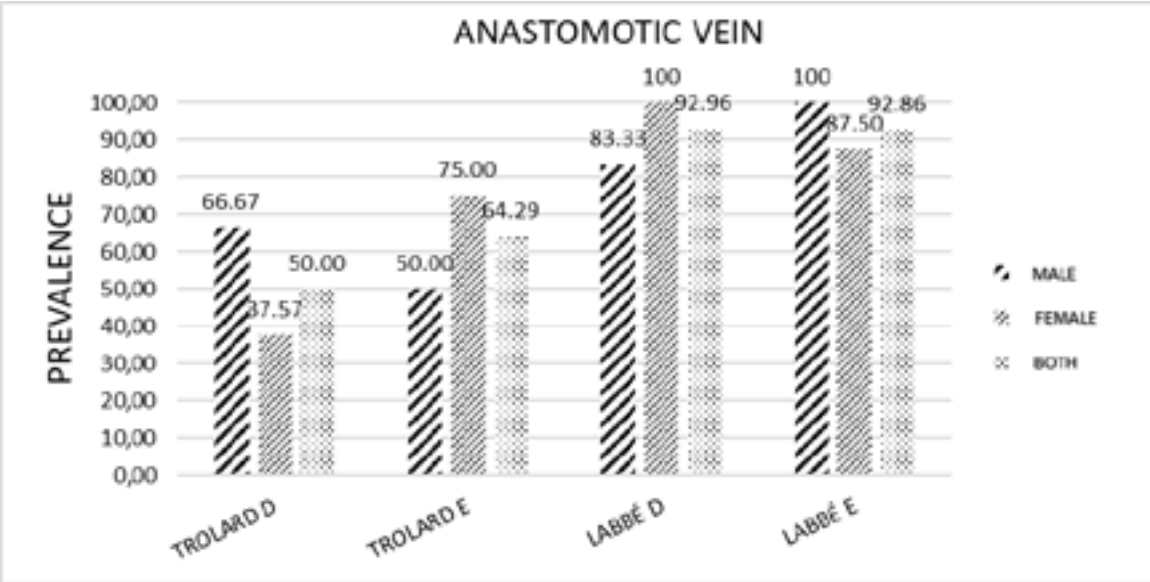

Figure 1. Distribution of anastomotic veins of Labbé and Trolard considering the laterality and sex.

DISCUSSION

Neurological consequences due to injuries to IAV and SAV during surgeries are documented in the literature, such as cerebral edema, venous infarction, hemorrhage, and ischemic complications in the central region of the cerebral hemisphere and temporal lobe. In addition, thrombosis may occur after prolonged or forced retraction of the IAV[2,3,7].

A study using computed tomography described distinct patterns of venous drainage based on impaired venous flow involving IAV and SAV[4]. Therefore, the present study aimed to analyze and associate these patterns considering the laterality and predominance between sexes. This highlights the importance and originality of the study, allowing a new approach with detailed information and analyses.

The main patterns observed in the previous study were (i) absence of connection between IAV, SAV, and the superficial middle cerebral vein; (ii) double connection (superficial middle cerebral vein connected to the IAV, superficial middle cerebral vein connected to the SAV, and connection between IAV and SAV); and (iii) triple connection (all three veins connected) [4]. Considering these results, the present study focused on differences between laterality and sexes.

Furthermore, findings observed during arteriography analysis were the absence of predominance between the right and left sides regarding the IAV and the prevalence of the SAV on the left. Additionally, the IAV was prevalent on the right and the SAV on the left in females; in males, the opposite was observed, the IAV was prevalent on the left and the SAV on the right.

The prevalence of IAV and SAV was accessed in a study using magnetic resonance, which showed prevalence of the IAV, followed by the middle cerebral vein and the SAV. However, the simultaneous presence of the three veins was higher than the presence of the IAV alone. Comparing these data with the arteriography analysis reaffirmed the prevalence of the IAV in exams and in both sexes, which reinforced the high risk of injuries during neurosurgical procedures[5].

The proportion of IAV and SAV corroborates the literature. However, the number of arteriographies did not allow for an inferential statistic, and a larger sample is suggested for significance tests.

This analysis reinforces the importance of pre-surgical mapping of IAV and SAV, considering the laterality and sex. These measures might reduce the risk of venous congestion or infarction due to surgical maneuvers that may cause vein stretching or stenosis. Those benefits mainly concern the IAV, the most frequent-

ly injured vein in neurosurgeries. Such injuries include permanent lesions leading to aphasia, hemiplegia, disorientation, loss of consciousness, and death, enhancing the necessity for neurosurgeons to evaluate patterns of venous drainage, highlighting the relevance of the present study[6,8].

CONCLUSION

Studies on the superficial cerebral venous system lack attention in the neurosurgery area. An extensive literature review showed limited systematic description and numeric distribution of IAV and SAV. Thus, the present study aimed to provide data for the practical planning of surgical procedures, presenting a systematic description and numeric distribution of IAV and SAV and the main differences between laterality and sex, using cerebral arteriography. Understanding the unique pattern of venous drainage present in each individual should be considered during the planning of neurosurgeries.

This study also described possible secondary complications due to venous iatrogenic lesions, which may impair the blood supply in brain regions and cause cerebral ischemic events.

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THE IMPORTANCE OF EARLY DIAGNOSIS OF RHEUMATOID ARTHRITIS TO REDUCE THE RISK OF UNFAVORABLE OUTCOMES: A CASE REPORT

A IMPORTÂNCIA DO DIAGNÓSTICO PRECOCE DA ARTRITE REUMATÓIDE PARA MINIMIZAR AS CHANCES DE DESFECHOS DESFAVORÁVEIS: RELATO DE CASO

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ABSTRACT

Rheumatoid arthritis (RA) is characterized by inflammation of the synovial tissue in multiple joints, leading to tissue destruction, pain, and deformities. This case report aimed to describe the case of a 68-year-old female patient with polyarthritis diagnosed 17 years ago. The physical examination revealed joint deformities in elbows and knees, decreased range of motion in elbows and hands, and edema in ankles and knees. Complementary exams showed positive rheumatoid factor and x-ray with severe joint destruction. These results, combined with clinical history and physical examination, lead to the diagnosis of RA. Early diagnosis of RA is crucial to avoid physical and psychosocial damage, medication costs, joint prostheses, and work absenteeism.

Keywords: Delayed diagnosis; patient harm; rheumatoid arthritis; rheumatic disease.

RESUMO

A artrite reumatoide (AR) é caracterizada pela inflamação do tecido sinovial de múltiplas articulações que leva à destruição tecidual, dor e deformidades. É apresentado o relato de caso de uma paciente de 68 anos que possuía história de poliartrite havia 17 anos e cujo exame físico revelava deformidades articulares de cotovelos e joelhos, bloqueio parcial da mobilidade de cotovelos e mãos, além de edema nos tornozelos e joelhos. Exames complementares evidenciaram fator reumatoide (FR) positivo, e a radiografia indicou severa destruição articular, que, combinados com a história clínica e a avaliação corporal, permitiram o diagnóstico de AR. A fim de evitar danos físicos, psicossociais e gastos com medicações, próteses articulares e afastamentos laborais, o diagnóstico precoce da AR é imperativo.

PALAVRAS - CHAVE: Artrite reumatoide; doenças reumáticas; dano ao paciente; diagnóstico tardio.

INTRODUCTION

Rheumatoid arthritis (RA) is characterized by inflammation of the synovial tissue in multiple joints, leading to tissue destruction, pain, deformities, and reduced quality of life[1]. Although its etiology is complex and largely unknown, studies suggest that genetic and environmental factors influence its pathogenesis[2]. Hands and wrists are the most frequently affected joints, causing pain and morning stiffness when inflamed. The articular cartilage is destroyed as the disease progresses, and patients may develop deformities. Due to the strong genetic influence, family members of patients with RA are considered a risk group for developing the disease, especially in severe forms[3].

The early stage of RA can be considered a therapeutic window since the appropriate therapy

can modify the disease course, resulting in a better prognosis than diagnosis at later stages[4]. To address this issue, the 1987 classification criteria of the American College of Rheumatology, which includes suboptimal characteristics of early RA (e.g., x-ray changes [erosions] and rheumatoid nodules), were reevaluated, leading to a new American College of Rheumatology/ European League Against Rheumatism (ACR-EULAR) criteria in 2010, which focused on the early phase of the disease[5]. The new criteria encompass four domains: joint involvement, serology, duration of symptoms, and acute-phase reactants. Items in each domain are scored, and a minimum score of six is required for classification. Additionally, the criteria can be applied to any patient if two requirements are met: evidence of active clinical synovitis in at least one joint during the physical examination and the absence of other diagnoses that better explain the synovitis[5,6]. The number of affected joints may use ultrasonographic imaging methods and magnetic resonance imaging if doubts remain[5].

Non-pharmacological treatments (e.g., occupational therapy, physical therapy, psychosocial support, and patient and family education) are crucial for managing patients with RA[7]. From a pharmacological perspective, the Brazilian Society of Rheumatology recommends the use of disease-modifying antirheumatic drugs (DMARDs), such as methotrexate and leflunomide, which can be combined with biological disease-modifying antirheumatic drugs and Janus kinase inhibitors[6].

Despite the high potential for disability, the course of RA can be modified with early diagnosis and proper management, reducing physical, social, and occupational damage that leads to prolonged periods of work absenteeism that directly (or indirectly) impact socioeconomic costs[8]. However, the heterogeneity of the clinical manifestations hamper the early diagnosis of RA, which delays the initiation of treatment.

