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EDITORIAL

CULTURA DE INVESTIGACIÓN CIENTÍFICA

La condición básica de la investigación científica es la existencia de investigadores con capacidades de pensamiento lógico, crítico y creativo, además, de habilidades comunicativas y con prácticas bioéticas[1]. Esta afirmación, que a primera vista podría parecer redundante, cobra especial relevancia cuando nos referimos al Dr. Eduardo Couve Montané (1952 - 2020), se tituló como Cirujano Dentista (1976) en la Universidad de Chile Sede Valparaíso (actual Universidad de Valparaíso), y que luego enriqueció con una amplia formación en destacados centros universitarios internacionales como Canadá, Alemania y EEUU; lo recordamos hoy, como un investigador científico de destacada y prolija trayectoria, que encarnó esas capacidades y prácticas señaladas, cuyo legado esta Revista Científica reconoce y aspira a continuar.

Dr. Couve como ayudante de biología - 1974

Dr. Couve jefe de laboratorio de Microscopia Avanzada UV.

Cabe señalar que el Dr. Couve, mostró su vocación científica en etapas tempranas de su formación universitaria, siendo ayudante de alumnos de la asignatura de Biología Celular de la Carrera de Medicina, y, recorriendo toda la carrera académica hasta consolidarse como Profesor Titular de la Facultad de Ciencias y del Instituto de Biología de la Universidad de Valparaíso.

Como científico mostró una prolífica producción de investigaciones científicas, que presentó en innumerables instancias nacionales e internacionales, tales como ponencias en más de 30 artículos de revistas de impacto, congresos, conferencias, asesorías, guía de tesis, receptor de fondos concursables por sus propuestas de investigación, revisor de artículos, editor de revista científica, etc.

Particularmente, queremos distinguir sus portadas aportando imágenes a la reconocida Revista Científica Journal of Dental Research de Julio 2012, agosto 2014 y en especial el 2018 con su publicación titulada "Schwann Cell Phenotype Changes in Aging Human Dental Pulp", en cuya portada aparecen las muestras fotográficas de procesos biológicos a nivel celular y molecular, que elaboró él mismo utilizando técnicas microscópicas de última generación, a propósito de su estudio. Lo cual pone de manifiesto las dotes de fotógrafo avanzado, transitando en imágenes entre la microscopía tradicional y la microscopía de fluorescencia evolucionada.

Además de lo precedente, nuestra Editorial busca mostrar los rasgos del hombre tras el científico, que sólo se comprenden uno articulado con el otro. El Dr. Couve, era una persona de gran rigurosidad crítica, desafiante de las ideas establecidas como verdades científicas, no sólo desde el discurso, sino que sobre todo desde su quehacer caracterizado por las inagotables horas que dedicó al trabajo de laboratorio, donde sometió sus hallazgos y la forma de obtenerlos a pruebas sistemáticas de validez. También, se caracterizó por su imparcialidad, receptividad humana y apertura a observaciones y proposiciones emergentes, que incluso cuestionaban su propio hacer. Esta flexibilidad que podría verse contradictoria con relación a sus altos niveles de rigurosidad y disciplina, lo hacía abierto a compartir con otros todo conocimiento científico, dado que siempre defendió la idea de la ciencia como un acto público y sus hallazgos como propiedad pública que demostraba con total consecuencia en la docencia que impartía inagotablemente tanto a nivel de pregrado como postgrado en diferentes carreras de la salud. Por otra parte, merece la mención de sus prácticas alojadas en los más apreciados valores de la ciencia moderna, en tanto, consideraba que el rol más relevante de los científicos, es generar conocimiento de calidad y veracidad que sirva al bienestar y desarrollo humano.

Este perfil, que refleja claramente las condiciones de investigador sencillo pero formidable del Dr. Couve, asociadas a su cautivante comunicación con las audiencias asistentes a conocer y reconocer sus hallazgos y conocimientos, representa, definitivamente, un ejemplo digno de imitar por todos y todas nosotros y nosotras, y en especial por las generaciones universitarias actuales en formación.
Finalmente, este sentido homenaje a este insigne investigador, que sirva a esta Revista, a su equipo y los investigadores que publican y publicarán en esta, de inspiración para aportar desde este espacio al desarrollo de una cultura científica e investigativa fértil y rica en creatividad y valores.

1. Como investigar con éxito en ciencias de la salud Bobenrieth M. Escuela Andaluza de Salud Publica Serie Monográfica EASP N°50, Cap 1. Granada España, 2012

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RESEARCH ARTICLE

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VASCULAR AND NERVOUS CHANGES IN DENTAL PULP OF HUMAN TEMPORARY TEETH ASSOCIATED WITH THE PHYSIOLOGICAL ROOT RESORPTION PROCESS

Eduardo Couve†[1], Diego Escudero[2], Macarena Orellana[3], Daniela Encalada[1]

ABSTRACT

Objective: To determine vascular and nervous changes during the progression of physiological root resorption, in three stages.

Materials and methods: 21 healthy temporary canines with indication of extraction, conducted on the Faculty of Dentistry of the University of Valparaiso were analyzed. Samples were processed for indirect immunofluorescence using Alexa Fluor 488 goat anti-mouse fluorochromes for Schwann cells (CS); Alexa fluor 555 goat anti-rabbit for endothelium. By confocal microscopy, a subodontoblastic portion, greater than the amelocemental junction, and an apical portion related to the root resorption area were analyzed. They were classified in initial, middle, and advanced stages. To process images, Ez-C1 3.90 and ImageJ programs were employed, and through visual analysis, researchers described pulpal changes.

Results: As the tooth was resorbed, nervous tissue degraded and disorganized with angiogenesis around it during middle stage. The same was observed in the coronal section, but with more predominance in the apical section. As physiological root resorption happened, the vascularity of temporary teeth increased.

Conclusion: As physiological root resorption ensues, simultaneously occurs degradation and disorganization of nervous tissue, along with angiogenesis.

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KEYWORDS:

Tooth, deciduous; root resorption; wallerian degeneration; blood vessels; fluorescent antibody technique.

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INTRODUCTION

The transition from temporary to permanent teeth is a unique and dynamic process in which the development and eruption of permanent teeth coordinate with physiological root resorption (PRR) of temporary teeth[1], this complex phenomenon is yet to be fully explained, and it is still of great clinical and biological interest[2].

Many pulp histological changes occur in temporary teeth because of this phenomenon. Changes associated with innervation and inflammatory infiltrate have been previously described[2,3], but, regarding pulp vasculature, both blood and lymphatic, there is not enough information that allows creating an overview of what happens during this process.

The purpose of this study is to determine vascular changes through comparative immunohistochemical analysis and confocal microscopy to achieve a further understanding of vascular-nervous changes during PRR on human temporary teeth, so that is possible to interpret the resorptive phenomenon and extrapolate its factors to pathological scenarios that affect the primary dentition.

MATERIAL AND METHODS

A cross-sectional, descriptive observational study was conducted on vascular-nervous changes in the dental pulp of temporary canines undergoing PRR.

Twenty-one asymptomatic temporary canine teeth without caries with different degrees of PRR were collected between August 2017 and January 2018. Prior donation of the teeth, informed consent was asked to the adult caretaker and the assent from the child was obtained, all canines had indications for extraction. Inclusion criteria were systemically healthy patients with temporary or mixed dentition, normal chronology, temporary healthy teeth with an indication of extraction endorsed by an orthodontist, with its crown intact and superficial lesions-free. Regarding the exclusion criteria applied, these were: teeth undergoing pulp necrosis, irreversible pulpitis, residual root, temporary teeth with a history of dental trauma, or avulsion, temporary teeth suffering from active/arrested caries, ankylosed canine or with an altered chronology of eruption.

All donors were patients of the Faculty of Dentistry of the University of Valparaíso, attended in the General Children's Dentistry clinics (I and II), aged 5 to 12 years, inhabitants of the Valparaíso Region, Chile.

The samples were classified into 3 groups, according to their PRR: initial stage, middle stage, advanced stage, based on stages described by Moorees in 1963 (4). The groups were obtained by the difference between maximum root length (13.25 mm) and minimum (1.5 mm) of the teeth, and dividing it into 3 (13.25 - 1.5 / 3), with this, it was obtained a 4mm approx. variability in each stage. For the sample selection, the PRR had to be restricted mainly from apical, with at least 1mm of root remnant. In contrast, all samples that presented poor fixation posterior processing, oblique or horizontal PRR, or teeth showing loss of pulp tissue, were excluded.

For the gathering of the samples, the investigator present during the exodontia took the tooth for examination, in search of pulp exposure. In those teeth who didn't present exposure, a transversal cut was performed mid-crown using a high-speed diamond bur. Then it was deposited with the pressure tweezers in a conical tube containing 20ml of 4% paraformaldehyde plus 0.5% picric acid for its fixation during 6 hrs., The tubes were identified employing an alphanumeric code (i.e.: A1, A2, ...). Once completed the fixation time, the samples were demineralized in a 4.13% EDTA solution, which was changed once a week, repeating this process for 3 months, until it was possible to start the necessary processes for cutting tests, immunohistochemistry and confocal microscopy.

Demineralized samples were washed with a phosphate-saline buffer solution (PBS) and cryoprotected in PBS solution with 15% sucrose for 2 hours, then this step was repeated in the same 30% solution for 24 hours.

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Samples soaked in a cryopreservative solution were mounted in tissue freezing medium (Tissue-Tek OCT Compound, Sakura Finetek, Torrance, CA, USA) and frozen at -25C°. 25um cuts were performed in a cryostat (Leica CM-1900) operated at -25 ° C and then mounted on slides previously positively charged with poly-lysine; these sections were rehydrated in PBS and incubated for 1 hour in a blocking solution composed of 1% bovine serum albumin (BSA), 1% horse serum, and 0.3% Triton X-100. Then the primary antibodies were added, which were diluted in a blocking solution and left to act for 12 hours at a temperature of 4 ° C. Samples went through 3 PBS washes of 10 minutes each. Then the secondary antibodies were added, also diluted in a blocking solution (1: 500) they were left to act for 1 hour at room temperature; later, it was washed again with PBS to remove excesses.

Subsequently, the nuclei were marked with DAPI (0.25µg / ml) for 10 minutes, finally, the samples were washed and covered with a mounting medium (Dako Industries, Carpenteria, CA, USA). For their maintenance, the samples were kept in plastic boxes at 4 ° C. Processed samples were analyzed with a Nikon C1 Plus confocal microscope.

The investigators used three lasers of different emission wavelengths (405, 488, and 555nm). The fluorochromes associated with the secondary antibodies were Alexa Fluor 488 goat anti-mouse (in green), Alexa Fluor 555 goat anti-rabbit (in red), and DAPI (405nm).

Digital records were processed employing Ez-C1 3,90 (Nikon Corporation) and Image J (NIH, Bethesda, MD, USA) programs for maximum intensity projections. For means of contrast and brilliance, Adobe Photoshop CS4 (Adobe Systems, Mountain View, CA, USA) was selected. For a standardized analyze, it was decided to use two base zones of 400 x 400 µm, zone 1 corresponded to the coronal pulp, located in the sector of the subodontoblastic portion superior to the amelo-cemental limit; meanwhile, zone 2 corresponded to an area of the same size in the most apical portion of the tooth, related to the root resorption zone itself.

Using the ImageJ processing program for multidirectional scientific images, the CS and endothelial images were quantified in an analog way by the researchers using confocal images, which were obtained from longitudinal sections of the dental pulp of twenty-one teeth.

In these immunoreactive profiles, it was fixed an expression threshold in all images obtained; At threshold fifty, the background elimination of the photographs of this investigation was achieved. The colors seen in the images correspond to red endothelial cells and green nerve cells.

The variables of this study were the type of tooth (temporary teeth, teeth that erupt between approximately 8 to 30 months of age, which after undergoing the physiological exfoliation process, were to be replaced by permanent teeth), the degree of radicular resorption (identifying the stage of PRR of temporary teeth, established by 3 stages (initial, middle, advanced)), vascular markers (identified by antibodies CD31, CD34, vWF, CD105), and markers of nerve components (identifiable due to the immunoreactivity of the CS against the S100 marker).

This study was approved by the Institutional Bioethics Committee for Research in Human Beings of the University of Valparaíso and complies with all the principles and updates of the Declaration of Helsinki.

RESULTS

After the inclusion and exclusion criteria were applied, 21 eligible samples were obtained, confirmed for processing.

Subsequent to the qualitative analysis, important changes were evident during the progression of the PRR when comparing the three stages. Vascular-nervous tissue increased significantly in this zone, especially when going from medium PRR to an advanced PRR. (Figure 1, Table I)

By schematizing the findings, considering the amount and density of tissue in each sample and

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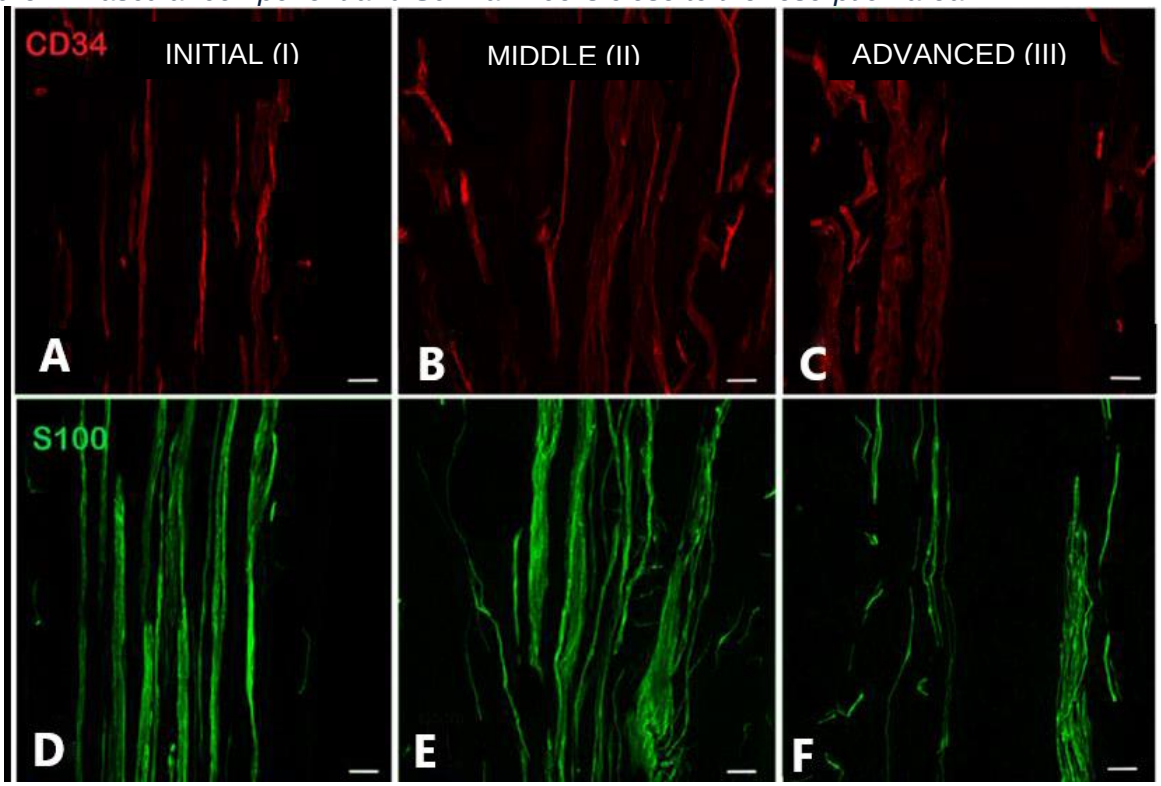
Figure 1: Vascular component and Schwann cells close to the resorption area



Vascular component (CD34, red) and Schwann cells (S100, green). Close to the resorption area, the fascicular degeneration process of the nervous component is observed as the resorption process progresses. In the initial PRR, a linear and neat fascicular arrangement of the axons is observed (D); In the middle PRR, an increase of the vascular caliber is observed (B) this persists in the advanced PRR (C) Furthermore, in this stage a fragmented fascicular disposition is observed (F). The increased vascular caliber is distributed around the areas of axonal degeneration (E, F). Scale bars: 50 µm.

Table I: Schematization of the samples close to the resorption area

Radicular Pulp	Initial PRR	Middle PRR	Advanced PRR
Schwann Cells (S100)	+ +	+ + +	+
Vascular Components (CD34)	+	+ + +	+ +

The schematization of the samples considering the quantity and density of tissue according to their PRR stages in the area close to the reabsorption. The more (+), the more predominance of the tissue.

classifying it based on its stage of PRR; the nervous tissue becomes more prevalent in the middle stage; this is due to its fascicular disorganization and because of its degradation it decreases towards the advanced stage. As for the vascular component, it increases on the middle stage, the same as SC, and in the advanced stage decreases its distribution the same as nervous tissue. In the areas with no nervous tissue, there is also no predominance of vascular tissue, establishing a correlation (Table I).

