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Impact of Propolis on the Oral Health

SUMMARY

Propolis is a natural resinous substance collected by honey bees from buds and exudates of plant species, mixed with bee enzymes, pollen and wax. It has a complex composition with a wide range of effects, including antibacterial, antiviral, antifungal, antiphlogistic, antioxidant, hepatoprotective, carcinostatic and immunomodulatory properties. It is often applied in the treatment of diseases involving the oral cavity and gums. The aim of this paper is to describe the therapeutic properties of propolis, chemical composition and its application in the oral cavity. Literature and systematic information on the composition and the effects of propolis on health were collected, with particular reference to the use in the treatment of oral cavity diseases. The chemical composition of propolis is very complex. The health impact depends on the biologically active components it contains. A particularly important application is in the treatment of diseases of the oral cavity. Studies show that propolis can help prevent dental caries and control gingivitis and plaque. It reduces halitosis (bad breath) and symptoms of periodontosis. It is also effective in fighting viruses. It can have significant application in orthodontics and restorative dentistry. A wide range of effects allows the multiple uses of propolis-based products. Recent research has been increasingly focused on diseases of the oral cavity. The development of novel propolis-based pharmaceutical forms could significantly reduce the use of antibiotics in conventional treatment of diseases of the oral cavity.

Key words: Propolis, Oral Health, Apitherapy, Oral Cavity

Introduction

Propolis, also referred to as bee glue, is a natural, non-toxic, resinous and sticky substance produced by honey bees through the mixing of hypopharyngeal gland secretions with digestive resin products collected from buds and bark of trees, flowers, leaves and other botanical sources. Propolis most often originates from trees such as poplar, willow, beech and wild chestnut. Bees use propolis to protect and strengthen their hive. With this natural product, the hive is protected from rain and pests such as insects and rodents. Propolis maintains aseptic conditions and the appropriate temperature within the hive. Propolis acts as a biocide that successfully fights bacteria, mushrooms and larvae of certain pests. In order to eliminate the potential infection within the hive, the bodies of dead pests are covered with propolis and thus prevent their decomposition.

Propolis is a lipophilic material that is firm and fragile at lower temperatures, while flexible and sticky at higher temperatures. Its melting point is between 60°C and 70°C, and for some samples the temperature can go up to 100°C. The best solvent used for the preparation of propolis is ethanol. Other solvents such as ethyl ether, water, methanol and chloroform can be used for the extraction and identification of propolis components. Gloverin and propylene glycol are used in the preparation of propolis for the pharmaceutical and cosmetic industries. It has a pleasant aromatic scent and can be, depending on origin and botanical origin, red, brown, yellow or green in colour.
Chemical composition of propolis

The chemical composition and content of the biologically active compounds of propolis depends on the geographical and botanical origin, the type of bees, and the seasons in which propolis is collected. Raw propolis consists of about 50% resin, 30% wax, 10% essential oils, 5% pollen and 5% of various organic compounds. Techniques for separating and purifying the mixture, such as high pressure liquid chromatography - HPLC, thin layer chromatography - TLC, gas chromatography-GC, as well as identification techniques such as mass spectrometry - MS, nuclear magnetic resonance-NMR, gas chromatography in combination with mass spectrometry- GC-MS, has identified several compounds within propolis including flavonoids, terpenes, phenols and their esters, sugars, hydrocarbons and mineral elements. In contrast, relatively frequent phytochemicals such as alkaloids and iridoids have not been detected.

According to research, Propolis is much more effective in combating Gram-positive bacteria than against Gram-negative bacteria. This is due to the fact that the membrane of Gram-negative bacteria exhibits greater complexity in the material than the Gram-positive bacteria. In the suspension of bacteria, certain components of propolis can be destroyed by hydrolytic enzyme of the bacteria. Some of the mechanism of action by which propolis exhibits its antimicrobial effect, is through the suppression of permeability of the bacterial membrane and inhibition of bacterial mobility. Propolis can affect the flow of ions through the inner bacterial membrane and lead to disturbance of the membrane potential, that affect the electrochemical gradient that is necessary for the production of adenosine triphosphate (ATP) required for the smooth flow of the membrane transport and for maintaining its mobility. The polypeptide directly affects the organisms in vitro. On the other hand, propolis can indirectly participate in the destruction of microorganisms by stimulating the in vivo immune system and activating the mechanisms responsible for killing microorganisms. The polypis can be combined with antimicrobial drugs because it has been proven that propolis reduces bacterial wall resistance on antibiotics and has a synergistic effect with antibiotics that work on ribosome’s, but does not show interaction with antibiotics that affect DNA or folic acid.

Table 1. Biological activity of different propolis components

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<tr>
<th>Component, propolis type</th>
<th>Biological activity</th>
<th>References</th>
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|                          |                                                                                   | Havsteen, 2002
|                          |                                                                                   | Ghisalberti, 1979 |
| Caffe-phenethyl ester acids (Topola, Baccharis) | Antioxidant, antitumor, antiinflammatory, antibacterial, antiviral, fungicidal, cardioprotective immunomodulatory. | Bankova, 2009
|                          |                                                                                   | Bankova et al., 2007 |
| Artepelin C (Baccharis) | Antioxidant, antiinflammatory, antitumor                                                                 | Bankova, 2009
|                          |                                                                                   | Bankova et al., 2007 |
| Terpene (terpens) (Greece, Croatia, Brazil) | Antibacterial, antimicrobial                                                                 | Bankova, 2009 |
| Essential oils (Brazil, Poland) | Antibacterial                                                                                                | Bankova et al., 1995

The action of propolis on human health

Propolis exhibits a wide range of biological properties, and one of the most significant characteristics is its antimicrobial activity. This is supported by a large number of publications. Table 1 below describes the biological activity of propolis components.

Research has shown that propolis extracts, which are composed of many polyphenols, have a strong antioxidant activity. Geographically and botanically different propolis samples have a different chemical composition, which directly affects their effect as antioxidants. Fabris et al. (2013) showed that ethanolic extracts of Russian and Italian propolis have a similar antioxidant effect because they have a similar polyphenol composition, while, on the other hand, the ethanolic extract of Brazilian propolis exhibits significantly less antioxidant effect because it contains a smaller amount of polyphenols in its composition. Phenolic acids and flavonoids exhibit a strong antioxidant effect, which is closely related to the chemical structure of the components.

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Propolis has also been shown to have a significant antiviral effect. It acts at different levels and impedes the replication of certain viruses such as herpes simplex type 1 and 2, adenovirus type 2, influenza virus, human immunodeficiency virus (HIV), and others. By research, propolis can exhibit antiviral activity by causing partial blocking of viral penetration into the cell, affecting the steps in the viral replication cycle, and leading to degradation of the RNA virus before penetration in a cell or after its release into the supernatant\textsuperscript{42}. Certain clinical trials in the male and female populations have shown that application of fat containing propolis can result in faster healing in genital herpes compared to conventional acyclovir treatment\textsuperscript{42}.

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<th>Table 2. Pathogenic bacteria, fungi, viruses and parasites on which propolis acts\textsuperscript{19}</th>
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<td><strong>Gram-positive bacteria</strong></td>
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<tr>
<td>Bacillus cereus, Bacillus mesentericus, Corynebacterium sp.,</td>
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<tr>
<td>Corynebacterium diphtheriae, Diplococcus pneumoniae, Enterococcus sp., Mycobacteria sp., Mycobacterium tuberculosis, Staphylococcus aureus,</td>
</tr>
<tr>
<td>Streptococcus: cricetus, epidermis faecalis, mutans, pyogenes, viridans, sobrinus</td>
</tr>
<tr>
<td><strong>Fungus</strong></td>
</tr>
<tr>
<td>Aspergillus sp., Candida: albicans, guiliermondi, parapsilosis, tropicalis; Cryptococcus sp., Cryptococcus neoformans; Histoplasma capsulatum; Madurella mycetomi; Microsporum: audoiini, canis, cepleo, distortum, fersregenium, gypseum; Piedra hortae, Phiaphthora jeanesmei, Saccharomyces sp., Trichophyton: sp., Mentagrophytes, rubrum, Trichosporon cutaneum</td>
</tr>
<tr>
<td><strong>Parasites</strong></td>
</tr>
<tr>
<td>Giardia lambia, Giardia duodenalis, Trichomonas vaginalis, Trypanosoma cruzi, Trypanosoma evansi</td>
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Propolis has been shown to exhibit antifungal action against \textit{C. albicans}, \textit{C. tropicalis} and \textit{C. krusei}\textsuperscript{45}. Propolis acts on the aflatoxigenic types of \textit{Aspergillus} such as \textit{Aspergillus flavus} by inhibiting the condom germination\textsuperscript{48}. Propolis also acts on numerous parasites (Table 2).

Antiparasitic and immunomodulatory activity of Brazilian propolis on \textit{Leishmania braziliensis} was investigated\textsuperscript{49}. Propolis directly affected the parasite and exhibited immunomodulatory effects on murine macrophages, although it has been shown that the parasite continues to affect the activation pathways of the cell. Components important for the antiparasitic action are phenolic compounds (flavonoids, aromatic acids, benzopyrene), di- and triterpenes; and essential oils found in the propolis sample\textsuperscript{49}.

Various in-vitro studies have demonstrated the cytotoxic effect of propolis on tumor cells. In-vivo studies have also shown that there is potential for the development of new antitumor drugs; showing no adverse effects when tested on rats\textsuperscript{50}. This natural product is able to block oncogenic signaling pathways, which in turn leads to decreased proliferation and cell growth. It also reduces the population of tumor stem cells by increasing apoptosis, preventing angiogenesis and modulating tumor micro-circulation\textsuperscript{51,52,53}. Caffe-phenethylether acids and artepealin C are distinguished as components possessing antitumor properties. Other distinguished components include: chrysine, nemesone, galangin and cardano\textsuperscript{51}.

Propolis modifies non-specific immunity. Propolis and its constituents, such as cinnamic acid and its p-coumarinic derivative, stimulate the production of TNF-α (tumor-alpha necrosis factor) and interleukin (IL)-1β in mouse macrophages\textsuperscript{54,55}. The expression of cellular receptors such as toll-like receptors TLR-2 and TLR-4 was also increased in peritoneal macrophages of propolis-treated mice\textsuperscript{56}. A study on the effect on reactive oxygen species also showed that propolis stimulates the formation of hydrogen peroxide (H$_2$O$_2$) through mouse macrophages while reducing the production of nitric oxide (NO)\textsuperscript{38}. In a second study however, the inhibitory effect on the production of superoxide anions by rabbit neutrophils was demonstrated by the propolis compounds\textsuperscript{57}. In humans, propolis can exhibit immunomodulatory effects on cellular receptors, as well as in the production of cytokines and the fungicidal activity of monocytes, depending on the concentration. It increases the expression of TLR-4 and CD80 receptors, influences the production of TNF-α and IL-10 and increases the fungicidal activity of monocytes\textsuperscript{58}. Cinnamic acid reduces the number and activity of TLR-2, HLA-DR and CD80 receptors, and increases the activity of TLR-4 receptors. High concentrations of cinnamic acid inhibit the production of TNF-α and IL-10, while the same concentrations encourage stronger fungicidal activity...
against C. albicans. Propolis stimulates the production of antibodies; independent of the year of propolis collection and its origin. This was confirmed by a 2005 study in which scientists used Brazilian and Bulgarian propolis as an auxiliary in rats immunized with bovine serum albumin.

Propolis can exhibit pro-inflammatory and anti-inflammatory effects depending on the concentration, enter period and experimental conditions and can stimulate or inhibit certain processes. However there is still little evidence of the clinical efficacy of propolis in this manner.

Propolis also has a beneficial effect in the treatment of wounds due to its antifungal and antibacterial abilities in view of the presence of certain components such as: flavonoids, phenolic compounds, terpenes and enzymes. It reduces the amount of free radicals (reactive oxygen species) and thus facilitates the wound healing process. It participates in collagen metabolism by increasing the synthesis of collagen type I and type III collagen in tissues. Propolis is a potential apitherapy agent that has the ability to modify the metabolism of fibronectin. It develops a fibrous network of extracellular matrix and inhibits the fibroblast disintegration. Components such as quercetin and resveratrol inhibit the fibronectin biosynthesis and TGF-β (transforming growth factor β) dependent production of fibroblast in C2C12 myoblasts. Both components play an important role in the expression of fibronectin. Studies have shown that the mobility of epithelial cells depends on the reduced content of fibroblast in the extracellular matrix. Reduced amounts of this glycoprotein allow propolis to better cure wounds and produce granulation tissue.

The oral cavity

The oral cavity represents the proximal part of the digestive system and plays a role in chewing and ingestion of food, as well as speech. The major components of the oral cavity are the teeth, tongue and the salivary glands. The mucous membrane of oral cavity has the role of protecting organs by allowing absorption and resorption of the substances, preventing the non-physiological change of substances and stimulating the secretion of harmful substances from the organism.

The most common diseases of the oral cavity include:

1. Dental caries (tooth decay)
2. Gingivitis – inflammation of the gums
3. Periodontitis - inflammation of periodontium (tissue that supports the teeth)
4. Other diseases of the oral mucosa: angular cheilitis, oral herpes, oral candidiasis, exfoliative glossitis, prosthetic stomatitis (denture stomatitis), aphthous ulcer, and others.

Propolis and oral cavity health

Early animal studies have shown that propolis significantly reduces dental caries in rats as a result of multiple effects on the bacterial flora. It limits the number of microorganisms, slows down the synthesis of insoluble glucans, and slows down the activity of glucosyltransferase enzyme. This natural product has oral cavity activity due to its high fatty acid content such as oleic, linoleic, palmitic and stearic acids which slows the production of acid by Streptococcus mutans and reduces the tolerance of microorganisms to the acidic pH. Propolis also have a lower cytotoxic effect on fibroblasts (found in gums) compared to chlorhexidine (also used in caries prevention), suggesting that propolis can be used as an ingredient in mouthwash.

Majority of studies uses propolis as a mouthwash in the form of aqueous and alcoholic solution or in the form of toothpaste. Propolis can be used in the form of a solution for the decontamination of fibers on the toothbrush.

Bacteria such as Tannerella forsythiensis, Porphyromonas gingivalis and Treponema denticol make up a red complex of microorganisms that increases the depth of the periodontal pocket and causes bleeding of the gums. The propolis extract shows high efficacy in preventing the growth of bacteria belonging to the red complex. Clinical examination has shown that the 3% ethanolic propolis extract in the form of gel and paste slows down and ultimately prevents pathological changes in patients at an increased risk of occurrence of dental plaque gingivitis. Based on a clinical trial of 25 patients, a non-alcoholic mouthwash containing 5% Brazilian green propolis proved effective in controlling plaque and gingivitis, suggesting its use in treatment and prevention periodontal diseases. However, a double-blinded randomized trial is needed before final clinical use in the dentistry industry. The preventative effect of propolis on periodontal tissues implies a slowing down of the calcium phosphate precipitate formation process and can therefore be used as ingredient in mouthwash and toothpaste, hence limiting the accumulation of dental plaque.

Halitosis is a frequent or permanent existence of bad breath from the mouth, and is closely related to the hygiene of the oral cavity. By-products from the degradation of microorganisms in the oral cavity are one of the main causes of halitosis. The microbes most commonly responsible for the onset of aching and halitosis include Prevotella intermedia, Porphyromonas endodontalis and from the bacteria of the genus Eubacterium. By measuring the content of volatile sulfur compounds in the exhaled air through the halometer, it was concluded that propolis significantly reduces halitosis.

Propolis has been shown to affect certain etiological factors that lead to the development of periodontal
disease. Therefore, some researchers use it as part of their therapeutic protocol in the treatment of periodontitis\textsuperscript{77}. A micro-adhesive hydrophilic gel containing propolis, when applied to gingival pockets was also shown to be efficacious\textsuperscript{78}. Based on clinical and microbiological parameters, the subgingival flushing with propolis extract as an auxiliary agent in the treatment of periodontitis proved more effective than the conventional method of scraping and pollinating roots\textsuperscript{1}.

