# Dental and Medical Problems

QUARTERLY ISSN 1644-387X (PRINT) ISSN 2300-9020 (ONLINE)

www.dmp.umed.wroc.pl

# 2018, Vol. 55, No. 1 (January–March)

Ministry of Science and Higher Education – 11 pts. Index Copernicus (ICV) – 113.75 pts.



# Dental and Medical Problems

#### ISSN 1644-387X (PRINT)

QUARTERLY 2018, Vol. 55, No. 1 (January–March)

#### **Address of Editorial Office**

Marcinkowskiego 2–6 50-368 Wrocław, Poland Tel.: +48 71 784 11 33 E-mail: dental@umed.wroc.pl

#### **Publisher**

Wroclaw Medical University Wybrzeże L. Pasteura 1 50-367 Wrocław, Poland

Polish Dental Society Montelupich 4 31–155 Kraków, Poland

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Online edition is the original version of the journal

#### ISSN 2300-9020 (ONLINE

www.dmp.umed.wroc.pl

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Indexed in: Scopus, DOAJ, GFMER, Index Copernicus, Ministry od Science and Higher Education, CiteFactor, WorldCat, Directory od Research Journal Indexing

This publication has been co-financed by the Ministry of Science and Higher Education

Typographic design: Monika Kolęda, Piotr Gil Cover: Monika Kolęda DTP: Wroclaw Medical University Press Printing and binding: EXDRUK

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QUARTERLY 2018, Vol. 55, No. 1 (January-March)

ISSN 1644-387X (PRINT) ISSN 2300-9020 (ONLINE) www.dmp.umed.wroc.pl

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# Acknowledgements

Editors of Dental and Medical Problems would like to take this opportunity to thank the following people for their long-lasting contribution to the journal and for their work on the Editorial Board:

- Prof. Leszek Paradowski initiator of changing the character of Dental and Medical Problems into more scientific journal in 2002 and Editorial Board member in 2005–2017;
- Prof. Jerzy Sokołowski Editorial Board member in 2016–2017 and the Thematic Editor for Dental Materials in 2016–2017;
- Dr hab. Jerzy Sokalski Editorial Board member in 2006-2017;
- Prof. Maria Chomyszyn-Gajewska Editorial Board member in 2009-2017;
- Prof. Jakub Gołąb Editorial Board member in 2010–2017;
- Prof. Krzysztof Woźniak Editorial Board member in 2011–2017;
- Dr Anna Paradowska-Stolarz Editorial Secretary in 2009–2017.

## **New Members of the Editorial Board**



#### Monika Łukomska-Szymańska

Professor Łukomska-Szymańska is an associate professor at the Chair of General Dentistry, Division of Dentistry, Medical University of Lodz, Poland. She received PhD with honours in field of diagnostic method of premature contacts, as well as Rector's Award for doctoral dissertation. Her habilitation thesis entitled "The evaluation of antibacterial properties of different bonding systems modified with antibacterial agents" was defended in 2013. She actively participates in congresses and publishes scientific works (180 scientific articles and conference communication till March 2018).

Professor Łukomska-Szymańska is a specialist in prosthodontics and restorative dentistry with endodontics. She developed her skills in Germany, Spain and USA. Her main fields of scientific and professional experience are dental materials, diagnosis and treatment of TMJ disorders, as well as microscopic endodontics and adhesive prosthodontics, with focus on interdisciplinary approach.

Professor Monika Łukomska-Szymańska is a Vice-Dean for Dental Education at Faculty of Medicine, Medical University of Lodz. She has created and supervised educational programme in ergonomics, dental materials, and integrated adult dentistry. Her scientific and teaching activity also encompasses education of young researchers (including PhD programme) and lecturers. She has lectured in English at Medical University of Lodz for 15 years. She carries out postgraduate training courses and lectures for dentists.

She has been a reviewer of many foreign and Polish scientific journals from ISI Master Journal List, as well as an active member of the Polish Dental Association, Polish Biomedical Engineering Association (Vice-President) and Association for Medical-Dental Education in Poland "SAPIENTIA".



#### **Piotr Fudalej**

Professor Piotr Fudalej studied dentistry at the Warsaw Medical University, Poland, and biology at University of Warsaw, Poland. He completed a postgraduate orthodontic program at the University of Washington, Seattle, Washington. He has a PhD degree from Radboud University Medical Center, Nijmegen, the Netherlands. Currently, he is affiliated with Palacký University, Olomouc, Czech Republic, and University of Bern, Switzerland. His main research areas are effects of orthodontic treatment of periodontal health and cleft lip and palate. Both research lines have been conducted in collaboration with Radboud University Medical Center. He received the 2010 Samuel Berkowitz Long-Term Outcomes Study Award for the best long-term outcomes study published in 2009 in the Cleft Palate Craniofacial Journal. Prof. Fudalej published more than 80 scientific papers in peer-reviewed journals.



#### Marcin Mikulewicz

Professor Marcin Mikulewicz graduated from Faculty of Dentistry, Wroclaw Medical University, Poland. He obtained PhD degree at the same university, and habilitation at Collegium Medicum, Jagiellonian University, Krakow, Poland. He has completed the orthodontic postgraduate program, earning title of a specialist. As an associate professor, he is the Head of Division of Facial Abnormalities at the Department of Dentofacial Orthopaedics and Orthodontics, Wroclaw Medical University. Professor Mikulewicz is involved in National Program for Care of Cleft Patients.

Professor Mikulewicz's main research interests are evaluation of biocompatibility and development of dental biomaterials, as well as orthodontic treatment of cleft patients. He is an expert for Polish National Centre for Research and Development, holds 3 patents, and has published 94 research papers and 7 book chapters, which have been cited more than 300 times. Prof. Mikulewicz is a member of European Orthodontic Association, Polish Orthodontic Society, Italian Orthodontic Society, and Polish Dental Association.

# Alveolar process changes associated with administration of nucleos(t)ide analogue (sofosbuvir) in rat model

# Zmiany wyrostka zębodołowego związane z zastosowaniem analogu nukleozydów (sofosbuwiru) w badaniu u szczurów

Dina Badawy Elsayed Farag<sup>A–D,F</sup>, Dalia Abdel-Hameed El-Baz<sup>A,C,D,F</sup>, Zeinab Amin Salem<sup>A,B,E,F</sup>

Department of Oral Biology, Faculty of Dentistry, Cairo University, Egypt

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):11-16

Address for correspondence Zeinab A. Salem E-mail: abodarwish2005@yahoo.com

Funding sources None declared

Conflict of interest None declared

Received on October 14, 2017 Reviewed on November 17, 2017 Accepted on December 27, 2017

**DOI** 10.17219/dmp/81592

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#### Abstract

**Background.** Sofosbuvir is a nucleotide compound that has proved to be among the most potent orally available antiviral treatments. Infrequent but serious adverse events have been reported with the use of oral nucleos(t)ide analogues.

**Objectives.** Investigating sofosbuvir-induced alterations in the rat mandibular alveolar process.

**Material and methods.** In the study, 30 adult male albino rats were used and divided randomly into following groups: group 1, group 2 and group 3 (10 rats per group). Group 1 served as a control, group 2 received sofosbuvir through oral gavage at a dose of 40 mg/kg/day for 6 weeks, and group 3 was similar to group 2 but received sofosbuvir for 12 weeks. The animals were sacrificed at the end of the experiment. The mandibles were dissected and examined histologically as well as by scanning electron microscope (SEM) and energy dispersive X-ray unit (EDX).

**Results.** Histologically, group 1 showed normal alveolar process. In group 2, histopathological changes occurred as bone trabeculae demonstrated obvious Howship's lacuna of osteoclasts. In group 3, bone trabeculae exhibited multiple degenerated areas as well as apparent vacuolization. Scanning electron microscopic examination revealed smooth alveolar bone architecture in group 1. On the other hand, groups 2 and 3 demonstrated irregular bone architecture with formation of multiple pores. EDX analysis demonstrated the highest calcium concentration in the control group, while the lowest was found in group 3. Statistical analysis of the EDX results revealed a statistically significant difference among the studied groups as the p-value was <0.01.

**Conclusions.** It has been concluded that sofosbuvir induced apparent alterations in the rats' alveolar bone. This effect was exaggerated in a longer period of drug administration. The sofosbuvir-induced alterations might be attributed mainly to mitochondrial toxicity. The effect had been clearly shown histologically and morphologically as well as in bone mineral (calcium) content.

**Key words:** hepatitis C virus, alveolar bone, sofosbuvir, nucleos(t)ide analogues, scanning microscopy

Słowa kluczowe: wirus zapalenia wątroby typu C, kość wyrostka zębodołowego, sofosbuwir, analogi nukleozydów, mikroskopia skaningowa

#### Introduction

Nucleos(t)ide inhibitors are analogues of the naturally occurring polymerase substrates. They form the basis of antiviral therapy for a number of chronic viral diseases such as herpes simplex virus (HSV), human immunodeficiency virus (HIV), and hepatitis B virus (HBV).<sup>1,2</sup> Nucleos(t)ides analogues resemble physiological nucleos(t)ides in terms of uptake and metabolism, and are incorporated into newly synthesized DNA. The incorporation of these compounds into DNA may induce either the termination of chain elongation or the accumulation of mutations in viral offspring. Some of these drugs also inhibit key enzymes involved in the generation of the purine and pyrimidine nucleotides and RNA synthesis, and directly activate caspase cascade. All of these effects may lead to cell death.<sup>3,4</sup>

Infrequent but serious adverse events have been reported with the use of oral nucleos(t)ide analogues. All nucleos(t)ide analogues have a "black box warning" regarding potential mitochondrial toxicity in their product labeling. The differences in analogue toxicity might result from a combination of cellular uptake, organellar transport, metabolic activation, incorporation and removal or degradation from the system.<sup>5</sup>

Sofosbuvir, acyclovir and tenofovir are nucleotide compounds proved to be among the most potent orally available antiviral treatments. These drugs exhibit high efficacy and a wide therapeutic index, with demonstrated utility in a number of chronic viral infections.<sup>6</sup>

Considering the published data regarding the adverse events associated with the use of oral nucleos(t)ide analogues, the present study was undertaken to evaluate the possible effects of sofosbuvir – a nucleo(s)tide analogue – on the mandibular alveolar process in albino rats through light microscopy, scanning electron microscopic examination and energy dispersive X-ray analysis (EDX).

#### Material and methods

#### Ethical statement

All experiments were conducted in the animal house of the Faculty of Medicine, Cairo University in Egipt according to the recommendations and approval of the Ethics Committee on animal experimentation of the Faculty of Oral and Dental Medicine, Cairo University in Egipt.

#### Experimental design

A total of 30 rats were used in the current study, weighing approx. 200 g. The animals were kept in a 12-hour light/dark cycle, at a temperature of  $22^{\circ}C \pm 2^{\circ}C$  with relative humidity 50% ±20%. The animals were fed

on standard chow pellets and tap water ad libitum for the entire test period. The rats were randomly assigned into 3 groups of 10 rats each. Group 1 served as a control and the rats received distilled water through oral gavage. Group 2 received sofosbuvir (Gratisovir<sup>®</sup> tablets, Pharco Pharmaceuticals, Alexandria, Egypt) dissolved in distilled water (8 mg/mL) through oral gavage at a dose of 40 mg/kg/day for 6 weeks.7 Group 3 received sofosbuvir dissolved in distilled water (8 mg/mL) through oral gavage at a dose of 40 mg/kg/day for 12 weeks.<sup>7</sup> All animals were sacrificed by ketamine overdose and the mandibles were dissected. The right side of each mandible was used for light microscopic examination, forming a total of 30 bone segments (10 specimens from each group). The other 30 bone segments (10 specimens from each group) of the left sides of the mandibles were used for scanning electron microscopic (SEM) examination as well as analysis of the constituent element (calcium concentration) using an energy dispersive X-ray unit attached to the SEM.

#### Light microscopic examination

All the specimens were fixed in 10% neutral formalin for 48 h, washed, soaked in 10% ethylene diamine tetraacetic acid (EDTA) for decalcification for 4 weeks, and then rinsed in distilled water. The specimens were dehydrated in ascending grades of alcohol and embedded in paraffin. Mesiodistal sectioning of the right side of the jaw was carried out. Histological sections were prepared of 5  $\mu$ m thickness. The sections were subjected to hematoxylin and eosin staining according to the conventional method. Histopathological examination was performed using light microscope.

# Scanning electron microscopic examination

The specimens were fixed in 2.5% glutaraldehyde in phosphate buffer (pH 7.4) for 6 h. The specimens were then dehydrated in ascending grades of ethanol for 10 min at each passage and 20 min twice in absolute ethanol, subjected to critical point drying according to standard procedures for SEM processing, and left to dry in air at room temperature for 3 days. The specimens were then mounted on scanning electron microscope stubs and studied with a Quanta 250 Field Emission Gun (FEG) SEM (Thermo Fisher Scientific, Waltham, USA).

#### **Energy dispersive X-ray analysis**

The quantitative composition of calcium in the studied samples was determined by EDX spot measurement, EDX line scan and element mapping. The EDX analysis unit works as an integrated feature of the Quanta 250 FEG SEM.

#### Statistical analysis

All the data obtained from the EDX analysis was statistically evaluated. This data was represented as mean values  $\pm$ SD, which was then statistically compared among the 3 studied groups. One-way analysis of variance (ANOVA) test for more than 2 independent samples was used, followed by Tukey's post hoc test for pair wise comparison. The results were expressed in the form of p-values that were considered significant when the probability value was  $\leq$  0.05 and highly significant when the probability value was  $\leq$  0.01. All statistical calculations were performed using following computer programs: Microsoft Excel 2007 and Statistical Package for the Social Science (SPSS) (SPSS Inc., IBM Corp., Armonk, USA) v. 15 for Microsoft Windows.

#### Results

#### **Histological results**

Group 1 (control group) showed normal alveolar bone architecture. The spongiosa consisted of interconnecting bone trabeculae containing osteocytes filling their lacunae. The bone trabeculae enclosed marrow cavities that were intensely lined with osteoblasts and filled with fibrocellular tissue (Fig. 1).



**Fig. 1.** Photomicrograph of group 1 (control): (A) showing normal interdental alveolar bone (AB) consisting of interconnecting bone trabeculae (bt), osteocytes filling their lacunae (arrows) and marrow spaces (asterisks) (A: H&E ×100, B: H&E ×400)

Group 2 demonstrated bone trabeculae with randomly distributed osteocytes as well as scalloped reversal lines. Howship's lacuna of osteoclasts were also clearly observed (Fig. 2).

Group 3 exhibited bone trabeculae with multiple degenerated areas as well as apparent vacuolization. The osteocytes were irregularly arranged. Moreover, some osteocyte lacunae were seen empty. Multiple reversal lines were clearly demonstrated (Fig. 3).



Fig. 2. Photomicrograph of group 2: (A) showing interdental alveolar bone (AB), (B) showing bone trabeculae (bt) containing reversal lines (black arrows) and Howship's lacuna (red arrows) (A: H&E ×100, B: H&E ×400)



Fig. 3. Photomicrograph of group 3: (A) showing interdental alveolar bone (AB), (B) showing bone trabeculae with multiple vacuoles (black arrows) and degenerated areas (red circle), multiple reversal lines (red arrows) and empty osteocytes lacunae (curved black arrows) (A: H&E ×100, B: H&E ×400)

#### Scanning electron microscopic results

The SEM images of the control group showed normal smooth alveolar bone architecture (Fig. 4).

SEM examination of group 2 demonstrated alterations of normal bone architecture. The bone surface revealed roughness as well as pore formation. Peeling off of the bone surface with some broken trabeculae was also observed



Fig. 4. Scanning electron microscopic photomicrograph of group 1 (control) showing normal smooth alveolar bone architecture (asterisks) (SEM  $\times$ 600)

in some areas. Other specimens displayed areas of disintegrated bone architecture with apparent mineralized collagen bundles. These collagen bundles seemed relatively well-organized (Fig. 5).



Fig. 5. Scanning electron microscopic photomicrograph of group 2 showing: (A) rough bone architecture (asterisks), peeling off of the bone surface (black arrow) and broken trabeculae (red arrows); (B) areas of disintegrated bone architecture with apparent mineralized collagen bundles (red arrows) (SEM A&B ×600)

SEM examination of group 3 showed a rough porous eroded bone surface with irregular disorganized bone architecture. The mineralized collagen fibers were seen forming a meshwork. A crack along the bone surface was also observed (Fig. 6).



Fig. 6. Scanning electron microscopic photomicrograph of group 3 showing eroded disorganized bone architecture (asterisks), mesh work of mineralized collagen fibers (red arrows) and a crack along the bone surface (white arrow) (SEM ×600)

#### Energy-dispersive X-ray analysis

The EDX elemental analysis demonstrated the peak of the chemical element – calcium (Ca) concentration – found in the samples in each group. The calcium content in the alveolar bone was evaluated in weight percent (wt%). EDX quantifies the relative contribution of Ca element to 100%. The mean values  $\pm$ SD for total weight percentage of calcium in groups 1, 2 and 3 were calculated (Table 1).

Table 1. Comparison of mean values (mean  $\pm$ SD) of calcium weight percentage in the studied groups

Studied groups	Ca weight [%]
Group 1	85.79 ±0.3725
Group 2	73.468 ±0.3182ª
Group 3	69.638 ±0.3669 <sup>a,b</sup>
p-value*	≤0.01

\* ANOVA test; Tukey's post hoc test; <sup>a</sup> statistically significant with group 1 ( $p \le 0.01$ ); <sup>b</sup> statistically significant with group 2 ( $p \le 0.01$ ).

Mean total weight percentage of calcium was highest in group 1 while the lowest was demonstrated in group 3. ANOVA test demonstrated a statistically significant difference among the 3 groups (p-value  $\leq 0.01$ ). Post-hoc multiple comparisons between 2 groups revealed that the difference in the mean value was statistically significant between groups 1 and 2, groups 1 and 3, and groups 2 and 3 (p-value  $\leq 0.01$ ) (Table 1).

#### Discussion

Nucleotide inhibitors (NIs) have proved great promise as direct-acting antivirals with broad genotype coverage, lack of preexisting alternatives with reduced susceptibility, a high barrier to resistance, and the ability to produce potent and durable antiviral responses.<sup>8</sup>

The hepatitis C virus (HCV) infects approx. 3 million individuals in the United States and 180 million worldwide per year.<sup>9</sup> NIs of HCV RNA synthesis served as alternate substrates and inhibitors of the viral RNA-dependent RNA polymerase HCV nonstructural protein 5B (NS5B). The binding of NIs to the highly conserved NS5B active site resulted in activity that maintained across genotypes and substantial loss of viral fitness upon the infrequent development of resistance mutations.<sup>10</sup>

Sofosbuvir is an oral NI of the HCV NS5B RNA dependent RNA polymerase enzyme, and the mediator demonstrated this activity across all genotypes of HCV.<sup>11</sup>

Although sofosbuvir showed a good secure profile in clinical trials, common side effects have been reported with sofosbuvir treatment in humans. These included headache, insomnia, fatigue, nausea, dizziness, pruritis, upper respiratory tract infections, rash, back pain, grade 1 anemia, and grade 4 lymphopenia.<sup>12</sup> Thus, we conducted this study to investigate the possible effects of sofosbuvir on alveolar bone histology and surface structure in a rat model.

In the current study, the sofosbuvir dose was calculated so that it was equivalent to the clinically administered dose. The usual total human adult dose of sofosbuvir per day is 400 mg.<sup>13</sup> According to Shin et al., this dose was equivalent to 8 mg per day for a rat weighing about 200 g.<sup>7</sup>

The alterations in the alveolar process were evaluated in the ongoing research following administration of sofosbuvir for 6 and 12 weeks in groups 2 and 3, respectively. These chosen time intervals were based on data from clinical trials reporting that the recommended treatment course duration with sofosbuvir 400 mg once daily is 12 weeks.<sup>14</sup> However the treatment duration was reported to be guided by on-treatment response and it might be extended to 24 weeks.<sup>15</sup>

The histological results of the present study revealed histopathological changes in the rat alveolar process in both groups 2 and 3 as compared to group 1. In group 2, the bone trabeculae exhibited irregularly arranged osteocytes. Multiple Howship's lacunae were also clearly seen denoting osteoclastic activity. Moreover, group 3 showed more pronounced changes as compared to group 2, where multiple vacuoles as well as degenerated areas in bone trabeculae were observed, indicating deteriorated bone structure. Empty osteocyte lacunae were observed, which indicate degeneration of osteocytes. Areas of post osteoclastic activity were reflected by the presence of multiple reversal lines in both groups 2 and 3.

Osteocytes in the current study displayed various changes in both group 2 and 3. These changes suggested bone pathology, as it was reported that osteocytes represent the key responder to various stimuli that regulate bone formation and remodeling as well as one of the key endocrine regulators of bone metabolism.<sup>16</sup>

The scanning electron microscopic results in the research provided good support for the histological findings. In group 2, the alveolar process bone architecture was rough. Moreover, group 3 showed a porous eroded bone surface with irregular disorganized bone architecture.

The relative content of calcium is critical for maintaining mineral homeostasis and bone metabolism.<sup>17</sup> Ca content was considered a suitable biomarker for the assessment of bone health.<sup>18</sup> EDX is a sensitive qualitative and semiquantitative technique for the assessment of the mineral content variations in calcified tissue.<sup>19</sup> The EDX results in the herein study support the SEM findings. The analysis of the Ca concentration (%) revealed the highest concentration in the control group while the lowest was found in group 3. Moreover, statistical analysis of the EDX results revealed a statistically significant difference among the studied groups.

It would appear from the reported results that there were obvious alterations in the alveolar bone accompanying sofosbuvir administration. The exact mechanism behind sofosbuvir's effect on the alveolar process is not clear but it may be attributed to the fact that sofosbuvir is a member of the nucleos(t)ide analogues.

Early generations of NIs were reported to cause toxicity. All HCV NIs are ribonucleotide analogues and, therefore, are more likely to attack host RNA polymerases than DNA polymerases.<sup>20</sup>

The mechanisms for toxicity of ribonucleotide analogues were not well characterized but they could be attributed to mitochondrial toxicity. NIs that were incorporated by the mitochondrial RNA polymerase (POLRMT) inhibited mitochondrial protein synthesis and showed a corresponding decrease in mitochondrial oxygen consumption in cells.<sup>21</sup> It has been reported that the use of sofosbuvir for 5 weeks resulted in swollen mitochondria with apparent cristolysis in the rat visual cerebral cortex.<sup>22</sup>

Mitochondrial dysfunction was reported to be the main factor in the formation of excess reactive oxygen species (ROS). The imbalance between these ROS and natural antioxidants created the conditions for oxidative stress.<sup>23</sup>

In bone tissues, recent studies have demonstrated that ROS generation is a key modulator of bone cell function and that oxidative status influences the pathophysiology of mineralized tissues. ROS can adversely affect bone homeostasis so that a proresorptive environment is favored. It was reported that ROS function as a signal mediator in osteoclast differentiation.<sup>24,25</sup>

Cadmium (Cd) has been shown to induce caspaseindependent apoptosis through a mitochondria–ROS pathway.<sup>26</sup> Zhao et al. investigated the effects of ROS in response to cadmium exposure on osteoblasts in rats.<sup>27</sup> They clarified that Cd had direct cytotoxic effect on osteoblasts, which mediated by caspases and mitogen activated protein kinase (MAPK) pathways.

A common dermatologic side effect reported with sofosbuvir was pruritis and rash.<sup>28</sup> Yonova has reported that pruritis was found to be caused by some chemical substance including histamine.<sup>29</sup> Another proposed cofactor that might be attributed to the alveolar bone histopathological changes demonstrated in the current study is the effect of histamine. Histamine was reported to be involved during the early phases of strong osteoclast resorption.<sup>30</sup>

#### Conclusions

It has been concluded that sofosbuvir induced apparent alterations in rats' alveolar bone. This effect was exaggerated upon a longer period of drug administration. The sofosbuvir-induced alterations might be attributed mainly to mitochondrial toxicity. The effect has been clearly shown histologically and morphologically as well as in bone mineral (calcium) content.

Further clinical and experimental studies are recommended to evaluate the effect of using sofosbuvir on various oral tissues.

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### Comparison of finite element results with photoelastic stress analysis around dental implants with different threads

# Porównanie wyników metody elementów skończonych w ocenie naprężeń fotoelastycznych wokół wszczepów zębowych o różnym gwincie

Maryam Geramizadeh<sup>1,B–D</sup>, Hamidreza Katoozian<sup>1,E,F</sup>, Reza Amid<sup>2,A,F</sup>, Mahdi Kadkhodazadeh<sup>2,A,E,F</sup>

<sup>1</sup>Department of Biomechanical Engineering, Amirkabir University, Tehran, Iran <sup>2</sup>Dental Research Center, Research Institute of Dental Sciences, Dental School, Shahid Beheshti University of Medical Sciences, Tehran, Iran

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):17-22

Address for correspondence Mahdi Kadkhodazadeh E-mail: Kadkhodazadehmahdi@yahoo.com

Funding sources None declared

Conflict of interest None declared

Received on November 29, 2017 Reviewed on February 2, 2018 Accepted on February 6, 2018

DOI 10.17219/dmp/85077

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#### Abstract

**Background.** The finite element method (FEM) has been used to analyze stress and strain distributions around 3 suggested dental implants with newly-designed thread parameters and the optimal shape of the implant was introduced considering the response surface optimization method sensitivity analysis. Experimental tests seemed necessary to confirm the results of the FEM.

**Objectives.** The aim of this study was to use experimental tests to prove the results of a finite element analysis of 3 dental implants with different thread designs under axial loads. Photoelastic stress analysis was chosen due to the similarity of analysis with FEM.

**Material and methods.** Two-dimensional models of 3 dental implants were built of grade 4 titanium to be tested in the polariscope. Model 1: A tapered implant with V-shaped threads; Model 2: A tapered implant with micro-threads in the upper area and V-shaped threads in the rest of the body; Model 3: A tapered implant with reverse buttress threads in all areas. Axial loading of 100 N was applied to the top of the implants and stress patterns and the maximum stress were evaluated for each implant.

**Results.** The minimum Huber-Mises-Hencky stresses of cortical bone were recorded in model 2, a tapered implant with micro-threads in the upper area and V-shaped threads in the rest of the body. The value for 100 N loading was 15.25 MPa, which was in agreement with the FEM.

**Conclusions.** Considering the stress patterns and values obtained from experimental tests of photoelasticity, the tapered implant with micro-threads in the upper area and V-shaped threads in the rest of the body has the most uniform and desirable stress distribution in the surrounding cortical bone and is preferred to be used in future applications.

**Key words:** dental implant, biomechanics, finite element, photoelasticity

**Słowa kluczowe:** implanty stomatologiczne, biomechanika, metoda elementów skończonych, fotoelastyczność

#### Introduction

Engineering techniques used to evaluate strain-stress fields are useful tools in the study of biomechanical applications. These experimental and numerical engineering tools were imported to biomechanics, in particular in dental biomechanics, a few decades ago. Several experimental techniques have been used in dental biomechanics, like photoelasticity, ESPI (Electronic Speckle Pattern Interferometry), strain gages, and other kinds of transducers. Photoelasticity is an experimental technique used for stress analysis of different objects under various loading conditions. It relies on transparent materials that show fringes when load is applied. This effect is the result of refraction of the polarized light by internal deformations due to stresses occurring in the model. Interpretation of these fringes shows the stress distribution and allows the measurement of their direction and magnitude. In the photoelastic method, a model similar to the studied structure is made in transparent material that has photoelastic properties. This model is placed under loading of the work conditions, which results in a deformation and color patterns.<sup>1–3</sup>

Polariscope is an optical system that shows the establishment of the main stress directions, as well as the difference between the 2 components of main stress. It is made up of polarizer, analyzer, and wave plates. The polarized light crosses the wave plates and arrives at the observer as an image of the optic parameters.<sup>4,5</sup>

The study of photoelasticity in the specific area of dental implant systems is of great interest since it can be useful in analyzing the stress distribution in both abutments (different designs and types) and implants screws (different types and angulations).<sup>6</sup>

Zielak et al. used 8 different designs of implants from 2 manufacturers, which were connected to their abutments, placed into epoxy resin blocks and observed under a polariscope coupled to a universal testing machine while subjected to axial loads of 5 N.<sup>7</sup> The obtained images were quantitatively analyzed by image analysis software. By this biomechanical study, it was possible to demonstrate a correlation of some implant characteristics to the colored fringe areas of tension distribution, a colorimetric method that can be used in comparative studies of photoelastic analysis.<sup>7</sup>

Udae et al. studied the photoelastic analysis of stress distribution on parallel and angled implants after installation of fixed prostheses.<sup>8</sup> The purpose of this study was to compare stress distribution in the fixed prosthesis with 3 parallel implants, to the stress distribution in the same prosthesis in the existence of an angled central implant. The presence of applied tensions was observed in the models after applying the torque to retention screws.<sup>8</sup>

Goiato et al. has done a photoelastic stress analysis in prosthetic implants of different diameters: mini, narrow, standard or wide.<sup>9</sup> Six photoelastic models were fabricated in PL-2 resin as single crowns or splinted 3-unit piece. This experiment shows that under axial loading, the number of fringes is inversely proportional to the diameter of the implants in the single crown models. The author concluded that the standard implant diameter always gives better stress distribution than the narrow and mini diameter implants. Additionally, the splinted crowns show more uniform stress distribution.<sup>9</sup>

Geramizadeh et al. used the response surface optimization method (available in finite element software packages) to optimize the parameters of designed dental implants, including thread depth and pitch, in order to have a uniform and desirable stress distribution.<sup>10</sup> In addition, the sensitivity of the simulation to different mechanical parameters was investigated and it was concluded that micro-thread parameters have greater effects on stress and strain values than the rest of the body's parameters.

Among the different aspects of design and analysis of dental implants, thread parameters are considered very important due to their effect on stress distribution. In the previous study,<sup>11</sup> 3 different dental implant designs were investigated using FE analysis, and model 2 was chosen as the best all around. In the present study, experimental tests were used to prove the former results and give better insight into the issue.

#### Material and methods

In our previous studies, modeling of the implants was done in Solidworks® 2014 and imported to ANSYS® Workbench software (ANSYS WB 2.0 Framework, v. 12.0.1, SAS IP®, Cheyenne, USA). All screws had the same geometry in terms of length and diameter (10.5 mm and 3.8 mm) in order to focus exclusively on the effect of thread design. The implant neck was beveled with a 22° angle relative to the vertical axis. This part had a 0.5 mm height and had to remain outside of bone. The area beneath the beveled margin had a 1 mm height with no threads and was to be placed in cortical bone. The rest of the body had specific thread designs as follows, to evaluate the effect of thread design on stress distribution: implant model 1 had V-shaped threads in the whole body; implant model 2 had 1.3 mm micro-threads on the upper part and the rest of the body was the same as model 1; implant model 3 had reversed buttress threads in all areas. The pitches of the V-shaped threads and micro-threads were 0.8 and 0.26 mm, respectively. The reversed buttress thread had a 0.8 mm pitch as well. The implant bodies had an 8° taper. A fixed rigid abutment with a 5 mm height was placed on top of the screw, where the force was applied. A bone block, 17 mm high and 12 mm wide, was built around the imported implants representing the surrounding bone in the second premolar region. A core of cancellous bone was covered with a thick layer of cortical bone with a constant width of 2 mm. The 3 models are shown in Fig. 1. Also, the whole model is shown in Fig. 2. Static loading



Fig. 1. Implant models with (1) V-shaped threads, (2) micro-threads and V-shaped threads, and (3) reversed buttress threads  $^{11}$ 



Fig. 2. Whole model including the implant and cortical, and cancellous bones

involved application of 100 N force at 0 and 25° angles relative to the vertical axis. The implants were considered to be made of grade 4 titanium. The behaviors of the materials were represented with linear isotropic material models. The mechanical properties of the materials used in this study are shown in Table 1. All materials were assumed to be homogenous, isotropic, and linearly elastic.<sup>11,12</sup>

In a plane stress problem, the thickness of the prototype is small in relation to the other dimensions; therefore, it can be assumed to function as a two-dimensional model. The 3 models of designed dental implants were built using a wire cut method (Fig. 3). Recently, these models were analyzed by means of the FEM and in this study the experimental tests were performed to prove the obtained results.<sup>11,12</sup> The prototypes were placed in a box with small depth compared to the other dimensions, so it can be considered a 2-dimensional model. This box was filled with photoelastic material, which was a combination of EPL 215 (epoxy oil) and EPH 5161 (hardener of epoxy oil). The mixture was liquid shaped at first and after mixing, it solidified in a few hours and the implant was fixed in it.

Materials	Young's modulus [MPa]	Poisson's ratio	Density [gr/cm³]	
Cortical bone	13700	0.3	1.85	
Cancellous bone	1370	0.3	0.9	
Grade 4 titanium	103400	0.35	4.5	



Fig. 3. Three models of dental implants with different thread designs



Fig. 4. Polariscope from 2 views

100 N axial loading was chosen to be applied on the top of the implants, which results in stress distribution in the whole model. The maximum amount of stress in the surrounding material around the implant is of interest in this study, which can be calculated using the following procedure.

First, the polariscope, which is shown in Fig. 4, is calibrated and, to do so, a disc in compression with the same material as the photoelastic material used is placed in the system. The stress-optic equations employed in photoelasticity always include a sensitivity coefficient. This coefficient represents the proportional factor between fringe order and stress level, and its magnitude varies with the type of photoelastic plastic. Loads are applied diametrically across the disc thickness and, from the theory of elasticity, the stresses at the center of the disc are:

$$\sigma_1 = \frac{2P}{\pi b d} , \quad \sigma_1 = -\frac{6P}{\pi b d} ,$$
  

$$\sigma_1 - \sigma_2 = \frac{c}{b} \times N = \frac{8P}{\pi b d} \rightarrow c = \frac{8P}{\pi d N}$$
(1)

Considering the color pattern, the center of the disk is green, which has 3.1 as N in the equation according to Table 1. Loading (P) of 3.5 N is applied and the prototype has a diameter (d) of 9 cm, which results in:

$$c = \frac{8P}{\pi dN} = \frac{8 \times 3.5}{\pi \times 0.09 \times 3.1} = 31.85 \ Pa \times m \tag{2}$$

This parameter will be used to calculate the stresses in each point of the model. In order to calculate maximum stress in each model, the color pattern is considered in 2 ways: normal view and oblique view.  $\alpha$  is the angle of the 2 views, which will be achieved using a prism light and, in this study, it is fixed at 30° in all tests. The parameter of fringe order (N) must be found in both views and finally the principle stresses can be calculated using the following equations. In these equations  $N_n$  and  $N_o$  are the fringe orders in normal and oblique views, respectively. The parameter d is the thickness of the model, which is 1 mm here.<sup>13,14</sup>

$$\sigma_1 = \frac{c}{d} \frac{1}{1 - \cos^2 \alpha} \left( N_o \cos \alpha - N_n \cos^2 \alpha \right)$$
(3)

$$\sigma_2 = \frac{c}{d} \frac{1}{1 - \cos^2 \alpha} \left( N_o \cos \alpha - N_n \right) \tag{4}$$

In FEM, the achieved stresses in the software are Huber-Mises-Hencky stress. Therefore, in order to compare the results, the principle stresses  $\sigma_1$  and  $\sigma_2$  must be converted to Huber-Mises-Hencky stress using this equation:

$$\sigma_{Von-Mises} = \sqrt{\sigma_1^2 - \sigma_1 \sigma_2 + \sigma_2^2} \tag{5}$$

Therefore, in the next step, each prototype should be fixed in the system and the loading condition will be applied. The color pattern is photographed in the normal and oblique situation to be used in calculations of stresses.

#### Results

The maximum amounts of Huber-Mises-Hencky stress in each model were calculated using the above-mentioned equations. The distribution of colors in Fig. 5 for each implant was chosen using Table 2, which gives the fringe order in normal view. In addition, color orders in oblique views are found in Fig. 6, which will result in fringe orders of the oblique view from Table 1. These parameters are used in equations 3 and 4 to calculate the principle stresses in each point. Equation 5 gives the Huber-Mises-Hencky stress, which makes it possible to compare the results with FEM. In this study, the maximum amounts of stresses are of great importance and examined carefully for each model. These values and the results of the FEM are reported in Table 3 to be compared easily.



Fig. 5. Stress distribution in the models in normal view

Table 2. Fringe orders obtained from different color patterns

Color	Fringe order
Black	0.00
Gray	0.28
White	0.45
Pale yellow	0.60
Orange	0.80
Dull red	0.90
Purple	1.00
Deep blue	1.08
Blue-green	1.22
Green-yellow	1.39
Orange	1.63
Rose red	1.82
Purple	2.00
Green	2.35
Green-yellow	2.50
Red	2.65
Red/green transition	3.00
Green	3.10
Pink	3.65
Pink/green transition	4.00
Green	4.15

Table 3. Comparison of the results of FEM and photoelastic analysis

Maximum Huber-Mises-Hencky stress in cortical bone [MPa]	Model 1	Model 2	Model 3
FEM	17.9	17.77	18.7
Photoelasticity analysis	16.75	15.25	20.12

FEM – finite element method.