The present study aimed to highlight the importance of early diagnosis of RA through a case study of a patient with a delayed diagnosis who developed irreversible consequences, joint destruction, and reduced quality of life.

CASE REPORT

MJS is a 68-year-old female retired dressmaker. She was referred to the Rheumatology Clinic at the School Clinic of the Faculty of Medicine of Olinda (FMO) due to polyarthralgia and a diagnosis of “arthritis” and “osteoarthritis” made 17 years ago, occasionally treated with prednisone. After tapering off the medication in February 2021, she attended her first consultation with the specialist without any medication. The patient reported that the pain started insidiously, associated with swelling in the elbow, knee, and ankle joints. The pain had worsened over the past three months, resulting in an inability to walk due to heat and a swollen joint. Regarding factors that reduced or increased the arthralgia, the pain was partially reduced with the use of dipyrone and prednisone but worsened after physical efforts (e.g., household chores). She also reported joint stiffness after resting for more than 30 minutes, which improved throughout the day, and denied experiencing cramps, muscle weakness, or other general symptoms.

During the physical examination, the right elbow and knee and the left ankle presented edema with limited extension in the right elbow and knee; additionally, edema and Bouchard’s nodes were observed on the third proximal middle finger of the right hand (Figures 1 to 4). On palpation exam, she reported pain in the knees, right elbow, shoulders, wrists, and left ankle. On the visual analog scale for pain, she rated it as eight out of ten over the past week. The squeeze test on the left foot was positive.

The x-ray of the hands and wrists revealed reduced joint space, cortical erosions, and the presence of bone cysts. The laboratory tests showed rheumatoid factor (RF) at 45 IU/mL (reference value [RV]: 20 IU/mL); C-reactive protein (CRP) at 53 mg/L (RV: 6 mg/L); 25-Hydroxyvitamin D at 25 ng/mL (RV: 20 ng/mL). Treatment was started with methotrexate 15 mg per week, prednisone 5 mg (half a tablet in the morning), folic acid 5 mg (to be taken the day after methotrexate), calcium carbonate, and vitamin D 50,000 IU.

Considering the diagnostic hypothesis of RA, the 2010 ACR/EULAR criteria were applied, resulting in nine points (involvement of more than ten joints, positive RF, duration longer than six weeks, and altered acute-phase reactants) [9]. At a follow-up appointment three months after starting the treatment, the patient reported improvement in pain (six out of ten over the past

week) and joint swelling, and CRP was normalized. The patient continued with quarterly follow-up.

This case report followed the guidelines of Resolution 466/12 and Circular Letter No. 166/2018 of the National Health Council and was approved by the research ethics committee of the FMO (no. 5.151.987).

Figure 01: Deformity of the right elbow, with limited joint extension and swelling in the 3rd proximal interphalangeal joint of the right hand.

Figure 02: Swelling of the knees (right greater than left) and ankles (left greater than right) with decreased range of motion.

Figure 03: X-ray of the hands showing joint space narrowing and erosions in the carpus; the red arrow indicates narrowing space and erosions in the 3rd proximal interphalangeal joint of the right hand.

Figure 04: X-ray of the elbows showing joint destruction in the right elbow (arrow) with significant decreased range of motion.

DISCUSSION

Advances in the diagnosis of RA facilitate early detection and monitoring of disease activity, contributing to appropriate and effective treatment[2]. Despite these advances and the available treatments, many patients affected by autoimmune rheumatic diseases progress to unfavorable outcomes. These outcomes can be influenced by intrinsic (e.g., genetic predisposition, sex, and family history) or extrinsic factors (e.g., delayed recognition by general physicians and difficulty accessing specialists)[8].

The duration of symptoms required to define early RA varies according to the literature[5]. Historically, RA was considered "early" if symptoms lasted less than five years; however, this period was reclassified to less than two years in the 1990s, emphasizing the first 12 months of clinical manifestations, which classify RA as "very early"[6]. The diagnostic criteria are based on clinical findings and complementary exams, including the disease progression, the presence of autoantibodies RF, anti-citrullinated peptide antibodies, elevated inflammatory markers (CRP and erythrocyte sedimentation rate), and compatible changes in imaging exams[5]. Besides the articular symptoms, the disease can cause extra-articular manifestations, such as rheumatoid nodules, interstitial lung disease, or cutaneous vasculitis[10]. In this case report, the lack of early diagnoses led to joint destruction with a significant limitation in the range of motion and walking ability and reduced quality of life. Patients with RA can also develop cardiac complications and infections, which increase mortality rates[11]. Estimates suggest a poor long-term prognosis, as 80% of affected patients will have some degree of disability after 20 years, and their life expectancy is reduced by 3 to 18 years[12].

The European League Against Rheumatism considers the risk factors for unfavorable progression listed in Chart 1. Monitoring these factors is crucial for identifying patients at higher risk of severe and debilitating disease progression and indicates a need for more rigorous clinical surveillance and more aggressive treatment[6].

Chart 01 - Factors for a bad prognosis in rheumatoid arthritis
Persistently moderate or high disease activity despite DMARD treatment
Presence of initial erosions
Failure of two or more csDMARDs
Presence of RF* or anti-CCP** (or both), especially at high levels
High number of swollen joints
High levels of acute-phase reactants

*rheumatoid factor ** anti-cyclic citrullinated peptide

Given the need for early identification and monitoring of poor prognosis factors in RA, access to specialist physicians, especially rheumatologists, must be ensured for the population. However, the training of general physicians is essential since they are the first line of healthcare assistance for the population. They need to be capable of recognizing the disease, initiating early therapy, or referring patients as needed.

FINAL CONSIDERATIONS

RA is an autoimmune polyarthritis that can lead to joint destruction and significantly impact quality of life. Despite advances that facilitate early diagnoses, some patients still progress to unfavorable outcomes, possibly due to modifiable factors, such as difficulties faced by general physicians in establishing the diagnosis, limited access to specialists, and failure to recognize poor prognosis factors. This aspect also included the lack of individualized treatment based on local realities and difficulties accessing complementary exams. However, factors inherent to the patient (e.g., genetic predisposition) influence the outcome.

Searching for goals that enhance early diagnosis of RA will contribute to reducing morbidity in affected patients.

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ANGIOARCHITECTURE OF THE MIDDLE MENINGEAL ARTERY: AN INTEGRATIVE REVIEW

Angioarquitetura da artéria meníngea média: uma revisão integrativa da literatura

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ABSTRACT

Introduction: The middle meningeal artery is the most important artery of the dura mater and has a complex embryological origin, resulting in anatomical variations. Knowledge of its anatomy, morphometric aspects, and anatomical variations is important in surgeries and in understanding clinical conditions. **Objective:** To integratively review the angioarchitecture, organization, variations, and morphometric aspects of the middle meningeal artery. **Methods:** An integrative review was conducted following the Preferred Reporting Items for Systematic Reviews and Meta-Analyzes (PRISMA) recommendations. The search was conducted in PubMed, Virtual Health Library, and MEDLINE databases considering the following terms: middle meningeal artery, anatomical variations, and anatomy. This study considered articles published in English. **Results:** After duplicate removal, 89 studies were identified and 10 met the eligibility criteria and were included in the synthesis. **Conclusion:** A relationship was observed between the aspects of the middle meningeal artery and various clinical conditions, with anatomical variations being the conditions best documented in the literature. Furthermore, morphometric data on the middle meningeal artery is scarce, reiterating the importance of promoting studies to understand this vessel.

Keywords: Middle meningeal artery, anatomy, variations, morphometric aspects.

RESUMO

Introdução: A artéria meníngea média, uma das mais importantes artérias da dura máter, possui uma complexa origem embriológica. O conhecimento de sua anatomia, aspectos morfométricos e variações anatômicas são importantes em situações cirúrgicas e no entendimento de condições clínicas. O presente artigo buscou revisar de maneira integrativa a organização anatômica, as variações e os aspectos morfométricos da artéria meníngea média. **Objetivo:** Revisar, de maneira integrativa, a angioarquitetura da artéria meníngea média. **Métodos:** Tratou-se de uma revisão integrativa da literatura redigida conforme às recomendações do *Preferred Reporting Items for Systematic Reviews and Meta-Analyzes* (PRISMA), a qual analisou estudos publicados na língua inglesa tendo como referência as bases de dados PubMed (*Public Medline* ou *Publisher Medline*), Biblioteca Virtual em Saúde e MEDLINE empregando os seguintes descritores: *middle meningeal artery*, *anatomical variations* e *anatomy*. **Resultados:** Dos 89 estudos identificados após a remoção de duplicatas, dez atingiram os critérios de elegibilidade e foram incluídos na síntese. **Conclusões:** Observou-se uma grande relação entre os aspectos desse segmento arterial e uma variedade de condições clínicas, sendo as variações anatômicas as condições mais bem documentadas na literatura. Por outro lado, existe uma escassez de informações no que diz respeito às informações sobre dados morfométricos desse vaso sanguíneo, fato que reitera a importância da promoção de estudos direcionados ao entendimento desse vaso.

Palavras chaves: Artéria meníngea média, anatomia, variações, aspectos morfométricos.