Studies have also been carried out on animals where propolis was given per os to determine if its systemic effect through circulation has positive action on oral cavity health. Morphological and histological pictures showed that oral propolis administration in rats prevents the loss of alveolar bone from periodontitis\textsuperscript{79}.

Herpes simplex type 1 is a virus that causes skin and mucous lesions on the membranes of the mouth. It is one of the most common human pathogens\textsuperscript{80}. Propolis is used locally in the treatment of oral cavity lesions caused by viruses, in studying its antiviral capabilities. As a propolis mixture, it is more effective in combating viral diseases compared to its individual components separate\textsuperscript{81}. Propolis slows down changes in skin and virus growth in the early stage of infection with Herpes simplex type 1 and is not cytotoxic on healthy cells\textsuperscript{82}.

Bee glue also is used in the treatment of recurrent aphthous stomatitis (canker sores). Although aphthous stomatitis is a relatively common disease whose symptoms are visible in the form of aphthous ulcers in the mouth; the etiology of the disease has not yet been established which significantly complicates the treatment\textsuperscript{83}. Propolis reduces the frequency of the disease and improves the quality of life in patients\textsuperscript{84}.

Dental avulsion is the traumatic displacement of a tooth from its socket in the alveolar bone. In oral surgery, propolis is used in the replantation of a broken permanent tooth and provides support in the healing process after surgery. Maintenance of periodontal cells is one of the key factors to determine success of tooth replantation. A research was carried out to determine the medium that provides the best protection during tooth replantation. Propolis as a transport medium showed positive results\textsuperscript{77}. According to the research by Ozan et al., a 10% propolis solution showed better results compared to a 20% propolis solution, Hank’s balanced saline solution (HBSS) or milk. A new study also showed the extraordinary effectiveness of propolis in not only reducing apoptosis of periodontal cells, but also increases metabolism and cell proliferation\textsuperscript{86}. Margo-Filho and Carvalho\textsuperscript{87} have proven that locally applied propolis helps to heal wounds after surgery in the oral cavity, reduces inflammation and also acts as an analgesic. Propolis accelerates the epithelization and formation of granulation tissue in the area of healing\textsuperscript{88}.

In the case of mal-occlusion followed by narrowing of the upper jaw, it is necessary to use an orthodontic device for the expansion of the palatinal suture. During treatment, bone remodeling occurs in the area of palatinal suturing\textsuperscript{77}. Research on rats showed that propolis solution helps in bone formation during treatment with orthodontic appliances that lead to the spread of palatinal suture. The results of this study showed an increased amount of osteoblastic activity in rats who received propolis in treatment and faster bone remodeling\textsuperscript{89}.

In restorative dentistry, propolis is used to reduce the permeability of dentine and in direct overlapping of the pulp to form reparatory dentin\textsuperscript{77}. Ahangari et al.\textsuperscript{90} have proven that propolis acts more efficiently in direct overlapping of the pulp compared to products of calcium hydroxide most commonly used for this purpose. It stops the inflammatory reaction, the infection of the microbes and the necrosis of the pulp and encourages the formation of high-quality tubular dentin by stem cell stimulation. The stimulatory effect on tooth pulp is conditioned by the presence of flavonoids in propolis extracts\textsuperscript{91}.

One of the aims of endodontic treatment is the elimination of microorganisms in the root canals of teeth\textsuperscript{92}. The efficacy of drugs is reflected in the Enterococcus faecalis test that is resistant to adverse conditions and can survive in the root canal system despite the use of certain medicines\textsuperscript{93}. The study has shown that it significantly reduces the number of cultured bacteria Enterococcus faecalis, but that it is not superior to chlorhexidine\textsuperscript{84}. However, due to the low level of periapical tissue and protective effect on periodontal cells, propolis can be used in the disinfection of the root canals of the teeth\textsuperscript{95}.

Prosthetic stomatitis is a common disease in people using dental prostheses. The etiological factors of the disease are: infection with Candida albicans, improper hygiene of the oral cavity and excessive use of prosthesis\textsuperscript{96}. Products based on propolis show strong anti-fungal effects on various types of Candida, and the most sensitive to propolis is Candida albicans\textsuperscript{87}. The most commonly used form of propolis for prosthetic stomatitis is as a mouthwash\textsuperscript{87} or as a gel for local application. Acrylic resin is one of the materials used to make dental prostheses. Da Silva et al.\textsuperscript{98} showed that propolis in the form of a gel can adversely affect the acrylic resin in a way that makes it rough and more prone to adherence to microorganisms.

**Potential adverse reactions of propolis**

Apart from being a resinous substance with multiple usages (as described above), propolis is also a known sensitizer; as highlighted by Menniti-Ippolito et al.\textsuperscript{100} in their report. There were 18 suspected adverse reactions involving propolis-based products that were reported between April 2002 and August 2007 to the Italian National Surveillance System.
It is advised to not be used by patients with predisposition to allergies, especially towards pollen and honey, as well as by individuals with atopy or asthma. The study concluded that healthcare practitioners and the general public must be made aware of the potential risk of allergic reactions of consuming products derived from bees, and that a warning label should be visible on product packaging.

In addition, a case report by Hay and Greig also supported the antigenic property of propolis and further suggests to consider delayed contact sensitivity reactions by propolis as a differential of oral mucosal lesions. Apart from the case report by Hay and Greig, another case report by Budimir et al. also noted adverse effects by individuals using self-prescribed propolis products. These adverse effects include oral mucositis, contact cheilitis and perioral dermatitis. However these case reports highlight the self-treatment with propolis-based products rather than through prescription.

It is concluded that despite several case reports stating the adverse reactions experienced by self-prescribing individuals, proper patient education, prescription and treatment monitoring of propolis-based products has a bigger benefit potential.

**Conclusions**

With the development of modern methods of analysis, new knowledge about propolis activity on human health has emerged. Its mechanisms of action are still being investigated, which will likely lead to the development of new products that affect the health of the oral cavity. Coupled with patient education, proper prescription and treatment monitoring, the benefits of propolis-based products, such as, antibacterial, antiviral, antifungal, anti-inflammatory, antioxidant and chemopreventive actions can be utilised. This may significantly reduce the use of conventional treatments and antibiotics, shifting towards the usage of propolis in the management of oral cavity conditions.

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How to Maintain Oral Health in Children with Respiratory Diseases – Literature Review

SUMMARY
The most frequent chronic respiratory problems in childhood are asthma and cystic fibrosis (CF). The purpose of this paper is to review basic knowledge and recent advances in oral health and associated dental morbidities in children with asthma and CF. This review considered clinical trials and systematic reviews related to oral health in children with CRD. An online base Medline was searched to determine relevant papers, using the combination of the following terms: “asthma”, “cystic fibrosis”, “caries”, “dental erosion”, and “oral health”. Oral health problems in children with chronic respiratory diseases (CRD) may be influenced by natural course of the disease, pharmacotherapy (inhalation therapy with bronchodilators and inhaled corticosteroids in asthmatic patients, systemic antibiotics and pancreatic enzyme replacement therapy in CF patients), medication administration technique and nutritional habits. Children with CRD may have higher prevalence of oral diseases. Patients and their parents, but also general paediatricians and pulmonologists, should be aware of importance of good oral health. Dental practitioners should be more informed about risk factors and specificities of oral health in these patients. Preventive measures, early diagnosis and effective treatment strategies in children with CRD can reduce occurrence of oral diseases and improve patient’s quality of life.

Key words: Asthma, Cystic Fibrosis, Oral Health, Dental Caries, Periodontal Disease, Dental Erosion

Introduction
Acute and chronic respiratory diseases (CRD) are the most frequent medical problems in childhood. Prevalence of CRD is increasing worldwide, why dental practitioners should be more aware of the specificities of oral health in these patients. Specificities of particular clinical problems in patients with CRD come from natural course of the disease, pharmacotherapy and nutritional risk factors that have not favourable impact on the appearance of oral diseases. Risk factors associated with oral diseases in children are presented in Table 1.

The purpose of this paper is to review basic knowledge and recent advances in oral health and associated dental morbidities in two most common chronic respiratory diseases in childhood - asthma and cystic fibrosis (CF).

Material and Methods
This review considered clinical trials and systematic reviews related to oral health in children with CRD. An
online database Medline was searched to determine relevant papers, using the combination of the following terms: “asthma”, “cystic fibrosis”, “caries”, “dental erosion”, and “oral health”. The last search date was March 31st 2017.

**Asthma**

Asthma is a chronic inflammatory disease, characterised by episodes of cough, wheezing, chest tightness and difficult breathing. Its prevalence in childhood is increasing worldwide, causing hospital admissions, school and work absenteeism, decreased quality of life and even asthma-related death. Latest Global initiative for asthma (GINA) estimated that the number of people with asthma in the world may be as high as 334 million. Although prevalence of asthma varies widely between the countries, it is estimated that about 14% of the world’s children were likely to have had asthmatic symptoms1. Besides GINA, numerous national asthma-reduction plans established useful diagnostic criteria and different treatment modalities which resulted in proper diagnosis and decrease of asthma exacerbations.

The cornerstones of modern therapy are inhaled bronchodilators (mostly β2 agonists) and anti-inflammatory drugs (inhaled corticosteroids (ICS)). Systemic adverse effects such as decreased growth velocity, adrenal suppression or osteoporosis are not related with chronic use of low and moderate doses of ICS. Local adverse effects- mostly dysphonia and oral diseases are associated with frequent use of inhaled bronchodilators or chronic use of ICS. Proper techniques of inhalation, use of spacers with metered-dose inhalers (MDI) and good oral hygiene may decrease the incidence of these complications. Most of the children with asthma have a mild disease treated intermittently with inhaled β2 agonists. Excessive and/or prolonged use of either nebulised or β2 agonists from MDI can lead to a reduced salivary flow which is essential for oral health. Alteration of protective role of saliva leads to increased number of cariogenic microorganisms2-4. A dry powder inhaler (DPI) is a device that delivers asthma medication in size of respirable particle (from 1-5μm in diameter) in mixture with excipients which carries the active drug. Most commonly used carrier is carbohydrate - lactose monohydrate. During inhalation from DPI, drug particles separate from the carrier and carried into small airways in the lungs. Larger, lactose particles, are deposited on the oropharyngeal mucosa which can contribute to elevated caries risk3.

It has been suggested that asthmatic patients may have a higher risk for oral diseases, either as a result of the medical condition or as adverse effects of medication. A higher prevalence of caries in school children5 and adolescents7-9 with asthma compared with healthy children has been reported. Ersin et al.5 demonstrated that the duration of therapy and seriousness of the disease had significant influence on the caries risk in asthmatics.

It was speculated that mouth breathing and higher intake of sweet drinks, which are commonly used in an attempt to eliminate the bad taste of the inhaled medication or to reduce the desiccating effect of mouth breathing and reduced salivary flow, may be related to worse oral health in primary dentition17-19. On the other hand, several studies with comparable methodology showed no relationship between asthma and caries incidence, regardless the age of the participants10-16.

It has been reported that asthmatics are in a higher risk to develop dental erosions17,18. This was explained by negative influence of frequent consumption of acidic soft drinks, acidity of some nebulised solutions (pH<5.5), and possible presence of gastroesophageal reflux disease, which show the erosive effect. On the other hand, Dugmore and Rock19 showed no differences in prevalence of dental erosions between asthmatic and healthy children.

Respiratory disorders may also be associated with enamel developmental defects. Guergolette et al.20 reported higher prevalence of enamel defects in asthmatic than in healthy children with demarcated diffused opacities being the most prevalent. Since ameloblasts are highly sensitive to the lack of oxygen, authors assumed that enamel defects are attributable to the episodes of hypoxemia during amelogenesis.

Chronic therapy with ICS, oral dryness due to mouth breathing, and proinflammatory cytokine release in persistent asthma have been shown to lead to greater incidence of gingivitis in asthmatic patients9,21,22. Children with allergic asthma phenotype frequently have associated allergic rhinitis, manifested with various degree of nasal obstruction. Partial nasal obstruction and reduced nasal clearance may cause pronounced mouth breathing with reduced salivary flow. This leads to subsequent bigger accumulation of dental plaque23. McDeera et al.22 showed raised prevalence of dental calculus in asthmatic children which can be contributed to the increased concentrations of salivary calcium and phosphate ions24.

Asthmatic children, especially those with associated allergic rhinitis, may have facial dysmorphism with increased facial height, higher palatal vaults, overjets and posterior crossbites25,26. It is probably caused by difficulty breathing, preferable mouth breathing due to nasal obstruction and diminished respiratory reserve in those patients with severe airway obstruction.

From the above, it is clear that attitudes about the oral health of children with asthma are not consistent. Some epidemiological and clinical studies showed no relationship between asthma and oral diseases, while other studies demonstrated an increased risk for oral diseases in asthmatic patients. However, authors suggesting increased incidence of caries, gingivitis, candidiasis, tooth erosion, changes in the salivary flow and composition, etc. emphasized difficulties in finding the exact reason for higher prevalence of oral diseases in children with asthma27-29. One of the possible
explanations for the differences between the studies may be the nonhomogeneity of the samples, i.e. severity of the disease and specificity of the inhalation drugs used by the subjects in diverse studies.

Cystic Fibrosis

CF is the most common autosomal-recessive disease in Caucasians, with an incidence of 1:1,700-1:40,000 newborns. It is caused by a mutation in a gene located in the long arm of chromosome 7, coding for the complex protein called cystic fibrosis transmembrane regulator (CFTR). The main function of CFTR is transepithelial chloride transport. In case of nonfunctional ion membrane transport, secretions in the exocrine glands (including salivary glands) become thick and dehydrated. This results in systemic illness with dominant obstructive, suppurative lung disease that leads to diffuse bronchiectasis and chronic respiratory insufficiency as a main cause of death. In most of the cases, CF patients also have maldigestion due to exocrine pancreatic insufficiency that leads to malnutrition which correlates to unfavourable outcome. In the last decades, there have been numerous improvements in early diagnosis and different treatment modalities, including lung transplant, with increased life expectancy and quality of life in patients with CF.

Therapy consists of high-calorie diet, pancreatic enzyme replacement therapy (PERT), fat soluble vitamins supplementation and medications that lead to increased mucus clearance (bronchodilators, mucolytics). In addition, patients are treated with inhaled and systemic antibiotic therapy in order to control chronic bacterial colonization in lower airways or treat exacerbation of suppurative lung disease.

Children and adolescents with CF are thought to be at an increased risk for oral diseases. This was thought to be related to the high calorie diet with frequent in-between sugar-rich meals, which may contribute to the high caries risk. In order to preserve lung function and increase mucus transport, secretions in the exocrine glands (including salivary glands) become thick and dehydrated. This results in systemic illness with dominant obstructive, suppurative lung disease that leads to diffuse bronchiectasis and chronic respiratory insufficiency as a main cause of death. In most of the cases, CF patients also have maldigestion due to exocrine pancreatic insufficiency that leads to malnutrition which correlates to unfavourable outcome. In the last decades, there have been numerous improvements in early diagnosis and different treatment modalities, including lung transplant, with increased life expectancy and quality of life in patients with CF.

Conclusions

Children with CRD may have higher prevalence of oral diseases influenced by numerous contributing factors in compare to healthy peers. Dental practitioners should be more informed about risk factors and specificities of initial bacterial infection or chronic colonization of lower airways in infancy and childhood (mostly Staphylococcus aureus), consists of chronic use of β-lactam antibiotics. It may also reduce cariogenic flora, i.e. Streptococcus mutans which is susceptible to β-lactams, and decreases plaque pathogenicity. Over the ages, predominant pathogen in CF lungs becomes Pseudomonas aeruginosa. Usual therapies are chronic treatments with inhaled tobramycin or colomycin, which are not effective against Streptococcus mutans. Therefore, adolescents and adults may lose protection against caries.

Increased salivary pH and higher concentration of calcium in saliva may result in increased calculus formation. The prevalence of dental calculus formation was not found to be significantly different between children with CF and other chronic respiratory diseases.