The same vascular-nervous changes observed in the area close to the resorption are observed in the coronal pulp, above the amelo-cemental junction, but more attenuated. (Figure 2 and 3)

By schematizing the findings, considering the quantity and tissue density, it was observed that nervous tissue increases in the middle stage, due to disorganization of the fibers, also it was observed their degradation, which in the advanced stage is almost complete. Vasodilation and vascular neoformation characterized the

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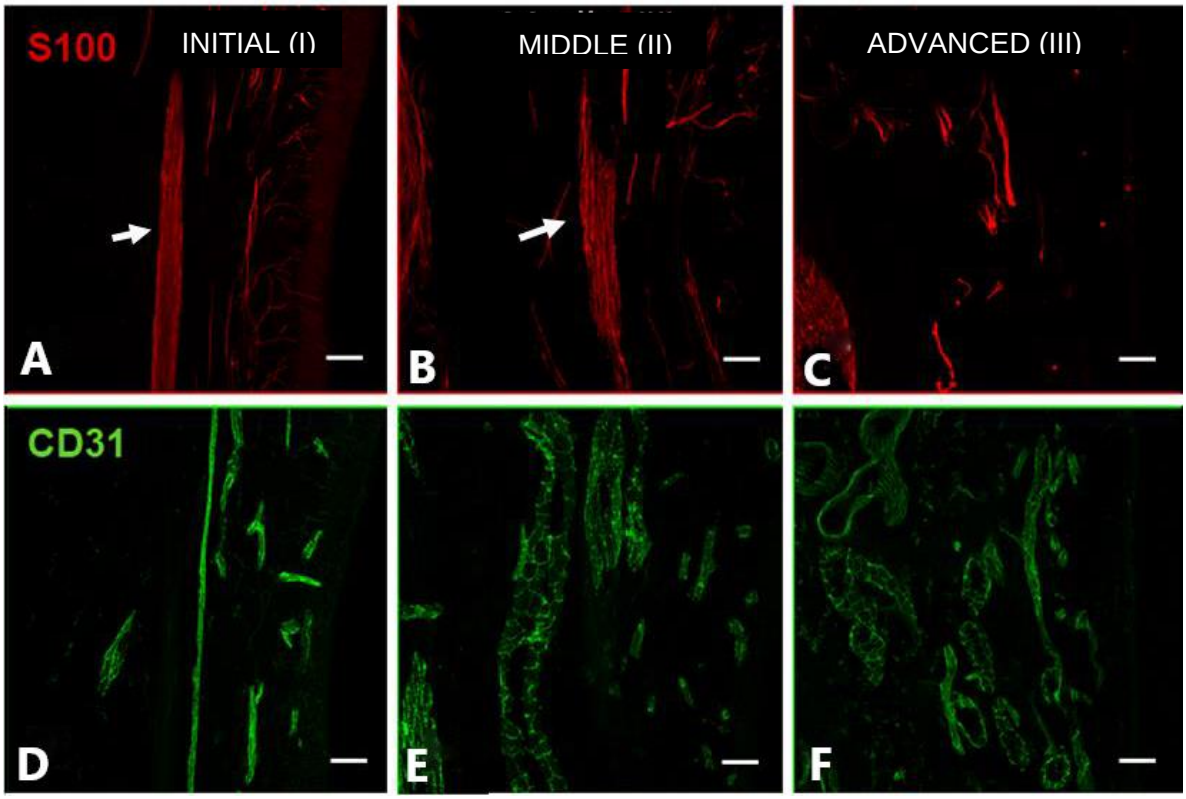
Figure 2: Vascular components and Schwann cells of coronal pulp above the amelo-cemental junction during PRR



Characterization of vascular components (CD31, green) and Schwann cells (S100, red) of coronal pulp above the amelo-cemental junction during PRR. In the initial stage, an integrated and conserved axonal band is observed (A, arrow); In the middle stage, myelinated axons bands are disintegrated (B, arrow) as the process progresses. Vasodilation is observed, with foci of angiogenesis (E); in advanced PRR, the axonal band degenerates losing its structure (C) also it is observed marked vasodilation, changes in the vascular pattern with distribution around areas of axonal damage (F). Less presence of nervous tissue. Scale bars 50 µm.

Table II: Schematization of the samples considering the quantity and density of tissue in the coronal pulp, over the amelocemental junction, according to their PRR stages.

Coronal P.	Initial PRR	Middle PRR	Advanced PRR
Schwann Cells (S100)	++	+ + +	+
Vascular Components (CD34)	+	+ + +	+ + +

The more (+), the more predominance of the tissue.

passage to the middle stage of PRR, together with the beginning of the degradation of the myelinated axon bundles, increasing its preponderance, this vasodilation and growing angiogenic zones develop fully in the advanced stage. In advanced PRR, angiogenic foci are observed in the reabsorption zone, and axonal degradation in the center and periphery (Table II).

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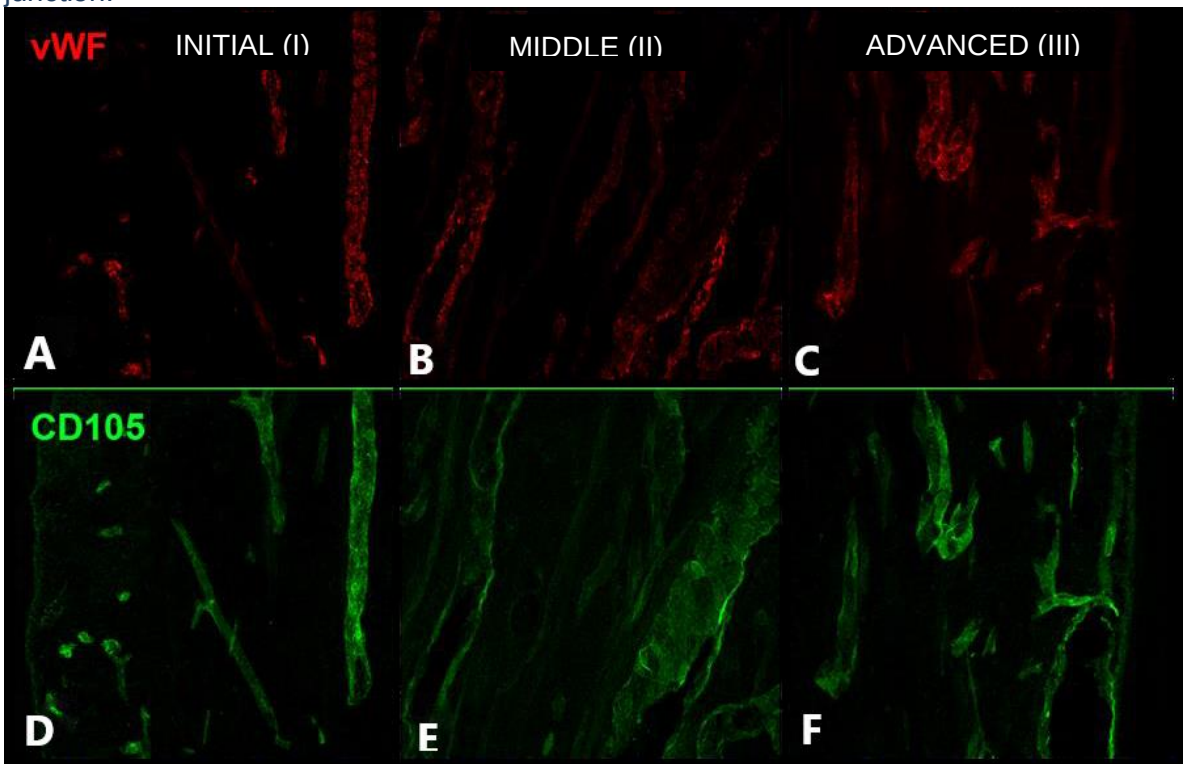
Figure 3: Double labeling for endothelial cells of the coronal pulp on the amelocemental junction.



Double labeling for endothelial cells (vWF, red; CD105, green) of the coronal pulp on the amelocemental junction. In the middle stage of PRR, the greatest increase in vascular diameter (B, E) is shown, accompanied by the proliferation of endothelial cells, which is also present in the advanced stage (C, F).

DISCUSSION

The main finding of this study was the variation in marking at the level of endothelial and nerve cells.

As the PRR progresses, the behavior of the SC changes in its distribution and increases in its concentration in the initial and middle stages of the PRR process, the progress of this phenomenon is associated with an asymptomatic chronic inflammatory process, as described by Angelova et al.[17]. This process comprehends an axonal degeneration and progressive denervation of the dental pulp in temporary teeth, which reflects a loss of the myelin sheath, leading to a differentiation of SC, which allows activation of the phenotypic plasticity of these cells, towards a repairing phenotype (RSC)[8] RSC promotes the remodeling of the degenerated axons of the dental pulp, through an autophagic pathway for the removal of myelin debris from axonal degeneration[9], this phenomenon is observed in the initial and middle stages during PRR, this matches with what was witnessed in this study, where a higher SC marking was obtained in the middle stage, being comparable to the research by Suzuki et. Al.[3], considering a similar methodology was employed with statistically significant results achieved. This higher concentration of SC (both coronal and apical of the dental pulp), is influenced by a greater degradation of the myelin sheath, therefore, a greater manifestation of RSC in an attempt to remodel the nerve fibers of the dental pulp.

In advanced stages, there is a larger expression of RSC for the correct remodeling of damaged axons, centering this phenomenon in the coronal area of the pulp according to what was stated by Couve et al.[10], is because of this that temporary teeth undergoing PRR may present pain when presented to some type of noxa or stimulus, for this, it is recommended the use of local

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anesthetic for dental procedures on temporary teeth experiencing an advanced PRR[11].

As for the endothelial cell marking, what stands out is that there is a similarity with the marking of degenerative areas of nerve cells, this is because along with nerve degeneration occurs angiogenesis, which leads to qualitative differences in the distribution and morphology of blood vessels, concentrating these changes in the coronal area, according to Monteiro Et. Al[11], that study differs in methodology and does not produce statistically significant results, compromising any extrapolation of their results, but in terms of biological plausibility, greater angiogenesis reflects a greater metabolic requirement during the PRR process, since it is required a greater blood flow for the subsistence of this phenomenon and, subsequently, the eventual dispose of metabolic residues[12].
Among the limitations of this study was the limited number of samples and the non-randomness of this, despite of that, the processing of the samples was carried out by the ImageJ software, which has been used by numerous investigations at an international level because it allows an objective and finished processing of the images obtained, in addition, the analysis of the samples was jointly carried out with the aid of an expert observer on the subject, who is credited with vast years of experience and multiple investigations validated in the scientific community.

Another limitation of this study was the PRR categorization employed, that, even though it has been previously used in past studies[4], was modified and is deeply influenced by the collected samples of the study, because of that this classification was drastic and broad (⅓, ⅓ - ⅔, ⅔ - > ⅔), this leads to possible correlations between the exact percentage of root resorption and the pulp state being masked, which causes a selection bias. For posterior investigations, we suggest the implementation of an internationally validated guide or one validated in another study.

Erroneously, it is considered that temporary teeth undergoing PRR are less sensitive to pain, thus, performing dental treatments avoiding the use of local anesthesia, however, current scientific evidence attests that these teeth preserve their regeneration and defensive capabilities well into PRR, this must be taken into consideration in the management of pain in pediatric dentistry, to preserve the primary dentition using conservative methods[3], moreover, nowadays temporary teeth are one of the main sources of stem cells, which are forebears of tissues with therapeutic expectations in many diseases in the future[4,6].

This study was focused on temporary teeth with different degrees of PRR, for this reason, it would be necessary to execute further investigations on how teeth suffering from caries, trauma or lesions, besides undergoing PRR ,behave when subjecting them to clinical procedures, to compare pulp resistance (which directly influences the prognosis of the tooth).

CONCLUSION

In conclusion, as PRR ensues, simultaneously occurs degradation and disorganization of nervous tissue, along with angiogenesis based on the results obtained, this implies that dental pulp retains its vitality during PRR unto more advanced stages. With this knowledge, we can employ new guidelines in the decision of the treatment plan in teeth undergoing PRR.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest with respect to this article.

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IN MEMORIAM EDUARDO COUVE

"Trabajar con Dr. Eduardo Couve fue un privilegio. Su capacidad innata para contagiar la pasión por la ciencia y el aprendizaje en cualquier categoría es una habilidad que nunca antes vimos en otro docente. Agradecemos profundamente su dedicación, su amor por lo que hacemos y sobretodo su actitud cercana, que propició un ambiente académico casi familiar, que jamás olvidaremos. Su partida tan prematura es una real pérdida para la universidad, siempre será extrañado. QEPD querido Dr. Couve" (Diego Escudero, Macarena Orellana, Daniela Encalada).

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RESEARCH ARTICLE

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KNOWLEDGE CONCERNING EXPLICIT HEALTH GUARANTEES FOR ADULTS AGED 60 IN PRIMARY CARE DENTIST OF VALPARAISO

Paulina Cortés[1], Tamara Espinoza[2]

ABSTRACT

Objective: To evaluate the knowledge degree in primary health care Odontologists regarding the clinical guide for Explicit health guarantees for 60 years old adults, evidence-based recommendations for the optimization in clinical attention of this group.

Material and Methods: A transversal descriptive study was conducted, surveying Primary Care professionals from the Family Health Centers of the Municipal Corporation of Valparaíso, with at least a 6 months old work contract. An informed consent was signed by all participants before an instrument was applied to them, where general data were consulted: 15 clinical-guide related questions, and 4 opinion related questions.

Results: The sample consisted of 41 professionals, belonging to 12 Family Health Centers, of which, 41% worked full time. 36 professionals acknowledge the guide, 31 of them have read it, and 71% of them have accessed it via the web. The mean of knowledge was 71.23% of correct answers, with a range between 40% and 92%. The recommendation of electric toothbrushes was correct in all, meanwhile, the wrongest answers were those concerning the normal characteristic of the mucosa and Xerostomia Treatment. The highest scores were obtained by professionals with less than 5 years of working experience. 58% recognize barriers and limited time available for their implementation, lack of supplies, difficulty in accessing the guide, among others. 57,58% believe that the guide is a good way of standardization.

Conclusion: 71% of professionals surveyed possess adequate knowledge of the guide.

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KEYWORDS:

Health Policy; Oral Health; Knowledge; Adults Aged 60; Primary Health Care; Chile.

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INTRODUCTION

Worldwide, the elderly population is a priority in public policies, in Chile, this is no different.
A milestone in this matter was the GES (Explicit Health Guarantees) program, formerly known as AUGE, that provides access guarantees, quality, opportunity, and financial protection to every FONASA (National Health Fund) or ISAPRE (Provisional Health Institutions) affiliates, and their custody[1]. In July 2005, this program started operating, offering coverage to 56 health issues, and rising to 80 since 2013, however, only 5 of them comprise dentistry issues[2]. One of these is the "60 years old Oral Health Program" also known as GES-60, whose intent is to educate, prevent, recover and rehabilitate oral health in 60 years old patients, trying to give a solution to 20 health problems recognized in this population, observable in the care basket.

The Chilean Ministry of Health (MINSAL) establishes a guide as a reference for dental treatment in primary health care, as in Endodontics, Periodontics, and Oral Rehabilitation, encompassed in the GES-60 program[3]. Its aim is to deliver recommendations based on available evidence, for the treatment of caries, periodontal disease, and partial/total edentulism, contributing to improving the quality of life of the elderly. While they emphasize that it is just a helping tool for clinical decision making, it must be considered all patient's preferences and personal values. According to both, MINSAL and FONASA statistics, the coverture of GES-60 is scanty, 27% of the population registered in 2016, in comparison to the "Integral oral health program" for 6 years old children or pregnant women, each with 80% and 72% respectively, of coverture that same year[4]. It has been given as the reason of this scanty coverture the retirement age of older adults, usually over 65 years old, their low pensions and high cost of living, these last the reason on why they must continue work until an older age and in schedules not compatible with access to Primary health care (APS) attention[2]. It is suggested an increase in the GES-60 policy range since although socioeconomic inequities have decreased in the Chilean adult population that utilizes dental care, it is still very meager the number of elderly - especially males- who consult[2]. Another measure proposed to tackle this problem is to financially incentivize all primary health care workers to upgrade their performance[5].
The need for treatment of these patients is high, being evident in, for example, how 82,3% of the total of GES-60 patients treated in APS of Villa Alemana requires complex periodontal treatment (6), or the 17,4 COPD in women, and 17 in men of Maipú[7]. The need for prosthetics in this group is 98%.
The aim of this study is to evaluate the degree of knowledge of primary health care Odontologist in Valparaíso regarding the clinical guide of the GES 60 years old Oral Health Program.

MATERIAL AND METHODS

A cross-sectional descriptive study was conducted, where 41 Odontologists of the Family Health Centers of the Municipal Corporation of Valparaíso, Chile were surveyed in the first semester of 2016. This sample size, randomly selected, is calculated based on a total of 69 Dentists belonging to this Corporation in 2017, with a confidence level of 95%, and a sampling error of 5%. For the inclusion in this study, the participant had to present their status of contractual quality: plant, contract, or fees with at least 6 months in office, excluding those in replacement. All signed an informed consent attached to the survey.

The survey created was applied to different professionals from different departments of the University of Valparaíso, until it was standardized. Finally, the instrument consisted of a general information section for all participants such as age, sex, educational degree, length of office and municipal workload; a second section consisted of 15 multiple selections problems that evaluated knowledge regarding the guide's info on oral hygiene, technique, frequency, and length, prosthesis disinfection, use of fluorides and most common oral lesions present in the elderly. A final section consisted of 4 questions about their personal opinion on the survey. The survey was evaluated in percentage and also scored with grades from 1.0 to 7.0, using a cut-off score of 4.0 and a minimum acceptable

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requirement of 60%.

The investigation protocol, measurement instrument, and informed consent were all presented and approved by the Municipal Corporation. The heads of dental programs were contacted and asked to define a schedule where the survey could take place without interrupting the normal attention to patients; the aim of the study was then explained and the survey was applied to them.

The register of the data obtained was executed using the Microsoft 2013 Office Excel program, in order to obtain a database, tables and figures.

For the description of the variables and descriptive analyzes, the STATA® (Statistics / Data analysis) from StataCorp, Texas – U.S.A. 11 version program was employed.