As can be seen, there is a negligible difference between the results of the FEM and experimental tests. Model 2, a tapered implant with micro-threads in the upper area and V-shaped threads in the rest of the body, had the lowest stress in critical points and seems to be safer.

Stress distribution in all 3 models in the normal and oblique views are shown in Fig. 5 and 6. Obviously, the most critical points in these pictures are the contact surface of threads and cortical bone. The first thread especially is the most important one since it has the biggest stress value.



Fig. 6. Stress distribution in the models in oblique view

#### Discussion

In this study, the maximum Huber-Mises-Hencky stresses in cortical bone were calculated using photoelastic stress analysis to be compared with the results of the FEM. The results are represented in Table 3, which shows enough agreement between these 2 methods. However, there is negligible difference between the stress patterns in the 2 methods, which can be acceptable due to the limitations of the experimental tests and assumptions made in the FEM. The experimental tests can represent results acceptably close to the real conditions. Therefore, they can be used to prove the results of the software analysis.

As can be seen in Fig. 5, the maximum stress is happening in the neck of the implant, almost in the first thread that has contact with the bone. Therefore, this area has the biggest risk of failure and the danger of overstressing in the surrounding cortical bone should be considered especially in this area. This was also the case in the FEM, so the design of the threads in the upper area is of great importance in the design and analysis of dental implants.<sup>15</sup> Additionally, modeling the exact geometry of the implant complex, including the thread details, is essential for this setting.<sup>16</sup> In the present analysis, all implants had the same taper and bevel angles, which enabled accurate comparison of the effects of thread design. They were built using cut wire that has an acceptable accuracy in this case.

As seen in Table 3 and Fig. 5 and 6, implant model 2 caused lower stress in cortical bone and the most homogenous stress distribution, which made it the best option amongst all, during the insertion procedure as well as during the loading of implants. This model had micro-threads in the neck, which helped reduce the stress values in cortical bone. It also had V-shaped threads, which are stronger for cutting the jawbone during the procedure of screw placement. This result was also found in previous FEM, and the experimental tests show great agreement with them.<sup>11,12</sup>

Ideal function of dental implants in the human body depends on the ranges of the stress and strain in the surrounding bone. These values should be within an optimal range. Exceeding this range might result in degeneration and fraction of the bone, and bone atrophy might occur in the case of stress values which are too low.<sup>17</sup>

There are some limitations and simplifications in this study. The polariscope used was only working in 2 dimensions. The 3<sup>rd</sup> dimension, the thickness of the implant, was reduced to 1 mm and stress distribution is neglected in this dimension. This is obviously not the case in reality but it can be considered reasonable due to the similarity of the model in all directions. In addition, the only type of loading in this device is axial loading; therefore, other kinds of tensions such as shear stress cannot be analyzed. Another important factor is the assumption of isotropic and homogenous materials, which is not the case in the human body and may be responsible for the differences in results compared to other studies. Moreover, the absence of some components

such as crown, which was not included in the model, may cause different effects on stress/strain patterns. However, these assumptions are acceptable in experimental tests and the results show reasonable agreement with FEM results. Future studies may focus on 3-dimensional photoelastic analysis of dental implants, which is a more realistic test, and of course a much more complex situation. In that case, shear stresses can be included in the loading conditions, which makes the analysis closer to reality.

In this study, it is concluded that a tapered implant with micro-threads in the upper area and V-shaped threads in the rest of the body has the optimum shape for the most desirable stress distribution. These tests proved that this thread design is the best one for the purpose of uniform and optimal stress distribution and is suggested to be used in future applications.

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## Chronic kidney disease in children: Assessment of oral health status

## Przewlekła niewydolność nerek u dzieci – ocena stanu zdrowia jamy ustnej

Claudio Andaloro<sup>1,A–F</sup>, Concetto Sessa<sup>2,B,C</sup>, Nicolò Bua<sup>3,B,C</sup>, Ignazio La Mantia<sup>4,A–C,E,F</sup>

<sup>1</sup> ENT Unit, Santa Marta e Santa Venera Hospital, Acireale, Italy

<sup>2</sup> Nephrology and Dialysis Center "Tike", Syracuse, Italy

<sup>3</sup> Dental Unit, Department of Medical Surgery Specialities, University of Catania, Italy

<sup>4</sup> Department of Medical Sciences, Surgical and Advanced Technologies, University of Catania, Italy

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):23-28

Address for correspondence Claudio Andaloro E-mail: cla.anda@gmail.com

Funding sources None declared

Conflict of interest None declared

Received on November 20, 2017 Reviewed on December 19, 2017 Accepted on January 3, 2018

#### **DOI**

10.17219/dmp/81747

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#### Abstract

**Background.** Oral disease may be more prevalent in people with chronic kidney disease (CKD) due to the underlying pathology and its treatment. In children, it can elicit a wide spectrum of oral manifestations, including saliva changes, mucosal lesions, oral infection, gingivitis, and dental anomalies (mostly hypoplasia).

**Objectives.** The aim of the study was to determine the oral health status in a group of children with CKD and to compare with that from healthy controls.

**Material and methods.** A clinical cross-sectional study was conducted between June 2016 and September 2017 on 126 pediatric patients. Oral findings in a group of 65 children suffering from CKD were compared with a control group consisting of 61 individuals, free of any disease. We obtained an assessment of the oral health status by collecting carious, debris, calculus, gingival conditions, hypoplasia, and salivary flow rate data by using specific indexes. Medical and dental history was obtained for each subject.

**Results.** We found that children with CKD have a lower prevalence of caries for both permanent dentition (p = 0.019) and primary dentition (p = 0.008), while the prevalence of calculus, debris, gingivitis and enamel hypoplasia seems to be higher (all p < 0.005). A significantly reduced salivary flow rate was also noted in the CKD children, both in unstimulated (p = 0.037) and stimulated (p = 0.026) conditions.

**Conclusions.** CKD pediatric patients are more likely to present oral and dental changes than healthy children, so proper oral care and preventive measures should be taken to avoid potentially severe dental problems.

Key words: oral health, enamel hypoplasia, chronic kidney disease

Słowa kluczowe: stan zdrowia jamy ustnej, hipoplazja szkliwa, przewlekła niewydolność nerek

#### Introduction

Various medical conditions can affect the oral health of patients. Patients affected by chronic kidney disease (CKD), a progressive and irreversible deterioration of nephrons, which causes a decline in the glomerular filtration rate, may present a wide spectrum of oral manifestations in the hard and soft tissues as a results of the disease itself or/and the side effects of its treatment.<sup>1,2</sup> Many oral manifestations can be observed in the oral cavity, such as an ammonia-like smell, gingival enlargement secondary to drug therapy with cyclosporine and/or calcium channel blockers, enamel hypoplasia, dental calculus and dry mouth, uremic stomatitis, mucositis and glossitis, gingival inflammation due to plaque accumulation caused by poor oral hygiene, and a wide range of oral mucosal lesions, mainly white patches and/or ulceration such as lichenoid disease as a consequence of the associated drug therapy, like diuretics and beta-blockers, or oral hairy leukoplakia, observed with uremia.<sup>3–9</sup>

Singularly, the prevalence of dental caries in patients with CKD has been reported as being lower compared with healthy children, mostly due to the massive amount of urea in the saliva of nephropathic patients, which led to less caries with its neutralizing plaque formation capacity and antibacterial properties.<sup>10</sup> As there are numbers of studies describing and helping to understand the orodental manifestation and relevant treatment for adult patients with this disease, there are relatively few systematic studies of oral health status including pediatric patients.

Moreover, according to some studies, nephropathic children seem to use more health care services than other children.<sup>11</sup> Therefore, there is a need for more research regarding the oral health of special care children to improve their standard of life through a proper oral care treatment.

The aim of this study was to determine the oral health status by collecting carious, debris, calculus, gingival, hypoplasia scores, and salivary flow rate in a group of children with CKD, and to compare the findings with those collected from healthy control groups.

#### Material and methods

A clinical cross-sectional study was conducted between June 2016 and September 2017 in the Otolaryngology Unit of the Santa Marta e Santa Venera Hospital in Acireale, Italy, after the approval by the ethical committee of the Unità Operativa Complessa (UOC) Otorinolaringoiatria – ASP 3 CT. The study included children with a diagnosis of CKD, based on the definition that combined estimated glomerular filtration rate (eGFR) values <60 mL/min/1.73 m<sup>2</sup> and albuminuria ≥30 mg/g, before they had dialysis treatment, referred to our unit by the Nephrology and Dialysis Center "Tike", Syracuse, Italy.<sup>12</sup> In addition, we included a group of healthy control participants for comparison. Patients who fulfilled the following exclusion criteria were not included in the study: age higher than 18 years or lower than 4 years, noncooperative behavior, other systemic disorders, and dental treatment received earlier. Signed written informed consent was obtained from the parents or guardians of all the children participating in the study.

In each child, demographic information, medical and dental history, and oral hygiene habits (tooth brushing and flossing) were based on patients' medical files and parents' replies.

All participants underwent a standardized oral examination by a dentist trained in periodontology, according to the World Health Organization (WHO) criteria.<sup>13</sup>

Caries status was determined by the decayed missing and filled teeth (DMFT) index in the permanent dentition or the dmft index in the primary dentition (the sum of decayed, missing or filled permanent teeth).<sup>14</sup>

With regard to oral hygiene habits, we decided to use the Simplified Oral Hygiene Index (OHI-S) for assessing individual levels of the oral health status. The OHI-S values may range from 0 to 6, and the index has 2 components, the Debris Index and the Calculus Index. Each of these indexes is based on numerical determinations representing the amount of the debris or calculus found on the tooth surfaces. Possible scores for the Debris Index or the Calculus Index ranged between 0 (no debris or calculus present) and 3 (debris or supragingival calculus covering more than 2/3 of the tooth surface).<sup>15</sup>

The Modified Gingival Index (MGI), a non-invasive (no probing) method to rate normal gingival status (score 0), a very mild (score 1), mild (score 2), moderate (score 3), and severe (score 4) inflammation, was used to assess the gingival status.<sup>16</sup>

The presence and severity of enamel defects were classified according to the most severe hypoplasia found in any tooth. The findings were scored according to the following scale: 1. no hypoplasia; 2. horizontal white discoloration, no missing enamel; 3. horizontal yellow-brown discoloration, no missing enamel; 4. pitted; 5. horizontal fissures; 6. whole enamel stained; 7. whole enamel hypoplastic; 8. morphologic anomalies.<sup>17</sup>

Differences of stimulated and unstimulated salivary flow rates were assessed through a test described elsewhere.<sup>18</sup>

The results obtained in the present study were analyzed using the SPSS software v. 20.0 (SPSS Inc., Chicago, USA). Quantitative variables were presented as mean ±SD, whereas categorical variables as numbers and frequency. The Kolmogorov-Smirnov test was used to evaluate the distribution of variables. Fisher's exact test or the  $\chi^2$  test were used to study the categorical variables between patients and control subjects. The 2-sided t-test or the Mann-Whitney U test were used to study the quantitative numeric variables between the 2 groups. Statistical significance was set at p < 0.05.

#### Results

The present clinical study comprised of 126 children, whose general characteristics are presented in Table 1. The participants in each of the 2 groups were statistically similar in terms of age and gender, and different with regard to the oral hygiene habits. Indeed, in the CKD group, the prevalence of children brushing teeth twice a day or more and using dental floss is significantly lower than in the control group (p = 0.047 and p = 0.024, respectively). The number of non-brushing children is much higher in the CKD group than in the control group (p = 0.031) (Table 1).

The analysis of the differences in oral variables values between the 2 groups (Table 2) indicated a significant difference both in DMFT index for permanent dentition (p = 0.019) and dmft index for primary dentition (p = 0.008), with lower scores in the CKD group. Moreover, the CKD group showed a significantly higher Debris Index, Calculus Index and OHI-S overall index compared to the control group (p = 0.036, p = 0.028 and p = 0.033, respectively).

Table 1. Demographics and oral hygiene habits of the 2 study groups

Regarding the gingival status, the CKD group showed a higher prevalence of severe gingival inflammation (p = 0.018) and a lower prevalence of normal gums (p = 0.026) compared to the control group, as revealed by MGI values. The severity of enamel hypoplasia was significantly higher in the CDK group as compared to controls (3.57 ±0.51 vs 0.09 ±0.25; p = 0.002).

A significantly reduced salivary flow rate was observed in the CKD children, both in unstimulated (0.07  $\pm$ 0.01 vs 0.15  $\pm$ 0.02; p = 0.037) and stimulated (0.13  $\pm$ 0.03 vs 0.36  $\pm$ 0.11; p = 0.026) conditions (Table 2).

#### Discussion

The results showed that the CKD group had a higher proportion of caries-free children than healthy children, free of any disease; indeed, the prevalence of caries is significantly lower in the CKD group than in controls for both DMFT and dmft scores, though the 2 studies did

Characteristics	CKD group n = 65	Control group n = 61	Overall n = 126	p-value
Age, in years mean ±SD median range	9.92 ±2.75 9 5-16	9.34 ±2.43 9 5-15	10.11 ±3.31 9 5-16	0.756 _ _
Gender, n (%) male female	35 (53.8) 30 (46.2)	32 (52.5) 29 (47.5)	67 (53.2) 59 (46.8)	0.624 -
Oral hygiene habits, n (%) toothbrushing never once a day twice a day or more	25 (38.5) 23 (35.4) 17 (26.2)	14 (23.0) 24 (39.3) 23 (37.7)	39 (31.0) 47 (37.3) 40 (31.7)	0.031* 0.893 0.047*
dental floss use	12 (18.5)	22 (36.1)	34 (27.0)	0.024*

\* Significant difference at p < 0.05.

Table 2. Oral variables of the 2 study groups

Characteristics	CKD group n = 65	Control group n = 61	Overall n = 126	p-value
DMFT index	6.86 ±1.69	10.69 ±1.81	8.78 ±1.75	0.019*
dmft index	7.02 ±1.75	12.11 ±2.03	9.57 ±1.89	0.008*
Debris index	1.33 ±0.43	0.56 ±0.29	0.95 ±0.36	0.036*
Calculus index	1.52 ±0.52	0.32 ±0.11	0.92 ±0.32	0.028*
OHI-S	2.85 ±0.95	0.88 ±0.40	1.87 ±0.68	0.033*
MGI, n (%) normal very mild mild moderate severe	11 (16.9) 10 (15.4) 13 (20.0) 16 (24.6) 15 (23.1)	20 (32.8) 14 (23.0) 11 (18.0) 11 (18.0) 5 (8.2)	31 (24.6) 24 (19.0) 24 (19.0) 27 (21.4) 20 (15.9)	0.026* 0.044* 0.687 0.091* 0.018*
Hypoplasia	3.57 ±0.51	0.09 ±0.25	1.83 ±0.38	0.002*
Salivary flow rate unstimulated (mL/min)	0.07 ±0.01	0.15 ±0.02	0.11 ±0.02	0.037*
Salivary flow rate stimulated (mL/min)	0.13 ±0.03	0.36 ±0.11	0.25 ±0.07	0.026*

\* Significant difference at p < 0.05.

not show any statistically significant differences in caries prevalence between the 2 groups.<sup>5,19</sup> Our results are in agreement with those reported by Nakhjavani et al. and Sobrado Marinho et al.<sup>20,21</sup> This may be due to the presence of highly buffered and alkaline saliva due to the elevated urea and phosphate concentrations in patients with renal disease, despite poor oral hygiene, a carbohydrate-rich diet (necessary to reduce the renal workload), in addition to disease-related debilitation, hypoplastic enamel and low salivary flow rate, usually seen in these patients.<sup>22</sup> In order to establish the Oral Hygiene Index in children with CKD, we collected the debris and calculus scores. Both scores were significantly greater in the CKD group compared to healthy individuals, in accordance with a study by Martins et al.<sup>5</sup> Elevated salivary pH, decreased salivary magnesium, and high levels of salivary urea and phosphorus, probably due to the decrease in the glomerular filtration rate and urinary excretion, lead to the precipitation of calcium-phosphorus and calcium oxalate, and, thus, dental calculus formation.<sup>23</sup> The issue of gingival inflammation in CKD patients appears to be very controversial in the literature. Several reports suggest reduced gingivitis in these patients, caused by immunosuppression and uremia, associated with the renal disease that may alter the inflammatory response to bacterial plaques in the gingival tissue.<sup>10,22,24</sup> Also, it was said that a decreased level of hemoglobin can lead to paleness and mask the inflammatory signs in the gingival.<sup>2</sup>

However, other reports indicate opposite results.<sup>25</sup> In experimentally induced gingivitis in CKD patients, Kitsou et al. concluded that chronic uremia had no effect on the defense of periodontal tissues against microbial plaque.<sup>26</sup>

In our study, the gingival status showed a higher prevalence of severe gingival inflammation in patients with CKD (23.1%) than in those from the control group (8.2%) (p = 0.018). The greater gingival scores in our study may be attributed to the higher plaque rate among CKD patients and the increased inflammation in the involved tissues due to the systemic conditions that change the local tissue homeostasis.<sup>27</sup>

Enamel hypoplasia is a well-known phenomenon in CKD patients.<sup>10</sup> Accordingly, in the present study, enamel hypoplasia was significantly more extensive and more severe in patients with CKD than in the control group. Calcium reduction together with renal disease that occurs during the mineralization of dentition can result in developmental defects of enamel as shown in several studies.<sup>22,28</sup> A putative reason for the finding of poor dental health is the lower stimulated salivary secretion rate observed in the CKD group when compared with the control group. This supports previous findings indicating that a decrease in salivary flow is an important risk factor for poor dental health in hemodialysis patients.<sup>5</sup>

However, the present study has some limitations and the results should be interpreted in the context of its design. There is no data about the patients' classification of CKD. The 5 different CDK stages may affect the oral health status of the study patients in various ways, so it could be useful to create a study with all CKD stages (I–V). An additional limitation of this study was that the participants in this analysis were already diagnosed with CKD and before progression to the dialysis treatment, so we do not know how our results would relate to the length of the disease or dialysis therapy. A study over a longer period, evaluating these factors, may address these issues.

Another limitation is that a number of indexes and scores have been developed for assessing individual levels of the oral health status, so different score methods can lead to different results.

#### Conclusions

Within the limitations of the study, our data suggests that CKD children are more likely to present oral and dental changes than healthy ones. Therefore, a better understanding of the systemic and oral abnormalities in those individuals may help clinicians obtain effective oral care and plan preventive routines according to individual needs.

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### In vitro effect of changing the horizontal angulation of X-ray beam on the detection of proximal enamel caries in bitewing radiographs

### Wpływ zmian poziomego kąta padania promienia centralnego X na wykrywanie próchnicy szkliwa na powierzchniach stycznych na zdjęciach skrzydłowo-zgryzowych – badania in vitro

Abbas Shokri<sup>1,A</sup>, Shahin Kasraei<sup>2,B</sup>, Elham Shokri<sup>3,C</sup>, Maryam Farhadian<sup>4,E</sup>, Bahareh Hekmat<sup>1,D,F</sup>

<sup>1</sup> Dental Implant Research Center, Department of Oral and Maxillofacial Radiology, Dental School, Hamedan University of Medical Science, Iran

- <sup>2</sup> Department of Restorative Dentistry, Dental School, Shahid Beheshti University of Medical Sciences, Tehran, Iran
- <sup>3</sup> Dental Research Center, Department of Oral and Maxillofacial Radiology, Dental School, Hamedan University of Medical Science, Iran

<sup>4</sup> Modeling of Noncommunicable Diseases Research Center, Department of Epidemiology, School of Public Health, Hamedan University of Medical Sciences, Iran

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation;

D- writing the article; E- critical revision of the article; F- final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):29-34

Address for correspondence Bahareh Hekmat E-mail: bahar\_hekmat70@yahoo.com

Funding sources None declared

Conflict of interest None declared

Received on November 21, 2017 Reviewed on December 27, 2017 Accepted on January 28, 2018

#### DOI

10.17219/dmp/84868

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#### Abstract

**Background**. Bitewing radiography is an important modality useful for the evaluation of teeth in patients of various ages and in different stages of tooth eruption. Clinical examination of proximal surfaces for caries may result in false negative results, especially in tight contact areas. Thus, radiography, as an adjunct to clinical examination, is used as a routine diagnostic modality for caries detection.

**Objectives.** The objective of this study was to assess the in vitro effect of changing the horizontal angulation of X-ray beam on the detection of proximal enamel caries in bitewing radiographs.

**Material and methods.** This in vitro study was conducted on 150 caries-free human premolars (code: p/16/35/9/210). The teeth were randomly divided into 3 groups (n = 50). Group 1 served as the control group and no carious lesions were induced in this group. Teeth in groups 2 and 3 were immersed in demineralizing solution for 2 and 4 months, respectively. After induction of caries and its radiographic confirmation, the teeth were mounted in wax in groups of 3 and bitewing radiographs were obtained at 0°, 5°, 10° and 15° horizontal angles.

**Results.** The sensitivity values of bitewing radiographs at 0°, 5°, 10° and 15° horizontal angles were 88%, 90%, 88%, 92% in group 2 and 88%, 94%, 94% and 94% in group 3, respectively. The specificity values of bitewing radiographs at 0°, 5°, 10° and 15° horizontal angles were 92%, 86%, 84% and 76%. The accuracy of bitewing radiographs at 0°, 5°, 10° and 15° horizontal angles was 89.3%, 90%, 88.7% and 87.3%, respectively. The highest diagnostic accuracy was obtained at 5° horizontal angle for caries detection; however, the difference in this regard among the tested horizontal angles was not statistically significant (p = 0.846).

**Conclusions.** Based on the results of this study, changing the horizontal angulation has no significant effect on the detection of proximal enamel caries in bitewing radiographs.

Key words: bitewing radiography, approximal caries, angulation, X-ray beam

**Słowa kluczowe:** zdjęcie skrzydłowo-zgryzowe, próchnica na powierzchniach stycznych, kąt promienia centralnego, wiązka promieniowania rentgenowskiego

#### Introduction

Dental caries is a microbial infectious disease characterized by demineralization and destruction of the mineral structure of teeth. Carious lesions develop as a result of the activity of acidogenic bacteria since they provide a suitable environment for the demineralization of tooth structure.<sup>1</sup>

Carious lesions confined to the enamel are referred to as incipient caries. Incipient caries is characterized by an intact surface with an underlying porous structure, which is reversible and can be remineralized. Clinically, an incipient carious lesion appears as a white spot lesion detectable after drying the tooth surface.<sup>1</sup>

Proximal tooth surfaces are susceptible to caries. Proximal caries develops at the contact area between 2 adjacent teeth. Clinically, proximal carious lesions have an opaque appearance and decrease enamel translucency below the contact area and above the free gingival margin.<sup>2</sup>

Carious lesions are primarily detected by a clinical examination. Visual inspection is the first step in the clinical examination of teeth. A dental explorer can be used for tactile examination. However, inappropriate use of the sharp tip of an explorer can create a cavity on the sound surface of an incipient carious lesion. Radiography is another diagnostic modality for this purpose. Properly obtained radiographs can provide valuable diagnostic information as a supplement to a clinical examination.<sup>1</sup>

Bitewing radiography is an important modality useful for the evaluation of teeth in patients of various ages and in different stages of tooth eruption. Bitewing radiographs often visualize the crowns of maxillary and mandibular teeth as well as the alveolar crest; thus, they are suitable for visualization and inspection of proximal areas and the detection of approximal carious lesions, which have yet to manifest clinically.<sup>3</sup> In bitewing radiography, X-ray beams pass through the teeth parallel to the occlusal plane, and thus the contact area is well visualized (as an open contact) on radiographs.<sup>4</sup>

Clinical examination of proximal surfaces for caries may result in false negative results, especially in tight contact areas. Thus, radiography, as an adjunct to clinical examination, is used as a routine diagnostic modality for caries detection.<sup>4,5</sup> In order to visualize a carious lesion on a parallel radiograph (0° angle), it must have a minimum depth of 0.5 mm; a cavity has yet to be formed in 60% of such carious lesions.<sup>1</sup> The absence of a horizontal and vertical overlap is a primary requirement for an optimal bitewing radiograph, which is helpful for the correct detection of caries.<sup>6</sup>

Despite the fact that the contact area of teeth must be open and overlaps must be absent on bitewing radiographs, in the current study bitewing radiographs were obtained at 0°, 5°, 10° and 15° horizontal angles to evaluate the effect of changing the horizontal angulation on the accuracy of detecting proximal enamel caries, since early detection of caries is absolutely necessary to take measures to cease the progression of carious lesions. In other words, we hypothesized that we might be able to disregard some degrees of overlap (caused by changing the horizontal angulation) in favor of enhancing early detection of proximal carious lesions shallower than 0.5 mm.

Digital radiography was used in this study, since digital software programs allow easy manipulation of contrast to enhance visualization of a particular area; whereas, if the respective contact area is completely open (no overlap), increased contrast may cause burnout of enamel margins and, consequently, incipient caries may be masked and remain undetected. Thus, this study aimed to assess the effect of changing the horizontal angulation of X-ray beams for the detection of artificially induced proximal enamel caries on bitewing radiographs.

#### **Objectives**

The objective of this study was to assess the in vitro effect of changing the horizontal angulation of X-ray beam on the detection of proximal enamel caries in bitewing radiographs.

#### Material and methods

This in vitro, experimental study was conducted in Dr. Abbas Shokri Oral and Maxillfacial Radiology Clinic (Hamedan, Iran). The study was conducted on 150 sound (caries-free) extracted human premolar teeth (code: p/16/35/9/210). The teeth had been extracted for orthodontic or periodontal reasons. The teeth were cleaned and immersed in 0.2% sodium hypochlorite solution for 20 min for disinfection. Next, they were stored in saline solution until the experiment. The teeth were inspected visually and then with a dental explorer to ensure that there were no caries, enamel defects or cracks. The tooth surfaces were coated with 2 layers of nail varnish. Then, the nail varnish on the mesial/distal surface was randomly removed to create a varnish-free window measuring  $2 \times 2$  mm (Fig. 1).

TenCate demineralizing solution with a pH of 4 was used to artificially create carious lesions. The chemical composition of this solution included 2.2 mM calcium chloride (CaCl<sub>2</sub>), 2.2 mM potassium dihydrogen phosphate (KH<sub>2</sub>PO<sub>4</sub>), 0.05 M acetic acid and 1 M potassium hydroxide (KOH).<sup>7</sup> The teeth were randomly divided into 3 groups (n = 50). Group 1 served as the control group and artificial carious lesions were not induced in teeth in this group. Teeth in groups 2 and 3 were immersed in demineralizing solution for 2 and 4 months, respectively. Teeth in groups 2 and 3 were radiographed periodically every 30 days using Minray<sup>®</sup> intraoral X-ray unit (Tuusula, Helsinki, Finland) with the exposure set-



Fig. 1. Teeth coated with nail varnish with a varnish-free window on one of their proximal surfaces

tings of 60 kVp, 7 mA and 0.1 s time at 0° angle. Development of incipient caries was radiographically confirmed as such (Fig. 2).

After the formation of carious lesions and their radiographic confirmation, the teeth were coded and randomly mounted in wax (3 teeth in a row per each wax mold) such that the carious proximal surfaces were positioned in contact with the adjacent teeth (Fig. 3). Photostimulable phosphor plate (PSP) digital sensors (Optime®, Soredex, Helsinki, Finland) were used to take digital radiographs of the teeth. After placing a size 2 PSP sensor in the desired position, bitewing radiographs were obtained using Minray X-ray unit with the exposure settings of 60 kVp, 7 mA and 0.1 s time at 0° (perpendicular to tooth surface), 5°, 10° and 15° horizontal angles. To adjust the horizontal angle (since the horizontal angle of the X-ray unit was not adjustable and the tube could only be adjusted in different vertical angles), the X-ray tube was rotated such that the vertical angulation options were positioned horizontally. Horizontal angulation was adjusted as such and bitewing radiographs were taken at the aforementioned angles (Fig. 3, 4).



Fig. 2. Radiographs obtained at 30 (a), 60 (b) and 90 (c) days following immersion of teeth in demineralizing solution to ensure development of carious lesions



Fig. 3. Teeth mounted in wax



Fig. 4. Adjusting the X-ray tube angulation to take radiographs at different horizontal angles

All radiographs were evaluated by 2 oral and maxillofacial radiologists for the presence of proximal carious lesions and performed clear detection using a 1–4 scoring system as follows:

- 1. No carious lesion detectable,
- 2. Carious lesion not clearly visible (indefinite diagnosis),
- 3. Carious lesion clearly visible,
- 4. Presence/absence of carious lesion could not be determined due to significant overlap.<sup>5</sup>

Observers were blinded to the carious/sound state of teeth and the horizontal angulations at which the radiographs were taken (Fig. 5).





Fig. 5. Bitewing radiographs taken at 0° (a), 5° (b), 10° (c) and 15° (d) horizontal angles



Fig. 6. Teeth longitudinally sectioned by a diamond disc for evaluation under a stereomicroscope for definite detection of carious lesions

The teeth were then sectioned parallel to their longitudinal axis in buccolingual direction (Fig. 6) and evaluated under a stereomicroscope (Olympus<sup>®</sup>, Hamburg, Germany) at  $\times 20$  magnification to determine the presence/ absence of carious lesions.

#### Results

Table 1 shows the inter- and intraobserver agreements for detection of caries on bitewing radiographs taken at 0°, 5°, 10° and 15° horizontal angles. The highest interobserver agreement (93.5%) was noted for radiographs taken at 10° horizontal angle while the highest intraobserver agreement (78%) was noted in radiographs taken at 5° horizontal angle.

 Table 1. Intra- and interobserver agreements for caries detection on

 bitewing radiographs taken at different horizontal angles by calculation

 of the kappa statistic

Interobserver agreement [%]	Intraobserver agreement [%]	Angle
85.4	73.9	0°
92.2	78.5	5°
93.5	76.5	10°
93.3	76.3	15°

Table 2 shows the sensitivity, specificity, accuracy, positive predictive value and negative predictive value of bitewing radiographs for detection of proximal enamel caries. In group 2, radiographs taken at 15° horizontal angle had the highest (94%) and those taken at 0° horizontal angle had the lowest sensitivity (86%). In group 3, radiographs taken at 10° and 15° horizontal angle had the highest (94%) and those taken at 0° horizontal angle had the lowest (80%) sensitivity. The difference in sensitivity values among different angulations was statistically significant (p = 0.026). Specificity values decreased by an increase in the horizontal angle. The highest specificity (94%) was noted at 0° horizontal angle, while the lowest specificity (84%) was noted at 15° horizontal angle. The difference in this respect was not statistically significant (p = 0.288). The highest accuracy (90%) was noted at 15° horizontal angle while the lowest accuracy (86%) was recorded at 0° horizontal angle. The difference in this regard among different horizontal angles was not statistically significant (p = 0.593).

#### Discussion

Nowadays, dental caries is a major health problem and one of the most common chronic diseases throughout the world. In has a great effect on the life quality and major indirect impact on the economy.<sup>8,9</sup> One of the most problematic parts of a dental examination is the detection of caries lesions on tooth surfaces which are hard to reach. Nowadays, in clinical practice, besides clinical examination, subsidiary methods, such as radiovisiography or laser in identifying proximal caries, are recommended.<sup>10</sup> Valid and reliable methods detecting proximal caries lesions are required for the appropriate treatment of dental caries.<sup>11</sup> Different techniques for detecting proximal caries have been studied and the bitewing radiography is reported as the most efficient modality for the detection of proximal caries.<sup>2,10</sup>

Early detection of incipient non-cavitated caries lesions is important since their progression can be stopped by preventive interventions with minimal invasion to the tooth structure and no need for restorative treatments.<sup>5,11</sup> Shallow carious lesions detected early (before forming a cavity) can be reversed by preventive measures such as fluoride therapy, oral hygiene instruction and decreasing the consumption of carbohydrates.

Table 2. Sensitivity, specificity, accuracy, positive predictive value and negative predictive value of bitewing radiographs taken at different angles for detection of proximal enamel caries

Angle	Sensitivity [%]	Spacificity [0/]		False negative [%]			
	group 3	group 2	specificity [%] Faise positive [%]	group 3	group 2	Accuracy [%]	
0°	86	80	94	6	14	20	86.7
5°	90	90	90	10	10	10	90
10°	88	94	86	14	12	6	89.3
15°	94	94	84	16	6	6	90.7
p-value	0.5	93	_	-	0.2	88	0.026

Horizontal angles adjusted for taking bitewing radiographs from the premolar and molar areas are slightly different. In the clinical setting, incipient carious lesions confined to the enamel in premolar teeth are often better visualized on radiographs taken from the molar area. Thus, a question arises whether changing the horizontal angle when taking bitewing radiographs can enhance the detection of proximal enamel caries, or on the contrary, lesions detected on such radiographs are not caries and changing the horizontal angle may create a caries-like appearance and lead to false positive results.

The current study evaluated the effect of changing the horizontal angle when taking bitewing radiographs on the detection of proximal enamel caries. The results showed that the sensitivity of bitewing radiographs increased by an increase in horizontal angle (compared to 0° angle); however, the difference in this respect among the tested horizontal angles was not statistically significant. Chadwick et al. evaluated the effect of changing the horizontal angulation of X-ray beam and buccolingual cavity width on radiographic depth of approximal cavities and reported that alterations in the horizontal angle significantly affected the depth of a cavity such that an increase in horizontal angle led to an increase of the radiographic depth as well. By changing the horizontal angle of the X-ray beam, carious lesions are visualized to a greater extent.<sup>12</sup>

In our study, the sensitivity of bitewing radiographs for detecting proximal enamel caries was found to be 88%, which was higher than the values obtained by Pontual et al. and Abesi et al.<sup>4,5</sup> This difference in the results may be attributed to the following reasons:

- 1. The current study was conducted on extracted teeth collected from several centers. Three teeth were mounted next to each other in a row in wax, and a simulation of dental arch and natural contact of teeth in the oral cavity was not feasible. This might have increased the ability of observers for detecting caries.
- 2. In our study, soft tissue was not simulated when taking radiographs while in the oral cavity, gingival soft tissue and structures such as nasolabial fold may be superimposed on teeth and decrease the accuracy of detection of caries.
- 3. In the clinical setting, following several consecutive demineralization and remineralization cycles, enamel caries occurs due to the dominance of the demineralization process. Thus, enamel carious lesions in the clinical setting are irregular and have a relatively low contrast; as a result, they are not easily detectable radiographically. However, the current study had an in vitro design; the teeth were sound and only demineralizing solution was used to induce caries. Thus, the resultant carious lesions had a more regular structure, which enhanced their detection using radiographs.

In the current study, specificity was calculated to be 92%, which was similar to the results of Kamburoğlu et al., Pontual et al. and Abesi et al. $^{2,4,5}$  In our study,

specificity decreased with an increase in the horizontal angle, although no statistically significant difference was noted in this regard among the tested horizontal angles. Changing the angle results in an overlap of adjacent structures and, due to superimposition of the enamel of the 2 adjacent teeth, this area appears more opaque on radiographs. Margins of the overlapped area absorb less X-ray beam due to the lower density and appear radiolucent on radiographs mimicking carious lesions. This decreases the ability of observers to detect sound surfaces. As a result, observers may misdiagnose sound surfaces as carious. In our study, vertical angle was 0° when taking radiographs and the highest diagnostic accuracy was noted in radiographs taken at a 5° horizontal angle; however, the difference in this respect among different horizontal angles was not statistically significant. Thus, if incipient carious lesions are not detected on standard bitewing radiographs but the patient complaints of tooth hypersensitivity at the area, horizontal angle can be changed when taking bitewing radiographs to better evaluate the area for possible caries. Moreira et al. evaluated the effect of vertical angle of X-ray beam on the detection of secondary caries developed under esthetic restorations and showed that 10° vertical angle yielded the highest diagnostic accuracy, although the difference among the tested angles was not statistically significant.<sup>13</sup> In the current study, the sensitivity for detecting carious lesions was higher in group 3 (longer immersion of teeth in demineralizing agent). This finding was in agreement with the results of previous studies reporting that a greater depth of caries results in the easier detection of carious lesions.<sup>5,12,14</sup> However, Pontual et al. reported results contrary to our findings and those of some other studies.<sup>4</sup> In our study, radiographs taken at 5° horizontal angle yielded the highest intraobserver agreement, while the highest interobserver agreement was noted in radiographs taken at 10° horizontal angle.

#### Conclusions

This study showed no statistically significant difference in the detection of proximal enamel caries in bitewing radiographs taken at different horizontal angles. However, based on the obtained results, in cases suspected of proximal caries which is not visualized in a standard bitewing radiograph taken at 0° angle in the clinical setting, horizontal angulation can be changed by 5° to 10° to enhance diagnosis.