Children with CF may have dental maturation delay and higher risk for development of systemic enamel defects. It was speculated that this was probably caused by metabolic and nutritional disorders and frequent use of antibiotics, which can have influence on teeth development. Ferrazzano et al. found enamel defects in 55.6% of CF patients, while they were present in 22.7% of healthy persons. In addition, more severe enamel defects with hypoplasia and partial loss of enamel had been noted in the CF group. Azavedo et al. showed no difference in occurrence of lesions on deciduous teeth, but enamel defects on permanent teeth were more prevalent in CF children than in healthy controls. High prevalence of tetracycline discolorations associated with frequent use of these drugs in the past is not common nowadays.

One of less frequent indicators of current malpractice in CF treatment is an inadequate administration of PERT, which happen because of improper use of product that is commercially available. Exocrine pancreatic insufficiency is clinically manifested by symptoms of maldigestion (greasy stools, flatulants, abdominal pain, rectal prolapses) when residual pancreatic function is <10%. Mainly of porcine origin, enteric-coated microsphere preparations were designed to avoid inactivation by acidic environment in the stomach. It dissolves in the duodenum when pH becomes alkaline. In case of crushing or swallowing and not chewing the medications, or mixing them in foods.
oral health in these patients. Patients and their parents should be educated about importance of oral health and possible severe general health consequences in case of presence of oral diseases or its complications. It seems that the occurrence of oral changes could be influenced by improper use of inhalation and PER therapy. Therefore, it is important to educate children and their parents on the proper use of these medicines. Although strict clinical protocols for the prevention of oral diseases in patients with CRD have not been defined so far, it would be of great importance if general paediatricians and pulmonologists would be aware of importance of good oral health suggesting regular dental examinations every three months. Preventive measures, early diagnosis and effective treatment strategies can reduce occurrence of oral diseases and make their complications less frequent which all may have significant impact on possible co-morbidities and patient’s quality of life.

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Change in Patients’ Self-Reported Quality of Life before and after Dental Implantation

SUMMARY

Background/Aim: The loss of teeth and its consequences for health, as well as the psychological discomfort it entails, have a negative impact on both self-reported health state and quality of life (QoL). Dental implantation aims to increase patients’ health and satisfaction and to improve all aspects of QoL. The purpose of this cross-sectional correlational study was to compare the patients’ QoL before and after dental implantation.

Material and Methods: The study comprised 62 patients aged between 24 and 77, including 28 (45.16%) women and 34 (54.84%) men, who reported to a private dental clinic in Szczecin, Poland to replace missing teeth with implants. The survey was carried out twice: prior to the treatment, when the decision to use implants had been made, and three months after implantation, during the first check-up. QoL was measured using the 36-Item Short Form Health Survey (SF-36), and sociodemographic data were collected using a questionnaire of the author.

Results: There were statistically significant differences between the assessment of health and QoL before and after implantation treatment. Both the patients’ health and comfort of life improved after therapy.

Conclusions: Replacement of missing teeth with dental implants brought overall improvement in patients’ QoL, social comfort, and general health. Dental implantation should be recommended in the early phase of edentulism, after tooth loss.

Key words: Dental Implantation, Oral Health-Related Quality of Life, Health Survey

Introduction

The loss of teeth is a serious life event that impairs two important functions, eating and speaking, and may be accompanied by problems such as pain and bleeding associated with gum disease. It thus affects various aspects of Quality of Life (QoL). Patients experience pain and psychological discomfort1-2. Somatic diseases may cause pathological changes within the oral cavity, potentially leading to partial or complete edentulism and, consequently, to the need to replace the missing teeth with implants3,4,5,6,7.

QoL and general health can serve as important indicators of the effects of a treatment, and should be taken into account during oral diagnosis, interventions, and the application of procedures5-7. Recent studies have revealed significant relationships between QoL, overall oral status, orthodontic treatment, and treatment with prosthetic implants3,4,6,7,8,9.

QoL is defined as an individual’s perception of his or her position in life, within the cultural context and value system he or she lives in, and in relation to his or her goals, expectations, parameters, and social relations6,9. QoL is regarded as a dynamic construct that changes over time, and which can be affected by health problems that occur2. As a vital outcome of therapy, QoL is rated among the so-called soft effects of treatment, since it modifies the internal and external conditions affecting the individual.

Researchers examining the effect of edentulism on the oral health-related quality of life (OHRQoL) have demonstrated that patients suffering from this problem had lower levels of QoL, higher levels of functional
limitation, more severe physical and psychological pain, and more serious sleep and digestive disorders. The purpose of prosthetic treatment is to alleviate tooth loss-related functional and aesthetic problems, and to improve QoL. According to Levi, one factor that motivates people to choose a particular method of tooth replacement is the aesthetic aspect. Patients who decide to undergo dental implantation feel a difference in their own perception of themselves. Dental implants are made of titanium and are inserted directly into the jawbone. On account of the osseointegration and mechanical stability they offer, they serve as pillars for prosthetic filling. Bone grows up to the implant surface, meaning that the implant can function as a natural tooth. Providing that hygiene principles are obeyed, implants should continue to function properly.

Implantation treatment aims to reconstruct missing teeth, thus restoring the normal functioning of the stomatognathic system, providing lips with support and, consequently, restoring facial profile and improving appearance. Implant-based definitive dental prostheses are widely applied as a highly efficient prosthetic treatment method. Some researchers maintain that the QoL related to edentulism, as well as its health, functional, psychological, and social consequences, the therapy employed, and oral hygiene during implantation treatment has not yet been fully described. An important contributor to QoL is the overall health and oral hygiene status. QoL assessment is widely used in dental medicine, since it provides information about changes in patients’ self-perceived health and QoL level in the course of the pathological process and after dental treatment. Nevertheless, as Riordain et al. pointed out, there is still too little evidence for improvement in QoL after implantation treatment.

The aim of this study was to compare the QoL of patients subjected to dental implant treatment before and after the treatment. We assumed that this method of replacing missing teeth has an effect on QoL and the self-reported health state.

Material and Methods

Study Design

This was a longitudinal clinical study with selected predictor variables. We formulated the hypothesis that, the patients’ QoL would be considerably improved following implantation. The survey was carried out twice during 2015 using the same questionnaire: once at the initial visit (V1), during which the doctors and patients established the protocol of dental implant treatment (including treatment date), and then on the first check-up (V2), after dental implantation (this usually took place three months after the treatment).

The criteria for inclusion in the study were age at least 18 years, committed to dental implantation, agreement to take part in both surveys, successfully completed implantation treatment, and attending the check-up. Each patient was examined by a dentist using a mirror and a probe under artificial lighting. The dentist assessed the patients’ needs for treatment and the oral health status.

Procedure

Our study was approved by the Bioethics Committee of the Pomeranian Medical University in Szczecin, Poland (approval no. KB-0012/41/05/15). The patients participated in the study voluntarily and gave their informed consent. The study was conducted in accordance with the guidelines of the Helsinki Declaration and the principles of Good Clinical Practice, as well as with respect for the rights and dignity of the person.

Evaluation of oral health-related quality of life (OHRQoL)

The research instruments used in this survey-based study were the 36-Item Short Form Health Survey (SF-36) and a questionnaire of the author’s devising (AQ). We obtained permission from Quality Metric, Inc. to use the authorized Polish Version of the SF-36. The SF-36 questionnaire consists of 36 questions divided into eight subscales: role physical (RP), mental health (MH), bodily pain (BP), general health (GH), physical functioning (PF), vitality (V), social functioning (SF), role emotional (RE), and one additional question concerning health change. Score on the Likert scale for each of these areas ranges from 0 to 100, with 0 denoting the worst and 100 the best possible health state. SF-36 is not time-consuming to use, and its usefulness, repeatability, and ability to reveal changes in QoL have been demonstrated. The author’s questionnaire contained 20 simple structured questions aimed at (1) demographic data collection (age, sex, place of residence, education, marital status, financial income), (2) selected clinical data concerning implantation treatment.

Statistical Analysis

The normality of variables distribution was verified using the Shapiro-Wilk test. The variables were characterized by arithmetic means (X), standard deviations (SD), medians (M), and extremes (min–max). Statistical analysis was performed using the chi-square test, Student’s t-test, analysis of variance (ANOVA), the correlation coefficient, and Cronbach’s alpha, in order to assess the reliability of the results in particular SF-36 domains. All tests were performed at a statistical significance level of α = 0.05.
Results

Participants

The study comprised 62 participants, including 28 (45.2%) women and 34 (54.8%) men, aged between 24 and 77. The mean age of the participants was $M_{\text{AGE}} = 55.77$ years, and the standard deviation (SD) was 12.01 for the entire group; $M_{\text{Female}} = 52.61 \pm 12.88$ (range: 24–77), $M_{\text{Male}} = 58.38 \pm 10.73$ (range: 28–73). Sociodemographic data are shown in Table 1. The majority of the participants had tertiary (43 - 69.35%) or secondary (18 - 29.03%) education; one person, a man (1.62%), had no higher than vocational education.

Table 1. Sociodemographic data of the participants (n=62)

<table>
<thead>
<tr>
<th>Variables</th>
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<tr>
<td>Age mini-max</td>
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<td>Gender mini-max</td>
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<td>Men X±SD</td>
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Tooth loss among the study subjects was mostly caused by dental caries (tooth decay) (24 - 38.71%), neglect of oral hygiene (19 - 30.65%), and periodontal disease (20 - 32.26%). The prevailing causes among the women were dental caries (11 - 39.29%) and periodontal disease (10 - 35.71%), and among the men, dental caries (13 - 38.24%) and oral hygiene neglect (13 - 38.24%). The main factor motivating the patients as a whole (50 - 80.65%) to choose implantation treatment was the esthetic aspect; the main factor motivating the women (23 - 82.14%) was psychological comfort, and the main factor motivating the men (30 - 88.24%) was the esthetic aspect.

Health assessment before and three months after implantation treatment (on the check-up):

1. The participants described their health as excellent (3 - 4.84%) or very good (25 - 40.32%), The mean score was $8.77 \pm 4.29$. The women more often described their health state as excellent or very good (16 - 57.14%), and the men as good or average (22 - 64.7%).

2. There was a statistically significant relationship between health assessment three months before treatment ($X= 6.78 \pm 1.85$) and three months after treatment ($X= 8.64 \pm 1.50$) (Table 2),

Table 2. Self-assessment of patients’ health status before and after treatment

<table>
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<th>After treatment</th>
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<td>SD</td>
<td>p</td>
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<tr>
<td>6.78</td>
<td>1.85</td>
<td>F=-7.502</td>
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</table>

3. Health assessment statistically significantly depended on economic status ($p= 0.04$); the higher patient’s economic status, the better health assessment,

4. Patients observed positive changes in their comfort of life, which were reflected in higher health assessment ($p= 0.05$).

Quality of life

1. The highest score was obtained for the physical functioning (PF) ($X= 89.68 \pm 14.43$), and the lowest for the general health (GH) ($63.87 \pm 18.34$) and vitality (V) ($73.63 \pm 16.17$) domains. Cronbach’s alpha was calculated for each of the SF-36 scales. For the physical functioning (PF), mental health (MH), vitality (V), bodily pain (BP), and general health (GH) domains, alpha was > 0.7, which suggested high reliability of the scales; for the role physical (RP) and role emotional (RE) domains, alpha was < 0.7,

2. The women scored higher for physical functioning (PF) than the men ($p= 0.03$), and the younger patients (aged up to 60 years) scored higher than those over 60 ($p= 0.01$),

3. The correlation coefficient for the physical functioning (PF) and role physical (RP) domains was 0.718 ($p< 0.001$), while for the mental health (MH) and vitality (V) domains, this was 0.756 ($p< 0.001$),

4. The lowest scores for general health (GH) were obtained by the patients aged between 56 and 60 years, while the highest was obtained by those younger than 55 ($p= 0.005$). The lowest scores were obtained by patients with vocational and secondary education only, and the highest by those with third-level education ($p< 0.001$),

5. The largest changes in health status were reported by patients over 60, and the smallest by patients aged up to 55 years ($p= 0.05$),

6. Higher economic status was associated with an increase in the average score for the vitality (V) QoL domain ($p= 0.03$),
7. City dwellers assessed their social functioning (SF) higher than their counterparts from rural areas (p= 0.03).

Changes in comfort of life

1. The majority of both the women (18 - 64.29%) and the men (25 - 73.53%) felt that their eating comfort had considerably improved,
2. After treatment, 89.29% of the women (n= 25) and 94.12% of the men (n= 32) observed that their health status and their comfort of life significantly improved - a total of 91.94% (n= 57),
3. Both the women and the men were very satisfied with the effects of implantation treatment. The mean score achieved by the women was 9.14, and by the men 9.24 (total 9.19),
4. The results concerning comfort of life before and after implantation were compared. Health status after dental implantation was assessed as lower by those patients whose comfort of life did not change and assessed highest by those who did see a significant change (p= 0.04).

Discussion

The loss of teeth poses both health and esthetic problems. Patients can obtain satisfaction through advanced implant treatment methods. Apart from the obvious esthetic advantages, dental implantation improves speaking and increases the comfort of biting and gustatory sensation, which translates into psychological and physical well-being and a better health state. Irrespective of age, reconstruction of missing teeth helps patients regain their self-confidence. Polish-language medical literature does not contain publications on the self-reported health state of implantology patients, as assessed by the SF-36. Such studies can, however, be found in the English-language literature. The mean age of the participants in our study was 56 ± 11.8 years. Hence, we can conclude that dental implant treatment is appropriate for patients of all ages, and is worth recommending, considering the ease of maintaining good oral hygiene and the psychological comfort associated with the alleviation of digestive problems.

Patients’ major hesitation in deciding on dental implantation is related to its cost. Nevertheless, patients value their comfort and health, and so they often choose this treatment method, despite the cost.

Our results demonstrate that the health state of our participants improved after implantation. They scored higher in the social functioning (SF) and vitality (V) domains, and suffered less from emotional problems and limitations on their activity. Other studies have compared changes in the QoL of patients with dentures and those with dental implants. Following dental implantation, patients found their QoL to be noticeably higher, whereas the QoL of patients with dentures was definitely lower. Similar results, confirming the alleviation of physical pain and psychological discomfort, have been reported by Yoshida et al. We observed a statistically significant relationship between the patients’ health assessment before the treatment and three months after it. We found that the assessment of patients’ health changed significantly, after implantation it was significantly higher by 2 units on average.

In his study, Yoshida measured chronological change in the QoL level during implantation treatment in a group of 20 patients with a small number of missing teeth (less than 4 teeth), who underwent implantation treatment. The patients completed the shortened Japanese version of the Oral Health Impact Profile (OHIP-J14) before surgery (T0), one week after surgery (T1), one week after interim prosthesis placement (T2), and 1 week after definitive prosthesis placement (T3). Although a temporary functional limitation was observed after implant placement, overall OHRQoL improved after placement of the definitive prosthesis. What is more, implantation treatment was more effective in the unilateral free-end edentulous space. Similarly, in the study of Pavel et al., the most significant associations on the functional scale (FS) were observed with the number of front teeth replaced with implants, followed by the presence of chewing problems and marital status.

Conclusions

This study has demonstrated the significant effect of implantation, as a method of treating missing teeth, on the self-reported health state. Implantation treatment improved patients’ health, QoL, and comfort of life. QoL, as measured by the SF-36 was higher after the implantation treatment than before it. Dental implantation should be recommended in the early phase of edentulism, after tooth loss.

Abbreviations

SF-36 - 36-Item Short Form Health Survey
AQ - Author’s Questionnaire
QoL - Quality of Life

References


Conflict of Interests: Nothing to declar.

Financial Disclosure Statement: Nothing to declar.

Human Rights Statement: All the procedures on humans were conducted in accordance with the the Helsinki Declaration of 1975, as revised 2000, and with national ethical committee. Consent was obtained from the patient/s and approved for the current study by national ethical committee.

Animal Rights Statement: None required.