RESULTS

The sample consisted of 41 professionals, 56,1% were female and 43,9% male, all from 12 different Family Health Centers (CESFAM) from the city of Valparaiso. 78% of this sample were aged 26-35 years old, with an in-office experience of fewer than 5 years, only 41% were working full time. 36% have subsequent studies, related to a specialty diplomate and/or post-title. In regard to the GES-60 clinical guide, 36 professionals (88% of the sample) acknowledged its existence, and 31 of them had read it. The most common method accessing it was via the web (71%). Only seven professionals claimed to have the guide physically at their workplace for access.

The percentage of correctly answered answers, that evaluated knowledge in all participants, had they read or not the guide, it's presented in table I. All answered correctly when to indicate an electrical toothbrush, and 85,63% when to indicate fluorides. The least correct answers were those concerning the normal characteristic of the mucosa in the elderly, and Xerostomia Treatment, each with 55% out of the total sample. The average of the knowledge, obtained from the total of correct answers by the professionals, was 71.21%, with a minimum of 40% and a maximum of 92%. The average grade of the group was 4.9 (in a 1.0 to 7.0 scale) with a minimum of 3.0 and a maximum of 6.4. Grades were higher in those professionals that had been

Table I: Percentage of correct answers regarding the GES-60 Clinical Guide.

Concept to evaluate	Total of the sample (n = 41)	Read the clinical guide (n = 31)	Didn't read the clinical guide (n=10)
Characteristics of toothbrush	69,17%	73,12%	55,56%
Use of toothbrush	100,00%	100,00%	100,00%
Technique, frequency and length of oral hygiene	65,00%	69,35%	50,00%
Interproximal hygiene method	65,00%	66,67%	59,26%
Prosthetic cleaning and disinfection	69,50%	74,84%	51,11%
Use of fluorides	85,63%	85,48%	86,11%
Characteristics of normal mucosa	55,00%	54,84%	55,56%
Oral lesions more prevalent	79,50%	77,42%	86,67%
Xerostomia etiology	77,50%	78,23%	75,00%
Treatment of Xerostomia	55,00%	59,68%	38,89%
Drugs associated with xerostomia	63,75%	68,82%	46,30%
Etiology of subprosthetic stomatitis	77,50%	78,23%	75,00%
Aims of a treatment plan in the elderly	65,00%	64,52%	66,67%
Treatment of gingival and periodontal diseases	63,33%	64,52%	59,26%
Criteria for referral to the specialty of Periodontology	77,50%	79,35%	71,11%

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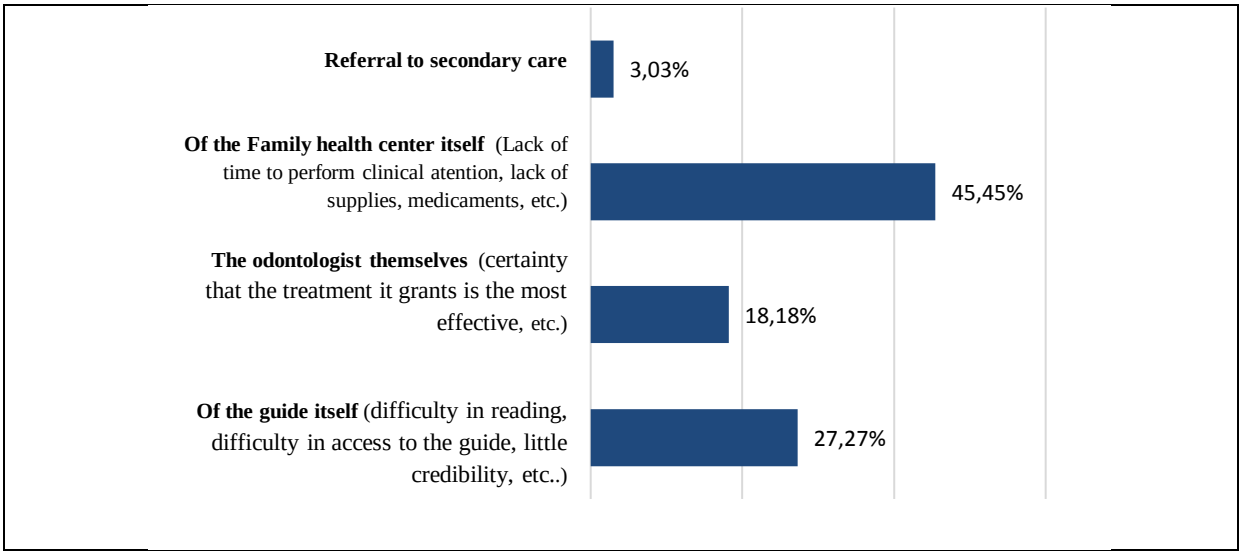
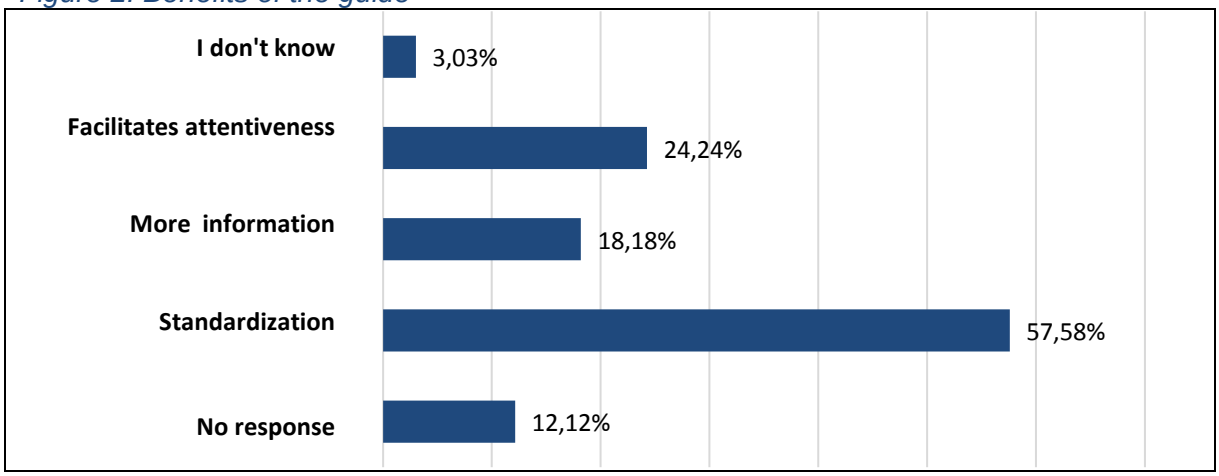
Figure 1: Barriers presented when following the guide's recommendations



Percentages of responses regarding the "Perceived Barriers" for the implementation of the GES-60 Clinical Guide (n = 18 professionals).

Figure 2: Benefits of the guide



Percentages of responses regarding the "Potential Benefits" of the GES-60 Clinical Guide (n = 31 professionals).

working in office less than 5 years in the CESFAM, and there was no difference between those who had a specialty and those who didn't.

Regarding those who reported having read the clinical guide, all of them answered correctly when to indicate an electrical toothbrush, and 85.48% when to indicate fluorides (Table I). The least amount of correct answers was achieved regarding the characteristics of normal oral mucosa in elderly patients, and the treatment of Xerostomia, 54,84%, and 59,68% respectively. The average of the knowledge obtained by the professionals was 73.00% of correct answers, with a minimum of 46.00%, and a maximum of 92.00%.

Converting to a rating scale, the group's average mark was 5.0, with a minimum of 3.3 and a maximum of 6.4. Results were slightly higher in those professionals with less than 5 years of working experience in the CESFAM, there was no difference among those who had a specialty and those who didn't.

Regarding those who claimed to have read the guide, all 10 professionals answered correctly when to indicate an electrical toothbrush, followed by 86,67% that answered correctly the most prevalent lesions in the elderly (Table I). The least percentage of correct answers were obtained concerning Xerostomia, its treatment,

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and associated pharmaceutics, 38.89%, and 46.30% respectively.

The average of knowledge obtained by these professionals was 65.01% of correct answers, with a minimum of 40.00% and a maximum of 74.12%. Converting to a rating scale, the average grade for the group was 4.45, with a 3.0 minimum and a 5.06 maximum.

There were no professionals, practicing for more than 10 years at CESFAM, who have not read the clinical guide. There was no difference found among those who had a specialty and those who didn't.

Of all 31 professionals who had read the clinical GES-60 guide (3), 58% recognize barriers for its implementation being mainly evidenced within the CESFAM itself (Figure 1). Regarding the potential benefits of this Guide, 57.58% of these professionals believe that it is a good way to standardize care (Figure 2).

DISCUSSION

This report had participation of 59.42% of the dental professionals belonging to the CESFAM of the Municipal Corporation of Valparaíso. Those who refused to participate claimed that they had little available time due to their busy schedules, both clinical and administrative. This clinical guide created by MINSAL mainly focused on its application in APS, delivers the best available evidence on the situation of this population (60 years old) and the most prevalent oral lesions the suffer, seeking to standardize their diagnosis and therapeutic approach. This could not be achieved if one of every four dentists in a working team hadn't read it. The benefits of this guide, perceived by the same professionals, are mainly the standardization of the processes (57,58%), facilitate care (24.24%), and provide more information (18.18%).

There is no previous evaluation of the GES-60 executing staff, odontologists from APS, as there is for the "Integral Oral Health program" for 6 years old children[8]. In that said executive review done by the MINSAL in 165 professionals from the public and private sectors, it is stated that 97.2% of dentists in the public sector and 85.7% in the private sector know that it exists and have read the guide. Thirteen questions were applied based on recommendations and concepts present in the guide, with 52.8% of these having more than ten correct answers, and 25.4% less than six.

The concept they least handled was the risk classification of patients. The sufficient level of knowledge was set at 60% of correct answers, note 4.0, obtaining mean values of 5.0 in the group that declared having read the guide and 4.5 those who did not.

In the present study, the best results were found associated with workers with less than 5 years of experience in APS, Attributing this to a greater updating of public policies in their study houses, due to their short time of graduates, and the greater incentive to this type of evidence-based guidelines, as evidenced in a study that reported that 71% of these people access to these type of guidelines via the web.

Regarding the level of knowledge of the same, the treatment of Xerostomia is one of the less correct answers, when 51.61% said they would recommend the use of artificial saliva when in fact the GES-60 guide considerate it to be ineffective. The usage of this therapy is controversial in the literature[3,9,10]. Only 61.29% of those surveyed recognized that the mucosa of the elderly presents less resistance to pressure and less ability to respond to external influences. This consideration is relevant given the possibility of the development of irritations, hyperplasia, and infections in the mucosa, as well as the need for a special design in rehabilitation treatments.

58% recognize barriers and limited time available for their implementation, lack of supplies, difficulty in accessing the guide, among others, being these mainly evidenced within the CESFAM itself. On the contrary, the GES-60 guide declares that the greatest barrier is the knowledge of the professionals, 71.23% of what was evaluated, followed by their attitudes towards the guide, and external barriers[3]. The least discussed barrier described by the

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professionals of Valparaiso was the referral to specialty care.

CONCLUSION

71% of professionals surveyed in the CESFAM of the Municipal Corporation of Valparaíso possess adequate knowledge of the GES-60 guide, although one in every four professionals haven't had read the guide.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest with respect to this article.

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RESEARCH ARTICLE

ASD JOURNAL Applied Sciences in Dentistry

EFFICIENCY OF INFILTRATIVE ARTICAINE V/S NERVE BLOCK LIDOCAINE IN MANDIBULAR THIRD MOLARS SURGERY

Baeza Solange[1], Rojo Felipe [2], Bialostocki Ebner[1]

ABSTRACT

Objective: To compare the efficiency of infiltrative articaine versus nerve block lidocaine in pain management during mandibular third molar extraction.

Material and methods: Randomized clinical trial. 25 patients with symmetrical semi-included third molars were analyzed, to which infiltrative Articaine and Nerve block Lidocaine were randomly applied to each demi-side. The proposed variable was the efficiency of the anesthesia protocol. Results were analyzed with a proportional comparison Z test, analyzed with a confidence level of 95%.

Results: The infiltrative articaine efficiency was 92% for B-group, existing no statistically significant difference to Nerve block lidocaine ($p<0.05$).

Conclusion: Infiltrative articaine achieves pain management comparable to that of nerve block lidocaine.

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KEYWORDS:

Oral Surgery; Articaine; Lidocaine; Anesthetics; Third molar.

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INTRODUCTION

Dental Anesthesia is fundamental to achieve painless dental therapies, the patient's opinion concerning the procedure is strictly associated with their previous experiences with anesthesia. Local anesthetics are the most employed drugs in dental attention, for this reason, its study is relevant to expand knowledge about these fundamental medications used in dentistry.

Among the nerve block procedures, Spix-technique is widely used to achieve anesthesia of the inferior alveolar nerve, however, there is an error range that varies from 5 to 15%, or 15% to 20%, reaching even higher percentages when achieving dental pulp anesthesia, being the main reason for this the anatomical differences among patients.

Failure rates of up to 15-20% can be associated with poor technique, caused by the difficulty to accurately localize the neurovascular bundle[1]. Another limitation is the risk of intravascular injections, which can lead to systemic complications such as, cardiovascular and Central Nervous System toxicity, tachycardia, and hypertension[2].

15,3% of inferior alveolar block nerve injections could be related to a positive aspiration, it has been observed intravascular injection in 14.2% of cases when direct Spix-technique was used (where the needle moves directly to the site of contralateral block nerve) and 23.3% of cases when using indirect Spix-technique (where the needle penetrates from the same side of injection)[3].

The introduction of anesthetics with greater fat-solubility, such as articaine, makes one think that it could be a good alternative its infiltrative usage in any zone of the mandible, in contrast to other anesthetics that don't have the ability to penetrate through the compact mandibular bone, also it is metabolized by plasmatic esterases, so its life expectancy is shorter, avoiding systemic or toxic complications[4]. On the other hand, it would be an ideal technique for patients that suffer from blood dyscrasias, hemophiliac patients, or emergency attention in anticoagulated patients, where the use of nerve block techniques could signify a greater risk of complications[5,6].

The aim of this study is to evaluate whether articaine used with infiltrative technique is effective for mandibular third molar extraction that requires osteotomy and odontosection, compared to lidocaine used with nerve block technique.

MATERIAL AND METHODS

In vivo experimental design, parallel non-inferiority. It was conducted in the minor surgery ward of the Faculty of Dentistry of the University of Valparaíso. It was approved by the bioethics committee of the Faculty of Dentistry of the University of Valparaíso in 2015.

Participants were individuals with full mental capabilities who signed informed consent. This study adjusted to ‘‘The Council for International Organizations of Medical Science’’ (CIOMS)[7] research standards, and to the Declaration of Helsinki[8], also, it had the authorizations of the school director and the headquarters of the minor surgery ward.

50 surgical sites were studied, corresponding to 25 patients without comorbidities that attended the Surgical Ward of the University of Valparaíso with an indication of extraction of both inferior third molars, that required osteotomy and odontosection. Patients allergic to anesthetics drugs, lighter than 50kgs, with cholinesterase deficiency or atypical pseudocholinesterase, acute local pathologies or psychiatric disorders were excluded.

The main variable of this study was the effectivity of the anesthetic technique in the management of pain during the procedure. For this, the anesthetic protocol must allow beginning the procedure after 7 minutes of waiting after the injection of the anesthetic, and also, must not exceed the maximum amount of reinforcement determined for each anesthetic protocol. Additionally, age, sex and anxiety score were registered and evaluated by modified Corah test [9]. An anesthetic protocol was administered for the intervention of the right side of the mandible, and another for the right side, assigned randomly.

The first corresponds to nerve block technique using 2% Lidocaine with 1:100.000 epinephrine utilizing 1 tube and a half for the Spix-technique,

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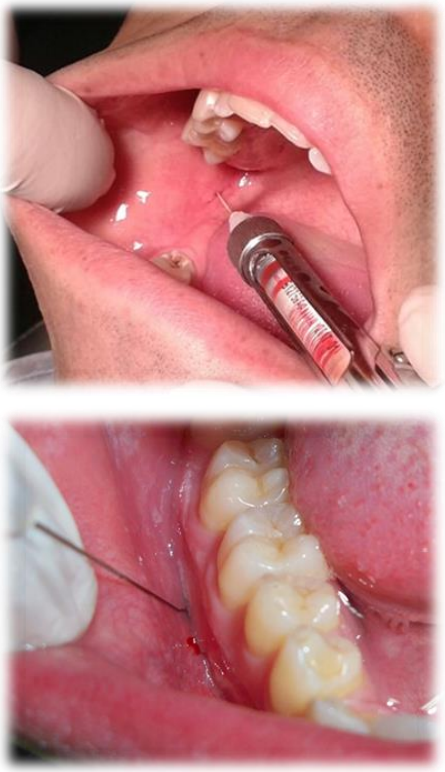
and a half tube for the buccal nerve, preserving maximum 1 tube for reinforcement. The second corresponds to Infiltrative technique using 4% Articaine with 1:100.000 epinephrine, injecting 1 tube and a half at the vestibule at the level of the first molar of the lower hemiarch , and a half tube at the level of the local lingual mucosa of the third molar, preserving maximum 1 tube for reinforcement. (Figure 1)

Figure 1: Anesthesia Tecnique



A

B

Lidocaine Nerve block protocol (A) and Articaine infiltrative protocol (B). Self-made images obtained with the prior informed consent and acceptance of the individual to take photographic records.

In case of not achieving anesthesia of the nerve posterior to 7 minutes after the injection, or in case of exceeding the maximum amount of reinforcement, the "failure protocol", consisting of the anesthetic injection of 2% Lidocaine, was activated, with a nerve block Spix-technique.

After reviewing the medical history, explaining the procedure, and reviewing the informed consent, the Modified Corah Test was conducted, to size anxiety caused by dental attention. Only those with low or mild anxiety could participate in this study.