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# Influence of crown design and material on chipping-resistance of all-ceramic molar crowns: An in vitro study

# Wpływ kształtu i materiału pełnoceramicznych koron trzonowców na odporność na odpryskiwanie – badanie in vitro

Majed Alsarani<sup>1,2,A–D</sup>, Grace De Souza<sup>3,A,C,E</sup>, Amin Rizkalla<sup>4,A,C,E</sup>, Omar El-Mowafy<sup>3,A,C,E–F</sup>

<sup>1</sup>Faculty of Dentistry, University of Toronto, Canada

<sup>2</sup> Faculty of Applied Medical Sciences, King Saud University, Riyadh, Saudi Arabia

<sup>3</sup> Department of Restorative Dentistry, Faculty of Dentistry, University of Toronto, Canada

<sup>4</sup> Faculty of Dentistry, Schulich School of Medicine & Dentistry, University of Western Ontario, London, Canada

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):35-42

#### Address for correspondence

Majed Alsarani E-mail: majed.alsarani@dentistry.utoronto.ca

#### Funding sources

This research was supported in part by King Saud University Riyadh, Saudi Arabia.

Conflict of interest None declared

Received on January 19, 2018 Reviewed on January 21, 2018 Accepted on February 2, 2018

#### DOI

10.17219/dmp/85000

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# Abstract

**Background.** All-ceramic restorations have become popular and the trend is ongoing. However, the incidence of chipping within the veneering layer has been a commonly reported failure in clinical practice.

**Objectives.** The aim of this in vitro study was to evaluate the effect of ceramic crown design (monolithic vs bi-layered) and material on the chipping resistance of molar crowns submitted to compressive cyclic loading.

**Material and methods.** Fifty identical epoxy resin replicas of a mandibular 1<sup>st</sup> molar with crown preparation were divided into 5 groups (n = 10) as follows: the MLD group – monolithic CAD/CAM lithium-disilicate glass-ceramic (LDGC) crowns; 30 zirconia cores were veneered with either feldspathic porcelain by hand-layering technique (ZHL) or by heat-pressing technique (ZVP), or with milled LDGC veneers and subsequently fused to the cores (ZLD); 10 porcelain-fused-to-metal (PFM) crowns acted as a control group. All crowns were cemented using Panavia<sup>®</sup> F2.0 resin cement (Kuraray Dental, Tokyo, Japan). After storage in water at 37°C for 1 week, the specimens were subjected to compressive cyclic loading at the mesiobuccal cusp which was tilted at 30°. A load cycle of 50–450 N was used and specimens were maintained in an aqueous environment throughout 500,000 cycles in a universal testing machine (Instron, Norwood, USA). The data was statistically analyzed at 5% significant level with Fisher's exact test and Kaplan-Meier survival analysis.

**Results.** Significant differences in survival rates of the specimens used in the groups (p < 0.001) were found. Specimens of the PFM, ZHL and ZVP groups underwent failures at different stages of the 500,000 fatigue cycles, while specimens of the MLD and ZLD groups survived the entire fatigue test. ZHL and ZVP crowns had the worst chipping-resistance, while PFM crowns performed slightly better. The Kaplan-Meier test revealed significantly higher survival rates for the MLD and ZLD specimens compared to the other 3 groups.

**Conclusions.** The use of LDGC as a monolithic molar crown and as a veneer over a zirconia core resulted in superior resistance to cuspal chipping.

Key words: fatigue, crown, chipping, monolithic, posterior

Słowa kluczowe: badanie zmęczeniowe, korona, odpryski, monolityczny, tylny

# Introduction

Computer-aided design/computer-aided manufacturing (CAD/CAM) technology enables precise milling of crowns and fixed dental prostheses (FDP) from a variety of ceramic-based blocks including high-strength materials such as lithium-disilicate glass-ceramic (LDGC) and zirconia.<sup>1</sup> Because of the ongoing trend toward more esthetically-pleasing and biologically-compatible restorations, different all-ceramic systems have been developed.<sup>2</sup> The use of yttrium partially-stabilized zirconia polycrystal (Y-TZP) has gained popularity due to the superior mechanical properties of the material such as high flexural strength (>900 MPa) and compressive strength (2000 MPa).<sup>3</sup> This material is capable of providing a strong framework for dental restorations which have a reportedly low failure rate.<sup>4,5</sup>

Originally, the application of high-crystalline zirconia in dental restoration was limited to substructures due to its high opacity. Veneering is typically applied over zirconia cores to provide a more natural appearance.<sup>6</sup> In such a complex, the veneering porcelain is the weaker component, and a high incidence of occlusal chipping of posterior bi-layered crowns has been reported.<sup>7,8</sup> The failure rate observed for both tooth-supported and implant-supported bi-layered zirconia crowns has varied between 3% and 50%.<sup>9,10</sup> Traditionally, veneering porcelain is hand-layered over the zirconia core, however a pressed-on veneering technique is an alternative. It has been reported that the pressed-on veneering technique minimizes chipping in zirconia crowns compared to the conventional (handlayering) technique.<sup>11,12</sup>

There are different hypotheses for the high chipping rates of all-ceramic restorations, including the mismatch of coefficient of thermal expansion (CTE) between the core and the veneer material, uneven thickness of the porcelain veneer, and the quality of the bonded interface between the veneer and the core material.<sup>13</sup> Significant CTE mismatch would potentially create stresses at the core/veneer interface, which might cause the porcelain veneer chipping. To enhance the overall strength of the core/veneer complex, the veneering porcelain should ideally have lower CTE compared to that of the core in order to create compressive stresses during cooling of the restoration.<sup>14</sup> In addition to variability in CTE, the thicknesses of the veneering porcelain and the underlying core also have an impact on the chipping behavior of the veneering porcelain. To that end, studies have suggested that applying a minimum veneer thickness to enhance the adequate esthetic and functional needs would increase the strength of bi-layered restorations.<sup>6,15</sup> Furthermore, inadequate bond strength between the veneering porcelain and the zirconia core could be the major underlying factor that causes chipping.<sup>16</sup> Reportedly, factors that lead to inadequate bond strength between the zirconia core and the veneering porcelain include

flaws in the veneering porcelain created during layering, liner material application and variability in zirconia surface preparation.<sup>17</sup>

As an alternative to zirconia cores veneered with porcelain, monolithic (full-contoured) crowns made of LDGC may be employed due to their relatively high flexural strength (360 MPa) and better translucency compared to high crystalline zirconia. This seems to be a reliable alternative to bi-layered zirconia-based crowns.<sup>2,18</sup> The short-term clinical performance of monolithic crowns after 24-month observation was reported to be promising, with a survival rate of 98–100%.<sup>19,20</sup> An in vitro study demonstrated that bulk fracture occurred at higher load levels for the monolithic LDGC crowns compared to ones made of hand-layered veneers over zirconia cores due to better stress distribution.<sup>21</sup> Alternatively, CAD/ CAM milled LDGC veneers applied over zirconia cores increased the mechanical stability of the restoration and seemed to be a promising alternative to minimize chipping and fracture.<sup>18</sup>

The purpose of this study was to evaluate the effects of crown design (monolithic vs bi-layered), material and layering technique on the chipping resistance of all-ceramic molar crowns. The null hypothesis was that there is no effect of crown design, material or layering technique on the chipping resistance of all-ceramic crowns.

# Material and methods

A ceramic crown preparation was made on an epoxyresin mandibular 1<sup>st</sup> molar with axial wall reduction resulting in a 1 mm shoulder finishline. It was located 1 mm above the cementoenamel junction. Occlusal surface reduction was at least 1.5 mm. Line angles between occlusal and axial surfaces were prepared rounded. The prepared tooth was used to fabricate 50 replicas using a highly filled epoxy-resin (Viade Products Inc., Camarillo, USA). The replicas were placed in a dentiform with adjacent teeth on both proximal sides to simulate a clinical situation of a molar needing crown. The materials used for crown fabrication and cementation are listed in Table 1.

Tooth replicas were divided into 5 crown groups (n = 10) as follows: MLD – monolithic LDGC crowns; ZHL – zirconia copings veneered by hand-layering technique; ZVP – zirconia copings veneered by heat-pressing technique; ZLD – zirconia copings veneered with milled LDGC; PFM – porcelain-fused-to-metal crowns (control group).

For the MLD group, the prepared tooth, adjacent and opposing teeth were coated with a thin layer of optical reflective powder (IPS Contrast Spray, Ivoclar Vivadent, Schaan, Liechtenstein) and a CEREC 3D intra-oral scanner (Sirona Dental Systems, Bensheim, Germany) was utilized to capture optical images. A full-contour crown was virtually-designed (CEREC 3.84, Sirona) and milled out of an IPS e.max CAD block using a CEREC milling

#### Table 1. Material properties (according to the manufacturer's instructions data)

Material and lot	Compositions	Fabricating technique	Flexural strength (MPa)	CTE (×10 <sup>-6</sup> )
IPS e.max Ceram (S00837)	nano-fluorapatite glass-ceramic	manual application	90	9.5
IPS e.max ZirPress (P76153)	fluorapatite glass-ceramic	pressing technique	110	9.8
IPS e.max CAD (R67755)	lithium-disilicate glass-ceramic	CAD/CAM	360	10.2-10.5
IPS e.max ZirCAD (R71099)	yttrium stabilized zirconium oxide	CAD/CAM	900	10.8
Argely NP Supreme (35052 03/10)	Co: 61%, Cr: 27%, Mo: 6%, W: 5%	laser sintering technique	475	14.1
IPS d.Sign (R73590)	leucite glass-ceramic	manual application	80 ±25	12.6
IPS e.max CAD crystal/connect (R66132)	fusion glass-ceramic	manual application	160	9.5

CTE - coefficient thermal expansion; CAD/CAM - computer-aided design and computer-aided manufacturing.

unit (Sirona). The milled crowns were then subjected to a crystallization firing cycle in a Programat furnace (Ivoclar Vivadent). Glazing paste was applied to the outer surfaces of the crowns before placement in the furnace.

For the ZHL, ZVP and ZLD groups, replicas were digitally-scanned as described above and copings were virtually-designed. Copings of a uniform thickness of 0.5 mm were then milled from Y-TZP blocks (IPS e.max ZirCAD, Ivoclar Vivadent) using CEREC inLab 3.84 (Sirona). The milled copings were then subjected to final sintering in a furnace following the manufacturer's instructions. The sintered copings were then assigned to 3 groups according to the porcelain veneering technique. Ten zirconia copings were veneered with manually-added porcelain (IPS e.max Ceram, Ivoclar Vivodent) (ZHL), while another 10 copings were veneered with heat-pressed porcelain (IPS e.max ZirPress, Ivoclar Vivadent) (ZVP). IPS e.max ZirLiner was applied to establish a bonding between the veneer materials and the zirconia copings. For the remaining 10 zirconia copings, CAD/CAM-milled LDGC veneers (IPS e.max CAD) were fused to the zirconia copings using a glass fusion bonder (IPS e.max CAD crystall/connect) (ZLD).

For the PFM crowns, copings were virtually-designed (CEREC 3D 3.84, Sirona) with a 0.5 mm uniform thickness. They were then fabricated with laser-sintering technology using a non-precious metal alloy (Argely NP Supreme, IdentAlloy, Glastonbury, USA). The metal copings were manually-veneered (IPS d.Sign, Ivoclar Vivadent). One experienced dental technician fabricated all PFM crowns.

All crowns were tried-in onto their corresponding tooth replicas in order to confirm proper seating and adequate marginal fit. The crowns were then cemented to their replicas with dual-cured resin cement (Panavia F2.0, Kuraray Dental, Tokyo, Japan). For all metal and zirconia copings, the intaglio surfaces were grit-etched with 50  $\mu$ m aluminum oxide powder under 1 bar pressure for 5 s.<sup>22</sup> For the LDGC crowns, the intaglio surfaces were etched for 20 s with hydrofluoric acid (IPS Ceramic Etching Gel, Ivoclar Vivadent). The crowns were cleaned with distilled water in an ultrasonic bath. The surfaces were then coated with a layer of silane-coupling agent (Mono-Bond Plus, Ivoclar Vivadent). Each crown was seated onto its corresponding replica, excess cement was removed and Oxyguard II (Ku-

raray) was applied to cover the margins for 3 min. A 10 N load was applied onto the occlusal surface for 15 min. Light-curing was performed for 20 s on each of the crown surfaces. Overall length of a specimen (tooth + crown) was measured before and after cementation with a digital caliper to ensure complete seating of the crowns. The specimens were then stored in distilled water at  $37^{\circ}$ C for 1 week prior to the compressive cyclic loading test.

Cyclic loading was applied to each specimen in a universal testing machine (Instron 8501, Instron, Norwood, USA). The specimens were embedded in a special acrylic holder to ensure that loading was applied to the mesiobuccal (MB) cusp incline set at 30°. A testing chamber was filled with distilled water with the specimen secured at its center. Cyclic loading was applied using a cone shaped indenter applied at the center of the MB incline (Fig. 1). Each specimen was subjected to compressive cyclic load-



Fig. 1. Schematic illustration of load application at the incline of the mesiobuccal cusp

Sample	ZHL	MLD	ZVP	ZLD	PFM
1	8,513	500,000	2,291	500,000	40,957
2	29,240	500,000	19,347	500,000	98,984
3	1,120	500,000	9,852	500,000	49,279
4	3,514	500,000	41,191	500,000	320,712
5	1,692	500,000	2,133	500,000	1,458
6	24,009	500,000	31,467	500,000	8,504
7	13,924	500,000	47,169	500,000	17,096
8	4,484	500,000	1,282	500,000	4531
9	10,329	500,000	3,203	500,000	9,106
10	5,206	500,000	23,520	500,000	236,896

Table 2. Number of cycles at which each specimen failed. The lowest and highest numbers of cycles at which failure occurred are marked in bold. Specimens that reached 500,000 cycles did not fracture

ZHL – zirconia copings veneered by hand-layering technique; MLD – monolithic LDGC crowns; ZVP – zirconia copings veneered by heat-pressing technique; ZLD – zirconia coping veneered with milled LDGC; PFM – porcelain-fused-to-metal crowns; LDGC – lithium-disilicate glass-ceramic.

ing at 20 Hz for 500,000 cycles. Each cycle started at 50 N and completed at 450 N. Where possible, the machine was stopped after 250,000 cycles and the specimens were examined under light microscope to check for the presence of cracks. If no defects were detected, 250,000 additional cycles were applied and the specimens were then microscopically re-inspected. When chipping or fracture of the specimen occurred before the completion of the cycles, the specimen was deemed a failure and the number of cycles at which the event occurred was recorded.

The data was statistically analyzed using Fisher's exact test. Statistical significance was set at 0.05. The log-rank test was performed for comparing Kaplan-Meier survival curves for the failed specimens.



Fig. 2. Survival results for all groups. MLD and ZLD withstood the fatigue test with no failures, while ZHL, ZVP and PFM crowns underwent failure at different stages of the fatigue test

MLD – monolithic lithium-disilicate glass-ceramic crowns; ZLD – zirconia coping veneered with milled lithium-disilicate glass-ceramic; ZHL – zirconia copings veneered by hand-layering technique; ZVP – zirconia copings veneered by heat-pressing technique; PFM – porcelain-fused-to-metal crowns.

### Results

Monolithic LDGC crowns (MLD) and crowns made with zirconia copings and veneered with LDGC veneers (ZLD) survived the entire fatigue test without any failures. In contrast, all specimens in the PFM, ZHL and ZVP groups underwent failure at different points of the cyclic loading test (Table 2). Fisher's exact test revealed a statistically significant difference among the groups (p < 0.001). Therefore, the null hypothesis was rejected.

Bi-layered zirconia crowns veneered either by the handlayering technique or the press-on technique had the worst performance to chipping resistance. These were followed by the PFM crowns, which all failed before the fatigue testing was completed; however, after a number of cycles greater than those in the ZHL and ZVP groups. The Kaplan-Meier test revealed significantly higher survivability of the LDGC crowns (MLD) and zirconia crowns veneered with milled LDGC veneers as compared to the remaining 3 groups (Fig. 2).



Fig. 3. Chipping of the veneering porcelain of a specimen in the ZVP (zirconia copings veneered by heat-pressing technique) group (cohesive failure)



Fig. 4. Core/veneer interface delamination in a specimen in the ZHL (zirconia copings veneered by hand-layering technique) group (A) and PFM (porcelain-fused-to-metal crowns) group (B)

Bulk fracture was not observed with any of the specimens; however, cohesive failure within the veneering porcelain was frequently observed (Fig. 3). Core/veneer interface separation was observed in 2 ZHL specimens and 4 PFM specimens (Fig. 4).

# Discussion

Ceramic materials are prone to slow crack growth during cyclic loading in an aqueous environment. The combination of a moist environment and stresses during functioning increases the potential for crack propagation and reduces the load required for failure.<sup>23</sup> The test used in the present study was performed while the specimens were maintained under water in order to counteract heat buildup at the point of contact due to friction, and to keep the test conditions clinically-relevant. The load cycle selected (50-450 N) is within the range of occlusal biting forces encountered in the posterior region.<sup>24</sup> Considering that 2,700 chewing cycles per day are reported as the average for a young adult, this would add up to 1 million cycles per year.<sup>25</sup> However, since not every chewing cycle is as active as the one selected in the present study, it was suggested that the total number of cycles should be divided by a factor ranging from 5 to 20.26 Thus, 500,000 load cycles equate to 5-10 years of functioning. Based on this, it may be assumed that monolithic molar crowns made of LDGC and bi-layered molar crowns made of zirconia cores veneered with LDGC veneers would be expected to resist cuspal chipping from 5 to 10 years under the mechanical conditions of the oral environment. However, this finding must be interpreted with caution since posterior teeth are subjected to a variety of forces during functioning, and not only compressive ones. This includes shear forces that occur during lateral excursions of the mandible, which are

an integral component of the chewing cycle. In addition, there are other factors in the oral environment that may influence the performance of ceramic restorations such as temperature and pH fluctuations, enzymatic challenges, and muscular volume. Combined, these factors may result in a shorter survival term to cuspal chipping; however, only further research can determine their exact effect.

The incidence of veneering porcelain chipping or delamination in a bi-layered zirconia crown has been reported as a major complication in the dental literature.<sup>19,27</sup> Two reasons that could explain this incidence are core thickness and design. In the present study, the metal and zirconia coping were designed with even thickness. This results in an uneven porcelain veneer layer when anatomical features of the occlusal surface were recreated during the fabrication of the veneer. For a thick porcelain layer supported by zirconia coping with low thermal diffusivity, there is a higher risk of buildup of residual tensile stresses within the veneering porcelain layer.<sup>28</sup> Such stresses may promote crack propagation, and hence increase the veneering porcelain susceptibility to undergo chipping.

Both types of bi-layered zirconia crowns veneered with hand-layered veneering porcelain and pressed porcelain veneer failed prematurely at a comparable mean number of load cycles. This may be due to inadvertent inclusion of voids within the veneering porcelain during fabrication; however, this would be less likely to happen in the case of heat-pressed veneer. It may also be due to the low bond strength between the zirconia and the veneering porcelain.<sup>11,29</sup> Preis et al. observed outstanding fracture resistance of heat-pressed porcelain because of the improvement in the microstructure of the material.<sup>30</sup> In agreement with the findings of the present study, Stawarczyk et al. found a slightly better or similar fracture resistance when comparing press-on and manually-layered veneering porcelain irrespective of the material used.<sup>7</sup>

The fact that 1 experienced technician applied the porcelain may have maintained the incidence of voids within the manually-layered veneering porcelain at the same level. However, the variability in CTE between the veneering porcelain and zirconia has a detrimental effect on the bond strength and strongly influences the resistance ability of the veneering porcelain to chipping.<sup>12</sup> Ideally, the veneering porcelain should have a slightly lower CTE than that of the zirconia core in order to create slight compressive stresses within the veneering layer. These stresses might increase the bond strength between zirconia and the veneering porcelain.<sup>13</sup> Accordingly, the CTE mismatch between the veneering porcelain and zirconia coping used in the present study (Table 1) resulted in the optimal bonding between the 2 structures; however, this was not enough for the crowns to adequately resist cuspal chipping under conditions of fatigue testing.

In the present study it was observed that porcelain-veneered zirconia crowns are more susceptible to chipping than PFM crowns, and both were less resistant to mechanical fatigue than zirconia crowns veneered with LDGC veneers. This result is in agreement with findings reported in several clinical studies that compared the longevity of zirconiabased and PFM crowns.<sup>8,27</sup> The similarity of the mechanical properties and composition of the 2 veneering materials, IPS e.max Ceram and ZirPress, could explain their comparable chipping behavior. In contrast, the void-free and stronger veneering material (LDGC) resisted chipping in CAD-on crowns for the entire length of the mechanical fatigue test.

In the present study, cohesive failure occurred within the veneering layer in all crowns in the ZVP group. This indicates the presence of adequate bonding at the interface between zirconia coping and the porcelain veneer. Fischer et al. stated that porcelain chipping takes place within the porcelain layer rather than at the porcelain/ zirconia interface.<sup>31</sup> Adhesive fracture is less often observed in zirconia bi-layered restorations, and in the present study it was observed with in 2 crowns in the ZHL group.<sup>7</sup> On the other hand, PFM crowns showed higher chipping resistance compared to zirconia-based ones, and the fracture took place at the metal core/veneer interface in almost half of the specimens. However, PFM crowns resisted chipping a little better than the crowns belonging to the 2 bi-layered zirconia groups (ZHL and ZVP).

LDGC material with a flexural strength of 360 MPa increased the chipping resistance of monolithic and bilayered crowns compared to bi-layered ceramic crowns in which the veneering porcelain had a much less flexural strength of only 100 MPa.<sup>2,18</sup> In a recent study that compared the chipping behavior of manually-veneered zirconia crowns with CAD-on veneered ones under thermocycling and chewing simulation for 1.2 million cycles, 88% of the manually-veneered zirconia crowns failed during the chewing simulation, test while no failures were observed in the CAD-on crowns.<sup>18</sup> Generally, this is in agreement with the findings of the present study.

In the present study, monolithic LDGC crowns showed higher cuspal chipping resistance compared to crowns belonging to the ZHL, ZVP and PFM groups. This is in agreement with findings reported by Guess et al., in spite of variations in the test design.<sup>21</sup> The superior chipping resistance of monolithic LDGC crowns may be attributed to a number of factors. The e.max CAD blocks are manufactured under ideal manufacturing conditions, including operating in a vacuum, which results in minimizing the formation of voids or flaws. In addition, their microstructure includes fine grain lithium disilicate, which results in superior homogeneity.<sup>21</sup> Furthermore, the monolithic configuration of LDGC crowns eliminated the interface between coping and veneer, which is the weak link in the bi-layered complex, where many failure modes are located.<sup>32</sup> In a short-term clinical trial study, monolithic LDGC crowns (e.max CAD) showed successful outcomes with no technical complications such as occlusal chipping or fracture.<sup>20</sup>

Some of the limitations of the present study include lack of periodontal ligament simulation in the specimens. The specimens were rigidly attached to resin bases. This would not allow any mobility during the cyclic loading test. Having a simulated periodontal ligament in the specimen could have acted as a cushion and resulted in better stress distribution. In addition, the cyclic loading test was performed at a relatively high frequency (20 Hz) compared to what would be expected to occur in the oral environment, and indeed in comparison to the 1-2 Hz reported in other studies. However, Zahran et al. investigated the fatigue resistance of 2 all-ceramic crown systems where the compressive load cycles ranged from 50 N to 600 N at 20 Hz, and their results were comparable to those reported in other studies, where a lower cycle frequency was followed.<sup>33</sup> Therefore, perhaps the relatively higher cycle frequency followed in the present study had little or no effect on the outcome.

# Conclusions

Within the limitations of the present in vitro study, crown chipping occurred with all specimens of the bi-layered crown groups: zirconia copings veneered with manually-added porcelain, zirconia copings veneered with heatpressed porcelain and PFM crowns. All monolithic LDGC crowns and zirconia crowns veneered with LDGC veneers survived the entire 500,000-cycle compressive fatigue test without any failures. Therefore, it is concluded that the latter 2 types of all-ceramic crowns would be expected to perform clinically better in terms of resistance to chipping and fracture under occlusal loads of mastication.

For bi-layered crowns, core material (zirconia vs metal) had an effect on the resistance of the crowns to cuspal chipping, with metal copings providing better resistance to cuspal chipping.

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# Effect of water quantity and quality on the properties of alginate impression materials

# Wpływ jakości i ilości wody na właściwości alginatowych materiałów wyciskowych

Zbigniew Raszewski<sup>1,A,B,D,F</sup>, Agnieszka Nowakowska-Toporowska<sup>2,C,E,F</sup>, Joanna Weżgowiec<sup>3,C,E,F</sup>, Danuta Nowakowska<sup>2,A,D,F</sup>

<sup>1</sup> Spofa Dental Kerr Company, Jičin, Czech Republik

<sup>2</sup> Department of Dental Prosthetics, Wroclaw Medical University, Poland

<sup>3</sup> Department of Experimental Dentistry, Wroclaw Medical University, Poland

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):43-48

Address for correspondence Joanna Weżgowiec E-mail: joanna.wezgowiec@umed.wroc.pl

#### **Funding sources**

The study was financed from statutory funds of Wroclaw Medical University, Poland.

Conflict of interest None declared

Received on October 5, 2017 Reviewed on November 26, 2017 Accepted on January 8, 2018

DOI 10.17219/dmp/82179

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## Abstract

**Background.** Alginates are impression materials commonly used in prosthodontics and orthodontics. However, all these materials have some disadvantages, such as limited elasticity, tearing resistance and low dimensional stability.

**Objectives.** The aim of this research was to investigate the effect of various water quantities and qualities on changes in the properties of alginates.

**Material and methods.** Two alginates, Neocolloid and Tulip, were mixed with different volumes of water, water with calcium ions, or sparkling water with CO<sub>2</sub>. The dimensions, setting times, and hardness of the specimens were measured and Young's modulus was calculated. The significance of the difference between the mean values of different groups and the control group was assessed by Student's t-test or the Mann-Whitney U test.

**Results.** The dimensional stability changes of both alginate impression materials were statistically dependent on the quantity of water used for mixing. Sample storage over 24 h of samples prepared with +15% water led to 5.00% shrinkage for Neocolloid and 4.41% for Tulip. The setting times of Neocolloid and Tulip were significantly prolonged when the alginates were prepared with +15% water; the addition of calcium ions shortened the setting times of both alginates. Specimens mixed with the water containing Ca<sup>2+</sup> ions were characterized by greater hardness and Young's modulus values when compared to the alginate mixed with distilled water.

**Conclusions.** For mixing alginates, it is necessary to use the manufacturers' recommended mixing ratios between powder and water. To obtain the right setting time, hardness and elasticity, the application of distilled or demineralized water is advised.

**Key words:** hardness, dental impression materials, irreversible hydrocolloids, storage conditions, dimensional changes

**Słowa kluczowe:** twardość, dentystyczne materiały wyciskowe, nieodwracalne hydrokoloidy, warunki przechowywania, zmiany wymiarów

# Introduction

Alginates are impression materials commonly used in prosthodontics and orthodontics for preliminary or final impressions for acrylic appliances, preparation of provisional restorations, impressions of opposite dentition, fabrication of diagnostic and orthodontic models, sports mouthguards, bleaching trays, and more. The popularity of alginates results from the satisfactory reproduction of details, ease of use, very good hydrophilicity, and low cost. However, these materials have some disadvantages, such as limited elasticity, tearing resistance, and low dimensional stability upon storage. Despite the recommendations of most of the manufacturers, in clinical practice impressions are often not poured immediately after removal from the mouth, especially when the dental surgery and dental prosthetics laboratory are not in the same place. Such procedure influences the final precision, since, over time, alginates shrink as a result of the evaporation of water from their structure (dehydration) and spontaneous syneresis.<sup>1-3</sup> From the beginning of their use in dentistry, there have been many studies concerning a wide range of properties of alginates. Many authors have investigated the effects of different factors and conditions on the mechanical properties, dimensional accuracy and stability after storage on the basis of examinations of casts made from this type of impression material.<sup>4-12</sup> Additionally, the influence of the mixing method (manual or automatic) and disinfection procedure on the reproduction of details, elastic recovery and tensile strength of alginates have been analyzed.<sup>13</sup> Farzin and Panahandeh have shown that low storage temperature (4°C) prolonged the dimensional stability of alginate impressions.<sup>14</sup>

Alginates, like irreversible hydrocolloids, are very sensitive to the mixing ratio between powder and water. Manufacturers add suitable spoons for powder and measuring cups for water, but it is possible to use these accessories imprecisely, i.e., due to a problem with the meniscus of water or because of using powder which has been packed or settled in the container, particularly after storage over a long period of time. In such conditions, the obtained mixing ratio may differ from the recommended one, resulting in as much as 15% excess or shortage of water. Obviously, different mixing proportions between powder and water may change the setting characteristics and the viscosity of alginate impression materials.<sup>15</sup>

Additionally, not only the quantity, but also the quality of water has an impact on the properties of alginates. There are studies describing the influence of gypsum on the setting time of alginates. Calcium sulfate dehydrate is present in the powder of alginate impression material to enable cross-linking of alginic chains. However, in excess it may accelerate the setting process.<sup>16</sup> Moreover, despite the fact that alginate manufacturers strongly recommend distilled water without other ions, which could influence the setting time, tap water is frequently used in common dental practice. Due to a possibly high concentration of different ions, using tap water may also increase or decrease the setting time of alginates.

# **Objectives**

The aim of this study was to investigate the effect of various quantities and qualities of water on dimensional changes during storage, as well as changes of setting time, hardness and elastic modulus of alginate impression materials.

# Material and methods

#### Material

Two commercial alginates were used: Tulip<sup>®</sup> (Cavex, Haarlem, Holland) – a chromatic material, which changes its color during the mixing process, and Neocolloid<sup>®</sup> (Zhermack, Badia Palesine, Italy) without a color indicator. Tulip is an example of alginate with a lower mixing ratio between powder and water (17 g/39 mL) compared to Neocolloid (19.5 g/39 mL). Neocolloid is used for precise impressions. Powders and water were measured using a laboratory balance KPZ 2-05-3 (KPZ, Czech Republic).

#### Sample preparation

In total, 7 groups of samples (5 groups prepared with different quantities of water, 1 group with water with gypsum and 1 group with water with a higher concentration of carbon dioxide) were prepared for Tulip and Neocolloid alginates. For each experimental group, 3 samples were prepared and tested. In the first part of the experiment, the samples of 9 g of powder (Tulip or Neocolloid) were mixed with distilled water with 10% and 15% less water and 10% and 15% more water, respectively, than recommended by the manufacturer. However, preparation of the alginates with a 15% shortage of water was not possible because of a lack of sufficient volume of water to mix it properly with powder. For this reason, only the setting time of the sample prepared with 15% less water was measured, and the measurements of the other parameters (dimensional changes and Shore A hardness) were not performed. In the next part, the samples of both the alginates with the same weight, were mixed with water with calcium ions. Water saturated with calcium sulfate was obtained by immersion of 10 g of calcium sulfate dehydrate (Sigma-Aldrich, St. Louis, USA) in 200 mL of distilled water, and storage of this solution for 24 h in 100% humidity at the laboratory temperature. This kind of water saturated with calcium sulfate can reflect the situation of using the same bowl for mixing gypsum and alginate without a thorough washing. In the last part, the alginate samples were mixed with sparkling water Żywiec (Żywiec Zdrój Corp., Żywiec, Poland) saturated with 600  $\mu$ g/dm<sup>3</sup> of CO<sub>2</sub>, as an example of tap water quality occurring in some geographical regions.

The alginates were mixed manually in a rubber bowl for 45 s. The temperature of the powder and the water was 23°C. Metal molds were used to form rectangular samples with dimensions of 10 mm × 60 mm × 3 mm. To obtain the samples, the metal molds were placed on a glass plate. After that, open spaces in the molds were filled with the mixed alginate and then covered with a second glass plate. After the alginate was set, the glass covers were removed and the samples were removed carefully. All the samples were stored in polyethylene (PE) string bags with a wet cotton ball inside to maintain 100% humidity for the whole testing period and protect the samples from additional drying. For each group, 3 samples were prepared.

#### Dimensional changes

An optical microscope at a magnification of  $\times 20$  (Delta Optical Genetic, Mińsk Mazowiecki, Poland) was used to measure dimensional changes of the samples. Two metal pins (Renfert, Hilzingen, Germany) were inserted into the alginate samples at a distance of 5 mm from the ends of the sample on each side (Fig. 1). The distance ( $\pm 0.01$  mm) between the pins in the sample was measured after 30 min and 1, 2, 3, 4, 5, 6, and 24 h of storage of the alginates after the setting time. The first measurement was conducted after 30 min to allow the material to come back to its original dimensions after being removed from the metal form. This time interval was used to simulate clinical conditions with the time needed in dental practice for the recovery of the impression material after being removed from the patient's oral cavity.



Fig. 1. Measurement of distance between 2 pins inside the alginate sample for evaluating dimensional changes of the sample upon storage

#### Setting time

The cone & plate CAP 2000+ viscometer (Brookfield Engineering Laboratories, Middleboro, USA) was used to determine the setting times of the alginate materials. This instrument subjected the sample to an oscillating stress of constant amplitude and frequency and it measured the resulting oscillating strain profile. All procedures were made under standardized conditions at room temperature. Before starting the experiment, the incubator was set to 23°C and allowed to stabilize. After 30 s of mixing the powder with water, the material was loaded between the cones of the viscometer and the time was measured till the material set completely. The same procedure was adapted for the alginates mixed with gypsum and sparkling water. For determination of setting time, 3 samples were used for each group. The setting time was defined as the length of time taken for obtaining 40% reduction in chart width plus 30 s for the mixing process.

#### **Shore hardness**

Metal molds with a diameter of 50 mm and thickness of 6 mm were placed on to a glass slab, filled with the alginate and covered by the second slab. After 15 min, the alginate discs were carefully removed. Shore A Hardness Durometer (Sauter GmbH, Balingen, Germany) was used to determine the hardness of the samples loaded with 10 N for a period of 15 min.<sup>13</sup>

#### Calculation of Young's modulus

Young's modulus of the tested samples was calculated using Gent's equation, which determines the relationship between Shore hardness (s) and Young's modulus (E) for alginates: 6 measurements for each sample at each time point were performed and the mean value was calculated.<sup>17</sup>

#### Statistical analysis

The results were expressed as a mean value of 3 measurements for each sample. The same materials prepared with mixing ratios recommended by the manufacturer, with distilled water, were used as the control sample. The data was analyzed with the use of STATISTICA v. 12 (StatSoft Inc., Tulsa, USA) software. The significance of the difference between mean values of different groups and the control group was assessed using Student's t-test (for data distributed normally), with p-value of  $p \le 0.05$ , or the Mann-Whitney U test (for data not distributed normally) with p-value of  $p \le 0.05$ , to show the statistical significance.

# Results

# Effect of water quantity on dimensional changes

The results of the experiments showing the effect of water quantity on dimensional changes of the samples during storage are shown in Fig. 2 for Tulip and Fig. 3 for Neocolloid. The statistically significant differences between the dimensional changes of the tested samples prepared with -10%, +10% and +15% water and the control sample prepared with the recommended mixing ratios were observed for both the alginates at most of the time points. The dimensional changes observed for samples prepared with the water excess (+10% or +15%) were larger than with lack of water (-10%) both for Tulip and Neocolloid. After 24 h of storage, the Tulip samples prepared with +15% water shrank by  $-4.41\% \pm 0.02\%$  (for the control  $-1.57\% \pm 0.03\%$ ) and Neocolloid samples changed dimensions by  $-5.00\% \pm 0.02$ (for the control  $-1.57\% \pm 0.01\%$ ).



Fig. 2. Dimensional changes during storage of Tulip mixed with different volumes of water; grey color of markers denotes the control group for each time point, black color denotes differences significant at the 0.005 level, white color denotes differences not significant at the 0.05 level



**Fig. 3.** Dimensional changes during storage of Neocolloid mixed with different volumes of water; grey color of markers denotes the control group for each time point, black color denotes differences significant at the 0.005 level, white color denotes differences not significant at the 0.05 level

# Effect of water quality on dimensional changes

Both alginate impression materials mixed with non-distilled water in mixing ratios recommended by the manufacturer had different properties upon storage. The results of these tests are presented in Fig. 4 (for water with a higher concentration of calcium sulfate) and Fig. 5 (for acidic water with a higher concentration of carbon dioxide).



**Fig. 4.** Dimensional changes during storage of the alginates mixed with water with calcium sulfate; data related to the control group (the alginates mixed with the recommended volume of water, without calcium sulfate), black color of markers denotes differences significant at the 0.005 level, white color denotes differences not significant at the 0.05 level



Fig. 5. Dimensional changes during storage of the alginates mixed with water with carbon dioxide; data related to the control group (the alginates mixed with the recommended volume of water, without carbon dioxide), black color of markers denotes differences significant at the 0.005 level, grey color denotes differences significant at the 0.05 level, white color denotes differences not significant at the 0.05 level

After 24 h, for Tulip mixed with water with a higher concentration of calcium sulfate a contraction of -1.45% was observed and for Neocolloid it was -1.39% (Fig. 4). For the same materials mixed with water with higher contractions of carbon dioxide, the contraction was -2.18% for Tulip and -2.37% for Neocolloid (Fig. 5).