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Evaluation of Permanent First Molar Tooth Loss in Young Population from North Turkey

**SUMMARY**

**Background/Aim:** Rate of missing permanent teeth in a population is important for oral health indicators. The aim of this retrospective study was to evaluate the prevalence of previously missing permanent first molar (PMF) teeth in a young population. **Material and Methods:** 1204 healthy patients who received panoramic radiographs were selected randomly at their first visiting to Samsun Ondokuz Mayis University Faculty of Dentistry. The patients' age ranges were 7 to 17. Information about extracted permanent first molar teeth, missing regions, patients' ages, and genders was recorded. The data were statistically analyzed using chi-square tests. **Results:** 1,204 patients, of these, 608 (51%) were female, and 596 (49%) were male. 4,816 PFM teeth were evaluated in this study, and 128 of them (2.66%) had extracted from 97 different patients. There were statistically differences between groups in terms of age and gender ($p<0.05$). It was observed that mandibular teeth were more frequently extracted than maxillary ones, and lower left permanent first molar teeth had more extracted than the others. **Conclusions:** The first permanent molar teeth could be extracted different reasons. However, these teeth should be protected by both dentists and patients.

**Key words:** Age, Child, Permanent First Molar, Tooth Extraction

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**Introduction**

Oral health care is part of general health, and it is considered essential to an individual’s quality of life. Therefore, tooth loss is considered a public health problem. Dental health programs are aimed to decrease dental plaque, tooth decay, periodontal disease and loss of teeth. Although many alternative practices have been developed to protect oral health, early tooth loss is still a big problem.

Permanent first molar (PFM) teeth are the first developing permanent teeth in posterior region. However, permanent first molar teeth have been characterized as most caries-prone teeth in the mixed dentition. Additionally, 10-19% of PFM teeth were hypomineralization. Consequently, PFM teeth may be lost at an early age. Other reasons for PFM teeth loss were poor prognosis, caries, orthodontic reasons and periodontal diseases.

Early extraction of PFM teeth may cause undesirable rotation and mesial drifting of secondary permanent molars. PFM teeth play an important role in balanced and normal occlusion. Because of this, early extraction of PFM may affect whole occlusion and development of both jaws. Additionally, asymmetry and temporomandibular joint problems may be observed. For making a decision about extraction of PFM, dental pain, excessive material loss, parental attitudes and toleration of dental treatment may affect indication. All conditions, in the developing dentition, should be assessed before extraction of PFM teeth.

In the present retrospective study was evaluated prevalence of missing permanent first molar teeth in young patients first admitted to Ondokuz Mayis University, Pediatric Dentistry Clinic for examination.
Material and Methods

This study was performed in the north region of Turkey (Middle Black Sea Region). Ethics approval was obtained from the Ethics Committee of Medical Research of Ondokuz Mayis University, Samsun, Turkey (2015/02). This study was performed in Ondokuz Mayis University, Faculty of Dentistry, and included 1,204 healthy patients (608 females and 596 males) who were admitted for the first time to Pediatric Dentistry Clinic for a routine dental control at first quarter of 2012. The patients’ age ranges were 7 to 17, and they had received panoramic radiographs. Patients with edentulism due to a systemic disease were excluded from the study. Demographic information was recorded, including age and gender. Extracted PFM teeth, jaws, right or left side of the oral cavity were determined from panoramic radiographs. Extraction required teeth were not recorded. The data were collected retrospectively by the same physician.

Statistical analyses were performed using SPSS version 22.0 (SPSS Inc., Chicago, IL, U.S.A.). All data were evaluated as frequency and percentage. Chi-square tests were used to compare relative differences in extracted PFM, gender, age, and jaws. P<0.05 was accepted as statistically significant in comparisons.

Results

In all, 1,204 patients were assessed. Of these, 608 (51%) were females, and 596 (49%) were males (p>0.05). It was determined that 128 PFM teeth (2.66%) from a total of 4,816 had been extracted from 97 (8%) of 1,204 patients. Seventy patients had only one PFM tooth loss, 23 patients had two, and four patients had three missing PFM teeth. No patient had four missing PFM teeth. The distributions by age, gender, and extracted first molar in the subjects are summarized in Table 1 and Table 2.

Table 1. Distributions of patients according to age and gender

<table>
<thead>
<tr>
<th>Age</th>
<th>Female Extraction</th>
<th>Male Extraction</th>
<th>Total Extraction</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Yes*</td>
<td>No</td>
<td>Yes*</td>
</tr>
<tr>
<td>7</td>
<td>0</td>
<td>49</td>
<td>0</td>
</tr>
<tr>
<td>8</td>
<td>1</td>
<td>64</td>
<td>1</td>
</tr>
<tr>
<td>9</td>
<td>0</td>
<td>75</td>
<td>1</td>
</tr>
<tr>
<td>10</td>
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<td>46</td>
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<tr>
<td>11</td>
<td>2</td>
<td>45</td>
<td>6</td>
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<tr>
<td>12</td>
<td>7</td>
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<td>1</td>
</tr>
<tr>
<td>13</td>
<td>8</td>
<td>62</td>
<td>11</td>
</tr>
<tr>
<td>14</td>
<td>12</td>
<td>47</td>
<td>4</td>
</tr>
<tr>
<td>15</td>
<td>14</td>
<td>61</td>
<td>8</td>
</tr>
<tr>
<td>16</td>
<td>11</td>
<td>53</td>
<td>4</td>
</tr>
<tr>
<td>17</td>
<td>3</td>
<td>4</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>59</td>
<td>549</td>
<td>38</td>
</tr>
</tbody>
</table>

*The number of patients having at least one first molar tooth loss

Considering the number of extracted teeth, gender, age and regions of jaws were all statistically significant (p<0.05). There was 49 (2.05%) extracted PFM teeth from 38 males and 79 (3.25%) extracted PFM from 59 females (p<0.05) (Figure 1).

Distribution of extracted PFM teeth according to the jaws and sides was showed in Table 3. There was statistical difference between the jaws but no in the sides.

Table 3. Distribution of extracted PFM in the jaws and sides

<table>
<thead>
<tr>
<th>Categories</th>
<th>N (%)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Jaw</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Maxilla</td>
<td>87 (68%)</td>
<td>p&lt;0.05</td>
</tr>
<tr>
<td>Mandible</td>
<td>41 (32%)</td>
<td></td>
</tr>
<tr>
<td>Side</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Right</td>
<td>68 (53%)</td>
<td>p&gt;0.05</td>
</tr>
<tr>
<td>Left</td>
<td>60 (47%)</td>
<td></td>
</tr>
</tbody>
</table>

Discussion

For a long time, extraction of PFM has been subject to debate. The extraction of PFM should be planned with an orthodontist before the eruption of second and third permanent molars. In recent years, orthodontists have favored extraction and also have given indications for extraction of PFM\textsuperscript{12,13}. Pediatric dentists have to perform complicated fillings that result in excessive material loss in children who have dental anxiety and behavior-management problems\textsuperscript{14,15}. Also, these molar teeth must be kept in the mouth for preventive reasons. Sometimes, orthodontists may want to preserve these teeth because of orthodontic procedure\textsuperscript{13}. Consequently, it is difficult...
to decide about PFM tooth extraction. Therefore, in the present study, only the frequency of extracted PFM teeth was investigated.

Despite the researches, early tooth loss is still a big problem, especially since the early loss of permanent first molars plays a key role in the asymmetry of dentition\textsuperscript{6,8,10}. Therefore, determination of the number of patients with early loss of permanent first molars in the community will be beneficial. In the present study, although many cases required tooth extraction etiologically, only extracted first molar teeth were investigated. In this study, the number of previously extracted permanent first molars was 128 (2.66\%) of 4,816 PFM teeth in 97 (8\%) of 1,204 patients. The data were compared with previous studies\textsuperscript{16-18}. Alves et al.\textsuperscript{16} performed a study among 12-year-old schoolchildren from South Brazil. The researchers observed that tooth loss rate in 1,528 patients was 5.81\%. This rate was lower than in this study (8\%) because they included only patients who were 12 years of age. George et al.\textsuperscript{17} reported that rates of all missing permanent teeth in children and young people of 6, 12 and 15 ages were 5.7\% 22\% and 28.3\%, respectively. These rates were higher than in this study because they included all permanent teeth. Atieh\textsuperscript{19} reported that the rate of tooth loss in 484 patients aged 14-19 were 40.9\% (198 patients). This rate was higher than in this study because they had included all permanent teeth. Demirbuga et al.\textsuperscript{18} performed a study on 31,580 permanent first molar teeth from 7,895 patients’ panoramic radiography, and reported that missing teeth rates from 19,488 teeth in 6-11 age groups were 0.47\% (122) and in 12-16 age groups from 12,092 teeth were 4.14\% (501). These results were very similar to the results in this study.

Several studies\textsuperscript{17,18,20} determined that gender may influence tooth loss, which agrees with findings of present study. Demirbuga et al.\textsuperscript{18} reported that of the 15,008 teeth examined in the boys’ group, 1.84\% (276) were missing, and in the girls’ group, of 16,572 teeth, 347 (2.09\%) teeth were missing. George et al.\textsuperscript{17} determined that tooth loss rates in males (42.9\%) were lower than in the females (47.9\%). Barbato and Perez\textsuperscript{20} claimed that this finding could be explained by the fact that females use more dental services due to their deeper health or aesthetic concerns, which may lead to overtreatment. On the contrary, Bhat et al.\textsuperscript{21} reported that the percentages of extracted teeth in males were 53.1\% and in females were 46.9\%. Jafarian and Etebarian\textsuperscript{2} assessed that males comprised 48.7\% of patients, but they had more extracted teeth (56.1\%) than females (43.9\%). On the other hand, Susin et al.\textsuperscript{22} claimed that tooth loss was affected more by the age factor than by gender. They stated that the prevalence of tooth loss increased markedly with age from 26\% to 60\% in the age groups 14-19 and 25-29 years, respectively\textsuperscript{22}.

Some studies stated that most missing teeth were in the mandibular\textsuperscript{20,21}. Bhat et al.\textsuperscript{21} determined that the rate of missing mandibular first molar teeth was 21\%, while the rate of missing maxillary first molar teeth was 10\%. Barbato and Perez\textsuperscript{20} showed that most missing teeth in jaws were mandibular first molars. Demirbuga et al.\textsuperscript{18} reported that rates of missing permanent first molar teeth in mandibula were 2.77\% (438) and 1.17\% (185) teeth in maxilla. In the present study, the number of extracted PFM teeth in the lower arch was 87 and in the upper arch was 41. This situation can be explained by several factors. One of them is that more nutrients remain in the lower jaw than upper. Others are early eruption of mandibular teeth, caries, increased hypomineralization level, different effects of saliva and different anatomical structures of the teeth.

In the literature, there is limited information about early missing teeth on the left and right sides of the oral cavity in pediatric patients\textsuperscript{18}. It was claimed that hand selection when tooth brushing may affect rates of extraction on left or right sides of the oral cavity\textsuperscript{18}. Similarly, chewing and cleaning habits and the residence time of food in the mouth may play important roles in rates of extraction on the left or right sides of the jaws. In contrast, Bhat et al.\textsuperscript{21} found no significant difference between rates of extraction on the left and right sides of the oral cavity. Demirbuga et al.\textsuperscript{18} reported that right side missing teeth numbered 302 (1.91\%) and left side missing teeth amount to 321 (2.03\%), and they did not find statistical differences between the right and left sides of jaws. Similar to previous studies, this study’s results showed that the numbers of formerly extracted PFM teeth were 60 in the right side and 68 in the left side and showed no statistical differences.

The ideal time for extraction of a PFM is at a chronological age of 8-10 years\textsuperscript{11}. Gill et al.\textsuperscript{23} claimed that extraction of PFM with poor prognosis in this time interval should facilitate mesial movement of the permanent second molar into the PFM area. Otherwise, extraction at a later age may result in unsatisfactory and inadequate space closure, condylar problems and orthodontic malocclusion\textsuperscript{24}. In the present study, according to age, the rate of missing teeth was 5.5\% in 8-10 age and 57\% in 13-15 age, but the time of extraction of the missing teeth was not known exactly.

Nowadays, dental materials and treatment choices have rapidly evolved. Direct/indirect pulp capping, root canal treatment, post-core, inlays/onlays, porcelain and ceramic crowns are good alternative treatments\textsuperscript{25}. The results of the study showed that a large number of teeth had extracted between the ages of 11-16. Preventive treatments should be increased for preservation of natural dentition, especially, in females.

Conclusions

In this retrospective study, mandibular first molar teeth were more frequently missing than maxillary teeth,
and females had more missing teeth than males. No difference existed between right and left side teeth loss. Even if PFM tooth loss is common in community, these cases are preventable. Therefore, several factors such as dental education of the community, brushing habits, specialized dental care and conservative treatments should be improved. Further studies with different parameters (socio-economic status, educational status) are necessary.

References


A Prospective Clinical Study of the Efficacy of Hyflex CM Rotary Instruments in an Endodontics Undergraduate Program

SUMMARY

Background/Aim: To investigate the incidence of procedural errors with the use of a novel nickel-titanium rotary system (Hyflex CM, Coltene/Whaledent, Altstätten Switzerland), evaluate the technical quality of root canal treatments and assess a questionnaire completed by the participants themselves in an undergraduate dental clinic between 2014 and 2017 (Department of Endodontology, School of Dentistry, Aristotle University of Thessaloniki). Material and Methods: 118 undergraduate students in their first year of clinical practice performed a root canal treatment on a patient’s molar (maxillary/mandibular). None of the participants had previous experience in rotary instrumentation. The periapical radiographs were taken with the use of the paralleling technique for standardization and were collected and evaluated by the investigator. After the root canal treatment was performed the students completed a questionnaire in order to evaluate their training on rotary instrumentation. Results: The overall incidence of instrument separation, apical perforation, root perforation, straightening and ledges was 0.8%, 4.4%, 2.3%, 5.5% and 29% respectively on root canal level. Ledges were detected more often in mandibular mesiobuccal canals. The frequency of root canals with an ‘acceptable’ filling was 68.4%, while overfilled and underfilled canals were found to be 8.6% and 16.2% respectively. The response rate was high (94.9%), 35% of the participants encountered no difficulty in the use of rotary instrumentation and 98.2% would use it again. Conclusions: The incidence of procedural errors was considerably low and the technical quality of the filled root canals was superior to that of similar studies. The responses of the questionnaire demonstrated a positive attitude toward rotary instrumentation.

Key words: Hyflex CM, Procedural Errors, Technical Quality, Undergraduate Dental Clinic, Questionnaire

Introduction

The advent of Ni-Ti rotary instrumentation has improved the quality and speed of cleaning and shaping procedures; simultaneously procedural errors such as transportation, ledging, zipping and perforations are decreased1,2. Despite their indisputable advantages, Ni-Ti instruments are still susceptible to separation3. Their widespread use among clinicians and the need to reduce procedural errors for more predictable results, have led to a rapid development of new endodontic rotary systems while formers are updated at an exponential rate.

Hyflex CM rotary files (Coltene/Whaledent, Altstätten, Switzerland) are manufactured from a novel type of Ni-Ti wire, namely Controlled Memory (CM) wire, which has undergone a unique proprietary thermomechanical processing4, that is not disclosed by the manufacturer. These instruments, as stated by the manufacturer, possess a lower percentage in weight of

Original Paper (OP)

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nickel (52.1 Ni% wt) in comparison to the vast majority of commercially available Ni-Ti rotary instruments (54.2-56.2 Ni% wt). Due to their special manufacturing process, Hyflex CM files are highly flexible, more resistant to cyclic fatigue, and partially or fully regain their original shape after sterilization. Rotary instrumentation is an integral adjunct to endodontic practice, thus undergraduate programs have gradually included it in their curricula. The purpose of this prospective study that was conducted in the Undergraduate Clinic of the Department of Endodontontology at the Aristotle University of Thessaloniki, Greece was two-fold. The primary objective was to investigate the frequency of root and apical perforations, ledges, straightening, separated instruments and the quality of root fillings when rotary instrumentation was used for the first time by undergraduate students. The second objective was to gain an insight on the undergraduates’ self-assessment concerning the root canal treatment and the evaluation of the rotary experience based on a questionnaire. To the best of our knowledge, there is no published data evaluating the efficacy of Hyflex CM rotary instruments clinically. Specifically, this is the first prospective clinical study to assess the efficacy of Hyflex CM rotary instruments in an endodontics undergraduate program and the first to use a questionnaire completed by the participants themselves to evaluate their training on rotary instrumentation.