INTRODUCTION

The middle meningeal artery (MMA) is one of the most important dural arteries, irrigating more than two-thirds of the dura mater. MMA originates from the internal maxillary artery, one of the largest branches of the external carotid artery, and is clinically and surgically relevant due to its intimate relationship with the cranium.[1,2]

When entering the cranial fossa through the spinous foramen, the MMA laterally crosses the bone crest and curves above the upper wing of the sphenoid, bifurcating into anterior and posterior branches. MMA presents a complex embryological origin, which occurs through the stapedial artery early in embryological development. The complex nature of this process allows for the appearance of several anatomical variations and anastomoses.[2]

Morphological variations are of clinical importance, evident in cranium base fractures, epidural hematomas, and during endovascular and surgical interventions.[3] Therefore, this study aimed to summarize the literature on the anatomical organization of the meningeal artery, its variations, and morphometric aspects due to the significant clinical and surgical importance of this information.[4]

METHOD

This study is an integrative literature review considering studies with various methodologies investigating the anatomical and morphometric aspects and the prevalence of anatomical variations of MMA. The main objective of this review was to summarize the morphometric and anatomical aspects of the MMA and its relationship with clinical and surgical practice.

This review was conducted and reported according to the PRISMA recommendations.[5] The search was conducted in November 2021 in the PubMed, Virtual Health Library, and MEDLINE databases. The following terms were used: “middle meningeal artery,” “anatomical variations,” and “anatomy”. The search was performed by combining the descriptors with the Boolean operator “AND.”

The inclusion criteria comprised i) articles; ii) with full text available for free; iii) addressing the specific theme of anatomical and morphometric aspects and variations of the MMA; and iv) written in English. Studies not addressing the guiding question and duplicates were excluded, according to Figure 1.

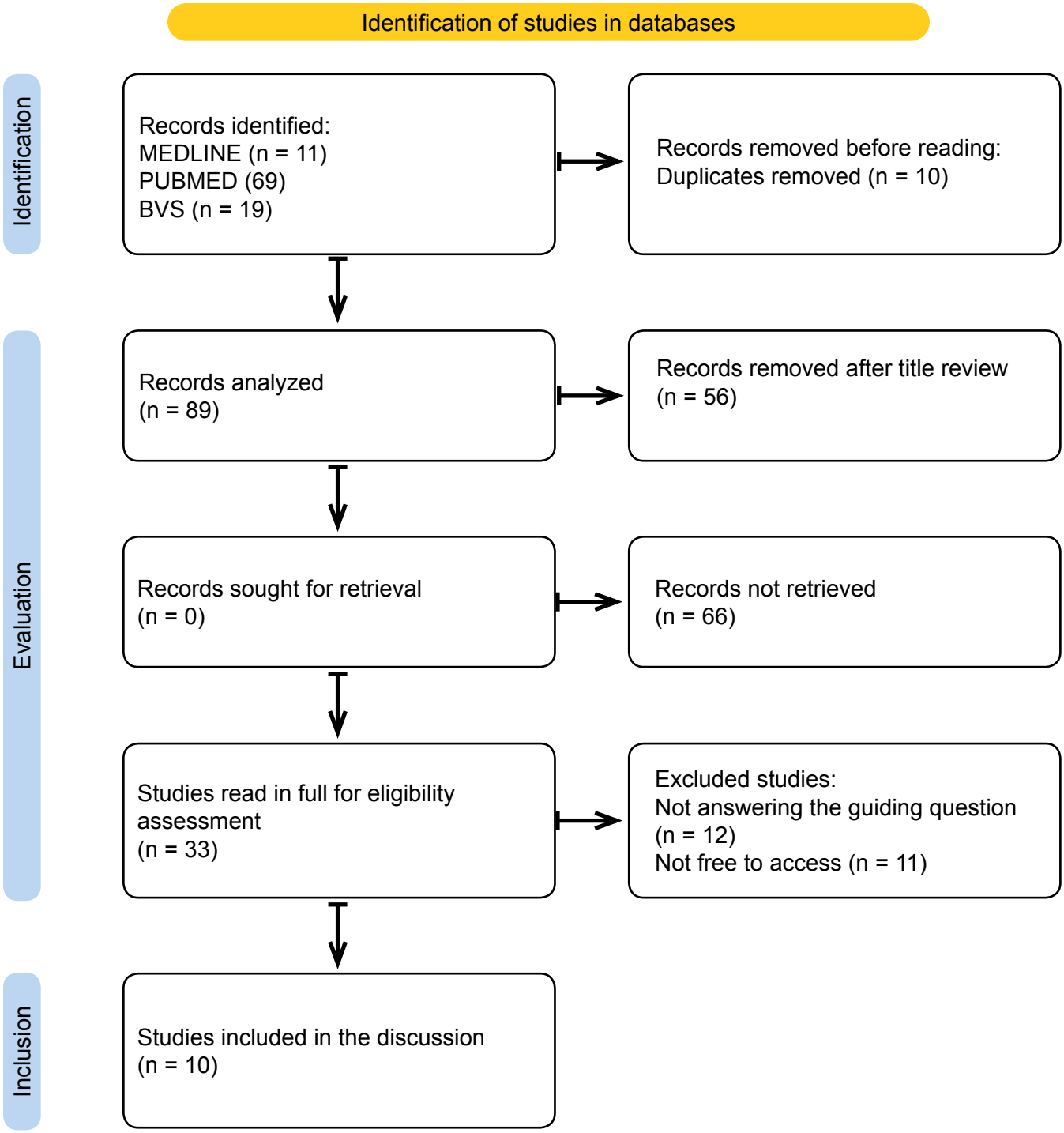

Figure 1. The selection of studies flowchart according to the PRISMA.

RESULTS

Table 1. Studies from search refinement.

	Title	Author	Objectives	Conclusions
1º	Middle Meningeal Artery: Anatomy and Variations.	Bonasia *et al.* (2020)	To review and analyze the anatomical aspects and variation of the MMA.[1]	The MMA has numerous anatomical variations due to its complex embryological origin.[1]
2º	Neuroanatomy, Middle Meningeal Arteries.	Natali *et al.* (2020)	To review the neuroanatomy of the MMA, citing clinical repercussions related to this arterial segment.[2]	Understanding the vascular anatomy of MMA is important in clinical practice for neuroradiologists and neurosurgeons due to its relationship with pathologies (e.g., epidural and chronic hematomas, and meningiomas).[2]
3º	Abnormality of the Foramen Spinosum due to a Variation in the Trajectory of the Middle Meningeal Artery: A Case Report in Human.	Ellwanger *et al.* (2013)	To report the occurrence of an anatomical abnormality of the spinous foramen due to variations in the trajectory of the MMA, and review aspects of the MMA.[3]	Many anomalous origins of the MMA have been observed in the literature. In this study, the right foramen ovale was significantly larger than the left, except for an unusual form of the spinous foramen. This difference might be explained by the MMA widening the foramen ovale, on specific occasions,.[3]
4º	Morphometric analysis of the middle meningeal artery organization in humans-embryological considerations.	Harthmann *et al.* (2013)	To analyze the morphometric aspects and embryological considerations of the MMA.[4]	Important morphometric parameters of the MMA were demonstrated, including the difference between the right and left sides in the length of the parietal branch of the MMA.[4]
5º	Morphometry of organization of middle meningeal artery through the analysis of bony canal in human's skull: A clinical-anatomical and embryological insight.	Honnegowda *et al.* (2019)	To identify the course of the bone canals of the MMA and their morphometric and structural aspects to allow safer explorations during surgeries.[6]	A significant difference was identified between the right (12.8 ± 4.5 mm) and left (13.7 ± 9.3 mm) length of the bone canal produced by the MMA. Furthermore, the bone canal was always located around the pterion, posterior to the coronal suture, and inferior to the temporal line.[6]

6º	Middle meningeal artery arising from the basilar artery: report of a case and its probable embryological mechanism.	Kumar S *et al (2014)*	To report and describe a case of anomalous origin of the MMA.[7]	Despite the rarity of the aberrant origin of the MMA from the basilar artery, these variations demonstrate the importance of planning any neurovascular procedure to reduce the risk of complications.[7]
7º	Bony Tunnel Formation Associated with the Distal Segment of the Frontal Branch of the Middle Meningeal Artery.	de Campos D *et al* (2019)	To investigate the incidence of a bone tunnel originating from the distal segment of the frontal branch of the MMA in the adult cranium.[8]	A higher prevalence of the bone tunnel formed by the distal segment of the MMA on the left side of the cranium (about 5.88%) was identified compared with the right side (about 1.18%).[8]
8º	Variations of the ophthalmic and middle meningeal arteries: relation to the embryonic stapedial artery.	Dilenge D *et al* (1980)	To analyze the relationship between the stapedial embryonic artery and the development of variations of the ophthalmic and MMA.[9]	Several anomalies were identified in the MMA and ophthalmic artery, proving the importance of knowledge of these structures for medical practice.[9]
9º	Normal craniovascular variation in two modern European adult populations.	Eisová, S *et al* (2019)	To analyze the set of craniovascular impressions in two populations of European dried cranium with different morphological characteristics.[10]	Geographic and genetic factors may influence macroanatomic characteristics (e.g., anteroposterior vascular distribution). Endocranial foramina is influenced by morphology. Among the functional applications of these variations, the involvement with endocranial thermal regulation is highlighted.[10]
10º	The foramen spinosum: a landmark in middle fossa surgery.	Krayenbühl N *et al* (2008)	To analyze the anatomical and vascular relationships of the foramen spinosum and define an external framework for its early identification.[11]	The spinous foramen, through which the MMA, middle meningeal veins, and other structures flow, is an important anatomic landmark in surgeries of the cranial fossa. Knowledge of the anatomical variations of this structure may help to identify and preserve important neurovascular structures during surgeries.[11]

Source: the authors. MMA: middle meningeal artery.