#### Setting time

The results of the measurements of setting time for both the alginate impression materials are presented in Fig 6. Preparation of alginates with a mixing ratio different than the ratio recommended by the manufacturers also had an impact on the measured setting time of both materials. A prolonged setting time was observed with the water excess, while water loss resulted in a reduced setting time. The most significant differences were observed when both alginates were prepared with +15% water.

The presence of calcium ions from calcium sulfate shortened the setting time of both materials most significantly – for Tulip 81 s as compared to 153 s for the control prepared with the manufacturer's recommended mixing ratio. A similar reduction of the setting time was observed



Fig. 6. Changes in setting time of Tulip and Neocolloid; data related to the control group (the alginates prepared with the recommended mixing ratios  $p \le 0.05$ ; \*\*  $p \le 0.005$ )

for Neocolloid (101 s as compared to 300 s for the control). The use of sparkling water caused significant prolongation of the setting time from 300 s  $\pm$ 3 s to 642 s  $\pm$ 3 s for Neocolloid material, while for Tulip, the setting time was reduced from 153 s  $\pm$ 3 s to 99 s  $\pm$ 2 s. For both materials the observed differences were statistically significant.

#### Hardness and Young's modulus

The results of the Shore A hardness measured 15 min after mixing and Young's modulus of both the alginate impression materials are presented in Table 1. The measured decrease of hardness and elasticity was related to the water excess. Samples mixed with water containing gypsum ions were characterized by increased hardness (32 Shore A for Neocolloid and 23 Shore A for Tulip). The same relationship was observed for Young's modulus – 0.78 MPa for Neocolloid and 0.44 MPa for Tulip, in relation to the alginates mixed with the recommended mixing ratios: 0.54 MPa and 0.40 MPa, respectively.

# Discussion

Alginate materials, as gel forms, contain more than 60% of water in their structures. During storage, water can evaporate and this process is responsible for the shrinkage of alginate impressions.<sup>1,2</sup> The impact of water quantity and quality on the setting time and mechanical properties of alginate impression materials have not been sufficiently clarified in previous studies.<sup>4-16</sup> In the present research, it was shown that the quantity of water had a significant influence on the setting process and viscoelastic behavior of both impression materials. Changing the recommended mixing ratios between powder and water caused changes of the setting time, Shore A hardness and Young's modulus. Additionally, the quality of water used for mixing with alginate powder is crucial for obtaining impressions of adequate quality. If water contains calcium ions, the setting time was reduced. This experiment revealed a reduced setting time from 300 ±2 s to 102 ±3 s for Neocolloid and from 153 ±2 s to 81 ±3 s for Tulip. This effect can be explained because of the fact that ions from the

second group of the periodic table can easily react with sodium alginate to form a gel. Calcium sulfate dehydrate is even added to the formulation of alginate powder (usually as 7–10% of the total weight) to cross-link the alginate. However, if more is added, the reaction may be disturbed, which should be avoided. For this reason, in dental practice it is necessary to use separate bowls, one for mixing alginate and the second for mixing gypsum products, to prevent the contamination between calcium ions and salt of alginic acid, i.e., sodium or potassium alginate, which is present in the alginate powder. In the presence of water, the monovalent ions (Na<sup>+</sup> or K<sup>+</sup>) are replaced by divalent calcium ions, which crosslink the alginate.

The changes of setting time in the group where sparkling water was used for mixing were statistically significant for both the alginates, but for Tulip the setting time was shortened, and for Neocolloid it was prolonged. The significant prolongation of the setting time of Neocolloid may be explained by the fact that sparkling water contains CO<sub>2</sub>. Being a week acid, it can form salts with ions, worsen their solubility in water and increase the setting time of Neocolloid. Therefore, from the clinical point of view, the use of water containing  $CO_2$  in the mixing procedure of alginates is not recommended. However, according to information from patents (US patent 2008/0057465 A1 and US patent 6,559,200 B1) and Buchan & Peggie publications, some producers have used magnesium oxide for preparing alginates, since it can easily react with CO<sub>2</sub> and neutralize it.<sup>18-20</sup> Quantity and quality of water also had an effect on the hardness and elastic modulus of the alginates. Increasing the water quantity resulted in a decrease of the material hardness and its elastic modulus. This may be related to the effect of water as a plasticizer. An excess of Ca(2+) can cross-link alginic chains more effectively and, as a result, the final hardness and elastic modulus could increase while the setting time could be reduced.

# Conclusions

The results of the present study suggest that for mixing alginate impression materials, it is necessary to use the manufacturers' recommended mixing ratio between pow-

Table 1. Hardness and Young's modulus of Neocolloid and Tulip prepared using different mixing procedures); data related to the control group (the alginates prepared with the recommended mixing ratios)

Group	Neoc	colloid	Tulip		
Group	hardness [Shore A]	Young's modulus [MPa]	hardness [Shore A]	Young's modulus [MPa]	
Recommended mixing ratio	25.6 ±0.5	0.53 ±0.02	21.8 ±0.4	0.40 ±0.01	
–10% water less	28.4 ±0.5**	0.63 ±0.02**	26.0**	0.54**	
+10% water excess	21.8 ±0.4**	0.40 ±0.01**	19.8 ±0.8**	0.33 ±0.03**	
+15% water excess	20.0 ±0.7**	0.34 ±0.02**	18.2 ±0.8**	0.28 ±0.02**	
Water with calcium ions	31.8 ±0.4**	0.77 ±0.02**	22.8 ±0.4*	0.43 ±0.01*	
Water with carbon dioxide	24.8 ±0.8*	0.50 ±0.03*	22.0 ±0.7	0.40 ±0.02	

\* p ≤ 0.05; \*\* p ≤ 0.005.

der and water. An excess of water increases changes of the alginate mass dimensions and reduces hardness and elasticity of these materials. A shortage of water causes the opposite effects. Moreover, to get the right setting time, hardness and elasticity of the final impression, the use of distilled or demineralized water is advised. The addition of water rich in calcium ions improves the mechanical parameters of the studied alginates but also accelerates their binding.

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# Estimating tooth retention by the number of present teeth in a middle-old-aged population: 3-year follow-up study in Korea

# Prognoza zachowania zębów na podstawie liczby obecnych zębów w populacji w wieku średnim – 3-letnie badanie kohortowe w Korei

Ga-Yeong Lee<sup>1,A-D,F</sup>, Sang-Baek Koh<sup>2,3,A,E,F</sup>, Nam-Hee Kim<sup>4,A,C,E,F</sup>

<sup>1</sup> Department of Dental Hygiene, Graduate School, Yonsei University, Seoul, Korea

<sup>2</sup> Department of Preventive Medicine, Wonju College of Medicine, Yonsei University, Korea

<sup>3</sup> Institute of Genomic Cohort, Yonsei University, Wonju, Korea

<sup>4</sup> Department of Dental Hygiene, Wonju College of Medicine, Yonsei University, Korea

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):49-56

Address for correspondence Nam-Hee Kim E-mail: nami71@yonsei.ac.kr

Funding sources None declared

Conflict of interest None declared

#### Acknowledgments

We sincerely thank 13 dental hygiene researchers who collected data from 2010 to 2014: Bo-Hyeon Ku, Hyo-Won Kim, Min-Young Lee, Hye-Yeon Seo, Ji-Eun Jeon, Hyun-Kyung Kim, Ji-Hyun Beack, Da-Yee Jeung, Yeun-Ju Kim, Eun-Mi Choi, Seong-Hyeon Chae, Hee-Jin Kim, and A-Ram Lee.

Received on October 11, 2017 Reviewed on November 10, 2017 Accepted on December 6, 2017

**DOI** 10.17219/dmp/81033

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# Abstract

**Background.** The number of individual teeth decreases with age, resulting in a decrease in masticatory capacity, and is an important indicator of oral health. However, it is difficult to estimate the number of present teeth on the basis of age alone.

**Objectives.** We aimed to determine whether tooth retention could be estimated by the number of present teeth in middle-old-aged individuals.

**Material and methods.** We used data from the Korean Genome and Epidemiology Study on Atherosclerosis Risk of Rural Areas in the Korean General Population (KoGES-ARIRANG). Subjects (40–75 years old) were invited to participate in a 3-year prospective follow-up survey conducted from 2010 to 2014. A total of 557 individuals (219 men and 338 women) took part in the study. Tooth retention was estimated from the number of present teeth by multivariate logistic regression analysis using SPSS v. 20.0.

**Results.** In total, 294 (52.8%) subjects retained teeth during a 3-year follow-up period. The number of present teeth and the proportion of subjects with complete tooth retention after 3 years decreased with increasing age. A greater number of present teeth in the baseline year was associated with complete tooth retention after 3 years in a greater proportion of subjects (p < 0.001). Logistic regression analysis revealed that the proportion of subjects with tooth retention in the Q4 quartile (28 teeth) was 9.17 times that in the Q1 quartile (less than 26 present teeth), even after adjusting for sociodemographic factors and oral health behavior in middle-aged individuals. In elderly individuals, tooth retention in the Q4 quartile (28 teeth) was 4.50 times that in the Q1 quartile (less than 17 teeth).

**Conclusions.** The number of present teeth could be used to estimate tooth retention over a 3-year period. This highlights the importance of promoting oral health care in middle-aged individuals before tooth loss occurs.

Key words: oral health, tooth loss, cohort study, preventive dentistry

Słowa kluczowe: stan zdrowia jamy ustnej, utrata zębów, badanie kohortowe, stomatologia prewencyjna

# Introduction

The number of teeth is an important indicator of oral health, particularly in the elderly population. The number of teeth decreases with age, resulting in a decrease in masticatory capacity.<sup>1</sup> Elderly individuals often choose to use a dental prosthesis, such as dentures or implants, to compensate for the loss of masticatory capacity.<sup>2,3</sup> However, these prostheses do not always fully restore mastication, and patients still experience discomfort.<sup>4-6</sup> Consequently, elderly patients commonly limit or exclude certain food items from their diet,<sup>7</sup> which leads to malnutrition and an increased risk of systemic disease.8 The number of teeth is also associated with accurate pronunciation during speech.9 Elderly individuals with few present teeth have difficulties in pronunciation, which could lead to social exclusion. These findings indicate that tooth loss can lead to a decrease in the quality of life in elderly individuals.<sup>10,11</sup> Therefore, retaining present teeth by preventing tooth loss is necessary for maintaining health during old age.

There have been many studies on the risk factors associated with tooth loss. Both sociodemographic factors, such as age, sex, marital status and cohabitation status,<sup>12,13</sup> as well as socioeconomic factors, such as educational level and income level,<sup>14,15</sup> have been associated with tooth loss. Poor lifestyle habits,<sup>16</sup> including smoking and drinking, and poor oral health behavior (e.g., improper tooth brushing, lack of deep cleaning and lack of dental checkups)<sup>17</sup> have also been associated with tooth loss. However, recently, the paradigm has shifted from identifying risk factors of disease to investigating disease-prevention factors.<sup>16–19</sup> Evidence has accumulated showing that prevention factors could help healthy people retain a healthy status.<sup>20,21</sup> From the perspective of oral health, it is important to investigate the factors preventing tooth loss.

The number of present teeth is associated with age and is an important indicator of oral health;<sup>22,23</sup> however, it is difficult to estimate the number of present teeth on the basis of age alone. A previous study reported that the number of present teeth in individuals at the ages of 50–59 years and 56–65 years was associated with tooth loss 6 years later.<sup>24</sup> However, in that study, it was not possible to estimate future tooth retention from the number of present teeth at baseline, because the authors used 2 separate cross-sectional or longitudinal studies to examine the effect of present teeth on tooth loss, but did not investigate the issue from the perspective of tooth retention.

# **Objectives**

The aim of this 3-year prospective cohort study was to determine if tooth retention in elderly individuals could be estimated from the number of present teeth during middle age.

# Material and methods

#### Study design and participants

The study protocol was approved by the Institutional Review Board of Wonju Christian Hospital in Korea (CR105024-026). All participants provided written informed consent.

We used data from the Korean Genome and Epidemiology Study on Atherosclerosis Risk of Rural Areas in the Korean General Population (KoGES-ARIRANG), which is a population-based, prospective cohort study assessing the prevalence of and risk factors for systemic and dental disease. In 2005, the KoGES-ARIRANG recruited individuals residing in rural Wonju in South Korea. Demographic shifts in the 8 districts involved are infrequent, and the population could thus be followed over a long time. The population was invited to participate in this study through the media, conferences, and telephone calls, among other methods.<sup>25</sup>

The survey was performed between March 2010 and August 2014 and included 1,726 adults (711 men and 1,015 women), aged 40–75 years, who underwent an oral examination and interview. Out of these, 899 (52.1%) subjects were selected for the study. We then excluded 296 subjects with an error in the number of lost teeth, 35 (3.9%) subjects who were edentulous at baseline, and 11 (1.2%) outliers who lost more than 9 teeth for unknown reasons during the follow-up period. The final sample size for the present analysis was 557 participants (219 men and 338 women) (Fig. 1). The participants completed a standardized dental history and lifestyle questionnaire, and underwent a comprehensive oral examination including counting the number of present teeth according to standard procedures at baseline and follow-up examinations.





#### Main outcome and exposure measures

The main outcome measure was tooth retention, i.e., without the loss of 1 or more teeth, during the 3 years following the baseline year. The main exposure variable was the number of present teeth. Present teeth were operationally defined as functional teeth with crowns and roots, excluding dental implants, dentures, and pontics. The number of present teeth was counted by trained dental hygienists. Trained dental hygienists also carried out the oral examination, using a tongue depressor, with the subjects seated on a chair.

#### Covariates

Sociodemographic variables were based on questions used during the interview: "What is the level of your education?" (elementary school or lower, middle school, high school or higher); "What is your marital status?" (married or unmarried, including being divorced and widowed); and "Who do you live with now?" (alone, with a spouse, with children).

Oral health behavioral variables were based on the following questions: "How many times did you brush your teeth yesterday?"; "Do you use any additional oral hygiene devices, such as dental floss or an interdental brush, mouth rinse products, an electronic toothbrush, or a waterpik?"; "Have you visited a dental clinic within the last 3 years?"; and "If so, what type of treatment did you receive?" (no treatment, oral examination, dental scaling and periodontal treatment, restorative treatment, endodontic treatment, prosthetics treatment, dental implant, or not sure). In the present study, we only analyzed the responses on prevention treatment, such as dental scaling and oral examination, i.e., we did not include the data on the other treatment types. Two to 4 weeks after the survey, the surveyor randomly invited 20% of the initial participants to complete the survey again, in order to evaluate test-retest reliability; the kappa  $\kappa$  value was 0.967.

#### Statistical analysis

In this study, as there were significant differences in the sociodemographic and baseline oral health characteristics between different age groups, the population was divided into "middle" (<60 years) and "older" (≥60 years) age groups for analysis.

A  $\chi^2$  analysis was performed to determine differences in tooth retention distribution according to the quartiles for the number of present teeth after the baseline year. A multivariate logistic regression analysis was used to identify the factors associated with tooth retention, and the odds ratio (OR) and 95% confidence interval (CI) were calculated. A crude model and 3 models adjusted for covariates were used to investigate how the number of present teeth in the baseline year affected tooth retention. The magnitude of the effect of exposure variables on tooth retention was identified using the crude model. Similarly, model 1 was used to identify the effects of exposure variables on tooth retention after adjusting for the sociodemographic characteristics; model 2 – after adjusting for oral health behavior; and model 3 – after adjusting for sociodemographic characteristics and oral health behavior. All statistical analyses were performed using SPSS v. 20.0 for Windows (IBM Corp., Armonk, USA), and a p-value <0.05 was considered statistically significant.

# Results

#### Participant characteristics

The mean age of the 557 participants at baseline was 57.9  $\pm$ 8.14 years (men: 59.5 years; women: 56.9 years); more women (n = 338, 60.7%) than men (n = 219, 39.3%) took part in the study. The sociodemographic characteristics of the participants are listed in Table 1.

Table 1. Sociodemographic and oral health behavior characteristics
of subjects in the baseline year

	Variables	n (%)
Total		557 (100.0)
Soci	odemographic characteristics	
Cau	male	219 (39.3)
Sex	female	338 (60.7)
	40–49	95 (17.1)
Anoluceral	50–59	233 (41.8)
Age [years]	60–69	177 (31.8)
	≥70	52 (9.3)
	elementary school or lower	190 (34.1)
Educational level	middle school	102 (18.3)
	high school or higher	265 (47.6)
	married	513 (92.1)
Marital status	unmarried	44 (7.9)
	alone	34 (6.1)
Cohabitation status	with a spouse	279 (50.1)
	with children	244 (43.8)
	Oral health behavior	
	at least once	59 (10.6)
Daily tooth brushing	twice	304 (54.6)
	3 or more times	194 (34.8)
Using oral hygiene	yes	184 (33.0)
devices	no	373 (67.0)
Visit to a dental clinic	yes	384 (68.9)
within 3 years	no	173 (31.1)
Oral examination	yes	80 (14.4)
within the last 3 years	no	477 (85.6)
Dental scaling within	yes	142 (25.5)
the last 3 years	no	415 (74.5)

The data was analyzed with a frequency analysis.

Out of the 557 participants, 294 (52.8%) subjects did not lose any further teeth during the 3-year follow-up period. The mean number of present teeth at baseline was 24.33  $\pm$ 6.45; the mean number of present teeth in the following year was 22.79  $\pm$ 7.47, and the mean number of lost teeth was 1.54  $\pm$ 2.53.

# Tooth retention according to the number of present teeth at baseline

The number of present teeth at baseline in the age group 40 to 49 was 27.4, and that in the  $\geq$ 70 age group was 20.7. After 3 years, 68.4% of the age group 40 to 49 at baseline and 30.8% of the age group  $\geq$ 70, respectively, retained all of their teeth. These findings indicate that the number of present teeth and the proportion of subjects with complete tooth retention after 3 years decreased with age (Fig. 2).

The distribution of present teeth in middle-aged individuals (<60 years) is shown in Table 2. For this analysis, we grouped the participants into quartiles ( $Q_{1,2,3,4}$ ) according to the number of present teeth at baseline. A higher num-



Fig. 2. Present teeth in the baseline year and tooth retention 3 years later \* tooth retention after 3 years means no tooth loss during the 3 years of follow-up; \*\* present teeth means the mean number of present teeth in the baseline year.

Table 2. Quartiles of present teeth by patient age group and tooth retention



Fig. 3. Tooth retention after 3 years according to the number of present teeth in the baseline year

 $^{\dagger}$  difference in tooth retention according to the quartile of present teeth using  $\chi^2$  tests (p < 0.001);  $^{\dagger}$  one or more teeth lost (%) represents the proportion of subjects who lost 1 or more teeth during the 3-year follow-up;  $^{\$}$  tooth retention (%) represents the proportion of subjects who did not lose 1 or more teeth during the 3-year follow-up.

ber of present teeth at baseline was associated with better tooth retention after 3 years (p < 0.001). In  $Q_{3,4}$  81.3% of the subjects showed complete tooth retention, i.e., the proportion of subjects with no tooth loss was greater than that of subjects with the loss of 1 or more teeth (Table 2, Fig. 3).

The distribution of present teeth in older individuals ( $\geq$ 60 years) is also shown in Table 2. Similar to the middle-aged individuals, a higher number of present teeth at baseline was associated with better tooth retention after 3 years (p < 0.001). In Q<sub>4</sub>, 61.5% of subjects showed complete tooth retention, i.e., the proportion of subjects with no tooth loss was greater than the proportion of subjects with the loss of 1 or more teeth (Table 2, Fig. 3).

# Association between tooth retention and the number of present teeth in the baseline year

In the middle-aged group, there was an association between tooth retention after 3 years and the number of present teeth at baseline in the  $Q_{3,4}$  group, after ad-

Quartiles of present teeth in the baseline	<b>n</b> (0/)	Moon (CDt	Too	th retention after 3 y	ears
year (the number of teeth)	n (%)	Mean ±50.	no	yes	p-value <sup>‡</sup>
Middle-aged (<60 years)	-	26.16 ±4.18	-	-	<0.001
total	-	_	212 (64.6)	116 (35.4)	_
Q <sub>1</sub> (2–25)	63 (19.2)	_	44 (69.8)	19 (30.2)	_
Q <sub>2</sub> (26–27)	72 (22.0)	_	36 (50.0)	36 (50.0)	_
Q <sub>3</sub> , Q <sub>4</sub> (28)	193 (58.8)	_	36 (18.7)	157 (81.3)	_
Older-aged (≥60 years)	-	21.70 ±8.05	_	-	<0.001
total	-	_	82 (35.8)	147 (64.2)	_
Q <sub>1</sub> (1–17)	55 (24.0)	_	41 (74.5)	14 (25.5)	_
Q <sub>2</sub> (18–25)	53 (23.1)	_	42 (79.2)	11 (20.8)	_
Q <sub>3</sub> (26–27)	56 (24.5)	_	39 (69.6)	17 (30.4)	_
Q <sub>4</sub> (28)	65 (28.4)	_	25 (38.5)	40 (61.5)	_

 $^{\dagger}$  mean ±SD – the mean number of present teeth and standard deviation;  $^{\ddagger}$  p-values are obtained from  $\chi^2$  tests.

Quartilas of patural tos	th in the baceline year		Tooth retentio	n after 3 years			
Qualities of natural tee	en in the baseline year	Q <sub>1</sub> (2–25)	Q <sub>2</sub> (2)	6–27)	Q <sub>3,4</sub> (28)		
	crude model	1	2.32* (1.	14–4.71)	0.10** (5.28–19.32)		
Middle aged (x60 years)	model 1	1	2.01 <sup>ns</sup> (0.	96–4.22)	9.55** (4.82–18.91)		
Mildule-aged (<00 years)	model 2	1	2.21* (1.	07–4.55)	9.70** (5.01–18.75)		
	model 3	1	1.90 <sup>ns</sup> (0.	89–4.04)	9.17** (4.60–18.30)		
		Tooth retention after 3 years					
	$Q_1 (1-17)$ $Q_2 (1-17)$		Q <sub>2</sub> (18–25)	Q <sub>3</sub> (26–27)	Q <sub>4</sub> (28)		
	crude model	1	0.77 <sup>ns</sup> (0.31–1.89)	1.28 <sup>ns</sup> (0.56–2.93)	4.69** (2.14–10.28)		
Older aged (>60 years)	model 1	1	0.69 <sup>ns</sup> (0.27-1.75)	1.28 <sup>ns</sup> (0.54–3.03)	5.03** (2.21-11.45)		
Older-aged (200 years)	model 2	1	0.76 <sup>ns</sup> (0.30–1.91)	0.92 <sup>ns</sup> (0.37–2.27)	3.99** (1.76–9.03)		
	model 3	1	0.69 <sup>ns</sup> (0.27-1.77)	1.04 <sup>ns</sup> (0.40–2.54)	4.50** (1.93–10.52)		

#### Table 3. Logistic regression analysis for the association between tooth retention and present teeth during follow-up

\* p < 0.05; \*\* p < 0.001; <sup>ns</sup> – non-significant; crude model – unadjusted; model 1 – adjusted for quartiles of present teeth in the baseline year and sociodemographic status (age, sex, educational level, marital status, cohabitation status); model 2 – adjusted for quartiles of present teeth in the baseline year and oral health behavior (tooth brushing, using dental hygiene devices, visiting dental clinic, oral examination, and dental scaling within 3 years); model 3 – adjusted for quartiles of present teeth in the baseline year, sociodemographic status, and oral health behavior.

justing for the sociodemographic characteristics (model 1). There was also a linear association after adjusting for oral health behavior (model 2). After adjusting for both sociodemographic characteristics and oral health behavior (model 3), the proportion of subjects showing complete tooth retention after 3 years in the  $Q_{3,4}$  group (28 teeth) was retained their present teeth about 9 times more than Q1 group (less than 26 teeth) (OR = 9.17; 95% CI, 4.60–18.30) (Table 3).

In the older age group, there was an association in the  $Q_4$  group between tooth retention after 3 years and the number of present teeth at baseline, even after adjusting for the sociodemographic characteristics (model 1) and oral health behavior (model 2). After adjusting for the sociodemographic characteristics and oral health behaviors (model 3), the proportion of subjects showing tooth retention after 3 years in the  $Q_4$  group (28 teeth) was retained their present teeth about 5 times more than Q1 group (less than 26 teeth) (OR = 4.50; 95% CI, 1.93–10.52) (Table 3).

## Discussion

Oral rehabilitation refers to the restoration and treatment of defective teeth.<sup>26</sup> The aim of replacing a missing tooth with a prosthesis such as an implant or a denture is to restore the esthetics and speech function of the patient. However, patients with prosthodontics have a lower ability to chew than those who have natural dentition, and hence quality of life decreases as well.<sup>6</sup> In other words, it is preferable to preserve natural teeth rather than perform restorations.

Death is inevitable for all, even in the absence of disease. However, tooth loss due to aging is not inevitable; i.e., tooth loss is not a normal phenomenon in elderly individuals, although many studies have reported that tooth loss is an outcome of aging and that age is an indicator of tooth loss.<sup>22,23</sup> Age alone cannot be used to predict the number of present teeth. In this prospective cohort study, we found that the number of present teeth in middle-aged/elderly participants was associated with tooth retention at an older age, even after adjusting for sociode-mographic status and oral health behavior. The number of present teeth might therefore help to estimate future tooth loss in elderly individuals.

The mean number of present teeth in the baseline year was  $26.16 \pm 4.18$  for the middle-aged group, and  $21.70 \pm 8.05$  for the elderly group. After adjusting for sociodemographic factors, such as age, sex, educational level, marital status, and cohabitation status, and for oral health behavior, such as tooth brushing, use of oral hygiene devices, oral examinations, and dental scaling, middle-aged subjects who had 28 present teeth at baseline retained 9.17 times more teeth than subjects who had <26 teeth. On the other hand, elderly subjects who had 28 present teeth at the baseline year retained 4.50 times more teeth than subjects who had <18 teeth in the baseline year. These findings suggest that the number of present teeth at baseline influenced tooth retention over the following 3 years.

Worthington et al. reported that fewer teeth at baseline was associated with greater tooth loss over 5 years in adults who regularly attended a dental clinic.<sup>27</sup> However, after adjusting for age, sociodemographic factors and lifestyle habits, such as smoking, no significant association was observed. In a nationwide cross-sectional study, Yoshino et al. demonstrated an association between the number of present teeth and tooth loss over a period of 6 years.<sup>24</sup> A smaller number of teeth in subjects aged 50–59 years and 56–65 years were associated with greater tooth loss after 6 years. Similarly, Eklund et al. reported that a higher percentage of subjects with fewer teeth became edentulous over a subsequent 10-year period, than did subjects with more teeth.<sup>28</sup> Our results correspond with these findings: the fewer present teeth an individual had at baseline, the greater the tooth loss that person would experience over the subsequent 3 years, even after adjusting for sociodemographic status and oral health behavior, which are known factors associated with tooth loss. Therefore, age per se was not the sole indicator of tooth loss. In fact, the number of present teeth in middle age influenced tooth retention to a greater extent than the number of present teeth at an older age. However, most oral health education programs target elderly individuals rather than middle-aged individuals. In addition, dental care is often not considered a public priority for government spending.<sup>29</sup> On the other hand, our findings suggest that preventive health policies should focus on the middle-aged or younger age groups, who have not yet experienced tooth loss.

This study has 4 notable limitations. Firstly, this study may have interview bias, as it was conducted using one-on-one interviews. The questionnaire comprised numerous questions (50), and the survey answers may have been affected by the interview environment. In addition, the participants answered the questions from memory, and recall bias may have occurred. However, the surveyors evaluated the test-retest reliability ( $\kappa = 0.967$ ). Secondly, the study participants consisted only of middle-aged and elderly individuals living in rural Korea, so it is unclear whether the data has the representativeness of the samples. However, it has been presented that the KoGES data could be generalizable to the Korean population by comparison between the disease prevalence in the subjects from the Korean Genome and Epidemiology Study (KoGES) data and the Korea National Health and Nutrion Examination Survey (KNHANES III) data.<sup>30</sup> Also, it should be noted that useful and meaningful data which revealed a relationship between sleep duration and the incidence of hypertension used the same data as in this study.<sup>25</sup> Thirdly, this study considered only sociodemographic characteristics and oral health behavior variables, and other known risk factors for tooth loss, including systemic conditions, such as diabetes, cardiovascular disease, metabolic syndrome, and general health status and lifestyle factors, such as smoking and drinking habits, were not considered. Finally, the confidence interval was not precise, because this study was conducted using subgroups of middle-aged and elderly individuals; hence, the sample size was small.

Nevertheless, this study focused on tooth retention, rather than tooth loss, to determine the preventive factors for tooth loss, and investigated the association between the number of present teeth and tooth retention over a 3-year period, using cohort data after adjusting for sociodemographic status and oral health behavior. We found that the number of present teeth could be used to predict tooth retention after 3 years.

# Conclusions

Our findings provide helpful information for developing a health policy for middle-aged individuals with no tooth loss; however, further studies are necessary to identify the factors underlying tooth longevity and to establish measures that can prevent tooth loss.

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# Comparison of oral-health-related quality of life in patients in the short- and long-term period following lower-facial injury and fractures – preliminary report

# Porównanie jakości życia związanej ze stanem jamy ustnej w obserwacji krótko- i długoterminowej u pacjentów po urazach i złamaniach dolnej części twarzy – obserwacje wstępne

Bogumił Lewandowski<sup>1,2,A,E,F</sup>, Ewa Szeliga<sup>3,A,B</sup>, Ewelina Czenczek-Lewandowska<sup>3,D</sup>, Dorota Ozga<sup>4,E</sup>, Aldona Kontek<sup>3,B,D</sup>, Małgorzata Migut<sup>1,2,C</sup>, Grzegorz Magoń<sup>3,C</sup>, Justyna Kosydar-Bochenek<sup>2,C</sup>, Adrian Kużdżał<sup>3,C</sup>

<sup>1</sup> Department of Maxillofacial Surgery, Frederic Chopin Clinical Voivodeship Hospital in Rzeszów, Poland

<sup>3</sup> Institute of Physiotherapy, Faculty of Medicine, University of Rzeszów, Poland

<sup>4</sup> Department of Didactics in Intensive Medical Care and Emergency Medicine, Faculty of Medicine, University of Rzeszów, Poland

A - research concept and design; B - collection and/or assembly of data; C - data analysis and interpretation;

D - writing the article; E - critical revision of the article; F - final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):57-62

Address for correspondence Dorota Ozga E-mail: gdozga@poczta.fm

Funding sources None declared

Conflict of interest None declared

Received on November 30, 2017 Reviewed on December 30, 2017 Accepted on February 12, 2018

**DOI** 10.17219/dmp/85255

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## Abstract

**Background.** Post-traumatic craniofacial injuries associated with bone fractures lead to serious morphological, functional and aesthetic complications which may negatively affect the physical and mental condition of the patient throughout the recovery period.

**Objectives.** The aim of this study was the evaluation of complaints and well-being in patients during the short- and long-term period following injury to the lower face, as well as an assessment of the effects of age and sex in the examined parameters.

**Material and methods.** The research group included 42 patients with injury to the lower face. The patients' well-being and the most common functional problems following treatment were assessed using Oral Health Impact Profile-14 (OHIP-14). Statistical analyses were performed using the Mann-Whitney U test and Spearman's rank correlation coefficient, with significance level assumed at p < 0.05.

**Results.** A period of 8 months following injury of the lower face was sufficient for the patients to achieve significant improvement in the quality of life. Improvement in well-being, according to OHIP-14, by approx. 11.9 points  $\pm$ 11.7 points, was highly significant (p < 0.001). The most frequent complaints included pain in the maxillofacial area, difficulties with consumption of food and dissatisfaction due to the necessary change of diet. The factors of age (p = 0.2839) and sex (p = 0.6047) did not significantly affect improvement in well-being.

**Conclusions.** During both the short- and long-term period following injury of the lower face, the most frequent complaints included pain in the maxillofacial region, problems with eating food and dissatisfaction due to change in diet. The study has shown that during the period of 8 months after the injury, there was a significant improvement in the quality of life assessed with OHIP-14. Age and gender do not significantly affect the quality of life after injury to the lower face.

Key words: quality of life, oral health, injuries of lower face

Słowa kluczowe: jakość życia, stan zdrowia jamy ustnej, urazy dolnej części twarzy

<sup>&</sup>lt;sup>2</sup> Department of Emergency Medicine, Faculty of Medicine, University of Rzeszów, Poland

# Introduction

Post-traumatic craniofacial injuries in adults and recovery thereafter are important clinical issues in contemporary medicine. According to statistics, in highly-developed countries injuries of this type are most commonly incurred during road traffic accidents.<sup>1,2</sup> It has been estimated that up to 60% of all traffic accidents lead to craniofacial injuries, and 62% of these involve motorcyclists.<sup>3</sup> Other frequent causes include falls, assaults, and sporting injuries.<sup>4,5</sup> Based on epidemiological data, it has been suggested such injuries are most frequently incurred by males in their twenties and thirties.<sup>6,7</sup>

Regardless of their intensity and origin, craniofacial injuries rank among the most dangerous, and frequently life-threatening traumas, and often require immediate surgery. This is linked with a significant risk of damage to the vital organs of the body located within the head and neck region, i.e. the central nervous system (CNS), eyes, ears, trachea, larynx and esophagus.<sup>8</sup> Severe craniofacial trauma, accompanied by bone fractures, results in serious morphological, functional and aesthetic complications, directly affecting the quality of the patient's performance in daily life and recovery.<sup>9</sup>

In the literature, we can find numerous reports related to the effects of conservative and surgical treatment applied to patients with oral and maxillofacial injuries, yet there are only a few studies discussing the problems encountered by patients in the post-operative period, including those related to both the physical condition of the stomatognathic system and mental well-being.<sup>10</sup> Additionally, it is important how patients understand the possible consequences, and relationship between maxillofacial impairments and daily functioning.<sup>11</sup>

This study was designed to evaluate the most frequent symptoms and complaints affecting the well-being of patients in the short- and long-term period following injury to the lower face. The aim was also to assess the effects of age and sex in the parameters examined.

# Material and methods

Selected from among 66 subjects, the study group comprised 42 patients, including 13 females (31%) and 29 males (69%) ranging from 17 to 59 years of age (mean 34.5 years ±13.2 years). The patients had surgical treatment of mandibular fractures at the Maxillofacial Surgery Clinic of the Fryderic Chopin Clinical Voivodeship Hospital in Rzeszów, Poland. The most common area of fracture was the body, condyle and angle of the mandible. Inclusion criteria were as follows: 1) an informed consent to participate in the research, 2) injury of the lower face as a result of trauma, 3) surgical treatment of the lower face, i.e. the mandible. The study was approved by the ethics committee of the Medical Department of the University of Rzeszów.

The assessments were carried out at 2 days and then 8 months after the injury to the lower face, with the use of the Oral Health Impact Profile (OHIP-14) questionnaire and additionally included questions specially designed for this purpose and addressing the characteristics of the study group and the patients' complaints preceding the surgery. The OHIP-14 scale developed by Slade and Spencer is used to assess the effects of impairments related to the oral cavity and their impact on the patient's quality of life. The questions focus on 7 domains, defined by the authors as: functional limitation, physical pain, psychological discomfort, physical, psychological and social disability, and handicap. The questionnaire contains 14 questions (2 in each of the 7 categories), addressing e.g. co-existing complaints, i.e. orofacial pain, discomfort during eating and problems with falling asleep. Responses are assessed on a 4-point Likert scale, where 0 means "never", 1 - "hardly ever" 2 - "occasionally", 3 - "fairly often" and 4 - "very often". Patients' score may be in the range from 0 to 56 points, and the highest number of points reflects the poorest oral health and well-being.<sup>12-13</sup> The results acquired were subjected to statistical analyses, with significance level assumed at p < 0.05 (marked with \*). The analyses performed using the Mann-Whitney U test and Spearman's rank correlation coefficient were verified with the Shapiro-Wilk test and Levene's test.

# Results

# Complaints related to the maxillofacial region in the short post-traumatic period

During the short period after the injury, i.e. 2 days after trauma, the patients' complaints mainly related to physical pain, followed by physical disability and psychological discomfort. Painful aching in the mouth, experienced very often – 4, or often – 3, was reported by a total of 50% of the subjects, while discomfort during eating by as many as 66.7%, which may have been linked to the lack of satisfaction due to the necessary change in diet, expressed by 54.8% of the subjects. A significant part of the group manifested poor psychological well-being, which was particularly linked with the awareness of the existing problems in the maxillofacial region, expressed by 54.8% of the patients. The data is shown in Table 1.