Material and Methods

Selection of cases

During three academic years (2014-15, 2015-16, 2016-17), a total of one hundred and eighteen undergraduate students on their first year of clinical practice participated in the study. None of the participants had a previous experience in rotary instrumentation; however all of them had performed two or more root canal treatments on patients with hand instruments. The participants performed a root canal treatment on one molar (maxillary/mandibular).

Instrumentation technique

The single-length instrumentation sequence of Hyflex CM system (Coltene/Whaledent, Altstätten, Switzerland) was demonstrated to each student on resin blocks by the investigator (ZDT), according to the manufacturer’s recommendations. After the access cavity was prepared, the working length was determined with a radiograph and in a few cases with the combination of an electronic apex locator (CanalPro Apex Locator, Coltene/Whaledent, Altstätten, Switzerland) and a radiograph. The canals were negotiated to the working length with stainless steel K-files up to size 20. The enlargement of the root canals was accomplished with the following order of Hyflex instruments, 25.08, 20.04, 25.04, 20.06 and 30.04. File size 40.04 taper was used only at the distal canals of mandibular molars and at the palatal canals of maxillary molars. All files were used in a slow speed handpiece (CanalPro CL Endodontic Handpiece, Coltene/Whaledent, Altstätten, Switzerland), at a setting and speed recommended by the manufacturer (2.5 Ncm, 500 rpm). As an irrigant between the files and after instrumentation 3% sodium hypochlorite (CanalPro, Coltene/Whaledent, Altstätten, Switzerland) was used. To remove smear layer 17% EDTA solution (CanalPro, Coltene/Whaledent, Altstätten, Switzerland) was applied. The canals were obturated with tapered master cone (0.04) and accessory points, combined with Roeko Seal sealer (Coltene/Whaledent, Altstätten, Switzerland) using the lateral condensation technique. Each file was used at a maximum of three times and was discarded either when it did not regain its form after sterilization or when a distortion or deformation were detected under magnifying loupes 3x. The incidence of each procedural error was calculated on root canal level.

Radiographic evaluation

The periapical radiographs were taken with the use of digital imaging technology (DIGORA® Optime digital imaging plate system, Soredex Tuusula, Finland). In order for the projections to be standardized the paralleling technique was used. Superimposed canals and canal fillings, working length radiographs with incorrect estimation of the working length, missed canals, calcified canals, and cases of radiographs without depiction of the apices were excluded. The data were collected and observed by the investigator.

Detection of procedural errors and evaluation of the technical quality of the root fillings

The criteria for the detection of procedural errors were as follows:

- Separated instruments were diagnosed during the time of the incidence and their location was determined radiographically,
- Ledge formation was diagnosed when the root filling was at least 1mm shorter than the working length and/or deviated from the original canal curvature,
- Apical perforation was diagnosed when the filling material extruded through the apical foramen,
- Root perforation was diagnosed when the filling material extruded in the lateral walls of the root,
- Straightening was diagnosed when a deviation from the original canal curvature was evident between the working length and the cone-fit radiograph.

The radiographic evaluation of the quality of the root fillings was based on the length and the density of the root filling and its adaptation to the canal walls. The categorization of the criteria was as follows:
A length of $\leq 2m$ from the apex with no voids ('Acceptable' filling)
- A length of $\leq 2m$ from the apex with voids
- Overfilling with no voids
- Overfilling with voids
- A length of $>2mm$ from the apex with no voids
- A length of $>2mm$ from the apex with voids. \(^{16}\)

**Questionnaire survey**
A hand-delivered questionnaire was designed including 15 closed-ended questions concerning hand instrumentation experience, degree and reason of difficulty with rotary instrumentation, familiarity with terms of rotary instrumentation, preparation’s and obturation’s quality, identification of procedural errors and predisposition to rotary instrumentation. An external pilot survey was conducted amongst 20 undergraduate students to evaluate the questionnaire’s reliability. The questionnaire was administered to the participants after the root canal treatment was performed and only those with all questions completed were included in the data analysis.

**Ethical consideration**
The study was approved by the Ethical Committee of the School of Dentistry of Aristotle University of Thessaloniki, Greece (protocol no.44/24-06-2016).

**Statistical analysis**
Cohen’s kappa coefficient was used to calculate interobserver and intraexaminer reliability regarding ledge formation and root filling’s quality. Interobserver agreement was determined by the scores of the radiographs of 30 randomly selected cases, while intraexaminer agreement was obtained by rescoring the radiographs of 40 randomly selected cases one month after the first evaluation. The reliability of the questionnaire was measured with the test-retest model. Twenty randomly selected undergraduate students were delivered the questionnaire for a second time approximately one month after their first response and the scores between the two time intervals were compared using the k-coefficient. The responses of the examiners and participants were calculated using the Statistical Package for Social Sciences (IBM SPSS v.24, Armonk, NY, USA). Qualitative data analysis was carried out using descriptive statistics for observed values and frequencies, Chi Square test and Fisher’s Exact test.

**Results**

**Reliability**
The k-values for interobserver reliability were 0.87, 0.88 and 0.87 for root filling’s length, presence of voids and ledges respectively. The k-values for intraexaminer reliability were 0.88 and 0.89 for root filling’s length and presence of voids and 0.95 for ledges. Because of the near perfect agreement, the scores of one author (ZDT) were used for the radiographic evaluation of the study. Similarly, the k-values obtained from the test-retest reliability were 0.89.

**Procedural errors**

**Separated Instruments**
The overall incidence of rotary instrument separation on root canal level was 0.8%. Of the 3 separated instruments one was located in a distal canal while the remaining two in mesiobuccal canals of maxillary and mandibular molars. Canal location did not have an effect on instrument separation ($p>0.05$) (Figure 1).

**Ledges**
Ledges were found in 29% of the root canals (96/330). 4.5% of the ledged canals was accompanied by straightening and/or root perforation. A statistical significant correlation was found between canal type and the incidence of ledges ($p< 0.05$). Statistical significant differences were found between distobuccal and mandibular mesiobuccal, distal and mandibular mesiobuccal, maxillary mesiobuccal and mandibular mesiobuccal and between palatal and mandibular mesiobuccal root canals.

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**Figure 1.** Example of instrument separation located in the apical third of the distal canal on tooth #36: A) Preoperative radiograph, B) Working length radiograph, C) Cone-fit radiograph, D) Postoperative radiograph.
Perforations
Root perforations and apical foramen damage were found to be 2.3% and 4.4% respectively. Root canal type did not affect the incidence of root perforation (p>0.05).

Canal Straightening
Canal straightening was detected in 5.5% of the root canals. No statistical significance was found between the type of the root canal and the incidence of canal straightening (p>0.05) (Figure 2, Table 1).

Table 1. Incidence (%) of procedural errors

<table>
<thead>
<tr>
<th>Type of procedural error</th>
<th>Percentage</th>
<th>Number of root canals</th>
</tr>
</thead>
<tbody>
<tr>
<td>Instrument separation</td>
<td>0.8</td>
<td>3</td>
</tr>
<tr>
<td>Ledge formation</td>
<td>29.0</td>
<td>96</td>
</tr>
<tr>
<td>Root perforation</td>
<td>2.3</td>
<td>8</td>
</tr>
<tr>
<td>Apical perforation</td>
<td>4.4</td>
<td>15</td>
</tr>
<tr>
<td>Canal straightening</td>
<td>5.5</td>
<td>18</td>
</tr>
</tbody>
</table>

Quality of the root filling
Root filling classified as “acceptable” was observed in 68.4% of the filled root canals, while 87.6% exhibited no voids and 75.2% were filled adequately. The frequency of overfilled and underfilled canals was found to be 8.6% and 16.2% respectively. Root canal type did not have an effect on the quality of the root filling (p>0.05) (Table 2).

Table 2. Percentages (%) of filled root canals according to classification's criteria

<table>
<thead>
<tr>
<th>Classification</th>
<th>Number of root canals</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.2 mm with no voids</td>
<td>232</td>
<td>68.4</td>
</tr>
<tr>
<td>0.2 mm with voids</td>
<td>23</td>
<td>6.8</td>
</tr>
<tr>
<td>Overfilling with no voids</td>
<td>27</td>
<td>8.0</td>
</tr>
<tr>
<td>Overfilling with voids</td>
<td>2</td>
<td>0.6</td>
</tr>
<tr>
<td>&gt;2mm with no voids</td>
<td>38</td>
<td>11.2</td>
</tr>
<tr>
<td>&gt;2mm with voids</td>
<td>17</td>
<td>5.0</td>
</tr>
<tr>
<td>Total</td>
<td>339</td>
<td>100.0</td>
</tr>
</tbody>
</table>

The response rate was 94.9%. The respondents were mostly familiar with the step-back instrumentation technique (70.5%). Most participants (92%) reported that the use of rotary instrumentation varied from easy to very easy while only 0.9% described the experience as “hard”. As the reason for the encountered difficulty-if any, the majority identified their inexperience, while more than 1/3 of the participants experienced no difficulty. More than...
half of them characterized the quality of their preparation as “very good”, while 0.9% as poor. Additionally, 70.5% considered “speed” as the major advantage of rotary instrumentation followed by “quality of the preparation”. The vast majority (98.2%) was positively predisposed to using rotary instrumentation in the future, while 74.1% considered rotary instrumentation superior to hand instrumentation (Table 3).

Discussion

This prospective clinical study evaluated the frequency of iatrogenic errors during rotary instrumentation and obturation by undergraduate students in a 3-year period. Our results indicate that the overall incidence of separated rotary instruments is lower than those of similar clinical and case-control studies. Previous studies reported instrument separation by undergraduate students that ranged between 1.0 and 2.09% 17,18. However those results derive from data consisting of all tooth types and the incidence of separation refers to hand and not rotary instruments. Studies referring to rotary instrument separation report results between 1.33-4.44% 10,19,20. The incidence reaches 2.5% when only molars are concerned10. Nevertheless, the root canal treatments in the latter studies were performed by postgraduate students and specialists with greater experience in rotary instrumentation. Mesiobuccal canals of maxillary and mandibular first molars exhibited the highest incidence of separation (0.56%). Those canals are narrow with great primary and apical curvatures21,22, thus susceptible to instrument separation. The low separation rate in our study may be attributed to the establishment of a manual glide path with a No 20 K-file. A direct comparison of those results is difficult to achieve since clinical studies investigating instrument fracture use different study designs and instruments.

Regarding the presence of ledges our results are lower than those of other investigators12,13,16,23 who detected ledges in 33-51.5% of molar cases. Dentina chips or residual debris which result in apical canal blockage can also affect the obturation length16, thus an overestimation of ledges might have occurred. It is also speculated that ledging was a result of inappropriate usage of hand files, overusage or loss of the determined working length. However, due to the highly controlled conditions and the fact that the canals were rotary instrumented, a lower percentage was expected.

Similarly low is the frequency of this study regarding root perforation compared to those of similar studies that range between 2.7% and 18.8% 14,16,24. When only molars are considered our observations are similar to those of Balto et al. (2.2%)13. To a certain extent root perforations might have derived from the misusage of K-files when inserted in severely curved canals, followed by rotary instrumentation of the newly-created path. The incidence of apical perforation in other studies by Balto et al.13 and Khabbaz et al.14 is 10.6% and 25.7% respectively, which is higher compared to ours. A loss of the working length during instrumentation or canal transportation could be responsible for the incidence of apical perforations.

The percentage of canals maintaining their original shape and curvature was 94.5%. It is accepted that canal deviations are minimized when rotary Ni-Ti instruments are used25,26. In vitro results indicate that Hyflex CM instruments produce less canal deviations compared to other Ni-Ti rotary instruments15-27. In our study the results are relatively low, but it must be considered that the use of two-dimensional radiographs to evaluate the straightening of a three-dimensional structure may be inaccurate.

The radiographic evaluation of the technical quality of root fillings showed that 68.4% of the root canals fulfilled the criteria of an “acceptable” filling. Previous studies that assessed the technical quality of root fillings performed by undergraduate students exhibited a lower percentage (13-55.3%)13,16,28-30. When only molars were considered, the latter studies indicated results ranging between 6.1% and 54.6%. When each root canal was considered as one unit those results reached 37.6% and 46.7% 16,28. Despite the superiority of our results, it is difficult to compare these studies due to the differences in the categorization criteria, the tooth type selection and the evaluation of each tooth as a unit or of each root canal individually. Moreover, the length of the root canal filling was estimated more precisely in our study, since the radiographs were taken with the paralleling technique and the distance of the root filling from the radiographic apex was calculated with digital measuring technology.

The response rate on the questionnaire was high (94.9%), thus the results can be considered representative of the population31. This survey could be considered innovative since it is the first to evaluate the performance of Hyflex CM rotary instruments after clinical use. Rotary instrumentation was found to be superior to hand instrumentation by the majority of the participants, which can be attributed to the fact that “speed” was suggested to be the greatest advantage of the former. The positive perception of the students about rotary techniques could be related with the very low degree of difficulty. Rotary training is not included in the preclinical courses of the undergraduate students and this justifies not only the lack of acquaintance with the geometrical traits and terms of rotary instruments but also the fact that the encountered
difficulty was mainly related to the students’ inexperience. Presumably due to their inexperience, most students were not able to identify the procedural errors of their cases. Instrument separation and ledge formation were the most easily recognizable errors. Those findings demonstrate the positive attitude of the undergraduate students toward rotary instrumentation and the necessity for rotary education in preclinical training.

On the basis of the results of the present study, the incidence of procedural errors by undergraduate students was considerably low, despite their inexperience in rotary instrumentation. The quality of the filled root canals was maintained to the highest standards. Rotary instrumentation was followed easily and received a positive feedback according to the participants’ responses.

Conclusions

The incidence of procedural errors was considerably low and the technical quality of the filled root canals was superior to that of similar studies. The responses of the questionnaire demonstrated a positive attitude toward rotary instrumentation.

References


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Efficiency of XP Endo Shaper (XPS) and Irrigation Protocol on the Quality of Cleaning the Apical Third of Root Canal: SEM Study

SUMMARY

**Background/Aim:** The aim of this study was to evaluate the efficacy of new rotary NiTi instrument XP-endo SHAPER (XPS) used with two irrigation protocols on the root canal cleaning in the apical area. **Material and Methods:** The research was conducted on 30 single-rooted teeth extracted for orthodontic reasons which were divided into the two groups. Instrumentation of the canals was conducted with XPS instrument and 2% solution of NaOCl was used as irrigant. Instrumentation in the first group was performed using a conventional continuous irrigation, in the second group, protocol of final irrigation was performed intermittently in 3 cycles. The SEM analysis of the apical third of the canal was performed on longitudinal root cross-section standardized photomicrography with a magnification of 2000X. **Results:** Results showed that a thicker smear layer was observed in the first group and with continuous irrigation protocol (2,10) in relation to the intermittent irrigation protocol in 3 cycles (1,96), but without significant differences. The walls of the root canal in the apical third of the samples of the second group were slightly cleaner (73.3%) in comparison with the teeth of the first group (64.7%), but also without significant differences. **Conclusions:** The use of XPS and 2% solution of NaOCl in the root canal enables efficient cleaning of the apical third of tooth. The final irrigation protocol in three cycles improves the efficiency of the smear layer removal in the apical segment of the canal.

Key words: XP-endo Shaper, Smear Layer, Irrigation Protocol, Apical Third

Introduction

Cleaning and shaping the root canal system is the most important phase in an endodontic treatment. However, complex anatomy of a canal often prevents adequate cleaning of this space using actual instruments and techniques. A particular problem is the irregularity of the canal system (isthmus, ramifications, and additional lateral canals) or apical third of the root canal, which cannot be reached by most of the instruments, and almost 30-50% of the surface of the canal walls remains untouched.

