

Participation of micro-organisms of dental interest in the etiology of Burning Mouth Syndrome

Participação dos micro-organismos de interesse odontológico na etiologia da Síndrome de Ardência Bucal

Felipe Eduardo OLIVEIRA¹, Janete Dias ALMEIDA¹, Cristiane YUMI KOGA-ITO².

1 - Institute of Science and Technology – UNESP – Univ Estadual Paulista – School of Dentistry – Department of Biosciences and Oral Diagnosis – São José dos Campos – SP – Brazil.

2 - Institute of Science and Technology – UNESP – Univ Estadual Paulista – Department of Environmental Engineering – São José dos Campos – SP – Brazil.

ABSTRACT

Objective: The Burning Mouth Syndrome (BMS) is a pathologic entity characterized by the presence of chronic symptoms of burning or pain in normal oral mucosa. It mainly affects women in the postmenopausal period, and its cause is unknown, but as there is an association with biological and psychological factors, it may assume a multifactorial etiology. Considering the unclear etiology of BMS, studies that contribute to its understanding are of great importance. In order to achieve a better understanding regarding microbial etiological factors of this disease, the aim of this review is to compile studies on possible involvement of micro-organisms of dental interest in the etiology of BMS. Studies have reported that patients with BMS harbor greater amount of intra-oral *Candida* and *Enterobacteriaceae* than patients without clinical manifestations of this disease.

Methods: Different sources such as articles, books and journals, published in the world literature were used in this research. These sources were accessed by databases like PubMed, SciELO, Scopus or search of full text. **Conclusion:** Studies in the literature have suggested that *Candida* and *Enterococcus* were correlated with BMS, although they might not necessarily be considered as an etiological factor but a predisposing factor. However, further studies that aim to elucidate relation between BMS and infectious factors are necessary.

KEYWORDS

Candida; *Enterobacteriaceae*; Burning mouth syndrome.

RESUMO

Objetivo: A síndrome da ardência bucal (SAB) é uma entidade patológica caracterizada pela presença de sintomas crônicos de ardor ou dor na mucosa oral clinicamente normal. Afeta principalmente mulheres no período pós-menopausa, sendo sua causa desconhecida, mas sua relação com uma completa associação de fatores biológicos e psicológicos nos faz supor uma etiologia multifatorial. Considerando a etiologia ainda não esclarecida da Síndrome da Ardência Bucal, estudos que contribuam para o esclarecimento desta são de extrema relevância. Visando maior entendimento com relação aos fatores etiológicos microbianos da doença, o objetivo desta revisão de literatura foi compilar estudos sobre possível participação de micro-organismos de interesse odontológico na etiologia da SAB. Estudos afirmam que pacientes com SAB abrigam maior quantidade de *Candida* e *Enterobactérias* intraorais que pacientes sem manifestações clínicas dessa patologia. **Métodos:** Nesta pesquisa foram utilizadas diferentes fontes, como artigos, livros e revistas publicados na literatura mundial. Estas fontes foram acessadas por bases de dados como (PubMed, Scielo, Scopus) ou por pesquisa do texto integral. **Conclusão:** Estudos na literatura sugerem que *Candida* e *Enterococcus* possuem correlação com a Síndrome de Ardência Bucal, embora não possam ser necessariamente considerados como um fator etiológico e sim um fator pré-disponente. No entanto, mais estudos com o objetivo de elucidar como ocorre essa relação entre a SAB e os fatores infecciosos são necessários.

PALAVRAS-CHAVE

Candida; *Enterobacteriaceae*; Síndrome da Ardência Bucal.

CRITICAL REVIEW

Burning Mouth Syndrome (BMS) is a pathologic entity characterized by the presence of chronic symptoms of burning or pain in normal oral mucosa. It mainly affects women in the postmenopausal period, and its cause is unknown, but its relation with a complete combination of biological and psychological causes us to assume a multifactorial etiology. Although it has been found effective treatments in some cases, the search for an effective treatment continues in most cases [1]. Almeida et al. [2] and Ros Lluh et al. [1] also stated that the etiology of BMS is difficult to diagnose, and there may be more than one etiological factor. Among these factors, systemic and local, psychogenic or idiopathic causes are described in the literature [3].

The main complaint of BMS carriers is a burning pain of varying intensity in the mouth [4]. Patients describe it as an itching sensation, stinging, burning, tingling or numbness in the mucosa, without a physical cause detected [5,6]. It may be located mainly on the sides or tip of the tongue, and it can also be seen on the lips, palate or involving the entire oral cavity [7]. Affected patients often have multiple oral complications, including burning, dryness and taste alterations. There is a pain increase during the day, in the case of stress, fatigue, excessive speaking or by the intake of spicy and hot food and recovery occurs with cold food, work and leisure [8,9].

The most common generic denomination is Burning Mouth Syndrome (BMS). It presents the following terms as synonyms: oral dysesthesia, stomatodynia, stomatopyrosis, glossodynia, glossopyrosis [2], glossalgia and psychogenic oral paraesthesias [10].

Only a few information is available about BMS variations in different populations in the world, and the same applies to its prevalence. The wide range of values, from 0.01 to 0.7% in the general population or 26% in elderly

people reflects the lack of exact and accurate definitions [5]. In America, there is an estimate of one million people suffering with burning mouth [11]. The sensation of burning mouth is reported more often in women, especially after menopause [12]. According to Almeida [2], several authors demonstrate that the prevalence of BMS varies from 1 to 5% of the population, being 3-6 times more common in women than in men, occurring mainly in pre and postmenopausal periods in women.

Aas et al. [13], through molecular study, showed that there are more than 700 bacterial species or phlotypes colonizing the oral cavity, and 50% of these micro-organisms have not been cultivated yet. According to these authors, the oral cavity presents several habitats with different nutritional and physicochemical characteristics, such as the mucosa of the cheek, tongue, gingival sulcus and teeth surfaces, which favor the adhesion and growth of certain species of micro-organisms. Carranza [14] reported that up to 600 different species of bacteria that colonize the oral cavity can affect the delicate balance of bacteria-host interactions that lead to health or disease.

Lamey et al. [15] states that the condition of the syndrome needs to be differentiated from the complaint of burning mouth associated with clinical abnormalities in the mucosa, such as lichen planus or geographic tongue. According to ADA [16], the term BMS should only be used when a definite cause is not found.

Patients usually have a characteristic profile. They are mostly postmenopausal women with an average age between 50 and 60 years, with a prevalence rate among males and females that can range from 1:3 to 1:16 [17]. Zeller et al. [18] found that even 30% of psychiatric disorders are associated with BMS, such as anxiety disorders and depression.

In 1994, Lamey et al. [15] divided BMS into three types in order to group different types of patients, according to the most common features presented (Table 1).

Table 1 – Subdivisions of BMS

Types	Clinical features	Association
1	Daily pain, not present on waking, worsening throughout the day	Non-psychiatric
2	Constant pain	Psychiatric, chronic anxiety
3	Intermittent pain in unusual sites (floor of mouth)	Stomatitis by allergic contact to conserving and additives

Source: Lamey et al. 1994 [15].

BMS pathophysiology remains under the shade of several hypotheses. Recently, the major site of pathology in BMS was not identified and, hence, no diagnostic test was available for this disease. However, a previous study investigated the innervation of the tongue epithelium in 12 cases of chronic BMS and 9 healthy controls using the tongue tissue biopsies to assess whether the damage of the peripheral nerve fibers form the basis of the pathogenesis of the disease. Researchers used microscopic and immunohistochemical methods to examine the nerve in the tongue. They reported a significantly lower density of nerve fibers in the epithelium in patients with BMS than the controls. The authors described changes in epithelial and sub papillary nerve fibers as suggestive of axonal degeneration, concluding that BMS can be caused by a trigeminal sensory small-fiber neuropathy [19].

There is also a possible relationship between the BMS and gustatory activity [20]. There is an increased prevalence “supertasters” (people with increased ability to detect flavors) among patients with BMS. These patients have a higher chance of being affected by this burning due to its higher density of taste buds, which are surrounded by a collection of trigeminal nerve sensory bundles [21]. This hypothesis would explain why Hormone Replacement Therapy is not effective once the nerve damage has already occurred. In addition, Lucchina et al. [22] states that the ability to detect bitter taste

decreases with menopause. This reduction in taste sensitivity to bitter flavors in the chorda tympani branch of the facial nerve, results in intensification of taste sensations from the area innervated by the glossopharyngeal nerve and produces “ghost” tastes [23].

Petruzzi et al. [24] states that these painful stimuli are detected by nociceptors, receptors where capsaicin acts. In a comparative study, triple blinded, it was observed that after administration of capsaicin for 4 weeks, there was reduction in mouth pain in 84% of patients, compared with the control group. The therapeutic efficacy of capsaicin strengthens the hypothesis of a neurogenic cause for BMS.

Etiological possibilities for BMS can be grouped into four groups (Table 2).

Table 2 – Etiological possibilities of BMS

Systemic	Local	Psychogenic	Idiopathic
Changes of the salivary glands (Sjögren, FM, medication, radiation, anxiety or stress)	Dental (prosthesis, trauma, parafunctional behavior changes)	Depression, anxiety, cancer phobia, OCD	---
Endocrine (DM, hypothyroidism, menopause)	Allergen (additives, colorants conservings, food)	---	---
Drugs (ACE, ATB, antiretroviral drugs, tricyclic antidepressants)	Infectious (<i>Candida</i> spp., nonspecific bacteria, fuso spirochetes)	---	---
Neurological (trigeminal and glossopharyngeal neuralgia)	---	---	---
Nutritional (iron, B complex, folic acid and zinc)	---	---	---

(FM) fibromyalgia, (DM) diabetes mellitus (ACE) inhibitor of angiotensin converting enzyme (ATB) antibiotic (OCD) obsessive compulsive disorder.

Source: Almeida A., Gago M.T., 2004 [2].

Among many possible etiologic factors, this literature review focused on local infectious factors. Thus, the most important aspects will be addressed about some micro-organisms of dental interest on their role in the possible etiology of this disease that still remains poorly understood.

The focusing micro-organisms of this review were *Candida* spp. and *Enterobacteriaceae*. The choice of these two genera is due to studies reporting candidiasis as an etiological factor of BMS [8,25,26]. Although this fact may be due to poor adaptation of xerostomia or dental prosthesis, a possible microbial origin of BMS is suggested since there are reports that show burning sensation remission after antifungal treatment [27]. Allied to this, a study from Samaranayake et al. [28] states that the prevalence of *Candida* spp. and coliforms (*Enterobacter* and *Klebsiella*) is higher in BMS patients than healthy patients.

Candida Genus

According to Jorge [29], this genus includes about 200 species of yeast that do not produce endospores. Due to the inability of this genus in presenting sexual forms, they were classified as imperfect fungi of the subdivision Deuteromycotina. However, for some species of this genus, it was shown the sexual state (teleomorph). *C. albicans*, *C. tropicalis* and *C. glabrata* are the most frequently isolated species from candidiasis, totaling about 80%.

Tortora [30], reports that the bacterial flora of the mucous membranes of the mouth and genitourinary generally suppresses the growth of fungi as *C. albicans*. Other *Candida* species as *C. tropicalis* or *C. krusei* may also be involved. Since the fungus is not affected by antibacterial drugs, sometimes it grows excessively in mucosal tissue when antibiotics suppress the normal bacterial flora. Changes in pH of normal mucosa may have similar effect.

Candida genus has been proposed as one of the most important in the etiology of

BMS among local causes, as revealed in the study of Samaranayake et al. [28], in which 130 patients (114 women and 16 men) with a history of BMS and 103 (90 women and 13 men) healthy were studied. Eighty per cent of both groups wore dentures or partial dentures, none of them receiving antibiotic or steroid treatment. The technique of mouth rinse was used to detect *Candida* count. The frequency of isolation of *Candida* species in the study population was 32% compared with 21% in the control population. The prevalent species isolated in both groups was *C. albicans* while *C. glabrata* and *C. tropicalis* were isolated with lower frequency. A variety of other *Candida* species were isolated, especially in the test group. It was shown that patients with BMS harbour more intra-oral *Candida* than patients considered healthy, especially concerning *C. albicans*. Moreover, the counting rate of these micro-organisms were also presented higher in the test group than the control group.

Many factors predispose to fungal infection in the mouth are mentioned in the literature, however, it is rare the occurrence of infection without the presence of one or more of these factors is rare. Among the local factors, xerostomia, changes in oral microbiota by the use of antibiotic, high-carbohydrate diet, use of prostheses and orthodontic appliances and associations with other injuries may be included. Among the systemic factors are endocrine (diabetes), nutritional and immunological factors [29].

According to Osaki et. al. [31], candidiasis affect mainly the elderly, patients with some type of immune deficiency, prolonged use of antibiotics, immunosuppressants, anti-retroviral drugs or corticosteroids. This infection can appear with or without inflammation clinical signs in the mucosa and patients report a burning sensation in the mouth, which leads to dysphagia and drooling.

Erythematous and pseudomembranous candidiasis have been associated with this

syndrome [32]. Pseudomembranous type is easily recognized by its white color, slightly elevated plaques having a milky appearance and they can be easily removed, being most commonly found on the cheeks and palate. Erythematous type is characterized by redness of the mucosa, tongue and palate. The diagnosis of candidiasis is commonly presumed, made in response to antifungal drugs and rarely based on histological or cytological studies.

Gorsky et al. [8] reported that in patients with BMS and without clinical signs of candidiasis, 86% showed clinical recovery after using antifungal tablets and 13% reported complete elimination of their symptoms.

Vitkov et al. [33] suggests that the burning sensation may be related to the multiplication of *Candida*. The detection of painful stimuli occurs in sensory nerve endings called nociceptors. These small-diameter neurons express mechanical, chemical and temperature variation signals in the central nervous system, giving the perception of pain or discomfort, acting through capsaicin receptor (vanilloid). Capsaicin, when applied to the tongue causes reproducible burning sensations [34]. Likewise, fungal products also induce such sensations [35]. The induction of burning sensations by fungal products also indicates a possible multiplication of *Candida* spp. [31].

In contrast, Cavalcanti et al. [4], in their study, which examined a possible relation between BMS and the frequency of *Candida* species isolated from the oral cavity of these individuals, observed no association between the BMS and the prevalence of yeasts. In this study, cultures were found positive for *Candida* 45.16% of patients, or in other words, a higher prevalence than that one observed for Samaranayake et al. [28], but the difference for the control group did not achieve significant results.

A possible explanation for these differences could be related to regional differences among the Brazilian population and individuals from

other countries, mainly because of the increased use of prostheses (mostly disabled) in the Brazilian population, which is an important factor for presence of *Candida* species in the oral mucosa. The control group, which had a higher frequency of users of removable dentures, also showed a higher frequency of *C. albicans* when compared to the study group, although the count was higher in the study group (113.8 ± 46.0 CFU/mL) compared to the control group (95.2 ± 40.8 CFU/mL). For these reasons the authors suggest it is not possible to confirm the presence of *C. albicans* as an associated factor in the BMS etiology.

***Enterobacteriaceae* family**

Jorge [29] states that the *Enterobacteriaceae* family constitutes a heterogeneous group of Gram-negative bacilli. Many species are part of the normal microflora of the intestinal tract of animals and man and they are also present in soil, vegetation and water.

Tortora, [30] reports that the bacterial flora of the mucous membranes of the mouth and genitourinary generally suppresses the growth of fungi as *C. albicans*. Other *Candida* species as *C. tropicalis* or *C. krusei* may also be involved. Since the fungus is not affected by antibacterial drugs, sometimes it grows excessively in mucosal tissue when antibiotics suppress the normal bacterial flora. Changes in pH of normal mucosa may have similar effect.

They have pili on their cell surfaces, which in some species confers adherence to epithelial surfaces. They also present sexual pili, an important factor in the transference of antibiotic resistance and it may happen between the same or different members of this family. Besides, these bacteria have on their cell wall constitution lipopolysaccharides (LPS), known as endotoxin. Many species of *Enterobacteriaceae* produce bacteriocins. Some genres of this family belong to a group known as coliforms, inhabitants of the human gut and responsible for causing some diseases. As examples, we have the genres *Enterobacter*, *Klebsiella*, *Escherichia*, etc. [36].

Samaranayake et al. [28], in the same study already cited, also showed the relation between BMS and *Enterobacteriaceae*. It was observed that 66% of the samples in the study group showed coliforms, while 42% of the control group samples harboured these micro-organisms. These data indicate that, proportionally, patients with burning mouth have a larger number of intraoral coliforms than the healthy ones. Furthermore, it was found coliforms coexisting with *Candida* spp. on 9 occasions, and on 1 occasion in the test and control groups respectively. Studies indicate that patients with burning mouth harboured more intraoral coliform than healthy ones. Moreover, the counting rate of these micro-organisms is higher in the test group than the control group.

In another study, Kuc et al [37] evaluated the oral health of a group of elderly, long-term care, institutionalised residents in a facility in Edmonton, Canada. It was also evaluated the occurrence of oral coliform to examine the relation between oral health status and the prevalence of this opportunistic infection. Thirty percent of the analyzed individuals presented oral coliforms and a significant number exhibited poor oral hygiene.

DISCUSSION

Based on the reviewed studies, we noted that there is an inaccuracy between the local infectious factors and their real role in BMS, in other words, if these micro-organisms are part of the etiology of this disorder, if there are factors that accelerate its onset or exacerbate its symptoms. On the other hand, we can predicate that there is a relation between these factors and BMS.

It should be highlighted that the studies about this syndrome follow many different paths, which make it difficult to come to a consensus on the etiology of this disease. That is why it is considered multifactorial, because there are many evidences for different causes of its etiology.

More studies need to be done so that the real etiological factors of BMS will be discovered.

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Janete Dias Almeida
(Corresponding author)

Institute of Science and Technology
Department of Biosciences and Oral Diagnosis
Av. Eng. Francisco José Longo, nº 777
Bairro: Jardim São Dimas
12245-000 - São José dos Campos, SP

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ORIGINAL ARTICLE

The use of bisphosphonates in dental clinic: a review of the last five years

O uso de bisfosfonatos na clínica dentária: uma revisão dos últimos cinco anos Ü

Yamba Carla Lara PEREIRA¹, Glauce Crivelaro do NASCIMENTO²

1 - School of Dentistry of Ribeirão Preto - University of São Paulo - Ribeirão Preto – SP – Brazil.

2 - School of Philosophy, Science and Literature of Ribeirão Preto - University of São Paulo - Ribeirão Preto – SP – Brazil.

ABSTRACT

The bisphosphonates are synthetic drugs used for treatment of neoplasms and bone, Paget's disease and more reason for excitement in cases of postmenopausal osteoporosis. These drugs present some known side effects, however, a new complication with oral manifestation was recently identified, called Osteonecrosis. When this necrosis of the bone bases is associated to the use of Bisphosphonates, it is named Osteonecrosis by Bisphosphonates. The type of bisphosphonate, route of administration, and duration of treatment with these drugs seem to have direct relation with the incidence of osteonecrosis associated with bisphosphonates. The aim of this work was to present a review of the last five years about the use of bisphosphonates, the available bisphosphonate groups in the Brazilian market emphasizing those used in the treatment of Osteonecrosis, and exploring possible diagnostic aspects of the disease from image Diagnostics. It is concluded the impact of current world life stimulates new research areas focusing on important illnesses, their manifestations, and on searching for better treatment protocols and faster diagnosis protocols, aiming reduce possible treatment unwanted side effects.

KEYWORDS

Biphosphonates; Bisphosphonate associated Osteonecrosis of the jaw; Radiography, dental

RESUMO

Os bisfosfonatos são drogas sintéticas usadas no tratamento de neoplasias de osso, doença de Paget e nos casos de osteoporose pós-menopáusia. Estas drogas apresentam alguns efeitos colaterais conhecidos, no entanto, uma nova complicação com manifestação oral foi recentemente identificada, chamada osteonecrose. Quando a necrose das bases ósseas está associada à utilização de bisfosfonatos, é nomeada osteonecrose por bisfosfonatos. O tipo de Bisfosfonato, a via de administração e duração do tratamento com estes medicamentos parecem ter uma relação direta com a incidência da osteonecrose. O objetivo deste trabalho foi apresentar uma revisão dos últimos cinco anos sobre o uso de Bisfosfonatos e os grupos de Bifosfonatos disponíveis no mercado brasileiro, enfatizando aqueles usados no tratamento da osteonecrose, além de explorar possíveis aspectos de diagnóstico da doença por meio de imagem. Conclui-se que novas áreas de pesquisa, com foco em doenças importantes e as suas manifestações, estão buscando melhores protocolos no tratamento e diagnóstico rápido, visando reduzir possíveis efeitos colaterais indesejados.

PALAVRAS-CHAVE

Bifosfonatos; Osteonecrose da arcada osseodentária associada a bisfosfonatos; Radiografias dentárias.

INTRODUCTION

The first synthesis of bisphosphonates occurred in the mid-nineteenth century, in 1865 in Germany, for application in industry. However, in humans, the use started about 40 years ago [1]. Bisphosphonates are drugs used

in the treatment of metastatic bone disease, Paget's disease, osteoporosis / osteopenia [2]. These drugs have varying potencies and dosages, directly proportional to the risk of bone disorders such as osteonecrosis. Bisphosphonates have been used with good results in children and little or no effects in several diseases: Osteogenesis

imperfecta, corticosteroid-induced osteoporosis and idiopathic juvenile disuse, metabolic bone disease, heterotopic calcification in soft tissues resistant hypercalcemia, hypervitaminosis D and fibrous dysplasia (FD) syndrome McCune Albright (SMA) [3]. Bisphosphonates are synthetic analogs of pyrophosphate that inhibit stable growth and dissolution of hydroxyapatite crystals of bone from the reduction of osteoclastic activity [4]. The parenteral administration seems to be associated with a significant number of cases of osteonecrosis of the jaw, whereas only a small percentage of cases were attributed to the use of these drugs orally [5]. Osteonecrosis of the jaw is a pathological change that can result from a complex interaction between bone metabolism, local trauma, infection, and hypovascularization [6]. Systemic factors such as diabetes mellitus, immunosuppression, concomitant use of other medications, such as chemotherapy agents, radiotherapy and corticosteroids, or complication, orthognathic surgery, hematological disorder, especially in patients with sickle cell disease may also develop framework of avascular necrosis of the jaw [7].

In the early stages of the disease, the patients have no specific symptoms, so it is necessary to stay alert to signs like constant pain and throbbing exacerbated by movement of the joint, headache, earache, pain and spasm of the jaw muscles, limitation of mouth opening, crepitus [8]. Osteonecrosis is often progressive and can create extensive areas of exposed bone and dehiscence. When tissues are severely affected, patients may complain of pain and lack of sensitivity [9].