\DISCUSSION

Many authors highlight the clinical importance of understanding the anatomy, variations, and morphometrics of the MMA.[2,3,4] Studies included different perspectives, such as the approach and relationships of MMA with nearby structures (e,g.,foramen spinosum) and its complex embryological origin, widely discussed in the literature.[3,11]

Natali *et al*. (2021) showed the importance of knowing the angioarchitecture of MMA for treating pathologies of the dura mater and for access in otorhinolaryngological surgeries. Anomalous origins of the MMA (origin from the basilar artery, internal carotid artery, and occipital artery) may have clinical implications and complex embryological explanations.[1]

From its origin in the internal maxillary artery, the MMA enters the cranial fossa through the spinous foramen, crosses the bone crest laterally, and curves anteriorly above the superior wing of the sphenoid. Before bifurcating into anterior and posterior divisions, the MMA emits the petrous and cavernous branches that irrigate the dura mater of the temporal fossa. The petrous branch runs through the petrous apex and irrigates the dura mater of this region and the upper part of the tympanic cavity. The cavernous branch irrigates the lateral wall of the cavernous sinus. In addition, the MMA supplies most of the dura mater of cranial convexity, and its anterior and posterior divisions still contribute to vascularizing the superficial half of the sickle of the brain.[1]

Bonasia *et al.,* (2020) reported the origin of the MMA, which develops similarly to other vessels via angiogenesis and is influenced by vascular endothelial growth and other growth factors. During MMA development, it is closely related to the stapedial system, which is present in the embryonic phase and usually degenerates in the tenth week of embryonic development.

Because of this complex origin, many anatomical variations and anastomoses may be observed. Surgical considerations involving MMA highlight the formation of epidural hematomas due to the close relationship of the meningeal vessels with the cranial vault, which may fragilize them in situations of injury.[2]

Ellwanger *et al.* (2013) described a case of morphological abnormality of the spinous foramen resulting from variations in the MMA course and showed the prevalence of anatomical variations of the blood vessel. Clinical repercussions related to the MMA highlight that 85% of the bleeding sources causing epidural hematomas are due to this vessel.[3]

Hartmann da Silva *et al*. (2013) studied morphometric aspects of the MMA and showed the scarcity of morphometric data in the literature. No significant differences were found between the morphometric measurements of the left and right sides of the MMA, except for the length of the parietal branch. The study highlights the importance of statistical analysis of these data, which may elucidate approaches in surgery and radiology. Among the clinical considerations, the study shows that MMA might be related to the development of pain in migraine crises, resulting either from neurogenic inflammation or cranial vasodilation.[4]

When analyzing the morphometric organization of the MMA, Page *et al*. (2021) identified significant differences between left and right-side parameters (extension of the frontal branch, length of the parietal branch, and length of the bone tunnel). Also, no variations were identified in the measurements of the angle and dimension of the main trunk of the artery and between the angle formed by the frontal and parietal branches. The study

evidenced the lack of information related to the morphometry of the MMA.[6]

Kumar *et al.* (2012) reported an anomalous origin of the MMA from the basilar artery and the importance of this arterial segment in various devascularization neoplastic processes of the dura mater and the healing of dural arteriovenous fistulas. Some factors of the MMA genesis were addressed.[7]

The stapedial artery, from which MMA originates, has three branches distributed around the divisions of the trigeminal nerve: mandibular, maxillary, and supraorbital. The internal maxillary artery, originating from the external carotid artery, is incorporated into the trunk of the maxillary and mandibular branches of the stapedial artery. The proximal part of this union forms the root of the MMA, and the distal part derives from the supraorbital artery mentioned above.

During embryological development, the stapedial artery regresses and its remnants contribute to the formation of other branches, which predisposes to variations in the angioarchitecture of the MMA. In addition, anomalous origin from the basilar artery is extremely rare. Knowledge of this anatomical variation is crucial for planning neuroendovascular procedures.[7]

Campos *et al.* (2018) reported a rare formation of a bony path related to the distal segment of the frontal branch of the MMA, revealing higher predominance on the left side (5.88% of the cranium studied) than the right side (1.18%). The lack of anatomical knowledge about this arterial segment, which is predominantly periosteal, might be a hindering factor during surgeries due to its location and anatomical relationships.

A correlation between the morphological characteristics of the MMA and the shape of the cranium was described in the literature, which makes the study a possible tool to predict variations in the MMA. In addition, a lack of information regarding the anatomical variation studied was identified in the literature.[8] Dilenge et al. (1980) reaffirmed the importance of knowledge of the anatomy of arteries, such as the MMA and ophthalmic artery, in angiographic studies for the resolution of clinical problems.[9]

Vascular trait analysis may be useful in forensic science, biological anthropology, evolutionary studies, and medicine. MMA development increases in the first and second years of life and reaches a stable morphology around the fifth and sixth years; however, MMA becomes fully functional only in adult life. The main branches of these vessels divide into smaller branches, apparently following geometric patterns, which may be partially identified in the cranial impressions.[10]

Krayenbühl *et al.* (2008) studied the spinous foramen, an important anatomic landmark in microsurgery, and showed that understanding its anatomical variations and the surrounding areas is crucial for preserving neurovascular structures during procedures in the median fossa. The spinous foramen size varies greatly and may be attached to the sphenomandibular ligament, a remnant of the first brachial arch. Variations in this site might be important for the vascular supply of the dura mater due to its close relationship with the MMA, and in anomalous cases, the artery may travel through different locations.[11]

CONCLUSION

The knowledge of the angioarchitecture of the MMA in the clinical, surgical, and scientific environment is important. A relationship

was observed between the aspects of this arterial segment and various clinical conditions, with anatomical variations being the most widely documented in the literature. Also, a lack of information regarding the morphometric data of MMA may hinder the planning of surgeries and reinforce the importance of further studies to understand this vessel, which is essential for dural irrigation.

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DIAGNOSIS AND MANAGEMENT OF WILMS TUMOR IN CHILDREN

Diagnóstico e manejo do Tumor de Wilms na população infantil

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ABSTRACT

Wilms tumor (WT), or nephroblastoma, is a malignant embryonic abnormality of the mesodermal lineage mostly identified during childhood. Initially, the clinical sign is an asymptomatic abdominal mass with subtle appearance. **Objective:** to analyze the diagnostic method and treatment of WT in children. Methods: an integrative review was conducted in Medline, Portal de Periódicos CAPES, Lilacs, and Scopus databases. The search was performed from March 1 to May 20, 2022 using the descriptors "*Tumor de Wilms*", "*Nefroblastoma*", "*Diagnóstico*", and "*Tratamento"*. Inclusion criteria considered publications from the last five years written in Portuguese English, and Spanish. **Results and discussion:** a total of 20 studies were selected and included in the discussion: 70% were retrieved from CAPES Periodicals, 5% from Medline, 15% from Lilacs, and 10% from the Scopus database. To ensure safe and effective diagnosis, studies used protocols from the International Society of Pediatric Oncology and guidelines from the Child Oncology Group. Although a positive prognosis was observed in most cases, it might be affected by the histological stratification of the tumor and the treatment onset period. **Conclusion:** The multidisciplinary team was crucial for early diagnosis, which directly affects treatment efficacy. Also, results from imaging and immunohistology are essential to tumor stratification and therapy adjustment.

Keywords: Wilms Tumor, Diagnosis, Therapeutics, Pediatrics.

RESUMO

O tumor de Wilms (TW), também conhecido como nefroblastoma, é uma alteração embrionária maligna da linhagem mesodérmica identificada predominantemente ao longo da infância. A apresentação clínica inicial mais comum é sutil e cursa com o surgimento de uma massa abdominal assintomática. **Objetivo**: Analisar a abordagem de diagnóstico e tratamento do TW em crianças. **Método**: Tratou-se de uma revisão integrativa realizada nas bases de dados Portal Periódicos CAPES, Medline, Lilacs e Scopus, em artigos com os descritores "Tumor de Wilms", "Nefroblastoma", "Diagnóstico" e "Tratamento", no período entre 01 de março e 20 de maio, 2022. Filtros de busca adotados: estudos em português, inglês e espanhol publicados nos últimos 5 anos. No total, 20 artigos foram incluídos. **Resultados e discussão**: Do total de artigos selecionados, 70% são procedentes da CAPES, 5% da Medline, 15% da Lilacs e 10% da Scopus. Os autores estão em consonância quanto à adoção de protocolos como o do *International Society of Paediatric Oncology* e as diretrizes do *Children's Oncology Group* para a definição segura e eficaz do diagnóstico. O prognóstico permanece positivo na maioria dos casos. No entanto, algumas variáveis podem influenciar a evolução, como o tipo histológico do tumor e o período de início do tratamento. **Conclusão**: A atenção da equipe multidisciplinar no processo de diagnóstico precoce influencia diretamente na qualidade do tratamento prestado ao paciente. O resultado de exames de imagem e a avaliação imuno-histológica são essenciais ao estadiamento e à adequação terapêutica.

Palavras-chave: Tumor de Wilms; Nefroblastoma; Diagnóstico; Tratamento; Pediatria.

INTRODUCTION

Wilms tumor (WT), or nephroblastoma, is the most prevalent abdominal neoplasm during childhood, representing about 90% of renal cancers in children. Despite the restricted access to specialized services, low socioeconomic conditions, and tumor severity, survival rates in developing countries range from 50% to 89% [1,2].

Renal embryogenesis is the core of WT pathophysiology. During healthy nephrogenesis, the metanephric mesenchyme differentiates into epithelial mesenchyme. Then, the nephric vesicles are formed, generating most of the differentiated cells in the fully-developed kidney. However, WT might disrupt renal development, resulting in a mixture of blastemal, epithelial, and stromal cells, and causing several renal impairments[3].