Following the application of the anesthetic technique, 7 minutes passed and it was verified that there was no painful sensation at the puncture at the level of the mucosa and periodontium of the second molar (buccal nerve and inferior alveolar nerve), nor the canine (inferior alveolar nerve) nor lingual of the third molar (lingual nerve). After the first surgical procedure was completed, the other surgical protocol for the intervention of the contralateral tooth was executed.

Characterization of the sample was performed using means and proportions. The comparison of effectivity among anesthetic protocols was evaluated with a Z test for comparison of proportions, as well as its comparison with literature. Shapiro-Wilk normality test was conducted for the score variable in the anxiety test, where the groups where the protocol used failed were compared with those where it was successful, for which a comparison of means of independent samples was carried out. The results were analyzed with a confidence level of 95%.

RESULTS

The sample consisted of 25 individuals, where the mean age was 22.4, with a range of 16-35. A total of 50 surgical interventions were performed, 25 of those using infiltrative Articaine, and 25 using nerve block Lidocaine (Table I)

Table I: Characterization of the Sample

Sample	Individuals n=25	
Sex	Female 22 (88%)	Male 3 (12%)
Age	Mean 22,4 (16-35)	
	Interventions n=50	
	Tooth 17	Tooth 32
Infiltrative Lidocaine	12	13
Nerve block Articaine	13	12

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When comparing both anesthetic protocols, it was found that the effectiveness of nerve block lidocaine was 76%, meanwhile, infiltrative Articaine was 92%, not existing a significative difference among them. (Table II)

Table II. Comparison of Effectiveness of Anesthesia Protocols

Effectiveness	Anesthesia Protocol		
	Nerve block Lidocaine	Infiltrative Articaine	P Value
Efective	19 (76%)	23 (92%)	0,12
Non Efective	6 (24%)	2 (8%)	

When comparing the proportions of cases where both protocols were effective with what is described in the literature, there were no significant differences. (Table III)

Table III. Comparison of effectiveness with Literature

Anesthesia Protocol	Result	Literature	P Value
Nerve block Lidocaine	76%	85%	0,12
Infiltrative Articaine	92%	93%	0,69

When comparing means of anxiety test scores of cases where the technique was effective, independently of the technique used, there was no significant difference between the scores of those where the technique used failed. (Table IV)

Table IV. Pre-Surgical Anxiety and Effectiveness of the Anesthetic Technique

	Efective	Non Efective	P Value
Anxiety Test Score	10,45	11,6	0,65

DISCUSSION

The main finding of the present study is that the anesthesia protocol was successful in 92% of the infiltrative articaine group, against 76% of the nerve block lidocaine group, equivalent to a difference of (+ 16%) in favor of articaine, which has no significant differences.

The findings of this study are related to those reported by Berlin ((+12%), Mikesell (+9%), Claffeley (+15%), Khoury and Sierra (+6%), who did not report significant differences; however, Costa (0%) and Ruprecht (0%) reported same success rate. Although, this differs from that reported by Kanaa (+26%), Robertson (+30%) [10].

Regarding the failure rate of the nerve block technique, literature has reported an error rate that varies between 5 and 15%[11], and 10 to 15%[12]. In this study, the failure rate was 25%, greater than those reported in the literature, although this is not statistically significant. Of the 6 cases in which the technique failed, 4 failed anew when repeating the technique.

Regarding the infiltrative technique failure rate, literature has reported a failure of 7%[13]. In this study, the failure rate was 8%, with no significant difference.

There are diverse reasons that explain why a local anesthetic technique could fail when in dental treatment, among them are: Patient's anxiety, incorrect technique, anatomic variations, and bone density[14].

Amid those patients classified as anxious, 30% could state that the anesthesia is not enough when in fact it has been successful[14]. However, in this study, there was no significant difference in the rate of success of the technique between those individuals with the highest score on the anxiety scale.

Anxiety could be related to lack of depth of the anesthesia or the anesthetic block sensitivity, according to the type of fiber: motor, touch, pressure, pain, and its characteristics[15]. As a non-methodological observation, it was found that many patients are unsure or find it difficult to discriminate the sensation of pain or pressure they are feeling.

There were no immediate complications reported such as intravascular injection, or in the posterior appointments, which could be related to the little variability of age, the sterile surgical environment in the ward, and surgeons with experience regarding the anesthetic technique, which reduces risks.

Among the limitations of this study, the sample size was adjusted to the number of patients cared for in the ward. Sex was determined by the profile of the patients treated in the minor surgery ward at the University of Valparaíso. The investigation was carried out with a single-blind,

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given that the surgeon knew anatomical sites and where infiltrative lidocaine is contraindicated.

Amongst the methodological suggestions, it is recommended to carry out the test with larger physical spaces that allow patient comfort posterior to the intervention; It would be ideal to keep the patient in a place to clinically evaluate the attenuation of the anesthetic effect over time. Additionally, it is recommended to use specific pain scales for a deeper understanding of the subjective feeling of pain.

CONCLUSION

Infiltrative articaine achieves pain management comparable to that of nerve block lidocaine.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest with respect to this article.

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RESEARCH ARTICLE

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IN VITRO DETERMINATION OF MINIMUM LENGTH PENETRATION OF A 27G MONOJECT NEEDLE TO IRRIGATE THE MAIN CANAL, AVOIDING EXTRUSION.

Carlos Marchant[1], Fernando Aguirre[2], Eduardo Márquez[3], Marcelo Tapia[4]

ABSTRACT

Objective: To determine the minimum length penetration required of a monoject27G needle to irrigate the main canal, avoiding extrusion.

Materials and methods: 52 teeth were used, decoronated and worked at equal lengths at a MAF 40. They were randomly assembled in 3 molds and irrigated with diluted Omnipaque. Central trend values were calculated using descriptive statistics. Barlett, Shapiro Wilk and ANOVA one-way tests were applied to analyze statistically significant differences. Subsequently the logistic regression of Oswell-Lemeshow was calculated to look for causality between variables.

Results: Statistically significant results show that at higher needle penetration, the probability of extrusion is greater; regarding minimal length penetration to prevent extrusion, the safest length to irrigate is working length (WL) -4mm.

Conclusions: The minimum length penetration to irrigate preventing extrusion is WL-4 mm.

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KEYWORDS:

Endodontic irrigation; sodium hypochlorite; extrusion; endodontic treatment; endodontic.

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INTRODUCTION

Cleaning and shaping are the most important goals in endodontic treatment; however, it has been proved that there is a 35% of untreated root areas inaccessible to mechanical instrumentation. To achieve the optimal cleansing of remaining tissues and bacterial biofilms, the root canal must be shaped so it can allow an effective flow of irrigating solutions, without damaging periapical tissues.[1,2]

Nowadays, the most used irrigating solution is sodium hypochlorite (NAOCl), although reports of its toxicity on soft tissue is well known.[3] Using NAOCl implies, inherently, an accident risk during treatment. Extrusion to the periapical tissue might be one of those, causing severe pain and tissue edema, that could extend to lips, cheeks, and the infraorbital region. In addition, there is a possibility of secondary infection or the dissemination of a pre-existent one.

A recent poll showed that almost half of all endodontists inquired (42%) in the United States had experienced at least one NaOCl related accident during their professional practice.[4]

To know an optimal penetration length of the needle of endodontic syringes may help to minimize the risks of clinical actions.

This study aims to determine in-vitro the minimum length penetration required of a monoject 27G needle to irrigate the main canal (using sodium hypochlorite), avoiding extrusion. As hypothesis, it was solved that the minimal length required is a WL -2mm.

MATERIALS AND METHODS

The design of this study was a double-blind, random cluster in vitro experimental analytical type.

Apical extrusion is considered an undesired event during clinical procedure, it provokes pain and damage to the patients, which violates the non-maleficence bioethical principle, so this study was conducted in-vitro.

The sample was composed of extracted tooth of patients of the School of dentistry of the University of Valparaíso, with an indication for tooth extraction, from June to July of 2017. The patients were asked to sign an informed consent to participate in this study.

The inclusion criteria were: single rooted permanent and mature teeth, both maxillary and mandibulary, with straight canals, of patients over 18 years old with decision-making abilities and an indication for tooth extraction, regardless of prior systemic health issues.

Exclusion criteria were: crown-root length minor to 15 mm, internal root resorption, apicoectomised teeth, calcified or atresia-compromised root canals, previously treated teeth, teeth with an Initial Apical File over #30 and severely curved canals according to Schneider's classification (1971).

The variables of the study were: needle penetration (independent variable) measured in millimeters, and apical extrusion (dependant variable) measured in its occurrence with a yes/no.

The sample size of this study for each group was 13 teeth, this number was obtained by a hypothesis contrast calculation for two means. In total, there were 4 groups, so the total number of teeth studied was 52.

Finally, the sample obtained was of 52 maxillary teeth that fulfilled the inclusion criteria. In the first instance, each tooth was disinfected and partially de-crowned with a steel disc using low-speed creating a cut transversal to the tooth mayor axis, at the pulp chamber. Using this method a 15mm working length was obtained in every case. In those cases where the working length was lost due to excessive cutting, it was reestablished using the restorative adhesive technique.

Later, the apexes were sealed with wax, allowing its biomechanical preparation. K-files (Dentsply Maillefer) were used with Roanne's balanced forces technique until a #40 MAF (master apical file) was reached, lastly the final irrigating protocol of the University of Valparaíso was used.[5]

Each tooth was placed with a 2x2mm pH paper in

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its apical foramen, formwork in 5mm pink wax, and filled 10mm with cast and sawdust, obtaining a stable structure.

Once all units were included and executed by the standards set, all teeth were aleatorily assigned to the monoject27G's length that they would be irrigated at, conforming 4 groups: circle group (WL-1mm); square group (WL-2mm); cross group (WL-3mm) and triangle group (WL-4mm). After the lengths were assigned, new randomization was performed to determine each teeth position inside the matrices, in three of them, two of 17 teeth (matrix B and C) and one of 18 (matrix A).

As the irrigating agent, an iohexol contrast medium diluted in a 300 mg/ml solution was selected to equalize the viscosity of sodium hypochlorite.

Corresponding millimeters to each length were fixed with a rubber bumper attached to each needle, marked with its corresponding symbol. 1mml of the irrigating solution was used for each tooth, using a Monoject Endo 27 G handled by a third blind operator.

Extrusion measurement was carried on by a blind operator through optic microscopic of the pH paper, this allowed to observe whether there were any pH changes, indicating any extrusion of the contrast medium. Results were recorded in a simple incidence spreadsheet.

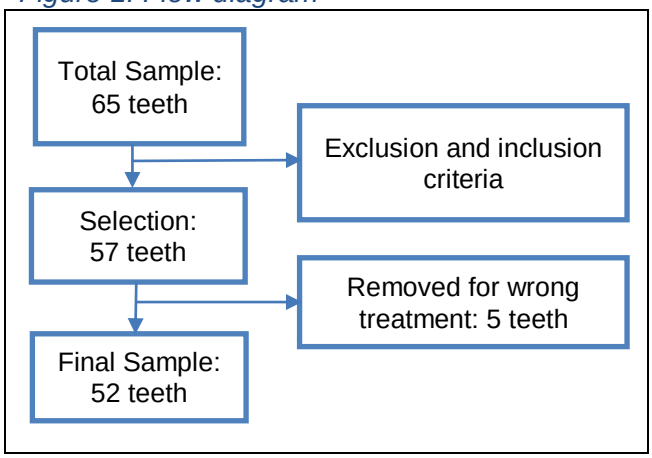
Collected data was automatically placed into Microsoft Excel 2010 by the Python software, obtaining a database. Later, this was complemented with data obtained with the microscope.

With this base, central tendency values were calculated using descriptive statistics. Then, Barlett Shapiro Wilk and one-way ANOVA test were applied to analyze statistically significant differences. Then, Post-hoc analysis of Tukey's test and Scheffe's test were used to evidentiate which groups presented differences. Oswell-Lemeshow regression logistic was calculated to look for any causalities between variables.

RESULTS

52 teeth were used; in the Figure 1, it is shown how the final sample was reached. (Figure 1)

Figure 1: Flow diagram



When evaluating teeth posterior irrigation, there were 11 extrusion cases observed in the WL-1 group, 5 cases in the WL-2 group, and 1 case in the WL-3, resulting in a total of 17 extrusions. There were not any extrusions in the WL-4 group.

By submitting the data of the extruded and non-extruded groups to one way ANOVA, there are statistically significant differences found in each group (non-extruded group p-value=0.00; extruded group p-value=0.0006). Post-hoc analysis of the non-extruded group revealed that the WL-4 group presented statistically significant differences when compared to all other irrigating groups.

Applying a logistic regression model with needle penetration and extrusion as variables, statistically significant results are obtained. (Table I)

When analyzing data through ROC curve, it is inferred that the experimental model serves to predict 80.9% of extrusion cases.
From the ROC curve, the model predicts that as the penetration of the needle into the conduct decreases, the range of extrusion probability decreases.
When analyzing data through ROC curve, it is inferred that the experimental model serves to predict 80.9% of extrusion cases. (Figure 2)
From the ROC curve, the model predicts that as

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the penetration of the needle into the conduct decreases, the range of extrusion probability decreases. (Figure 3)

Table I. Oswell Lemeshow logistic regression for Penetration-Extrusion variable.

Penetration	Odds Ratio	Standard Error	P value	95% confidence interval
WL-2	.1136364	.1087536	.023	.0174137 .7415560
WL-3	.0330579	.0359377	.002	.0039257 .2783737
				Pron>chi2 .0008 – Pseudo R2 .2635

Figure 2. ROC curve

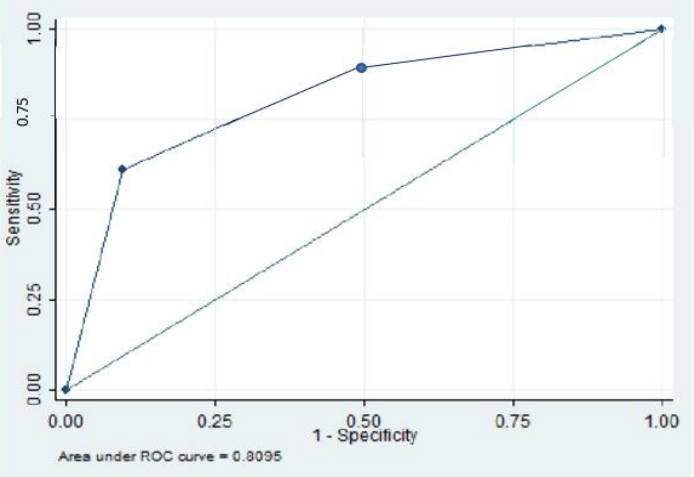
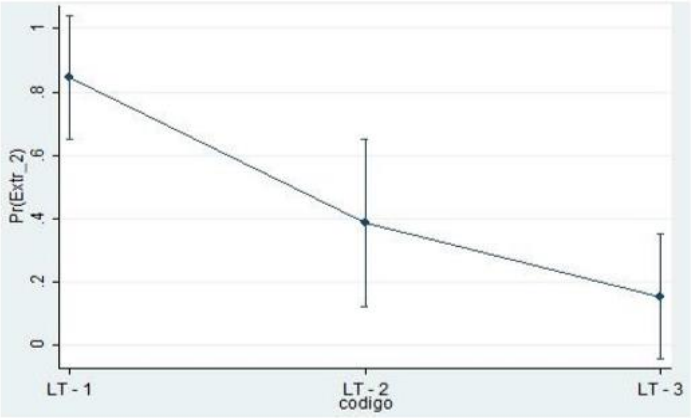


Figure 3. Probability of extrusion per group of needle penetration.



**LT = WL*

DISCUSSION

According to our findings, the higher the needle penetration, the higher is the probability of extrusion, being WL-4mm the safest length for irrigating since it did not present any positive case of extrusion. This response is explained by the location of the bubbles generated inside the canal. E.E Zukoski (1966) evidences the presence of the vapor lock effect with the bubble in an intimate relationship with the total wall, preventing the passage of liquid: in these cases, there would be no extrusion. The bubble may also not contact 100% of the canal surface, allowing the irrigating flow towards the apex.[6] This is the probable situation for those cases where there was a positive extrusion.

According to Boutsioukis et al. (2013), as the needle is closer to the apex there is an increase in the pressure exerted by the liquid with an increase in the probability of extrusion: the results are consistent with this statement, added to the air lock factor that does not obliterate the canal.[7, 8]

The work with freshly extracted teeth made it possible to reproduce, as closely as possible, a real clinical condition as to what it is the superficial energy present in a root canal. In regards to periapical tissue and its simulation, this study used an in vitro mounting configuration similar to those used in previous investigations[9]. This consisted of extracted human teeth with its root submerged into wax and cast, simulating periapical tissues, and thus creating a closed working system that has been endorsed in the literature.[10, 11, 12] However, the mounting structure used in this study was standardized in a way that all samples were equally affected. Besides, the development of a contrast medium with similar viscosity to sodium hypochlorite allows equating its physical characteristics, eliminating errors attributable to the instrument.

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On the other hand, the diameter of the root canal ensures a bigger width than that of a monoject 27G, that is, the obliteration at the time of downloading the medium is not a factor to consider, added to the fact that the tip has a lateral discharge that reduces the probability of extrusion.

One of the limitations of this study is when extrapolating its results to an in vivo scenario. The adverse reaction to the extrusion of sodium hypochlorite, and its sequelae, make randomized controlled clinical trials unethical unless chemically inert irrigants are used.[13] Also, a common limitation, both in this and others, in vitro studies[14, 15] is that it is unknown whether the resistance exerted by the materials is greater, less than or equal to that exerted by periapical tissues in vivo.