Mechanical instrumentation with manual or mechanical instruments leads to the formation of smear layer and dentin debris on the walls of the canal, and often to their accumulation in inaccessible areas of the canal system. In this way there is significant efficiency reduction of irrigant on the residual bacteria and significant linkage material disturbance for obturation of the canal walls.

In order to clean the root effectively, type and quantity of the irrigant or irrigation techniques and protocol are significant. These intracanal solutions provide a lubricating effect during instrumentation, have an antibacterial effect, remove debris and smear layer from the root canal walls and from inaccessible areas of the canal system.
The most frequently used endodontic irrigant is sodium hypochlorite (at various concentrations), primarily due to its antibacterial and solvent effect, although higher concentrations can cause irritation of the periapical tissue. Exceptional and prolonged antibacterial effect of chlorhexidine on the large number of bacteria in the canal, makes this irrigant often used in endodontic treatments. Researchers have also shown that it is possible to increase the efficiency of the irrigant, or to achieve better penetration and better cleaning of the canal walls using some forms of solution activation like ultrasound, a new instrument XP-endo Finisher or laser.

Chemo-mechanical procedure provides a significantly lower number of bacteria in the canal, but not complete disinfection of the canal system. In addition, any available set of manual or mechanical burnishing tool results in the formation of smear layer and dentin debridement on processed canal walls. For the success of this endodontic treatment, it is necessary to remove this layer, and the removal efficiency is significantly affected by irrigating solutions on the basis of the chelating agent, which effectively remove the smear layer from all areas of the canal.

The aim of this study was to evaluate the efficiency of the apical third of the canal after the instrumentation of the NiTi rotating instrument XP-endo Shaper and the application of two final irrigation protocols using SEM analysis.

**Material and Methods**

The study was conducted on 30 premolars extracted for orthodontic reasons. Teeth were stored up to the experiment in a 0.2% solution of thymol, at a temperature of 4 °C.

**Root Canal Instrumentation**

In all teeth the access cavity was formed and established initial passage with K-file #15. The working length is determined to be 1 mm shorter than the length at which the tip of the hand file appears on the apical foramen. To prevent leakage of the solution for irrigation during instrumentation, a wax ball was placed at the apex of each root.

The teeth were randomly divided into two groups (each of 15 teeth) and the complete mechanical instrumentation was performed by one researcher. As an irrigant, a 2% solution of NaOCl (Cloraxid 2%, Cerkamed, Poland) was used, and the canal was washed with plastic syringes, a volume of 2 ml, and needle size 27. A 4 ml solution for irrigation was used for each canal, and the flushing protocol lasted 150 sec.

The canal instrumentation was carried out in both groups by the new NiTi rotating instrument XP-endo Shaper (FKG, Dentaire, Swiss) (dimensions 30/04). This instrument represents a new generation of NiTi rotating instruments that, thanks to its extraordinary superelasticity, can change its shape in the canal and thus, reach inaccessible areas of the canal. A special production technique and specific geometry of the cutting part ensures the cleaning and shaping of the canal with only one instrument (800 rpm).

**GROUP 1** - In the first group, a conventional technique of continuous irrigation was applied. The canal is filled with an irrigant (0.5 ml) and then the XP instrument is inserted into the canal with gentle insertion and withdrawal placed to the working length (3-5 times for 30 sec). Then the irrigant (0.5 ml) was re-inserted into the canal and usage of XP provided the final apical preparation 8-10 times over 30 sec. The final irrigation with 2% NaOCl solution was performed after the completion of the instrumentation with another 3 ml solution for 90 sec.

**GROUP 2** - In the second group, XPS instrumentation was done in the same way as in the first group, but the final irrigation was performed intermittently in 3 steps (3 times 1 ml for 30 sec).

**SEM analysis**

After finishing the instrumentation, the crowns of all teeth were cut at the cement-enamel junction and then the roots were cut with diamond disc (without penetration into the canal) longitudinally in the vestibulo-oral direction and separated into two halves with sharp chisel. Obtained halves were placed on a carrier, gold-coated and analyzed by SEM (JOEL, JSM, 6460 LV, Japan).

Only an apical third (region 3 mm from the instrumentation border) was analyzed, so that, for each sample (half of the teeth) 5 standardized microphotographs were taken at 2000x magnification. SEM photographs of teeth samples (300 images) were analyzed by two researchers who independently rated each photo. In case of disagreement, it was discussed to reach a consensus. The evaluation of cleaning efficiency was based on qualitative estimation of residual smear layer in the apical segment of the canal with the criteria presented by Hülsmann et al.

**Grade 1** - no smear layer, dentinal tubules open,
**Grade 2** - small amount of smear layer, several dentinal tubules open,
**Grade 3** - homogenous smear layer covers the canal wall, small number of dentinal tubules open,
**Grade 4** - the entire wall of the canal covered with smear layer, no open dentinal tubules,
**Grade 5** - non-homogeneous smear layer covering the entire wall of the canal.
The clean wall canal included ratings 1 and 2, and the wall with the present smear layer grades 3, 4 and 5. The obtained results were processed in the SPSS 20 (IBM, CHICAGO) program. Methods of descriptive statistics and Mann Whitney test were used in statistical analysis.

Results

The results of the analysis of SEM photographs after the instrumentation and canal irrigation are shown in Tables 1 and 2 and in Figures 1 and 2. The analysis of the apical thirds samples indicated mainly clear canal walls, without the smear layer in both tested groups (Figure 1).

Samples of the first group where the technique of continuous irrigation (grade 2) was applied. SEM 2000x, B) Samples of the second group where the final irrigation was performed by intermittent technique in 3 steps (grade 1). SEM 2000x.

A slightly smear layer was registered in the first group where the instrumentation was performed using the XPS instrument and with the continual irrigation protocol (2,10) in comparison to the second group where the instrumentation was performed using XPS and an intermittent 3-step irrigation protocol (1,96), but without statistically significant differences (Table 1).

Table 1. Evaluation of smear layer in the apical third of the root canal

<table>
<thead>
<tr>
<th>Smeared layer ratings</th>
<th>N</th>
<th>Mean</th>
<th>SD</th>
<th>Med</th>
<th>Min</th>
<th>max</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Continuous irrigation</td>
<td>150</td>
<td>2.10</td>
<td>1.03</td>
<td>2.00</td>
<td>1.00</td>
<td>4.00</td>
</tr>
<tr>
<td>Irrigation in 3 steps</td>
<td>150</td>
<td>1.96</td>
<td>0.98</td>
<td>2.00</td>
<td>1.00</td>
<td>4.00</td>
</tr>
<tr>
<td>Total</td>
<td>300</td>
<td>2.03</td>
<td>1.00</td>
<td>2.00</td>
<td>1.00</td>
<td>4.00</td>
</tr>
</tbody>
</table>

The obtained results also indicated cleaner walls in the apical third of the samples of the second group (73,3%) compared to the canal walls of the first group (64,7%) (Figure 2, Table 2).

Table 2. Evaluation of cleaning quality in the apical third of the root canal

<table>
<thead>
<tr>
<th>Cleaning ratings</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Good</td>
<td>N</td>
<td>97</td>
<td>53</td>
<td>150</td>
<td></td>
</tr>
<tr>
<td>With smeared layer</td>
<td>%</td>
<td>64.7%</td>
<td>35.3%</td>
<td>100%</td>
<td></td>
</tr>
<tr>
<td>Group</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Continuous irrigation</td>
<td>N</td>
<td>110</td>
<td>40</td>
<td>150</td>
<td></td>
</tr>
<tr>
<td>Irrigation in 3 steps</td>
<td>%</td>
<td>73.3%</td>
<td>26.7%</td>
<td>100%</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>N</td>
<td>207</td>
<td>93</td>
<td>300</td>
<td></td>
</tr>
</tbody>
</table>

Discussion

Although there were earlier controversies, today there is a generally accepted consensus among endodontists about the necessity of removing the smear layer from the walls of the root canal. This layer significantly influences the success of endodontic treatment because it can contain bacteria and its presence can reduce the efficacy of intra canal medicaments, or reduce the adhesion of endodontic sealers to canal walls during obturation.

The possibilities of light microscopy in the debris and smear layer identification after chemo-mechanical canal preparation are quite high, but SEM analysis is certainly the most reliable and most popular method, primarily because of the possible magnification and high image resolution.

Studies have confirmed that the smear layer from the canal walls is easier to remove from the coronal and middle third, while the cleaning problem is particularly pronounced in the area of the apical third.
These research were realized by an identical protocol (all canals were processed by one researcher), with one type of instrument (XPS), with the same amount and duration of irrigation and with two final irrigation protocols (2% NaOCl). The obtained results indicated a very efficient cleaning of the apical segment of the canal and the walls mostly without a smear layer in both tested groups. A more efficient removal of the smear layer and better cleaning was observed after the intermittent final irrigation protocol in three steps in comparison to continuous irrigation protocol.

Clean canal walls in apical third of the nearly all samples could be explained primarily by the simplicity of canal morphology, but also by the effects of the new XPS instrument, or by its specific design, at a speed of 800 rpm, and by the fact that it can change its shape in the canal during the instrumentation and thus, reach the inaccessible canal areas. In addition, the extreme flexibility of the XPS and the working end with 6 cutting edges (with minimal torque) ensure efficient cleaning of all canal walls and the apical segment. The formed dentine micro debirs is easily removed due to the pronounced “turbulence” of irrigants during instrument rotation in the canal.

The application of XPS in the chemo – mechanical canal preparation provides the necessary and sufficient diameter of the apical preparation (30/04), which is also a precondition that facilitates cleaning of this part of the canal. Diameter of the apical preparation formed in this way allows the tip of the needle to easily reach the apical terminus, which also improves the efficiency of the irrigant in the removal of the smear layer. One of the problems of such deep application of irrigation needles in clinical conditions can be the conveyance of irrigant (NaOCl) into periapex, which can cause adverse effects on periapical structures.

The results comparison of various studies on the effects of cleansing the apical third is quite complicated because of both, the different techniques of instrumentation and irrigation, and different evaluation methods. In this study, a 2% NaOCl solution was used as an irrigant with two final irrigation protocols.

The largest number of researchers agree that the quality of the cleaning of the canal system depends largely on irrigants, its quantities, irrigation techniques and the time of exposure of the canal walls to the irrigants solution. The fact is that the solution for irrigation, quantity and time of action were identical in both groups, nonetheless, better results could be obtained primarily in the final irrigation protocol.

An intermittent final irrigation protocol (in 3 steps) has shown somewhat better results than the conventional irrigation protocol. During the conventional protocol, the irrigation solution is mixed with the remains of the smear layer and debris, which significantly reduces the efficiency, while the fresh solution during each cycle provides better canal cleaning. It has also been confirmed that 3 cycles of fresh NaOCl solution increase its cumulative efficiency as well as efficacy in canal cleaning quality.

A smaller amount of the smear layer in the apex part of the canal could be due to the fact that XPS due to the specific working part of the instrument and higher speeds during the canal treatment, leads to considerably less transport of the cut dentine into the apex part of the canal.

Efficient cleaning of the apical third and clean walls without smear layer were also observed after the application of the specific self-adjusting file (SAF-Self Adjusting File) in the canal instrumentation. The irrigation solution flows through the SAF file to provide a permanent freshness which is additionally activated by the movements of the file.

What is interesting in this study is the fact that the chemo-mechanical preparation of the canal was done with only one instrument and thus confirmed that the efficacy of cleaning does not depend on the number of used instruments, but above all from the diameter of apical preparation or from type, volume, concentration, and irrigation protocol.

The results of this study indicated that the apical segment of the canal was clean and without a smear layer in a high percentage, although only 2% NaOCl solution was used for irrigation. These findings are inconsistent with the findings of the authors who suggest that the removal of the smear layer from the canal walls is mainly dependent on the usage of chelating agents.

Conclusions

Within the limitations of this study, it can be concluded that the chemo-mechanical instrumentation of root canal using XPS and 2% NaOCl solution provides efficient cleaning of the apical canal segment, primarily due to the adequate diameter of the apex preparation and the specific effect of the new instrument during canal preparation. The intermittent final irrigation protocol in three cycles improves the removal of the smear layer in the apical part of the canal.

References


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Comparative Evaluation of Resistance to Cyclic Fatigue of Three Rotary Endodontic Ni-Ti Instruments

SUMMARY

Background/Aim: The present study examined the resistance to cyclic fatigue of three different rotary Ni-Ti instruments: K3XF (Kerr, Orange, CA), HyFlex CM (Coltene/Whaledent, Altstätten, Switzerland) and X7 EdgeFile (EdgeEndo, Albuquerque, New Mexico). Material and Methods: Thirty instruments (n=30) of each type were used with tip size 25 and 0.04 taper. All instruments were constrained to 60° of curvature with a radius of 5 mm by the use of two grooved stainless steel rods and rotated at a speed of 300 rpm and 3.0 Ncm of torque. The time until separation was recorded for each of the instruments and the number of cycles to fracture (NCF) was calculated. Statistical analysis was performed using R Programming language. Results: The X7 EdgeFile instrument showed significantly greater resistance to cyclic fatigue when compared to the HyFlex CM and the K3XF with mean NCF for each instrument 1046 ± 311, 707 ± 219 and 360 ± 96 respectively. HyFlex CM performed significantly better than K3XF. Conclusions: The X7 EdgeFile Ni-Ti file appears to be significantly more resistant to fracture, due to flexural fatigue, than the HyFlex CM and the K3XF.

Key words: Cyclic Fatigue, Nickel-Titanium Rotary Instruments, K3xf, Hyflex Cm, X7 Edgefile

Introduction

There are indisputable advantages in using nickel-titanium (Ni-Ti) rotary instruments1, resulting in their almost universal use among clinicians. However, during preparation, separation of these instruments can occur, exacerbating the difficulty of the case. Ni-Ti file separation is mostly associated with two phenomena; torsional failure and flexural fatigue of the instrument2–3. Torsional failure can occur due to the relatively low tensile strength of Ni-Ti alloy in comparison to stainless steel4. In this case, the jamming of the tip of the instrument in the root canal, while its shank continues to rotate, will lead to fracture when the torque applied by the handpiece exceeds the instrument’s torsional limit5. On the other hand, when a Ni-Ti instrument rotates within a curved canal, at any moment, the inner instrument surface is subjected to compression and the outer to tension. This will result in crack propagation and failure due to cyclic flexural fatigue6.

Advances in the metallurgy of Ni-Ti instruments have significantly improved the resistance to flexural fatigue7. In our study three different Ni-Ti instruments were used; K3XF (Kerr, Orange, CA), HyFlex CM (Coltene/Whaledent, Altstätten, Switzerland) and X7 EdgeFile (EdgeEndo, Albuquerque, New Mexico). K3XF files are the development of the earlier K3 files (Kerr, Orange, CA), maintaining the same design geometry, but now composed of R-phase heat treated Ni-Ti alloy7,8. The Ni-Ti alloy R-phase is an intermediate transformation phase with a rhombohedral crystalline structure between the austenite and martensite phases9. This crystalline structure is characterized by increased flexibility and reduced stresses on the instrument when rotating in curved canals, thus enhancing cyclic fatigue resistance10.

The EdgeFile is a relatively new rotary Ni-Ti file made of thermally treated nickel-titanium alloy, which...
The results were analyzed with the use of the R programming language. Data were analyzed for normal distribution and then statistical analysis was performed with independent samples t-test. The selected level of significance was 0.05.