WT etiology might be explained by potential interconnections between genetic mutations and alterations in the genitourinary tract during embryological development. However, literature on WT etiology is still scarce. The genes WT1, CTNNB1, WTX, TP53, and MYCN are among the genetic markers associated with WT. The worst prognoses are frequently associated with the TP53 suppressor gene and loss of heterozygosity on chromosomes 1p, 1q, and 11p15[4,5].

WT is more frequent in children with genetic syndromes, such as the Wilms tumor-aniridia syndrome, characterized by abnormal genitourinary tract function (50% of cases develop WT); Denys-Drash syndrome, a progressive renal condition (90% of cases develop WT); and Beckwith-Wiedemann syndrome, a genetic condition marked by abnormalities in organ growth leading to dysmorphisms (10% of cases develop WT)[1, 6].

Anatomical signs of WT are mostly characterized by an initial swelling in the renal cortex, starting the initial expansion. Then, the renal tissue surround the region with swelling, forming a pseudo capsule with necrotic cells that may extend into the renal pelvis, affecting vascular and lymphatic vessels and the right atrium via the renal vein. Metastases tend to occur predominantly in the lungs and lymph nodes[7].

The WT is a significant topic in global clinical practice. Since the first report in 1971, constant updates with substantial contributions from clinical trials and group studies in North America, Australia, New Zealand, Europe, and Brazil aimed to improve disease management and increase survival rates. Notable pioneers of WT study include the Renal Tumor Study Group of the International Society of Pediatric Oncology (SIOP) , the Children's Oncology Group (COG), and the National Wilms Tumor Study Group[1, 3].

Since WT onset is subtle and has various therapeutic strategies, healthcare professionals involved in pediatric care must develop a sharp and efficient clinical reasoning process. In this sense, coherent information on the topic must be disclosed to improve decision-making and clinical outcomes in children with WT. Also, the literature lacks reviews focusing on the diagnosis and treatment of WT, highlighting the need for clarifying information on associated issues and minimizing knowledge gaps. Therefore, this study aimed to analyze the diagnostic method and treatment of WT in children.

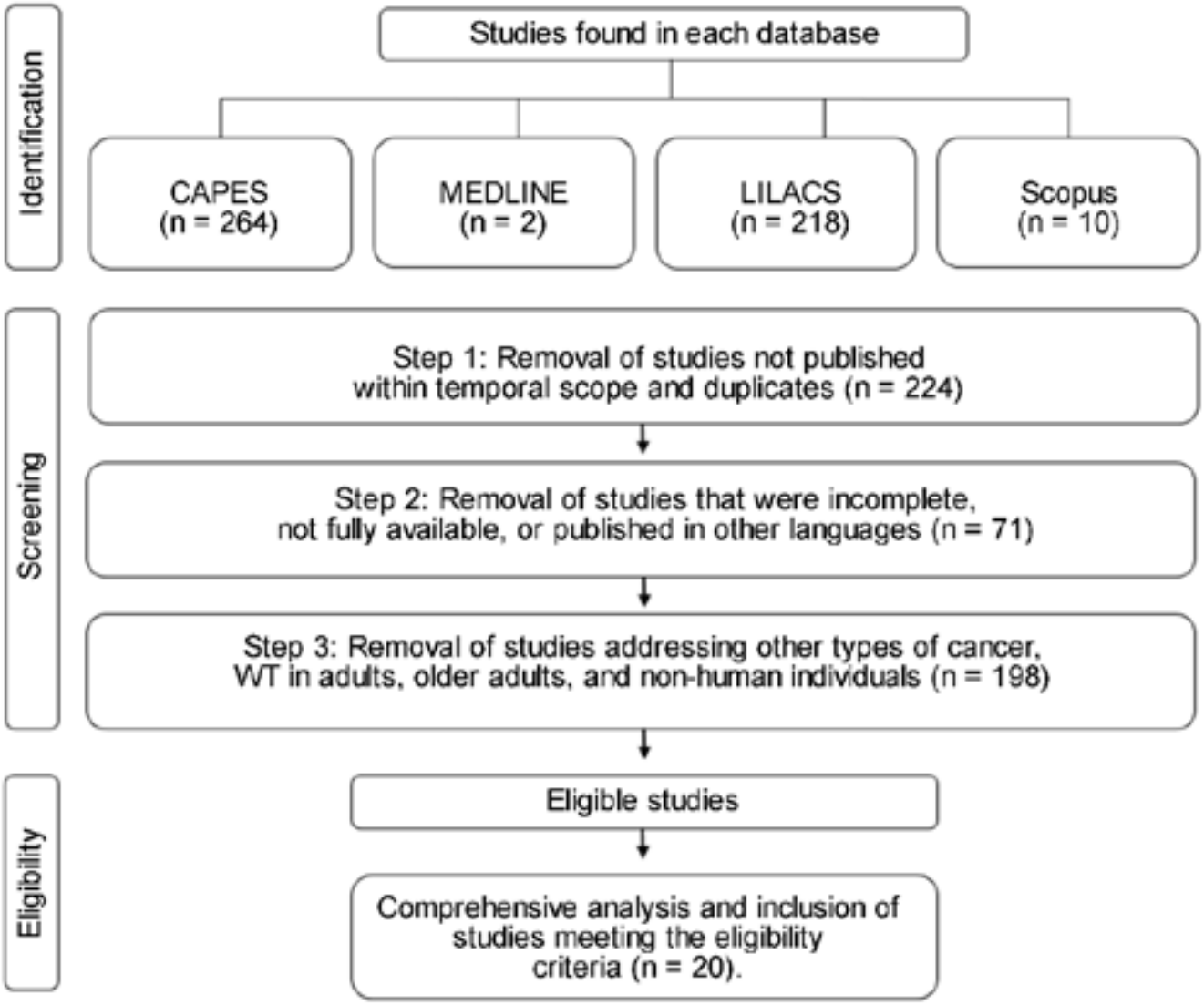
METHOD

An integrative review was conducted considering the need for consolidated information from primary studies, theoretical reviews, reports, and different types of research addressing the role of medicine in the identification and treatment of WT. This method allows a panoramic view of integral care by synthesizing knowledge from different authors [8, 9].

The search was conducted on CAPES Periodicals, Medline, Lilacs, and Scopus databases using a paired combination of the DeCS/MeSH descriptors "*Tumor de Wilms*", "*Nefroblastoma*", "*Diagnóstico*", and "*Tratamento*", between March 1 to May 20, 2022.

Inclusion criteria considered fully available scientific studies on WT in children published in the last five years and written in Portuguese, English, or Spanish. Exclusion criteria comprised studies published in other time frames, addressing divergent topics (other types of cancer, adults with WT, older adults, or studying other species), written in other languages, or not fully available. Figure 1 shows the flowchart of data screening.

Figure 1. Flowchart of data screening.



Source: Authors, 2022.

RESULTS

Table 1. Main findings of the selected studies

Title	Author	Conclusion
Bilateral Wilms tumour: a review of clinical and molecular features	Charlton J, Irtan S, Bergeron C, Pritchard-Jones K. (2017)	WT is an embryonal tumor that shows mimicry of cells of nephrogenesis. Unveiling the underlying genetic risk of bilateral WT offers a relevant challenge to maximize survival.
Dataset for the reporting of nephrectomy specimens for Wilms' tumour treated with preoperative chemotherapy: recommendations from the International Society of Paediatric Oncology Renal Tumour Study Group	Vujanić GM, D'Hooghe H, Vokuhl C, Collini P. (2021)	Due to their low occurrence, no international guideline by the International Collaboration on Cancer Reporting is currently available. COG preconizes primary surgery, while SIOP recommends preoperative chemotherapy as the initial step in management.
Histologia do Tumor de Wilms e seu impacto no prognóstico da doença	Gonçalves EVMC, Silva VLCD, Gonçalves VVMC. (2022)	WT features a unique cytoarchitectural pattern in which anaplasia adversely affects prognosis. Consequently, histopathological analysis is critically relevant.
Intra-tumor genetic heterogeneity in Wilms tumor samples	Pereira BM de S, Azevedo RM de, Neto JC de A, Menezes CF, et al. (2019)	The genetic heterogeneity of the intratumoral milieu plays a relevant role in the clinical outcomes of children. Identifying molecular markers is a current research topic by COG and SIOP groups.
Synchronous Bilateral Wilms Tumor: Surgical Evaluation and Survival	Silva JMM, Kipper ACS, Neves BH dos S, Borges DS, et al. (2021)	Protocols, such as COG and SIOP, are essential for developing WT treatment. Preservation of the renal parenchyma is advised upon the compatibility of patient variables.
Synchronous Bilateral Wilms Tumor: Surgical Evaluation and Survival	Oliveira PB, Grabols MF, Lima FFda S, Faria PAS, et al. (2018)	Conservative surgeries are recommended in bilateral WT cases managed in major referral centers.
The UMBRELLA SIOP-RTSG 2016 Wilms tumour pathology and molecular biology protocol.	Vujanić GM, Gessler M, Ooms AHAG, Collini P, et al. (2018)	The UMBRELLA protocol aimed to assess new prognostic factors. However, large international databases are needed for better follow-up of subgroups with negative prognoses.
Tumor de Wilms: análise das características clínicas e epidemiológicas	Turmina L, Voigt AD, Rodrigues AJS, Hata MM, Fiori CMCM. (2021)	WT is known to be the most common renal cancer in children (95%), resulting from a disturbed glomerular development of the kidney. Nonetheless, more reliable epidemiology data are needed.
Tumor de Wilms en riñon en herradura	Molina A, González F, Hernádez E, Bolaños J. (2021)	Few documented cases of horseshoe kidney associated with WT are available. However, nephroblastoma in this population is potentially associated with syndromic origins. Surgical intervention should prioritize the preservation of renal function.
Tumor de Wilms: Uma revisão de 15 anos de experiência recente	Illade L, Hernandez-Marques C, Cormenzana M, Lassaletta A, et al. (2018)	Studying WT is necessary, considering its epidemiological relevance. Protocols such as COG and SIOP sustain treatment practices.