In conventional radiographs structural changes are not observed, but it is possible to see morphological alterations. The magnetic resonance imaging or computed tomography may detect this type of pathology, even in early lesions [10]. The imaging findings can provide important information about the course,

magnitude and progression of the disease. The literature states that the radiographic image of the traditional BRONJ is a mixed lesion with areas of bone sclerosis, radiopaque and radiolucent areas of destruction around the teeth and the alveolar crest. Diffuse or regional osteosclerosis, thickened dental lamina dura subperiosteal bone deposition, low density of cancellous bone and bone healing following surgical procedures might also be associated with the condition [11].

The preventive intervention of patients taking bisphosphonate is a good option for management of these cases. Preventive action of health professionals who have access to routine radiographic examination can diagnose sub clinical stages of the osteonecrosis and establish a curative therapy, intercepting the course of the disease before it compromises the patient's life.

The purpose of this literature review was to explore the literature on the use of bisphosphonate. With a bibliographical survey of the literature reviews on the topic in the last five years we intended to present the theme, spread its seriousness as well as alerting the clinician to early manifestations of the osteonecrosis disease, preventing the worsening the clinical scenario.

MATERIAL AND METHODS

A search was made in PUBMED database [<http://www.ncbi.nlm.nih.gov/pubmed>] in order to raise all literature reviews and case reports using the keywords "bisphosphonates" and "osteonecrosis", and filter option being the last five years and humans.

LITERATURE REVIEW

Bisphosphonate drugs

Bisphosphonates (BPs) are synthetic drugs, analogues of pyrophosphate, in which the oxygen atom has been replaced by a carbon atom in the bisphosphonate molecule. These

compounds have been synthesized, and used in industry since the 19th century but it is only in the 1960s that their *in vitro* ability to inhibit the precipitation of calcium phosphate was applied clinically [12].

In this way, in the 60s, BPs have emerged as therapeutic tool in diseases with high bone resorption - neoplastic hypercalcemia, Paget's disease, bone metastases, multiple myeloma, primary and secondary hyperparathyroidism and Osteogenesis imperfecta [13]. Currently, these drugs are the first choice in the treatment of osteoporosis, since it is inexpensive and capable of reducing osteoporotic fractures by 60% [14] and more than two million people worldwide are treated with BPs [15]. This class of drugs has a significant effect on bone turnover, reducing osteoclastic activity and improving bone mineral density, what result in a reduced risk of osteoporotic fractures.

The BPs can be divided into two groups, nitrogen-containing and non-nitrogen-containing bisphosphonates, differing in the mechanism of action on osteoclasts [16]. Non-nitrogen-containing BPs are taken up by the osteoclasts and trigger intracellular mechanisms leading to apoptosis [17]. These kind of BPs such as etidronate [Didronel], clodronate [Bonefos, Loron], tiludronate [Skelid], are metabolized in the cell that replace the terminal portion ATP pyrophosphate, forming a non-functional molecule which competes with adenosine triphosphate (ATP) in cellular energy metabolism. The osteoclast initiates apoptosis and dies, leading to an overall decrease in the breaking of the bone [18].

In contrast, nitrogen-containing BPs, such as pamidronate (APD, Aredia), Neridronate, Olpadronate, Alendronate (Fosamax), Ibandronate (Boniva), Risedronate (Actonel) and Zoledronate (Zometa, Aclasta), act on bone metabolism, binding and blocking the enzyme farnesyl diphosphate synthase (FPP) in inhibitor of HMG-CoA reductase [also known as the mevalonate pathway] [19]. Nitrogen-containing BPs have a complex pathway of action resulting

in interference with the osteoclastogenesis, in apoptosis and changes in cytoskeletal dynamics [16]. They are not metabolized, where 50% are secreted in the urine unchanged, and the rest bind to bone and are slowly released into the circulation; therefore, their half-life in the bone could be as long as 10 years [20].

BPs have also the characteristic of specificity, in that the drugs in question come from two phosphonate groups and, possibly, a hydroxyl in R1, which works together to coordinate calcium ions. The molecules of this medicament have a preference for calcium and bind to it. The largest store of calcium in the human body is in the bones, so BPs tend to accumulate in high concentrations only in bones [18].

Biphosphonated - related osteonecrosis of the jaw (BRONJ)

Although BPs are the first choice for treatment of osteoporosis, they influence negatively in the clinic therapeutic that require consistency of bone metabolism. They can cause necrotic diseases in bones. The BPs-related osteonecrosis is a clinical entity relatively current. This secondary severely condition affects the quality of life, producing significant morbidity in affected patients [21].

Osteonecrosis is a clinical condition characterized by necrosis of the bone, resulting in systemic factors and sites that compromise the bone vascularity. BRONJ is a pathological change that can result from a complex interaction between bone metabolism, local trauma, infection, hypovascularization and bisphosphonate use. This pathology is a rare but serious complication that can be difficult to manage and may result in significant morbidity to the patient, including severe pain and loss of large portions of the mandible and/or maxilla [22].

Marx [23] first described the association between use of BPs and jaw osteonecrosis. Today, this relationship is well established and is defined as the presence of exposed bone for 8 weeks in patients undergoing treatment with

bisphosphonates and no history of radiotherapy [24]. Several factors have been implicated in the development of BRONJ, such as high doses of this drug for long periods, advanced age and invasive surgical procedures. The initiation of osteonecrosis can be understood as elicited via inhibition of bone remodeling. BPs incorporate local high bone turnover. The mucosa and periosteum are easily affected by infection or local trauma and the presence of any microtrauma, infection or iatrogenic damage can increase the demand for bone repair thus, exceeding the limits of the capacity of newly formed bones, resulting in osteonecrosis site [25]. Some authors believe also that osteonecrosis injuries are always associated with damage to the gum. This was attributed to the mechanism of apoptosis induced by BPs on osteoclasts and keratinocytes. There is a hypothesis which proposes that apoptosis of these cells result in a reduction and destruction of keratin barrier of the oral mucosa and this consists a pathway that leads to osteonecrosis of the jaws [26].

The American Association of Oral and Maxillofacial Surgeons (AAOMS) established in 2009 that should be considered cases of BRONJ those who submit all of the following [27]:

- 1 - current or previous treatment with bisphosphonates;
- 2 - necrotic bone in the jaw region that persists for more than eight weeks and
- 3 - no history of radiation therapy in maxillomandibular complex

The use of intravenous BPs, especially for a long period, and dentoalveolar surgical procedures are major risk factors for BRONJ. These data are described in a recent systematic review, where the prevalence observed from a sample of 39,124 patients was equal to 6.1% [28]. Local factors involved are the dentoalveolar surgeries such as tooth extractions, implants, periodontal surgery or periapical. The bisphosphonate-related osteonecrosis is more common in the mandible

in patients with previous dental problems [gum disease and tooth abscess] and in regions where the mucosa overlying the bone is thinner [29]. The time between the dental procedure and the development of osteonecrosis ranged between one month and one year [30].

The participation of dental radiographs

Clinically, BRONJ is similar to osteoradionecrosis and manifests itself as dehiscence and destruction of the oral mucosa with exposure of a necrotic jaw bone, yellow and irregular. This bone necrosis, painless or painful, can arise spontaneously or after dentoalveolar surgery. This pathology can be associated with gingival redness, edema, oozing intra and extra-oral, mobility adjacent teeth, sinusitis, purulent nasal discharge, paresthesia, enlarged lymph nodes, pathological fracture and spontaneous detachment of necrotic bone in the oral cavity [31].

Before the clinical manifestation of necrotic bone exposure, patients may develop an BRONJ with nonspecific symptoms, and may remain so for weeks or months, and at this early stage, little or no radiographic change can be observed and radiographic alterations of BRONJ are not specific [10]. There are characteristic clinical parameters in the diagnosis of BRONJ, but the imaging findings provide important information about the course, magnitude and progression of the disease.

An increase of radiopacity is observed before clinical evidence of the necrosis, mainly in areas of high bone remodeling. The radiologic findings of the BRONJ are found in other conditions such as osteomyelitis, osteoradionecrosis, cancer metastasis, periapical inflammatory lesions [10]. The early phase of BRONJ may not show any significant changes on panoramic and periapical films. In this stage, the imaging findings seem to be sclerosis with poor corticomedullary differentiation and involvement of the inferior alveolar canal which is clinically accompanied by tooth mobility [26]. It is possible to see in BRONJ's images also

widened periodontal ligament space, unhealed extraction space, osteoclerotic lamina dura and narrowing of the marrow space [27]. Bone sequestrum, fractures, cortical destruction, permeative appearance, areas of low attenuation and periosteal reaction are associated with the late disease. An important finding radiograph of BRONJ is persistent alveolar sockets. In this advanced phase can also be observed areas of mottled bone. In severe cases, the BRONJ creates an ill-defined radiolucency [32].

Radiographs may appear normal when lesions are smaller than 1 cm and are insensitive in demonstrating the extent of the lesions or complications [33]. The maxilla can be involved with necrosis and in these cases the radiographs may reveal abnormalities in the adjacent maxillary sinus and mucoperiosteal thickening to air-fluid levels [34].

Computed tomography can be useful, because it reveals the full extent of bone involvement and the presence of bone sequestrum. Magnetic resonance [MRI] and bone scintigraphy may also help in the early detection of this disease [35].

Treatment

There is general consensus in the literature on the fact that dental preventive measures in patients being candidate for or already receiving BP are extremely important to reduce the risk of BRONJ development. Preventive measures are applied during intravenous bisphosphonate treatment, with endodontic and periodontic therapy preferred over tooth extractions and with avoidance of dental implants [21]. It is important to educate the patient to become cognizant of certain things for example: maintenance of good oral hygiene, routine oral examination, removal of nonviable teeth, completion of any invasive dental treatment and achievement of optimal dental health [26].

The first aim of treating patients with BRONJ is to eliminate clinical symptoms such as pain, treat any infection of the soft tissues

or bone, and minimize the progression of bone necrosis [10]. The use of antimicrobial mouth rinses [chlorhexidin or hydrogen peroxide] and/or analgesia is proposed for patients with clinical evidence of BRONJ such as exposed bone but in the absence of any evidence of infection. They are used to reduce the risk of bone infection (AAOMS, Stage 1) [10]. In the second and third stage where symptoms and signs of infection are present, systemic antibiotics and analgesics are indicated in addition to antimicrobial mouth rinses [36]. Various antibiotic regimens have been tested in several studies such as penicillin, doxycycline, quinolones, metronidazole and clindamycin. Broad-spectrum antimicrobial therapy [phenoxymethylpenicillin, amoxicillin, clindamycin or metronidazol] is recommended although the correct duration of treatment is not clear [36].

The surgical treatment is necessary to remove necrotic bone and create soft tissue coverage of remaining healthy bone. The most commonly recommended approach is to remove symptomatic bony sequestra with minimal soft tissue disturbance and avoiding further bone exposure, although some authors prefer more extensive soft and hard tissue debridement and primary closure of the wound [28,36]. More radical surgical management is advocated where there are large segments of necrotic bone or where there is pathological fracture of the bone (AAOMS, stage 3) [36].

Adjunctive therapies suggested for the management of BRONJ include hyperbaric oxygen [HBO], parathyroid hormone, platelet rich plasma and lasers [37]. With the exception of HBO, the literature consists primarily of small case series and further studies need to be undertaken before any are considered for routine use [36].

DISCUSSION

The importance of the topic Bisphosphonates for health professionals is quite clear. It is essential to understand the risks that dental procedures

performed in patients who use bisphosphonates, and the impact of possible consequences on the quality of life of patients.

Since 2003, bisphosphonate therapy has been a source of concern in the field of dentistry. This was based on the reports of the first known cases of BRONJ associated with BP intravenous therapy administration. Most situations reported are typically associated with a surgical procedure, a simple extraction, which precipitates the onset of osteonecrosis. The number of dental appointments made by patients treated with BP may increase to facilitate prevention, diagnosis or needed treatment to osteonecrosis [38]. Most cases of BRONJ occur mainly in the posterior region of the mandible, Maxilla may also be affected at the posterior region. The simultaneous involvement of both the maxillary and mandibular bones may also occur [39]. The half-life of BFs is approximately 10 years and the long-term use result in substantial drug accumulation in the skeleton [28]. Thus, it would take a long period of discontinuation of the medication to eliminate the drug from the body. The liquid presentation of the medication is often not possible because of the benefits they provide for the drug treatment for the prevention of osteoporosis and bone metastases [14].

The individual benefit from BP treatment ultimately depends on the weight of risk factors for osteoporotic fracture — such as age, bone mineral density, race, family history and fracture history — and on the presence of risk factors for atypical fractures and other potential complications [40]. These may include treatment duration, pathway of administration and perhaps also the binding properties of the BP in question [40]. Therefore, the optimum duration of treatment is unlikely to be the same in all patients and the benefit of treatment will almost always be greater in patients who are at an elevated risk of osteoporotic fractures. Furthermore, the evidence of bone fracture risk reduction with BPs is somewhat stronger at the short term and the effect of BP treatment does not disappear immediately on pausing the drug or ceasing treatment entirely.

Undoubtedly, the clinical examination is sovereign in diagnosis of osteonecrosis after using BPs, but it is possible, through imaging analysis, to assess the course, magnitude and progression of the disease, allowing clinicians to recognize the risk, consider the action and perform dental procedures safely.

Osteonecrosis leads to physical damage, but also interfere with the quality of life of patients. Thus, it is important for patients who already use BPs or are initiating this treatment, especially by intravenously pathway, to be carefully evaluated by a medical and dental support to avoid the disease.

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Yamba Carla Lara Pereira

(Corresponding author)

Avenida Bandeirantes, 3900

Monte Alegre - Ribeirão Preto - SP

CEP: 14.040-900

e-mail: yambacarla@bol.com.br

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SHORT COMMUNICATION

May the flexural strength of ceramics be influenced by salivary pH?

A resistência à flexão de cerâmicas pode ser influenciada pelo pH salivar?

Caroline COTES¹; Vanessa Cruz MACEDO¹; Mariana Andrade CAMILLO¹; Bruna Pastro de LARA¹; Rodrigo Furtado de CARVALHO¹; Carolina da Silva Machado MARTINELLI¹; Estevão Tomomitsu KIMPARA¹

1 - UNESP – Univ Estadual Paulista – School of Dentistry – Department of Dental Materials and Prosthodontics – São José dos Campos – SP – Brazil.

ABSTRACT

Objective: This study purpose was to compare the three-point flexural strength of feldspathic ceramic after storage in artificial saliva for 30 days with different pH regimens, as acidic pH (3.5), neutral pH (7.0), basic pH (10) and alternating between acid/basic pH, for 15 days each. **Material and Methods:** The bars were luted with resin cement and subjected to storage in artificial saliva of different pH values. **Results:** The values of flexural strength were significantly higher for bars stored in distilled water, at neutral and basic pH, when compared with the results for bars stored in acidic pH and in acid/basic pH. **Conclusions:** Storage for 30 days in artificial saliva at acidic pH, or alternating between acidic and basic pH, can reduce the mechanical properties of ceramics.

KEYWORDS

Dental porcelain; Artificial saliva; Compressive strength.

RESUMO

Objetivo: O objetivo do estudo foi comparar a resistência à flexão três pontos de uma cerâmica feldspática após armazenagem em saliva artificial, durante 30 dias com diferentes regimes de pH: pH ácido (3,5), pH neutro (7,0), pH básico (10) e alternando entre pH ácido/básico durante 15 dias cada. **Material e Métodos:** As barras foram cimentados com cimento resinoso e submetidas a armazenagem em saliva artificial com os diferentes valores de pH. **Resultados:** Os valores de resistência à flexão foram significativamente maiores para as barras armazenadas em água destilada, em pH neutro e em básico, quando comparadas com os resultados para as barras armazenadas em pH ácido e em meio ácido/básico. **Conclusões:** A armazenagem durante 30 dias em saliva artificial, com pH ácido, ou alternando entre pH ácido e básico, pode reduzir as propriedades mecânicas das cerâmicas.

PALAVRAS-CHAVE

Porcelana dental; Saliva artificial; Força de compressão.

INTRODUCTION

Ceramics are widely used in dentistry because of their excellent properties. Chemical durability is one of the requirements for their intra-oral use, since dental prostheses must be resistant to degradation in a wide range of variable pH solutions. Although ceramics are considered to be the most inert of all dental materials used for restorations[1], in reality, some ceramics are not chemically inert, even in a neutral

aqueous environment[2], and storage in acid or basic pH can result in degradation of their flexural strength[1,3].

However, few studies have been performed to measure the effects of chemical 'attack' on ceramics over the entire pH range[1], and no study has evaluated the effects of pH changes on their mechanical properties. Therefore, the aim of this study was to evaluate the flexural strength of a feldspathic ceramic stored at different pHs.

MATERIALS AND METHODS

Fifty bars (20 mm x 2 mm x 2 mm) of ceramic (VITA VM7, VITA Zahnfabrik, Bad Säckingen, Germany) were fabricated according to the manufacturer's instructions. The bars were etched with 10% hydrofluoric acid for 20 s, washed, and dried. The silane (RelyX Ceramic Primer, 3M-ESPE, Seefeld, Germany) was applied to the etched surface, and after 60 s an air-spray was applied for 5 s. The luting agent (RelyX ARC, 3M-ESPE, Seefeld, Germany) was manipulated according to the manufacturer's instructions and applied to the treated surface. A load of 750 g was applied to standardize the luting layer, and the specimens were light-cured for 40 s on each side. Specimens were stored for 30 days in artificial saliva (except for control), at 37 °C, and the solutions were changed each 5 days. The bars were divided into 5 groups (n = 10) according to storage regimen:

Group 1(C): control, distilled water

Group 2(A): acidic pH (3.5) artificial saliva

Group 3(N): neutral pH (7.0) artificial saliva

Group 4(B): basic pH (10) artificial saliva

Group 5(A/B): alternating between acid/basic pH artificial saliva, for 15 days each.

The three-point flexural strength test was performed in 37 °C distilled water, and the flexural strength values (MPa) were subjected to one-way ANOVA and Tukey's test.

RESULTS

The results of the one-way ANOVA test showed statistically significant differences among the groups (p-value = 0.000) (see the Table 1).

Table 1 – Mean and standard deviation values the groups, and results of Tukey's test

Groups	Means and standard deviations (MPa)	Tukey's test*
C	95.63±12.45	A
A	61.27±14.35	B
N	87.71±14.70	A
B	89.16±16.74	A
A/B	62.71±18.80	B

*Different letters show statistically significant differences.

DISCUSSION

The mechanical properties of feldspathic ceramics were affected by storage in acidic artificial saliva for 30 days and 15 days with alternation. Shorter storage in acid pH did not cause surface changes [4], but after 7 days of acid immersion, surface deterioration could be observed [1,2], with numerous porosities and small cracks covering the feldspathic ceramic surface [2]. This surface alteration could reduce the ceramic's flexural strength, although the crack growth rates of existing flaws were similar over time, regardless of the salivary pH [3]. Furthermore, the hardness of feldspathic ceramic could decrease with an increase in the pH [3], but this might not be responsible for the decrease in flexural strength, since storage in basic and neutral pH showed no influence on the present data. It was observed that the acid pH was more detrimental to flexural strength, even when alternating acidic and basic storage media.

The use of luting agent before the storage is justified for standardization of tensile surface in tensile test and for simulation of ceramic clinical use. The combination of surface pre-treatment and luting switches the fracture origin from the porcelain/cement interface to cement surface, similar of what occurs clinically [5].

The observations from this study indicate that dietary habits including ingestion of highly acidic foods, like citric fruits, might result in lowered mechanical strength of ceramic restorations over the long term. Exposure of ceramics to gastric acid could also be harmful to their properties [4], but this effect may be reduced by dilution and through the action of buffering systems [2]. It should be taken into account that this study had limitations, since the conditions in which the samples were stored were not identical to those found in the oral cavity, which presents a more complex environment [2], such as more rapid temperature changes, masticatory stress, daily brushing, and pH changes for each food ingested, as well as for acidic by-products released by bacteria. However, clinicians should be aware that ceramics could have decreased resistance in patients who ingest acidic foods or have bulimia.

CONCLUSION

The flexural strength of feldspathic ceramic stored in acid or acid/basic pH might be decreased in comparison with that of ceramics stored in basic and neutral pH artificial saliva, or in distilled water.

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Vanessa Cruz Macedo
(Corresponding author)

Av. Eng Francisco José Longo 777
Jd. São Dimas – São José dos Campos
12245-000 – São Paulo – SP – Brazil

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ORIGINAL ARTICLE

Effects of typified propolis on mutans streptococci and lactobacilli: a randomized clinical trial

Efeitos da própolis tipificada nos estreptococos do grupo mutans e lactobacilos: ensaio clínico randomizado

Camilo ANAUATE NETTO¹; Maria Cristina MARCUCCI²; Niraldo PAULINO²; Andrea ANIDO-ANIDO¹; Ricardo AMORE¹; Sergio de MENDONÇA³; Laurindo BORELLI NETO¹; Walter Antonio BRETZ⁴

1 - Biomaterials Research Group – School of Dentistry – UNIBAN Bandeirante Anhanguera University – São Paulo – SP – Brazil.

2 - Professional Masters Program in Pharmacy – School of Pharmacy – UNIBAN Bandeirante Anhanguera University – São Paulo – SP – Brazil.

3 - Microbiology Research Group – Professional Masters Program in Pharmacy – UNIBAN Bandeirante Anhanguera University – São Paulo – SP – Brazil.

4 - Department of Cariology & Comprehensive Care – College of Dentistry – New York University – New York-NY – USA.

ABSTRACT

Objective: The aim of this study was to determine in a randomized, double-blind, placebo-controlled clinical trial the effects of typified propolis and chlorhexidine rinses on salivary levels of mutans streptococci (MS) and lactobacilli (LACT). **Methods:** One hundred patients were screened for salivary levels of MS >100,000 CFUs/mL of saliva. All patients presented with at least one cavitated decayed surface. Sixty patients met entry criteria. Subjects were adults 18-55 years old. After restoration of cavitated lesions patients were randomized to 3 experimental groups: 1) PROP-alcohol-free 2% typified propolis rinse (n = 20); 2) CHX- 0.12% chlorhexidine rinse; 3) PL-placebo mouthrinse. Patients rinsed unsupervised 15 mL of respective rinses twice a day for 1 min for 28 days. Patients were assessed for the salivary levels of MS (Dentocult SM) and LACT (Dentocult LB) at baseline, 7-day, 14-day, and at 28-day visits (experimental effects) and at 45-day visit (residual effects). General linear models were employed to analyze the data. **Results:** PROP was superior to CHX at 14-day and 28-day visits in suppressing the salivary levels of MS (p < .05). PROP was superior to PL at all visits (p < .01). The residual effects of PROP in suppressing the salivary levels of MS could still be observed at the 45-day visit, where significant differences between PROP and CHX (p < .05), were demonstrated. PROP was significantly superior than CHX in suppressing the levels of salivary LACT at the 28-day visit (p < .05). **Conclusion:** Typified propolis rinse was effective in suppressing cariogenic infections in caries-active patients when compared to existing and placebo therapies.