Material and Methods

For this study, thirty rotary nickel-titanium instruments were used for each system (K3XF, HyFlex CM and EdgeFile X7). All instruments were of equal length (25 mm), tip size 25 and a constant 0.04 taper. To test the resistance to fracture of each instrument under continuous rotation, the following model (Figure 1) was constructed: Two grooved stainless steel rods with a diameter of 2 mm, were used to constrain the apical part of each instrument in a curvature of 60° and a radius of 5 mm (Figure 2), in accordance with previous research model. Each instrument was rotated at a constant speed of 300 rpm and 3 Ncm of torque with the use of an X-Smart endodontic motor handpiece (Dentsply-Maillefer, Ballaigues, Switzerland). The time of rotation until fracture for each instrument was recorded with the use of a VMS-001 USB microscope (Veho, Hampshire, UK) connected to a computer with an Ubuntu (Canonical Ltd, London, UK) Linux operating system and measured in seconds with the use of VLC media player software (Softonic International, S.A., Barcelona, Spain). All the instruments in this study were tested at room temperature. Finally, the number of cycles to fracture (NCF) was calculated according to the mathematical formula: Number of Cycles to Fracture = Time until separation (in seconds) * 300 (rpm) / 60.

In this study, no wear was observable in the rods, and this correlated with no progressive change in the time to fracture over the 90 tests.

Figure 1. The flexural fatigue testing model

Figure 2. Each instrument was constrained to rotate at a 600 angle and 5 mm radius of curvature

Results

All instruments separated within the curved part of the file. The analysis of the data confirmed normal distribution (Shapiro-Wilk normality test). The mean NCF for the K3XF, HyFlex CM and X7 EdgeFile instruments were 360 (± 96), 707 (± 219) and 1046 (± 311), respectively. The independent samples t-test showed a statistically significant difference (p<0.05) between the NCF of all instruments tested, hence the null hypothesis was rejected.

Discussion

The ideal test model for flexural fatigue in clinical use should be the human tooth. However, the root canal morphology would be altered after instrumentation, thus rendering the conditions of the study different for each instrument. Testing in different canals would encounter
the same problem. Therefore, it seems reasonable to test Ni-Ti instruments in vitro in order to investigate resistance to flexural fatigue. The testing rig constructed for our study was similar to that of Zinelis et al. Alterations included a 60° curvature according to Pruett with a radius of 5 mm and a higher rotational speed (300rpm). In our model, special care was taken to ensure that the different instruments were constricted in exactly the same position, which is not the case when a relatively wide (1.2 – 2 mm) metal tube is used to simulate the canal. In that case, the individual bending properties and cross section design of different files lead to differing positioning in the artificial canal. Some newer study models use artificial canals that follow the size and taper of the instrument at a given curvature. However, the superiority of one laboratory study model design over another is relevant when attempting to extrapolate in vitro results to indicate potential clinical performance. K3XF rotary Ni-Ti files are known to exhibit improved results when tested for flexural fatigue in comparison with its predecessor K3.

The results of our study showed that the X7 EdgeFile and Hyflex CM demonstrate greater resistance to flexural fatigue than K3XF. Earlier research has shown that files made from controlled memory Ni-Ti alloy are extremely flexible when compared with conventional superelastic Ni-Ti files. The specific mechanical properties of X7 EdgeFile and Hyflex CM could be a possible reason for their superiority to K3XF. The X7 EdgeFile instrument can be deformed by light pressure, the characteristic also found in Hyflex CM, which exists in a martensitic state in use. Due to their crystalline structure, HyFlex CM instruments, when deformed, partially or fully recover their original shape after sterilization. However, X7 EdgeFile instruments do not regain their original shape when heated above 125°C. That fact has led us to assume that the X7 EdgeFile instruments exhibit a martensite/austenite composition, with the former constituent being in a greater proportion. This fact could explain the superior performance of X7 EdgeFile over Hyflex CM in this study. Up to date, the specifics of the metallurgy of the two aforementioned instruments remain, as yet, unpublished, and therefore our assumptions remain unverified.

**Conclusions**

Under the conditions of this in vitro study, it can be concluded that the X7 EdgeFile Ni-Ti file is significantly less susceptible to fracture due to flexural fatigue than the HyFlex CM and the K3XF. The HyFlex CM appeared significantly less susceptible to fracture when compared to the K3XF.

**References**

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Modern Trends in Prosthetic Implant Rehabilitation of Patients: Case Report with 5-Year Follow-Up

SUMMARY

Background/Aim: Implant treatment expands extensively the possibilities of prosthetic treatment, which provide benefits, bigger comfort as well as general improvement of the patient’s life quality. In cases with no possibility of implantation, it is possible to improve conditions by using modern methods for bone tissue repair. One of factors important for the long-term success is proper oral hygiene, as well as raising awareness of its importance to patients. The aim of the paper is to present a patient rehabilitated with multiple implants and followed-up for a five-year period, and to point out the importance of raising patient’s awareness and motivation in order to preserve the results of the treatment. Case Report: A 31-year-old patient was admitted to the oral surgery clinic for rehabilitation of a poor oral health status. After taking history, clinical examination and additional analysis, the following treatment plan was suggested: to remove impacted upper canines and to put an implant supported by fixed prosthesis in the upper jaw, to make two implants supported by bridges laterally and one dental supported by bridge in the inter-canine sector in the lower jaw. The treatment was carried out in several stages that involved extraction of residual roots and impacted teeth, augmentation of bone defects with bone substitutes and bio-absorbable membranes, placing implant, and prosthetic rehabilitation. By verbal communication with the patient, we pointed out the importance of proper oral hygiene and regular check-ups. The five year follow-up showed the absence of factors that could adversely affect the success of the treatment, and the patient was still highly motivated to maintain proper oral hygiene.

Conclusions: It is possible to achieve predictable results in complex cases by using a multiphase prosthetic treatment supported by implants. Concerning a long-term success, motivation, proper information and patient’s willingness to cooperate play an important role.

Key words: Dental Implants, Motivation, Quality of Life, Rehabilitation

Introduction

Implant treatment extensively expands possibilities of prosthetic rehabilitation, which becomes more acceptable to the patient, unlike conventional prosthetics compensations that may be the cause of dissatisfaction, causing decline in quality of life\(^1\). The patients’ decision to accept suggestion of undergoing implant treatment is based on several factors, primarily on expectation of improving function and aesthetics, as well as financial aspect. For many patients, prosthetic rehabilitation with implants presents a definite need and directly affects quality of life, patient satisfaction and psychological state of mind\(^3\). However, in some cases, due to the absence of anatomical and morphological conditions (vertical and horizontal dimensions of the alveolar ridge and the proximity of anatomical structures such as the maxillary sinus in the maxilla and the mandibular canal in the mandible) implant rehabilitation may be carried out only by use of some additional procedures that modern treatment methods ensure\(^4\). The concept of
inter-canine region. After presentation of the treatment plan, the detailed briefing of the patient took place. The complexity of the process and the durability of the multi-phase care were pointed out, as well as factors that are important for the success of the therapy and of the possible complications. Education also included the aspect of the importance of an adequate oral care hygiene maintenance in patients with implants. The need of coming to regular follow-ups, and responsibility of the patient in the prevention of possible complications due to non-compliance with the proposed measures.

Guided bone regeneration, which involves the use of a bone substitutes and bio resorbative membranes, allows vertical and horizontal bone augmentation in combination with a different surgical approach. Moreover, it is well ascertained that implantation into bone substitute is possible, and that implants do not behave differently from implants embedded in natural bone.

Patient’s expectations are high and, therefore, as fixed prosthetic rehabilitation offers several advantages over rehabilitation with any kind of mobile prosthetic appliances, it should be a treatment of choice whenever it is possible. However, there are several problems with implant supported fixed prosthetic appliances, especially the presence of excess cement, which is difficult to be removed, or additional difficulties in maintaining adequate oral hygiene, especially if possibility of self-cleaning is heavy. Therefore, patient education in maintaining oral hygiene is necessary for long-term success whenever an implant supported fixed prosthetics is planned. If patient’s cooperation and motivation is not present, a plan of rehabilitation should be focused on conventional methods.

The aim of this paper is to present a patient rehabilitated with multiple implants and followed-up for a five-year period, and to point out the importance of raising patient’s awareness and motivation in preserving the results of the treatment.

**Case Report**

Patient BG, 31 years old, came to the Clinic of Oral Surgery, Medical Faculty, University of Pristina with headquarters in Kosovska Mitrovica. Except wish to improve his oral health from the aspect of aesthetics and function, patient didn’t have any local complaint. After further interviewing and examining the patient, an extremely unfavourable oral health status and high anxiety for dental treatment was found (after completing a questionnaire suggested by Humphries et al.). Verbal monitoring and motivational interviews have established willingness of the patient to cooperate, enabling further clinical examination and implementation of supplementary diagnostic procedures.

Clinical examination revealed a lack of several teeth in the upper jaw and the advanced caries with extensive destruction of hard dental tissues at the rest of upper teeth (Figure 1), with multiple periapical lesions and the presence of both impacted upper canines. In the lower jaw, several teeth were missing or being carious.

Several options of prosthetic rehabilitation were offered to the patient. One of the options was an implant-supported fixed circular bridge in the upper jaw, two implant-supported bridges in both lateral regions of the lower jaw, and one dental-supported bridge in the inter-canine region. After presentation of the treatment plan, the detailed briefing of the patient took place. The complexity of the process and the durability of the multi-phase care were pointed out, as well as factors that are important for the success of the therapy and of the possible complications. Education also included the aspect of the importance of an adequate oral care hygiene maintenance in patients with implants. The need of coming to regular follow-ups, and responsibility of the patient in the prevention of possible complications due to non-compliance with the proposed measures.
Surgical Aspect of Implant-Prosthetic Treatment

A two-phase surgical protocol of implantation comprised implantation and covering the implant in the first phase. Three Bredent “Blue Sky” implants were installed at the right side of the maxilla in the region of teeth #13, #14, and #15, with dimensions 4.0 x 12mm, 4.0 x 12mm, and 3.5 x 12mm, while at the left side, three implants of the same dimensions were put in the region of teeth #24, #25 and #26.

In the lower jaw, on the left side, we installed two implants in the region of teeth #34 and #35 (Bredent Blue Sky, dimensions 3.5x12 and 4.0x10). On the right side, in the region of teeth #43 and #45, we also installed two implants (3.5x10 and 4.0x10). In the prosthetic phase, this arrangement would allow a production of two lateral implant-supported bridges and a dentally supported bridge in the inter-canine sector.

After a three-month period of osseointegration (meanwhile, the patient worn a mobile prosthetic device), a second surgical phase was carried out, which comprised detection of implants and setting of gingival-formers for a period of another two weeks to form a proper gingival profile.

Prosthetic Aspect of the Treatment

Firstly, a single-phase print was taken and implant transfers placed, which were spliced to increase the stability and accuracy during the printouts (Figure 3). A re-registration of vertical relations was also performed due to the loss of the vertical dimension of occlusion. After that, a laboratory phase of the metal construction followed and after clinical testing, the laboratory phase was finished by placing ceramics. For a period of one month, the patient was wearing temporarily cemented compensation in order to make any corrections that might reconcile to expectations of the patient concerning to colour, shape, size and other characteristics of the prosthesis. Definitive cementation of bridges was performed by glass-ionomer cement (Figure 4).

Maintaining Oral Hygiene

Measures related to the maintenance of oral hygiene were focused on mechanical and chemical control of dental plaque and food residues accumulation. The proposed methods of mechanical control included the use of soft toothbrushes, less abrasive tooth paste, interdental brushes and stimulants (“soft pick”), and an interdental aids adapted for patients with implants (“Superfloss”). Also, the patient was proposed to mechanically remove food residues by using water irrigation (“Water-pick” machine). For chemical control of dental plaque accumulation, non-alcoholic mouthwashes (0.12% chlorhexidine gluconate) were proposed, two to three times per day for a period of one month with pauses to avoid negative effects of the solution.

Follow-ups

At the follow-up six months after the submission of definitive prosthetic work, it was visible that the patient adhered to advices related to the maintenance of oral hygiene, and that the functionality and aesthetics were favourable, further motivating the patient to maintain a long-term advantageous result.

At the follow-up after one year, the gingiva did not show visual signs of inflammation. A plaque accumulation around implants was under control; clinical probing of the sulcus depth did not show noticeable resorption of bone tissue around the implants, and there was no bleeding on probing. By checking the occlusion, it was found that the occlusion was balanced, without traumatic contacts. Panoramic radiograms did not show signs of resorption of the peri-implant bone. The patient was satisfied with the result and expressed a high degree of motivation for further maintaining oral hygiene.

Five years after the treatment, everything was satisfactory (Figure 5). Further education on maintaining oral hygiene of the patient was not necessary since he adhered to everything that was proposed in the previous period. An analysis of panoramic radiogram revealed an absence of pathological bone resorption around implants (Figure 6).
preserve and augment the residual alveolar ridge by bone substitutes in combination with membranes, as bone defect was limited by bone tissue; in the case of major defects a solid non-corrosive membranes commonly made of titanium might be used to prevent deformation of the alveolar ridge. Studies have shown that implantation into augmented bone can give predictive results as implantation into natural bone, depending on bone region and density. Although several studies stressed a need for additional research, it seems that the use of bone substitutes in combination with bio-resorptive membranes can serve as an alternative to conventional protocols of implant insertion.

Several studies have also shown the importance of interlinking implants with fixed prosthetic works because due to the mode of force distribution behaves as a unique functional unit. Concerning rehabilitation with implant supported fixed prostheses, in addition to fulfilling local clinical conditions needed for a long-term favourable result, a proper cooperation of the patient is absolutely necessary, as well as motivation and raising awareness of the importance of maintaining oral hygiene. Several authors state that the prevention of late complications in the form of peri-implant mucositis and periimplantitis but also the prevention of pathological resorption of peri-implant bone tissue is the matter of respecting biomechanical principles, balancing occlusal forces and maintaining adequate oral hygiene.

The fact that cemented fixed prosthesis on implants cannot be removed during professional office maintenance, the maintenance of oral hygiene is a priority and a very important segment of preserving the health of peri-implant tissue. Therefore, more attention have been paid to the patient’s education concerning dental plaque accumulation, influence of pathogenic agents from bio film on peri-implant tissue, complications that can arise due to failure to maintain adequate oral hygiene, and the way in which the formation of deposits can be controlled.

Conclusions

It is possible to achieve predictable results in complex cases by using a multiphase prosthesis treatment supported by implants. Concerning a long-term success, motivation, proper information and patient’s willingness to cooperate play an important role.

References


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Mini-Implants and Zirconium Crowns in Treating Congenitally Missing Maxillary Lateral Incisors: Case Report

Introduction

Maxillary lateral incisors are the second most common missing teeth. Being located in the esthetic zone, congenitally missing laterals represent a challenging case for dentists as they affect the esthetics and general appearance of the patients. There are several options to treat these patients and the selection of the option depends on several factors. The space can either be closed with canine substitution for the lateral incisor or it can be opened for prosthetic replacement of the upper lateral incisor. Several options are available for prosthetic replacement including: resin bonded bridge, cantilever bridge, conventional fixed partial denture, or single crown implant restoration. The choice among these factors depends on occlusal relationship and other occlusal disharmonies, over-jet and over-bite, canine size and shape, condition of adjacent teeth, as well as patient’s desire and expectations.

Mini-implants have been reported intensively in literature with a long term high success rates to support both fixed and removable prosthesis. They can solve problem of reduced bone width in regions with anatomical limitations.

In the following case report, a space between the central incisors and canines was created orthodontically to restore the maxillary lateral incisors with single zirconium crowns on two mini-implants.

Case Report

A young female patient presented to the outpatient clinic at the Faculty of dentistry, Cairo University, with a chief complain of unpleasant appearance and spacing between upper anterior teeth. A thorough examination was performed including extra-oral and intra-oral examinations as well as panoramic X-ray. The patient had congenitally missing upper lateral incisors bilaterally (Figures 1-4).
was carefully examined. The patient had Angle’s class I molar relationship, with minor occlusal disharmonies. A multi-disciplinary approach combining both orthodontist and prosthodontist was required to restore the esthetics and function. The chosen treatment was space creation by orthodontic movement of the teeth, followed by mini-implant placement to support cemented zirconium crowns for optimum esthetics.