# Complaints related to the maxillofacial region in the long post-traumatic period

The findings of the assessment involving the same patients, and carried out 8 months after the trauma, are presented in Table 2. The most commonly persisting complaints, on the scale marked as experienced very often -4 or often -3, related to the same domains of the

Subscales	OHIP-14 (Oral Health Impact Profile)	n	Verv often	Fairly often	Occasionally	Hardly ever	Never
(domains)		(%)	veryonen		Occasionally		never
	1. Have you had trouble pronouncing any words because	n	4	9	9	7	13
Functional	of problems with your teeth, mouth or dentures?		9.50	21.40	21.40	16.70	31.00
limitation	2. Have you felt that your sense of taste has worsened because of problems with your teeth, mouth or dentures?		5	3	7	9	18
			11.90	7.10	16.70	21.40	42.90
	3. Have you had painful aching in your mouth?		7	14	9	6	6
Physical			16.70	33.30	21.40	14.30	14.30
pain	4. Have you found it uncomfortable to eat any foods because		16	12	2	7	5
	of problems with your teeth, mouth or dentures?	%	38.10	28.60	4.80	16.70	11.90
	<ul> <li>5. Have you been aware of problems with your teeth, mouth or dentures?</li> <li>6. Have you felt tense because of problems with your teeth, mouth or dentures?</li> </ul>		12	11	5	2	12
Psychological			28.60	26.20	11.90	4.80	28.60
discomfort			6	9	13	4	10
			14.30	21.40	31.00	9.50	23.80
	7. Has your diet been unsatisfactory because of problems	n	11	12	4	8	7
Physical	with your teeth, mouth or dentures?	%	26.20	28.60	9.50	19.00	16.70
disability	8. Have you had to interrupt meals because of problems		6	12	6	7	11
	with your teeth, mouth or dentures?		14.30	28.60	14.30	16.70	26.20
	9. Have you found it difficult to relax because of problems with your teeth, mouth or dentures?		5	9	10	6	12
Psychological			11.90	21.40	23.80	14.30	28.60
disability	tability 10. Have you been a bit embarrassed because of problems		7	10	8	8	9
	with your teeth, mouth or dentures?	%	16.70	23.80	19.00	19.00	21.40
	11. Have you been a bit irritable with other people because	n	4	6	4	9	19
Social	of problems with your teeth, mouth or dentures?	%	9.50	14.30	9.50	21.40	45.20
disability	12. Have you had difficulty doing your usual jobs because	n	5	7	5	5	20
	of problems with your teeth, mouth or dentures?	%	11.90	16.70	11.90	11.90	47.60
	13. Have you felt that life in general was less satisfying because	n	8	3	12	8	11
	of problems with your teeth, mouth or dentures?	%	19.00	7.10	28.60	19.00	26.20
Handicap	14. Have you been totally unable to function because	n	4	1	9	8	20
	of problems with your teeth, mouth or dentures?	%	9.50	2.40	21.40	19.00	47.60

Table 1. Complaints in the facial area of the cranium according to OHIP-14 - 2 days after trauma

quality of life which were most problematic during the short term period after the trauma; first of all, psychological discomfort linked with an awareness of the persisting problems, reported by 23.8% of the subjects, then physical pain, in particular in the oral cavity, reported by 16.6%, and dissatisfaction with the necessary change in diet by 14.3%.

## Comparison of OHIP's 14 parameters on the second day after trauma and 8 months later

The assessment conducted on the  $2^{nd}$  day after trauma showed the following results: out of the maximum possible score of 56 points on the OHIP-14 scale, the subjects' mean score was 25.1 points ±15.5 points. The lowest recorded value equaled 0 points (the best frame of mind) and the largest was 56 points (the poorest frame of mind). The median value in the first assessment was 24.5 points, which means that half of the subjects assessing their well-being on the OHIP-14 scale received a maximum of 24.5 points and the other half of the subjects scored at least 24.5 points.

The assessment performed with the OHIP-14 scale 8 months after the trauma showed the subjects on average scored 13.2 points  $\pm 13.8$  points. The median value in this case was only 9.5 points. The minimum and maximum values in the 2<sup>nd</sup> measurement were the same as in the 1<sup>st</sup> one, i.e. 0 and 56 points, respectively.

The identified improvement in well-being, according to OHIP-14, by approx. 11.9 points  $\pm$ 11.7 points (approx. 47.4% in comparison to the initial value) was highly significant, at the level of p < 0.001\*\*\* (p = 0.0000; Z = 4.87) in the Wilcoxon signed-rank test. The data is shown in Table 3.

A change in well-being between the measurement 2 days after the trauma and the measurement 8 months after the trauma was found in the case of 39 respondents (92.4%). Only 3 subjects (7.1%) were found with no change in the level of well-being.

Subscales (domains)	OHIP-14 (Oral Health Impact Profile)	n (%)	Very often	Fairly often	Occasionally	Hardly ever	Never
	1. Have you had trouble pronouncing any words because	n	2	2	5	8	25
Functional	unctional of problems with your teeth, mouth or dentures?		4.80	4.80	11.90	19.00	59.50
limitation	2. Have you felt that your sense of taste has worsened because of problems with your teeth, mouth or dentures?		2	3	1	10	26
			4.80	7.10	2.40	23.80	61.90
	3. Have you had painful aching in your mouth? I		4	3	7	10	18
Physical			9.50	7.10	16.70	23.80	42.90
pain	4. Have you found it uncomfortable to eat any foods because		3	5	6	15	13
	of problems with your teeth, mouth or dentures?	%	7.10	11.90	14.30	35.70	31.00
	5. Have you been aware of problems with your teeth,		6	4	6	10	16
Psychological	mouth or dentures?	%	14.30	9.50	14.30	23.80	38.10
discomfort	6. Have you felt tense because of problems with your teeth,	n	2	5	7	12	16
	mouth or dentures?	%	4.80	11.90	16.70	28.60	38.10
	7. Has your diet been unsatisfactory because of problems	n	2	4	2	14	20
Physical	with your teeth, mouth or dentures?	%	4.80	9.50	4.80	33.30	47.60
disability	8. Have you had to interrupt meals because of problems		2	2	6	6	26
	with your teeth, mouth or dentures?	%	4.80	4.80	14.30	14.30	61.90
9. Have you found it difficult to relax because of problems		n	2	3	3	14	20
Psychological with your teeth, mouth or dentures?	with your teeth, mouth or dentures?	%	4.80	7.10	7.10	33.30	47.60
disability	10. Have you been a bit embarrassed because of problems with your teath mouth or doptures?		3	3	6	13	17
	with your teeth, mouth or dentures?	%	7.10	7.10	14.30	31.00	40.50
	11. Have you been a bit irritable with other people because	n	3	1	3	11	24
Social	of problems with your teeth, mouth or dentures?	%	7.10	2.40	7.10	26.20	57.10
disability	12. Have you had difficulty doing your usual jobs because	n	2	2	4	6	28
	of problems with your teeth, mouth or dentures?	%	4.80	4.80	9.50	14.30	66.70
	13. Have you felt that life in general was less satisfying because	n	3	3	6	9	21
Handican	of problems with your teeth, mouth or dentures?	%	7.10	7.10	14.30	21.40	50.00
nanuicap	14. Have you been totally unable to function because	n	2	0	5	10	25
	of problems with your teeth, mouth or dentures?	%	4.80	0.00	11.90	2.80	59.50

Table 2. Complaints in the facial area of the cranium according to OHIP-14 – 8 months after trauma

Table 3. Comparison of the well-being of patients after injury of the lower face, after 2 days and 8 month later

Descriptive statistics [points]	2 days after surgery	8 months after surgery	Difference	Wilcoxon test
Mean ±SD	25.1 ±15.5	13.2 ±13.8	11.9 ±11.7	
Median	24.5	9.5	10.5	N = 39
Minimum	0.0	0.0	-13.0	Z = 4.87 p = 0.0000***
Maximum	56.0	56.0	32.0	

Fig. 1 illustrates the scores acquired by the respondents 2 days after the trauma, and 8 months later. It shows that the curve for the values 2 days after the trauma is located in the proximity of higher values on the "x" axis, and the results recorded 8 months after the trauma create a curve whose peak corresponds to lower values on the "x" axis. The distribution of the results confirms the statistically significant improvement in the patients' well-being after a longer period of time from the injury, as evidenced by the Wilcoxon test.



Fig. 1. Comparative assessment of patients' well-being, as measured with Oral Health Impact Profile (OHIP-14)

# Improvement in patients' well-being after injury of the lower face, depending on age and sex

Statistical analysis with the Spearman's rank correlation test has confirmed that there was no effect of age (p = 0.2839) or sex (p = 0.6047) of subjects in the level of improvement in their well-being after the trauma. The relationships were statistically insignificant, at p < 0.05, both for the factor of age in the Spearman's rank correlation test and for sex in the Mann-Whitney U test. The absolute values were in the range from |R = 0.0| to |R = 0.2|, which shows no correlations with these factors. The data is shown in Table 4.

## Discussion

Although they are characterized by significant strength and resistance to stress, skeletal structures of the facial part of the cranium, because of their location, are at high risk of traumatic injuries. Craniofacial injuries with bone fractures lead to various health-related consequences, including functional impairment of the local muscles and soft tissues. In view of the fact that normal functioning of the motor structures linked with the stomatognathic system is indispensable for such vital processes as breathing, articulation, mastication and swallowing of food, as well as sense of taste, any related dysfunctions may lead to significant discomfort experienced by patients, especially during the short post-trauma period.<sup>14,15</sup>

The present study was an attempt to identify the most common complaints connected with the physical and psychological well-being of patients following trauma located in the lower regions of the facial part of the cranium. From the clinical point of view, it seemed interesting to find out whether, and if so, what kind of, changes could be observed in patients' quality of life after traumatic injury; because of this, the assessment was performed 2 days and 8 months after the trauma. The results explicitly show significant improvement in the quality of life during that period, in comparison to the initial condition at the short post-trauma stage.

An assessment with the same research tool was carried out by Conforte et al. in a group of patients with craniofacial injuries, immediately after trauma, and then 30 and 60 days later.<sup>16</sup> The present findings corroborate those reported by Conforte et al., who also observed time-related improvements in the quality of life after mandibular fractures. Similarly, Kaukola et al. have reported significantly lower quality of life in patients with mandibular fractures in the short post-operative period in comparison to healthy controls.<sup>17</sup> Conforte et al. observed improvement in daily functioning within the period of 3 months after trauma, and they suggested that this length of time was sufficient for the patients to achieve a quality of life similar to that in the control group. The authors also reported that sensory impairments were the most burdensome consequence of the trauma.<sup>17</sup>

The present study shows slightly different findings. Analysis of the most common complaints in the short postoperative period shows that most patients reported strong pain in the injured area. The most commonly indicated problem was the functional impairment related to eating selected foods. A significant number of the subjects emphasized difficulties connected with eating and dissatisfaction resulting from adopting a liquid diet. Similar functional problems were pointed out by Omeje et al., who showed that in a group of subjects with mandibular fractures, during the short post-trauma period, the most significant complaints included problems with chewing and swallowing as well as limitations in the options of food to eat.<sup>18</sup>

In the literature, there are many publications discussing the effects of injuries to the mental state and the related risk of depression.<sup>19</sup> According to Rustemeyer and Gregersen, the most significant post-traumatic changes are connected with psychological discomfort and poorer social functioning.<sup>20</sup> The neuropsychological consequences of craniofacial injuries were also pointed out, e.g. by Snell et al., who reported a one-year follow-up observation of patients with severe injury to the craniofacial skeleton due to road accidents. They found that in 30% of the subjects, injuries to the facial skeleton may significantly impair the emotional sphere, and lead to personality changes and to poorer social performance. The authors also emphasized that a decrease in the quality of life could be observed not only in the victims but also in their close family.<sup>21</sup>

Notably, our study shows evidence that even after 8 months from the injury, the majority of the patients present an unsatisfactory mental condition, which suggests it is necessary to introduce early psychological support for these patients. The present study has not confirmed that gender affects improvement in the patient's well-being. Similar opinions were expressed by Rustemeyer and Gregersen, as well as Barros et al., who did not find significant differences in the assessment of the quality of life of female and male subjects following craniofacial surgery.<sup>21,22</sup>

Table 4. Improvement in patients' well-being after injury of the lower face relative to sex and age

Level of improvement	Age		Age		Spearman's R		p-value
Assessment of well-being – OHIP-14					-0.2	0.2839	
	Sex females males		Mann-Whitney U test				
Level of improvement			Z	p-value			
Assessment of well-being – OHIP-14	14.2 ±13.2	10.9 ±11.0	-0.52	0.6047			

Despite the observable improvement in oral-health related quality of life (OHIP-14), another important problem that should be investigated is the effect of duration of convalescence on the quality of life following craniofacial injury. Borgiel-Marek et al., basing on a study involving 89 patients with mandibular condyle fracture, have shown that favorable effects of treatment and recovery largely depend on the timing of rehabilitation, and only to a lesser degree on the method of surgical treatment.<sup>23</sup>

In the present study the authors have focused on assessing the quality of life resulting from complaints and general and psychological well-being of patients following injury of the lower face. It would seem justified to expand the assessment of the quality of life, especially following massive craniofacial injuries associated with multiple injuries, by examining the effects of specific treatment methods, and physical as well as manual therapy applied to the stomatognathic system, which may constitute a significant element of the multidisciplinary treatment process.<sup>24,25</sup> Complex collaboration between maxillofacial surgeons and physiotherapists may reduce the duration of convalescence and recovery.<sup>26,27</sup>

# Conclusions

During both the short- and long-term period following injury of the lower face, the most frequent complaints included pain in the maxillofacial region, problems with eating food and dissatisfaction due to change in diet. The study has shown that, during the period of 8 months after the injury, there was a significant improvement in the quality of life assessed with OHIP-14. Age and gender do not significantly affect the quality of life after injury to the lower face.

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# Factors associated with dental pain related to last dental visit among adult patients

# Czynniki związane z bólem stomatologicznym u pacjentów dorosłych podczas ostatniej wizyty w gabinecie dentystycznym

Muhammad Ashraf Nazir<sup>A,C–F</sup>

Department of Preventive Dental Sciences, College of Dentistry, Imam Abdulrahman Bin Faisal University, Dammam, Saudi Arabia

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):63-68

Address for correspondence Muhammad Nazir

E-mail: manazir@uod.edu.sa

Funding sources None declared

Conflict of interest None declared

Received on November 30, 2017 Reviewed on December 29, 2017 Accepted on January 17, 2018

**DOI** 10.17219/dmp/83039

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### Abstract

**Background.** Most dental visits are made to seek dental care to relieve dental pain. However, the factors associated with behavior regarding dental visits due to pain are not fully understood.

**Objectives.** The objective of this study was to assess the factors associated with dental pain related to last dental visit among adults.

**Material and methods.** The data was obtained from a cross-sectional study conducted on adults, aged 18–69 years, who visited public and private dental clinics in Dammam, Saudi Arabia. A pilot-tested, self-administered questionnaire was employed for data collection. Dental pain related to last dental visit was a dependent variable in the study. Gender, age, nationality, income and education levels, time since last dental attendance, type of dental clinic visited, and chronic systemic disease were important independent variables. The Pearson's  $\chi^2$  test and multiple logistic regression analysis were used.

**Results.** The participants returned 671 questionnaires. Most of the participants were females (75.4%), and the majority (68.0%) attended private dental clinics. The prevalence of dental pain as the reason of last dental visit was 70.4%. A logistic regression model showed that odds ratio for subjects who visited the dentist due to pain in  $\leq$ 1 year was 0.34 (95% CI = 0.24, 0.49) compared with patients who visited the dentist in >1 year. Similarly, male gender (OR = 0.64; 95% CI = 0.42, 0.97) subjects aged  $\leq$ 35 years (OR = 0.61; 95% CI = 0.40, 0.95) were less likely to make pain-related dental appointments than females and individuals over 35 years. The respondents who attended private dental clinics were 1.56 (95% CI = 1.12, 2.37) times more likely to visit the dentist because of pain than those who attended a public clinic.

**Conclusions.** Delayed dental visits, female gender, age over 35 years, and private clinic attendance were significant factors associated with dental consultations motivated by pain.

Key words: adult patients, last dental visit, dental pain

Słowa kluczowe: pacjenci dorośli, ostatnia wizyta u stomatologa, ból stomatologiczny

# Introduction

The utilization of dental care is critical to achieving, maintaining and promoting oral health, general health and well-being of individuals.<sup>1</sup> However, the past years have seen declining trends in the use of oral care services among adult populations, and it has decreased from 40.5% to 37% in the USA over a period of a decade (2001–2010).<sup>2,3</sup> Generally, patients avoid visiting dentists due to the cost of treatment, lack of perceived need for oral care, lack of insurance, transportation difficulty, lack of time, and dental anxiety.<sup>3–5</sup> Nevertheless, dental pain, a dental public health problem, is the predominant reason of dental attendance, and depending upon its duration and intensity, patients tend to take self-medication or visit a dental office, hospital emergency room, or physicians.<sup>6–8</sup>

Dental pain is the most common of different orofacial pains such as temporomandibular disorders, neuralgias and facial pains.<sup>9</sup> It can negatively influence chewing, swallowing, speaking, smiling, and productivity, and most dental visits are made to relieve dental pain.<sup>10</sup> In a national survey of Spanish general population, the prevalence of dental pain was 26.3% in adults aged 35–44 years.<sup>11</sup> The incidence of dental pain is also high, as a considerable percentage of individuals develop pain over a certain time period. For example, in a longitudinal study of a 2-year duration, 31.2% of subjects with no dental pain at the baseline developed dental pain.<sup>12</sup> Dental caries is the leading cause of dental pain.<sup>13,14</sup> In addition, pain frequently occurs as a result of abscess, loosening of teeth, bleeding gums, and broken teeth.<sup>14</sup>

According to Nuttal et al., 40% of dentate subjects suffered from pain during the last 1 year, which affected their activities of daily life.<sup>15</sup> Compared with 3.5% of Chinese adults who attended the dentist for routine dental care, 84.1% visited a dental office due to a toothache.<sup>16</sup> Fewer subjects (3%) who sought care for a routine dental checkup reported poor oral health than 15% of those who visited the dentist when in trouble.<sup>17</sup> Patients expressed poor satisfaction when they visited the dentist for pain relief.<sup>18</sup> Moreover, a recent study documented that dental attendance due to trouble with teeth was associated with the impact on individuals' quality of life.<sup>19</sup> Gender and race were found to be associated with the number of dental problems experienced during the last 10 years.<sup>6</sup> Being a single parent, an immigrant, self-perception of poor oral health, and low income are the predictors of dental attendance motivated by pain.<sup>20,21</sup> Likewise, male gender, younger age (<30 years) and high educational level were associated with preventive dental visits.<sup>16,22</sup>

While the factors related to different reasons of seeking dental care have been investigated, the available literature lacks a clear picture of the factors that influence oral care seeking behaviors of patients when in pain. Since most dental visits are made to relieve pain, understanding these underlying factors can help policy makers and other stakeholders develop appropriate oral health policies and programs. The present study was set out to assess the factors that are associated with dental pain related to last dental visit among adult patients in Dammam, Saudi Arabia.

# Materials and methods

This study presents a secondary analysis of data from self-administered questionnaires gathered from patients visiting private and public dental clinics in Dammam, Saudi Arabia. The ethical clearance (No. EA201713) was obtained from the Scientific Research Unit (an ethics committee) at the College of Dentistry, Imam Abdulrahman Bin Faisal University, Dammam, Saudi Arabia. Data collection started in March 2017 and was completed in August 2017. The study comprised a non-probabilistic sample, and patients who were willing to participate in the study were provided with questionnaires. The participants were informed about their right of voluntary participation and there were no consequences for them in case they refused to participate in the study. The objective of the study, including its potential benefits, was discussed with the participants. The study was conducted in full accordance with the World Medical Association Declaration of Helsinki. The patients were approached in person in the dental hospital of the college, and other public dental clinics and private practices in the city. They provided their consent by filling out the questionnaires. The questionnaire was developed in English and was translated into Arabic for its administration among Arab participants, while non-Arab study subjects received the English questionnaire. Piloting of the questionnaire was conducted on a group of patients to ensure its validity, feasibility and practicality. The privacy and confidentiality of patients were preserved during data collection, analysis and publication.

The participants' reasons of visiting a dental office were sought, and dental pain as the reason of last dental visit was the dependent variable in the present study. Age, gender, nationality, educational qualifications, income level, time since last dental visit, type of dental clinic visited, presence of chronic systemic disease, and family history of chronic systemic disease were the independent variables. Based on the participants' information, they were grouped into 3 income levels: low, middle and high. Educational qualification was categorized as high school education or less, and university education. The time since last dental visit included <6 months, 6 months–1 year and >1 year.

Field data was translated back into English and entered into a computer database. SPSS software (IBM SPSS Statistics for Windows v. 22; IBM Corp., Armonk, USA) was used for processing and analyzing the data. Percentages of participants were presented along with their confidence intervals (CI). The study variables were dichotomized. The Pearson's  $\chi^2$  test included bivariate analysis of participants' responses between those who visited the dentist because of dental pain and those with other reasons of dental attendance, and unadjusted odds ratios (OR) were calculated. Multiple logistic regression analysis was performed to calculate the adjusted odds ratios between 2 variables after controlling for other variables. Using backward stepwise logistic regression modeling, the final model with best predictive value was created. A 95% confidence interval and p-value <0.05 were used for statistical testing.

# Results

The study sample consisted of 671 participants (7.6% non-response or refusal). The age of the participants ranged from 18 years to 69 years with a mean age 33.6  $\pm$ 12.1 years. More females (75.4%) than males (24.6%) participated in the study. Saudi respondents made up the predominant proportion (89.9%) of the study sample. The prevalence of dental pain as the reason of last dental visit was 70.4%, and 52.7% visited a dental office in  $\leq$ 1 year. Slightly less than a half belonged to low and middle income groups individually, and 67.4% had undergraduate degree or higher education. Most subjects (68.0%) visited private dental clinics (Table 1).

Table 1. Distribution of responses of study participant
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Variables	Percentage (95% Cl)
Reasons of visiting dentist pain other reasons	70.4 (66.9, 73.8) 29.6 (26.1, 33.0)
Time since last dental visit <6 months 6 months–1 year >1 year	9.6 (7.4, 11.8) 43.1 (39.3, 46.8) 47.3 (43.5, 51.1)
Gender male female	24.6 (21.3, 27.8) 75.4 (72.1, 78.6)
Age ≤35 >35	60.1 (56.4, 63.8) 39.9 (36.2, 43.6)
Educational qualification ≤high school education ≥undergraduate degree	32.6 (29.0, 36.1) 67.4 (63.8, 70.9)
Income level low income middle income high income	46.0 (42.2, 49.8) 45.5 (41.7, 49.3) 8.5 (6.4, 10.6)
Nationality Saudi non-Saudi	89.9 (87.6, 92.2) 10.1 (7.8, 12.4)
Type of clinic private public	68.0 (64.5, 71.5) 32.0 (28.5, 35.5)
Presence of chronic systemic disease yes no	21.0 (17.9, 24.1) 79.0 (75.9, 82.1)
Family history of chronic systemic disease yes no	62.4 (58.7, 66.1) 37.6 (33.9, 41.2)

CI – confidence interval.

Bivariate analysis of the data shows that younger age ( $\leq$ 35 years), male gender and last dental visit in  $\leq$ 1 year had significantly lower OR of making last dental visit because of dental pain. On the other hand, attending a private dental clinic, presence of systemic chronic disease and family history of systemic chronic disease had significantly higher OR of pain-related dental attendance compared with those who visited a public clinic, who had no disease or family history of chronic systemic disease (Table 2).

Table 2.	Bivariate	analysis:	factors	associated	with	dental	pain	related
to last de	ental visit	i.						

Mandalah a	Unadjusted OR		
Variables	percentage (95% Cl)	p-value	
Time since last dental visit ≤1 year >1 year	0.37 (0.26, 0.52)*	<0.001*	
Gender male female	0.68 (0.47, 0.99)*	0.044*	
Age ≤35 >35	0.56 (0.39, 0.80)*	0.001*	
Educational qualification ≤high school education ≥undergraduate degree	0.93 (0.65, 1.32)	0.681	
Income level low ≥middle	0.77 (0.56, 1.08)	0.132	
Nationality Saudi non-Saudi	0.75 (0.42, 1.34)	0.333	
Type of clinic private public	1.57 (1.11, 2.22)*	0.010*	
Presence of chronic systemic disease yes no	1.69 (1.09, 2.62)*	0.018*	
Family history of chronic systemic disease yes no	1.40 (1.00, 1.97)*	0.048*	

OR - odds ratio; CI - confidence interval; \* statistically significant.

Almost similar estimates of OR were observed when multiple logistic regression analysis was performed. The male respondents, those aged  $\leq$ 35 years, and who visited the dentist in  $\leq$ 1 year had significantly lower OR of dental pain as the reason of last dental visit than female patients, individuals > 35 years and who performed dental visit in >1 year. The participants attending only a private dental clinic had significantly higher OR of seeking dental care due to pain than those who had a dental appointment in a public clinic. Income level, educational attainment, nationality, the presence of or family history of chronic systemic diseases were not significantly associated with pain-related patterns of dental attendance (Table 3).

Vestelae	Unadjusted OR	a valua
Valiables	percentage (95% Cl)	p-value
Time since last dental visit ≤1 year >1 year	0.34 (0.24, 0.49)*	<0.001*
Gender male female	0.64 (0.42, 0.97)*	0.033*
Age ≤35 >35	0.61 (0.40, 0.95)*	0.027*
Educational qualification ≤high school education ≥undergraduate degree	1.02 (0.70, 1.48)	0.930
Income level low ≥middle	0.88 (0.60, 1.31)	0.544
Nationality Saudi non-Saudi	0.62 (0.33, 1.16)	0.137
Type of clinic private public	1.63 (1.12, 2.37)*	0.010*
Presence of chronic systemic disease yes no	1.51 (0.92, 2.49)	0.101
Family history of chronic systemic disease yes no	1.27 (0.88, 1.83)	0.203

Table 3. Multivariate analysis: factors associated with dental pain-related last dental visits

OR - odds ratio; CI - confidence interval; \* statistically significant.

Interaction/moderation analysis demonstrated no significant interactions between different variables of the study (Table 4). The backward stepwise final logistic regression model revealed that OR for subjects who visited the dentist due to pain in  $\leq$ 1 year was significantly lower (OR = 0.34) compared with patients who had a dental appointment in >1 year. Likewise, male patients (OR = 0.64)

 Table 4. Interaction/moderation analysis between different variables of the study

Verielales	Unadjusted OR		
Variables	percentage (95% Cl)	p-value	
Time since last dental visit Gender	1.44 (0.65, 3.18)	0.365	
Time since last dental visit Age	1.59 (0.76, 3.34)	0.215	
Time since last dental visit Type of clinic	0.60 (0.28, 1.26)	0.179	
Age Gender	0.80 (0.37, 1.74)	0.583	
Age Type of clinic	1.04 (0.50, 2.16)	0.907	
Gender Type of clinic	0.86 (0.40, 1.86)	0.714	

OR - odds ratio; CI - confidence interval; \* statistically significant.

and age  $\leq$ 35 years (OR = 0.61) had significantly lower OR of making dental visits than females and patients >35 years. The participants who attended a private dental clinic were 1.56 times more likely to avail to dental services for pain than those who visited a public clinic (Table 5).

 Table 5. Multiple logistic regression final model: factors associated with dental pain-related to last dental visit

Variables	Unadjusted OR	n voluo
Variables	percentage (95% Cl)	p-value
Time since last dental visit ≤1 year >1 year	0.34 (0.25, 0.49)*	<0.001*
Gender male female	0.64 (0.43, 0.95)*	0.029*
Age ≤35 >35	0.56 (0.38, 0.84)*	0.005*
Type of clinic private public	1.56 (1.08, 2.26)*	0.016*
Presence of chronic systemic disease yes no	1.56 (0.95, 2.54)	0.076

OR - odds ratio; CI - confidence interval; \* statistically significant.

# Discussion

The present study described the factors that are associated with dental visits of patients suffering from dental pain. The understanding of these factors by health policy makers, educationists and dental professionals can improve access to oral care and minimize distressing sensations of pain experienced by most patients. It is known that poor utilization of dental services can delay a timely diagnosis, deteriorate oral conditions, compromise general health, and rarely lead to mortality.<sup>1</sup>

In the present study, dental pain as the reason of a dental visit was reported by 70.4% of subjects. This prevalent figure is lower than that reported by Liu et al., who found that 84.1% of adults visited a dental office due to a toothache.<sup>16</sup> On the contrary, the results of the study by Lacerda et al. showed that 18.7% of patients attended a dental clinic to seek oral care to relieve pain.<sup>13</sup> The authors pointed out that study participants had high access to dental services as they belonged to the working class, which could be the reason for such low prevalence. Similarly, Jaffar et al. observed that 1 in 4 industrial workers with pain seek dental treatment.<sup>23</sup> Moreover, Ekanayake and Mendis reported that 23% of patients who visited a dental teaching hospital required treatment because of dental pain.7 These discrepancies about prevalence estimates could be due to variations in data collection methods.<sup>13</sup> In addition, the distribution of oral diseases which are the underlying causes of dental pain varies in different populations.14

When the relation between a dental pain-related visit and the time since last visit was analyzed, there was a 34% lower chance of patients seeking dental attendance because of pain if individuals made a dental visit in  $\leq$ 1 year than in the case of a dental visit in >1 year. This finding is particularly significant, because the time duration between dental visits is essential for establishing evidence based recommendation for consulting a dentist in less than a year to avoid pain-related dental visits. Likewise, the previous study also demonstrated that individuals who performed regular dental visits for a routine checkup were 2.5 times less likely to experience dental pain.<sup>24</sup> Regular access to dental services is usually suggested to get an early diagnosis and prevent complications of oral conditions.

Our study revealed that the type of dental clinic is another predictor of visiting the dentist as a result of feeling dental pain. The individuals with access to private dental care were more likely (OR = 1.56) to perform a dental visit due to pain than those who used dental services in a public clinic. Dental pain is a disturbing experience that requires patients to seek immediate access to dental care and better quality of services. High patient satisfaction and improved quality of dental services were reported in private dental practices.<sup>25</sup> However, private oral care services are limited to a relatively small proportion of the population compared with the public sector that provides preventive services in addition to curative care to most people in the country.<sup>26</sup> On the contrary, the private sector broadly targets patients that require more esthetic, restorative and curative treatments. This might explain the reason why patients visiting private clinics have higher chances of making pain-related dental visits in Saudi Arabia.

The dental care utilization is more common in females than in males, and the female gender has higher chances for dental pain than their male counterparts.27,28 In contrast, men were 1.6 times more likely to report dental pain compared with women, and men were more likely to visit the dentist.<sup>20,24</sup> In the present study, males were less likely than females to visit a dental office to seek pain relief. Two explanations can be formulated to provide the basis for a discussion regarding these gender differences. Firstly, dental caries and tooth loss, common reasons of dental pain, are more prevalent in women than in men.<sup>29,30</sup> Secondly, pain perception varies between male and female genders as females tend to have a lower threshold and tolerance to pain than males. In addition, gender differences in pain can be related to social norms of pain expression and underlying biological mechanisms.<sup>31</sup> Therefore, women with a high prevalence of caries and tooth loss, and low pain threshold are susceptible to pain experience, and consequently perform frequent dental visits.

The findings of our study revealed that subjects aged  $\leq$ 35 years compared with individuals older than 35 years had lower odds (OR = 0.56) of dental consultation related to dental pain. In Saudi Arabia, lower caries experience (DMFT = 14.5) has been documented in adults aged

35-44 years than in older individuals of mean age 62 years (DMFT = 24.3).<sup>32</sup> Similarly, the prevalence of tooth loss increases with age, culminating in a sharp rise in individuals in their seventies.<sup>30</sup> It is reasonable to consider that a low distribution of caries experience and tooth loss in younger adults accounts for lower chances of seeking dental care because of pain. On the contrary, the studies have identified more young adults experiencing dental pain, which declined with advancing age.<sup>21,33,34</sup> These inconsistencies in age-related pain perceptions can be ascribed to the subjective nature of pain, which is influenced by patient's knowledge, belief, expectations, and culture.<sup>13</sup>

There was no significant influence of income and education on dental attendance in the present study. Patterns of dental visits are often associated with high education and income levels.<sup>16,20</sup> However, these factors may not have an influence on pain as the reason for dental visits. While our study provided valuable data, there are certain weaknesses that should be considered in interpreting study results. High estimates of prevalence of dental pain as a reason of dental visits in the present study can be attributed to data collection from dental clinics as opposed to a study with a random sample of adult population from the community. Typically for survey research, there is a possibility of recall bias due to inaccuracy in recollections about past experiences such as previous dental visits. Moreover, attention should be paid when generalizing these findings to patients who visit dental offices in other geographical locations of the country. It is suggested that a comparative study should be conducted using the same methods in another country in the future.

# Conclusions

Based on the data analyzed in the present study, it can be concluded that subjects who sought dental care in less than a year had lower chances of pain-related dental attendance. Similarly, males and young adults were less likely to receive dental consultation due to pain. Finally, there was a higher possibility of performing a dental visit because of dental pain if individuals had access to a private rather than public dental clinic. These findings particularly suggest enabling these subjects to use dental health services by improving accessibility of such services and oral health literacy, and by changing patient perception of the importance of oral care.

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# Risk factors in oroantral communication while extracting the upper third molar: Systematic review

# Czynniki ryzyka połączenia ustno-zatokowego podczas usuwania górnego trzeciego zęba trzonowego – systematyczny przegląd piśmiennictwa

Katarzyna Lewusz-Butkiewicz<sup>1,A-F</sup>, Kinga Kaczor<sup>2,A-F</sup>, Alicja Nowicka<sup>1,A-F</sup>

<sup>1</sup> Department of Conservative Dentistry and Endodontics, Pomeranian Medical University Szczecin, Poland <sup>2</sup> Doctoral Studies of the Faculty of Dentistry, Pomeranian Medical University, Szczecin, Poland

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):69-74

# Address for correspondence

Katarzyna Lewusz-Butkiewicz E-mail: kasia-lewusz@wp.pl

Funding sources None declared

Conflict of interest None declared

Received on September 15, 2017 Reviewed on November 13, 2017 Accepted on December 2, 2017

#### Abstract

The removal of the upper third molar is a procedure commonly performed in oral and maxillofacial surgery. Maxillary third molars are generally less difficult to extract than mandibular third molars. The surgical removal of maxillary third molars is usually associated with low complication rates and low morbidity. This procedure involves the risk of developing complications such as oroantral communication, displacement into adjacent anatomic spaces, fracture of the maxillary tuberosity, and root fracture. Orthopantomograms are the standard preoperative imaging modality, but there is no proven tool for predicting oroantral communication. New possibilities have been offered by cone-beam computed tomography (CBCT), which is increasingly used in dentistry and is an innovative technique that provides more information as it eliminates the superimposition of surrounding structures and allows the acquisition of 3-dimensional images and their qualitative assessment. The aim of this systematic review was to assess risk factors during the extraction of the upper third molar using orthopantomograms and CBCT.

Key words: cone-beam computed tomography, radiography, third molar, oroantral fistula

**Słowa kluczowe:** stożkowa tomografia komputerowa, radiografia, trzeci ząb trzonowy, połączenie ustno-zatokowe

#### DOI

10.17219/dmp/80944

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# Introduction

The development of the maxillary sinus begins in the fetal phase of life and ends with the eruption of the third upper molar. Maxillary sinus shape alterations may lead to the formation of a recess. Alveolar recesses are usually situated between the 2 laminas dura of the alveolar process. Less frequently, the recesses are located between the roots of the teeth, or the interradicular or interdental septa. An alveolar recess may cause a protrusion of the lateral tooth roots into the lumen of the maxillary sinus. Protruding roots may be only covered by a thin layer of mucosa as a consequence of bone lamina atrophy.<sup>1–3</sup>

The removal of the upper third molar (UTM) is a procedure commonly performed in oral and maxillofacial surgery. This procedure involves the risk of developing complications such as oroantral communication (OAC), displacement into adjacent anatomic spaces, fracture of the maxillary tuberosity, and root fracture.<sup>4–7</sup> The frequency of OAC after UTM extraction noted in the literature is 0.8%.<sup>8</sup> Oroantral communication of less than 2 mm may spontaneously heal within 48 h after extraction, OAC greater than 3–4 mm is not expected to resolve itself without intervention and requires surgical treatment.<sup>9</sup>

Orthopantomograms (OPG) are standard preoperative imaging modalities. However, the assessment of the exact position of UTM roots relative to the maxillary sinus using OPG can be confusing. This is due to the shortcomings of this imaging method, including magnification and distortion effects. Cone-beam computed tomography (CBCT) is an innovative technique that provides greater information as it eliminates the superimposition of surrounding structures, as well as allows the acquisition of 3-dimensional images and their qualitative assessment.<sup>7,10</sup>

Therefore, the aim of this systematic review was to assess any risk factors during the extraction of the UTM using orthopantomograms and CBCT.

# Material and methods

A systematic review (SR) of the literature concerning OPG and CBCT in the evaluation of risk factors of oroantral communication during the extraction of the third upper molar was executed using PubMed (MEDLINE), Scopus and ScienceDirect databases on February 24, 2017. The search equations used for different databases are presented in Table 1.

A time frame between January 1, 2007 and December 31, 2016 was applied. Only publications in English, including peer-reviewed journals, were considered. The titles and abstracts obtained during the electronic search were screened and evaluated by 2 observers for eligibility according to inclusion and exclusion criteria (Table 2). Studies not meeting the inclusion criteria were excluded from further evaluation.