KEYWORDS

Randomized clinical trial; Typified propolis; Chlorhexidine; Mutans Streptococci; Lactobacilli.

RESUMO

Objetivo: O objetivo deste estudo foi determinar, em um estudo randomizado, duplo-cego, placebo-controlado os efeitos da própolis tipificada e clorexidina sobre os níveis salivares de estreptococos do grupo mutans (EM) e lactobacilos (LACT). **Métodos:** Cem pacientes foram selecionados para níveis salivares de MS > 100.000 UFC/mL de saliva. Todos os pacientes apresentaram pelo menos uma superfície cariada com cavitação. Sessenta pacientes preencheram os critérios de inclusão. Os indivíduos eram adultos com 18-55 anos de idade. Após a restauração das lesões cavitadas, os pacientes foram randomizados para três grupos experimentais: 1) PROP- bochecho livre de álcool de 2% de própolis tipificada (n = 20), 2) CHX- bochecho de clorexidina 0,12%, 3) PL- bochecho placebo. Os participantes bochecharam sem supervisão 15 mL dos enxagatórios duas vezes por dia, durante 1 minuto, durante 28 dias. Os pacientes foram avaliados para os níveis salivares de MS (Dentocult SM) e LACT (Dentocult LB) na linha de base, e após 7 dias, 14 dias, 28 dias (efeitos experimentais) e 45 dias (efeitos residuais). Modelos lineares foram utilizados para analisar os dados. **Resultados:** PROP foi superior ao CHX nas visitas de 14 dias e de 28 dias na supressão dos níveis salivares de SM (p < 0,05). PROP foi superior ao PL em todas as visitas (p < 0,01). Os efeitos residuais de PROP na supressão dos níveis salivares de MS ainda foi observado na visita de 45 dias, onde diferenças significativas entre PROP e CHX (p < 0,05) foram demonstradas. PROP foi significativamente superior a CHX na supressão dos níveis salivares de LACT na visita de 28 dias (p < 0,05). **Conclusão:** O enxagatório de própolis tipificada foi eficaz na supressão de infecções cariogênicas em pacientes com atividade de cárie quando comparado a terapias existentes e ao placebo.

PALAVRAS-CHAVE

Ensaio Clínico Randomizado; Própolis tipificada; Clorexidina; Estreptococos do grupo Mutans; Lactobacilos.

INTRODUCTION

Fluorides and chlorhexidine are arguably the most common agents utilized for the prevention of oral diseases. These chemical agents have been available for use to the general population where chlorhexidine, particularly, has been used to promote gingival health for over 45 years [1]. The effectiveness of chlorhexidine rinses in fighting gingivitis has extensive documentation as its efficacy is evident from reports using the methodology of meta-analysis [2]. The use of chlorhexidine mouth rinses in the prevention of dental caries however is contradictory. Clinical evidence on the application of chlorhexidine gels and varnishes for the prevention of dental caries is also inconclusive [3].

Propolis is a resinous matter collected by honeybees from different plant exudates, which is used to seal beehives. At least 200 compounds have been identified in different propolis samples of different botanical geographic origins. The typified propolis has standardized constituents such as: prenylated phenolic acids derived from p-coumaric, including it [4]. The literature on propolis use in dentistry is extensive. There are numerous laboratory and clinical reports of propolis that include: suppression and inhibition of cariogenic [5] and periodontal organisms [6], prevention of respiratory infections [7] and gingival inflammation, [8] inhibitory activity against endodontic pathogens [9], and therapeutic action on oral ulcers [10]. These reports however lack evidence of propolis effectiveness because adequately designed randomized controlled trials have yet to be conducted.

Studies comparing propolis with chlorhexidine solutions have been limited to in vitro studies. These studies have suggested that

propolis solutions were equivalent to chlorhexidine solutions in inhibiting the mutans streptococci [11]. The primary aim of this investigation was to determine in a randomized, double-blind, placebo-controlled clinical trial the experimental and residual effects of typified propolis and chlorhexidine rinses on salivary levels of the mutans streptococci and lactobacilli.

MATERIALS AND METHODS

Inclusion/Exclusion Criteria

One hundred-fifty patients were screened from a patient pool attending the Dental Clinics at Bandeirante Anhanguera University – UNIBAN, São Paulo, Brazil. After signing informed consent approved by the Institutional Review Board (UNIBAN-Protocol N.0038/2007), patients were submitted to eligibility criteria. The main entry criteria for participants was to present with salivary levels of the mutans streptococci >100,000 CFUs/mL of saliva and to present with at least one cavitated decayed surface. Additional entry criteria included: the presence of at least 20 teeth, no clinical signs of periodontal disease, age range of 18 to 55 years-old, not being a current smoker, normal saliva secretion rate, not being pregnant, and not making use of any oral topical or systemic medication.

Subject Population/Demographics

This was a randomized double-blind placebo-controlled clinical trial. Sixty patients met entry criteria. These participants were 18-55 years old of both genders and in good general health. Table 1 depicts demographic and clinical characteristics of study participants. Study groups were well balanced at baseline for demographic variables and for the number of decayed and restored teeth.

Table 1 – Demographics and clinical parameters of study participants at entry

Parameter / Group	Chlorhexidine	Propolis	Placebo	Sig.
Age	41.6 (13.4)*	39.4 (9.8)	39.0 (11.7)	ANOVA NS
Gender				Chi-square Test NS
Male	7	8	9	
Female	13	12	11	
Race				Chi-square Test NS
White	15	13	14	
Black	2	4	4	
Parida	3	3	2	
Number of Decayed Teeth	3.0 (3.1)*	2.9 (1.9)	4.15 (3.41)	ANOVA NS
Number of Restored Teeth	10.1 (6.2)*	6.9 (5.0)	6.6 (5.2)	ANOVA NS

*mean (standard deviation).

Treatment Products and Protocol

After restoration of all cavitated lesions patients were randomized to 3 experimental groups: 1) alcohol-free, 2% typified propolis mouth rinse (n = 20). Propolis 2% rinse was manufactured at the laboratories of the Department of Pharmacology at Federal University of Santa Catarina, Florianópolis, Santa Catarina, Brazil. The formulation included 2% typified propolis, mint flavor, polioxyethelers, sorbitol, blue color and water; 2) a commercially available 0.12% chlorhexidine mouth rinse; 3) placebo mouth rinse that matched propolis mouth rinse without the active ingredient. Patients rinsed 15 mL of the experimental rinses twice a day for 1 min for 28 days. Rinsing was performed in the morning and before bedtime after ordinary oral hygiene procedures. Patients were assessed for the salivary levels of mutans streptococci (Dentocult SM, Orion Diagnostica, Espoo, Finland) and lactobacilli (Dentocult LB, Orion Diagnostica, Espoo, Finland) at baseline, 7-day, 14-day, and at 28-day visits (treatment effects) and at 45-day visit (residual effects). All adverse reactions were documented and patient accountability/continuance criteria were recorded at all visits.

Allocation Concealment

For allocation of groups a computer-generated list of random numbers was used. Rinses were prepared in dark-bottles, which were consecutively numbered according to the randomization schedule. Participants were randomized to one of the three test color-matched rinses. Study coordinator, examiners and participants were unaware of group allocation. The group identity was generated and kept in Florianópolis, SC, Brazil while the study was conducted in São Paulo, SP, Brazil.

Mutans Streptococci Assay

The Dentocult SM test was employed to determine the salivary levels of the mutans streptococci. Two thirds of a treated plastic strip was inserted into the mouth and rotated on the surface of the tongue about 10 times. This

strip was placed into a culture vial containing a well-mixed bacitracin solution and processed according to the manufacturer's instructions. Interpretation of test scores using a density chart was as follows: 0-1: <100,000 CFU/mL of saliva, 2: >100,000 to <1,000,000 CFU/mL and, 3: >1,000,000 CFU/mL.

Lactobacilli Salivary Levels

The Dentocult LB assay was employed to estimate the levels in saliva of lactobacilli. Saliva collected after stimulation was poured over agar surfaces, ensuring that they are well moistened. Excess saliva was allowed to drain from the slide. The slide was screwed tightly back into the tube and placed in an upright position in an incubator ($36 \pm 2^\circ\text{C}$) for four (4) days. The salivary levels of the lactobacilli were estimated as follows: 0- Non-detectable; 2- 1,000 CFU/mL saliva; 3- 10,000 CFU/mL saliva; 4- 100,000 CFU/mL saliva; 5- 1,000,000 CFU/mL saliva.

Product Satisfaction Questionnaire

Participants were asked to rank mouth rinses according to taste, breath improvements, nausea symptoms, perception of oral cleanliness, ease to use, and olfactory perception. Participants then ranked each item with scores ranging from 1 (excellent) to 5 (poor) for an overall score based on the range of acceptance for a particular mouth rinse.

Statistical Analysis

Univariate models were employed to analyze the data of treatment effects between study groups for the salivary levels of cariogenic bacteria. Analysis of co-variance was performed to compare treatment effects for all groups between baseline and 28 days and between baseline and 45 days for the salivary levels of the mutans streptococci and lactobacilli adjusted for age and gender. Chi-square tests were employed to analyze frequency distributions of demographic parameters. ANOVA was employed to estimate differences among study groups at baseline for age, and for the number of decayed and restored teeth. We employed SAS (r).

RESULTS

Adverse reactions were reported for the chlorhexidine and placebo groups at high frequencies with regards to flavor, burning sensations and alterations of taste. Patient satisfaction and acceptability was highest and excellent for the propolis mouth rinse (74%) followed by the chlorhexidine (68%) and placebo (45%) mouthrinses, respectively.

Analysis of co-variance revealed significant treatment effects from baseline to 28 and 45 days for both propolis ($p < 0.05$) and chlorhexidine ($p < 0.05$) groups for the salivary levels of mutans streptococci. These same findings were not observed for the salivary levels of lactobacilli. The propolis mouth rinse was superior to chlorhexidine and placebo rinses at 7-day, 14-day and 28-day visits (treatment effects) in suppressing the salivary levels of the mutans streptococci (Table 2). The chlorhexidine was superior to placebo at 7-day and 14-day visit. The propolis mouth rinse was superior to the placebo rinse at all visits (treatment period) in suppressing the salivary levels of the mutans streptococci. The residual effects of propolis mouth rinse in suppressing the salivary levels of mutans streptococci could still be observed after 17 days of product discontinuation, where significant differences between the propolis rinse and chlorhexidine and placebo rinses, were demonstrated.

Very little information is available on the efficacy and superiority of suppression of salivary levels of lactobacilli by means of antimicrobials. The data presented in Table 3 shows that propolis mouth rinse was significantly different than chlorhexidine mouth rinse in suppressing the levels of salivary lactobacilli at the 28-day visit.

DISCUSSION

Upon search of the literature it is apparent that this is the first randomized double-blind placebo-controlled trial on the effects of propolis on cariogenic bacteria. Although there are several in vitro studies confirming the inhibitory activity

of propolis against the mutans streptococci and in vivo studies attesting the efficacy of chlorhexidine on the suppression of the mutans streptococci, our study design does not permit comparisons with the existing literature as data is not available with study design and product evaluation similar to our protocol.

Despite the high number of initial decayed and restored teeth present in our study population (Table 1), the propolis and chlorhexidine rinses were effective in suppressing the salivary levels of the mutans streptococci from baseline up to 45 days after a 4-week twice-a-day daily use. Similar results were not found for the placebo group. These results need to be put in perspective as a high number of restorations allows for rapid re-colonization of the mutans streptococci [12;13] and, therefore, had our study design been of a longer duration we are unsure if results presented here would have been extended for a longer period of time.

Group analysis at the various point-visits revealed superior suppression of the mutans streptococci for the propolis rinse when compared to placebo and chlorhexidine rinse at days 7, 14 and 28. Chlorhexidine rinse was superior to placebo at day-7 and day-14 visits but not at day -28 visit (Table 2). The residual effects of the rinsing protocols clearly show that propolis rinse could sustain suppression of the mutans streptococci after 17 days of rinse discontinuation. Notably, we would have expected chlorhexidine rinse to exert similar effects because of chlorhexidine substantivity.

We are unaware of any clinical studies on the effects of propolis rinses on salivary levels of the lactobacilli. Our study has demonstrated that after 4-week use of propolis rinse a significant suppression of the salivary levels of lactobacilli was evident when compared to chlorhexidine and placebo rinses (Table 3). This is added benefit for the propolis rinse as suppression of lactobacilli is hard to attain as recently shown in comparative studies employing chlorhexidine rinses [14,15].

Table 2 – Effects of rinses on mutans streptococci salivary levels

Group	Baseline	7 days	14 days	28 days	45 days
Propolis 2% (n=20)	21 (0.3)	16 (0.5) ^a	13 (0.5) ^{ab}	12 (0.4) ^a	14 (0.5) ^a
Chlorhexedine 0.12% (n=20)	23 (0.5)	17 (0.6) ^b	18 (0.5) ^a	17 (0.7) ^a	19 (0.5) ^a
Placebo (n=20)	22 (0.4)	22 (0.7) ^{ab}	19 (0.5)	26 (0.6) ^a	26 (0.7) ^a
ANOVA p-value	0.177	0.008	0.001	0.000	0.000
	Treatment Period				Residual

a, b - Numbers with same superscripts are significantly different by Tukey's pairwise comparisons.

Table 3 – Effects of rinses on lactobacilli salivary levels

Group	Baseline	7 days	14 days	28 days	45 days
Propolis 2% (n=20)	2.9 (0.9)	2.8 (1.0)	2.5 (0.7)	2.5 (0.7) ^a	2.7 (0.8)
Chlorhexedine 0.12% (n=20)	3.5 (0.9)	3.3 (1.1)	3.1 (0.8)	3.1 (0.8) ^a	3.1 (0.8)
Placebo (n=20)	2.7 (0.9)	2.9 (0.8)	2.7 (1.0)	3.0 (0.9)	3.1 (1.0)
ANOVA p-value	0.021	0.265	0.108	0.033	0.220
	Treatment Period				Residual

a, b - Numbers with same superscripts are significantly different by Tukey's pairwise comparisons.

Limitations of this study include the non-determination of the power of our sample size prior to the commencement of the study. Although our study groups were well-balanced at baseline for various parameters (Table 1) and the fact that we were able to demonstrate superiority of propolis rinses, no sample size calculations were performed during design of this protocol.

Lastly, our questionnaire survey showed higher acceptance of propolis rinse for various factors when compared to chlorhexidine and placebo rinses. One recent study evaluated the compliance and acceptability of a 5% propolis rinse [16], and although most subjects reported the unpleasant taste of the rinse, they said they were satisfied with the rinse and would recommend its use by others. Only 24% of individuals reported difficulties in following the study protocols.

CONCLUSIONS

Typified propolis rinses may be of value in suppressing cariogenic infections in caries-active patients when compared to existing and placebo therapies.

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Dr. Camillo Anauate Netto
(Corresponding author)

Programa de Pós-Graduação em Odontologia
UNIBAN Universidade Bandeirante Anhanguera
Rua Maria Cândida, 1813 - Vila Guilherme
02071-013 São Paulo, SP
Tel: 55(11)2967-9145
e-mail: anauatenetto@uol.com.br

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ORIGINAL ARTICLE

Mechanical behavior of NiCr and NiCrTi alloys for implant prosthetic components

Comportamento Mecânico de ligas NiCr e NiCrTi utilizados para componentes protéticos de implantes

Ivete Aparecida de Mattias SARTORI¹, Carla Müller RAMOS², Fernanda FAOT¹, Luciano Monteiro da SILVA³, Laiz VALGAS⁴, Ana Flávia Sanches BORGES²

1 - Latin American Institute of Dental Research – ILAPEO – Curitiba – PR – Brazil.

2 - University of São Paulo – Bauru School of Dentistry – Department of Dentistry – Endodontics and Dental Materials Bauru – SP – Brazil.

3 - School of Science – São Paulo State University – Bauru – SP – Brazil.

4 - Neodent® – Curitiba – PR – Brazil.

ABSTRACT

Objective: The aim of this study was to evaluate the tensile and hardness mechanical properties, as well as the composition and microstructure of three different alloys used for implant prosthetic components casting. **Methods:** The alloys were divided into three groups: Tilite (Tilite), Vera (Verabond) and Malloy (DanCeramalloy). For the tensile test, the specimens (n = 10) of each group were evaluated in “alter” form and the maximum load fracture, the deformation at maximum load and Young’s modulus were determined. The data was subjected to one-way ANOVA and Turkey’s test. For the hardness test, five discs from each group were evaluated for Vickers hardness. The data was analyzed using multiple regression ANOVA followed by the Turkey’s test. The significance level was set at 5% ($\alpha = 0.05$). The composition and microstructure was determined through analysis of two specimens from each group by metallographic analysis (MEV/EED). **Results:** With regards to maximum load tensile, the deformation and the Young’s Modulus the three alloys evaluated were statistically similar. Regarding hardness, Tilite showed significant higher values than the others alloys. **Conclusion:** All the examined alloys can be used in implant prosthetic components and the presence of the element Ti did not influence the mechanical behavior of the alloy.

KEYWORDS

Prostheses and implants; Hardness tests; Alloys; Tensile strength.

RESUMO

Objetivo: O objetivo deste estudo foi avaliar as propriedades mecânicas de tensão e dureza, assim com a composição e microestrutura de três diferentes ligas utilizadas em componentes protéticos de implantes. **Métodos:** As ligas foram divididas em três grupos: Tilite (Tilite), Vera (Verabond) e Malloy (DanCeramalloy). Para o teste de tensão, os espécimes (n = 10) de cada grupo foram avaliados em forma modificada e a força máxima de fratura, a deformação em carga máxima e o módulo de Young foram determinados. Os dados foram submetidos ao teste ANOVA a um fator e teste de Tukey. Cinco discos de cada grupo foram avaliados para o teste de dureza Vickers. Os dados foram analisados utilizando o teste ANOVA de regressão múltipla, seguido do teste de Tukey. O nível de significância adotado foi de 5% ($\alpha = 0,05$). A composição e microestrutura foram determinados em dois espécimes de cada grupo por meio de análise metalográfica (MEV/EED). **Resultados:** Não houve diferenças estatísticas entre as três ligas estudadas em relação à força de tensão máxima, à deformação e ao módulo Young. Em relação à dureza, Tilite mostrou valores estatisticamente superiores em comparação às outras ligas. **Conclusão:** Todas as ligas examinadas podem ser utilizados em componentes protéticos de implantes e a presença do elemento Titânio não influenciou o comportamento das ligas.

PALAVRAS-CHAVE

Próteses e implantes; Ligas; Testes de dureza; Resistência à tração.

INTRODUCTION

In The NiCrMo alloys are the most utilized for the casting of prosthetic abutments as well as for metaloceramic crowns in Brazil, due to their low cost and satisfactory properties, when compared to other alloy compositions [1]. However, these alloys are not totally accepted for casting because they contain low percentages of beryllium and other chemical elements such as nickel, which could cause damage [2]. In addition, due to their high hardness, the NiCrMo alloys present difficulties in their laboratory manipulation. Some specific procedures are required before ceramic firing [3,4]. NiCrMoTi alloys are commercially available and may be used in replacement of the NiCrMo alloy. Titanium (Ti) is a chemical element that presents satisfactory mechanical properties [5-7] and corrosion an absence of corrosion in the oral environment due to its titanium oxide passive layer [8,9].

An important property is the mechanical strength, such as hardness and tensile strength. Hardness is the first strength, like a “surface protection”, against the first contact with external loads that can cause plastic deformation [10]. An alloy with high hardness protects the abutment against surface plastic deformations, which is very important, considering that abutments need to resist loads from other materials with a different Young’s modulus and pass on the implants. Many prosthetic ceramic crowns, based on lithium disilicate and leucite reinforced, present different fracture resistance values under fatigue [11]. Furthermore, the tensile strength can identify future behavior patterns of alloys, such as like their rigidity (Young’s modulus), plastic deformation under load and their strength in the maximum tensile load, which predicts how resistant the alloy is before a fracture. NiCrMo and NiCrMoTi comparisons are needed in order to predict their clinical behavior for basic mechanical requirements.

METHODS AND MATERIAL

The metallic alloys evaluated are three that are commercially available in Brazil. Trademarks, composition and manufacturers are described in Table 1. The tensile and hardness specimens were casting from resin material (Neodent, Curitiba, PR, Brasil).

Table 1 – Sintering cycles of studied porcelains

Groups	Trademark	Composition	Manufacturers
Tilite	Tilite®	Ni – 60 to 76% Cr – 12 to 21% Mo – 4 to 14% Ti – 4 to 6%	Talladium Inc., Valencia, CA, USA
Vera	Verabond®	Ni – balanced Cr – 14% Mo – 8.5% Al – 1.7% Be – 1.8%	VeraBond ; Aalba Dent Inc, Cordelia, Ca, USA
Malloy	Dan ceramalloy®	Ni – 65.7% Cr – 20% Mo – 8% Others – 6.3%	Osaka, Japan

*Chemical composition from manufacturer´s, however, according to Bezzon et al. (1998) the Ni quantity varies from 68 to 80% for NiCr alloys.

Tensile test

The tensile specimens (n = 10) of each group were constructed in “alter” form, with 3 mm (\pm 0.1) of diameter in the tensile area, according to #1562 ISO specifications (Figure 1a, 1c and 1d).

The tensile test was carried out on a universal test machine INSTRON 3382 (Instron Corporation, Norwood, MA, USA). The maximum load fracture, the deformation at maximum load and the Young’s modulus were determined. The data was subjected to one-way ANOVA and Turkey’s test.

Hardness test

Five discs measuring 5 mm in diameter and 2 mm in thickness (Figure 1b) from each group were evaluated for Vickers hardness (kg/mm²), recommended by the manufacturer without prior surface treatment, in a micro hardness tester (HMV-2, Shimadzu, Tokyo, Japan). The mean values were calculated by means of three indentations with a distance of 150 μ m between each of them, with a load of 100 g for 10 s. Data was analyzed using a multiple regression ANOVA test followed by the Turkey’s test. The significance level was set at 5% (α = 0.05).