In addition, it is necessary to consider the variation that exists among files used in Biomechanical Preparation (BMP). It's of our knowledge that files used in this study are #40 with a 0.2 taper and ±0.02mm tolerance (ADA specification N°20), for that reason the diameter can vary ±0.02 mm between one file # 40 and another, depending on its fabrication and cannot be handled in any clinical nor experimental level; If we consider that our study performed 52 BMP and used 13 different #40 files, there is a high variety in the diameters of the prepared ducts that cannot be objectified. Even if this variety might be millimetric, it directly influences when comparing flow dynamics in two conduits that were prepared by different files of the same number, since according to Ohm's Law, if the diameter of a duct increases twice, the flow increases 16 times, reaching important differences in the resistance.

Besides, the model of this study does not evaluate the sodium hypochlorite extrusion during the treatment, since only extrusion posterior irrigation was measured. According to Hulsmann et al (2009)[16], the extrusion of the irrigant can clinically occur in small amounts during instrumentalization of the root canal, independently of the type of instrument and technique of preparation, not being limited to the moment of irrigation.

To this, we reckon that the adverse effects of sodium hypochlorite have been evaluated in the literature dependant of different concentrations[17,18] but further studies are necessary that relate this effect with the amount of irrigant extruded, allowing to establish in what degree adverse effects are volume dependant and what is the minimal amount that could generate symptoms and physical signs.

Considering that our study detects the presence of extrusion but not its volume, we suggest that further studies are conducted on this topic using extrusion as a quantitative variable, thus being able to determine if the volumes of extruded hypochlorite have significance from the clinical point of view, and to what extent the volume is attributable to the adverse effects described in the literature.

If we analyze the anatomical variability of the duct system, either in diameter or presence of accessory and/or lateral channels, we can deduce that even when results prove that the presence of extrusion is closely related with needle penetration, there might be other factors such as tolerance among prepared canal roots, offering a variable range of resistance for the same file, keeping in mind the resistance exerted by the periodontal tissues that must be overcome.

CONCLUSION

The initial hypothesis was rejected since as the needle is closer to the apex there is an increase in the pressure exerted by the liquid with an increase in the probability of extrusion. This statement added to the air lock factor that does not obliterate the canal,[17] enables establishing that the minimal length required for irrigation without extrusion is WL-4mm, not WL-2mm.

CONFLICT OF INTEREST.

The authors declare not having any conflict of interest.

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RESEARCH ARTICLE

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IN VITRO EVALUATION OF DENTINE ADHESIVE RESISTANCE AFTER APPLYING TWO IRRIGATION SEQUENCES

Carlos Marchant[1], Brito Daniela[2], Bruno Durán[2], Juan Carlos Vidal[3], Ángel Navarrete[5]

ABSTRACT

Objective: To evaluate the bond-strength (MPa) of an adhesive system to intracameral dentin after 2 different endodontic irrigation sequences.

Material and Methods: An in vitro double-blinded experimental cross-over study was conducted. 23 teeth were extracted, and sectioned, exposing the pulp chamber. Samples were separated into three groups: the first group (control) used a 0.9% saline solution; in the second group, the conventional sequence of irrigation of the University of Valparaíso was employed (5% NaOCl - 18% EDTA-0.9% saline solution); and finally the third group, the experimental sequence was employed (5% NaOCl, 18% EDTA and 15% saline solution). The sealing technique was then conducted using universal 3M ESPE Single Bond adhesive and Filtek Z350 resin. Subsequently, the bond-strength test was performed in a microtensile, until fracture.

Results: The control group had the highest values of frequency of adhesive and cohesive failure compared to the conventional and experimental group but presented a statistically non-significant association. As for the irrigation protocols, they did not show a major difference in their adhesive bond-strength when compared.

Conclusion: The results of this study conclude that the adhesive resistance is not significantly modified by different irrigation protocols.

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KEYWORDS:

Endodontics; irrigators; coronal sealing; adhesive systems; hybrid layer

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INTRODUCTION

Cleaning and shaping of the Root Canal System (RCS) is one of the main predictors of endodontic success[1], however, it also may alter the proper coronal sealing. Immediate coronal sealing after endodontic treatment is a powerful tool to avoid microleakage and to enhance prognosis. Nowadays, coronal sealing is based on different adhesive systems allowing more dental tissue's conservation, increment the fracture resistance of the remaining dental structure, optimize the coronal restoration's retention and a significant reduction of microleakage through micromechanical bonding of restorative materials to previously conditioned dentin. Nonetheless, dentinal adhesion might be jeopardized by a numerous factors, being the effect of irrigating solutions on teeth during the endodontic treatment one of the steps which may alter the chemical composition of the intracameral and radicular dentinal surface, affecting its interaction with the adhesive materials and their sealing[2]. Adhesive failures can also be found in different levels. Cohesive failures are the most important type of them, because they can occur at the composite-adhesive level[5], on the other hand, some adhesive failures may occur among two different structures (dentin-adhesive interface or dentin-resin interface)[6]. Sodium Hypochlorite (NaOCl) is a well-known endodontic irrigant which can cause degeneration of the dentin due to dissolution of collagen, carbonic bonds' breaking and protein's structure disorganization. Decrease of bonding force between dentin and adhesive systems is also described, this might be explained because of the collagen's removal from dentin surface.

Ethylenediaminetetraacetic acid (EDTA), is another well-known endodontic irrigant, which has a chelating effect, enhancing dentin demineralization. This compound is used mainly during the final irrigation of endodontic treatment to permeabilize canals and to remove the smear layer once the biomechanical preparation is concluded[7].

Coronal sealing and bonding among the final restoration and remaining dental tissues are still problem to practitioners[2,3,4]. There is lack of information of dentin surface and adhesive system reaction after different endodontic irrigation protocols, so it is possible to think, different protocol sequences will have diverse effects and therefore, various bond strength outcomes. This study aims to evaluate the adhesive bond strength (MPa) of an adhesive system to intracameral dentin after the application of 2 different endodontic irrigation protocols.

MATERIAL AND METHODS

An in vitro double-blind experimental cross-over study was conducted to estimate the adhesion resistance of the adhesive bond after the application of two irrigation protocols and a control group. (Figure 1)

23 teeth were extracted for therapeutic reasons at the surgery clinics of the Faculty of Dentistry of the University of Valparaíso, informed consent was obtained from all participants before the extractions, to donate the teeth to this study.

All the extractions were effectuated within a period of no more than 6 months, during 2017, they did not present vertical fractures, previous endodontic treatment, or large coronary destruction that compromised the cervical third. They were placed in distilled water posterior the extraction. Each tooth was cut into thirds in mesiodistal and crown-apical directions, taking the middle third of each tooth as a reference, to expose the roof and floor of the pulp chamber. Two more additional cuts were performed, the first one 0.5mm apical to the pulp chamber ceiling mesio-distally and horizontally, the second one 0.5mm coronal to the floor of the pulp chamber. The teeth were sectioned with a low speed diamond disc saw to expose the walls of the pulp chamber. (Figure 2)

With the pulp chamber exposed, all pulp was eliminated using a sharp end manual instrument. Next, samples were divided into three groups of 23 cuts each. Two of the groups were operated on

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with a different irrigation sequence and the remaining group (control) received only irrigation with 0.9% saline solution (Figure 1). Following this, the adhesive system and composite resin were applied.

Walls of the pulp chamber were washed with 10mL of distilled water at room temperature and blown dried for 5 seconds. Later, they were irrigated with water for 30 seconds and dried with sterile gauze on the surface, a layer of 3M ESPE Universal Single Bond Adhesive (self-etching) was applied with a microbrush, eliminating excesses with a jet of air, later then each layer was photopolymerized for 20 seconds with an Ivoclar Vivadent LED curing lamp model Bluephase NMC with a light intensity of 800 MW / cm2 calibrated between 430-490 nm. The insertion of the composite resin, Filtek Z350 (3M) was carried out with a manual instrument for resin (Hu-Friedy), placing layers of 2 mm thickness to seal the pulp chamber and each layer was light-cured for 40 seconds with the same lamp of light curing. Then a cut with a low-speed diamond saw was performed, pieces of 1mm by 1mm at the level of the walls of the pulp chamber. The cut was made in a vestibular side-lingual side direction (horizontal), including dentin and composite resin (Figure 2 and 3). Each specimen was labeled according to an endodontic irrigating group; then the bond-strength test took place in a microtensile microtensiometer OM 100, which is a tool manufactured by BISCO Inc, used to test the resistance of adhesives.

Samples were obtained in an environment with controlled temperature and humidity.
The specimens were fixed at their ends to a test device, using cyanoacrylate. Each one of these were attached to the fixtures of the universal testing machine; tension was performed, and adhesive failure was calculated until the specimen fractured. Each failure was expressed in megapascals (MPa), being able to evaluate the adhesive bond strength. After the experimental phase, the teeth were destroyed according to the biosecurity protocol.

Figure 1: Study groups distribution

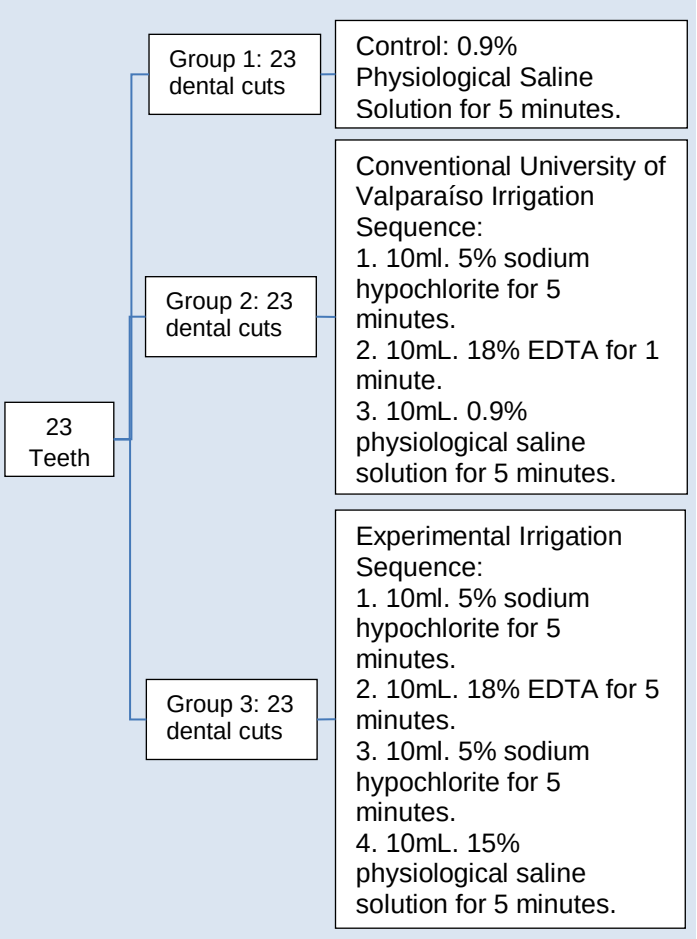
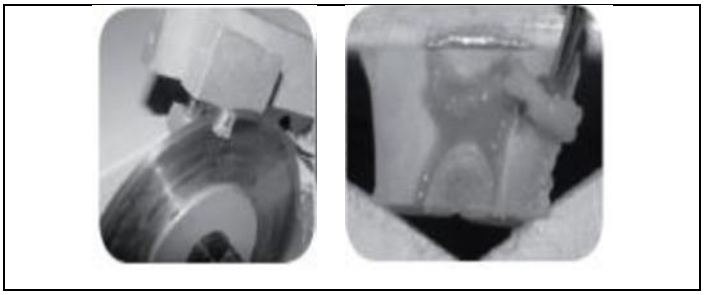
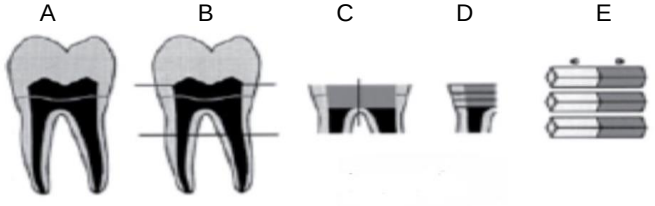


Figure 2: Sectioned teeth



Left: low speed diamond disc; Right: exposure of the walls of the pulp chamber.

Figure 3: Schematic Representation of the sample collection



(A) Mesiodistal cut of the teeth. (B) Pulp chamber cut. (C)Placement of intracameral composite. (D)Cutting of specimens. (E)Finished sample, 1mm area.

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Table IV: Chi-square value for association of variables.

Failure	Control	Conventional Sequence	Experimental Sequence	Total
Adhesive	0.0552	0.0464	0.0936	0.1953
Cohesive	0.0170	0.0091	0.0233	0.0494
Mixed	0.0398	0.0416	0.0755	0.1569
Total	0.0112	0.0971	0.1924	0.4016

The variables to be measured in this study were irrigants, adhesive failure, and adhesive bond strength (MPa). Data were registered into an excel database, and the statistical analysis was carried out later with the Stata 13.0 software. The Bartlett and Shapiro-Wilks test were used to determine homogeneity and normality and then a test of equality of medians to determine if there were significant differences between groups. Hypothesis testing for two means was employed. A significance level of 95% was used with a statistical power of 90%. The standard deviation was 1.27 MPa and a minimum difference of 0.315 MPa.

The evaluators were calibrated on what should be considered an adhesive failure, based on operational definitions. Two were the evaluators of the failure, for this reason, the Kappa concordance test was performed, and a minimum of 0.81 was considered to be acceptable.

For the bonding strength test, there was not required the calibration due to the use of microtensile.

RESULTS

During the experimental phase occurred losses both in the conventional irrigation sequence and control group, attributed to fractures of the test bodies at the time of their installation in the microtensile. The data obtained from the measurements with the microtensile, posterior to the bond-strength test in Mpa. (Table I)

The collected data was grouped according to the type of failure they presented, as shown in Table II. (Table II)

Table II: Frequency of failures associated to the irrigation sequence.

Failure	Control	Conventional Sequence	Experimental Sequence
Adhesive	67	76	85
Cohesive	15	2	6
Mixed	18	22	9

The chi-square test was performed to determine the association between the failures and the irrigation protocol and to obtain the observed and expected frequencies (Table III).

With these data, it was possible to arrive at the calculated chi-square value, which was calculated with a significance level of 5% to compare it with the critical chi-square.

The data obtained (Table IV) indicate that the calculated chi-square value is less than the critical chi-square value, for that reason there is not a statistically significant association between variables.

Table III: Observed and expected frequencies for association between variables.

Failure	Control		Conventional Sequence		Experimental Sequence		Total	
	F.E	F.O	F.E	F.O	F.E	F.O	F.E	F.O
Adhesive	0.50	0.67	0.59	0.76	1.18	0.85	2.28	2.28
Cohesive	0.20	0.15	0.24	0.20	0.49	0.60	0.95	0.95
Mixed	0.28	0.18	0.33	0.22	0.67	0.90	1.30	1.30
Total	1.00	0.22	1.18	1.18	2.35	2.35	4.53	4.53

F.O = Observed frequencies; F.E = Expected frequencies

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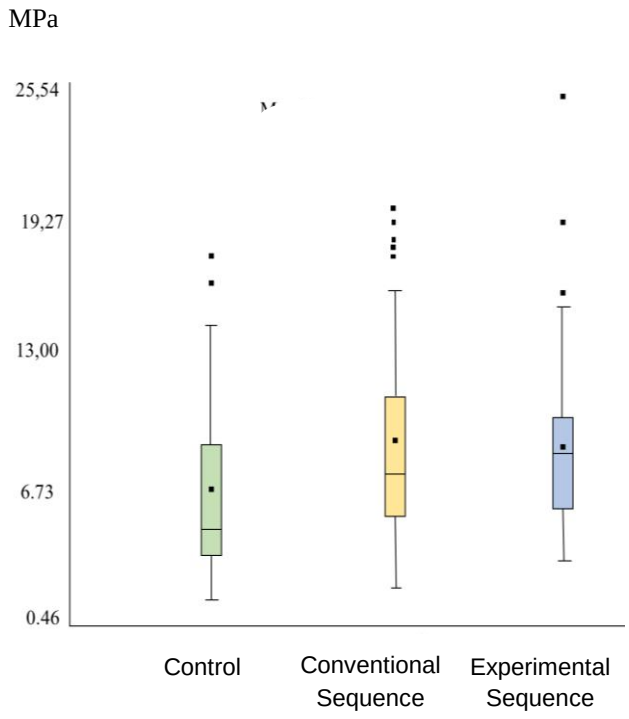
Figure 4 shows the data of the three groups did not follow a normal distribution. Control group and the conventional group are asymmetric upwards and the experimental group asymmetric downwards, however, there is no statistically significant difference among bond-strength. (Figure 4)

Figure 4: Distribution of the sample data



DISCUSSION

The main finding of this study is that the implementation of different sequences of irrigations during endodontic treatment has no clinical relevance on bond-strength (Mpa).

The design of this study had the benefit of using the same body of experimentation subjected to the two irrigation sequences used, plus a control, therefore selection biases were avoided. Furthermore, investigators were calibrated on means of bond-strength obtaining a concordance of 0.81, also it was used a OM 100 microtensile, which is considered a low cost, safe, easy to handle, and widely used in dentistry investigation tool[8].

The results of this study indicated that the control group had the highest values in the frequency of adhesive and cohesive failure compared to the conventional and experimental group, but even when it has a statistically non-significant association, its clinical relevance must not be disregarded, due to this irrigation protocol may be altering negatively dentin by the use of less NAOCl, avoiding a coronal sealing, diminishing prognosis, furthermore, independent of the use of NaOCl, adhesion force tends to decrease after an endodontic treatment, reporting a decrease of up to 23% in the bond-strength[4].