Tumor de Wilms y otros tumores renales	Sánchez C, Ciordia TC (2021)	Physical examination often leads to the accidental discovery of an asymptomatic mass. In specialized services, diagnosis is given by clinical findings radiological, and histological information. The treatment typically involves chemotherapy and surgery.
Unmet needs for relapsed or refractory Wilms tumor: Mapping the molecular features, exploring organoids, and designing early phase trials – A collaborative SIOP-RTSG, COG, and ITCC session at the first SIOPE meeting	Brok J, Mavinkurve-Groothuis AMC, Drost J, Perotti D, et al (2021)	The recurrence of WT should undergo a comprehensive molecular characterization and be enrolled in protocols or trials including systematic data collection and reporting. It is crucial to improve the enrollment rates of children with WT in trials at the initial phase.
Update on Wilms Tumor. Journal of Pediatric Surgery	Aldrink JH, Heaton TE, Dasgupta R, Lautz TB, et al.(2018)	New insights into current diagnostic evaluation, surgical standards, and the impact of surgical intervention on risk stratification require further discussion.
Outcome of Nephroblastoma Treatment According to the SIOP-2001 Strategy at a Single Institution in Argentina.	Cafferata C, Cacciavillano W, Galluzzo ML, Flores P, et al. (2017)	The SIOP-01 protocol proposes a viable treatment strategy, presenting positive results for the institution.
Wilms Tumor	Haegner MJK, Villegas AZ, Calvo DM, Chavarría (2020)	The tumor, with sporadic clinical presentation, has been associated with genetic syndromes.
Wilms Tumor	Spreafico F, Fernandez CV, Brok J, Nakata K, et al. (2021)	The prognosis for children with WT is currently based on clinical, pathological, and molecular findings. In some cases, conventional cytotoxic agents, chemotherapy, surgery, and radiotherapy are considered.
Wilms tumor: 15 years of experience at a children's hospital in Córdoba	Seminara C, Planells MC, Pogonza RE, Morales M. (2019)	The most relevant prognostic factor identified was histology; however, children required long-term nephrological follow-up to confirm this hypothesis.
Wilms' Tumor – an unusual disease	Duarte ML, Duarte ER, Ferrreira JB de AF (2020)	Although clinical and imaging evaluation facilitates the diagnosis of pediatric renal tumors, histopathological examination remains indispensable.
Wilms Tumor and its recent advances	Ostrowski RV, Valentini MGT, Silva AR da, Trindade AF. (2022)	The tumor occurs mostly in children under 10 years old, resulting from abnormal proliferation of metanephric blastema cells.

WT: Wilms tumor, SIOP: International Society of Paediatric Oncology, COG: Children's Oncology Group.

DISCUSSION

Since the most frequent sign of WT is an asymptomatic abdominal mass, most children are diagnosed during routine pediatric consultations [3,4,10,11]. However, other signs may be associated with WT, such as abdominal pain and distension, gross hematuria, varicocele, hypertension or hypotension (normalized after nephrectomy), fever, anemia, and dyspnea (in cases of pulmonary metastasis).

Imaging exams (e.g., computed tomography and magnetic resonance imaging) are accurate for identifying, differentiating, and staging the tumor. Literature shows that complementary assessment is relevant for presumptive diagnosis, preoperative planning, and metastasis assessment. [2,6,12] Due to the absence of radiation and facilitated access, ultrasound is usually one of the first tests requested. Computed tomography facilitates visualizing lymph nodes and adjacent organs, further contributing to tumor staging.

Histology is often considered an essential examination to confirm the diagnosis of WT. Among the microscopic characteristics, a triphasic tumor pattern predominates, in which blastemal, mesenchymal, and epithelial cells are arranged in multiple proportions. In rare cases, tumors may be presented in a biphasic or monophasic pattern, characterized by the occurrence of anaplastic cells providing an important histological marker in risk stratification[10,13,14]. Thus, tumors are classified as low, intermediate, or high risk[1].

A new study protocol for pediatric renal tumors has been established based on evidence from the SIOP. Risk stratification is structured into five stages: stage I (limited to the renal area and complete surgical resection is facilitated), stage II (tumor expansion covers adjacent areas, but resection is still possible), stage III (tumor spread to adjacent structures and complete surgical resection impracticable; abdominal vascular or lymphatic beds are potentially affected and tumor rupture may be present), stage IV (hematogenous metastases or affecting lymph nodes outside the abdomen area), and stage V (bilateral tumor)[15]. Thus, histological stratification and tumor characterization (type, size, invasion of adjacent structures, metastases, and lymph node affected) are mandatory for the final diagnosis[16].

Before starting the treatment, differential diagnosis is essential to exclude neuroblastoma and other renal tumors (including clear cell renal sarcoma), congenital mesoblastic nephroma, renal cell carcinoma, and medullary renal carcinoma[1 6,17].

Regarding the type of therapy applied, the SIOP (widely used in Europe) and the COG (widely used in the USA) guidelines present divergencies. The COG indicates nephrectomy early in WT onset, while the SIOP initially suggests pre-surgical chemotherapy (except in children under six months of age) to prevent tumor rupture and the need for radiotherapy[12, 18]. Both guidelines present similar survival rates

In the postoperative period, indications vary according to the staging grade[19]. In stage I chemotherapy is not required in low-risk cases. However, vincristine and actinomycin for four weeks are prescribed in intermediate-risk cases. High-risk cases are treated with vincristine, actinomycin, and doxorubicin for 27 weeks[1].

In stages II and III, treatment with vincristine and actinomycin for 27 weeks is indicated for low or intermediate-risk tumors. However, for high-risk tumors, cyclophosphamide, doxorubicin, etoposide, and carboplatin are indicated. In stage IV, The combination of vincristine, irinotecan, cyclophosphamide, carboplatin, etoposide, and doxorubicin is recommended [1, 20].

Adjuvant radiotherapy is indicated in high-risk stage II, intermediate or high-risk stage III, preoperative or intra-operative tumor rupture, and cases presenting macroscopic peritoneal deposits. Pulmonary radiotherapy is also indicated for children stratified with high-risk cases and pulmonary metastases[1].

Children with bilateral WT (5% to 8% of cases[21]) are treated with preoperative chemotherapy followed by conservative surgery (resection of tumor leaving a safety margin and best preservation of the organ) and postoperative chemotherapy. Radiotherapy may also be indicated in some cases. Therefore, nephron-sparing surgery is performed to avoid intense nephrectomy. This approach might control the disease and protect the kidneys prone to relevant renal

failure[12, 22].

Children with WT may present higher risk of other chronic conditions than the general population. These conditions include secondary tumors, especially breast cancer, sarcomas, and lymphomas. The risks of renal failure, pulmonary fibrosis, ototoxicity, fertility problems, and cardiac abnormalities are also present[1, 19, 23].

The WT prognosis remains positive in most cases; however, it may vary according to histological type, disease stage at diagnosis, and adherence to the multidisciplinary treatment (psychologists, pediatricians, social workers, pediatric surgeons, anesthesiologists, radiologists, oncologists, and nursing staff)[24]. Also, the survival rate for children in stages I and II may reach 90%, while stages III and IV may reach 70%[10].

CONCLUSION

Early diagnosing WT affects its prognosis on children, highlighting the importance of physical examination in routine assessments. Clinical and complementary assessment (using imaging exams) by clinicians, immunologists, and histopathologists is essential to the diagnostic and to obtain descriptive tumor information. Available treatments include chemotherapy and conventional (or conservative) nephrectomy, which may be combined with radiotherapy.

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STORYTELLING AS MINIMIZER OF PAIN IN HOSPITALIZED PEDIATRIC PATIENTS: AN EXPERIENCE REPORT

A CONTAÇÃO DE HISTÓRIAS COMO MINIMIZADORA DA DOR EM PACIENTES PEDIÁTRICOS HOSPITALIZADOS: RELATO DE EXPERIÊNCIA

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ABSTRACT

Hospitalization is a delicate moment that involves pain, physical discomfort, and emotional instability, requiring patience from the hospitalized pediatric patient due to restriction to bed for a long period. During hospitalization, storytelling may promote moments of leisure and culture and reduce discomfort by allowing the patient to enter the world of the story. Therefore, storytelling is important for pediatric hospitalization due to its potential to temporarily reduce pain, facilitating the hospitalization process.

Keywords: Hospitalization; humanization; story; pain perception pediatrics.

RESUMO

A hospitalização é um momento delicado que envolve condições como dor, mal-estar físico e instabilidade emocional. Portanto, exige paciência do paciente pediátrico hospitalizado devido à restrição ao leito por um longo período. Durante a hospitalização, a contação de histórias pode promover momentos de lazer e cultura e amenizar o desconforto por permitir que o paciente adentre no mundo da história. Assim, a contação de histórias se torna importante para a hospitalização pediátrica, dado seu potencial de minimizar as reações à dor, mesmo que temporariamente, facilitando o processo da hospitalização.

Palavras-chave: Percepção da dor; hospitalização; humanização; história; pediatria.

INTRODUCTION

Humanization is a constant theme in debates and research in the health field due to its relevance in improving care and consolidating the values and principles of the Unified Health System.[1] Humanization involves the promotion of well-being and the appreciation of social circumstances and psychic, educational, and ethical aspects present in human beings.