Metallographic analysis

For the metallographic analysis two randomly chosen hardness specimens from each group were mounted on a semi-automatic mounting press for hot compression (LaboPress 1, Struers, Ballerup, Denmark) with phenolic resin (MultiFast, Struers, Ballerup, Denmark). The basic steps for the metallographic specimen preparation included: mounting, planar grinding, rough polishing, final polishing and etching, followed by an optical microscopic analysis. The plane grinding was performed with 220- to 1000-grit silicone carbide paper (Struers, Ballerup, Denmark) under water cooling in a LaboPol-21 grinding/polishing machine (Struers, Ballerup, Denmark). The rough polishing was performed with a LaboPol-5 polishing machine (Struers, Ballerup, Denmark) and a 6 μm diamond suspension (Struers, Ballerup, Denmark), and the final polishing with 3 and 1 μm diamond pastes (Arotec, São Paulo, Brazil) at 200 rpm without refrigeration. To prevent any contamination between abrasives, the surfaces were washed with propyl alcohol. In order to maintain a shiny surface, the alloys were chemically and mechanically polished using a mixture of colloidal silica (OP-S, Struers, Ballerup, Denmark) and hydrogen peroxide. The polishing pattern was produced by dipping the samples for 20 seconds into an acid mixture of Kroll's reagent (100 mL water, 6 mL nitric acid, 3 mL hydrofluoric acid), and the reaction was interrupted with 70% alcohol. After drying, the surface microstructure was examined by a light microscope (Olympus BX60, Hamburg, Germany) 200x magnifications and photographed by a digital camera (Olympus SC35 Type 12, Hamburg, Germany).

After that, the two specimens from each group were chemically analyzed by Energy Dispersive Spectroscopy (EDS) (Scanning Electron Microscopic JEOL JSM-5600LV, Tokyo, Japan).

The results of the morphological and microstructural analysis were submitted to further descriptive analysis and the chemical components identified had their spectra arranged as well as their regions analyzed.

RESULTS

The metallic alloys evaluated are three that are commercially available in Brazil. Trademarks, composition and manufacturers are described in Table 1. The tensile and hardness specimens were casting from resin material (Neodent, Curitiba, PR, Brasil).

For the maximum load tensile, the three alloys were similar. The values were 756.25, 776.03 and 722.05 kgf for Tilite, Vera and Malloy, respectively (Figure 1). Deformation at maximum load among the three alloys were similar. The values were 9.91 (2.98), 12.37 (3.63) and 9.99 (2.04) for Tilite, Vera and Malloy, respectively (Figure 2). The results of Young's modulus showed higher values for Vera, 57.50 (48.87) GPa, in comparison to Tilite values, 52.08 (40.15) GPa. Malloy values, 55.57 (38.18) GPa did not differ from the other two groups (Figure 3). Regarding hardness, Tilite showed statistically significant higher values (433.32 ± 11.77) than the others alloys; which were 357.24 ± 14.93 for Vera and 377.72 ± 32.99 for Malloy) (Figure 4). The metallographic images and EDS data were described (Figures 5 and 6).

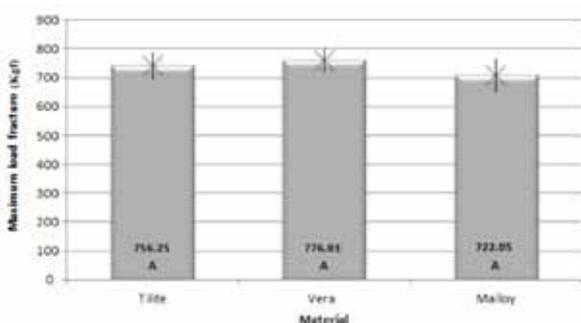


Figure 1 – Ultimate load fracture under tensile (Mean \pm SD) of the evaluated alloys.

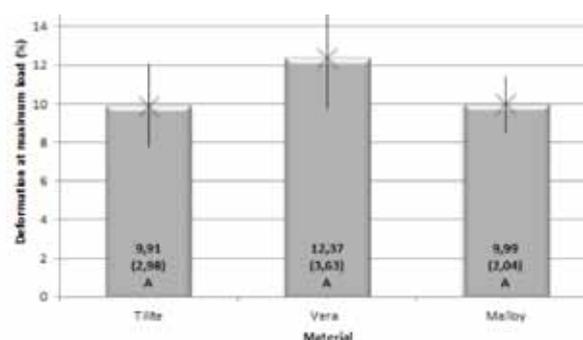


Figure 2 – Deformation at maximum load (Mean \pm SD) of the evaluated alloys.

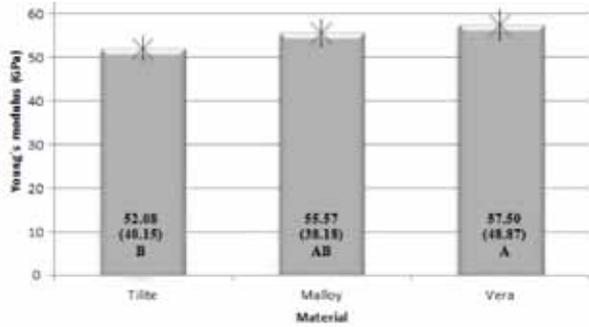


Figure 3 – Young's modulus (Mean ± SD) of the evaluated alloys.

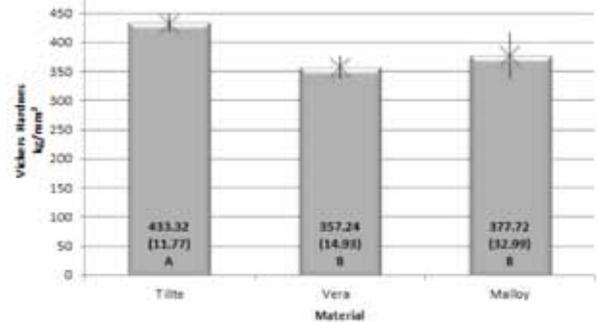


Figure 4 – Vickers Hardness (Mean ± SD) of the evaluated alloys.

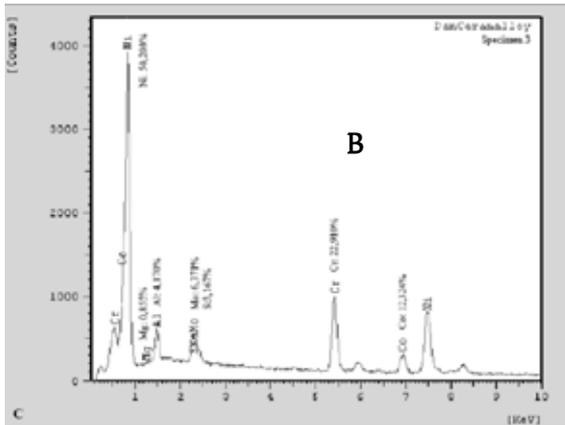
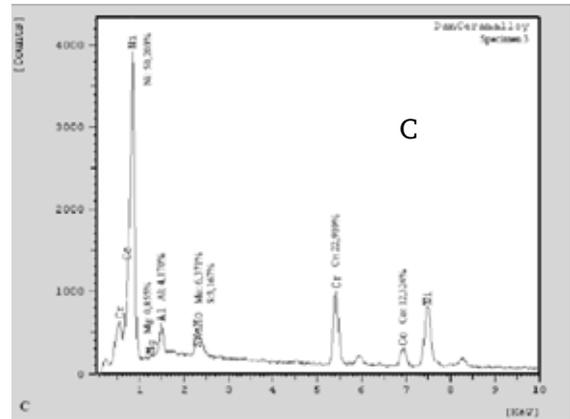
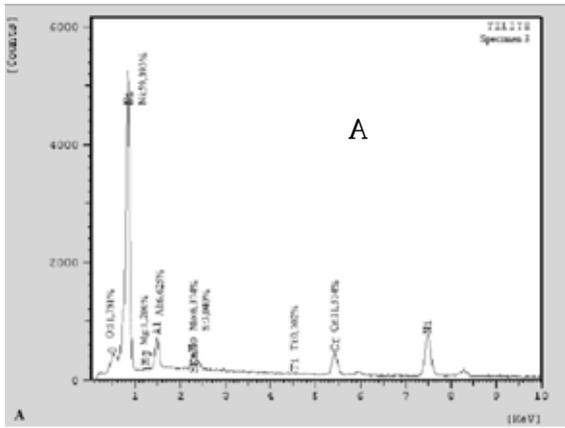


Figure 5 – The most representative Chemical Analysis of A) T tilite, B) Vera and C) Malloy. Percentage by weight of each chemical.

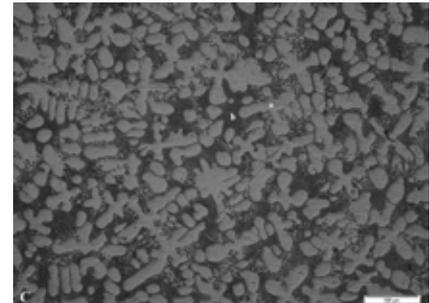
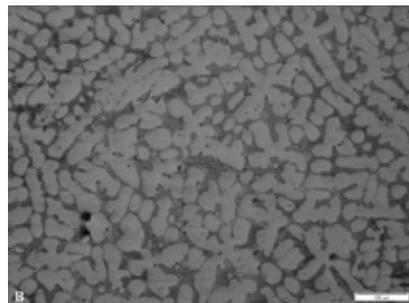
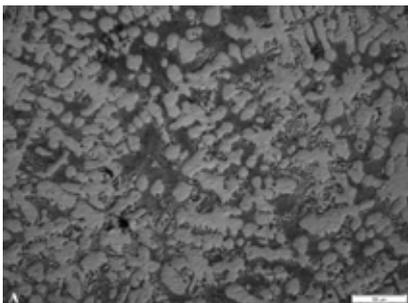


Figure 6 – Metallographic Analysis A) Tilite, B) Vera and C) Malloy. a) phase gamma (γ) and b) an intermetallic phase (δ').

DISCUSSION

It is relevant to emphasize the clinical mechanical properties for selection between the different types of alloys available in the market for use in prosthetic components. The value of the modulus of elasticity must be high for bridges of the various elements, whereas the deflection of the alloy should be minimized and the fracture strength is essential for restoration to be subjected to substantial functional loads [12]. Additionally, the laboratory manipulation of these alloys for a more precise adjustment of the castings, will be dependent on the relationship between the elastic modulus, elongation and strain.

The quality of an elastic material, which is presented by the interatomic or intermolecular force is related to the forces of attraction of a material which is similar when it is subjected to tensile or compression is the case with prosthetic components [13]. Young's modulus is the tension divided by deformation up to the elastic limit of each material under load [10]. This property is generally independent of any thermal or mechanical treatment, but depends largely on the composition of the material [13]. In the present study, the three alloys presented similar results of elastic modulus to each other and with values indicated by the manufacturers, confirming the findings of chemical elements, that are also similar.

The maximum load fracture, the deformation at maximum load and the Young's modulus of the three alloys are not affected by the concentrations of their elements. Deformation at maximum load (%) is calculated by the formula $= [(L_f - L_i) / L_i] \times 100$; L_f = Final length of the specimen body after the test start until its fracture; L_i = Initial length of the specimen body. The presence of high amounts of plastic deformation characterizes the material as a ductile fracture. This fracture is often preferred because the deformation gives a warning and preventive measures are taken, in addition, a higher strain energy is required to induce this type of fracture [23].

When a metal is stretched beyond its yield point, hardness and resistance to deformation increases as the discrepancies are concentrated along the intergranular boundaries and thus, a plastic deformation area becomes more difficult [10].

Concerning hardness, Tilite alloy was the hardest of the three alloys. These values were similar to those related in previous studies that evaluated under the same conditions [17,24]. Analyzing the surface hardness of the different materials sit is important, not only to explain the risk of surface roughness of the prosthetic component during the cleaning work or even during normal oral hygiene procedures [14], but also may show the need for heat treatments in order to optimize properties such as tensile and fatigue [15]. In addition, alloys with higher hardness properties can hinder the finishing and polishing, requiring more time to run and altering the routine prosthesis laboratory [16,17].

Tilite is the only one that has titanium in its composition, suggesting that the presence of this chemical element could be related to the higher hardness. Ti shows a satisfactory mechanical performance and excellent corrosion resistance [5,21]. However, according to the results of chemical analysis, the Ti while present in small amounts (0.524% by weight), did not justifying the higher hardness of Tilite.

The metallographic has an important role in ensuring that products have the correct microstructure, determine whether a material was processed correctly and may explain why the alloy faile [18]. In the metallographic analysis of the three alloys, no differences were observed between the structures. Figure 6 shows that there is a similarity in size and direction of the dendrites formed during the solidification of the alloys. All the alloys had a microstructure of two equilibrium phases, as shown in Figure 7, consisting of gamma (γ) and gamma-prime (γ') and the phases resulting in a transition zone between the matrix and the training inter dendritic eutectic [19]. The γ' ,

an intermetallic phase, is responsible for the high temperature resistance of the material and its resistance to deformation. However, it was observed that the alloy Vera presented a more homogeneous matrix can be sufficiently low because on the quantity of chemical elements present in the alloy and all of these elements have been incorporated into a solution nickel.

The amount of γ'' depends on the chemical composition and temperature, as shown in the quaternary phase diagram [20]. The addition of Cr in the alloy forms Ni-Cr dendrites and is essential for oxidation resistance, due to the formation of a Cr-rich film, that is highly resistant to acid attack, while adding the molybdenum-based alloy NiCr increases resistance to localized corrosion [2,22]. These elements (Cr and Mo) are also enhancers, both in a solid solution γ and γ' phase [20]. With regards to the different chemical composition based alloys of the Ni-Cr studied, some scientific findings are important in regard to critical interpretation. For general applications in dentistry, the Ni-Cr base alloys added with 12% Cr, 2-5% Mo are recommended for improving corrosion resistance [2]. For the Ti oxide layer and its reactive and protective properties, usually based on TiO₂ oxide, an excellent range of Ti in the passive film of the NiCr-based alloy is still unknown [2,15].

Within the limitations of this study, it was concluded that all three alloys can be used to fabricate prosthetic components with appropriate physical and microstructural properties. Higher hardness and lower Young's modulus of Tilitite predict a higher initial resistance to better absorption of strain and stress to prosthetic abutments. The presence of the element Ti did not influence the mechanical behavior of the alloy.

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**Carla Müller Ramos
(Corresponding author)**

Rua Jacarezinho, 656
Bairro Mercês - Curitiba
80710-150 – Paraná - PR
Email: carla_muller@yahoo.com.br

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ORIGINAL ARTICLE

Micro-hardness of acrylic resin utilized for provisional crowns: Effect of different polymerization techniques and pH-Cycling.

Avaliação da microdureza de resina acrílica para coroas provisórias: Efeito de diferentes técnicas de processamento e ciclos dinâmicos de pH.

Cesar Augusto Zanlorenzi NICODEMO¹, Carlos Eduardo Edwards REZENDE¹, Rafael Tobias MORETTI-NETO², José Henrique RUBO¹

1 - Bauru School of Dentistry - University of São Paulo - Bauru - SP - Brazil.

2 - Alfenas Federal University - Alfenas - MG - Brazil.

ABSTRACT

Objective: This study aimed at evaluating the micro-hardness of an acrylic resin used for provisional crowns. **Materials and methods:** Five different processing techniques (direct and indirect) were assessed: (I) auto polymerizing resin in sandy stage; (II) auto polymerizing resin in plastic stage; (III) bead-brush technique; (IV) auto polymerizing resin under pressure; (V) heat-cured acrylic resin under pressure. Five specimens were made for each test group. For the initial micro-hardness test, the specimens were immersed in deionized water for 48 hours. For the analysis of the final micro-hardness, the specimens were subjected to pH-cycling to simulate the changes in the pH level which occur in the oral cavity. **Results:** Tests revealed that the micro-hardness was decreased after the pH-cycling. However, no statistical difference was found among the different types of acrylic resin polymerization techniques. **Conclusion:** It was concluded that the micro-hardness is not directly related to the polymerization technique for making provisional crowns.

KEYWORDS

Acrylic resins; Dental crowns; Dental prosthesis; Temporary dental restoration.

RESUMO

Objetivo: Avaliou-se a dureza superficial de uma resina acrílica (PMMA) utilizada para a confecção de coroas provisórias. **Material e Métodos:** Cinco diferentes técnicas de processamento (Direta e Indireta) para polimerização da resina foram testadas: I - autopolimerização após mistura (pó + líquido) em pote dappen e inserção em matriz de silicóna na fase arenosa; II - autopolimerização após mistura em pote dappen com inserção em matriz de silicóna na fase plástica; III - autopolimerização utilizando a técnica do pincel; IV - autopolimerização sob pressão em matriz de silicóna; V - polimerização térmica utilizando líquido de polimerização rápida em mufla. Foram confeccionados cinco corpos-de-prova para cada grupo. Para o teste de microdureza inicial os espécimes foram imersos em água deionizada por 48 horas. Para a análise da dureza final, os espécimes foram submetidos a ciclos dinâmicos de pH, para simular a variação de pH ocorrida na cavidade oral. **Resultados:** Os testes revelaram que houve diminuição da dureza do material após a ciclagem ácida, porém não foi verificada diferença estatística entre os diferentes tipos de processamento da resina acrílica. **Conclusão:** Concluiu-se que a microdureza não está relacionada diretamente com a técnica de confecção dos provisórios.

PALAVRAS-CHAVE

Resinas acrílicas; Coroas dentárias; Prótese dentária; Prótese parcial temporária.

INTRODUCTION

The rehabilitation of partially edentulous patients often requires a long-term provisional stage that makes it possible for the

professional to foresee the success of the final restoration in its mechanical, aesthetic and functional aspects [1,2]. Provisional crowns work as a diagnostic device [3] and allow for the correction of the occlusal plane and vertical

dimension [2], establishment of occlusal guidance and posterior occlusal contacts, evaluation and conditioning of the gingival tissues and prediction of the shape, size and color of the restoration [4].

The most common material used in provisional crowns is the auto-polymerizing acrylic resins, more precisely poly-methyl methacrylate (PMMA). Because of the long-term use of provisional restorations, superficial wear can occur due to tooth-brush abrasion [5] and parafunction attrition [2]. Only the long-term use of provisional crowns is a determinant factor to promote superficial wear [6]. The dietary solvents are also an important factor on the superficial wear of the provisional restorations [7].

Provisional crowns can be made either chair-side (direct technique) or on a cast model with the help of a dental laboratory (indirect technique) [3]. The option for one of these techniques is based on factors like the type or the extension of the prosthesis and the expected treatment longevity. The polymerization technique can modify the acrylic resin properties [8,9]. It may compromise the quality of the provisional crowns and can contribute to the treatment failure [1,3,10]. Thus, it is important to evaluate, among the other properties, the superficial hardness of the acrylic resin when submitted to different processing protocols.

Superficial micro-hardness can be used for density indication, so that denser materials should have more resistance to superficial wear [2,11]. Thus, evaluating the micro-hardness of the acrylic resins signifies evaluating this material's capacity of maintaining the diagnostic elements provided by the treatment up to the definitive prosthesis cementation. There are few studies regarding the acrylic resin micro-hardness modification, mainly on the different techniques of resin processing. Thus, the aim of this study is to verify this micro-hardness alteration while varying the different technique.

MATERIAL AND METHODS

The present study consisted of two stages: 1st - evaluation of the micro-hardness considering five different techniques for provisional crowns production; 2nd - evaluation of the final micro-hardness after pH-Cycling.

For the first stage, 25 specimens were made in acrylic resin (Dencor® - Artigos Odontológicos Clássico LTDA, São Paulo – SP, Brazil), which is an auto-polymerizing PMMA for provisional crowns fabrication. The specimens consisted of a disc with 2 cm of diameter and 3 mm of thickness and were standardized from an impression made in silicone (Zetalabor® - Zhermack, Badia Polesine/ RO - Italy) which was used as a matrix. These specimens were equally divided into five different groups which were composed of different techniques of acrylic resin processing.

Except for group III (bead-brush technique), the mixing proportion used was: 1.50 g of polymer, measured by weight in a high precision scale (Sauter®, model K 1200, Switzerland) and 0.70 mL of monomer, measured by volume with a pipette (Pyrobras®) i.e equivalent volume ratio 3:1 as indicated by the manufacturer. The liquid and the powder were dispensed in a Dappen dish and smoothly mixed for five seconds.

Groups:

- Group I: for this group the liquid was saturated with the powder to obtain a smooth texture. The mold was filled with the acrylic resin immediately after saturation, in its sandy stage.

- Group II: the liquid was saturated with the powder to obtain a smooth texture. The resin was allowed to reach its plastic stage (1.5 to 2 minutes after mixing) keeping a glass slab on top of the Dappen dish to avoid excessive monomer evaporation. Then, the acrylic resin was inserted into the mold with a spatula.

• Group III: the acrylic monomer and polymer were dispensed in separate Dappen dishes in order to use the bead-brush technique. The tip of the brush was moistened by the liquid and put in contact with the powder. The silicone mold was continually filled with the resin until completely full.

• Group IV: the specimens were made by manipulating the acrylic resin in a Dappen dish and pouring it in the silicone mold when the resin was in the plastic stage. This mold was placed against a plaster platform and stabilized with rubber bands. The mold and plaster platform set was immersed in water under pressure ($3 \times 10^5 \text{ N/m}^2$) at 70°C for 15 minutes.

• Group V: five wax blocks were flaked with dental stone (Durone IV, Dentsply, Rio de Janeiro, Brazil), according to the conventional procedures. After the stone was set, the wax was removed and the acrylic resin was manipulated. When the plastic stage was obtained (1.5 to 2 minutes after saturation in Dappen dish), the resin was inserted into the flasks. The flasks were then taken to a hydraulic press and immersed in water under pressure ($3 \times 10^5 \text{ N/m}^2$) and heated to 70°C for 15 minutes. De-flasking was performed by routine laboratory procedure.

The specimens were finished in a lathe (APL 4, Arotec, Cotia, Brazil) using a sequence of 320, 600 and 1200 grit wet sandpaper. This system allows the automatic polishing of 6 specimens simultaneously. The final polishing was made by a felt disc impregnated with $0.3 \mu\text{m}$ alumina solution for 4 minutes in high speed and a load of 215 g. The goal of this stage was to standardize the surfaces of the specimens before the micro-hardness measurements.

Digital microscopy analysis

To test the initial micro-hardness, the specimens were embedded in deionized water for 48 h and then dried at 37°C for 24 h in a stove. After this procedure, the Knoop micro-

hardness measurements were made with a microdurometer (HMV-2000/ Shimadzu Corporation, Japan), which was linked to a computer with a specific software used for the image analysis (Cams-Wins-New Age Industries/ USA). The microdurometer has a diamond tip which penetrates the specimen with a 0.025 Kg load for 10 seconds and a 10x objective lens was used to read the micro-hardness parameters (Fig 1).



Figure 1 - Microdurometer linked to a Desktop Computer.