The results of this study revealed that the control group showed significantly lower adhesive bond strength. As for the irrigation protocols, they did not show a major difference in their adhesive bond-strength when compared, this is consistent with what was expressed by Nagpal et al, 2014, where the EDTA-Hypochlorite combination showed a higher adhesive bond strength than controls with physiological saline solution in the use of self-etching adhesive, this may be due to the effect on the hybrid layer formed on the dentin surface, which would help improve adhesion[9]. However, these results are discordant with Carvalho et al. 2017, who stated that adhesive systems were not influenced by any endodontic irrigating protocol; Only the "adhesive system" factor was statistically significant. Compared to our results, where there is a difference between experimental groups, not being able to affirm that endodontic irrigation protocols affect the bond strength[10]. This could have happened because only the self-etching adhesive system was used. In different studies, conventional adhesives report high levels of bond strength compared to sixth and seventh-generation self-etching adhesives, due to the formation of water vesicles at the adhesive interface[11].

Cecchin et al., 2010, proposed that higher adhesion values are obtained using adhesive systems on dentin that has not previously been treated with NaOCl and EDTA irrigation solutions; the results on said study revealed the irrigation with 1% NaOCl during 1 Hrs (applied every 5 minutes) decreased the bond-strength 13. One possible explanation for this is due to the superficial morphology of NaOCL treated dentin,

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where the smear layer is not eliminated, and thus, dentinal tubules are not exposed, added to the formation of free radicals and oxidation produced by NaOCl on the tooth surface, where the presence of oxygen limits the properties and polymerization of adhesive systems.

Coinciding with what was described by Barutcigil et al., 2012 on the fact that if there is a relationship between the application of EDTA in different concentrations with a reduction in the adhesive bond strength, considering that any alteration of calcium (Ca^{+2}) concentration may cause a modification of the dentin composition and therefore in its mechanical characteristics and bond-strength to biomaterials[14].

Adhesion to dentin depends on the presence of residual Ca^{+2} in the bonding area, as evidenced by a significant reduction in bond-strength of some adhesive materials caused by partial depletion of superficial Ca^{+2}.[15] This is why the adhesive bond strength values are expected to be lower after endodontic irrigation protocols have been performed, this could happen due to differences in the methodology implemented[16].

Because this is an in-vitro study, results obtained do not represent conditions present in the oral environment, although dehydrated dentin shows a lower resistance to fracture[14], when conducting the study in a clinical context, multiple factors could also increase the possibility of adhesive failures, such as the filling materials used, or the application of absolute isolation.

It is suggested to expand the sample size and their characteristics to allow better evidence of coronal sealing after endodontic treatments. Also, it is recommended to consider the storage time in distilled water, because it could influence the results of bond strength and adhesive failure. It is advised to use different adhesive systems since their various compositions would alter dentin differently.

CONCLUSION

It is concluded that bond strength is not modified by different irrigation protocols, even when between groups there are significant differences. Factors that could explain these results were not using different adhesives systems that could have acted differently based on their different properties and interactions with dentin previously treated by the irrigation protocols, hence altering the function of the hybrid layer during postendodontic treatment restoration.

CONFLICT OF INTEREST

The authors declare not having any conflict of interest.

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RESEARCH ARTICLE

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PREVALENCE AND CLINICAL CHARACTERIZATION OF MOUTH BREATHING PATIENTS IN VIÑA DEL MAR AND QUILPUÉ, CHILE.

Finger Valentina[1], Henríquez Cristina [2], Muñoz Daniela[3], Alan Barraza[4]

ABSTRACT

Objective: This study aimed to clinically characterize and determine the prevalence of mouth breathing in the pediatric population.

Materials and methods: 383 students, aged 6 to 13 years, from public and private subsidized schools were analyzed. Clinical criteria were applied, and measurement of maximum nasal inhalation flow (PNIF) was performed to determine their breathing condition (Mouth, Nasal, or Mixed). Statistical analysis of the data was carried out through Stata SE 10.1, R-Cran 2.13.1, and Minitab 15.

Results: The prevalence of mouth breathers was 18,80%, mixed breathers 17,49%, and nasal breathers 63,71%. The most common facial characteristic was the presence of eye bags (82%) and dry lips (78%). The maximum nasal inhalation flow (PNIF) average registered in mouth breathing patients was 54,4 L/min, meanwhile in nasal breathing patients was 84,7 L/min.

Conclusion: Mouth-breathers are a relevant percentage of the examined population. Early intervention of pediatricians is transcendental for the diagnosis, derivation, and treatment of this syndrome in order to limit future complications.

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KEYWORDS:

Mouth breathing; prevalence; nasal inhalation flow; Chile

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INTRODUCTION

Human breathing is a basic function of life. If there is any obstacle that prevents nasal-breathing, survival will depend on the adaptation to oral or mouth-breathing. Mouth-breathing corresponds to a mechanism through which air is inhaled through the mouth instead of the nose.[1]

The nasal-breathing function not only concerns general doctors, but also pediatricians, otolaryngologists, allergy specialists, language-speech therapists, pneumonia-specialists, orthodontists, and maxillofacial surgeons; in other words, all those specialists involved in the nasal-breathing function and facial growth[2]. The early detection of the mouth-breathing syndrome is relevant since alterations of the anatomical structures involved, affect not only the orofacial condition of the child but also their systemic health, aesthetics, body posture, mastication function, language-speech function and personality disorders that could affect them for the rest of their lives.

Each member of the health team who examines the child in due course must be prepared to detect mouth-breathing and explain the parents the consequences if the syndrome is not corrected on time, considering that if the habit persists throughout the patient growing period, the alterations will worsen, and correction will become more complex.

The purpose of this study was to determine the prevalence of mouth-breathing in children and make a clinical characterization of it. However, the practical application of this article is to guide the general doctor, and especially pediatricians, for early diagnosis and treatment of this pathology.

MATERIAL AND METHODS

A prevalence study was conducted with students attending public and private subsidized schools in the communes of Viña del Mar and Quilpué in Chile. 6 to 13-year-old children were included, who at the moment of the examination did not have a cold, were not depressed, did not suffer from attention deficit disorder or mental retardation. Informed consent was obtained from the parents or guardians of the students, in addition to the consent of the children.

The examination included the clinical evaluation of the following intra and extraoral characteristics: Dark circles below the eyes (bilateral change of coloration of lower eyelid and of the periorbital socket, ranging from a violet tone to dark brown), Dry lips (dry and cracked lips), Forward posture of head (distance from cervical spine greater than 6 cm from the vertical tangent to the thoracic spine), Hypertrophy of tonsils (size enlarged tonsils), Paleness (abnormal loss of skin coloration), Lip gap (distance from the lowest part of upper lip to the uppermost part of the lower lip), Eversion of the lower lip (voluminous and outwards lower lip), Exposition of upper incisors (measured from the lowest point of the upper lip to the upper incisive edge), compression of maxilla (high palate or narrow base), Deep palate (high or ogival palate dome), Interposition of tongue in phoneme (usual forward push of tongue during speech), Protrusion of upper incisors (incisors in vestibular position), Gingivitis (pathological inflammation of gums), Oral biofilm (mixed heterogeneous microbial community -aerobic and anaerobic- surrounded by an inter-cellular matrix of polymers of salivary or microbial origin adhered over teeth surface), and Nasal permeability These were measured as can be appreciated in figure 1, by means of the maximum nasal inhalation flow using a Portable Nasal Inhalation Flowmeter or PNIF (In-Check Nasal® manufactured by Clement-Clarke International) imported from England. (Figure1)

Figure 1: Collection of clinical characteristics to reach a diagnosis:

A. Clinical examination; B. Nasal permeability record. A + B = Diagnosis.

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By nose-breathing it was considered the free flow of air through the nasal and nasal-pharynx canals; by mixed-breathing, the partial flow of air both through nose and mouth –being this condition temporary or permanent, and by mouth breathing, the mechanism through which air is inhaled through the mouth instead of the nose (1) in a permanent way. The diagnosis of mouth-breathing was carried out according to the clinical criteria presented by Treviño-Salinas (3), in other words, if a child manifested ten or more of the clinical characteristics described above (mouth breathing always present) they were considered a mouth-breather. The qualitative variables were expressed by tables of absolute and relative frequency. Odds Ratio (OR) values were estimated, with their respective confidence intervals at 95% as an association measure for the condition of mouth-breathing, according to the different variables measured. For quantitative variables, descriptive measures were estimated. Fisher's exact test, Pearson's Square-Chi, Mood's median, and Kolmogorov-Smirnov test were applied, according to the behavior of the variables. They were considered significant when the p-value was less than 0.05. The analysis of the database was done using Microsoft Excel 2007, R-Cran 2.13.1, and Minitab 15 software.

RESULTS

The examined sample was composed of 383 students, of which 51% were male and 44.9% female. The average age of children was 9.9 years (SD=2,3). 73.1% of these children attended private subsidized schools, and the remaining 26.7% public schools.

This study determined a prevalence of mouth-breathers of 18.8% [14.9 – 22.7%], nasal-breathers 63.7% [58.89 – 68.52%] and mixed-breathers, 17.5% [13.69 – 21.30%]. The prevalence of characteristics and odds ratio showed on table I. (Table I)

The clinical examination revealed that the most common alterations in mouth breathers were the following: Regarding the maximum inhalation flow (PNIF) and the breathing condition of students, it was observed (Figure 2) an average

Figure 2. Graphic of the relationship between PNIF and respiratory condition

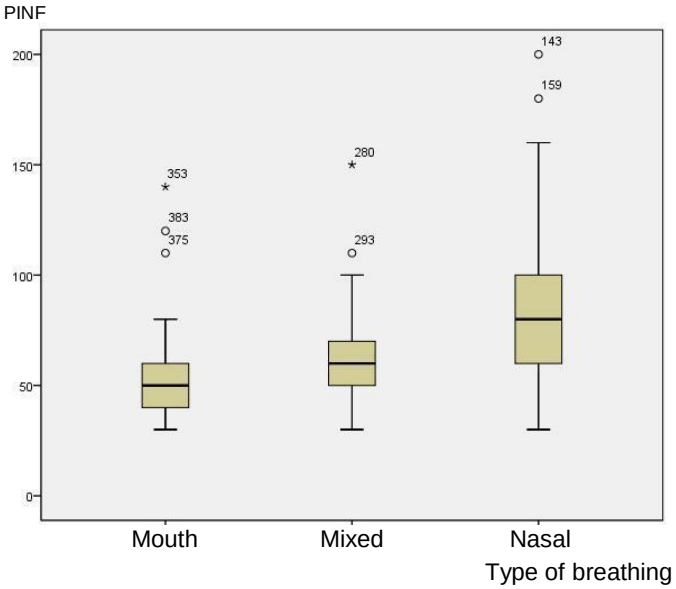

Table I: Prevalence of characteristics and odds ratio

Characteristic	%	OR	IC of 95% for OR
Oral Biofilm	94	6,9	[2,4 - 27,2]
Tongue interposition in phoneme	89	3,9	[1,8 - 10,0]
Lip Gap	88	129,7	[50,6 - 374,5]
Dark circles under eyes	82	4,8	[2,4 - 10,1]
Dry lips	78	4,6	[2,4 - 9,1]
Deep palate	72	4,3	[2,4 - 8,2]
Forward posture of head	65	4,6	[2,5 - 8,5]
Protrusion of upper incisors	63	5,9	[3,3 - 11,0]
Hypertrophy of tonsils	58	4,6	[2,5 - 8,5]
Gingivitis	56	3,8	[2,1 - 6,8]
Eversion of lower lip	51	33,6	[13,5 - 96,6]
Maxillary compression	49	3,3	[1,9 - 8,2]
Paleness	40	6,1	[3,1 - 12,2]
Exposition of upper incisor	18	19,5	[10,2 – 39,1]

PNIF of 5.4 L/min (SD=±20.6) in mouth-breathing children, 84.7 L/min (SD=±30.5) in nose-breathing children, and 62.5 L/min (SD=±21.8) in mixed-breathing children.

In PNIF it was measured no statistically significant differences among mouth and mixed-

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breathers (P-value=0.114).
However, there was a statistically significant difference in the distribution of PNIF measures between mixed and nasal-breathing children (P-value=0), and in the distribution of PNIF measures between mouth and nasal-breathers (P-value=0).

DISCUSSION

The syndrome of mouth-breathing is corroborated as a frequent condition in 6 to 13-year-old students, although this pathology could be easily detected at an early age by diverse medical professionals.

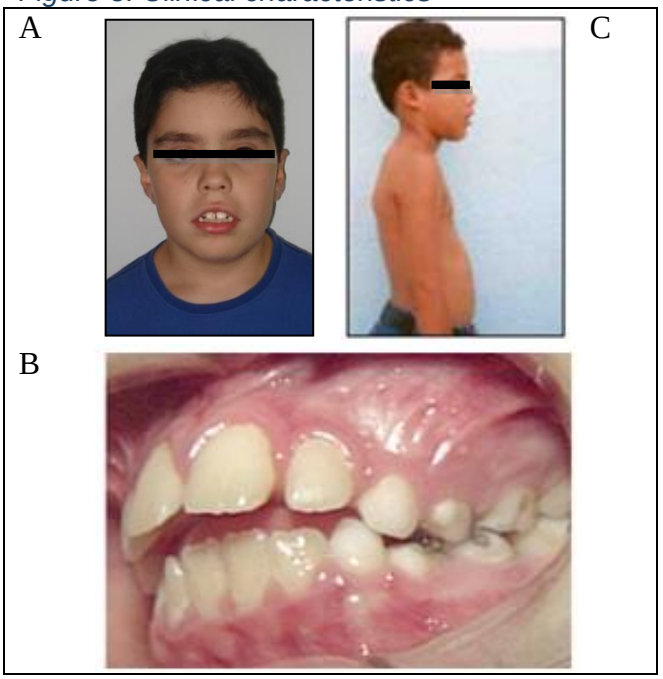
This study considered a standardized procedure for the examination and gathering of data, using a tester calibrated by experienced dentists to ensure a right and reliable diagnosis for each tested student[1,3]. Children sick with cold were excluded so as not to incorporate bias to the sample, as they show a momentary mouth-breathing pattern conditioned by the temporary obstruction of their respiratory airways.
Our results are similar to the ones detected by other studies carried out in Chile: in 1999 it was determined a prevalence of 23% of mouth-breathers among 1878 students evaluated4; meanwhile, in 2001 it was reported a 34.5% (5) of the prevalence of mouth-breathing in 5 to 17-year-old children in Santiago de Chile.

At an international level, a Cuban study carried out in 2009 reported a percentage of 24.7% of mouth-breathing children from 3 to 14-year-old. The most affected group was those from 6 to 11-year-old children[3].

Mouth-breathing is a syndrome that can have its etiological origin in obstructive problems, habits, and/or anatomy[6]. Thus, we see that it can be originated due to an atypical swallowing habit encouraged by finger sucking or prolonged use of pacifiers. In the beginning, this action is done consciously but afterward in an unconscious and repetitive way causing damage and setting up mouth-breathing in a permanent way4. Thus, if it is detected before the conscious stage the best therapeutic effect will be reached.
Some children breathe by the mouth due to obstructions and/or anatomy as they have a deviation in nose septum, enlarged turbinate bones, chronic inflammation, and congestion of the pharynx mucous membrane, allergies, and tonsils hypertrophy[7]. These children will need a more complex treatment, probably complemented with surgery or orthodontics to achieve correcting the bone and teeth alterations.

The consequences of mouth-breathing may be divided into extraoral, facial, and body clinical characteristics. Among the former, the following stand out: hypoplasia of the middle third of the face or maxillary atrophy, mandibular prognathism, weakness of facial muscles, dark circles below the eyes, narrow nostrils, hypo-tonicity of the upper lip, upper lip short and inefficient, lower lip thick and everted, and dry lips8 (figure 3A). Among the extraoral non-facial characteristics most frequently found in mouth breathing children the following stand out: abnormal body posture[8], a forward posture of the head, lordosis, protrusion of shoulders (figure 3C), elevation of scapulas and poor growth of thorax[9] which are easy to be detected and professionals must take into consideration to make a prompt diagnosis.

Figure 3: Clinical characteristics



A. Facial extraoral clinical characteristics; B. Protrusion of upper incisors; C. Protrusion of shoulders[11]

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Our results coincide with those reported by other studies, which conclude that the most common intraoral characteristics are the presence of oral biofilm, irritation of oral mucosa, hypertrophy of tonsils[9], maxillary compression, vis a vis bite (overjet and overbite equal to 0mm) or crossed bite2, descended tongue, deep palate, protruded upper incisors (figure 3B), atypical swallowing, gingivitis, and tendency to forward open bite[10].

It is extremely important to recognize the signs of mouth-breathing at an early stage, to give prompt treatment that will prevent further consequences, not only in the oral and maxillofacial zone but also, others described in the literature, such as hypoxia[12], speaking alterations, neck and/or back pain, the sensation of lack of fresh air, tiredness during physical activities, daily drowsiness and poor school performance[3].

Based on the correlation between low nasal permeability and the breathing pattern of the child (Figure 2), it would be recommended to perform follow-up examinations to achieve a more specific diagnose of mouth-breathing.

If detected prematurely, and derivation occurs promptly, the intervention of the pedodontist or orthodontist will be fundamental since these specialists will prescribe the required treatments to hinder and correct the alterations in orofacial growth and development. However, if the syndrome is detected late, a greater amount of health professionals will be required, and eventually, permanent consequences could remain, demanding higher economic costs.

CONCLUSION

Mouth-breathers are a relevant percentage of the examined population. Early intervention of pediatricians is transcendental for the diagnosis, derivation, and treatment of this syndrome in order to limit future complications.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest with respect to this article.

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Los autores interesados en publicar en Applied Sciences in Dentistry deben seguir las siguientes instrucciones:

1.- NORMAS GENERALES

1.1 Los trabajos enviados deben ser inéditos y no se permite su presentación en otra revista simultáneamente. Applied Sciences in Dentistry tiene todos los derechos autorales de los trabajos publicados, permitiendo su posterior transcripción citando la fuente (Appli Sci Dent). Ninguno de los autores será remunerado.