Hospitalization is a delicate moment that involves pain, physical discomfort, and emotional instability, requiring patience from the hospitalized pediatric patient due to restriction to bed for a long period. This condition temporarily limits access to leisure and cultural activities.[2,3]

Humanized practices involve creativity and sensitivity for producing and promoting health. These moments can be promoted by storytelling during hospitalization and diseases. Furthermore, the attention received by the patient can reduce the discomfort of hospitalization since the physical or emotional pain is momentarily forgotten during the story.[4,6]

OBJECTIVE

This case report aimed to show the practice of storytelling in pediatric hospitalization, which identified the minimization of patient impacts and suffering.

METHODOLOGY

The methodology included training the authors in storytelling and hospitalization, storytelling in wards of a hospital in the city of Recife, the preparation of individual reports, and debates between students and the supervisor about their experiences.[4,5]

RESULTS

The hospitalization period refers to fragility, discomfort, pain, anguish, and fear, differing from other social contexts.3 Therefore, storytelling in this environment becomes more careful since the presence of the accountant causes interest, fun, and curiosity, contributing to the receptivity of children and their caregivers to this activity that promotes leisure and culture.[2,6]

Patients can become discouraged, withdrawn, and emotionally shaken during hospitalization. In this sense, a good story changes moods, stimulates smiles, and encourages speech, reducing suffering and favoring an exchange relationship. The thanks and requests for an encore at the end of each story reinforce the importance of this moment for the hospitalized pediatric patients.

FINAL CONSIDERATIONS

Comprehensiveness is one of the fundamental principles of the Unified Health System[1] and refers to the integral condition of understanding of the human being. Therefore, the union of healthcare professionals is essential to develop health practices that consider the biopsychosocial spheres of the individual.

Even when the patient does not verbally express what they are feeling, their look can communicate a lot, highlighting the importance of this attention for pediatric hospitalization patients. Although the storytelling reduces pain only temporarily, it facilitates the hospitalization process.

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"DESCARTES' ERROR: EMOTION, REASON, AND THE HUMAN BRAIN"

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António Rosa Damásio was born in Lisbon (Portugal) and has been based in the United States since 1975. He is a neurologist who serves as director of the Department of Behavioral Neurology and Cognitive Neuroscience and researches in the Alzheimer's Disease Research Center at the University of Iowa. He also teaches neuroscience at the University of California. From his writing and research, Damásio became known as one of the great names in world medicine. His research concerning the decision-making process, from which humanity is forced many times, establishes him as a great thinker within the intersection of social psychology with active and investigative medicine.

Among the exponents of his work, "Descartes' error" stands out, alluding to the well-known Cartesian dualism that postulates the mind, the sublime reason that resides in the soul, as independent of the body and emotions, and ceasing to occupy a physical space. This idea strongly influenced scientific research and Western philosophical thought (like Kant) and is opposed by Damásio's writing, which is still unfavorable to the French Enlightenment philosopher's mechanistic method of analysis, proposing the union of neuropsychological investigation with neurobiological study.

The beginning of the work highlights the case of Phineas Gage, known worldwide in medicine, in which a young adult had his skull transfixed by an iron bar. The chapter narrates in a fluid language how the case had repercussions. Although the functional motor and language impairment was minimal, the character and personality of the victim changed abruptly, as in the consolidated sentence: "Gage stopped being Gage."

Therefore, this case encouraged several debates at the time, as some scientists argued that psychological functions could not be attributed to a specific brain region, while others focused on the specialization of certain parts for generating different mental functions. The text continues to attribute a role to the knowledge tools of that time, such as "organology", which gave rise to the field of phrenology.

The book continues to present the case of the devastating accident using a neuroanatomy approach, which was also incorporated to reveal Broca's discoveries in Gage's time, a challenging aspect for the attempt to understand John Harlow, the doctor responsible for the case. The chapters denote the relationship between the limbic system and the frontal cortex.

Making this relationship, it is clear that an interruption in the judgment of future actions, as occurred with Phineas Gage after the injury, demonstrates that individuals with an impairment in integrating functions from the frontal cortex to the limbic system are unable to contain secondary emotions, reducing emotional learning.

Following the proposed ideas, the author introduces the somatic marker hypothesis, a focus that also plays a central role in the theme. Defined by a neuropsychological elaboration of the theory of emotion by the American psychologist William James, Damásio states the occurrence of primary and secondary emotions, as well as the feelings associated with them, showing how these elements interact to create mental images.

In this way, Damásio's elegant work delves into the intention of elevating the reader's understanding by countering an established philosophical impression that emotion hampers the development of the rational process, being essential to it. The historical survey of a landmark case, neurobiological research, the explanation of the intuitive mechanism, the proposition of somatic markers, as well as the unraveling

of reasoning with emotional input and decision-making capacity in a personal and social context remove the idealization of the immaterial soul of its apotheosis and practically force us to change this fundamental point that Descartes left, going from “Cogito, ergo sum”, from the Latin, “I think, therefore I am” to “I exist, I have emotions, therefore I think”.

HOW TO WRITE SCIENTIFIC ARTICLES: WITHOUT FUSS AND WITHOUT FEAR OF ABNT

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How to write scientific articles: without fuss and without fear of Brazilian Association of Technical Standards (ABNT) was written by Italo de Souza Aquino, who presented a Post-doctorate in Entomology from Oklahoma State University. Of the books published by the author, three best-sellers on scientific methodology stand out: How to read scientific articles: from undergraduate to doctorate (three editions); How to speak at scientific meetings: from classroom seminars to international congresses (five editions); and How to write scientific articles: without fuss and without fear of ABNT (nine editions), the latter stands out most among students. In this book, the author states that scientific methodology is important for the training of a researcher, and a good article must present objective, judicious, and ethical language.

In its 20 carefully selected chapters, the book clearly and directly describes the step-by-step process for accurate writing, covering all the aspects of writing scientific articles in the health field that result in high-quality reading.

The book also covers other productions, such as course conclusion works, monographs, dissertations, and theses. It also includes observations, the most frequent errors that cause the rejection of manuscripts, what the reviewer expects in a scientific article, plagiarism, and ethical standards in publishing scientific articles.

Lightly, the author seeks to “calm down” the reader-researcher regarding the ABNT standards, encouraging him to start writing before dwelling on them. Aquino states that the practice leads researchers to start to follow the ABNT standards without even realizing it. Moreover, the ABNT provides a website with all the needed information for the production of manuscripts.Clear scientific writing generally follows a specific format with key sections: introduction to a specific topic, hypotheses, description of methods, main results, and a discussion that links these results to a conclusion.

For the introduction of a scientific article, Aquino suggests writing in three parts: present by describing the importance of the research, highlighting the problem that demonstrates the relevance of the article to the reader; past by presenting what has already been done and what the scientific community reports on the topic covered; and future by proposing possible solutions that culminate in the study aims. The author indicates that the methodology should be well-written, considering that other people will be able to replicate the study.

The results must be presented in a way that justifies your initial questions without the need to publish all the data, exposing only the relevant or unexpected secondary findings that deserve mention. Aquino states that the researcher must keep the writing simple and impartial despite the presence of fascinating results. The book also describes how to create figures, graphs, and tables with good formatting and easy assimilation.

In chapter 12, we find how to write the discussion, report the interpretation, and comment on the meaning of the results. The author explains that the researcher has more freedom of expression in this section, with a personal position on the theme. Two important points covered in this chapter are the way and the need for researchers to be impartial.

Chapter 13 discusses the last part of the scientific article: the conclusion. In this section, the researcher must state their position consistent with the objectives and the report.

Currently, we see a great demand from undergraduate students to prepare and write articles. Books that promote greater awareness and access to information are important for students to become more effectively involved in research activities. Despite the simplicity of the writing,

we missed a more attentive presentation of the general standards and formatting for the composition of articles and scientific projects, considering the ABNT, the American Psychological Association, and Vancouver standards and the specific formatting of some journals.

How to write scientific articles: without fuss and without fear from ABNT provides an accessible, comprehensive, and essential resource for anyone seeking guidance on how to develop their research work. Students who are interested in publishing scientific articles during their undergraduate and postgraduate studies will find all the needed steps in this book.

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SCOPE

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The Journal Annals of Olinda Medical School - Health Social Responsibility, was created in 2018. Since then, it has been the official vehicle of the Olinda School of Medicine to support its principles, especially those related to encouraging research, teaching, and professional medical practices. It is an important instrument for disseminating knowledge, allowing exchange with other areas that favor medicine and the community, and enabling improvement of the standard care provided to the population. Since its inception, Anais FMO has faithfully complied with the requirements for biannual online and printed periodicity for scientific publication, following the recommendations of the International Committee of Medical Journal Editors (www.icmje.org) , which are commonly used in the areas of medicine and related sciences. Currently, Anais FMO is duly registered as a journal in the ISSN system. Articles are published in a continuous flow and all are free and open access, offered through the link https://afmo.emnuvens.com.br. By publishing their article in Anais FMO, authors transfer copyright to the journal and grant it the right of first publication. Manuscripts are submitted online through the platform, available at https://afmo.emnuvens.com.br/afmo/about/submissions.

POLICIES OF THE JOURNAL ANNALS OF OLINDA MEDICAL SCHOOL

Research Ethics Committee Approval

All publications submitted to the Annals of Olinda Medical School must have followed the research ethics recommendations of the Declaration of Helsinki and the standards of Resolution no. 466/2012 (http://conselho.saude.gov.br/resolucoes/2012/Reso466.pdf http://conselho.saude.gov.br/resolucoes/2016/Reso510.pdf) of Brazilian National Health Council. Studies that analyze aggregated data without identifying participants, such as those available in official databases in the public domain are exempt from research ethics committee approval.