When the microdurometer is activated, the diamond tip presses the specimen surface generating a lozenge shaped geometric figure which can be visualized by the contrast between the impression and the specimen surface. The lozenge allows for the determination of the superficial micro-hardness by the measurement of its major diagonal. This value is applied to a mathematical equation to obtain the results. The microdurometer used in the experiment automatically performs calculations, from two dotted marks overlapping to the sharp corners of the lozenge. (Fig 2)

The specimen's surface elected to the micro-hardness reading was divided into quadrants and one indentation was made in the center of each quadrant for all the specimens. On the other surface, markings were performed to allow the identification of the specimens during and after the pH cycling procedures. (Fig 3)

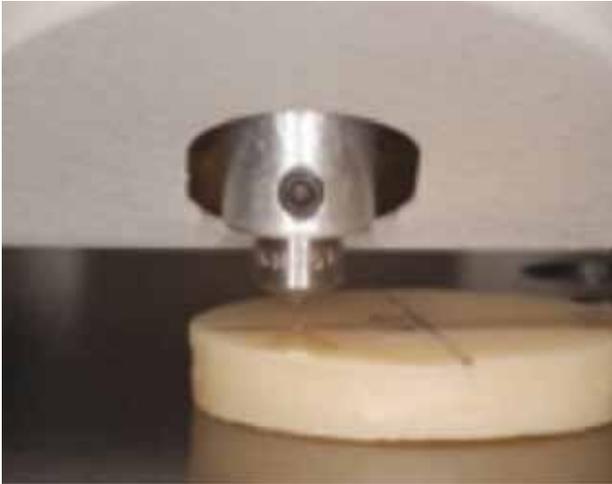


Figure 2 - Microdurometer diamond tip penetrating the specimen surface. The specimens were divided in quadrants.



Figure 3 - Specimens immersed in artificial saliva.

pH Cycling

In the second stage, the final micro-hardness of the specimens was analyzed after its submission to a pH dynamic cycling, according to the method proposed by Featherstone et al (1990) [12] and modified by Carvalho and Cury (1999) [13], to simulate the change of acidity occurring in the oral cavity. The pH cycling corresponded to the specimen's immersion in an acid solution for six hours and intercalated with artificial saliva for 18h during 15 days (Fig 3).

The acidic solution consisted of 2 mM calcium chloride and 2.0 mM potassium phosphate in a solution of 75 mM acetic acid at pH 4.3. The artificial saliva consisted of 1.5 mM calcium chloride, 0.9 mM potassium phosphate and 150 mM Potassium Chloride in a 2 mM

solution of hydroxymethyl-aminomethane at pH 7.0.

Statistical Analysis

After obtaining the data, the mean values for the micro-hardness, before and after the pH cycling, was calculated to classify the hardness reduction that occurred for the different groups. These values were grouped and compared with the 2-way variance analysis (ANOVA) for the variables processing technique and pH cycling and the Tukey Test was used for the comparison among the groups.

RESULTS

The statistical analysis revealed a significant difference before and after the pH cycling. The Knoop micro-hardness test measurements revealed a reduction in the superficial hardness after the pH cycling for all the evaluated specimens (Table 1).

Regarding the acrylic resin processing technique, there was no statistical significant difference found among the studied groups, thus, the technique used to fabricate temporary crowns does not influence the superficial hardness. The interaction between the variables included in this experiment was verified with the 2-way Anova test, adopting a significance level of $p < 0.05$. No statistical significant difference was found for the interaction of the studied variables (Table 2).

Table 1 – Mean and standard deviation (SD) values of micro-hardness for the different methods of processing before and after the pH cycling

Groups	Before pH Cycling	After pH cycling
	(Mean ± SD)	(Mean ± SD)
I	14.06 ± 0.61A	13.03 ± 0.48 B
II	13.47 ± 0.48A	12.71 ± 0.44 B
III	14.12 ± 0.65 A	12.93 ± 0.34 B
IV	13.73 ± 0.87 A	12.65 ± 0.35 B
V	13.48 ± 1.01A	12.73 ± 0.29 B

* Different letters indicate statistical significance.

Table 2 – Variances analysis (ANOVA) between the tested groups and phases (before and after de pH Cycling) and the interaction between both factors

	Df Effect	MS Effect	Df Error	MS Error	F	P
Group	4	0.506593	20	0.326566	1.551273	0.226
Phase	1	11.57767	20	0.384614	30.10206	0.000*
Interaction	4	0.096787	20	0.384614	0.251647	0.905

* Statistical significant difference. ($p < 0.05$)

DISCUSSION

The term hardness is related to the resistance of the material for penetration [11]. Thus, the material hardness is determined by standardized tests that promote the penetration of a tip into this material with the use of a specific instrument known as a durometer. For the analysis of the polymeric materials, the Knoop hardness mensuration is more recommended [2,14,15], because measures the major diagonal lozenge length which is maintained without dimensional changes, since the elastic recuperation and dimensional alterations occur on the shorter diagonal. Thus, the Knoop hardness value is virtually independent of the tested material ductility due to the action of tearing on the major diagonal [11].

Studies concerning the acrylic resin hardness often obtain the measurements after the immersion of the specimens in distilled water, disinfectant solutions [16] or alcohol [14]. However, the use of these solutions does not simulate the oral environment dynamics, which is characterized by constant variations in the bacterial biofilm's pH levels [13].

According to Cate (1990) [17], the pH variations in the oral environment are due to the bacterial biofilms action on the consumed fermentable carbohydrates (fast reduction of the pH levels) and to the saliva's buffering capacity (gradual elevation of the pH levels), characterizing the de-mineralization phenomena. Throughout the day, this phenomenon results in a series of reductions in the pH levels, interspersed with resting periods. The simulation of this pH variation in the oral environment was made in vitro through the daily immersion of the specimens for limited periods in alternate

solutions which promote demineralization and remineralization. The laboratory model of pH cycling allows for the in vitro analysis of the influence of de-mineralization process on the dental materials [17]. In this study, the pH cycling protocol first developed by Cate (1990) [17] for in vitro study and later modified by Carvalho and Cury (1999) [13], proved to be relevant for the evaluation of the pH dynamics on the acrylic resin hardness.

In the present study, all specimens showed a reduction of micro-hardness after the dietary simulating, in accordance with the studies of Yap et al (2004) [7] and Akova et al (2006) [18], who demonstrated a reduction in the micro-hardness after the storage of the specimens in heptane and ethanol solutions with different concentrations, in order to simulate the human diet. Some similarities were also found with other studies, in which there was a continuous decrease in hardness related to the storage time of specimens in water [16] and artificial saliva solution [6]. Conversely, the in vitro test performed by Whitman et al (1987) [14] stated that the conventional acrylic resin, IPN resin and Isosit stored in water had no significant variations between the initial and final hardness values, i.e., no loss of material hardness was found. However, when stored in ethanol, there was a reduction in the values, indicating a loss of hardness [7,14,18].

The superficial hardness reduction can occur due to the water sorption phenomenon by polymeric materials. The water excess can cause a filler-matrix debonding [18]. In addition, the absence of cross-linked bifunctional acrylates in the methyl methacrylate based materials could further the softening effects of dietary acid solvents [7]. Another factor that

may have influenced the hardness reduction for all the specimens is the presence of residual monomer, which adversely affects the mechanical properties due to a plasticizing effect that decreases the interchain forces and allowing the deformation to occur easily under load [9]. Regarding the storage of the specimens in artificial saliva, it may also contribute to the reduction of Knoop Hardness, because the saliva can act like water, causing the phenomenon of plasticizing and reduction of acrylic resin hardness [19].

In the present study, the five evaluated groups represent the main techniques for provisional crowns fabrication, in which two of them were made in laboratory (Groups IV and V) and the others three are direct techniques (groups I,II and III). Based on studies evaluating other properties, such as roughness and porosity [1,3], a statistical difference among these techniques was expected, however, this evidence was not demonstrated. Thus, based on the results obtained in this study, is not possible state that the hardness of an acrylic resin is related to the technique used for provisional crown fabrication, either laboratorial or directly made. This is in contradiction with the results of studies by Lee et al. (2002) [10] and Jo et al. (2011) [2], who found a higher micro-hardness for techniques in which the specimens were immersed in water under heating, as the specimens of the groups IV and V of the present study. This difference in results is probably attributed to the distinction of the methods used.

Evidently, only the observation of the micro-hardness is not sufficient to indicate a better technique for provisional crowns fabrication. The evaluation of others physical properties must be considered, such as roughness and porosity, wear after brushing simulation, flexural strength and others. Furthermore, the evaluation of these properties in different brands of acrylic resins and simulating other conditions present in the oral environment, like thermal variation, will enable the Dentist to more accurately display the technical and material that best suits his need.

CONCLUSION

In conclusion, the pH cycling reduces the hardness of the acrylic resin, independently of the processing technique, while the different processing techniques of the acrylic resin did not seem to influence the material hardness.

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**Carlos Eduardo Edwards Rezende
(Corresponding Author)**

Alameda Octávio Pinheiro Brizolla,
9-75 - 17012-101 - Bauru - SP - Brazil

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ORIGINAL ARTICLE

Evaluation of dental staining using a dentifrice containing chlorhexidine and zinc acetate. A double blind randomized clinical trial

Avaliação de dentifricio contendo clorexidina e acetato de zinco. Estudo clínico controlado randomizado duplo cego

Emanuel da Silva ROVAI¹, Tábata de Mello TERA¹, Andrea Carvalho DE MARCO¹, Maria Aparecida Neves JARDINI¹, Mauro Pedrine SANTAMARIA¹, Warley David KERBAUY¹

1 -Institute of Science and Technology – UNESP – Univ Estadual Paulista – School of Dentistry – São José dos Campos – SP – Brazil

ABSTRACT

Objective: The aim of this study was to evaluate whether the association of chlorhexidine with zinc acetate in dentifrices formulations could reduce the emergence of extrinsic tooth stain. As second outcome check the clinical gingival parameters. **Methods:** 30 volunteers were randomly divided into three groups: CHX+Z, consisting of 10 participants who used a dentifrice with 0.8% chlorhexidine gluconate and zinc acetate 1.16%; CHX group, with 10 participants who used a similar formulation dentifrice without zinc acetate, and the Placebo group also with 10 participants who used a dentifrice formulation similar but without the chlorhexidine and zinc acetate. Patients were assessed at baseline and 60 days when the indexes of plaque, gingival bleeding and staining were collected. **Results:** The results showed that by day 60, there was a decrease of both plaque index(PI) and the gingival index(GI) for all groups. Additionally, it was observed that both groups using dentifrice containing chlorhexidine, showed more stain than placebo. The CHX+Z group showed less stain compared to the CHX group, but the difference was not statistically significant. The difference between Placebo and Chlorhexidine Groups was statistically significant ($p < 0.05$) when considered the stain intensity and area plus intensity scores. The CHX+Z group was as efficient in PI and GI reduction as the CHX group. **Conclusions:** The association of chlorhexidine with zinc acetate showed no additional benefits regarding reducing the staining. The dentifrices containing chlorhexidine presented higher reduction of GI and PI when compared to Placebo group.

KEYWORDS

Chlorhexidine; Zinc acetate; Staining index; Plaque index; Gingival index.

RESUMO

Objetivo: O objetivo deste estudo foi avaliar se a associação de clorexidina com acetato de zinco em dentifricios poderia reduzir o aparecimento de manchas dentárias extrínsecas. Como segundo objetivo verificar os parâmetros clínicos gengivais. **Materiais e métodos:** 30 voluntários foram divididos aleatoriamente em três grupos: CHX + Z, composta por 10 participantes que usaram um creme dental com 0,8% de gluconato de clorexidina e acetato de zinco 1,16%; grupo CHX, com 10 participantes que usaram uma formulação de dentifricio semelhante, porém sem acetato de zinco, e o grupo do placebo também com 10 participantes que usaram uma formulação dentífrica semelhante, mas sem a clorexidina e sem acetato de zinco. Os pacientes foram avaliados no início do estudo e após 60 dias, quando foram coletados os índices de placa, sangramento gengival e manchas. **Resultados:** Os resultados revelaram que ao dia 60, havia uma redução em ambos os índices, de placa (IP) e do índice gengival (IG) em todos os grupos. Além disso, observou-se que em ambos os grupos que utilizou dentifricio contendo clorexidina, mostrou um maior índice de manchas maior do que o placebo. O grupo CHX + Z apresentou um índice de manchas menor em relação ao grupo CHX, mas a diferença não foi estatisticamente significativa. A diferença entre os grupos quem continham clorexidina e placebo foi estatisticamente significativa ($p < 0,05$) quando considerado a intensidade e a área de manchas +intensidade das manchas. O grupo CHX + Z foi tão eficiente na redução IP e IG quanto o grupo CHX. **Conclusão:** A associação de clorexidina com acetato de zinco não mostraram benefícios adicionais em relação a redução de manchas extrínsecas . Os dentifricios com clorexidina apresentaram maior redução de IG e IP quando comparado ao grupo placebo.

PALAVRAS-CHAVE

Clorexidina; Acetato de zinco; Índice de manchas; Índice de placa; Índice gengival.

INTRODUCTION

Chlorhexidine is a cationic biguanide with broad antimicrobial activity. It is the most effectively antiplaque and anti-gingivitis agent used in dentistry due to its duration and spectrum of action.[1,2] It causes damage to bacteria cytoplasmic membrane, leading to lysis of cell, being bacteriostatic or bactericidal. It has an affinity for a large variety of substrates and persists in total surfaces, as a result of its high substantivity. [3]

Several formulations have been studied in different vehicles and concentrations, due to anti-plaque activity, prevention of periodontal and caries diseases. Although the literature shows several studies regarding chlorhexidine effectiveness, it is known that prolonged utilization of this substance might cause side effects, such as altered taste, burning sensation, irritation of the mucosa, and development of dental stains.[4] Reports on appearance of extrinsic dental stains have been described. This pigmentation occurs due to precipitation of chlorhexidine products, which interact with pigments derived from food, on oral surfaces.[5]

It was reported that 0.12% chlorhexidine formulation might cause adverse effects to users light, as staining on teeth. McCoy et al. noticed staining of teeth, tongue, and restorations in 18% of patients using 0.12% chlorhexidine for 14 days. In 2006, Guimarães et al. showed that 55% of patients who used rinse of 0.12% chlorhexidine associated with 0.05% presented dental stains. At the end of last century new formulations containing zinc emerged with the objective to reduce dental staining.[4,6,7]

Zinc is used in attempt to minimize dental stains caused by chlorhexidine. [8,9] However, factors such concentration of chlorhexidine and period of use might be related to the appearance of stainings. Another factor that may favor the appearance

of stains is the lack of dental plaque removal before the use of chlorhexidine.[10] Thus, the aim of the present study was to evaluate a dentifrice containing chlorhexidine digluconate and zinc acetate used for a period of 60 days, on the development of extrinsic tooth stains.

MATERIALS AND METHODS

This study was approved by the Ethics in Research Involving Human Subjects under protocol No. 022/2009-PH/CEP.

Study design

For this double-blind randomized clinical study, 30 volunteers of both genders aging 18-50 years were recruited among patients from São José dos Campos Dental College. All patients were informed about the purpose and procedures of the research to be performed, and signed a consent form.

Population

Inclusion Criteria

- a) Good general and oral health
- b) Have at least twenty-four teeth in mouth
- c) Do not smoke,
- d) Do not wear braces
- e) Do not use any type of mouthwash
- f) Do not use antibiotics 6 months prior to the study.

Randomization and allocation

The volunteers were allocated to groups (Figure 1), according to a computer-generated list. This process was accomplished by employing different people for treatment and exams. The randomization code was not broken until all data had been collected. The treatment groups were not revealed to the clinical examiner and professional statistician.

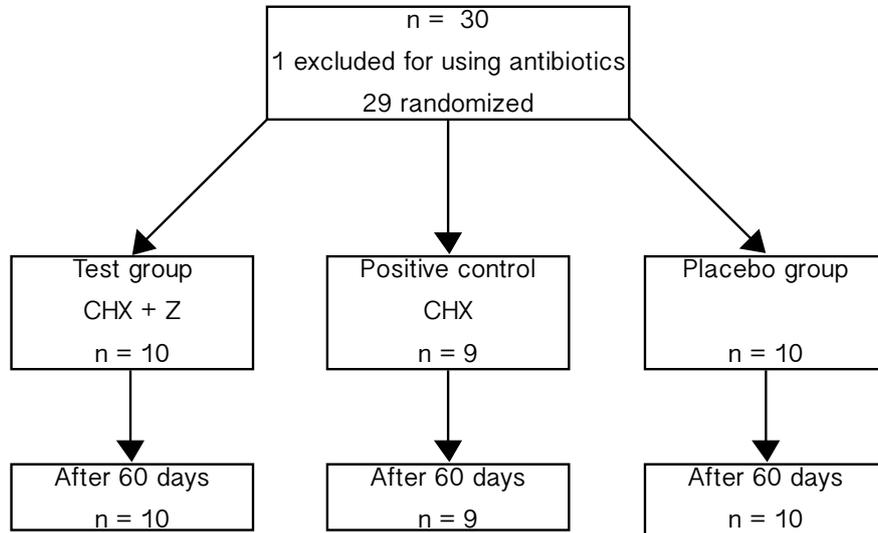


Figure 1 – Distribution of the patients in groups.

Treatment

Before the beginning of the study, all participants received oral hygiene instruction with atraumatic brushing technique and using dental floss, in order to standardize the way participants would make mechanical cleaning of the teeth. They received standardized toothbrushes and dental floss (Sorriso – Kolynos, Br). They were also instructed to put the same amount of toothpaste to perform brushing, which should be carried out for two minutes, three times per day for sixty days. All patients were asked about their consumption of foods and substances that might stain teeth, especially regarding the use of coffee, tea, red wine, and foods containing dyes.

Each participant was asked about the occurrence of any unusual reaction. At the end of the study, all stains that emerged were removed with professional cleaning.

The groups were as follows (Figure 1):

Group CHX+Z: 10 participants using a dentifrice with the following formulation: 2% Titanium Dioxide, 0.2% Aspartame, 0.04% Saccharin, 8% Glycerol, 12% Sorbitol, 1% peppermint menthol essence, 0.6% chlorhexidine gluconate, 0.8% Zinc acetate, and 1.16% Natrosol gel.

Group CHX: 10 participants using a dentifrice produced by the same compounding

pharmacy with similar formulation, except for the zinc acetate.

Group Placebo: 10 participants using a dentifrice produced by the same compounding pharmacy, but without chlorhexidine and zinc acetate.

Treatment

The following clinical parameters were evaluated:

Gingival Index (GI) according to Loe and Silness assessed at four sites: mesial-buccal, buccal, distal-buccal and lingual. All teeth were examined at baseline and 60 days, excluding third molars.[11]

The plaque index (PI) used was Turesky Index et al., a modification of the Quigley and Hein. Data were collected simultaneously to the gingival index assessments.[12,13]

Stains at buccal and lingual surfaces of incisors and canines were evaluated according to Macpherson et al. In the original article, canine teeth were not considered. The stain area, the intensity of staining and also the association of area and intensity of stains were considered for incisive tooth. Teeth were divided into three parts: body, approximal and gingival (Figure 2).[14]

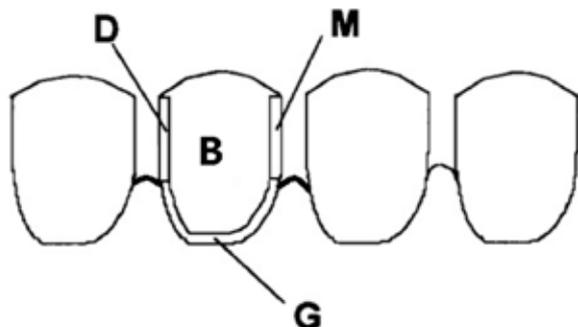


Figure 2 – Schematic drawing of the studied tooth areas. Figure adapted from MacPherson et al. (J Clin Periodontol 2000; 27:854–859) showing stain sites: B – body; D – distal area; M – mesial area and G – gingival area.

Briefly, the method used by Macpherson et al. was: Stain evaluations were performed by one examiner and recorded by a writer. Stain accumulation on the index teeth was scored using a modification of the Lobene stain scoring index. This involved visual stain assessment of the buccal/labial and lingual/palatal aspects of the index teeth. The modification consisted of dividing each aspect into 4 separate sites (Fig. 1) instead of only 2 (gingival and body): gingival (G): 2 mm wide strip running parallel to the gingival margin. The limit towards the incisal edge was given by the end of the interdental papilla; body of tooth (B): central area of buccal/lingual aspect, between gingival and distal/mesial sites, extending to incisal edge; mesial (M): visible area between line angle and adjacent tooth, ending at the interdental papilla (i.e. start of gingival site); distal (D): as for mesial (M) site. Stain was recorded using 2 separate characteristics, namely intensity and area (extent) as suggested by Lobene. The criteria for these 2 parameters were also slightly modified to provide better discrimination at the lower end of the scale and to take into account anatomical differences between the different sites.[14,15] The criteria and codes for intensity were:

- 0= no stain present, natural tooth coloration
- 1= faint stain
- 2= clearly visible stain, orange to brown
- 3= dark stain, deep brown to black

The area (extent) of the stain was recorded only if an intensity score of 2 or 3 was given. The area criteria and codes for approximal and gingival sites were:

- 1 = thin line, can be continuous
- 2 = thick line or band
- 3 = covering total area

The criteria and codes for area of the body of stained tooth are shown below.

At buccal/labial surfaces:

- 1 = Stain limited to pits/grooves.
- 2 = Stain outside pits/grooves, up to 10% of the area affected.
- 3 = Stain outside pits/grooves, more than 10% of the area affected.

In Lingual/palatal surfaces:

- 1 = Up to 1/3 of the area affected
- 2 = Between 1/3 and 2/3 of the area affected
- 3 = more than 2/3 of the area affected

Examiner Calibration:

The following data were collected at baseline and 60 days by a single calibrated examiner who was blinded in relation to the toothpaste used by each participant, and data were self-recorded.

The calibration was made by repeating a third of the sample 24 hours after the first examination. The parameters considered were: gingival index and tooth stains. The results were analyzed by intra-examiner Kappa test.

Statistical analysis:

Mean and standard deviation values of each parameter were described. The data showed normal distribution by Shapiro-Wilk. Statistical analysis was performed by Analysis of Variance test and the Tukey test as a post-hoc test using computer software (SIGMA PLOT for Windows, version 12.0 - State College, Pennsylvania, USA. Minitab Inc.).

To evaluate the differences within each group and among groups the level of significance was set at 5%.

RESULTS

In relation to Gingival Index, only the groups CHX and CHX+Z showed a statistically significant difference comparing baseline and 60 days, with p values < 0.001 , and the Placebo group with $p = 0.065$. When comparison between groups CHX+Z and CHX was performed, the differences were not statistically significant. When compared the same groups with Placebo the difference was significant. (Figure 3).