1.2 Applied Sciences in Dentistry recibirá artículos en idiomas español e inglés, siendo responsabilidad de los autores la correcta redacción del texto. Los artículos escritos en español deben enviar el resumen en inglés.

1.3 Los contenidos de los artículos publicados son de exclusiva responsabilidad de los autores.

1.4 Las fechas de recepción, revisión, aceptación y publicación online del artículo estarán al final del resumen, en la primera página de cada artículo al ser publicado.

2.- FORMATOS

2.1 Artículos de investigación: Incluye investigaciones en odontología y otras áreas complementarias y aplicadas para la disciplina. La extensión máxima del manuscrito será de 3500 palabras, además de hasta 40 referencias bibliográficas actualizadas y debidamente justificadas. Para estudios de carácter observacional (transversales, cohortes o casos y controles) se sugiere el uso de la Declaración de la Iniciativa STROBE (STrengthening the Reporting of OBservational studies in Epidemiology, Fortaleciendo el Reporte de Estudios Observacionales en Epidemiología). Para estudios de carácter experimental se sugiere el uso de la Guía CONSORT (CONsolidated Standards Of Reporting Trials, Estándares Consolidados del Reporte de Ensayos). No existe un máximo de autores.

2.2 Revisiones de la literatura: Pueden ser de tipo sistemática o narrativa. La extensión máxima del manuscrito será de 5000 palabras, incluyendo hasta 80 referencias bibliográficas actualizadas y debidamente justificadas. Para aquellas de carácter sistemático se sugiere el uso de la Declaración PRISMA (Preferred Reporting Items for Systematic reviews and Meta-Analyses, Ítems de Reporte Preferidos para Revisiones Sistemáticas y Meta-Análisis). No existe un máximo de autores para revisiones sistemáticas y meta-análisis. Para revisiones de tipo narrativa puede tener un máximo de 3 autores.

2.3 Reportes de caso: Casos clínicos o serie de casos deben ser de relevancia académica y científica, y debe cuidar el componente bioético. La extensión máxima del manuscrito será de 1500 palabras y hasta 20 referencias bibliográficas actualizadas y debidamente justificadas. Los casos clínicos pueden tener un máximo de 5 autores.

2.4 Cartas al Editor: Notas cortas, de un máximo de 1000 palabras, con opiniones de lectores sobre trabajos publicados o comentarios que se relacionan con la Revista, la investigación odontológica en general y/o algún artículo publicado en la revista. Puede tener hasta 5 referencias, no incluyen resúmenes y puede tener un máximo de 2 autores.

2.5 Comunicaciones breves: Una oportunidad para la presentación de observaciones preliminares o breves que no requieren de un artículo completo, con un máximo de 1000 palabras, y hasta 10 referencias. Un máximo de una tabla y/o una figura puede ser aceptada. No incluye resumen y puede tener un máximo de 3 autores.

3.- CONSIDERACIONES BIOÉTICAS

3.1 Todos los trabajos presentados que involucren estudios con seres humanos deben respetar las normas actualizadas de la Declaración de Helsinki.

3.2 En las imágenes, no se debe identificar el paciente, por lo que no deben aparecer nombres y/o iniciales. Adicionalmente se debe enviar una copia del consentimiento informado del paciente y/o responsable legal para su publicación.

3.3 Los estudios de carácter experimental en seres vivos deben incluir el número de aprobación por un Comité de Bioética en la sección de "Materiales y Métodos".

4.- PRESENTACIÓN DEL MANUSCRITO

Los manuscritos deben ser escritos en Microsoft Word con espacio interlineado de 1,5 líneas, en hoja tamaño carta (21,59 x 27,94 cm), con márgenes de 3 centímetros en sus 4 bordes, en fuente Times New Roman con tamaño 12, color negro y páginas numeradas en el ángulo inferior derecho desde la primera página.

4.1 PÁGINA DE TÍTULO:

Debe presentarse en un archivo separado.

El título debe tener un máximo de 15 palabras, y escrito en español e inglés, o solamente en inglés para artículos en lengua inglesa.

Debe identificarse a los autores con su nombre y apellido. La(s) afiliación(es) de cada autor se colocan a continuación de la lista de autores. Se debe indicar el nombre y datos de contacto del autor correspondiente.

Conflictos de interés, se debe indicar si existe una fuente de apoyo financiero si corresponde, identificando el proyecto e institución patrocinadora. Detallar los conflictos de interés por cada autor en el caso que existan. Si no existen se debe comunicar “Sin conflictos de interés”.

Agradecimientos, deben estar indicados al final de la página de título.

4.2 RESUMEN:

Debe incluirse al inicio del manuscrito, después del título.

Escrito en español e inglés, o solamente en inglés para artículos en lengua inglesa.

Máximo de 250 palabras.

Incluir 5 palabras claves en español e inglés, o solamente en inglés para artículos en lengua inglesa. Usar términos MeSH.

Estructura: Objetivos, Materiales y Métodos, Resultados y Conclusiones.

4.3 CUERPO DEL MANUSCRITO:

4.3.1 Para artículos originales y revisiones sistemáticas:

Introducción: Presentar contextualización del tema a tratar, indicando la importancia de publicar el estudio, finalizando con la exposición de la hipótesis y objetivo de la investigación.

Material y métodos: Detallar los métodos, equipos y procedimientos realizados, para que permitan su reproducción por otros investigadores. Métodos ya publicados pueden ser citados. Debe describir adecuadamente el diseño del estudio, población de interés, criterios de selección, variables, medición, análisis estadístico, y consideraciones bioéticas.

Resultados: Presentados en un orden coherente, sin comentarios ni explicaciones personales, resaltando los que resultaron más relevantes. Pueden ser acompañados por un máximo de 6 tablas, gráficos y/o imágenes cuando sea necesario.

Discusión: Enfatizar los aportes del estudio, derivados de la interpretación de los resultados obtenidos, relacionados al conocimiento publicado.

Conclusiones: Desarrollarlas de acuerdo con los objetivos propuestos y resultados obtenidos, respondiendo a la hipótesis de trabajo planteada.

4.3.2 Para casos clínicos o serie de casos:

Introducción.

Presentación del caso: Debe tener un máximo de 6 imágenes, tablas y/o gráficos.

Discusión.

Conclusiones: Incluidas en el caso que entreguen un aporte inédito a la literatura.

Consentimiento informado: Indicar que el paciente ha dado el consentimiento informado para la publicación del caso.

4.3.3 Para revisiones narrativas:

Debe ajustarse a las características del tema presentado. Los revisores pueden sugerir cambios a la estructura del artículo.

4.4 REFERENCIAS BIBLIOGRÁFICAS:

Deben citarse a medida que aparecen en el texto con números arábigos entre paréntesis al final de cada oración, utilizando la norma Vancouver modificada, adicionando el DOI si lo posee. Para más de 5 autores debe escribir et al. después del quinto autor. Puede utilizarse los programas EndNote o Reference Manager:

Artículo de Revista: Tietel Z, Plotto A, Fallik E, Lewinsohn E, Porat R. Taste and aroma of fresh and stored mandarins. J Sci Food Agric. 2011;91(1):14-23. doi: 10.1002/jsfa.4146.

Libros: Jiménez Murillo L, Montero Pérez FJ. Compendio de Medicina de Urgencias: guía terapéutica. 2ª ed. Madrid: Elsevier; 2005.

Capítulo de Libros: Mehta SJ. Dolor abdominal. En: Friedman HH, coordinador. Manual de Diagnóstico Médico. 5ª ed. Barcelona: Masson; 2004. p.183-90.

Comunicación presentada a un congreso: Castro Beiras A, Escudero Pereira J. El Área del Corazón del Complexo Hospitalario Universitario de A Coruña (CHUAC). En: Libro de Ponencias: V Jornadas de Gestión y Evaluación de Costes Sanitarios. Bilbao; Ministerio de Sanidad y Consumo, Gobierno Vasco; 2000.p. 12-22.

Informe científico-técnico: Organización Mundial de la Salud. Factores de riesgo de enfermedades cardiovasculares: nuevas esferas de investigación. Informe de un Grupo Científico de la OMS. Ginebra: OMS; 1994. Serie Informes Técnicos; 841.

Tesis: Muñiz García J. Estudio transversal de los factores de riesgo cardiovascular en población infantil del medio rural gallego [tesis doctoral]. Santiago: Servicio de Publicaciones e Intercambio Científico, Universidad de Santiago; 1996.

4.5 ILUSTRACIONES, TABLAS Y GRÁFICOS:

Tablas, cuadros, gráficos y figuras deben ser numeradas en forma secuencial según orden de aparición en el texto y deben estar citados sin excepción en el texto entre paréntesis. Las tablas (Tabla I, II, etc.) y el resto de las ilustraciones como (Fig. 1, 2, etc.). Deben ser presentadas todas en un mismo archivo, no son aceptables tablas sin edición que solo sean las salidas de los software estadísticos.

Las tablas y los gráficos deben enviarse en archivo word, como archivo complementario, cada uno en páginas aparte. Cuando se requieran notas aclaratorias, agréguelas al pie de Tabla o Gráfico.

Explique al pie de las Tablas o Gráficos el significado de abreviaturas utilizadas. Denomine Figura a toda ilustración que no sea Tabla (Ej: gráficos, radiografías, fotografías, ilustraciones, etc.). Las Figuras deben tener un tamaño adecuado para su publicación y una resolución de 300 dpi.

Aplique su juicio estético para imaginar cómo visualizará el lector una Figura que deberá reducirse de tamaño para imprimirla. Sus títulos y leyendas no deben insertarse en la Figura

sino que se incluirán en el texto del archivo word. Los símbolos, flechas o letras empleadas en las fotografías de preparaciones microscópicas deben tener un tamaño y contraste suficientes para distinguirlas de su entorno.

Si una Figura reproduce material ya publicado, indique su fuente de origen y obtenga permiso escrito del autor y del editor original para reproducirla en su trabajo.

La publicación de Figuras en colores debe ser consultada con la Revista, su costo es fijado por los impresores y deben financiarlo los autores. Las imágenes histológicas, fotografías de lesiones, imágenes intraoperatorias o similares, deben publicarse en colores.

Las fotografías de pacientes deben cubrir su rostro para proteger su anonimato. Los autores deben contar con una autorización escrita del paciente, o su representante legal.

5.- EVALUACIÓN

El tiempo de revisión desde la confirmación de recepción hasta la primera respuesta, será de aproximadamente 30 días corridos.

Todas las comunicaciones serán sometidas a la evaluación por pares, con excepción de las cartas al Editor, que serán evaluadas únicamente por el Editor.

Los manuscritos que no cumplan cualquiera de las normas aquí publicadas, serán devueltos para su corrección antes de ser evaluados.

Cada trabajo será revisado por revisores expertos de acuerdo a la sección a la que sean enviados.

Los revisores podrán sugerir el rechazo, cambios menores, cambios mayores o la aceptación del manuscrito. Se comunicará a los autores las apreciaciones de los revisores, así como la decisión editorial.

Si se solicitan cambios menores, el manuscrito será aceptado una vez que se incluyan las modificaciones solicitadas.

Si se solicitan cambios mayores, el manuscrito modificado será evaluado por uno de los revisores iniciales, lo que tomará un máximo de 30 días corridos.

Si el artículo es rechazado, el editor informará del rechazo definitivo al autor/es.

6.- ENVÍO

Los manuscritos deben enviarse vía online a través de la plataforma de la revista.

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ASD JOURNAL
Applied Sciences in Dentistry

AUTHOR GUIDELINES

Authors interested in publishing in Applied Sciences in Dentistry must follow the instructions below:

1. - General Rules:

1.1 Papers submitted must be original and its presentation in another journal simultaneously is not allowed. Applied Sciences in Dentistry has all copyrights of the published articles, allowing subsequent transcription quoting the source (Appli. Sci. Dent.). None of the authors will be paid.

1.2 Applied Sciences in Dentistry receive articles in Spanish and English, and is the responsibility of the authors the correct writting of the manuscript. Articles written in Spanish must submit an abstract in English.

1.3 The contents of the published articles are the exclusive responsibility of the authors.

1.4 The dates of receipt, review, acceptance and online publication of the article will be at the end of the abstract on the first page of each published article.

2. - Formats:

2.1 Rsearch articles: Includes research in dentistry and other complementary and applied areas to discipline. The maximum length of the manuscript is 3,500 words, in addition to until 40 references updated and justified. For observational studies (cross-sectional, cohort or case-control) the use the STROBE Statement (STrengthening the Reporting of OBservational studies in Epidemiology) is suggested. For experimental studies using the Guide CONSORT (CONsolidated Standards Of Reporting Trials) is suggested. There is not a maximum number of authors.

2.2 Literature reviews: Can be systematic or narrative reviews. The maximum length of manuscripts is 5000 words, in addition to until 80 references updated and justified. For systematic reviews, the use of the PRISMA Statement (Preferred Reporting Items for Systematic reviews and Meta-Analyses) is suggested. There is not a maximum of authors for systematic reviews and meta-analyzes. Reviews of narrative type can have a maximum of 3 authors.

2.3 Case reports: Clinical cases or case series that must have an academic and scientific relevance, and should take care of the bioethical component. The maximum length of the manuscript is 1500 words and 20 references updated and justified. Clinical cases can have a maximum of 5 authors.

2.4 Letters to the Editor: Short Notes, a maximum of 1000 words, with opinions of readers about published articles or comments related to the Journal, dental research in general and/or any article published in this journal. You can have up to 5 references, do not include summaries and can have a maximum of 2 authors.

2.5 Short communication: An opportunity for the presentation of preliminary or brief observations that do not warrant a full paper, with a maximum of 1000 words, and until to 10 references. A maximun of one table and/or one figure can be accepted. Do not include abstract and can have a maximum of 3 authors.

3. - Bioethical Considerations:

3.1 All articles involving human studies should respect the updated rules of the Declaration of Helsinki.

3.2 In the images, do not identify the patient, so names and/or initials should not appear. Additionally, you must send a copy of the informed consent of the patient and/or legal responsibility for publication.

3.3 The experimental studies on living beings must include the approval number of Bioethics Committee in section "Materials and Methods".

4. - Manuscript Presentation:

4.1 Manuscripts should be typed in Microsoft Word with 1.5 spacing lines, letter size (21.59 x 27.94 cm), with margins of 3 cm in the 4 edges, in Times New Roman size 12, black color and with pages numbered in the lower right corner from the first page.

4.2 Title page:

- Must be submitted in a separate file.
- The title should have a maximum of 15 words, written in Spanish and English, or only in English for English language articles.
- A short version of the title- of no more than 60 characters- will be required (including spaces), so that it can be used as a headline in every page .
- The authors must be identified with name and surname.
- The affiliation(s) of each author is placed after the list of authors.
- At the end you must indicate the name and contact details of the corresponding author.
- Acknowledgements must be placed at the end of the title page.

4.3 Conflicts of interest:

- Must be submitted in a separate file.
- Indicate if there is a source of financial support if appropriate, identifying the project and sponsoring institution.

- Detail conflict of interest by each author if any. If they do not exist should be communicated "No conflicts of interest".

4.4 Abstract:

- Must be included at the beginning of the manuscript, after the title.
- Written in Spanish and English, or only in English for English language articles.
- Maximum of 250 words.
- Include 5 key words in Spanish and English, or only in English for English language articles, using MeSH terms.
- Structure: Objectives, Materials and Methods, Discussion and Conclusions.

4.4 Text of the Manuscript:

4.4.1 For original articles and systematic reviews:

- Introduction: Contextualization of the topic, indicating the importance of publishing the study, ending with exposure of the hypothesis and research objectives.
- Methods: Detail the methods, equipment and procedures performed, to allow its reproduction by other researchers. Already published methods can be cited. Must describe adequately the study design, population of interest, selection criteria, variables, measurement, statistical analysis, and bioethical considerations.
- Results: Presented in a coherent order, without comment or personal explanations, highlighting those that were most relevant. It may be accompanied by a maximum of six tables, graphics and/or images as needed.
- Discussion: Emphasize the contributions of the study, regarding the interpretation of the results, related to the published knowledge.
- Conclusions: Develop them in accordance with the objectives and results, responding to the proposed hypothesis.

4.4.2 For case reports and case series:

- Must contain a maximum of 6 images, tables or graphics.
- Must contain an Introduction (brief, with proper references).
- Patient related information must include demographic data, main symptomatology, medical background, relevant comorbidities, past interventions and outcomes.
- Clinical Findings (Must include a description of all relevant findings).
- Diagnostic Evaluation (Must mention methods, reasoning, prognostic, challenges).

- Therapeutic Intervention (Type of intervention and administration).
- Follow-up and Results (Must include a clinical course review and follow-up).
- Discussion. (Strengths and limitations of the treatment of choice, scientifically based background analysis, main lessons).
- Conclusions: Included if the case gives an original contribution to literature.
- Informed Consent: Indicating that the patient gave the informed consent to the publication of the case. Present a patient signed document that proves so.

4.4.3 For narrative reviews:

- Must be written conform to the characteristics of the subject presented. Reviewers can suggest changes to the structure of the article.

4.5 References: Should be cited as they appear in the text by Arabic numerals in parentheses at the end of each sentence, using the modified Vancouver rules, in addition must be included the DOI number if it is possible. For 6 or more authors, must be written et al. after the fifth author. The EndNote or Reference Manager software can be used:

- Journal article: Tietel Z, Plotto A, Fallik E, Lewinsohn E, Porat R. Taste and aroma of fresh and stored mandarins. J Sci Food Agric. 2011;91(1):14-23. doi: 10.1002/jsfa.4146.