Treatment Planning

Upper and lower primary impressions were taken to obtain a diagnostic casts. This was followed by a face-bow (Bio-Art Equipamentos Odontológicos Ltda) record to mount the upper cast on a semi-adjustable articulator (Bio-Art Equipamentos Odontológicos Ltda), and a diagnostic bite to mount the lower one. Occlusion
Orthodontic phase

It lasted for about a year and a half. Distalization of the canines bilaterally was performed till a space of 6 mm was created to accommodate a lateral incisor in harmony with the patient’s dentition.

Surgical phase

A cone beam computed tomography (CBCT) was performed to determine the available width and height of bone for proper implant size selection. Two-piece mini dental implants were used as there was no enough bone width for regular implant placement. A size 3mm x 12mm was chosen to restore left lateral incisor and 3.3mm x 12 mm for the right one.

After administration of local anesthesia with 2.2 ml of Mepivacaine (Scandonest 2%, Mepivacaine HCl, USP) as a labial and palatal infiltration, a crestal incision and sulcular one were made around the central incisor and the canine using Bard Parker blade no. 15 (Hu-Friedy Mfg. Co., LLC). This was followed by a full thickness flap elevation and reflection with a sharp mucoperiosteal elevator. The implant osteotomy was created using a reduction hand piece (1:16) at a speed of 1200 rpm with a physio-dispenser and adequate flow of sterile saline solution. The implant was initially placed manually then continued with a ratchet till the implant flushed with the bone surface. The flap edges were repositioned and sutured. Patient was instructed to apply cold fomentation for 12 h post-surgically, take an anti-inflammatory drug (Cataflam® Novartis Pharma, S.A.E. Cairo-under license from Novartis Pharma AG., Basle, Switzerland) 3 times for 2 days, as well as broad spectrum antibiotic for 5 days and to follow the usual oral hygiene measures in combination with Chlorhexidine mouth wash (Antiseptol mouth wash; Chlohexidine gluconate, Kahira Pharmaceuticals) three to four times daily for 2 weeks. The patient was recalled after one week to remove the sutures.

Prosthetic phase

Implants were allowed to osseointegrate for a period of 3 months. After that, they were exposed by a crestal incision and healing collars were installed. They were left in place for 2 weeks to allow for mucosal healing (Figure 5), after which closed tray coping transfers were attached to the implants and their proper seating was verified with periapical X-rays (Figure 6). An implant level impression was made. Impression transfers were attached to fixture analogues and re-inserted into their place in the impression (Figure 7). The use of 2-piece implant system allowed for proper abutment selection for optimum esthetic results. Angled abutments were chosen to allow for correction of the axial inclination of the implants dictated by the available bone. They had titanium nitride coated collars. Impression was sent to the lab where a master cast with tissue mimic was obtained to construct the zirconium crowns (Figures 8 and 9). First, the lab sent zirconium cores for try-in. Then, they were built into full crowns. Abutments were screwed to the implant at a torque of 30 N/cm. Implant protected occlusion was assured to allow for long term success. Crowns were cemented temporarily to allow any further adjustments. After crowns installation, the patient was completely satisfied with the results (Figures 10 and 11).
malocclusion. Moreover, distalization of permanent canines to their proper positions might help in alveolar ridge development in the lateral incisor region. This also would enhance the final esthetic results rather than canine substitution for the lateral incisor. Besides, having the permanent canine in place would help to obtain a stable final occlusion.

The space required for maxillary lateral incisor is about 5-7 mm. The space created orthodontic treatment was about 6 mm bilaterally, which was sufficient for restoring lateral incisors. Since she was an adult patient, there was no problem for implant placement because growth of the maxilla was completed.

CBCT was performed to determine the needed implants’ dimensions. There was deficiency in the labio-palatal dimension. This was overcome by using mini-implants, which contribute the avoidance of additional bone grafting surgery. Mini-implants can be supplied either as single piece or two-piece. Although single piece implants have the advantage of strength and simple restorative procedures, they provide little flexibility in abutment angulation and customization to meet the esthetic requirements. Implants were placed with axial inclination labially to avoid labial fenestration, which was corrected later on using angled abutments. The use

**Discussion**

The chosen treatment plan for this patient was to open the space where the patient had a molar class I relationship and had no concomitant needs to treat
of two-piece implants allowed for the correction of the axial inclination with angulated abutment. The abutments had a titanium nitride coated collar giving a gold hue which allowed for optimal esthetics. Zirconium crowns were made to restore the lateral incisors which enhanced the final esthetic results. Implant protected occlusion is important to avoid un-necessary forces falling on the implants predicting a long-term success.

Conclusions

Oral rehabilitation of patients with congenitally missing lateral incisors can be achieved with orthodontic space opening combined with implant placement to support single crowns when other conditions permit. Mini-implants can be used successfully to over-come problem of insufficient bone.

References


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Papillon-Lefévre Syndrome:
Case Report and Genetic Analysis

SUMMARY

Background/Aim: Papillon Lefévre syndrome is a rare autosomal recessive genodermatosis. The characteristic findings of the disease are early loss of primary and permanent teeth and palmoplantar keratoderma. Notwithstanding that many etiologic factors like genetic mutations, bacterial agents, immunologic changes have been identified, the pathogenesis has not been fully understood. Although dentists play an important role in the diagnosis and treatment of Papillon Lefèvre syndrome, it is appropriate to treat the disease with a multidisciplinary approach. Case Report: In this case report, the clinical, radiological and genetic examination of the patient with Papillon Lefèvre syndrome who has a homozygous mutation in the CTSC gene will be presented. Conclusions: Dentists should have knowledge about treatment management of these patients. Teeth can be preserved longer with early diagnosis and appropriate treatment of the disease.

Key words: Papillon Lefèvre, Cathepsin C, Gene Mutation

Introduction

The syndrome was first described by Papillon and Lefèvre in 1924. The disease is characterized by diffuse palmoplantar keratoderma and prematurely starting aggressive periodontitis affecting both dentitions. It has an autosomal recessive inheritance pattern. The patients have cathepsin C (CTSC) gene mutation. The symptoms that may accompany are hyperhidrosis, aracnodactilia, intracranial calcification, tendency to infection and mental retardation. The incidence of the disease has been reported to be 1 to 4 in a million. The incidence is higher in societies where consanguineous marriages are common. Girls and boys are affected equally by the disease.

Cutaneous lesions usually begin to appear along with oral findings between 6 months to 4 years of age. Cutaneous lesions have been considered to develop due to the disorders of ectodermal and mesodermal components. Cutaneous lesions are seen in Papillon Lefèvre syndrome since CTSC gene is expressed in epithelial tissues. Hyperkeratosis and erythema are present in palms and soles. Keratinization may spread to the dorsum of hands and feet. Hyperkeratotic plaques developed might be associated with hyperhidrosis of the hands and feet and may end up with foulodor. The lesions may appear as patches or deep fissures with different colors and appearances. Eyelids, cheeks, knees and labial commissure might also be involved. The patients report that their complaints increase in cold weather. Horizontal growth and fissure formation might be seen in nails in advanced cases.

Vertical bone loss in first molars is seen in localized forms of PLS as radiographic findings, while this bone loss may include all teeth in the generalized form. “Floating in air” image might develop in radiography since bone support of the teeth is completely lost in very advanced stages.

Some cases are reported to have microdonti, root eruption and deterioration in root formation although there is no change in the form and time of tooth eruption of primary teeth. Upon eruption of primary teeth, an inflammatory picture begins in gingiva. Gingiva is hyperemic and edematous. Flow of pus from periodontal pockets is seen. After premature loss of primary teeth, gingiva returns to normal. The process repeats in conjunction with the eruption of permanent teeth. The patients with Papillon Lefèvre syndrome usually remain toothless at age of 14-15. Alveolar bone resorption is observed in radiographic assessment. Wisdom teeth are usually not affected by the disease.
Due to constant use of the same prosthesis during growth and development period and being toothless, there was inadequate development of jaws in horizontal and vertical planes. The vertical dimension decreased and the denture face appearance occurred in the patient. The success rate and cost of implant therapy were explained and the patient was notified that additional surgical procedures may be required. Renewal of the prosthesis was offered to the patient as the second treatment option. The patient rejected implant therapy since it has high cost and is an invasive treatment and he wanted his prosthesis to be renewed.

Discussion

Although the etiology of Papillon Lefèvre syndrome is not fully understood, the syndrome has been associated with cathepsin C gene mutation, various microbial agents and immunologic factors. Dipeptide peptidase 1, also known as cathepsin C, is a lysosomal cysteine protease and is encoded by the cathepsin C gene which is located on the 11q14.1-q14.3 chromosome. Seventy-five different mutations related to the CTSC gene have been described. Among the mutations, 75% are homozygous and among the homozygous mutations, 50% are lost mutation, 25% are meaningless mutation, 23% are frameshift mutation and 2% are other type mutations. Mutation of c.415 G>A, seen in this presented case was defined first by Zhang et al. as heterozygous in a PLS patient with Caucasian origin. Cases carrying homozygous c.415 G>A mutation have also been reported. CTSC, CTSG and elastase functions are almost completely lost in homozygous mutations.

Cathepsin C plays an important role in the activation of cytotoxic T lymphocytes, natural killer cells, mast cells and serine proteases in neutrophils. It also functions in collagen type I, III, IV and fibronectin degradation. In Papillon Lefève syndrome, as a result of the inactivation of serine proteases in inflammatory cells due to mutations in the CTCS gene, impairment occurs in the immune system. The periodontal disease and the tendency to infection are caused by the impairment of neutrophil, T lymphocyte and B lymphocyte functions. The first immune dysfunction developing in PLS is the impairment of the cytotoxic functions of natural killer cells.

Aggregatibacter Actinomycetemcomitans, Porphyromonas gingivalis, Fusobacterium nucleatum and Prevotella intermedia are the bacteria which are present in high numbers in periodontal pockets of the patients with Papillon Lefèvre syndrome and therefore they are held responsible for the pathogenesis of the disease. High levels of immunoglobulin against Aggregatibacter Actinomycetemcomitans have been
observed in individuals affected by the disease. In Papillon Lefèvre syndrome, there is a decrease in the neutrophil response to Staphylococcus spp. and Aggregatibacter Actinomycetemcomitans. Herpes viruses in addition to the pathogenic bacteria including Aggregatibacter Actinomycetemcomitans and also impairment of the host immune response are considered to play role in periodontitis developing in patients with Papillon Lefèvre syndrome.

There is no specific histopathological finding associated with Papillon Lefèvre syndrome. Hyperkeratosis, acanthosis, hypergranulosis or psoriasiform hyperplasia might be seen in gingival epithelium. Intense inflammatory infiltration in patients with PLS is observed in the subepithelial connective tissues of the periodontal tissues. Dominant cells of this inflammation are the plasma cells.

Dental treatment of patients with Papillon Lefèvre is challenging for both the patient and the dentist since the prognosis is poor and outcome is unpredictable. The main goal of the periodontal treatment in Papillon Lefèvre syndrome is to optimize oral hygiene and keep the teeth in the mouth as long as possible. For this purpose, oral hygiene education is given, scaling and root planning is performed and oral rinse with 0.2% chlorhexidine is recommended. Antibiotics may be used in the presence of active periodontitis. The most commonly used antibiotics for this purpose are erythromycin and tetracycline. There are also studies reporting that amoxicillin metronidazole or amoxicillin clavulanic acid combinations result in success. Some investigators defend the idea that cleaning the oral cavity from pathogen bacteria and then the tooth eruption of the permanent teeth uninfluenced from the infection increases the duration of permanent teeth stay in mouth. For the treatment of cutaneous lesions, retinoids, salicylic acid and steroid are applied. Use of retinoid was reported to increase alveolar bone height and periodontal attachment level, reduce periodontitis formation and improve cutaneous lesions.

There are studies reporting that the use of dental implants result in success for prosthetic rehabilitation of PLS patients. Senel et al. followed a PLS patient, to whom they applied implant therapy, for 3 years and reported successful results.

The role of the dentist is very important in the diagnosis and treatment of PLS. Duration of stay of the teeth in mouth can be prolonged when the disease is treated appropriately. This situation is important for the preservation of maxillary and mandibular bone height. Unfortunately, it was late for these treatment options when the patient was admitted to us. The most appropriate treatment options were presented to the patient and the decision was left to him. He was informed about his disease and possible complications were explained.

**Conclusions**

Dentists should have knowledge about treatment management of these patients. Teeth can be preserved longer with early diagnosis and appropriate treatment of the disease.

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Summary and key words

The second page should carry a structured abstract (250-300 words for original articles and meta-analyses) with the title of the article. In short, clear sentences the authors should write the Background/Aim, major procedures – Materials and Methods (choice of subjects or laboratory animals; methods for observation and analysis), the obtained findings – Results (concrete data and their statistical significance), and the Conclusion. It should emphasize new and important aspects of the study or observations. A structured abstract for case reports (up to 250 words) should contain subtitles Introduction, Case report, Conclusion. Below the abstract Key words should provide 3–10 key words or short phrases that indicate the topic of the article according to Index Medicus.

Text

The text of the articles includes: Introduction, Material and Methods, Results, and Discussion.

Introduction. After the introductory notes, the aim of the article should be stated in brief (the reasons for the study or observation), only significant data from the literature, but not extensive, detailed consideration of the subject, nor data or conclusions from the work being reported.

Material and Methods. The selection of study or experimental subjects (patients or experimental animals, including controls) should be clearly described. The methods, apparatus (manufacturer’s name and address in parentheses), and procedures should be identified in sufficient detail to allow other workers to reproduce the results. Also, give references to established methods, including statistical methods. State the approval of the Ethics Committee for the tests in humans and animals.

Results should be presented in logical sequence in the text, tables and illustrations. Emphasize or summarize only important observations.

Discussion is to emphasize the new and significant aspects of the study and the conclusions that result from them. Relate the observations to other relevant studies. Link
the conclusions with the goals of the study, but avoid unqualified statements and conclusions not completely supported by your data.

References

References should be numbered consecutively in the order in which they appear in the text. All references are identified, whether they appear in the text, tables, or legends, by Arabic numbers in superscript. Two references are cited separated by a comma, with no space. Three or more consecutive references are given as a range. The use of abstracts, secondary publications, oral communications, official and classified documents as references is strongly discouraged. Manuscripts accepted for publication may be cited and should include the manuscript’s DOI, if known. Material submitted, but not yet accepted for publication, should not be included in the reference list, but may be included in the text and referred to as "unpublished data" giving the names of the involved researchers. Obtaining permission to quote unpublished data from the cited colleagues is the responsibility of the author.

The list of references should only include works that are cited in the text. The style and punctuation of references should strictly conform to The National Library of Medicine Style Guide for Authors, Editors, and Publishers (NLM Style).


Several examples of references in NLM format are shown below.

For more samples of formatted references visit the NLM site (http://www.nlm.nih.gov/bsd/uniform_requirements.html).

Examples of references:

Journal title abbreviations should be those used by the U.S. National Library of Medicine. If you are uncertain about the correct abbreviation for a journal title, please search for the journal at http://www.ncbi.nlm.nih.gov/nlmcatalog.

Authors may wish to make use of reference management software to ensure that reference lists are correctly formatted.

Tables

Each table should be typed single-spaced, numbered in the order of their first citation in the text and supplied with a brief title each. Explanatory notes are printed under a table. Each table should be placed within the text at the appropriate points, rather than at the end. If data from another source are used, acknowledge fully.

Illustrations

All illustrations, labelled as figures (such as photographs, line drawings, charts or tracings) should be submitted as high-contrast prints, suitable for publications. Illustrations should have a final resolution of 300 dpi, and line drawings of 800-1200 dpi. They must be numbered with Arabic numerals in the same order as they are cited in the text. Photomicrographs should have the magnifications and details of staining techniques shown. Short explanatory captions of all illustrations should be typed on a separate sheet.

Abbreviations and symbols

Use only standard abbreviations. Avoid abbreviations in the title and abstracts. The full term for which an abbreviation stands should precede its first use in the text.

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