Related to plaque index, mean values and standard deviation of groups at baseline Placebo, CHX+Z and CHX were respectively 0.983 ± 0.894 , 1.185 ± 0.449 and 1.264 ± 1.089 , and the differences were not statistically significant. In the final evaluation period (60 days) no statistically significant differences were detected among them. There was a reduction of PI in all groups studied and when compared baseline and 60 days, all groups had statistically significant reduction with $p < 0.001$. The data are shown in Figure 4.

Stain index (SI) average values and standard deviations of the groups Placebo, CHX+Z and CHX at baseline and final assessment period are shown in Figure 4.

As can be observed in Figure 5, the Placebo group was the only group that did not show significant stain variation between baseline and 60 days, comparing the area, intensity and area+intensity.

At baseline the groups Placebo, CHX+Z and CHX showed average and standard deviation regarding the stained area of 0.1497 ± 0.1072 , 0.2891 ± 0.2314 and 0.1684 ± 0.2083 , respectively. Comparison among groups at baseline showed no statistically significant difference with $p > 0.05$. After 60 days, all groups showed increase in levels of staining, with mean and standard deviation of 0.2435 ± 0.2947 , 0.6906 ± 0.6505 and 0.8576 ± 0.4927 , respectively to Placebo, CHX+Z and CHX. For multiple comparison analysis, CHX versus Placebo group showed $p = 0.004$; group Placebo versus CHX+Z resulted in $p = 0.040$, and CHX+Z versus CHX presented $p = 0.620$. Thus, CHX+Z and CHX groups had statistically significant increase in the stained area as compared with the Placebo group, but the difference between the two was not statistically significant.

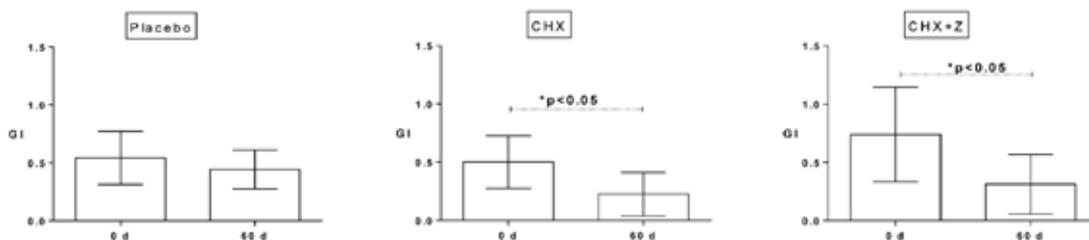


Figure 3 – Gingival Index at baseline and 60 days in Groups CHX+Z, Placebo and CHX. Group CHX+Z and CHX showed $p < 0.001$ and the Placebo Group $p = 0.065$

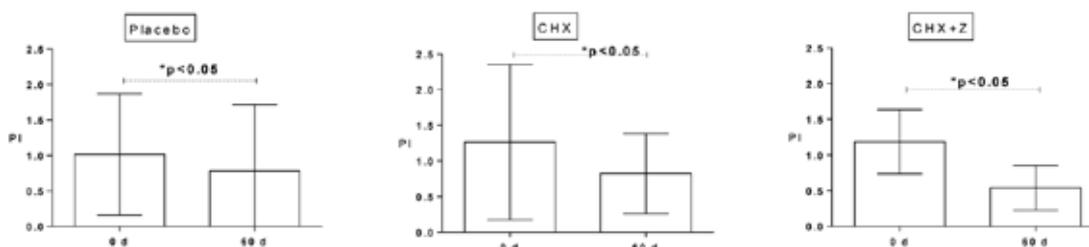


Figure 4 – Plaque Index at baseline and 60 days in Groups CHX+Z, Placebo and CHX. P-values comparing baseline and 60 days in all groups < 0.001 .

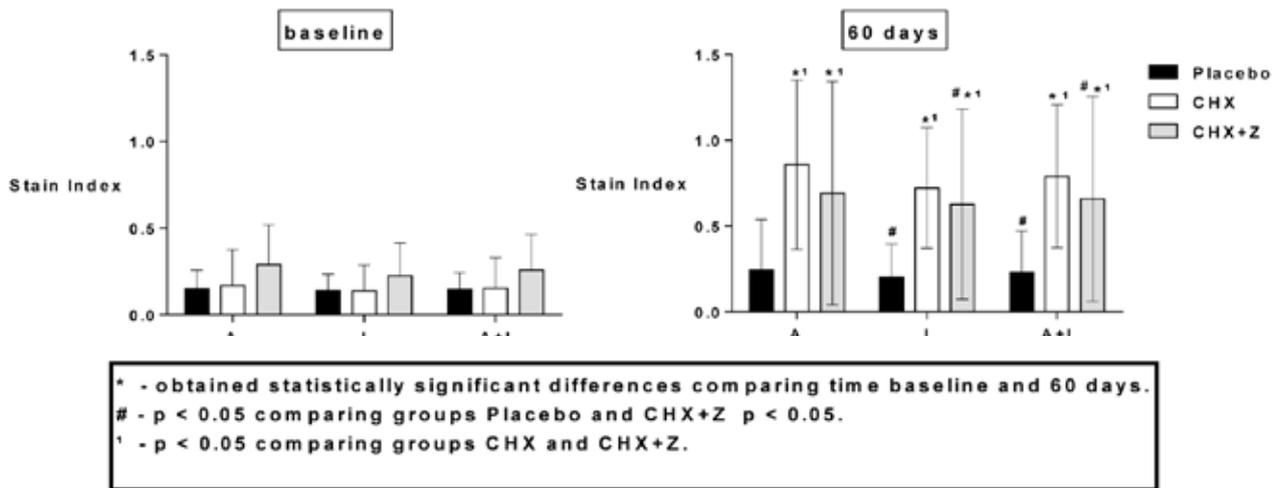


Figure 5 – Stain Index. Comparison of baseline and 60 days on the stained area (A), intensity stains (I) and area + intensity stains (A+I).

DISCUSSION

The use of dentifrices is an important tool against caries and periodontal disease. However, the association of mechanical action of brushing and flossing with the chemical control by chlorhexidine can be a great ally for plaque control. Nevertheless, studies have shown that the chronic use of chlorhexidine might cause side effects already described for McCoy et al., namely changes in dental surfaces such as stains, loss of taste, burning sensations or irritation of the mucosa, thus stimulating the search for new chemical compounds that may minimize the side effects and being affordable to the population. With this objective, we evaluated the effect of a dentifrice containing chlorhexidine associated with zinc acetate for a period of 2 months.[4]

During the period of the study we compared the results of the 3 groups: CLX + Z (test), CLX (positive control) and Placebo, and noted that group using dentifrice containing chlorhexidine associated with zinc acetate showed an improvement in periodontal clinical parameters and also showed a lower tendency to formation of extrinsic dental stains (intensity and area + intensity) compared to the positive control group, although this difference was not statistically significant.

The data showed that the PI had a statistically significant reduction in all groups when comparing baseline and 60 days, being slightly higher in the CHX + Z. This reduction occurred even in the placebo group due to the fact that all patients have been instructed about oral hygiene before the beginning of the study and also by monitoring every 15 days, leading to increased motivation for plaque control.

About 50% of the participants of this study were dental students with average of 23 years of age. This might explain the equivalence of gingival index, and stain index at baseline. Although we have not performed dental prophylaxis for the groups no interference on the results was detected, as there was no difference among groups at baseline.

In Sanz et al. study, the positive control received 0.12% chlorhexidine mouthwash while the test group used a dentifrice containing 0.4% chlorhexidine plus zinc. In the present study, the same concentration of dentifrices used in both groups prompted to an easier comparison, once the component zinc was the only variable in question.[7]

The ADS (Anti-Descoloration System) composed of ascorbic acid (vitamin C) and sodium metabisulfate has been associated with

chlorhexidine in order to reduce the formation of stains. Solis et al observed a decrease of close to 50% in formation of dental stains when compared with to conventional mouthwash containing chlorhexidine. However, it is important to note that the assessed period of 15 days and 0.2% in concentration were one fourth of those used in our study. Another study used the aluminum lactate, but Rathe et al. found no statistically significant difference when compared extrinsic stains between the test and positive control.[16,17]

Different studies have tested the prolonged use of mouthwash with lower concentrations of chlorhexidine in order to reduce the side effects, but Hoffman et al. concluded that to maintain the gold standard for this substance its concentration must be kept at least at 0.1% in mouthwash form.[18]

Oltramari-Navarro et al. tested dentifrices containing 0.5% and 0.75% chlorhexidine for 3 months and found that 0.5% chlorhexidine being more suitable for use in orthodontic patients because no significant increase in stain index and improvement in clinical parameters were observed. Group receiving 0.75% chlorhexidine showed a significant increase in stain indexes and also improvement in clinical parameters. In our study, we employed a similar concentration of chlorhexidine (0.8%), but associated with zinc acetate and also did not find a reduction in the appearance of stains when compared with the group that used only chlorhexidine [19].

At the end of this study, two subjects, one from CHX+Z group and other from CHX group showed SI index values significantly greater than the others. Knowing that the stains formation can vary according to the individual in the use of chlorhexidine these patients reported in their forms that had consumed foods with dyes in frequency and amount far above than the other participants, such as coffee, red wine and cola.[20]

Some limitations of our study should be considered. The first one is sample size. Although our study presented a similar sample size compared to other studies,

additional studies with larger sample size are recommended. Another limitation is the method of evaluation of the stains. We used a method that is considered subjective, even being consolidated by Macpherson et al. There are more objective methods that can be used, such as spectrophotometry.[14,16] Despite these limitations, studies aiming to evaluate tooth stains caused by chlorhexidine have used the same methodology.

CONCLUSION

Within the limitations of the present study, it can be concluded that in both groups that used chlorhexidine, the stains index was higher than placebo. The association of chlorhexidine with zinc acetate showed no additional benefits regarding reducing the staining. The dentifrices containing chlorhexidine presented higher reduction of GI and PI when compared to Placebo group.

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**Emanuel da Silva Rovai
(Corresponding address)**

Instituto de Ciência e Tecnologia
Curso de Odontologia – Depto de Dentística
Av. Francisco José Longo, 777, 12201-000 – São José dos Campos – SP, Brazil

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ORIGINAL ARTICLE

Incomplete vertical root fracture associated with lateral compaction technique: a microscopic analysis

Fratura radicular vertical incompleta associada com a técnica de condensação lateral: Análise microscópica

Gisselle Moraima CHÁVEZ-ANDRADE¹, Carolina ANDOLFATTO¹, Loise Pedrosa SALLES¹, Ana Livia Gomes CORNÉLIO¹, Gisele FARIA¹, Idomeo BONETTI FILHO¹

1 - Department of Restorative Dentistry – Araraquara Dental School – UNESP – Univ Estadual Paulista – Araraquara – São Paulo – Brazil.

ABSTRACT

Objective: This study evaluated the effects of root canal obturation employing lateral compaction technique and spreader load of 1.5 kg on the incidence of complete (CVRF) or incomplete vertical root fractures (IVRF). **Material and Methods:** Twenty-seven distal roots of extracted human mandibular molars were used. All root canals were prepared by biomechanical step-back technique and obturated by lateral compaction technique. The prepared roots were distributed into two groups: G1- experimental (n = 17) and G2- control (n = 10). During obturation, load of 1.5 kg was applied to a size # 30 finger spreader. Pre- and post-obturation images of the coronal portion of the roots were captured by inverted digital microscopy and analyzed by one trained examiner. Data were evaluated by Fisher's test ($p < 0.05$) using Graph Pad Prism 5.0. **Results:** No roots exhibited CVRF. All fractures observed before and after obturation were IVRF or "other defects". In G2 (control group), there was no increase of IVRF number. Interestingly, G1 presented an increase in the IVRF number to 70.59% in the 12 teeth out of 17 teeth studied. The statistical analysis showed that the mean of IVRF increased significantly in G1 when compared to G2 ($p < 0.05$). **Conclusion:** The application of a 1.5 kg spreader load during lateral compaction technique does not produce complete vertical root fractures, but may produce incomplete fractures or "other defects".

KEYWORDS

Endodontics; Root canal obturation; Vertical root fracture.

RESUMO

Objetivo: Este estudo avaliou os efeitos da obturação dos canais radiculares, empregando a técnica de condensação lateral com uma força do espaçador de 1,5 kg, na incidência de fraturas radiculares verticais completas (FRVC) ou incompletas (FRVI). **Material e Métodos:** Foram usadas 27 raízes distais de dentes molares mandibulares humanos extraídos. Todas as raízes foram instrumentadas por meio da técnica clássica ápice-coroa e obturadas pela técnica de condensação lateral. Após o preparo biomecânico, as raízes foram distribuídas em 2 grupos: G1- experimental (n = 17) e G2- controle (n = 10). Durante a obturação, uma força de 1,5 kg foi aplicada ao espaçador digital # 30. As imagens pré- e pós-obturação da superfície coronal das raízes foram capturadas por meio de um microscópio digital invertido e analisadas por um examinador treinado. Os dados obtidos foram avaliados por meio do teste de Fisher ($p < 0,05$) usando o programa Graph Pad Prism 5.0. **Resultados:** Não foram detectadas FRVC. Todas as fraturas observadas antes e após a obturação foram FRVI ou "outros defeitos". No G2 (grupo controle), não houve um aumento no número de FRVI. Curiosamente, G1 apresentou um aumento das FRVI em 70,59% (12/17 raízes). A análise estatística mostrou que a média de FRVI aumentou significativamente no G1 quando comparado ao G2 ($p < 0,05$). **Conclusão:** A aplicação de uma força de 1,5 kg ao espaçador durante a realização da técnica de condensação lateral não produz fraturas radiculares verticais completas, mas pode produzir fraturas incompletas ou "outros defeitos".

PALAVRAS-CHAVE

Endodontia; Fratura radicular vertical; Obturação do canal radicular.

INTRODUCTION

Complete or incomplete vertical root fracture is frequent occurrence in endodontically treated teeth (10.9 to 20%) [1-6]. In the study of Seo et al. [7], out of 107 of fractured teeth, 33 (30.8%) were treated endodontically and 14 (13.1%) were diagnosed with vertical root fracture (VRF). Touré et al. [8] reported the factors to extraction of 119 teeth with a root canal treatment and the VRF was found in 13.4%. These fractures are difficult to diagnose and treat [4,6,9-11]. In addition, they may start at the coronal portion of the tooth, at the root apex, or at any location within these structures [12,13].

Prognosis of VRF is unfavorable, and they often result in bone loss and periodontal defects [2,12,14,15]. Presently, there is still a limited amount of treatment in extraction of root or resection of fractured tooth [3-5,12,16]. VRF generally occurs as a result of trauma, root canal therapy, stress produced during post cementation [17], among other factors [4,16,18-20].

The scientific literature suggests that the most common cause of root fractures is application of excessive loads during lateral compaction of the gutta-percha during canal filling [2,10,19,21]. Previous studies have demonstrated that the main determining factors for the occurrence of root fractures are: the design of the spreaders [22], combined with the stress generated during compaction [18,23], canal width, canal taper, size and other characteristics of the tooth itself [23,24]. Moreover, pre-existing cracks, craze lines or incomplete fractures, especially those in direct contact with the root canal, may propagate and develop into fractures after the endodontic treatment [15].

Holcomb et al. [23] reported that the fracture load found to produce VRF was 1.5 kg. Accordingly, this was the smallest observed fracture load which could serve as a guideline for limiting clinical spreader forces in an effort

to avoid root fractures. However, new studies are necessary to confirm these data.

Therefore, the aim of this ex-vivo study was to evaluate by digital microscopy, the incidence of obturation of root canal employing lateral compaction technique and spreader load of 1.5 kg on the incidence of complete or incomplete vertical root fractures.

MATERIAL AND METHODS

Matrix construction

This study was submitted to and approved by the Ethical Committee of the Araraquara Dental School, UNESP (CEP 55/10). Twenty-seven extracted human mandibular permanent molars were selected and stored in 1% thymol solution. The length of the selected teeth was between 18 and 21 mm. These roots were observed at 8x magnification with a stereomicroscope (Leica Microsystems, Wetzlar, Germany) to exclude those with external cracks. Exclusion criteria were root canals allowing introduction of an instrument exceeding ISO size 15 at working length. After that, the working length was determined as 1 mm short of the length that a size 15 K-file was observed to exit the apical foramen. The teeth had their crowns sectioned and the mesial roots were discarded through a cutting machine Isomet 1000 (Buehler, Lake Bluff, IL, USA). Proximal periapical radiographs of each distal root were taken to verify the presence of a single canal.

Specimens were placed in a small flask containing silicone-based impression material (Zetaplus, Zhermack, Italy) and embedded up to 1 mm from the coronal surface of the sectioned root in order to simulate the periodontal ligament, in accordance with Wilcox et al. [25] and Bhuvra et al. [26], during instrumentation and obturation of the root canal.

Biomechanical preparation

The canal was instrumented to a size 40 K-file (Dentsply-Maillefer, Ballaigues, Switzerland) and step-back preparation

was performed up to a size 55 K-file. Each canal was irrigated with 3 mL of 1% solution of sodium hypochlorite (Ciclo Farma Indústria Química Ltda., EPP Serrana, SP, Brazil) between each file change, using a syringe and a 27G endodontic needle (EndoEze irrigator, Ultradent, USA). After completion of instrumentation, the root canals were washed with 2 mL of 17% EDTA for 3 minutes and subsequently rinsed with 5 mL of distilled water and dried by using size 40 paper points (Tanari Indústria Ltda., São Paulo, SP, Brazil). The prepared roots were distributed into two groups: G1- experimental (n = 17) and G2- control (n = 10).

Root canal obturation

In G1, the root canals were obturated by lateral compaction technique with standardized gutta-percha cone size 40 (Tanari Indústria Ltda., São Paulo, SP, Brazil) and AH Plus endodontic sealer (Dentsply De Trey, Germany) using a size C finger spreader (D1 diameter 0.3 mm, 0.04 taper) (Dentsply-Maillefer, Ballaigues, Switzerland). After that, four M-sized accessory gutta-percha cones (Tanari Indústria Ltda., São Paulo, SP, Brazil) were added in sequence, until the spreader could penetrate no more than 3 mm. The spreader load applied (1.5 kg) was monitored by a Emic DL testing machine (Emic Equipamentos e Sistemas de Ensaio, São José dos Pinhais, PR, Brazil) throughout the compaction procedure, which was carried out by a single trained operator, in accordance with Soros et al. [14]. The accessory cone was compacted by inserting the spreader mounted in the machine at constant speed of 5 mm per min. Obturation was finalized by trimming

the gutta-percha with a heated plugger. No vertical compaction was performed. In control group, root canal filling was performed in the similar way to the G1, but no forces were applied for placement of accessory cones.

Digital microscopy analysis

The coronal surfaces of the specimens were analyzed in digital microscopy images acquired after biomechanical preparation (pre-obturation) and after obturation (post-obturation). The images were captured with an inverted digital microscope (Olympus MIC-D, Philippines) and analyzed by a single blinder trained examiner. Complete vertical root fracture (CVRF) was defined as a fracture which extend continuously from the root canal to the external root surface and incomplete vertical root fracture or “other defects” (IVRF) was defined as a fracture or craze line / cracks observed that did not extend from the root canal to the root surface, according with Onnink et al. [27] and Shemesh et al. [21]. The number of fractures found before and after obturation was computed and the data were evaluated by Fisher’s test ($p < 0.05$) using Graph Pad Prism 5.0 (San Diego, CA, USA).

RESULTS

No roots exhibited a CVRF; all fractures observed before and after obturation were IVRF. In G2 (control group) there was no increase of IVRF number. In G1 there was an increase in the IVRF number to 70.59 % (12 teeth) of the cases (Table 1). The comparison of variation of number of the IVRF between G1 and G2 showed that there was increase statistically significant in G1 ($p < 0.05$) (Table 1).

Table 1 – Comparison of IVRF increased number after obturation

Groups	Total (n)	Roots with no IVRF increased number	Roots with IVRF increased number
G1- Experimental	17	5 (29.41%)	12(70.59%) ^a
G2- Control	10	10 (100%)	0 ^b

G, group; n, number of roots; **IVRF**, incomplete vertical root fracture. Different letters indicate statistically significant difference ($p < 0.05$).

DISCUSSION

This study demonstrated that spreader load of 1.5 kg during lateral compaction technique induces an increase of number of IVRF, compared with the group that was not employed load with the finger spreader. This result is in accordance with previous studies which have suggested lateral compaction technique as a possible causal factor for VRF [12-14,25,28,29].

Wilcox et al. [25] demonstrated that a load of 3.3 kg applied during lateral compaction technique caused VRF in 35.29% of cases. Soros et al. [14] found that spreader load required for IVRF varied from 4.3 to 26.9 kg. In another study, Shemesh et al. [21] evaluated ex vivo the incidence of fractures and defects in root dentine after root canal preparation and filling by lateral compaction technique, using a spreader load of 2 kg. The lateral compaction group had significantly more fractures and defects than all other groups (no preparation, only preparation and no compaction).

Recently, Barreto et al. [15] evaluated ex vivo the effects of root canal preparation and filling techniques on the incidence of dentinal defects and VRFs. In the lateral compaction group, the teeth were filled by using a size C spreader and FM gutta-percha cones controlling the load to a maximum of 2 kg. They found CVRF (13.3%) and other defects (46.6%) in the roots of lateral compaction group in comparison with prepared group (not obturation) that shown other defects in 53.3% and no CVRF.

Previous studies have shown that the mean maximum spreader load applied during lateral compaction ranges from 1 to 3 kg [12,30]. We applied a standardized load of 1.5 kg to the spreaders during root canals obturation, which was in accordance with the study of Holcomb et al. [23] where they suggested that 1.5 kg load to be regarded as a limit in clinical practice. Interestingly, in our study this load led to an increase in the IVRF number.

In the present study we used size C finger spreaders during lateral compaction because

they are compatible with the prepared root canal width. Thinner finger spreaders usually lead to spaces and sealer removal in the apical region without the accessory cone filling that gap in its entirety, leaving voids in the apical third [31].

VRF can be caused by a variety of factors and generally lead to a significant number of endodontic failures. Treatment of VRF is difficult and their prognosis is reserved. These findings are of great importance for the daily clinical practice: endodontists should be aware that application of excessive load during lateral compaction may potentially produce IVRF.

Within the limitations of this study, it was concluded that a load of 1.5 kg during obturation of root canal by lateral compaction technique does not induce the formation of complete vertical root fractures, but might be considered as a potential risk to produce incomplete vertical root fractures or “other defects”.

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**Gisselle Moraima Chávez-Andrade
(Corresponding author)**

Rua Humaitá 1680 - Centro, Araraquara
14801-903 – São Paulo – SP - Brazil.