Any discrepancies in the selection were settled through discussion. After verification with respect to the aforementioned criteria, an additional search took place focusing on the browsing references of the acquired studies. A Prisma diagram flowchart presents the selection scheme (Fig. 1).<sup>11</sup>

For the purpose of this study, a data extraction form was created (Fig. 2). One review author extracted data from the studies that were included, and another author checked the forms. Information obtained from the data extraction forms were as follows: the characteristics of the participants (age, gender, diagnosis, and previous treatment), imaging method, and imaging method reliability. This review was registered in the "PROSPERO international prospective register of systematic reviews" as CRD42017071690.

Table 2. Inclusion and exclusion criteria

Inclusion criteria	in vitro and in vivo human study articles in English orthopantomograms cone-beam computed tomography upper third molars imaging study group ≥5 full-text available journal articles measurements performed in metrical manner
Exclusion criteria	animal study articles without abstracts

Table 1. Search strategy in PubMed (MEDLINE), Scopus and ScienceDirect

PubMed	(((oroantral communication[TIAB] OR oroantral perforation[TIAB] OR sinus-perforation[TIAB] OR sinus membrane perforation[TIAB] OR "Oroantral Fistula"[Mesh]) OR ("Molar, Third"[Mesh] OR maxillary third molar[TIAB] OR maxillary wisdom teeth[TIAB])) AND ("Radiography, Panoramic"[Mesh] OR pantomography[TIAB] OR "Cone-Beam Computed Tomography"[Mesh])) AND ("2007/01/31"[PDat] : "2016/12/31"[PDat] AND "humans"[MeSH Terms] AND English[lang])
Scopus	((TITLE-ABS-KEY(oroantral AND communication) OR TITLE-ABS-KEY(oroantral AND perforation) OR TITLE-ABS-KEY(sinus-perforation) OR TITLE- ABS-KEY(sinus AND membrane AND perforation) OR KEY(oroantral AND fistula)) OR ((KEY(molar, AND third) OR TITLE-ABS-KEY(maxillary AND wisdom AND teeth)) AND (KEY(radiography, AND panoramic) OR TITLE-ABS-KEY(pantomography) OR KEY(cone-beam AND computed AND tomography)))) AND (LIMIT-TO(PUBYEAR, 2016) OR LIMIT-TO(PUBYEAR, 2015) OR LIMIT-TO(PUBYEAR, 2014) OR LIMIT-TO(PUBYEAR, 2013) OR LIMIT-TO(PUBYEAR, 2012) OR LIMIT-TO(PUBYEAR, 2011) OR LIMIT-TO(PUBYEAR, 2010) OR LIMIT-TO(PUBYEAR, 2009) OR LIMIT-TO(PUBYEAR, 2008) OR LIMIT-TO(PUBYEAR, 2007)) AND (LIMIT-TO(LANGUAGE, "English"))
Science Direct	pub-date > 2006 and pub-date < 2017 and (((tak(oroantral communication) OR tak(oroantral perforation) OR tak(sinus-perforation) OR tak(sinus-perforation) OR tak(sinus membrane perforation) OR key(Oroantral Fistula)) OR (key(Molar, Third) OR tak(maxillary third molar) OR tak(maxillary wisdom teeth))) AND (key(Radiography, Panoramic) OR tak(pantomography) OR key(Cone-Beam Computed Tomography)))


Fig. 1. Study flowchart according to the PRISMA statement<sup>11</sup>

Title of study						
Authors						
Vear of						
publication						
Imaging	orthonantomo	arams 🗆 — CB	CT 🗆			
nnaging Gi 1 1	ormopantomo					
Study aims						
Type of study	in vitro 🗌	in vivo 🗌				
Sample size	randomised 🗌 recruited 🗌					
BASELINE CHARACTERISTICS OF PATIENTS						
	Study group	Control group	Others	p-values		
Number of						
patients						
Age						
Mean/±						
Median/±						
Gender	female:	female:				
male: male:				Б		
BASELINE CHARACTERIZATION OF PATIENTS (continued)						
Information	Evaluated	Statistically	Notes			
		significant				
		differences				
Eastan A		between groups				
Factor A						
Into 1						
Validation		Var	No			
Validation		105	INO			
Number of observer						
Number of measurement						
(points angles plana etc.)						
(points, angles, plane, etc.)						
Does the conclusion correspond						
with the aims?						
Does the result correspond with						
the aims?	*					

Fig. 2. Data extraction form

#### **Data analysis**

The articles which were qualified to the study contained high heterogeneity in regard to the assessment method. Therefore, a meta-analysis was not conducted and qualitative synthesis was performed.

A quality assessment instrument (QAI) was used, utilized by Kuijpers et al., to assess the level of evidence for any retrieved studies (Fig. 3). Two reviewers used the QAI independent of each other, with disagreements between the observers being resolved through discussion.<sup>12</sup>

I. S	I. Study design			
A	<ol> <li>Objective – objectives clearly formulated</li> </ol>			
E	<ol> <li>Sample size – considered adequate</li> </ol>			
(	2. Sample size – estimated before the collection of data			
I	<ol> <li>Selection criteria – clearly described</li> </ol>			
E	$\dot{z}$ . Baseline characteristics – similar baseline characteristics			
F	. Timing – prospective			
(	B. Randomization – stated			
II. Study measurements				
H	I. Measurement method – appropriate to the objective			
I	Blind measurement – blinding			
J	. Reliability – adequate level of agreement			
III. Statistical analysis				
ŀ	<ol> <li>Dropouts – dropouts included in data analysis</li> </ol>			
I	. Statistical analysis – appropriate for data			
N	A. Confounders – confounders included in the analysis			
N	J. Statistical significance level – p-value stated			
(	<ol> <li>Confidence intervals provided</li> </ol>			
Maximum number – 15				
<ul> <li>✓ – should be checked if it satisfactorily fulfilled methodological criteria</li> </ul>				
0.	<ul> <li>– should be checked if it did not fulfill the methodological criteria</li> </ul>			
•	<ul> <li>– not applicable</li> </ul>			

Fig. 3. Level of evidence<sup>12</sup>

## Results

Out of 1154 articles initially identified, automatic rejection of duplicates by Mendeley Desktop v. 1.17.8 (Mendeley, London, England) resulted in 1123 articles. On verification and after taking into account the inclusion/ exclusion criteria, 1060 articles were excluded. Sixty-six full-text articles were read in their entirety by 2 observers. Fifty-four articles were excluded on the basis of exclusion criteria. The final number of articles included in the study was 8 (Table 3).

Authors	Year of publication	OPG/CBCT	Rating
del Rey-Santamaría et al. <sup>16</sup>	2006	OPG	molar angulation, surgical technique and radiological sinus proximity
Demirtas et al. <sup>19</sup>	2016	CBCT	position of the maxillary sinus relative to the maxillary third molars
Hasegawa et al. <sup>14</sup>	2016	OPG	proximity of the roots to the maxillary sinus floor (root-sinus [RS] classification), relationship between the maxillary second and third molars was classified according to the modified version of the Archer classification, depth of the maxillary third molar in the bone, angulation of wisdom tooth
Jung et al. <sup>21</sup>	2015	OPG and CBCT	eruption level of the maxillary third molars, available retromolar space, angulation, relationship to the second molars, number of roots, and relationship between the roots and the sinus
Lewusz et al. <sup>13</sup>	2015	OPG	position of teeth 18 and 28, distance between the apex of the maxillary third molar and the floor of the maxillary sinus, position of UTM in relation to the cementoenamel junction of the adjacent second molar
Lim et al. <sup>18</sup>	2012	OPG	modified version of the Archer classification of impacted maxillary wisdom, proximity to the floor of the sinus
Nedbalski et al.15	2008	OPG	distance between the most superior point and the most superior root of the tooth
Pourmand et al. <sup>17</sup>	2014	OPG	type of retention, the relation of the tooth root to the maxillary sinus

#### Table 3. Articles included

Out of 8 articles, 2 (25%) were evaluated as good according to the methodological quality score  $\geq 60\%$ .<sup>13,14</sup> The lowest level of evidence was 20% and the highest 60%.<sup>13–15</sup> One study was prospective, while the others were retrospective.<sup>16</sup>

Oroantral communication was found to be statistically significant (p = 0.0368) more frequently in higher age groups (above 40 years of age).<sup>17</sup> The risk of OAC oscil-



Fig. 4. Archer third molar classification

1 – mesioangular; 2 – distoangular; 3 – vertical; 4 – horizontal; 5 – buccoangular; 6 – linguoangular; 7 – inverted; a – the occlusal surface of the impacted tooth is approximately at the same level as the occlusal surface of the second molar – class A; b – the occlusal surface of the impacted tooth is at the middle of the crown of the adjacent second molar – class B; c–e – the occlusal surface of the impacted tooth crown is below the cervical line of the adjacent molar or even deeper, contiguously or even above its roots – class C<sup>20</sup> lated between 4.6 and 5.3%.<sup>16,17</sup> The risk of OAC was seen to increase with molar extraction complications, though the linear-trend  $\chi^2$  test failed to find any statistical significance ( $\chi_{LT}^2$  = 3.411; df = 2; p = 0.065). Surgical extraction without osteotomy carried a 4.2% risk and surgical extraction with osteotomy carried a 7.0% risk.<sup>16</sup>

The risk of OAC is higher for mesioangular teeth, ranging from 42.9 to 69.6%. It also increased in the case of Archer classification class B at 60.9%, type 3 root-sinus classification at 87% and modified root-sinus classifications, IV being 3.1% and V at 7.1%, respectively.<sup>14,17,18</sup>

Both Lim et al. and Demirtas et al. used Archer's classification to arrive at comparable results in evaluating UTM position, Lim et al., on the basis of OPG and Demirtas et al. on the basis of CBCT.<sup>18,19</sup>

The authors of the publications evaluated the risk factors for the occurrence of OAC during the extraction of UTM on OPG using several methods:

- evaluation of the position of UTM using Archer classification or its modification;<sup>13,14,18</sup>
- the position of UTM in relation to the floor of the maxillary sinus;<sup>13–15,17,18</sup>
- the position of UTM in relation to the adjacent tooth;<sup>13,14,16,18</sup>
- level of UTM retention.<sup>17</sup>

## **Results and discussion**

#### Archer classification and modification

Archer classification describes the position of the long axis of UTM relative to the long axis of the upper second molar (USM) and an evaluation of UTM retention (Fig. 4).<sup>20</sup> Position d matches class C (the occlusal surface of the impacted tooth crown is below the cervical



Fig. 5. The modified Archer classification class A – UTM is between the occlusal plane of USM and the cervical line of USM; class B – UTM is between the cervical line of the USM and the middle third of its root; class C – UTM is at or above the apical third of the root of USM.<sup>14</sup>

line of the adjacent molar or deeper, contiguously or even above its roots) showing a statistical significance associated with the occurrence of OAC during the extraction of UTM.<sup>13</sup> A similar relationship was obtained in the case of class B of the modified Archer classification according to Hasegawa et al. (Fig. 5).<sup>14</sup> Class B after modification was equal to class C before modification, and this meant that the results obtained in the articles could be confirmed.

# The position of UTM in relation to the floor of the maxillary sinus

The position of UTM relative to the maxillary sinus floor can be evaluated in a number of ways. A ruler or software to evaluate and edit OPG can be used to determine the length between the UTM apex and the maxillary sinus floor.<sup>13,15</sup> This method is not a particularly useful tool, and it does not show any statistical significance regarding the assessment of the risk of OAC during UTM extraction.



Fig. 6. Root-sinus classification

type 1 – clear distinction between the 2 features (the tooth and the sinus floor); types 2 and 3 – different degrees of radiographic superimposition of the sinus floor across the roots; type 4 – close proximity of the sinus and the roots but clear demarcation between the sinus floor and the roots; type 5 – indistinct relationship between the roots and the sinus floor.<sup>14</sup>

Root-sinus (RS) classification and its modifications can be a valuable tool in the prediction of OAC during UTM extraction. An increased risk of OAC occurs in type 3 of the RS classification (Fig. 6), and type 4, and type 5 (the sinus floor extends up to the trifurcation/tooth cervix) of the modified RS classification, according to Pourmand et al.<sup>14,17</sup>

# The position of UTM in relation to the adjacent tooth

The Archer classification is used to evaluate the position of UTM in relation to the adjacent tooth. In publications that used this tool, it was shown to offer no statistical significance for predicting OAC during the UTM extraction.<sup>13,14,18</sup>

The UTM angulation in relation to the long axis of USM can be described according to Winter's classification. Unfortunately, in this study it was not included in the description of the results.<sup>16</sup>

#### The level of UTM retention

The degree of UTM retention is described by the following classification: type 1 – tooth bud; type 2 – root growth still incomplete; type 3 – retention with normal positioning of the axes; type 4 – mesially angled tooth; type 5 – distally angled tooth; type 6 – tooth turned horizontally toward the alveolar process. The highest incidences of OAC were observed in type 5 (7.1%) and type 3 (4.9%), while no complication was observed in the other types.<sup>17</sup>

Before extracting UTM, it is important to thoroughly analyze the anatomy of the tooth and its relation to the maxillary sinus. It is a standard practice to take OPG before the planned procedure. If the root and sinus floor are superimposed on the OPG, the relative probability of OAC increases during extraction.<sup>17</sup> In this situation, it seems necessary to obtain more information about the maxillary sinus relative to UTM in order to avoid OAC, and carry out CBCT.<sup>21,22</sup> It is difficult to measure the distance between UTM and the floor of the maxillary sinus. OPG are related to defects such as overlapping anatomical structures, enables horizontal and vertical magnification, and a 2-dimensional representation of a 3-dimensional structure.<sup>8,15,18,23</sup> There was a significant difference between OPG and CBCT measurements.<sup>21</sup>

Unfortunately, there are no strict guidelines regarding the role of CBCT in dentistry. It has become a substitute for conventional radiography, including periapical, bitewing and OPG. The radiation doses from full FOV dental CBCT scans have been measured to be 4–42 times the dose from OPG. An increasing number of CBCT images are performed in children who are more sensitive to radiation, particularly in the thyroid gland, testes, and breast tissue as the cancer risk per Sievert is the highest at a younger age. It is proposed to modify the concept of ALARA (as low as reasonably achievable) to ALADA (as low as diagnostically acceptable).<sup>24</sup>

## Conclusions

Orthopantomogram assessment is not a reliable method for assessing the risk factors for oroantral communication, but CBCT seems a better tool for the assessment of the proximity of an UTM to the maxillary sinus. For this reason, if information from orthopantomograms is not clear and there is a risk of a complications, CBCT should be performed to verify the information.

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# Oral *Helicobacter pylori*: Interactions with host and microbial flora of the oral cavity

# Helicobacter pylori w jamie ustnej – interakcje z mikrobiotą gospodarza

Paweł Krzyżek<sup>1,A–D,F</sup>, Grażyna Gościniak<sup>1,E,F</sup>

Department of Microbiology, Wroclaw Medical University, Poland

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):75-82

Address for correspondence Paweł Krzyżek E-mail: krojcerpawel@gmail.com

Funding sources None declared

Conflict of interest None declared

Received on September 18, 2017 Revised on November 21, 2017 Accepted on December 16, 2017

#### Abstract

The role of the oral cavity as a reservoir for *Helicobacter pylori* is still a controversial issue. There is a great number of articles indicating the presence of this pathogen in the oral cavity, but discrepancies among techniques for *H. pylori* detection and the variations in the patients tested often make it difficult to formulate a final verdict. Outer membrane proteins (OMPs) are one of the most important factors determining colonization of *H. pylori* in the oral cavity. Among them, the key role is attributed to BabA, SabA and NapA, all of which promote adherence and retention within this area. The oral cavity is characterized by the co-existence of numerous microorganisms which may potentially affect the physiology and morphology of *H. pylori*. The presence of coccoid-stimulating factors and relatively low levels of Al-2 in the early- to mid-stages of supragingival plaque allow dental *H. pylori* to colonize this niche as nonculturable spherical forms. On the other hand, subgingival plaque characterized by high numbers of periopathogens, capable of synthesizing high concentrations of Al-2, may favor the presence of mixed populations of spiral and coccoid *H. pylori* to interact both with the host and the local microflora of the oral cavity.

Key words: biofilm, Helicobacter pylori, oral cavity, dental plaque, microflora

Słowa kluczowe: biofilm, Helicobacter pylori, jama ustna, płytka nazębna, mikroflora

DOI

10.17219/dmp/81259

#### Copyright

© 2018 by Wroclaw Medical University and Polish Dental Society This is an article distributed under the terms of the Creative Commons Attribution Non-Commercial License (http://creativecommons.org/licenses/by-nc-nd/4.0/) Every living organism, to maintain the stability of its internal environment, must constantly adapt at the molecular, cellular and physiological levels. The gastrointestinal tract is the largest surface of the body exposed to the external environment. Therefore, the presence of mucous membranes and a well-functioning immune system is essential to protect the host organism.<sup>1</sup>

The first barrier in the digestive system, including the oral cavity, is the mucus layer. The mucus matrix forms a strongly hydrophilic colloid, mainly composed of water (about 95%), but also lipids (phospholipids, cholesterol), nucleic acids, polysaccharides and proteins (antibodies, defensins, growth factors).<sup>2</sup> Antibodies found in saliva belong mainly to 2 classes, i.e., IgA (90-98%) and IgG (1-10%). Their presence contributes to the inactivation and opsonization of microorganisms, which are then detected by the antigen presenting cells (APCs) and phagocyted. In the oral cavity, antibody production comes through the stimulation of B cells, located within the mucosa-associated lymphoid tissue (MALT). The main region of their recruitment is nasopharynxassociated lymphoid tissue (NALT), which consists of the adenoids and palatine tonsils of Waldeyer's ring. Salivary defense proteins play an important role in the protection of the oral environment. These include, but are not limited to: lysozyme, bactericidal increasing proteins (BPIs), palate lung and nasal epithelial clone proteins (PLUNCs), salivary amylase, cystatins, proline-rich proteins, statherin and peroxidases. The function of these proteins is to adsorb to the microbial surface and form ion channels, which leads to the microorganism's destruction. In addition, some of these compounds also possess immunomodulating activity and the ability of chelating metal ions from the local environment (Fe<sup>3+</sup>, Ni<sup>2+</sup>, Cu<sup>2+</sup>).<sup>3</sup>

Another important component of mucus is the high molecular weight oligomeric glycoproteins, namely mucins. More than 20 mucins covering the epithelial surface of the respiratory, digestive and reproductive tracts have been detected in the human body.<sup>4</sup> MUC1, MUC4, MUC19, MUC5B and MUC7 are produced in the oral cavity, while the dominant and most important groups are the last two.<sup>4,5</sup> MUC5B is a compound with a molecular weight in excess of 1000 kDa and consists of disulfide-bonded subunits.<sup>3</sup> MUC5B is a member of the gel-forming mucin family, secreted by the submandibular, sublingual, palatine and labial mucous cells of the salivary glands.<sup>4</sup> MUC7 is a secretory mucin present in the form of monomers (180-200 kDa) or dimers. It is located mainly in the submandibular and sublingual salivary glands.<sup>3,4</sup> Mucins and defense proteins play a crucial role in the formation of acquired pellicle, i.e., a thin layer (0.5–1  $\mu$ m) of saliva components with the ability to bind to calcium hydroxide. These components have receptors for microbes and, thus, in the surface immune exclusion process, contribute to the immobilization of microorganisms and protection against host tissue invasion.<sup>3,5</sup> Despite the protective function, this process also has a disadvantageous aspect associated with the formation of biofilm by microorganisms colonizing the oral cavity.<sup>3,4</sup>

Populations of microbes often form highly organized complexes called biofilms. These structures are made up of microbial cells embedded within the extracellular matrix, which accounts for nearly 90% of biofilm and consists of exopolysaccharides, proteins, lipids and extracellular DNA (eDNA). Biofilm provides a protective environment against adverse conditions including temperature, pH and oxidation-reducing potential fluctuations, presence of antimicrobial agents (antibiotics, enzymes) and the action of innate and adaptive immune systems. Moreover, besides the protective function, biofilm also determines co-adhesion and aggregation of the cells, formation of a strongly hydrated environment (protection against drying) and sorption of organic and inorganic compounds from the surrounding environment.<sup>6</sup> The formation of biofilm is also related to another important process, supervising the physiology of microorganisms, called quorum sensing (QS). In the course of this mechanism, microbes communicate using small molecular signaling molecules, which allow them to sense their density and integrate actions that resemble multicellular organisms.7 With respect to the intestinal flora, the oral cavity is one of the most colonized areas in the human body. The number of bacterial species localized in this region is estimated at 600-800.8 In addition to the enormous diversity of microorganisms, the oral ecosystem is characterized by succession, microbial composition variations and changes in conditions within the different morphological regions of the oral cavity.9 For this reason, oral biofilm is a very dynamic environment modified by availability and type of nutrient substances, pH, microbial toxic metabolite concentration, host immune response and shear forces.<sup>10</sup> The surface of the teeth is covered by the saliva components forming the acquired pellicle.9 This area is colonized by microorganisms capable of linking to these components, referred to as early colonizers, i.e., Streptococcus spp., Actinomyces spp., Gemella spp., Veillonella spp., Selenomonas spp., and Capnocytophaga spp.<sup>8-11</sup> Fusobacterium nucleatum is known to act as the "bridge bacterium". This is due to the long, filamentous shape and numerous multivalent adhesins exposed on the surface of this bacterium. These features make it possible to co-adhere with Streptococcus spp. and recruit Gram-negative rods (late colonizers), including: Porphyromonas gingivalis, Tannerella forsythia, Treponema denticola, and Aggregatibacter actinomycetemcomitans.<sup>8,12</sup> In many scientific studies, it is also suggested that the oral cavity may be a reservoir site for Gram-negative, microaerophilic *Helicobacter pylori*.<sup>5,13–17</sup>

# Colonization of the oral cavity by *H. pylori*

*H. pylori* infections occur most often in childhood, and clinical symptoms may develop over many years. This microorganism inhabits the gastric mucosa, where it is able to survive despite the therapy used. Persistent infections induce chronic gastritis, which is associated with the risk of developing gastric or duodenal ulcers and gastric cancers.<sup>18</sup> Transmission from person to person seems to be the main mechanism of the bacteria spreading. It is suggested that environmental reservoirs, such as contaminated water and food or infected animals, also exist. The factors influencing the frequency of *H. pylori* infection include age, sex, genetic predisposition, immunological status, and hygiene level.<sup>16</sup>

The outer membrane proteins (OMPs) presence is a crucial factor determining host colonization by *H. pylori*. It is estimated that 4% of the genome of this bacterium encodes information for the production of these structures.<sup>19</sup> The ability of direct adhesion to host epithelial cells is a process responsible for persistent infections, because it contributes to the protection against the removal of microbes from the mucous layer and enables delivery of virulence factors to eukaryotic cells.<sup>19,20</sup>

One of the most important *H. pylori* adhesins is BabA (blood group antigen binding adhesin A), which allows interaction with fucosylated Lewis b (Le<sup>b</sup>) and H type 1 antigens. For this reason, the protein has a strong affinity to MUC5AC, mucin secreted in the stomach environment. Additionally, BabA also determines adhesion with saliva components, i.e., MUC5B, proline-rich proteins and salivary agglutinin (gp-340).<sup>20</sup>

The second essential adhesin produced by *H. pylori* is SabA (sialic acid-binding adhesin). This protein promotes the adherence of bacteria to the inflamed stomach tissue, which is characterized by increased mucin sialylation (the process of linking sialic acid residues to Lewis antigens). This property also affects the ability of *H. pylori* to associate with salivary glycoproteins, such as MUC5B, MUC7, carbonic anhydrase VI, zinc- $\alpha$ -2-glycoprotein or heavy chains of sIgA1. The expression of SabA is regulated by the pH of the environment, and the amount of *sabA* mRNA decreases under acidic conditions.<sup>20</sup> This feature makes it possible that in more neutral pH conditions (e.g. in the oral cavity) SabA production can play a key role in *H. pylori* oral mucosa colonization.

The third vital adhesin expressed by almost all clinical *H. pylori* strains is NapA (neutrophil-activating protein). This protein has the ability to stimulate myeloperoxidase secretion and the production of reactive oxygen species (ROS) by neutrophils. Therefore, this factor most likely contributes to the release of nutrients from destroyed tissues.<sup>21</sup> *H. pylori* NapA has been shown to be capable of binding to the sulfur oligosaccharides of saliva, i.e., sulfo-Le<sup>a</sup>, sulfo-galactose and sulfo-*N*-acetyl-glucosamine.<sup>22</sup>

The significance of these proteins was confirmed by an oral H. pylori virulence factors analysis, because the presence of all these adhesins was detected in this microbe. Furthermore, the ability of oral *H. pylori* strains to secrete factors associated with tissue damage, inflammation initiation and nutrient uptake, i.e., VacA (vacuolating cytotoxin A), CagA (cytotoxin-associated antigen A), and serine protease HtrA, was observed.<sup>23</sup> The ability of *H. pylori* to cause pathological changes in the oral cavity is uncertain. In the stomach environment, this bacterium has many factors facilitating persistent colonization of the gastric mucosa. These properties include the ability to invade eukaryotic cells, disrupt cell-cell contact, promote inflammation, inhibit immune cell activity (including T cells) and induce excessive host cell proliferation.<sup>24</sup> H. pylori colonizing the oral cavity is also suggested to be involved in the development of various ailments in this region, such as halitosis, glossitis, recurrent aphthous stomatitis and dental caries.<sup>25</sup> Studies on the human acute monocytic leukemia cell line (THP-1) have shown that these microorganisms may be responsible for causing pathological changes in the oral cavity. The main factor responsible for this effect was CagA, because strains producing this protein contributed to significant increases in inflammatory cytokine secretion (IL-6, IL-8, IFN- $\gamma$ ) as compared to non-producing strains.<sup>26</sup> The capacity of *H. pylori* to interact with the host and modulate the local environment was also demonstrated by the ability of this bacterium to induce elevated MUC5B and MUC7 levels by three- and twofold, respectively. The increase in the amount of these components, which are receptors for oral H. pylori, can promote colonization and retention within the oral cavity.<sup>27</sup> Despite this, there is a need to increase the number of studies showing a direct relationship between *H. pylori* infection/colonization and oral pathophysiology.

# Epidemiology of oral *H. pylori* infections

There are many diagnostic methods for the detection of H. pylori, but none of them are 100% specific and sensitive. Invasive methods require removal of the gastric mucosa. Histopathological examination allows for accurate assessment of the degree and type of the gastric mucosa inflammation. Hematoxylin and eosin staining are used for the visualization of inflammatory cells, while Giemsa or Genta staining serve for microorganism detection. The sensitivity of the method is 89-91% and the specificity is 91–100%. Culture methods have a very high specificity (about 100%), but much lower sensitivity (78–94%). In addition, the culture of this bacterium is necessary to determine the sensitivity of *H. pylori* to antibiotics. Rapid urease testing is the simplest, cheapest and fastest test determining urease activity in biopsy specimens. This method has a sensitivity of 82-94% and a specificity of 96-96%. PCR is a highly useful method because, apart from the detection of bacteria (the highest accuracy in 16S rRNA gene detection), it also provides information about the production of virulence markers, such as cagA or vacA. Improved PCR methods with colorimetric detection of *H. pylori* DNA using isothermal helicase-dependent amplification result in sensitivity and specificity of up to 95%. Non-invasive methods are divided into active (detecting active *H. pylori* infection) or passive (showing evidence of contact with bacteria). Among the active assays are: the fecal antigen test which is used to determine the presence of *H. pylori* antigens in feces (sensitivity and specificity 92% and 94%, respectively), and the urease test. The latter is based on the detection of *H. pylori* urease activity during active infection. The level of labeled CO<sub>2</sub> derived from the decomposition of the labeled urea <sup>13</sup>C is measured. Sensitivity and specificity are 91% and 94%, respectively. Serological tests belong to passive assays and are based on the detection of IgG anti-*H. pylori* antibody levels in the plasma. The biggest disadvantage of these tests is the lack of the ability to delineate between active infection and temporal exposure to H. pylori antigens. This method is characterized by a high sensitivity (90–97%), but a low specificity (50-96%).<sup>28,29</sup>

The role of the oral cavity as a reservoir for *H. pylori* is still a controversial issue. There are a great number of articles indicating the presence of this pathogen in the oral cavity, but discrepancies among techniques for H. pylori detection and the variations in patients tested often make it difficult to formulate a final verdict. One of the newest and more accurate meta-analyzes conducted by Sayed et al. suggests that dental plaque can be an alternative, extragastric site of *H. pylori* residence.<sup>14</sup> Briefly, the frequency of H. pylori detection depends on the diagnostic technique used. The prevalence of bacteria detection using urease tests in most studies ranged between 50% and 100%. PCR studies have even greater divergence in results, i.e., 0-100% (of which only 7/35 studies showed a frequency greater than 50%). Culture methods have a low sensitivity, with prevalence lower than 50% (half of studies with prevalence lower than 10%). Inhomogeneous results were obtained using immunoassays (2 studies with frequencies above 65% and 2 with very low prevalence, 0% and 11%).14

Namiot et al., in oral *H. pylori* detection, used immunoassays intended routinely for fecal antigen detection.<sup>30–32</sup> The authors suggest that these tests can be successfully used to detect *H. pylori* in the oral cavity after appropriate sample preparation (preincubation in a microaerophilic atmosphere). This procedure increases the incidence of *H. pylori* more than twice.<sup>31</sup> Using this method, a high prevalence of these microorganisms in dental plaque was reported, i.e.,  $58.7\%^{32}$  and  $65.6\%.^{30}$  It was suggested that plaque reduction and oral health had no effect on the amount of antigens of these bacteria.<sup>32</sup>

Interesting results were obtained using molecular techniques. The presence of *H. pylori* in the oral cavity of patients with gastric H. pylori was reported in 46% of the subjects. Moreover, a relationship between these bacteria and increased approximal plaque index (API) and bleeding on probing (BOP) values has been noted. Within strains colonizing the oral cavity, only 16% expressed *cagA*, suggesting that this virulence factor is not critical in causing pathological changes to the oral cavity mucosa.<sup>33</sup> In another study using PCR, the DNA of oral H. pylori was isolated more frequently in patients with oral ailments (leukoplakia and oral lichen planus; 20% and 23%, respectively) than in those without these diseases (0%). The potential of these microorganisms to promote inflammatory conditions in the oral environment was proposed on this basis.34

There is also a relationship between the presence of pathological gingival pockets and *H. pylori* infections. Umeda et al. found that in patients with gastric *H. pylori* infection, the detection frequency of these bacteria in the oral cavity was 4-fold higher (41.2%) when patients had gingiva with a pocket depth of  $\geq$ 4 mm than those with healthy periodontium (9.1%).<sup>35</sup> Similar observations were made in a large-scale epidemiological study of 4504 patients by Dye et al.<sup>36</sup> It has been shown that pocket depth greater than 5 mm correlates positively with *H. pylori* seroprevalence. Despite the ability of *H. pylori* to colonize dental plaque, this area is not the only site of colonization. Other regions include the tonsils and dorsal mucosa of the tongue.<sup>15,37</sup>

Many studies based on genetic or serological tests in H. pylori detection have shown the presence of this bacterium in the oral cavity or stomach, while the number of studies demonstrating the effective isolation of this microbe from the oral environment is still very low. Hirsch et al.<sup>23</sup> successfully managed to isolate live *H. pylori* from samples taken from dental canals. In the morphological observation, using field-emission scanning electron microscopy, the presence of a mixed population of spiral rods and aggregates of coccoids was observed. Cultivation of the samples taken from supragingival plaque was unsuccessful, suggesting that in this niche, *H. pylori* occurred only in the coccoid form. The presence of particular H. pylori morphological forms in various areas of the oral cavity may indicate that the physicochemical parameters and local microbiological flora may affect the transformation process of *H. pylori*.

# Interactions of oral *H. pylori* with microbial flora

The diversity of environmental factors in the oral cavity contributes to the development of specific microbiota in different regions.<sup>11</sup> Tooth-associated biofilm can be divided into the supragingival (on the exposed surface of the

enamel) and subgingival (below the gums and inside the periodontal pocket) regions.<sup>9</sup>

Supragingival plaque is characterized by a low level of species diversity and the dominance of *Streptococcus* spp., *Actinomyces* spp. and *Lactobacillus* spp.<sup>38</sup> The presence of these microorganisms is related to the intense saccharolytic metabolism associated with the fermentation of carbohydrates to organic acids, and the subsequent acidification of the local environment. In addition to the acidification, the process of supragingival plaque maturation is also linked with the formation of microaerophilic conditions. This is the result of the oxygen consumption by the NADH oxidase in the Embden-Meyerhof-Parnas pathway. This process is accompanied by the appearance of *F. nucleatum* and late colonizers, which are sensitive to high oxygen levels.<sup>11</sup>

In vitro studies have shown that *F. nucleatum* supports the growth of P. gingivalis in the presence of elevated oxygen concentrations (10% and 20%) and lack of  $CO_2$ . Under such conditions, the oxygen-sensitive capnophilic P. gingivalis was unable to survive in monoculture. During the co-culture with *F. nucleatum*, a strong growth of this bacterium was observed.<sup>39</sup> The ability to generate a capnophilic environment by F. nucleatum is a feature that could potentially promote the survival of another bacterium, for which the main factor limiting growth is access to  $CO_{2}$ , i.e., H. pylori.40 It has been observed that H. pylori has a strong capacity to co-aggregate with *Fusobacterium* spp. naturally isolated from dental plaques (F. nucleatum and F. periodontium).<sup>41</sup> The tendency to co-aggregate with F. nucleatum was also confirmed in another experiment.<sup>42</sup> In addition, it has been determined that *P. gingivalis* may also be involved in such interaction and that this is a fimbriae-dependent process.<sup>42</sup> On this basis, it can be concluded that the presence of *H. pylori* is strongly associated with the physiological functioning of these two bacterial species (F. nucleatum and P. gingivalis) within the dental plaque, and vice versa.

Streptococcus spp. is the dominant genus in supragingival plaque. The ability of these bacteria to ferment carbohydrates into acidic products (acetic, formic and lactic acid) reduces local pH values near 5, during high availability of fermentation substrates.9 These are the conditions in which P. gingivalis and F. nucleatum are unable to survive, because the minimum pH tolerated by these microorganisms is 6.5 and 5.5, respectively.<sup>11</sup> Hence the close association of microbes capable of buffering acidic pH is highly desirable. The presence of bacteria able to utilize lactic acid, the main fermentation product of Strep*tococcus* spp., may protect the supragingival plaque from excessive acidification.8 H. pylori has been shown to have genes responsible for the uptake and metabolic conversion of D- and L-lactose.43 The pH-buffering process in supragingival plaque can also be mediated in an ammonia-dependent manner. H. pylori produces urease, an enzyme which converts urea into CO2 and ammonia. The urease produced by *H. pylori* has a very high activity. It has been shown that the rate of urea hydrolysis by the enzyme of this bacterium is  $36 \pm 28 \,\mu$ M/min/mg of bacterial proteins. This result is twice as high as that of Proteus mirabilis and 10 times higher than in other urinary tract pathogens.44 In vitro studies have demonstrated that the ability to produce ammonia is a key contributor to the preservation of the species diversity in oral biofilm. The presence of physiological concentrations of urea in the culture medium (1-10 mM urea in saliva) determined the bacterial population heterogeneity. However, in a culture with strains defective in urease production or the absence of urea in the culture medium, there was a drastic decrease in the viability of periopathogens (P. gingivalis, F. nucleatum, Prevotella intermedia), Neisseria subflava and Streptococcus oralis, and the dominance of biofilm by cariogenic bacteria, i.e., *Streptococcus* spp. (other than S. oralis) and Lactobacillus spp.<sup>45</sup>

Supragingival plaque consists mainly of early colonizers, Streptococcus spp. and Actinomyces spp., which may potentially modulate the physiology of oral H. pylori. It has been observed that such microorganisms have the ability to inhibit *H. pylori* growth in vitro.<sup>42</sup> Diffusible compounds secreted by Streptococcus mutans<sup>46</sup> and Streptococcus mitis<sup>47</sup> contribute to a dramatic decrease in H. pylori viability. However, microscopic observation has shown that this effect was caused by the conversion of H. pylori to nonculturable coccoid forms. This mechanism is consistent with the lack of possibility to isolate this bacterium from supragingival plaque, in which Streptococcus spp. is the dominant microbial genus.<sup>9,23</sup> Streptococci are a source of SDSF (Streptococcus diffusible signal factors), which may be involved in the morphological transformation of *H. pylori* into coccoid forms.<sup>48</sup> In an experiment, Khosravi et al. determined the effect of *H. pylori* and *S. mitis* co-culture on the gene expression of both bacteria.47 It was observed that streptococci expressed genes encoding phosphoglycerate kinase (PGK) only when co-existing with *H. pylori*. PGK is a glycolytic enzyme responsible for the conversion of 3-phosphoglycerol aldehyde to 1,3-bisphosphoglycerate. PGK is a surface protein produced by streptococci in increased amounts during the biofilm phase and is responsible for cell-cell interactions.<sup>49</sup> During co-culture in *H. pylori*, significant reduction in oxidative stress proteins, including glutathione metabolism enzymes, thioredoxin, flavodoxin and thiol peroxidases, have been observed. On the other hand, the amount of proteins involved in RNA degradation and nucleotide excision repair were increased.47 The reduction of genes encoding *H. pylori* antioxidants may potentially be due to increased physical contact between the bacterial cells and PGK-dependent aggregation/biofilm promotion in *H. pylori*.<sup>49</sup> Despite the use of filters (pore size 0.22 μm) for the spatial separation of both microorganisms, it cannot be excluded that interactions between PGK, present on the surface of membrane vesicles secreted by Strepto*coccus* (<0.2  $\mu$ m), and *H. pylori* cells still may exist.<sup>50</sup> The *P. int* increase in the amount of *H. pylori* proteins responsible for the DNA rearrangement can be beneficial under stressful to the conditions by rapidly selecting well-adapted strains for

conditions by rapidly selecting well-adapted strains, for example to antibiotics presence.<sup>51,52</sup> Because of this, the co-aggregation of both bacteria in dental plaque can promote the survival of *H. pylori* in the oral cavity.