Following the guidance of the National Research Ethics Commission, National Health Council, Ministry of Health (CONEP/CNS/MS), no institution is superior to the research ethics committee to analyze the nature of research proposals. The research ethics committee that approves the research must be registered with CONEP.

Research ethics committee must also approve case reports, following the provision no. 166/2018, of the Research Ethics Committee/CONEP/CNS, (http://conselho.saude.gov.br/images/comissoes/conep/documentos/CARTAS/CartaCircular166.pdf).

Case reports involving cadaveric parts must also have a research ethics committee approv-

al. Reports that use parts from cadavers destined for medical schools or similar areas for teaching and research purposes, in addition to ethical approval, must have authorization from the responsible institution to conduct the research.

It is mandatory to send a copy of the research ethics committee approval before submission.

Peer Review

Annals of Olinda Medical Schools recognizes that peer review is important in the publication process.

Therefore, we analyze the submitted manuscripts with ethics and maximum scientific rigor, following the steps below:

Every manuscript received is analyzed for suitability to the scope of the journal, its contribution to knowledge advancement, its originality, the methodological rigor with which the study was conducted, and the adequacy of the conclusions in relation to the results presented. In addition, the formatting is evaluated according to the standards of the journal. If any inaccuracy is identified, the manuscript is returned to the corresponding author, indicating the necessary adjustment. Only manuscripts that meet all the criteria described in the “Author Guidelines” undergo peer review.

Peer review is performed by at least two reviewers with extensive competence to evaluate the manuscript. The entire process is double-blinded; that is, reviewers and authors do not know each other’s identities.

The editorial decision is made based on the comments from the reviewers. It may follow one of the following outcomes: (1) rejection, (2) new submission chance by addressing the comments received, or (3) approval with or without changes.

Manuscripts falling under outcome (1), rejection, will be returned to the authors. In the case of outcomes (2) and (3), more than one round of review may be necessary. Conflicting opinions will have a third opinion requested or will undergo editorial arbitration. Failure by authors to comply with the review deadlines stipulated by the journal may result in the submission being archived. Approved manuscripts may receive layout changes as long as they do not alter the merit of the work.

The final editorial decision is recorded and is the responsibility of the editorial board. The manuscript content is the sole responsibility of the authors.

TYPES OF MANUSCRIPTS ACCEPTED FOR PUBLICATION

Original article: a full paper of a clinical or experimental investigation with unpublished research results (limit of 3,400 words, seven authors, and 30 references).

Integrative, systematic review, and meta-analysis: they should address topics of interest in health. Narrative reviews will not be accepted. Authors must include the study motivation in the Introduction. Summary and abstract must be formatted as a single paragraph in a block format with up to 250 words (limit of 3,400 words, seven authors, and 45 references).

Short Communication: short communication of original research results. In general, short communications are leaner analyses with a brief discussion of the results (summary and abstract must be formatted as a single paragraph in a block format with up to 120 words; the manuscript must be up to 1,000 words with Introduction, Methods, Results, and Discussion sections; up to two tables/figures can be included in up to three pages combined; references are limited to six).

Case reports: description of clinical cases of interest due to their rarity, presentation, innovative diagnosis, or treatment (summary and abstract must be formatted as a single paragraph in a block format with up to 120 words; the manuscript must be up to 2,000 words with Introduction, Case Report, and Discussion sections; up to two tables/ figures can be included in up to three pages combined; references are limited to fifteen; limit of seven authors).

Experience reports: detailed description of a successful or unsucessful experience of an author or a team, which contributes to the discussion, exchange, and proposition of ideas

for improving health care. It must include an introduction with a theoretical framework for the experience, objectives of the experience, methodologies used (including a description of the context and procedures), results, and final considerations. Summary and abstract must be formatted as a single paragraph in a block format with up to 120 words; the manuscript must have up to 2,000 words and up to two tables/figures; limit of 15 references and seven authors).

Methodological paper and theoretical/technical essays: manuscripts that deal with techniques or theories used in epidemiological studies or that portray an original clinical observation or description of technical innovations. Manuscript should be concise, limited to 1,500 words, five references, two illustrations, four authors, summary and abstract in must be formatted as a single paragraph in a block format with up to 120 words.

Critical review: restricted to a book or film in the medical field and related sciences. Argumentative manuscript in which the author describes and analyzes a social production aiming to influence his readers by recommending the work for its qualities or rejecting it for its flaws. It must be presented as follows: (1) presentation - summary of the work analyzed with both technical information and information about the book or film content; (2) analysis – interpretation and analysis of the work highlighting its main points, whether positive or negative, and the critical analysis from the author; (3) conclusion - opinion on the work, resuming the main points analyzed (up to 1000 words and two authors);

Letters to the Editor: comments from readers on works published in the Annals of Olinda Medical School (500 to 700 words).

Editorial: It is the initial article of a volume and is generally requested by the Chief and Deputy Editors to guests with recognized technical and scientific skills.

The word count includes Introduction, Methods, Results, and Discussion (title page, summary, abstract, references, tables, and figures are not included in the word count).

Manuscripts submitted must be intended exclusively for the Annals of Olinda Medical School, and simultaneous submission to another journal is prohibited. The information and concepts presented in the manuscript, as well as the veracity of the research content, are the sole responsibilities of the author(s).

Formatting

Manuscripts are accepted in Portuguese or English and must have an abstract in the original language of the manuscript and English. Manuscripts in English must have an abstract in English and Portuguese.

Manuscripts must be sent in Word, double-spaced, and Arial font size 12. Do not use line breaks. Do not use force manual hyphenations. The full term must follow abbreviations cited for the first time in the document. Title and abstract must not contain abbreviations.

Title page

Title of the manuscript in Portuguese and English (up to 25 words for each title);

Author information (full name, email, ORCID, affiliation, city, state, and country — do not include title and position);

Indication of the corresponding author, with their full address and email;

Conflicts of interest, in accordance with the Resolution of the Federal Council of Medicine (CFM) no. 1595/2000, which prohibits the publication of works for advertisement purposes of medical products and equipment, available at https://sistemas.cfm.org.br/normas/visualizar/resolucoes/BR/2000/1595. Conflicts of interest must be presented as follows: “The author(s) (name them) received financial support from the private company (mention its name) to conduct this study”. If there are no conflicts of interest, the authors must declare: “The authors have no conflicts of interest to declare”.

Source of financing, stating whether public or private; if there is none, mention that the study was not funded;

Number of the Certificate of Presentation for Ethical Assessment (CAAE) or number of Research Ethics Committee approval;

Authors contribution to the manuscript.

On the following pages, always starting on a new page, the following sections must be presented:

Summary and Abstract

Summaries must comply with the recommendations for each category of manuscript. In general, it must contain up to 250 words and be in structured format, covering the sections Objective, Methods, Results, and Conclusion. The same rule applies to the abstract.

Authors must include a minimum of four and a maximum of six keywords in both English and Portuguese regardless of the language in which the manuscript was submitted.The keywords must be standardized according to the Health Sciences Descriptors (DeCS), available at http://decs.bvs.br/.

References

References must be numbered consecutively according to the first mention in the manuscript and using superscript Arabic numerals in accordance with Vancouver style (www.icmje.org). The reference list must follow the numerical order of the manuscript, ignoring the alphabetical order of authors. Journal titles must follow the Index Medicus/Medline. The name of the first six authors must appear, followed by the expression et al. when this number is exceeded. Whenever available, the Digital Object Identifier (DOI) must be provided (see examples below). Personal communications, unpublished or ongoing work, citations from books, thesis, and dissertations should be avoided. The accuracy of references is the responsibility of the authors.

EXAMPLES

Reference to a journal publication:

Ng OT, Marimuthu K, Koh V, Pang J, Linn KZ, Sun J, et al. SARS-CoV-2 seroprevalence and transmission risk factors among high-risk close contacts: a retrospective cohort study. Lancet Infect Dis. 2021 Mar; 21(3):333-343. doi: 10.1016/S1473-3099(20)30833-1

Jardim BC, Migowski A, Corrêa FM, Azevedo e Silva G. Covid-19 no Brasil em 2020: impacto nas mortes por câncer e doenças cardiovasculares. Rev Saude Publica. 2022; 56:22. https://doi.org/10.11606/s1518-8787.2022056004040.

Reference to a World Health Organization Report

World Health Organization. Clinical Care for Severe Acute Respiratory Infection—Toolkit—Update 2022. Genebra: World Health Organization; 2022.

Reference to electronic documents

Brasil. Casos de aids notificados no SINAN, declarados no SIM e registrados no SISCEL/SICLOM, segundo capital de residência por ano de diagnóstico. Brasil, 1980-2021 [Internet]. 2021 [acessado em 12 abr. 2022]. Available at: http://www2.aids.gov.br/cgi/deftohtm.exe?tabnet/br.def

Figures and tables

Figures and tables must be inserted at the end of the manuscript, followed by their respective captions. Submission in separate files is not permitted. There must be page breaks between each one, respecting the maximum number of three pages for tables and figures combined. Do not format tables using the TAB key.

Figures must be up to 15 cm wide in Portrait orientation and 24 cm wide in landscape orientation and be presented within the requested margin (Normal Word setting). Colored figures are accepted. Figures must be provided in high resolution, plots in editable format, and tables, equations, charts, and flowcharts must be sent in an editable file (Word or Excel), never as an image.

Contact Methods

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