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CASE REPORT

Multidisciplinary approach for crown-root fracture treatment after trauma: case report

Abordagem multidisciplinar para tratamento de fratura corono-radicular pós-traumatismo: relato de caso

Gisele Cristina Galdeano ANDRIOLO¹, Denise PEDRINI², Elizane Ferreira HAMANAKA¹, Eloá Rodrigues LUVIZUTO², Sônia Regina PANZARINI², Celso Koogi SONODA²

1 - UNESP - Univ Estadual Paulista - School of Dentistry of Araçatuba - Araçatuba - SP - Brazil.

2 - UNESP - Univ Estadual Paulista - School of Dentistry of Araçatuba - Department of Surgery and Integrated Clinic - Araçatuba - SP - Brazil.

ABSTRACT

The development of an accurate diagnosis and appropriate treatment plan can be a complex task, especially in cases of dentoalveolar trauma. The authors present a case report of crown-root fracture caused by trauma and highlight the importance of a multidisciplinary approach for the treatment. An eighteen year-old boy had a bicycle accident resulting in dental trauma. The upper right first molar showed a complicated crown-root fracture and the lower left second pre-molar showed an uncomplicated crown-root fracture. Endodontic treatment, controlled tooth extrusion, periodontal surgery for recovery of biological width, and porcelain crown and onlay restorations were performed. Esthetic and functional results were achieved. At the two-year follow-up it was observed that the tooth/onlay interface of the upper right first molar was stained and the onlay of the left lower second pre-molar was fractured. Therefore, the interface stained was repaired and a porcelain crown was made for the lower second premolar. The clinical case presented herein leads to the conclusion that a multidisciplinary treatment plan is extremely important for a proper resolution in cases of dentoalveolar trauma.

KEYWORDS

Crown-root fracture; Permanent tooth; Tooth injuries.

RESUMO

A elaboração de um diagnóstico preciso e de um plano de tratamento adequado pode constituir uma tarefa bastante complexa, especialmente nos casos de traumatismos dentoalveolares. O relato de caso teve por finalidade demonstrar a importância da abordagem multidisciplinar para o tratamento de fratura corono-radicular pós-traumatismo. Paciente de 18 anos de idade, gênero masculino, sofreu uma queda de bicicleta resultando em traumatismos dentários. O 16 sofreu fratura corono-radicular complicada e o 35 fratura corono-radicular não-complicada. Foi realizado tratamento endodôntico, extrusão dentária controlada, cirurgia periodontal para recuperação do espaço biológico, coroa de porcelana e restauração tipo onlay. O resultado estético e funcional foi alcançado. O paciente foi acompanhado por 2 anos onde foi observado um manchamento na interface dente/onlay do 16 e fratura da onlay do 35. Foi realizado um reparo na interface do 16 e confeccionada uma coroa de porcelana para o 35. Foi possível concluir que a multidisciplinaridade do plano de tratamento é de extrema importância para uma adequada resolução dos casos de traumatismos dentoalveolares.

PALAVRAS-CHAVE

Fratura corono-radicular; Dentes permanentes; Traumatismos dentários.

LITERATURE REVIEW

Crown fractures comprise the most frequent injuries in the permanent dentition (26-76% of dental injuries), while crown-root fractures comprise only 0.3 to 5% [1,2]. Dentoalveolar trauma presents epidemiological expression and becomes a public health problem due to the high incidence of physical violence, bicycle accidents, car accidents, falls, children's games, domestic violence, and the increasing popularity of extreme sports [1,3,4].

Dental traumatic injuries involve function and esthetics damage, ranging from minor loss of enamel to complex fractures involving pulp tissue. Technical knowledge and clinical experience are essential to establish an accurate diagnosis and provide appropriate treatment plan [5]. Most of these types of fracture require a multidisciplinary approach and the treatment of choice will depend on a number of factors such as: length of fracture [2,3,6-8], biological width invasion [2,5,9,10], presence of pulp exposure, stage of root development [2,5-8,11,12], soft tissue injuries [3,7,12,13], presence of concomitant periodontal trauma [3,7,8], presence / absence of fractured fragment and its adaptation, occlusion, esthetics, time, and available resources [2,8-11,14-17].

There are several strategies in the literature for the treatment of esthetic and

functional rehabilitation of crown and crown-root fractures: temporary or permanent use and maintenance of tooth fragment [2,8-11,14,15,17], orthodontic extrusion or periodontal surgery for recovery of biological width followed by direct resin restoration or prosthetic rehabilitation [3,7,10,16,19,20], and extraction followed by dental implant or conventional prosthesis installation [3,7].

Thus, this paper aims to present a clinical case of complicated and uncomplicated crown-root fractures treated with a multidisciplinary approach.

DESCRIPTION OF CASE

An eighteen year-old boy had dental injuries after a bicycle accident and had drawn attention at the Dentoalveolar Trauma Clinic at the Araçatuba Dental School – UNESP, complaining of pain at the upper right first molar. A chin laceration already sutured was observed at the extra-oral examination. Clinical and radiographic examination were observed: enamel crack on tooth 31; enamel fracture on teeth 13, 14, 15, 21, 23, 24, 26 and 36; complicated crown-root fracture (with pulp involvement) on tooth 16 (Figure 1 - A and B) and uncomplicated crown-root fracture (without pulp involvement) on tooth 35 which was not observed on initial radiographs (Figure 2 – A and B).

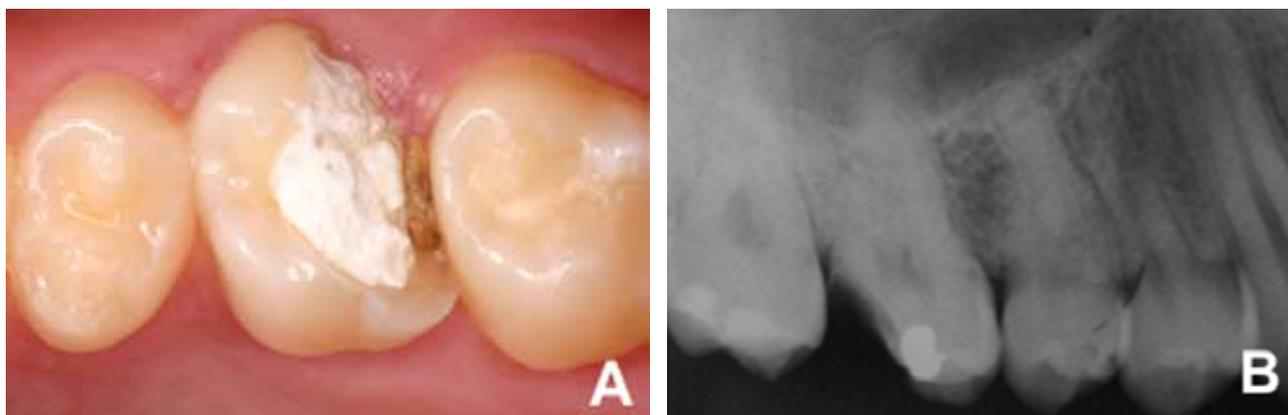


Figure 1 – Initial clinical and radiographic examination of tooth 16. A - Radiographic image of the crown-root fracture. B - Radiographic image.

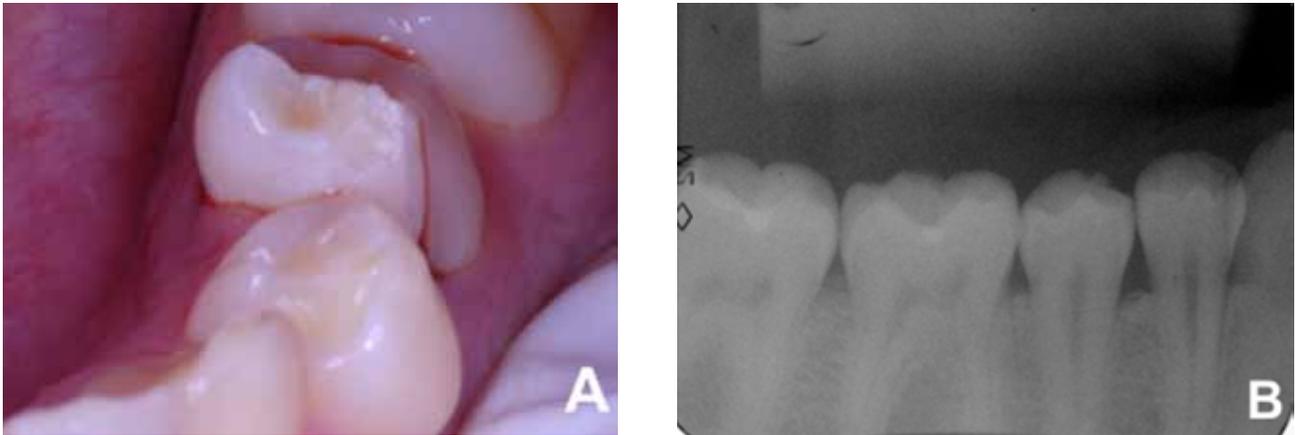


Figure 2 – Initial clinical and radiographic examination of tooth 35. A - Clinical aspect of the fragment slightly shifted. B - Radiographic image.

Endodontic treatment of tooth 16 was initiated and calcium hydroxide root canal dressing was conducted. The fragment of tooth 35 was removed (Figure 3) and a protection with glass ionomer cement (Fuji II LC, GC Corporation, Hasunuma-cho, Itabashi-ku, Tokyo, Japan) of the exposed dentin was performed. Fixed orthodontic appliance with brackets between tooth 35 and tooth 45 and single pipe on tooth 36 and tooth 46 (Figure 4A) was mounted in order to extrude tooth 35 to restore biological width. Occlusal wear of tooth 35 was performed every 21 days to allow space for tooth extrusion (Figure 4B). Total time required was six months: three months of activation and three months of containment.



Figure 3 – Removal of fragment of tooth 35.



Figure 4 – Orthodontic extrusion of tooth 35. A - Orthodontic device with fixed brackets in the lower arch. B - Occlusal crown abraded.

Root canal filling with calcium hydroxide cement (Sealapex, Kerr Corporation, Orange, California, USA) and gutta-percha (Tanari Industrial Ltda, Manaus, Amazonas, Brazil) by lateral condensation technique was held 14 days after the placement of intracanal dressing on tooth 16. The crown was temporarily restored with glass ionomer (Vidrion R, SS White, Rio de Janeiro, RJ, Brazil) and an interdental elastic wedge was placed on the distal surface (Figure 5A), in order to reestablish the interdental contact point. The interdental elastic wedge was removed after 1 week and another temporary restoration was performed to fill the interdental space gained followed by a periodontal surgery to recover biological width (Figure 5B). After 60 days, a ceromer indirect restoration was fabricated (Epricord, Kuraray Medical Inc., Tokyo, Japan).

Endodontic treatment of tooth 35 was held after recovery of the biological width through orthodontic extrusion. Since gingival margin follow tooth extrusion during

orthodontic extrusion, the gingival flap was apically positioned to properly recover the gingiva (Figure 6). Ceromer indirect onlay (Epricord, Kuraray Medical Inc., Tokyo, Japan) was performed after gingival healing.

Clinical and radiographic follow-up of tooth 31 was performed due to the presence of an enamel crack. Tooth 13 and 23 showed small enamel fractures, which had esthetic and function restored by polishing. The remaining teeth with enamel fractures were restored with light-cured composite resin (TPH, Dentsply Ind. e Com. Ltda, Brazil).

Clinical (Figure 7 - A and B) and radiographic (Figure 8 - A and B) evaluations at 2-year follow-up appointment were performed. It was observed that the tooth/onlay interface of the right upper first molar was stained (Figure 7A) and the onlay of the lower left second pre-molar was fractured (Figure 7B). So, the interface stained was repaired (Figure 9A) and a porcelain crown was made for the lower second pre-molar (Figure 9B).

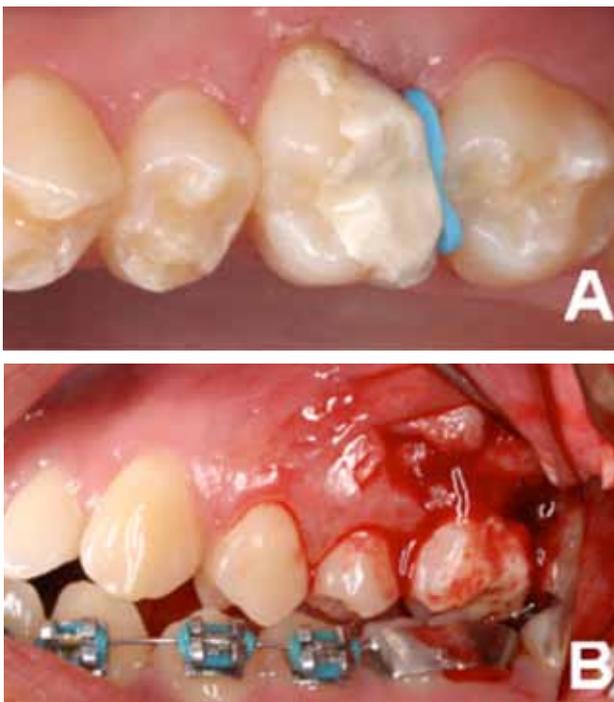


Figure 5 – A – Temporary restoration and interdental elastic wedge on tooth 16. B – Periodontal surgery to recover biological width.

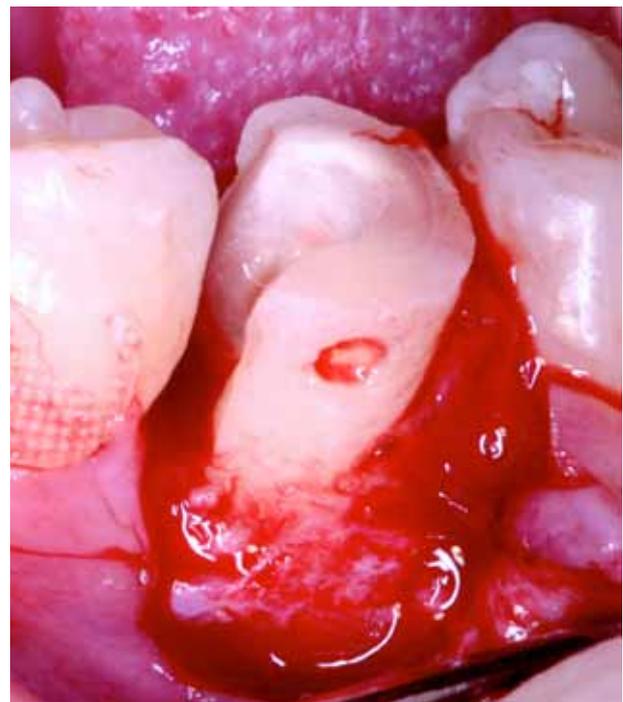


Figure 6 – Apically positioned gingival flap of tooth 35.

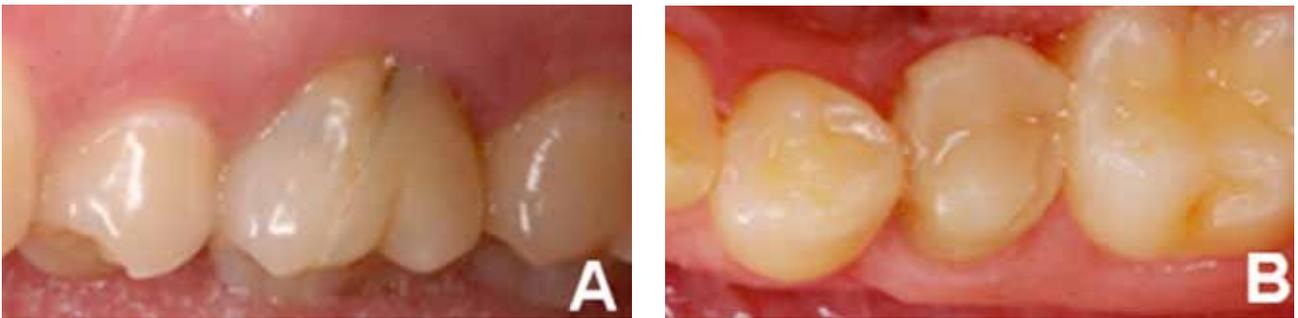


Figure 7– Clinical two years follow-up of tooth 16 (A) and 35 (B).

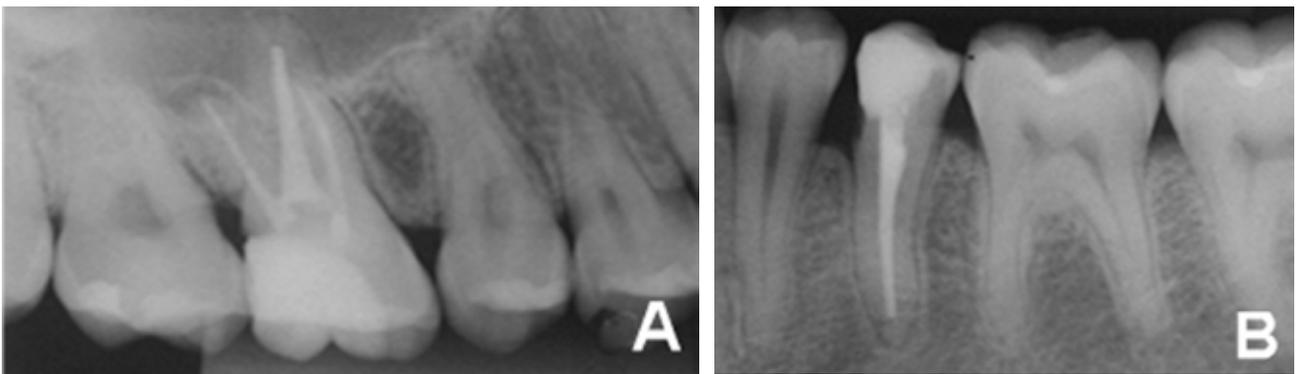


Figure 8– Radiographic two years follow-up of tooth 16 (A) and 35 (B).



Figure 9 – Final clinical aspects of teeth 16 (A) and 35 (B).

DISCUSSION

Teeth and supporting dental tissues can be traumatized in different ways. Although the incidence of dental trauma has reached epidemiological levels in recent years, its treatment still raises questions among professionals. The esthetic and functional recovery is a challenge in most cases and dentists should be well prepared to provide the best possible treatment without additional damage [5].

Fractures that invade the biological width, especially if invasion is located at the buccal and/or proximal regions and extending apically to the bone crest, are the most difficult to solve and, bonding the fragment is rarely a viable treatment [21]. In the present clinical case, tooth extrusion required occlusal wear in order to obtain occlusal space during extrusive movement and therefore, the tooth fragment was unsuitable.

Both teeth 16 and 35 had the invasion of biological width in at least one of their surfaces. Chronic inflammatory response occurs when teeth are restored without reestablishing the biologic width. Crest bone resorption with apical migration of connective tissue and junctional epithelium is also observed histologically. The consequences of the invasion of the biological width include gingival inflammation that persists even in the presence of an adequate plaque control, gingival pain and sensitivity to mechanical stimuli, gingival recession and/or periodontal pocket formation [21]. Moreover, when the fracture margin is exposed, clinical procedures can be performed with strict control of moisture and bleeding, optimizing the predictability of restorative treatment [19].

In such cases, the clinician should consider a few factors for selecting the best possible approach: subgingival lesion extension, lesion morphology, crown/root proportion, tooth position in the arch, prognosis of restorative treatment, esthetic and phonetic considerations, occlusal factors, location of furcation, root morphology and anatomy, endodontic considerations and treatment costs / benefits [19,21].

Orthodontic extrusion was the treatment chosen for the recovery of biological width of the uncomplicated crown-root fracture tooth. This treatment option may be considered more conservative to the esthetic [21] and organic [19] point of view, compared to resective surgery [21]. Before deciding on this treatment, the practitioner should be aware of some factors that affect final result such as bone root insertion and dentin thickness [3,9]. During orthodontic extrusion of tooth 35, the occlusal portion of the crown needed to be abraded resulting to pulp exposure. Therefore, endodontic treatment was performed.

Resective surgery is the most commonly used approach for posterior teeth due to greater bone support, greater root surface area, flat interproximal form, and less esthetic requirement. Another factor to be considered is the prominence of molar roots, and the intercuspatal unfavorable axial position of the posterior teeth, which can affect the extrusion of these teeth. Another problem is that the beginning of the furcation can be at the level of the cemento-enamel junction of the adjacent teeth, and if it is necessary to regularize bone architecture with osteotomy, furcation exposure might occur [21]. As tooth 16 showed biological width invasion only at the distal surface, the surgical procedure was opted.

The extraction procedure is a treatment only performed if all other treatments are not indicated. But, the clinician should be aware of the fact that the future prosthetic rehabilitation is considerably complicated if bone is lost with time [3,9].

Bonding dental fragment is a conservative treatment option when the tooth fragment is available. It reestablishes the form, contour, surface texture, color and alignment of the original tooth, and it is a simple and fast treatment. This treatment option leads to a positive emotional and social response of patients due to the maintenance of the natural tooth structure [11,22]. However, there are limitations as there is the possibility that the fragment may not reacquire the original color of the remnant tooth, there is a risk of

displacement of the fragment with increasing magnitude of the fracture and type of occlusion of the patient.

The need of crown resection in order to gain interocclusal space for tooth extrusion also modifies the original anatomy of the crown, not allowing the reattachment of fractured tooth fragment. Thus, the choice of prosthetic onlay constitutes appropriate indication for preserving the remaining coronary tissue. The prosthetic onlay also provides satisfactory cosmetic results. Masticatory and hygiene habits, which may change over time, might jeopardize the results. In this case presented herein, masticatory forces resulted in the necessity to perform a premolar full crown.

The clinical case reported herein demonstrates the importance of a multidisciplinary treatment plan for a proper resolution of dental trauma.

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Sônia Regina Panzarini (Corresponding address)

UNESP - Univ Estadual Paulista – Campus de Araçatuba
Rua José Bonifácio, 1193 - Araçatuba – SP - Brasil
CEP: 16015-050
E-mail address: panzarini@foa.unesp.br

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CASE REPORT

Effectiveness of low level laser therapy in the treatment of TMD myalgia: two case reports

Eficácia do laser de baixa potência no tratamento da DTM: dois relatos de casos

Natália Cortez GUTIERREZ¹, Heleine Maria hagas RÊGO¹, Maria Ângela Lacerda Rangel ESPER¹, Pedro Henrique CORAZZA², Carolina da Silva Machado MARTINELLI², Fernanda CAMPOS², Carlos Rocha Gomes TORRES¹, Sérgio Eduardo de Paiva GONÇALVES¹

1 - Institute of Science and Technology – UNESP – Univ Estadual Paulista – São José dos Campos (SP) – School of Dentistry – Restorative Dentistry Department – São José dos Campos – SP – Brazil.