- Book: Jiménez Murillo L, Montero Pérez FJ. Compendio de Medicina de Urgencias: guía terapéutica. 2ª ed. Madrid: Elsevier; 2005.

- Book chapter: Mehta SJ. Dolor abdominal. En: Friedman HH, coordinador. Manual de Diagnóstico Médico. 5ª ed. Barcelona: Masson; 2004. p.183-90.

- Communication presented in a congress: Castro Beiras A, Escudero Pereira J. El Área del Corazón del Complexo Hospitalario Universitario de A Coruña (CHUAC). En: Libro de Ponencias: V Jornadas de Gestión y Evaluación de Costes Sanitarios. Bilbao; Ministerio de Sanidad y Consumo, Gobierno Vasco; 2000.p. 12-22.

- Scientific-technical report: Organización Mundial de la Salud. Factores de riesgo de enfermedades cardiovasculares: nuevas esferas de investigación. Informe de un Grupo Científico de la OMS. Ginebra: OMS; 1994. Serie Informes Técnicos; 841.

- Thesis: Muñiz García J. Estudio transversal de los factores de riesgo cardiovascular en población infantil del medio rural gallego [tesis doctoral]. Santiago: Servicio de Publicaciones e Intercambio Científico, Universidad de Santiago; 1996.

4.6 Illustrations, tables and graphs: Tables, charts, graphs and figures should be numbered sequentially in order of appearance in the text and should be cited without exception in the text in parentheses. Tables (Table I, II, etc.) and the rest of the illustrations as (Fig. 1, 2, etc.). Each should be presented in a separate file.

- The tables and charts can be sent in word, tiff, png or jpg file, and must have a title. The abbreviations should be explained in the legend.
- Photographs and illustrations can be sent in tiff, png or jpg file and must be sized for publication in a resolution of 300 dpi. All illustrations must have a legend.
- All non-table illustration shall we called "figure" (In example, graphics, x-rays, photographs, illustrations, etc). Figures must be of a proper size for publication means (300dpi image resolution).

- Base on objective aesthetic judgement to visualize how a figure will look like after its size reduction when printed. Titles and legends should not be inserted into a figure but into the Word file. Symbols, arrows or letters of microscopic preparation photographs must be of a size that allows proper visualization.
- If a figure belongs to an already published work you must indicate its origin source and the author's and editor's permission to use it in your work.
- Publication of any colored figure must be consulted in advance with the magazine, its price is calculated by the printers and must be financed by the authors. Histologic, lesions, intra-operatorial images should be published in color.
- Patient photographs must be censured to ensure anonymity. Authors must present a written authorization of the patient (or its legal agent) allowing the use of their pictures.

5. Evaluation: Time for revision from confirmation of receipt to first response will be about 30 consecutive days.

5.1 All communications will be submitted to peer review, except letters to the Editor, which will be assessed only by the Editor.

5.2 The manuscripts that do not follow any of the rules published here, will be returned for correction before being evaluated.

5.3 Each article will be reviewed by expert reviewers according to the section to which they are sent.

5.4 The reviewers may suggest the rejection, minor changes, major changes or acceptance of the manuscript. Authors will be informed about assessments of the reviewers and the editorial decision.

- If minor changes are requested, the manuscript will be accepted once the requested changes are included.
- If major changes are requested, the revised manuscript will be evaluated by the original reviewers, it will take a maximum of 30 calendar days.
- If the item is rejected, the editor will leave open the possibility of a new submission of the modified manuscript or final rejection.

5.5 Accepted papers will be published immediately online on the website of Applied Sciences in Dentistry (http://www.asdjournal.cl and http://www.facultadodontologiauv.c/asd).

6. Sending: Manuscripts should be submitted via online through the platform of the journal

Submission Preparation Checklist

As part of the submission process, authors are required to check off their submission's compliance with all of the following items, and submissions may be returned to authors that do not adhere to these guidelines.

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1. I have read and the manuscript follows the stylistic and biliographic requirements outlined in the Author Guidelines.
2. The article has not been previously published and is not being submitted for consideration by any other journal (or has provided an explanation in the Comments to the Editor).
3. The manuscript is in Microsoft Word file.
4. References follow the modified Vancouver rules, and when possible the DOI is provided from those that possess.

APPLIED SCIENCES IN DENTISTRY JOURNAL se compromete a alcanzar y defender los estándares de comportamiento ético en todas las etapas del proceso de publicación, para lo cual se basa en los parámetros para buenas prácticas del Comettee on Publication Ethics (COPE).

De acuerdo a este código ético, editores/as, evaluadores/as y autores/as asumen las siguientes responsabilidades:

RESPONSABILIDADES ÉTICAS

Los trabajos enviados a la Revista deben contener una descripción detallada de los procedimientos experimentales y analíticos realizados, que permitan la replicación de los resultados reportados, y de forma objetiva y conmensurada con los resultados del estudio. Así mismo no deben contener afirmaciones falsas o distorsionadas, lo cual constituye un comportamiento no ético.

Cualquier idea o resultado descrito en un manuscrito enviado a la Revista y que pertenece a alguien que no es un coautor en el mismo, o que ha sido publicado previamente, debe ser apropiadamente referenciado.

Responsabilidades del Autor

Los autores deben presentar trabajos originales, no publicados incluso de manera parcial en otra revista, en consideración o aceptado para ser publicado, y deben ser el resultado del trabajo y/o ideas propias de los autores, el plagio no es aceptado.

Los autores, en particular el autor de correspondencia, se comprometen a mencionar a todas las personas que han colaborado y contribuido a la investigación y sus resultados, y a cerciorar que todas han conocido, estudiado y aprobado el contenido del artículo antes que éste sea sometido para publicación.

Es obligación de los autores indicar cualquier conflicto de intereses o interés financiero que podría interferir con los resultados o interpretaciones de la investigación.

Los autores deben involucrarse activamente en proceso de revisión por pares, y deben proporcionar oportunamente información veraz solicitada por los revisores o por el Comité Editorial de la Revista.

Si un artículo llegase a ser publicado con errores numéricos, conceptuales, ortográficos o tipográficos, y uno de los autores lo advierta; es su responsabilidad notificar inmediatamente al Comité Editorial y cooperar para publicar una fe de erratas, o retractarse del artículo, si se considera necesario.

Los autores que deseen retractar un artículo, deben enviar una carta al Comité Editorial manifestando su deseo y explicando las razones de su solicitud. Una

vez el Comité Editorial se reúna y analice el caso, se publicará una nota de retracción claramente identificable en la tabla de contenidos y los servicios de indexación.

Responsabilidades de los Revisores

Los revisores deben contribuir a la toma de decisiones para el proceso de publicación asistiendo el mejoramiento de la calidad de los artículos mediante revisiones objetivas, justas, y oportunas. Las revisiones tendrán que formularse en un tono respetuoso, constructivo, y libres de opiniones personales o juicios de valor. Si el revisor no se considera suficientemente experto o calificado para revisar un artículo, debe manifestarlo luego de leer el resumen del mismo

Los revisores deben mantener completa confidencialidad de la información proporcionada por el editor o el autor. Además tendrán que alertar al editor de cualquier contenido publicado o presentado que es sustancialmente similar a aquel que se está evaluando y que no ha sido advertido por el comité editorial.

Los revisores deben estar libres de cualquier potencial conflicto de intereses, relacionado con artículo, fuente financiera, instituciones, colaboraciones u otras relaciones entre el evaluador y el autor. Debe alertar al editor del mismo, si es necesario, retirando sus servicios para el manuscrito en cuestión.

Responsabilidades del Editor

El Comité Editorial asume la responsabilidad de velar por el cumplimiento de las normas y procedimientos contenidos en esta declaración, y de alertar a los otros miembros cuando exista un incumplimiento de los mismos.

Los editores deben actuar de manera balanceada, objetiva y justa mientras desempeñan sus tareas esperadas, sin discriminación en términos de género, orientación sexual, creencias políticas o religiosas, origen étnico o geográfico de los autores. Están obligados a considerar y aceptar artículos solamente por su mérito académico y sin influencia comercial.

El comité editorial asegurará que los evaluadores, bajo cualquier circunstancia, no intenten que el autor cite textos irrelevantes de revistas y/o autores con el único propósito de aumentar fraudulentamente el prestigio de dicha revista y/o autor.

El equipo editorial no revelará información sobre la autoridad de los artículos a los revisores, y así mismo debe seguir los procedimientos necesarios para garantizar que la identidad de los revisores se mantiene anónima. Tampoco deben revelar a nadie el contenido de los artículos sometidos para consideración de publicación.

Los editores deben adoptar y seguir siempre procedimientos razonables en la eventualidad de reclamos de naturaleza ética o conflictiva, de acuerdo con las políticas y procedimientos de la Universidad de Valparaíso. Dar a los autores la oportunidad razonable de responder a cualquier reclamo. Todos los reclamos deberían ser investigados sin importar cuándo la publicación original fue aprobada. La documentación asociada con cualquier reclamo de este tipo debiese ser conservada. Los editores deberían estar siempre dispuestos a publicar correcciones, clarificaciones, retractaciones y disculpas cuando fuere necesario.

Los Editores deben estar libres de cualquier conflicto de interés relacionado con los autores, el trabajo o sus financiadores, que incline su decisión hacia la aceptación o rechazo de un manuscrito en particular, y si tienen un impedimento insalvable, deben manifestarlo oportunamente y apartarse de la evaluación de un trabajo particular.

La decisión de aceptación, rechazo o aceptación condicional es de soberanía absoluta del Comité Editorial, basado en el juicio que realiza de la factibilidad, originalidad y contribución del trabajo.

Si un miembro del Comité Editorial advierte errores o imprecisiones en artículos que ya han sido publicados, es su responsabilidad informar a los demás miembros del Comité, para proceder a una corrección o retracción según lo amerite el caso.

PROCEDIMIENTOS PARA LIDIAR CON COMPORTAMIENTOS ANTIÉTICOS

Identificación de la conducta antiética

- Cualquier persona y en cualquier momento puede identificar un comportamiento antiético e informar al editor.
- Constituyen conductas antiéticas: el plagio, la omisión de coautorías; la falta de indicación del uso de fuentes de financiamiento; el envío total o parcial de escritos ya publicados; el uso de artículos no publicados por parte de algún árbitro, editor o de un miembro del comité editorial; la revisión del manuscrito existiendo conflictos de interés; y en general, cualquier otra que comprometa los parámetros para buenas practicas.

Investigación

- Quien informe al editor una conducta antiética, deberá acompañar antecedentes suficientes que se inicie la investigación. Todos los alegatos deben ser tomados en serio y tratados del mismo modo
- La investigación se mantendrá confidencial, evitando la divulgación de pruebas sobre la persona que se le reprocha una conducta contraria a la ética.
- Se velará que la persona a quién se le reprocha una conducta contraria a la ética tenga la oportunidad de formular sus descargos.

Infracciones

Las infracciones pueden ser leves, en tal caso, el Editor consultará a dos miembros del Comité Editorial. En infracciones graves, deberán informarse a todos los miembros del Comité Editorial y solicitar su opinión acerca de la necesidad de contar con un experto que evalúe la situación y la conveniencia de informar al empleador de quién ha incurrido en una conducta antiética.

Concluida la investigación, podrán aplicarse una o más de las siguientes medidas, de acuerdo con la gravedad de la infracción:

A. Para manuscritos en evaluación y proceso de publicación

   - Informar a la persona acerca de la falta a los estándares éticos de la publicación.
   - Advertir a la persona en términos más severos que su conducta no es aceptable y que no puede repetirse hacia el futuro. En caso de repetirse, la revista se reserva el derecho de rechazar el artículo.
   - Eliminar el manuscrito de la revista, aun cuando haya sido aceptada.

B. Para artículos publicados

   - Publicación en la misma revista de una nota o editorial, indicando la infracción a la ética.
   - Tratándose de la versión electrónica, incorporar una nota de retractación como primera página, antes del artículo, indicando las razones de retractación y colocando en todas las páginas del artículo una marca que indique “Artículo retractado”.

C. Para todos los manuscritos y artículos

   - En caso que la conducta corresponda a un ilícito penal, informarlo a las autoridades correspondientes.
   - Enviar una carta formal e informativa al empleador o a la institución a la que pertenezca la persona a quien se reprocha la falta.

ETHICS AND GOOD PRACTICES DECLARATION

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APPLIED SCIENCES IN DENTISTRY JOURNAL has a serious commitment of achieving and standing by the highest standards of ethics in every step of the publication process, for that, its parameters of publication ethics and practical resources are based on the Committee on Publication Ethics (COPE).

According to these, our editors, publishers and authors assume the following responsibilities:

ETHICS RESPONSABILITIES

All submissions must contain a detailed description of all experimental and analytical procedures/steps that took place, so that all results reported can be replicated, objectively and commensurately according to those reported. In addition, submissions must not contain any false nor distorted data; doing so will be considered as unethical behavior.

All ideas or results described in any manuscript sent to our journal whose authorship is different from a co-author, or has previously been published in other article must be properly referenced.

Author Responsibilities

All authors must submit original articles that haven't been partially/totally published or accepted in other journal or in the process of reviewing. All submissions must be the result of research and/or original planning, plagiarism is not accepted.

All authors, and specially the correspondence author, must mention all collaborators and contributors present in the process of research, just the same, they must make sure these have studied and approved the content of the article before its submission.

It is mandatory that authors inform of any conflict of interest that may interfere with the results of an article or the interpretation of these.

Authors must get actively involved in the peers reviewing process, and must opportunely deliver truthful information requested by the editorial board or reviewers.

If an article were to be published with numerical, conceptual, orthographical or typographical errors, it is the author's responsibility to make the notification to the editorial board and cooperate to launch an erratum or retract of the whole article if necessary.

Authors that wish to retract an article must send a letter to the editorial board manifesting his intention and explaining the reasons of the petition.

Once the editorial board analyzes the case, a retraction note will be published on the table of contents and indexation services.

General duties and Responsibilities of Reviewers

Reviewers must contribute throughout the publication decision making progress, assisting to the improvement of article's quality through objective, fair and opportune reviewing. All reviews are formulated so that they are respectful, constructive and bias-free. If the reviewer is unworthy or unqualified for an article, they must inform so immediately after reading the abstract.

Reviewers must keep complete confidentiality of all data proportionated by the author/editor. In addition, they must make note to the editor of any content – whether published or presented- similar to the one being reviewed if the editorial board has not taken notice.

Reviewers must be unbound of any conflict of interest related to the article, financial support, collaborations or other relationship with the author/s. They must alert the editor, if necessary, stepping down from the reviewing process for that article in specific.

General duties and Responsibilities of Editors

The Editorial board assumes responsibility of watching over the fulfilment of norms and procedures covered in this declaration, as well as notify other members in case of an infringing of these.

Editors must act in a balanced, objective and fair manner while they perform their role, avoiding any type of discrimination (regarding author's genre, sexual orientation, personal beliefs, political views, religion, ethnic/geographic background). They are compelled to consider and accept articles based solely by their academic value and free of commercial bias.

The editorial board will prevent reviewers from aiding authors by suggesting they add irrelevant citations with the sole purpose of rising that journal/article's prestige.

The editing team won't reveal information regarding an article's authorship to the reviewers, just the same, they will make sure to preserve the reviewer's anonymity. The content of any submitted article under no circumstances will be made public by the editing team.

Editors must always implement and follow reasonable measures in the eventual happening of any ethical/conflictual complaint against an article, according to University of Valparaiso's political policies, allowing the author/s the chance to defend themselves or reply back. All complaints should be looked up into, no matter when the article compromised was published. All documentation associated with any complaint shall be preserved. Editors must always be open to publish a retraction, correction, clarification or apology if necessary.

Editors must be free of any conflict of interest regarding its author or its financing that might influence their decision of whether accepting or rejecting a submission. In case there exists an insurmountable conflict, they should opportunely inform it and take a step back from the reviewing process of that particular article.

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PROCEDURES TO DEAL WITH UNSCRUPULOUS BEHAVIOR

Unethical behavior Identification

- Any individual, anytime, can notify an unethical behavior and inform the editor.
- The following constitute unethical behavior: plagiarism, co-authorship omission; lack of indication of the funding source; total/partial submission of articles already published; review of an article if there's a conflict of interest; in overall, any other that compromises good practice parameters.

Investigation

- Whoever informs the editor of an ethical conduct, will have to adjunct enough evidence for an investigation to take place. All complaints must be taken seriously and treated in the same way.
- The investigation will be kept confidential, avoiding the divulgation of evidence that might be related to the person being investigated.
- This journal has a pledge to ensure that all persons accused of unethical behavior has a chance to retort.

Infringements

In minor Infringements the Editor will consult two other members from the Editorial Board. In severe Infringements, all members from the Editorial board will be informed and their opinion -regarding the need of an expert's opinion to asses the situation and the convenience of notify the supervisor of whoever did the infringement- will be solicited.

Concluded the investigation, one or more of the next measures will take place, depending on the infringement severity:

A. In articles submitted for reviewing/revision in the publication process:
    - Notify the author about the article's unethical standards.
    - Inform the author that their behavior is unacceptable and that it cannot be repeated in the future. In case of recurrence, this Journal reserves its right of admitting any other articles from that person.
    - The elimination of the article involved from this Journal, even when previously accepted.

B. In Published Articles:
    - Publication in the Journal's broadsheet of an Editorial note, indicating the ethic infringement.
    - In the electronic version of this Journal, a retraction note will be incorporated on the first page, at the beginning of the article, indicating all reasons for retraction and inserting in all of the article's pages a water mark that will read "Retracted Article".

C. For all manuscripts and articles:
    - In case the misbehavior answers to a criminal offense, authorities will be informed.
    - A formal informative letter will be sent to the employer or institution that the accused person answers to.

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