One of the key signaling substances produced within dental plaque is autoinducer-2 (AI-2).8 This compound is a chemorepellent agent that promotes the dispersion of H. pylori aggregates/biofilms and initiates negative chemotaxis against the source of these signals.<sup>53</sup> Hence, the colonization of this niche by H. pylori should be impeded or even impossible. Commensal plaque bacteria secrete relatively low concentrations of AI-2 and respond to much lower levels of these substances in the environment (less than 100 pM) than periopathogens, considered to be strong AI-2 producers (nanomolar concentrations).9 The amount of AI-2 produced endogenously by H. pylori corresponds to the concentrations of signaling compounds secreted by periopathogenic microbes (~37 nM).<sup>53</sup> Therefore, the presence of coccoid-stimulating factors and relatively low levels of AI-2, in the early- to mid-stages of supragingival plaque allow dental H. pylori to colonize this niche as nonculturable spherical forms. On the other hand, subgingival plaque characterized by a high density of periopathogens, and thus also higher concentrations of AI-2, may favor the presence of mixed populations of spiral and coccoid *H. pylori* forms.

Subgingival biofilm is composed of many species of microbes, including the dominance of *Actinomyces* spp., *Tannerella forsythia, Fusobacterium nucleatum, Spirochetes* and *Synergistetes.*<sup>38</sup> In addition, the presence of *A. actinomycetemcomitans, Streptococcus parasanguinis, Filifactor alocis, Eubacterium* spp., *Prevotella* spp., *Porphyromonas* spp., and *Campylobacter* spp. is also detected.<sup>11,54</sup> Metabolism of the bacteria located within this niche is characterized by the breakdown of nitrogen compounds (amino acids, peptides, proteins) derived from gingival crevicular fluid (GCF). This niche possesses a neutral pH, abundance of secondary metabolites, short chain fatty acids (SCFAs), and ammonia.<sup>9,11,12</sup>

In a study, Henne et al. observed that in patients with periodontal disease, *Campylobacter rectus* could be detected at elevated levels in diseased subgingival sites compared to the healthy control group.<sup>55</sup> The reverse relationship was observed in the case of *Campylobacter concisus*. Such microbial shift was accompanied by more frequent isolation of "red complex" bacteria, including *P. gingivalis* 15.9% vs 0%, *F. nucleatum* 33.9%, vs 3.1%, and *T. forsythia* 28.2% vs 0.1%, respectively. A similar relationship was also observed in another Gram-negative bacterium from the Epsilonproteobacteria class, i.e., *H. pylori*. Hu et al. investigated the effect of *H. pylori* on inflammation severity and the presence of oral pathogens.<sup>26</sup> In samples taken from gingival lesions, the incidence of *P. gingivalis*,

*P. intermedia, F. nucleatum* and *T. denticola* was significantly higher when *H. pylori* co-existed. This was related to the exacerbation of periodontal disease. Interestingly, the inverse relationship between the presence of *H. pylori* and the pathogenic bacterium *A. actinomycetemcomitans* was also observed. The reason of this phenomenon is not known, but it cannot be ruled out that *H. pylori* can limit the amount of L-lactic acid in the local environment, which is a growth factor for *A. actinomycetemcomitans.*<sup>8</sup> The mechanism of competition for lactic acid availability is uncertain and should be confirmed in experimental studies.

# Influence of oral *H. pylori* on gastric infections and eradication efficacy

Colonization of the oral cavity by H. pylori can promote reinfections, even when antibiotic therapy has taken place. The ability to form biofilm and coccoid forms by oral H. pylori, and the presence of a relatively low biocide concentration in saliva often make it impossible to achieve therapeutic success. In a meta-analysis by Zou et al., it has been shown that in patients with gastric H. pylori, the incidence of oral H. pylori was significantly higher (45%, 490/1088) than in those without gastric infection (23.9%, 196/821). $^{13}$  It was also determined that the eradication rate of systemic therapy was significantly higher in cases of gastric H. pylori (85.8%, 187/218) and extremely low for oral H. pylori treatment (5.7%, 9/158). Gao et al. determined the effect of triple therapy (systemic therapy) and triple therapy combined with oral treatment, on the eradication rate of gastric H. pylori.<sup>15</sup> Combination therapy proved to be more effective, as one year after the end of treatment the incidence of these bacteria was twice as low (32.4%) as compared to the exclusive use of systemic therapy (62.8%). A similar relationship, showing an increased degree of H. pylori eradication after combination therapy, has been demonstrated in the meta-analyzes of Ren et al.<sup>56</sup> and Bouziane et al.<sup>57</sup> It was found that the relative chance of gastric H. pylori recurrence in patients who underwent oral and gastric therapy was reduced by 63%.57

## Conclusions

The role of the oral cavity as a reservoir for *H. pylori* is still a controversial issue. There are a great number of articles indicating the presence of this pathogen in the oral cavity, but discrepancies among techniques for *H. pylori* detection and the variations in patients tested often make it difficult to formulate a final verdict. Urease and PCR assays have a high level of positive results in oral *H. pylori* detection, while culture methods have an extremely low sensitivity. Despite large discrepancies in the results

obtained, it is suggested that the oral cavity may be an extragastric H. pylori reservoir, which contributes to reinfections after successful eradication therapies. This location is characterized by the co-existence of numerous microorganisms that naturally colonize the oral cavity. That may potentially affect the physiology and morphology of *H. pylori*. It appears that in the supragingival plaque, H. pylori occurs in the form of nonculturable coccoids, which often prevent the detection of such bacteria using standard culture techniques. Subgingival plaque is associated with high amounts of periopathogens, and thus also higher concentrations of AI-2, which determines the presence of H. pylori as mixed spiral and coccoid subpopulations. Based on the considerations presented in this review, it is suggested that there is a need to increase the number of studies that identify the relationship between *H. pylori* and other microorganisms, including oral flora. Identification of these mechanisms will make it easier to understand the physiology of oral H. pylori and will help to create new, alternative methods to increase the effectiveness of standard systemic therapy.

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# **Recurrent peripheral ossifying fibroma: Case report**

# Nawrotowy obwodowy włókniak kostniejący – opis przypadku

Harshita Nadimpalli<sup>D,F</sup>, David Kadakampally<sup>B–D</sup>

Department of Periodontology, Manipal College of Dental Sciences, Manipal Academy of Higher Education, Mangalore, India

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):83-86

Address for correspondence David Kadakampally

E-mail: david\_mahe2005@yahoo.co.in

Funding sources None declared

Conflict of interest None declared

Received on September 15, 2017 Reviewed on October 30, 2017 Accepted on November 28, 2017

#### Abstract

Most of the reactive lesions in the oral cavity arise from gingiva. Fibroma, focal fibrous hyperplasia, pyogenic granuloma, and peripheral ossifying fibroma are the commonly encountered lesions of gingiva. Peripheral ossifying fibroma (POF) frequently arises from the peripheral tissues like gingiva. It is commonly found in females, mostly in the anterior of the molar region. POF is predicted to arise from the cells of periodontal ligament due to close proximity of gingiva to periodontal ligament. Its exact origin is unclear.

The present article describes the case of recurrent peripheral ossifying fibroma located in the right lower premolar region in a 23-year-old female patient. Clinical, radiographic and histologic features as well as differential diagnosis, treatment and follow-up are discussed in this report. Early diagnosis along with surgical excision and curettage of surrounding tissue is important for the prevention of recurrences. Early conservative management of lesion reduces the risk of progression of lesion, and frequent follow-up visits are required to evaluate for recurrences.

Key words: peripheral ossifying fibroma, reactive gingival lesions, recurrent

Słowa kluczowe: obwodowy włókniak kostniejący, zapalne zmiany dziąsłowe, nawrotowy

DOI

10.17219/dmp/80863

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## Introduction

Gingiva is said to be a common site for localized reactive lesions but not for neoplastic lesions.<sup>1</sup> Peripheral ossifying fibroma (POF) is a reactive lesion of gingiva, which is commonly found in the interdental region. It is usually seen as pale pink to dark red in color, with a smooth or rough surface lying on a pedunculated or broad base by which it adheres to the underlying tissue. It accounts for about 9% of all gingival growths and 2% of all oral tumors.<sup>2</sup> The origin of peripheral ossifying fibroma is unclear. It occurs most commonly in 5–25-year-old patients, females are more susceptible than males.<sup>3</sup> It cannot be clinically separated from pyogenic granuloma.<sup>4</sup>

#### Case report

A 23-year-old female patient presented with a chief complaint of swelling in the right lower back teeth region. The patient gave a history of swelling 2 years ago in the same region, and it was excised; the recurrence of swelling was seen 1 week back, and the size was increasing, which was associated with slight pain in that region. No extra oral swelling was seen.

Intraoral examination revealed that there was swelling in the right lower premolar region, which is shown in Fig. 1. Swelling with the size of 6 mm  $\times$  5 mm was present interdentally between the right lower first and second premolars on the lingual side. It was reddish pink in color, with a rough surface and well-defined margins, and its consistency was firm and fibrotic on palpation. Swelling extended from the lingual side to the interdental region of both premolars.



Fig. 1. Clinical picture - pre-operative

A radiological examination revealed no evidence of bony involvement in intra oral periapical radiograph (IOPA). Routine blood tests were carried out and were found to be normal.

Scaling and root debridement were performed as phase I therapy. The patient was given an appointment 1 week after phase I therapy for reevaluation. Informed consent was taken before the surgical treatment. Local anesthesia was administered and excision of the lesion was done; the excised tissue was placed in formalin solution and sent for a histopathological evaluation.

A routine histological examination with hematoxylin revealed the presence of parakeratinized stratified squamous epithelium overlying a fibrocellular connective tissue stroma ( $4\times$  magnification) (Fig. 2). The connective tissue stroma exhibits haphazardly arranged proliferating spindle cells, dystrophic calcifications, mild chronic inflammatory cell infiltrate (mainly lymphocytes), and areas of hemorrhage ( $40\times$  magnification) (Fig. 3). The diagnosis was suggestive of peripheral ossifying fibroma. In the follow-up period, no recurrence was seen after 5 months after surgical excision (Fig. 4).

### Discussion

POF has been known by various other names, such as peripheral odontogenic fibroma (PODF) with cementogenesis, peripheral cemento-ossifying fibroma, peripheral fibroma with calcification, peripheral fibroma with osteogenesis, calcifying fibroblastic granuloma, fibrous epulis, etc.<sup>5</sup> The sheer number of names used for fibroblastic gingival lesions indicates that there is much controversy surrounding the classification of these lesions.<sup>6,7</sup>



Fig. 2. Histological picture of lesion showing central fibrocellular tissue and overlying epithelium (4× magnification)



Fig. 3. Histological picture of lesion showing dystrophic calcification (40× magnification)

Maxilla is the common site for POF compared to mandible. About 60% of the lesions occur in maxilla, mostly in the mesial to molars region. Dental plaque, dental calculus, microorganisms, dental appliances, and restorations act as triggering factors for the lesion to occur. The chances of high recurrence (8–20%) for POFs indicate that a careful treatment strategy should be recommended, involving the excision of the entire lesion witch surrounding healthy margins and debridement of



Fig. 4. Clinical picture - 5 months post-operative

the underlying bone and tooth.<sup>8,9</sup> The reason behind the recurrence rate of POF is probably due to the remnants of the lesion, persistence of local irritants or repeated injury.<sup>10</sup> POF has to be differentiated from other reactive lesions of gingiva, like pyogenic granuloma, peripheral giant cell granuloma (PGCG) and peripheral odontogenic fibroma.<sup>11,12</sup> The differentiation of POF has to be done on histological and sometimes by radiographic basis. Pyogenic granuloma presents as an erythematous overgrowth with surface ulceration. It has a tendency to easily bleed clinically. Microscopically it exhibits vascular proliferation resembling granulation tissue. PGCG can be differentiated by histological appearance, which shows scattered giant cells in a fibrous stroma. Peripheral odontogenic fibroma shows prominent islands of odontogenic epithelium in the histological picture. Though not significant in most of the cases, some alterations in bones are noted, like foci of calcifications, bony erosion, widening of the periodontal ligament space, and thickened lamina dura. Alteration in teeth position due to interdental bone loss is also seen.

The basic microscopic pattern of the POF is fibrous proliferation associated with different types of mineralized components.<sup>13</sup> The content of mineralized components varies from 23% to 75%. Butcher and Hansen reported 3 types of components in POF: bone (woven/lamellar), dystrophic calcifications, cementum.<sup>3</sup> In the present case, a histological examination revealed fibrocellular component with a mineral component as dystrophic calcification.

#### Conclusions

In the initial stages of lesion development, it is difficult to differentiate clinically a particular lesion. For confirmation, a histological examination has to be conducted. Even though the etiology is unclear, poor oral hygiene could be a predisposing factor in peripheral ossifying fibroma and the tissue has to be examined. Early diagnosis and conservative management is important in such lesions, since they can become more destructive over time if not treated. A regular follow-up is required after excision due to high growth potential of the lesion (8%–20% recurrence rate).<sup>14</sup>

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# Intraosseous migration of second premolar below the inferior alveolar nerve canal: Case report

# Wewnątrzkostna migracja drugiego zęba przedtrzonowego poniżej kanału dolnego nerwu zębodołowego – opis przypadku

Hamed Mortazavi<sup>A-F</sup>, Maryam Baharvand<sup>A-E</sup>, Kosar Rezaeifar<sup>F</sup>

Department of Oral Diseases, School of Dentistry, Shahid Beheshti University, Teheran, Iran

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):87-90

Address for correspondence Kosar Rezaeifar E-mail: kosar.rezaeifar@gmail.com

Funding sources None declared

Conflict of interest None declared

Received on September 24, 2017 Reviewed on October 26, 2017 Accepted on November 27, 2017

#### Abstract

Migration is a kind of eruption abnormality where a tooth grows far from its original site of development. Anomalies in dental eruptions are referred to as ectopia, which might be encountered in several regions around the oral cavity. The incidence of mandibular second premolar impaction has been estimated to be 2.1-2.7%. The frequency of its intraosseous distal migration is 0.25%.

We report a case of extremely distally intraosseous migration of mandibular second premolar, which was found in a routine radiography. A 28-year-old woman who attended a dental clinic to treat teeth caries was noticed to have her lower second premolar located horizontally in the homolateral mandibular angle below the inferior alveolar nerve canal on a panoramic view. As the patient was asymptomatic, she was advised to take follow-up radiographs to rule out any cystic/neoplastic changes. Taking a panoramic radiograph in patients with missing mandibular premolars should be considered, because in rare cases migration or transmigration of these teeth may happen.

Key words: migration, mandible, second premolar

Słowa kluczowe: ektopia, żuchwa, drugi ząb przedtrzonowy

DOI

10.17219/dmp/80829

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### Introduction

Anomalies in dental eruption are referred to as ectopia, which might be encountered in several regions around the oral cavity.<sup>1</sup> When teeth are grown in areas far from their regular place of development, the condition is named migration or transmigration.<sup>1,2</sup> According to Peck, dental migration occurs exclusively in the mandible and it refers to the horizontal movement of unerupted teeth.<sup>2</sup> This entity occurs in permanent dentition and usually involves mandibular lateral incisors, canines, and the second premolars. There is a female predilection with a female to male ratio of 1.7:1. Intrabony migration of unerupted teeth may take place in both distal and mesial directions. For example, the mandibular lateral incisor and, in rare instances, the first premolar tend to migrate distally, whereas the mandibular canine most often migrates mesially and even sometimes across the midline.<sup>3-5</sup>

The prevalence of premolar impaction in the adult population is 0.5%, which is reported in the range of 0.2–0.3% for mandibular premolars and 0.1–0.3% for maxillary premolars.<sup>5</sup> On the other hand, specific articles indicate that the second mandibular premolar alone accounts for 24% of total dental impactions.<sup>6</sup> The incidence of mandibular second premolar (MSP) impaction has been estimated to be 2.1–2.7%. The frequency of its intra-osseous distal migration is 0.25%.<sup>5</sup> It is noted that in most instances, intrabony migration of the MSP is idiopathic, unilaterally, and without any associated dental anomalies.<sup>3</sup> Both genetic and environmental factors are implicated in the etiology of this anomaly. Early loss of permanent first molar increases the chance of distal migration of MSP from 5% to 10%.<sup>1,7</sup> Management of this condition depends on the position of the malposed tooth, the degree of discomfort, and the associated pathologic lesions, if any.<sup>4–7</sup>

In this paper, we report a 28-year-old woman with an extremely distally intrabony migration of MSP, which was found in a routine radiography.

#### **Case report**

A 28-year-old woman attended at our clinic in Tehran (Iran) to treat dental caries. She had a panoramic radiograph performed 1 week prior to her appointment. No history of systemic disease, skeletal abnormalities or dental extraction was mentioned. In the oral examination, decay of the teeth 25, 36, 46 was shown. These caries were also detected by radiography. In the clinical examination of tooth 46, a class V filling on buccal side of tooth was detected. The panoramic view revealed a root canal therapy of 11 and 21, and an overhang of tooth 26. A second premolar was noticed horizontally in the homolateral mandibular angle below the inferior alveolar nerve canal (Fig. 1). The follicular space was seen as a pericoronal radiolucency with sclerotic borders around the tooth. The patient was referred to a routine exam after the discovery of the impacted teeth; because she had no symptoms, she was reluctant to attend a follow-up, and so advanced radiography (e.g., cone beam computed tomography) was not prescribed to her. The patient reported no discomfort and was not aware of this anomaly



Fig. 1. Panoramic view shows distal migration of right second mandibular premolar below the inferior alveolar nerve canal

prior to a dental examination. Therefore, surgical removal of the migrated tooth was not considered – the patient was asked to take periodic panoramic radiographs (twice a year) for early detection of any changes in size of dental pericoronal radiolucency due to the formation of cystic lesions, such as dentigerous or follicular cyst, and possible damages to the inferior alveolar canal.

### Discussion

Intrabony migration of premolars has a low occurrence compared to other teeth. There are some etiological factors involved in the migration of teeth, such as retention or premature loss of a primary tooth, ectopic growth of tooth buds, genetic factors, endocrine disorders, and trauma.<sup>8</sup> Distal migration is not yet completely understood, because the teeth tend to move mesially as a result of masticatory forces.<sup>3</sup> Distal migration of MSP can occur due to the development of the tooth bud with a variable degree of distal inclination under the distal root of the primary second molar. When deciduous root is resorbed and the permanent molar is extracted early, the second premolar may migrate distally. In rare instances, the MSP was reported to migrate as far distally as the mandibular angle and the coronoid process. Such a migration is slow and occurs during a period of several years.<sup>5</sup> The intrabony migration cannot be diagnosed by using routine periapical radiographs, because the tooth is usually horizontally under the root of other teeth and near the border of the mandible.<sup>5,7</sup> Therefore, a radiographic examination including a panoramic view and sometimes occlusal radiographs should be requested.7 In accordance with our case, it is noted that premolar migration is more common in females.9,10 It was demonstrated that 55.5% of the mandibular ectopic premolars were located on the right side and 44.4% were on the left side as well.<sup>10</sup> Intraosseous migration of MSP is often unilateral, horizontal and asymptomatic. It can appear at any age (ranging from 8 to 62 years old) and can happen mostly in adults older than 20 years of age, which corroborates our report.<sup>5,8,10</sup> Treatment options for migrated premolars include surgical exposure combined with orthodontic therapy. If not treated, impacted teeth can lead to problems, e.g., carious lesions, infections, destruction of adjacent teeth, periodontal disease, and even oral and maxillofacial cysts or tumors. In 16% of cases, cystic and neoplastic changes have been reported in relation to impacted teeth.<sup>11,12</sup> On the other hand, complications associated with impacted tooth surgeries include: pain, swelling, trismus, bleeding, cellulitis, abscesses, septicemia, wound dehiscence, bone sequestra, paresthesia, anesthesia, hematoma, alveolar osteitis, temporomandibular joint dysfunction, and jaw fracture.<sup>13</sup> In patients with no considerable sign or symptoms, a periodic radiographic observation should be suggested.<sup>5</sup>

In conclusion, taking a panoramic radiograph in patients with missing mandibular premolars should be considered, because in rare cases migration or transmigration of these teeth may happen.

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## Vital root submergence of immature permanent incisors after complicated crown-root fracture followed by orthodontic space maintenance: A presentation of two cases

# Pozostawienie "zatopionego" korzenia stałych siekaczy z niezakończonym rozwojem po ciężkim złamaniu koronowo-korzeniowym w celu ortodontycznego zachowania miejsca w łuku – opis dwóch przypadków

Adrian Strzecki<sup>1,A,D</sup>, Agnieszka Bruzda-Zwiech<sup>2,B-C</sup>, Renata Filipińska<sup>2,B,E</sup>

<sup>1</sup> Department of Orthodontics, Medical University of Lodz, Poland <sup>2</sup> Department of Pediatric Dentistry, Medical University of Lodz, Poland

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):91-98

Address for correspondence Adrian Strzecki E-mail: adrian.strzecki@gmail.com

Funding sources None declared

Conflict of interest None declared

Received on October 29, 2017 Reviewed on November 9, 2017 Accepted on January 24, 2018

#### Abstract

Complicated crown-root fractures are considered rare occurrences in young permanent dentition; however, they pose a particularly difficult clinical challenge, especially when the traumatized tooth is immature.

Two cases of complicated crown-root fractures of immature incisors are presented. In both cases, vital root submergence with no treatment to the exposed pulp was introduced as a stage in a complex treatment plan with the primary goal of preserving the shape of the alveolar ridge in the traumatized area until the root was completely formed or facial growth was finished. No inflammatory symptoms were detected either radiographically or clinically during the 15- and 16-month follow-up periods. In the chosen clinical cases, vital root submergence followed by orthodontic space maintenance can be beneficial to young patients if other treatment options are limited by the depth of the crown-root fracture or if the patient suffers from high dental fear and presents a particularly low level of compliance.

Key words: dental trauma, crown-root fracture, vital root submergence

Słowa kluczowe: uraz zęba, złamanie koronowo-korzeniowe, pozostawienie zatopionego korzenia

#### DOI

10.17219/dmp/83744

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## Introduction

Traumatic dental injuries (TDI) in patients in developmental age are considered a particularly difficult challenge, as the prognosis regarding tooth vitality is usually poor and the results of dental management difficult to predict. Clinicians need to make the correct clinical decision as quickly as possible while considering its life-long consequences. The early loss of a tooth prior to the pubertal growth spurt can have profound social, aesthetic, occlusal and functional consequences,<sup>1-3</sup> as the alveolar ridge in the traumatized, post-extraction areas ceases to develop and bone atrophy begins. Although the bone loss of the alveolar ridge is most significant in the horizontal aspect, vertical bone loss also occurs, with the buccal side of the alveolar ridge being most prone to resorption.<sup>4,5</sup> As a result, the alveolus is reduced, becomes more narrow and more lingually proclined.<sup>6</sup> What is of major concern in early adolescence is permanent anterior maxillary tooth loss due to alveolar bone arrest and aesthetic ridge deformation, which preclude the insertion of osseointegrated implant with a good emergence profile that is impossible to achieve without ridge augmentation.<sup>7,8</sup>

The central maxillary incisors are the teeth most likely to be injured, and account for 80% of all traumatized teeth.<sup>1</sup> Maxillary lateral incisors and mandibular incisors are vulnerable to injury to a lesser extent.<sup>1</sup> Crown-root fracture defined as a fracture involving enamel, dentin and cementum accounts for 5% of all dental traumata to permanent dentition,<sup>1</sup> with transverse fractures being observed less frequently,<sup>9</sup> especially in children<sup>10,11</sup> and when the root development is incomplete.<sup>10</sup> The fracture line can also expose the pulpal tissue, thus dividing the crown-root fracture into 2 groups: complicated and uncomplicated. Treatment options in cases of complicated crown-root fractures are complex, and require a multidisciplinary approach,<sup>12</sup> the choice of which largely depends on the position and direction of fracture line, pulpal involvement, tooth maturity, and the length of root remaining in the alveolus.<sup>2,3,13</sup> Even when the fracture is positioned subgingivally, bacterial invasion should be expected to the fracture region, leading in most cases to subsequent inflammation.<sup>1,14</sup> For this reason, most treatment protocols of deep crown-root fracture require removal of the coronal tooth fragment; however, it can be used for further prosthetic tooth reconstruction.

The treatment modalities described in previous studies can be divided into 2 groups: surgical and conservative. Surgical methods other than extraction focus on the immediate reposition of the intra-osseus tooth portion followed by endodontic treatment. It can be either intentionally replanted or surgically extruded and fixed in a more favorable, supragingival position. Surgical extrusion can be achieved with or without raising the mucoperiosteal flap. The semi-conservative approach involves gingivectomy and osteotomy in order to alter the spatial relations between the alveolar margin and remaining tooth portion. Orthodontic extrusion and submerging vital root portion in situ are procedures burdened with limited invasiveness.

Most case reports describe crown-root fractures occurring in teeth with closed apex or in adult patients. However, most traumatic dental injuries to the permanent dentition occur in schoolchildren.<sup>15</sup> When the clinician needs to manage crown-root fracture in an immature tooth, treatment modalities are limited by the stage of root development. As presented above, although a plethora of treatment options can be mentioned, almost every one of them requires non-delayed, permanent obturation of the root canal system at one of the clinical stages. For this reason, traumatized developing teeth with open apices are particularly challenging to clinicians, especially when the fracture line is located subgingivally. According to current guidelines regarding traumatic dental injuries (TDI) management,<sup>15</sup> "every effort should be made to preserve pulpal vitality in the immature permanent tooth to ensure continuous root development". Young age restricts the range of available treatment modalities, both due to the stage of root development and the limited level of patient compliance. Hence, in young children with immature permanent teeth demonstrating crown-root fracture, the main focus should be on choosing methods with minimal invasiveness to the surrounding bone and periodontium, retaining the ability of the root to continue its development and preventing the early loss of the tooth, thus preserving the height and width of the alveolus in the traumatized site, negating the possible consequences of the injury to the occlusion and providing a temporary aesthetic solution to minimize the social problems encountered by the child. The aim of this article is to present 2 cases of subgingival crown-root fracture of immature teeth treated by a minimally invasive multidisciplinary treatment approach which can be also introduced in patients with a high level of dental fear.

### **Case presentation**

#### Case 1

A 10-year-old male patient was referred to the Department of Pediatric Dentistry of our institution due to a dental injury suffered the previous evening after falling from a chair. His medical history was not relevant. Clinical examination revealed a fractured restoration of the medial corner of the left upper central incisor, as well as movable composite build-up of the left lateral incisor with marginal gum swelling and bleeding. Tooth #22 was very tender to percussion and the lower lip was swollen. The prior dental history revealed that he had suffered trauma to the same maxillary region 6 months before,

diagnosed as uncomplicated crown fracture of teeth #11 and #21, and uncomplicated crown-root fracture of tooth #22, with an oblique fracture line penetrating subgingivally. The teeth underwent immediate conservative restoration with no further complications. The second trauma led to a crown-root fracture of the lateral left incisor as the mobile composite reconstruction was attached to the fractured crown portion. The fracture line was horizontal and reached below the alveolar ridge margin. A radiographic examination confirmed the diagnosis of a crown root fracture with pulp exposure and additionally showed incomplete root development of tooth #22 (Fig. 1a). After a holistic assessment of clinical and radiographic findings, the decision was made to perform in situ submergence of the vital root fragment. The coronal part of tooth #22 was surgically removed under local anesthesia, and the fracture margin was exposed. The root fragment with the bleeding exposed pulp was gently rinsed with 0.9% saline, and the gingival tissue was soundly sutured. Antibiotic therapy with 150 mg clindamicine administered 4 times a day was prescribed.

This clinical decision had the following rationale: 1. as the crown root fracture occurred at the level of the alveolar ridge, the remaining apical portion of injured tooth #22 did not reach the criteria of 1:1 crown-root ratio and so could barely support a future permanent prosthetic build-up; gingivectomy/osteotomy with subsequent apexification or mineral trioxide aggregate (MTA) pacement in apical region and root canal obturation, followed by orthodontic extrusion would not alleviate this problem; 2. due to the patient's young age, extraction of the remaining root fragment would lead to severe alveolar atrophy in anaesthetically-sensitive region. The root was submerged with the intention of retaining the shape of the alveolar process for as long as possible, preferably until the facial growth was finished and a dental implant could be placed in the traumatized area. After 2 weeks of uneventful healing, the sutures were removed (Fig. 1b, c). Subsequently, the orthodontic



Fig. 1. A – radiograph taken at the day of trauma – crown root fracture of the upper left lateral incisor (#22). B, C – upfront view of traumatized area after 2 weeks of healing and suture removal



Fig. 2. A, B – upper lingual arch with acrylic tooth #22 on the dental cast. Orthodontic bands fitted on the upper second deciduous molars. C – upfront view of patient's anterior dentition without the prepared appliance. D, E – patient's dentition with the cemented lingual arch: upfront (D) and palatal view (E). F – dental radiograph taken after the placement of orthodontic appliance – 4 weeks after the trauma

bands were fitted to the deciduous upper second molars and dental impressions were taken to restore the missing crown of tooth #22 by the use of an upper lingual arch with an attached acrylic tooth (Fig. 2a, b). The patient did not have any orthodontic treatment needs besides the insertion of a space retainer after the dental trauma, and his first permanent molars were in I Angle Class intercuspidation on both sides of the dental arch. The lingual arch was cemented on the next dental visit with satisfactory aesthetic and functional effect (Fig. 2c-f). As the oral hygiene of the patient was sub-optimal, the deciduous second molars were chosen as a support for the lingual arch instead of the permanent first molars to protect the latter from the increased risk of caries associated with the presence of orthodontic bands. Therefore, the mentioned appliance should be perceived as a long-term solution, but not a permanent one: after root resorption occurs in the deciduous second molars, the lingual arch will be modified and fitted to the permanent first molars. No inflammatory symptoms could be detected either clinically or radiographically at any stage of treatment or during a 13-month follow-up (Fig. 3a). The submerged root manifested continued development. Although the shape of the alveolar ridge slightly altered vertically, the bone loss was not significantly pronounced in the horizontal aspect (Fig. 3b-d). The orthodontic appliance was well tolerated by the patient, who underwent check-ups every 6 weeks.



Fig. 3. A – dental radiograph taken after 13-month follow-up period. No inflammatory symptoms could be detected. B–D – bone loss in the injured area appears to be only slightly pronounced in vertical aspect and the overall esthetic outcome is satisfactory

After 15 months from trauma, the upper second deciduous molars became mobile due to the physiological root resorption and additional occlusal load connected with supporting the artificial crown of the tooth #22. The appliance was removed and the patient was referred to the surgical clinic to have teeth #55 and #65 extracted. Clinical examination revealed the retained shape of alveolar process with slightly pronounced vertical bone loss (Fig. 4a–b). No inflammatory symptoms could be observed (Fig. 4a–c). Closed root apex with no signs of chronic periapical inflammation can be observed on the dental radiograph taken during the appointment (Fig. 4d). The patient was highly anxious towards the planned surgical procedures and both his compliance and oral hygiene



Fig. 4. A, B – patient after 15-month follow-up period with and without the attached acrylic tooth #22. Slight inflammation of the mucosa observed in the injured region can be attributed to the poor oral hygiene combined with the contact with the acrylic resin. C – the bone loss in the horizontal aspect can be hardly observed. D – dental radiograph taken after 15-month follow-up period: apex of the root of the tooth #22 is closed; previously exposed pulp area is covered with radiopaque tissue

were unsatisfactory. Certain clarification of the observed situation was given, with the parents mentioning that our patient was recently diagnosed with Asperger's syndrome. The treatment focusing on retaining the space after the lost tooth #22 was continued by means of removable orthodontic appliance (modified Schwarz-type expander) with the attached acrylic crown.

#### Case 2

An 8-year-old female patient was referred to the Department of Pediatric Dentistry of our institution for traumatic dental injury suffered the previous day at school. The medical history was non-contributory. The maxillary anterior region was most severely injured, with the right central incisor subluxated and left central incisor crown horizontally fractured at the level of the alveolar ridge (classified as crown-root fracture with pulp exposure). The clinical examination also revealed poor oral hygiene and many carious lesions requiring immediate treatment. Tooth #21 was not visible in the mouth, and the dental socket in the site of the tooth was filled with a blood clot (Fig. 5b). The fractured coronal tooth fragment, brought in by the parents, was rinsed with 0.9% saline and stored in saline at 4°C for possible future adhesive build-up. The parents also provided clinicians with a dental radiograph taken on the day of the trauma; however, it did not show the whole apical region of tooth #21 (Fig. 5a). Tooth #11 was subluxated and manifested slight pathological mobility, along



Fig. 5. A – dental radiograph taken at the day of trauma. Tooth #21 is horizontally fractured at the cervical area. Periapical region of the tooth #21 cannot be assessed. B – missing clinical crown and the dental socket of the tooth #21 filled with a blood clot – clinical situation during the first appointment. C – semi-rigid splint connecting the teeth #11–#13 applied due to the subluxation of the tooth #11



**Fig. 6.** A – dental radiograph of the injured area taken after 7-week period. No inflammatory symptoms could be detected and the process of the closure of the root apex continued. Note the mineralized tissue covering the area of previously exposed pulp. B–E – view of the patient's dentition after 7-weeks from the dental injury. No inflammatory symptoms could be detected clinically

with tenderness to vertical percussion. The sensitivity of the pulp was positive when tested with a cold stimulus. The subluxated tooth was splinted with semi-rigid wire and composite resin for 2 weeks (Fig. 5c). The anatomic relationship between the fracture line and the alveolar ridge margin could be assessed, despite the parents not giving consent for another dental X-ray to be taken on the day of admission. Although the stage of root formation could not be evaluated, it was assumed on the basis of age of the patient that root development was not completed.

As the length of the root of tooth #11 seemed to be adequate for supporting post-retained crown reconstruction, immediate gingivectomy or even osteotomy followed by the subsequent MTA placement in apical region of the root to form a hard barrier prior to gutta-percha root obturation were considered. However, this treatment option was abandoned due to dental fear and low level of compliance by the patient. Vital root submergence in situ was discussed with the parents as a treatment option, which might allow for root apex closure of tooth #22 and the use of an apical fragment for later post-retained crown reconstruction.

The traumatized area with its remaining root segment was rinsed with 0.9% saline and soundly sutured. After 2 weeks of uneventful healing, the sutures and splint were removed. The patient failed to attend a scheduled check-up visit 4 weeks following the trauma, but did so after 7 weeks, when it was possible to assess the course of the treatment both clinically and radiographically (Fig. 6a–e). No inflammatory symptoms were present at the time of examination and the traumatized region had healed properly. A dental radiograph showed an incompletely closed apex root of tooth #21, the root portion appeared to be of sufficient length for future core build-up. The area of the previously-exposed pulp appeared to be covered with hard tissue resembling radiographically reparative dentine.

After the apex closure process is completed and patient compliance will be sufficient, it is planned to extirpate the pulp and extrude the root orthodontically above the gingival level, thus enabling a long-term temporary prosthetic build-up. It was also noted that the margin of the fractured root was visible above the mucosa in a very limited area. It is possible that the root either began to spontaneously erupt or the root exposure occurred due to alveolar ridge recontouring. Due to the patient's young age, poor oral hygiene and narrow maxillary arch, it was decided to combine a temporary replacement for the crown of tooth #21 with treatment using an orthodontic removable appliance. An active palatal plate with an acrylic tooth #21 and expansion screw positioned in the midline was designed (Fig. 7a-d). After a 10-month follow-up, no inflammatory symptoms could be observed either clinically or radiographically (Fig. 8a-d). The apexification of the submerged root continued, thus indicating that the pulp retains its vitality. The patient showed improved compliance and was pleased with the esthetics of the proposed clinical solution (Fig. 8e).