2 - Institute of Science and Technology – UNESP – Univ Estadual Paulista – São José dos Campos (SP) – School of Dentistry – Dental Materials and Prosthodontics Department – São José dos Campos – SP – Brazil.

ABSTRACT

Objective: To report two cases of low level laser therapy used in the treatment of muscle pain caused by temporomandibular disorders (TMDs). **Methods:** Two patients were selected and subjected to eight applications of low level laser therapy (diode - wavelength of 795 nm, energy density of 8 J/cm², power of 120 mW, 66 se per point). Laser was punctually applied on masseter and temporalis muscles bilaterally. Three methods were used to evaluate the effectiveness of treatment: pressure algometer, visual analog scale (VAS) and maximal mouth opening. Each measurement was performed before and after the laser therapy session. The results were subjected to statistical analysis (ANOVA two factors and Tukey's test, $\alpha = 0.05$). **Results:** For both patients, no significant difference was found between the results obtained with the algometer, before and after laser application, within each session. VAS results showed a tendency to lower values after laser application. In both cases, the highest values obtained by the pressure algometer were found between the days 9 (fourth application) and 16 (sixth application). Both patients had an improvement on mouth opening. **Conclusions:** The assessment methods used were quite practical to register the pain before and after treatment. Therapy with low level laser seems to have a beneficial effect for the masticatory muscles pain.

KEYWORDS

Maximal mouth opening; Pressure algometer; TMD; VAS.

RESUMO

Objetivo: relatar dois casos do uso do laser de baixo potência no tratamento da dor muscular causada pela disfunção temporomandibular (DTM). **Métodos:** Dois pacientes foram selecionados e submetidos a oito aplicações de laser de baixa potência (diodo - comprimento de onda de 795 nm, densidade de energia de 8 J/cm², potência de 120 mW, 66 segundos por ponto). O laser foi aplicado pontualmente nos músculos masseter e temporal bilateralmente. Três métodos foram utilizados para avaliar a eficácia do tratamento: algômetro de pressão, escala visual analógica (VAS) e abertura máxima da boca. Cada medição foi realizada antes e após a sessão de terapia com laser. Os resultados foram submetidos à análise estatística (ANOVA dois fatores e teste de Tukey, $\alpha = 0,05$). **Resultados:** Em ambos os casos, não foi encontrada diferença significativa entre os resultados obtidos com o algômetro antes e depois da aplicação do laser dentro de cada sessão. Os resultados com o VAS mostraram uma tendência a reduzir os valores após a aplicação do laser. Foram encontrados os maiores valores obtidos pelo algômetro de pressão entre os dias 9 (quarta aplicação) e 16 (sexta aplicativo), em ambos os casos. Ambos os pacientes tiveram uma melhora na abertura da boca. **Conclusões:** Os métodos de avaliação utilizados foram bastante prático para registrar a dor antes e após o tratamento. A terapia com laser de baixa potência parece ter um efeito benéfico para a dor músculos mastigatórios.

PALAVRAS-CHAVE

Abertura da boca máxima; Algômetro de pressão; DTM; VAS.

INTRODUCTION

Temporomandibular disorders (TMD) are characterized by tenderness of facial muscles, headaches, click of the joint and limitation of mandibular movements [1-3]. No factor should be considered individually as a TMD causer because its etiology is multifactorial. However, there are some risk factors: gender (female), age (between 20 and 40 years), genetic, and parafunction (presence of bruxism and clenching) [4-7]. TMD usually affects the masticatory muscles and/or temporomandibular joint, and is often associated with psychological disorders and malocclusion [1,2,8-12]. The American Academy of Orofacial Pain (AAOP) recently divided TMD into two groups: Muscular TMD and Articular TMD [13].

The TMD diagnosis is usually made by anamnesis, physical examination and laboratory tests. After the diagnosis, the professional should control the pain of the patient using medication, counseling (changing habits), heat therapy, transcutaneous electrical nerve stimulation (TENS), muscle exercises, low level laser therapy (LLLT) or combination of different treatments [2,8,14]. The low level laser therapy (LLLT) is a non-invasive treatment that does not cause side effects [2]. A biomodulatory effect occurs on cells and tissues with this treatment, promoting an analgesic effect, anti-inflammatory and muscle relaxant [14-17]. Therefore, it has been used especially in cases of pain of the masticatory muscles, neck and shoulders [18].

However, to prove the effectiveness of this therapy, it is necessary to use pain measurement methods, as visual analog scale (subjective method) [19-25], maximum mouth opening and the pressure algometer applied on specific points of the face called trigger points (objective methods) [19,26-29]. These points are described as sensitive circumscribed nodules that are part of a palpable band of muscle fibers. The pain of myofascial trigger point has been reported as the most prevalent cause of painful symptoms in TMD [30].

The aim of this study is to report two cases of low level laser therapy used on the treatment of muscle pain caused by TMD. Besides the difficulties, consistency and relationship among different methods of measurement will be discussed.

MATERIAL AND METHOD

This study was developed by post-graduate students and professors of the Laser discipline, using the facilities of prosthodontics clinic of Institute of Science and Technology of São José dos Campos – São Paulo State University (UNESP). Two patients already registered on the waiting list for treatment at Occlusion and Temporomandibular Joint Center were selected (patient 1 and patient 2). Criteria for inclusion in the study were: presence of muscle pain in the masseter and temporalis muscles, presence of at least 20 teeth in the mouth and availability to the treatment. Patients were excluded if there were presence of: cancerous lesion, hypo or hyperthyroidism, neurological disorders and pacemaker. Patients agreed to attend all appointments and refrain from using analgesics, anti-inflammatory, anxiolytics, anti-depressants and/or muscle relaxants during treatment period. Patients signed an informed consent form, accepting the terms of the investigation.

Firstly, the patients were submitted to anamnesis through a form composed by patient identification, medical and disease history. The clinical evaluation was performed through inspection, palpation of the muscles involved and functional examination of the temporomandibular joint (TMJ). The treatment was performed twice a week for four weeks. Each patient received a total of eight applications of low level laser. Laser was applied punctually, perpendicular and in contact with the skin surface. The procedure was done bilaterally on potential trigger points located in the masseter and temporalis muscles, as shown in Figure 1, totaling 10 points per patient. These points were selected based on the literature [30] and marked with a ballpoint pen by the same operator. The patients were treated with diode laser (Laser Easy, Clean Line Ind. e Com Prod. Odontológicos Ltda., Taubaté, SP, Brazil) with wavelength of 795 nm, energy density of 8 J/cm², average power density of 63.6 mW/cm², beam area of 5.3 cm² and power level of 120 mW. Continuous emission was done for 66 seconds per point of application [31]. Following the bio-safety rules for laser application, the equipment was protected by a plastic film, and the volunteer and the professional used safety glasses during the phototherapy.

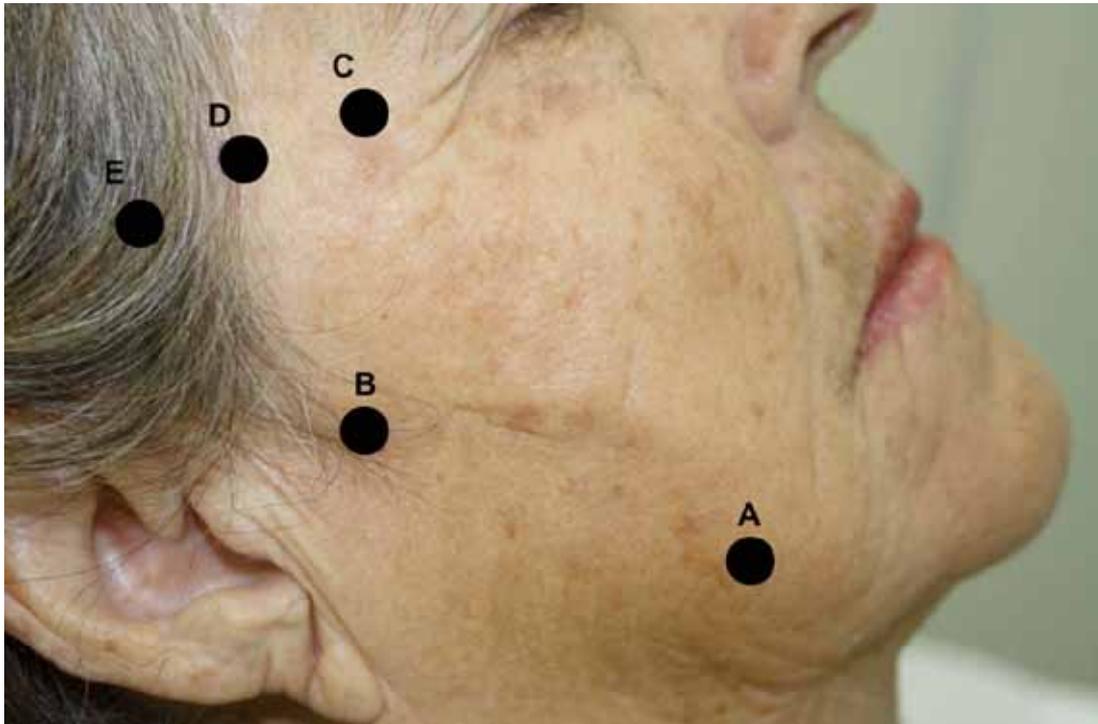


Figure 1 - Schematic trigger points of laser irradiation: (A) lower portion of the surface layer of the masseter muscle; (B) posterior upper layer of the deep portion of masseter, below TMJ; (C) anterior portion of the temporal muscle; (D and E) intermediate portion of the temporal muscle.

Three methods were used to evaluate the effectiveness of the treatment: pressure algometer, visual analog scale (VAS) and maximal mouth opening. Each measurement was performed before and after each laser therapy session.

Pressure Algometer

The pressure algometer (Force Dial Algometer – Wagner Instruments, CT, EUA) is an instrument composed by a pressure analog indicator (in kgf) and a pressure tip applicator,

which contact with the patient's skin (Figure 2). All measurements were performed by the same operator, at the same points where the laser was applied.

To measure the patient sensitivity to pressure pain, arithmetic average of the 10 obtained values (10 points) was calculated for each session, before and after laser therapy, totaling 16 averages for each patient. Data obtained on each day of measurement were statistically compared using 2-way ANOVA and Tukey test ($\alpha = 0.05$).



Figure 2 - Algometer being used for measurement.

Visual Analog Scale (VAS)

The Visual Analog Scale (VAS) consisted of a 100 millimeters long non-graded ruler with descriptors representing the extremities: left “no pain” and right “unbearable pain”. The distance between the point corresponding to the pain intensity, as indicated by the patient, and the left end of the scale determined the VAS value in millimeters. Higher scores indicate higher levels of pain intensity.

Maximal Mouth Opening

Linear measurements of maximum mouth opening were recorded with a digital caliper (727 Starrett, Itu, SP, Brazil). The distance between the incisal edge of the maxillary central incisors and the incisal edge of the mandibular central incisors (mm) was measured, after which the patient was asked to open his mouth as wide as possible, even if it generated pain. The obtained value was added to the vertical overlap length of the patient.

RESULTS

Patient 1

Table 1 presents the mean, standard deviation and homogeneous groups for the algometer measurement results of the patient 1. Figures 3 and 4 show the results of the algometer (load in kgf), VAS and maximal mouth opening measurements respectively.

There was no significant difference between results obtained with pressure algometer before and after low level laser therapy, at each appointment ($p = 0.995$). However, there was a significant difference when comparing the appointment days ($p = 0.000$). The highest values were obtained between the days 9 (forth application) and 16 (sixth application) (Table 1 and Figure 3). The patient reported lower results in VAS on these three days, both before and after laser application (Figure 4). The patient indicated a significant improvement of the pain after the laser application on days 21 and 23 (seventh and eighth applications), which was not detected by algometer (Figures 3 and 4). There was an improvement in maximum mouth opening of Patient 1. The highest value was registered on day 16 (Figure 4).

Table 1 - Means, standard deviations and homogeneous groups for the pressure algometer results of the patient 1

Day	0	2	7	9	14	16	21	23
Mean ± sd (Kg)	1.84 ± 0.81	2.21 ± 0.57	1.58 ± 0.61	2.84 ± 0.64	3.3 ± 0.74	3.07 ± 0.65	1.88 ± 0.52	1.64 ± 0.36
Homogeneous groups	BC	B	C	A	A	A	BC	BC

* Values followed by different letters are statistically different.

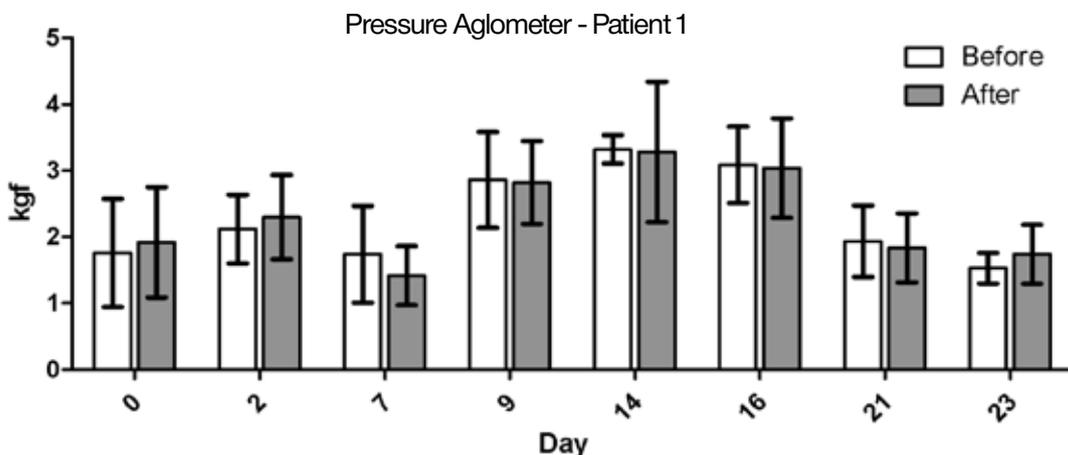


Figure 3 - Pressure algometer results for patient 1.

Patient 2

Table 2 shows the means, standard deviations and homogeneous groups for the pressure algometer results of the patient 2. Figures 5 and 6 illustrate the results of pressure algometer, VAS and maximal mouth opening measurements of the Patient 2 respectively.

There was no significant difference between results obtained with the pressure algometer before and after low level laser therapy, at each appointment ($p = 0.122$). The

figure shows slightly higher results after laser application, with the exception of day 14 (Figure 5), when comparing the appointment days ($p = 0.000$), and the highest values were obtained on day 14 (fifth application). Except for day 9, lesser values of pain (VAS) were reported in all appointments after the laser therapy (figure 6). An improvement in maximum mouth opening was reported for Patient 2, both before and after laser application, with the highest value on the day 16 (Figure 6).

Table 2 - Means, standard deviations and homogeneous groups for the pressure algometer results of the patient 2

Day	0	2	7	9	14	16	21	23
Mean ± sd (Kg)	1.54 ± 0.58	2.6 ± 0.42	1.32 ± 0.32	2.68 ± 0.34	3.22 ± 0.58	2.57 ± 0.33	1.68 ± 0.37	1.60 ± 0.30
Homogeneous groups	C	B	C	B	A	B	C	C

* Values followed by different letters are statistically different.

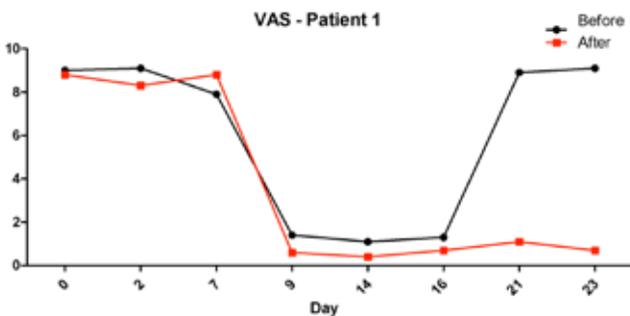


Figure 4 - VAS and maximum mouth opening results for patient 1.

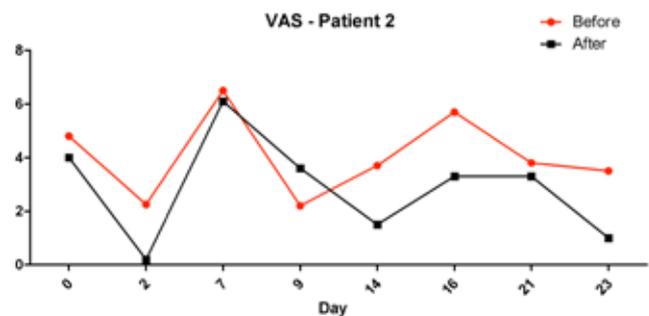


Figure 6 - VAS and maximum mouth opening results for patient 2.

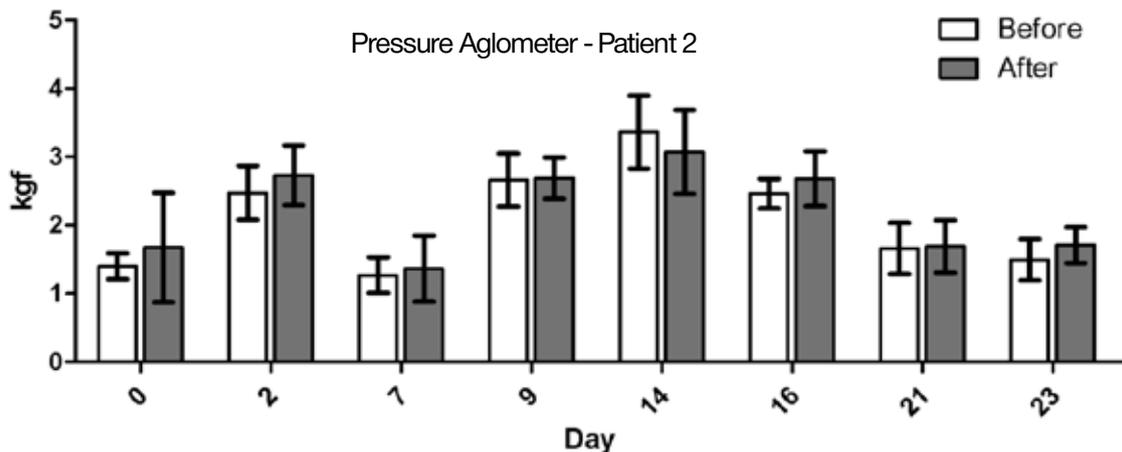


Figure 5 - Pressure algometer results for patient 2.

DISCUSSION

Low level laser application is a complementary treatment to pain caused by TMD due to its analgesic, anti-inflammatory and muscle relaxation properties [14-17]. The results of this study are in agreement with other studies [20,32-35] that also reported the low level laser as an effective therapy in minimizing the pain symptoms triggered by the TMD. Some mechanisms of laser action are cited in the literature, as increase on the endogenous opiates liberation, decrease on the permeability of the nerve cell membrane and increase on the ATP production [14]. The reduction on the levels of creatine kinase activity, a cytokine involved in the early phase of muscle damage, and C-reactive protein, a marker of systemic inflammation, is also reported in the literature [31].

The clinical results showed in VAS figures that pain relief tends to improve after laser application. Although there are some crossings of the lines "before" and "after" in some regions, the line symbolizing the patient after treatment is usually below the one symbolizing the pain before laser application. Although the VAS scale is quite subjective, this method was already validated by the literature. The results obtained by VAS in this study are in agreement with Shinozaki study [32], which verified that laser therapy promoted immediate relaxation of the masseter and temporalis muscles with consequential relief in painful symptoms of TMD.

Moreover, the effectiveness of the treatment is noted comparing the initial (day 0) and final (day 23) pain of patients according to VAS. The patient 1 reported, on the VAS, value 9 before starting the treatment and changed to value 1 at the end. Patient 2, reported value 5 on the VAS before starting treatment and value 1 after the last laser application. These results agree with Dostalová et al. [20], which found that TMD patients undergoing treatment with laser had a reduction in VAS values after five treatment sessions. The visual analog scale is easy to use, in accordance with some studies [26-29]. However, because it reflects a momentary feeling described by the patient, it should be used with caution.

Comparing the maximum mouth opening of the patients at the beginning and the end of the treatment, patient 1 had an increase of 54.25% in his maximum opening, while patient 2 increased 37.87%, which demonstrates again the efficacy of laser therapy, as previously reported [20,32-35]. In this study, patient 1 had an initial maximum mouth opening of 30 mm that turned to 50 mm at the end of the treatment. Patient 2 evolved from 28 mm to 39 mm. These results are in agreement with the findings of Dostalová et al. [20], who reported an improvement in mouth opening from 34 mm to 42 mm.

There was no significant difference in values before and after the laser application for both patients using the pressure algometer. However, some values were slightly higher after laser therapy (Figure 5). The greater results for both patients were reported between 4° and 6° application, and the highest values occurred at the 5° application. This similarity on the results can indicate the reliability of this method, in agreement with previous literature [22-25], which highlights this instrument as a valid way of measurement in patients with varied musculoskeletal pain syndromes, as well as in asymptomatic patients. It is suggested, nevertheless, an adaptation of the instrument for its use in the masticatory muscles, especially when these are symptomatic and recording lighter forces.

Comparisons between different methods of pain measurements should be made with caution, since pressure algometer values are specific and timely and VAS values are more subjective and correlated to the patient's emotional state. So the authors of this study believe that the three methods are complementary and provide important data. Another point worth noting is that the variations in the values obtained for the three assessment methods can be part of the natural evolution of the disease. A controlled clinical study would be required to say with greater certainty that the treatment is really effective.

The authors also believe the use of a placebo group is primordial for studies that want to evaluate the effectiveness of a therapeutic method, despite the difficulties in

approving a project using a placebo in Research Ethics Committees. This practice should be reconsidered by the committee since the motivation for treatment, even if it does not have an active vehicle, can change the posture of the patient towards the problem, generating distortions on the results.

FINAL EVALUATION OF THE CASES

The assessment methods used are very practical, for both scientific and clinical usage, in the registry of pain before and after laser therapy. The analog pressure algometer offers a precise numerical measurement data but causes uncomfortable feeling for the patient. The VAS has the disadvantage of subjectivity. The maximum mouth opening is an objective method, which can reflect the improvement of pain symptoms. Low Level Laser therapy seems to have a beneficial effect for muscle pain of TMJ.

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**Heleine Maria Chagas Rêgo
(Corresponding author)**

Av. Eng. Francisco José Longo, 777
Jd. São Dimas - São José dos Campos
12245-000 – São Paulo – SP - Brazil
Email: heleine.rego@hotmail.com

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