Certain difficulties occurred in further monitoring of treatment outcome, as the patient failed to show up for a pre-scheduled appointment at the 12-month stage of follow-up. It was not until 16 months after the trauma when we could get in touch with the patient's parents



Fig. 7. A, B – removable appliance fitted on the dental cast. C, D – upfront view of patient's anterior dentition without the appliance and with the appliance inserted into the oral cavity

and the control clinical and radiological examination could be performed. No inflammatory symptoms were either mentioned by the patient or discovered during the check-up (Fig. 9a, b). The crest of the alveolar process in the injured area was on the level of cervical gingival margin surrounding the tooth #11, which could be described as a major improvement (Fig. 9c). A radiographic examination showed closed root apex of the tooth #21 with no periapical lesions and the area of previously exposed pulp covered with radiopaque tissue (Fig. 9d). The patient and her parents were reluctant to the proposed plan of the elective endodontic treatment and a subsequent forced orthodontic eruption of the root and wished to continue the treatment with the current appliance. It was established that the treatment options will be re-discussed after the eruption of all premolar teeth, which will necessitate the change of the removable appliance. Nevertheless, sustaining the alveolar bone level and completed closure of the root apex made all of the permanent treatment options ranging from the combined orthodontic and prosthetic treatment to the implant placement possible.



Fig. 8. A – dental radiograph taken after the 10-month follow-up period. The apex of the root of the tooth #21 is closed and no sign of chronic inflammation could be detected. B-E - 10 months from the injury. No inflammatory symptoms could also be detected during the clinical examination



**Fig. 9.** A–C – clinical examination after 16-months. Both vertical and horizontal dimension of the alveolar ridge remained unchanged. No inflammatory state could be detected. D – dental radiograph taken after 16-month follow-up period: apex of the root of the tooth #22 is closed; previously exposed pulp area is covered with radiopaque tissue

#### Discussion

Although intentional submerging in situ of vital roots after crown-root fracture is mentioned in up-to-date guidelines regarding the management of traumatic dental injuries,<sup>15</sup> reports of cases presenting it are rarely encountered in scientific literature. A review of existing literature reveals only one such study by Mackie et al.<sup>16</sup> Although the main advantage of this approach is the possibility of retaining the shape and size of the alveolar ridge in the traumatized region until the facial growth is finished, it also offers limited invasiveness, which requires little compliance, continued root development and no additional iatrogenic trauma to the surrounding tissues. Furthermore, it offers other benefits for the patient in cases when the remaining root cannot support any post-retained crown due to inadequate length and needs to be extracted.

The fact that the untreated, exposed pulp showed no immediate or delayed inflammatory symptoms, either clinically or radiographically, can be expected to a certain extent. This can be probably attributed to the stage of tooth development and relatively good perfusion of pulpal tissue in immature teeth. Furthermore, radiographs taken at various treatment stages indicated that the pulp tissue remained vital. The rationale behind root submergence is derived from studies concerning fractured and retained roots following unsuccessful extractions of vital teeth. The vast majority of studies show that such retained roots remain either vital or uneventful.<sup>17,18</sup> Some authors suggest that the submerged root can be covered with new bone.<sup>19</sup> Attempts to preserve the volume of the alveolar ridge by means of vital root submergence in older prosthetic patients, when the crown-root ratio is unfavorable, have also been suggested.<sup>20</sup> As it was mentioned above, the decoronation of deeply impacted lower third molars bears a certain resemblance to the intentional submergence of traumatized roots. Despite obvious differences, both procedures treat the exposed pulp with 0.9% saline, which is then covered with a tightly sutured mucoperiosteal flap. The most important similarity is, however, the lack of subsequent symptoms of pulp inflammation despite the lack of any capping. Interestingly, in some cases, the passive eruption of retained teeth occurs.<sup>21</sup>

What should, and could, be done with the remaining apical tooth segment is clearly the main problem encountered in crown-root fracture management in the developmental age. The 4 key issues that need to be addressed are as follows: 1. whether the length of the remaining portion of the root is adequate and whether it may be used after surgical procedures/orthodontic extrusion for fixed prosthesis purposes; 2. what can be done and achieved at the current development stage of traumatized tooth root and patient growth stage; 3. the level of patient motivation and compliance and whether the clinician is likely to be limited by technical constraints or adolescent patient cooperation, and finally; 4. what can be done to protect the height and width of the alveolar process.

When the submergence of the vital root is performed for the preservation of the alveolar ridge prior to permanent prosthetic reconstruction with a dental implant, spontaneous eruption of the root may be regarded as an undesirable complication.<sup>22</sup> However, in our case of the submergence of a vital root with a length adequate to support further post-retained crown reconstruction, the fact that the root began to erupt can be beneficial, as described in a case presented by Johnson and Jensen,<sup>22</sup> in which spontaneous eruption eliminated the need for surgical exposure and orthodontic extrusion of the root fragment and allowed for successful root canal obturation and restoration with a post and porcelain fused to metal crown.

In the first of our cases, the possibility of using the root fragment for the prosthetic purposes, even after orthodontic extrusion, would be very limited as the crown to root ratio would be significantly higher than 1:1. In the second case, the remaining fragment of the central incisor is sufficient for prosthetic build up and forced eruption is planned. However, in our opinion, prosthetic reconstruction should be delayed, as the root shows no inflammatory symptoms, root development persists and mildly pronounced passive eruption can be observed. After apexification is completed, a forced eruption should be performed, thus enabling at least temporary prosthetic restoration of the fractured crown.

## Conclusions

Vital roots submergence could be considered as the first stage of a more complex treatment scheme in juvenile patients burdened with few possible complications and requiring very low level of compliance. The retained root may remain within the alveolus until the implant placement is possible or serve as a prosthetic post following delayed pulp extirpation. The possibility of delaying pulp extirpation may prove beneficial in certain clinical situations with the emphasis of the immature traumatized teeth. Orthodontic appliances with attached artificial dental crowns prove to be aesthetically acceptable, multi-purpose clinical solutions in cases of early clinical crown loss in growing patients. The root submergence procedure also allows for dental fragments that would otherwise be extracted to remain within the alveolus and at least partially contribute to retaining the shape of the alveolar ridge.

As the pulpal tissue in the submerged roots showed no inflammatory symptoms, despite being left untreated and only tightly covered with mucosa, its immune defensive capabilities in teeth with an open apex may be higher than commonly perceived. For this reason, root submergence is worth considering in young patients experiencing deep crown-root fracture, especially when they suffer from high dental fear and a low level of compliance.

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## Hand, foot and mouth disease as an emerging public health problem: Case report of familial child-to-adult transmission

# Choroba dłoni, stóp i jamy ustnej jako pojawiający się problem dla zdrowia publicznego – opis przypadku rodzinnego przeniesienia zakażenia z dziecka na dorosłego

Zuzanna Ślebioda<sup>A–D</sup>, Barbara Dorocka-Bobkowska<sup>A,E,F</sup>

Department of Gerodontology and Oral Pathology, Poznan University of Medical Sciences, Poland

A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation; D – writing the article; E – critical revision of the article; F – final approval of the article

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):99-104

Address for correspondence Zuzanna Ślebioda E-mail: zuzia\_slebioda@o2.pl

Funding sources None declared

Conflict of interest None declared

Received on November 8, 2017 Reviewed on November 26, 2017 Accepted on December 5, 2017

#### Abstract

Hand, foot and mouth disease (HFMD) is a highly contagious viral infectious disease that commonly affects small children. Typical clinical symptoms include low-grade fever, malaise and myalgia followed by a characteristic maculovesicular eruption on hands, feet and the oral cavity. In most cases, the disease is self-limiting, but some severe complications, including pneumonia, meningitis and encephalitis, may occasionally occur. The most severe outbreaks of HFMD have been observed in Asia-Pacific region; however, epidemics in Europe and America have also occurred in the past. The disease is caused by an infection with various members of *Picornaviridae* family in the genus enterovirus, most commonly by Coxsackievirus A6 (CVA6), Coxsackievirus A16 (CVA16), and Enterovirus 71 (EV71).

This report describes the intra-familial transmission of hand, foot and mouth disease between 2 sibling children (a 3-year-old girl and a 5-year-old boy) and their immunocompetent mother in Poland. Clinical presentation with signs, symptoms and a suggested treatment regime are discussed and illustrated.

Key words: oral mucosa, hand, foot and mouth disease, enteroviruses, Coxsackie A virus

**Słowa kluczowe:** błona śluzowa jamy ustnej, choroba dłoni, stóp i jamy ustnej, enterowirusy, wirus Coxsackie A

DOI

10.17219/dmp/80995

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## Introduction

Hand, foot and mouth disease (HFMD) is a common childhood illness most typically seen in children younger than 10 years of age and is characterized by fever, malaise and maculovesicular eruptions located mainly on the hands, feet and in the mouth, and less often on the buttocks. In some cases, nail matrix arrest may be affected.<sup>1–3</sup> However, the disease is usually self-limiting and complications such as pneumonia, aseptic meningitis, acute flaccid paralysis or brainstem encephalitis rarely occur.<sup>1,2,4,5</sup>

The disease is caused by an infection with various members of the *Picornaviridae* family in the genus enterovirus; most commonly by Coxsackievirus A6 (CVA6), Coxsackievirus A16 (CVA16) and Enterovirus 71 (EV71). These are small, non-enveloped viruses that consist of a single-stranded, positive-sense RNA. Over 110 genetically distinct enteroviruses responsible for infections in humans and nonhuman primates have been recognized.<sup>4,6</sup> Humans are the only known reservoir of the disease. Enteroviruses spread via fecal-oral, oral-oral and respiratory routes. The basic mode of transmission is the intra-familial path.<sup>1</sup> The incubation period ranges from 3 to 6 days; however, the virus can be shed in the stool and saliva for several weeks after the onset of the primary signs of the infection.<sup>4</sup>

HFMD was identified as an enterovirus infection in the late 1950s.<sup>4,6</sup> Outbreaks of the disease have been mainly due to 2 types of Enterovirus A species: CVA16 and EV71.<sup>7,8</sup> A series of EV-A71 epidemics was observed in the Asia-Pacific region between 1997 and 2010, while the recent outbreaks of severe HFMD in the United States were caused by CVA6, as in Finland.<sup>6,7</sup> Certain symptoms are associated with each of the 2 virus infections: CVA6 causes nail shedding during the convalescence period, while CVA16 leads to the formation of large, vesicular eruptions.<sup>4</sup>

Enterovirus infections remain an important public health problem, mainly due to the HFMD-associated complications during epidemics. Currently, there is no causative treatment or effective vaccinations available; however, several vaccines are being investigated for the HFMD application.<sup>4,6</sup> Due to the high infectivity, the restrictive hygienic procedures in limiting interpersonal spread and isolation of symptomatic cases are essential in preventing HFMD transmission and controlling the outbreaks.<sup>9</sup>

In this paper, we present a case of intra-familial transmission of hand, foot and mouth disease between 2 children attending kindergarten and their generally healthy adult relative in Poland. Prodromal signs, clinical presentation, diagnostics, prophylaxis, and treatment options are also discussed.

### **Case report**

A 3-year-old female child attended the oral pathology office due to maculovesicular skin eruptions affecting perioral region, accompanied by a sore throat and moderately severe discomfort while consuming sweet and acidic food and beverages. These symptoms were preceded by a mild fever and fatigue, which had appeared 2 days before the development of skin and mucosal lesions.

Despite the illness, the child was considered generally healthy. No allergies or nutritional intolerances were reported by the child's parents. The patient had not traveled abroad in the past year, but she had started to attend a kindergarten 2 weeks before.

Informed consent was obtained from the parent as a part of the routine protocol prior to the clinical examination. On examination, red macules, papules and vesicles where revealed in the perioral area, on the hands, the soles of the feet, and buttocks. The lesions where not itchy and moderate discomfort appeared only in the oral area (Fig. 1, 2). During the intraoral examination, the redness of the palatal arches and moderately coated tongue were observed with no other pathologic eruptions being evident.

Based on the characteristic presentation and localization of the lesions on hands, feet, buttocks, and oral region with no other area of the body involved, the diagnosis of hand, foot and mouth disease was established. Meanwhile, the staff of the kindergarten informed the parents of another case of HMFD in the facility.



Fig. 1. Maculovesicular eruptions with crusts in the perioral area of a 3-year-old female patient



Fig. 2. Maculopapular lesions on a hand of a 3-year-old female patient

About 2 days after the development of skin lesions in this 3-year-old girl, a fever accompanied by a sore throat appeared in her 5-year-old brother. These symptoms where followed by single reddish macules present on the plantar hand surfaces and the soles of the feet. Maculovesicular eruptions appeared also on his buttocks. The boy reported a transient tenderness of the palms and soles. No lesions developed in the perioral area, while intraorally, redness and edema, accompanied by a single vesicular eruption on the labial mucosa were observed (Fig. 3).

Like his sister, the boy was considered generally healthy without any relevant medical history. He had not visited any foreign countries in the past year, but he was attending the same kindergarten as his younger sister. Although, due to the absence of the perioral lesions, the clinical presentation in the boy was not as evident as in his sister, based on the symptoms and the history, a diagnosis of hand, feet and mouth diseases was made.

In both children, the acute symptoms of HFMD lasted for approx. 7 days and passed without any complications. Exfoliation on the palms and soles appeared during the healing stage of the infection. This sign was evident in the boy and insignificantly marked in his sister (Fig. 4, 5).

The evaluation of the full blood count, anti-streptolysin O (ASO) and C-reactive protein (CRP) levels performed on the 5-year-old boy 2 weeks after the primary signs of the infection did not show any abnormalities. Also, no pathologies were revealed in the urinalysis.

Approximately 5 weeks after the acute phase of the disease, onychomadesis appeared in both children (Fig. 6).

At the same time when the boy started to suffer the prodromal signs of the HFMD, flu-like symptoms, including malaise, fever, muscle pain, and sore throat, also appeared in the children's mother. It was followed by a very limited reddish rash on the palms and soles with no other pathologic findings. Simultaneously, she reported tenderness of the hands and feet. All the symptoms subsided within 3–4 days. The female was generally healthy; she did not report any complaints and was not using any dental appliances. Considering the history of HFMD in her children and based on very discrete, but rather characteristic clinical symptoms, this female patient was also diagnosed with HFMD.



Fig. 3. A vesicular eruption on the lower lip of a 5-year-old male patient



Fig. 4. Exfoliation of soles in a 5-year-old male patient



Fig. 5. Exfoliation of palms in a 5-year-old male patient



Fig. 6. Onychomadesis on the right thumb with visible Beau's line in 3-year-old female patient

No specific treatment was induced in the members of this family, apart from antipyretic drug containing paracetamol prescribed for the boy patient during the 1<sup>st</sup> day of the infection. Due to the high infectivity of the disease, a disinfecting mouth rinse and a restrictive hygienic regime was recommended.

#### Discussion

Greater exposure to microbial vectors due to changing travel habits and lifestyles, occupational migration, military conflicts, and climate changes, which has occurred in recent times, leads to an increasing frequency of infections caused by new or old, re-emerging viruses. The recurrences of known pathogens and the evolution of additional new variants should be considered. HFMD predominantly affects infants and small children. As a highly contagious disease, it spreads rapidly in childcare facilities and among family members. Although normally the course of the disease is not very dramatic, the morbidity and mortality associated with the recent enterovirus outbreaks has demonstrated the urgent need for effective antiviral treatment and for more education regarding the disease prophylaxis.<sup>1,6,9</sup>

The case series in this paper has demonstrated that HFMD affects not only children or immunocompromised subjects, but in favorable conditions may easily spread to a generally healthy adult person. According to some reports, approx. 11% of adults become infected after the exposition to the pathogen, although less than 1% of those develop the HFMD clinical symptoms.<sup>10,11</sup> Cases of immunocompetent adults suffering from HFMD have also been described by other authors.<sup>10–16</sup> Familial transmission between the child and an adult, similar to the situation observed in our case, was presented in 3 case series by Kaminska et al.<sup>12</sup> Viral transmission occurred faster between the infected pediatric patient and an adult than between 2 adults. Intra-familial transmission between a small child and immunocompetent adults was also the subject of case reports by Omaña-Cepeda et al. and Tai et al.<sup>13,14</sup> An unusual location of erythematous crusted macules on the scalp, which accompanied a typical spectrum of clinical symptoms in a generally healthy adult, was presented by Andreoni and Colton.<sup>15</sup> In that case, a resolution of oral and cutaneous eruptions was followed by onychomadesis. Regrowth of new fingernails appeared approx. 2 months post infection. A total of 5 adult cases of CVA16-confirmed HFMD with similar disease progression were presented by Ramirez-Fort et al.<sup>10</sup> In all the subjects, the prodromal symptoms were followed by the development of rash on the palms and soles, and to a lesser degree on the facial skin and the buttocks. The cutaneous lesions evolved from macules to vesicles, followed by erosions. The subsequent desquamation was than observed. The authors emphasized

that erythematous or pruritic macular lesions in HFMD may mimic those of secondary syphilis. No systemic complications, e.g., encephalitis or myocarditis, where observed in those patients. Meanwhile, in an adult case study of HFMD presented by Flor de Lima et al., typical oro-cutaneous eruptions were followed by the development of myopericarditis, rarely described as a complication of this condition.<sup>16</sup>

In most cases, HFMD in adults can be easily diagnosed based on clinical grounds and the patient's history. In differential diagnosis, several conditions must be considered, including varicella zoster, papular urticaria, impetigo, or syphilis.<sup>10,17</sup> In doubtful cases, rapid molecular diagnostic methods should be utilized to recognize an enteroviral disease.<sup>5</sup> Histopathologic findings, which typically include intense edema, necrotic or shadow keratinocytes, and neutrophilic exocytosis with T-cell infiltrate, may assist the diagnosis.<sup>18</sup> Currently, there is neither an effective antiviral therapy, nor an effective vaccine available for the disease. Supportive care, including maintenance of hydration and pain control, together with antipyretics and optionally antihistamines to reduce itching, are recommended for the management of patients with HFMD.<sup>1,4,6</sup>

It needs to be emphasized that the disease is very contagious and has the potential to spread very quickly through a large population. To avoid major outbreaks, containment of the disease once it is diagnosed is required. Strict implementation of basic protocols like monitoring cleanliness of the hands, utensils and drinking water, together with preventing affected children from attending school and other childcare facilities is recommended.<sup>2,4,9</sup>

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# 3<sup>rd</sup> Conference of the Polish Society of Periodontology Warsaw, Poland, 2–3 March 2018

Dental and Medical Problems, ISSN 1644-387X (print), ISSN 2300-9020 (online)

Dent Med Probl. 2018;55(1):105-114

### 1. Lack of association between the *IL17F* gene rs763780 and rs2275913 and periodontal disease

Małgorzata Mazurek-Mochol<sup>1</sup>, Elżbieta Dembowska<sup>1</sup>, Ryta Łagocka<sup>2</sup>, Małgorzata Kozak<sup>3</sup>, Marek Sawczuk<sup>4</sup>, Agnieszka Maciejewska<sup>4</sup>, Damian Malinowski<sup>5</sup>, Krzysztof Safranow<sup>6</sup>, Andrzej Pawlik<sup>7</sup>

<sup>1</sup>Department of Periodontology, Pomeranian Medical University, Szczecin, Poland

- <sup>2</sup> Department of Conservative Dentistry, Pomeranian Medical University, Szczecin, Poland
- <sup>3</sup> Department of Prosthetics, Pomeranian Medical University, Szczecin, Poland

<sup>4</sup> Faculty of Physical Education and Health Promotion, University of Szczecin, Poland

<sup>5</sup> Department of Pharmacology, Pomeranian Medical University, Szczecin, Poland

<sup>6</sup> Department of Biochemistry and Medical Chemistry, Pomeranian Medical University, Szczecin, Poland

<sup>7</sup> Department of Physiology, Pomeranian Medical University, Szczecin, Poland

**Background.** Periodontal disease is a common oral disease characterized by the degradation of the periodontal attachment and alveolar bone. Inflammatory and immune responses to oral microorganisms initiate the development of periodontitis. These responses protect gingival tissues against local microbial invasion, but also induce an inflammatory cascade. The pathogens induce the production of cytokines, such as interleukin 17 (IL-17), which enhances inflammatory response and progression of the disease. IL-17 is a proinflammatory cytokine and is produced by Th17 cells and neutrophils; it consists of IL-17A-IL17F. IL-17 family cytokines have been demonstrated to be crucial in host defense against microbial organisms. Several polymorphisms have been detected in *IL-17* gene and have been examined in various inflammatory diseases.

**Objectives.** In this study, we examined the association between the *IL17F* gene rs763780 and rs2275913 polymorphisms and periodontal disease.

**Material and methods.** This study enrolled 360 Caucasian subjects (age range: 25–69 years) from the West Pomeranian region of Poland. The subjects were submitted to anamnesis and to clinical and periodontal examination. The subjects were divided into 2 subgroups: patients with periodontal disease and healthy subjects without periodontal disease. The first group was comprised of 200 patients (87 men, 113 women), aged 26–69 years (mean 50.47  $\pm$ 9.09), with chronic periodontal disease, diagnosed using the periodontal disease classification system of the American Academy of Periodontology. All samples were genotyped in duplicate using allelic discrimination assays with TaqMan<sup>®</sup> probes (Applied Biosystems, Carlsbad, USA) on a 7500 Fast Real-Time PCR Detection System (Applied Biosystems).

**Results.** There were no statistically significant differences in the distribution of *IL17F* gene rs763780 and rs2275913 polymorphisms genotypes and alleles between patients with periodontal disease and control subjects, as well as between smoking patients with periodontal disease and smoking control subjects, and between non-smoking patients with periodontal disease and non-smoking control subjects.

**Conclusions.** The results of this study suggest there is no association between the *IL17F* gene rs763780 and rs2275913 polymorphisms and periodontal disease.

Key words: IL17F, polymorphism, periodontal disease

Address for correspondence: malgorzata.mazurek@poczta.onet.pl

### 2. Lack of association between the *CD28* gene rs3116496 polymorphism and periodontal disease

Małgorzata Mazurek-Mochol<sup>1</sup>, Elżbieta Dembowska<sup>1</sup>, Ryta Łagocka<sup>2</sup>, Małgorzata Kozak<sup>3</sup>, Marek Sawczuk<sup>4</sup>, Agnieszka Maciejewska<sup>4</sup>, Damian Malinowski<sup>5</sup>, Krzysztof Safranow<sup>6</sup>, Andrzej Pawlik<sup>7</sup>

<sup>1</sup>Department of Periodontology, Pomeranian Medical University, Szczecin, Poland <sup>2</sup>Department of Conservative Dentistry, Pomeranian Medical University, Szczecin, Poland

<sup>3</sup>Department of Prosthetics, Pomeranian Medical University, Szczecin, Poland

<sup>4</sup> Faculty of Physical Education and Health Promotion, University of Szczecin, Poland

<sup>5</sup> Department of Pharmacology, Pomeranian Medical University, Szczecin, Poland

<sup>6</sup>Department of Biochemistry and Medical Chemistry, Pomeranian Medical University, Szczecin, Poland

<sup>7</sup>Department of Physiology, Pomeranian Medical University, Szczecin, Poland

**Background.** Periodontal disease is a chronic inflammatory disease characterized by the infiltration of inflammatory cells. The immune system has developed several mechanisms regulating the activation of T cells. The main mechanism is based on the stimulation of T cells by a second signal in addition to that derived from the T cell receptor (TCR). As a stimulatory signal transducer, CD28 plays an important role in T cells co-stimulation. The binding of CD28 to its ligands promotes the activation of a TCR-stimulated T cell. Several polymorphisms have been detected in *CD28* gene and examined in various inflammatory diseases.

**Objectives.** In this study, we examined the association between the *CD28* gene rs3116496 polymorphism and periodontal disease.

**Material and methods.** This study enrolled 360 Caucasian subjects (age range: 25–69 years) from the West Pomeranian region of Poland. The subjects were submitted to anamnesis and clinical and periodontal examination, and then divided into 2 subgroups: patients with periodontal disease and healthy subjects without periodontal disease. The first group was comprised of 200 patients (87 men, 113 women), aged 26–69 years (mean age 50.47  $\pm$ 9.09 years), with chronic periodontal disease, diagnosed using the periodontal disease classification system of the American Academy of Periodontology. All samples were genotyped in duplicate using allelic discrimination assays with TaqMan<sup>®</sup> probes (Applied Biosystems, Carlsbad, USA) on a 7500 Fast Real-Time PCR Detection System (Applied Biosystems).

**Results.** There were no statistically significant differences in the distribution of CD28 rs3116496 genotypes and alleles between patients with periodontal disease and control subjects, as well as between smoking patients with periodontal disease and smoking control subjects, and between non-smoking patients with periodontal disease and non-smoking control subjects.

**Conclusions.** The results of this study suggest there is no association between the *CD28* gene rs3116496 polymorphism and periodontal disease.

Key words: CD28, polymorphism, periodontal disease

Address for correspondence: malgorzata.mazurek@poczta.onet.pl

#### 3. Clinical significance of periodontal indexes in patients with acute myocardial infarction

Anna Surtel<sup>1</sup>, Robert Klepacz<sup>2</sup>, Małgorzata Kiernicka<sup>1</sup>, Joanna Wysokińska-Miszczuk<sup>1</sup>

<sup>1</sup>Department of Periodontology, Medical University of Lublin, Poland <sup>2</sup>Department of Clinical Pathomorphology, Medical University of Lublin, Poland

**Background.** High morbidity and mortality caused by cardiovascular diseases (CVD) is a significant clinical and social challenge in highly developed countries. Many multidirectional and interdisciplinary researches aim to determine CVD risk factors. It has been confirmed that atherosclerosis leads to ischemic changes, and is directly responsible for CVD. Inflammation, both in atherosclerosis and periodontal disease, leads to increased inflammatory response, which is reflected into the value of inflammatory markers and mediators, and consequently in the clinical view of CVD.

**Objectives.** The aim of the paper was the assessment of the clinical significance on the periodontium condition in patients with acute myocardial infraction. The study was the attempt to determine the relationship between selected markers of heart failure and periodontal indexes.

**Material and methods.** In the study, 71 patients with diagnosed acute myocardial infraction hospitalized and treated in the Cardiology Clinic SPSK 4 in Lublin were qualified for the examination. The individuals who hadn't received anticoagulant therapy (clopidogrel) were excluded. Finally, the study included 68 patients (53 men and 15 women) with the mean age  $54.25 \pm 7.00$  years. The average body mass index (BMI) was  $28.37 \pm 4.18$  kg/m<sup>2</sup>. The selected components of lipid profile: total cholesterol (TC), low-density lipoprotein (LDL), high-density lipoprotein (HDL), and triglycerides (TG) as well as biomarkers of heart failure and acute myocardial infraction: high sensitivity C-reactive protein (hs-CRP), fibrinogen, N-terminal pro-B-type natriuretic peptide (NT-proBNP) were determined on the basis of the medical history. For the assessment on periodontium condition, following periodontal parameters were examined: approximal plaque index (API), bleeding on probing (BOP), pocket depth (PD), and clinical attachment level (CAL). The periodontal examination was performed on the 3<sup>rd</sup> day of hospitalization using a basic set and periodontal probe HuFriedy PCPUNC 15<sup>®</sup>.

**Results.** In the study group, the mean value of TC, LDL, blood glucose, hs-CRP, OB, fibrinogen, and NT-proBNP were elevated. The average periodontal parameters indicated poor oral hygiene (API – 78.16 ±25.7%), generalized inflammation (BOP – 43.81 ±23.5%) and moderate periodontitis (CAL – 3.14 ±1.97 mm). The rank (compartmentalized) level components of lipid profile and markers of heart failure (according to the references values) did not statistically differentiate the periodontal parameters in the patients with acute myocardial infraction.

**Conclusions.** There was no directly correlation between the periodontal indexes and markers of heart failure and acute myocardial infraction. Regular monitoring of the periodontium condition, alongside the components of lipid profile should be part of the CVD prophylaxis.

**Key words:** cardiovascular disease, periodontal status indicators, marker for cardiac dysfunction, lipid profile

Address for correspondence: asu1@op.pl
### 4. Oral health (dental and periodontal status) in multiple sclerosis patients: Preliminary report

Anna Kurhańska-Flisykowska<sup>1</sup>, Jacek Wencel<sup>2</sup>,

Kacper Nijakowski<sup>1</sup>, Jakub Doroszewski<sup>1</sup>, Włodzimierz Łojewski<sup>3</sup>, Marzena Wyganowska-Świątkowska<sup>1</sup>, Radosław Kaźmierski<sup>2</sup>, Anna Surdacka<sup>3</sup>, Anna Kowalska<sup>4</sup>

- <sup>1</sup>Department of Oral Surgery and Periodontology, Poznan University of Medical Sciences, Poland
- <sup>2</sup> Department of Neurology and Vascular Diseases of the Nervous System, Poznan University of Medical Sciences, Poland
- <sup>3</sup>Department of Conservative Dentistry and Endodontics, Poznan University of Medical Sciences, Poland
- <sup>4</sup> Institute of Human Genetics, Polish Academy of Sciences, Poznań, Poland

**Background.** Emerging evidence suggests that poor oral health influences the initiation and/or progression of diseases such as atherosclerosis (including myocardial infarction), stroke or diabetes mellitus. Specific bacterial ligands increase the expression of proinflammatory molecules, which activates the innate and adaptive immune systems. Recent studies have shown that systemic, peripheral infections also affect central nervous system (CNS). Periopathogens may be a risk factor for common diseases of CNS with neurodegenerative, vascular, and autoimmunological background. Evasion of pathogens from destruction by the host immune reactions leads to persistent infection, chronic inflammation, neuronal destruction, and amyloid beta (A $\beta$ ) deposition. A $\beta$  has been shown to be a pore-forming antimicrobial peptide, indicating that A $\beta$  accumulation might be a response to infection. Chronic infection can cause slow progressive dementia and cortical atrophy.

**Objectives.** Very little is known about the impact of periopathogens on the etiology and course of multiple sclerosis (MS). Therefore, to explore a link between periodontitis and MS, we performed a detailed analysis of dental and periodontal status among patients with the disease.

**Material and methods.** A cohort of 42 patients with MS (aged 22–57 years, with a low physical disability) was collected at the Clinic of Vascular Diseases of Nervous System, Poznan University of Medical Sciences. in 2016–2017. Patient's dental and periodontal status was assessed according to the WHO Oral Health Data chart including approximal plaque index (API), bleeding on probing (BOP), clinical attachment level (CAL), and probing depth (PD).

**Results.** Among patients with MS, 71% were treated with interferon, 12% with Gilenya, 10% with Copaxone, and 7% with Tecfidera. Regardless of the drug, 74% of patients complained of dry mouth. Examination outcomes were meanly: DMFT – 15, DMFS – 36.5. The average API was 11% and BOP – 8%. About 17% of patients presented gingivitis and 5% had periodontitis; 19% of examined group suffered from bruxism. We did not found any special difference in oral health status between the MS group and healthy controls. Among post stroke patients examined at the same time, we observed opposite results: a prevalence of oral pathologies was twice as high as in the age-matched control subjects. This may suggest that patients with MS have a very good motivation to avoid any infection or inflammation form – they are afraid of being excluded from treated group. **Conclusions.** Dental and periodontal examination performed among patients with MS revealed very strong patients' motivation to keep their oral health in good condition.

Key words: multiple sclerosis, oral inflammation, periodontitis, caries

Address for correspondence: anna.kowalska@igcz.poznan.pl

### 5. Oral cavity state in rheumatoid arthritis patients on the basis of self-assessment questionnaire investigation

Małgorzata Radwan-Oczko<sup>1</sup>, Irena Duś<sup>1</sup>, Anne Maria Thomsen<sup>2</sup>, Claus Rasmussen<sup>2</sup>

<sup>1</sup>Department of Oral Pathology, Wroclaw Medical University, Poland <sup>2</sup>Division of Rheumatology, Clinic Medicine, North Jutland Region Hospital, Hjørring, Denmark

Background. Rheumatoid arthritis (RA) and chronic periodontal disease (chronic periodontitis – CP) are 2 common chronic, destructive and progressive diseases. RA is an inflammatory autoimmune disorder affecting more than 1.5% of the world's population, with higher female percentage, characterized by chronic pain and joint destructions - primarily of the hands, wrists and feet. Apart from the evidence for a link between the oral health and general health related to the periodontal inflammation and cardiovascular diseases, diabetes, and respiratory diseases, also the association between rheumatoid arthritis and poor oral health has been investigated for last decades in epidemiological, clinical, and basic science research. Some of the clinical studies have shown an increased frequency of periodontal disease in patients with RA as compared to healthy individuals. Current findings strongly support the hypothesis that chronic periodontal disease could be a factor in the initiation and maintenance of the autoimmune inflammatory responses that occur in RA. In a systematic review and meta-analysis of 17 studies including a total of 153,492 participants, the relationship between RA and CP was found to be significant.

**Objectives.** The purpose of this study was to asses chosen parameters of RA patient's oral cavity state with the use of a self-assessment questionnaire.

**Material and methods.** Danish citizens have access to free pediatric dental care (since 1960's) and partial reimbursement for routine adult dental care. In the study, 300 patients under treatment for RA from Division of Rheumatology, Clinic Medicine, North Jutland Region Hospital, Hjørring, Denmark were included. Questionnaires were emailed to the patients, and 164 completed forms have been received.

**Results.** The mean age of the group of 164 patients was 65 years. As much as 47% of patients have never smoked, 41% were smokers in the past, and 12% were active smokers. Only 3.5% of patients pointed out to have frequent problems with dry mouth when chewing or swallowing. In 1.5% of patients, this symptom was present only sometimes. Patients estimated their status of the teeth as poor in 13%, good in 46%, and excellent in 40% of cases. Moreover, they estimated the status of gingiva as poor in 11%, good in 49%, and excellent in 40% of cases. About 15% of patients (often 1% and sometimes 14%) experienced spontaneous gingival bleeding, however gingival bleeding

during or after tooth brushing were observed frequently in 49% of patients (in 6% and 43%, respectively). Only 21% of patients rinsed their mouth (therein 6% used rinsing often and 15% only sometimes). A total of 14% of patients complained about feeling of loose/movable teeth. Any difficulties in biting or chewing were revealed in 10% of patients.

**Conclusions.** The status of the oral cavity revealed in investigation by Danish RA patients can be perceived as quite satisfactory. However, some of the parameters indicate the possibility of presence of gingival/periodontal infection which can make symptoms of rheumatoid arthritis worse and disrupt disease management.

Key words: rheumatoid arthritis, oral cavity state

Address for correspondence: malgorzata.radwan-oczko@umed.wroc.pl

### 6. Application of radiovisiography in periodontal therapy

Magdalena Batko<sup>1</sup>, Andrzej Miskiewicz<sup>2</sup>

<sup>1</sup> Students' Scientific Association, Department of Periodontology, Medical University of Warsaw, Poland

<sup>2</sup> Department of Periodontology, Medical University of Warsaw, Poland

**Background.** X-ray radiovisiography (RVG) is a modern diagnostic technique used in periodontal treatment and dental plaque retentive factors detection.

**Objectives.** The aim of the study was to establish the correlation between periodontitis local factors and the degree of the lesions they lead to. Moreover, we have evaluated the presence of the most frequent local factors for periodontitis and their impact on the course of the disease.

**Material and methods.** A retrospective analysis of 226 radiovisiographs was performed. We selected cases which illustrated overlapping dental restorations in permanent dentition. The analysis included RVG presenting a unilateral tooth in the same dental arch. Planmeca Romexis Viewer<sup>®</sup> was used to perform the research. The study covered the assessment of the patients' age and sex, general condition of the alveolar bone or alveolar mandible, the presence and the correct contact with the opposite arch, the state of the adjacent tooth, and the same information about the unilateral tooth. Inappropriate fillings were also assessed. The obtained results were subjected to statistical analysis by means of STATISTICA 13.1. Values for which the significance level was p < 0.05 were considered valid and statistically significant.

**Results.** The results of the study revealed that the state of periodontium was significantly worse in patients with class II (according to Black's classification) restorations on proximal and distal tooth walls (Mann-Whitney U test; p < 0.0078; U = 4.16). The average bone lesion measured by the RVG adjacent to the abovementioned class II restoration was 1.95 mm, whereas in teeth with a 3-wall restoration on the opposite side of the dental arch, the loss was significantly higher (4.01 mm; p < 0.0026; U = 7.12). The Spearman's rank correlation coefficient, used as a nonparametric measure, calculated between the size of overhanging filling and the edge of the alveolar crest was R = 0.64, thus confirming a strong correlation.

Key words: radiovisiography, periodontal therapy, overhanging dental filling

Address for correspondence: sluzowki@wum.edu.pl

### 7.

### The new porous matrix based on gelatin/hydroxyethyl cellulose blend for local treatment of advanced chronic periodontitis: A pilot study

Małgorzata Szulc<sup>1</sup>, Dorota Kida<sup>2</sup>, Aleksandra Sender-Janeczek<sup>1</sup>, Irena Duś-Ilnicka<sup>3</sup>, Bożena Karolewicz<sup>2</sup>

<sup>1</sup>Department of Periodontology, Wroclaw Medical University, Poland <sup>2</sup>Department of Drug Form Technology, Wroclaw Medical University, Poland <sup>3</sup>Department of Oral Pathology, Wroclaw Medical University, Poland

**Background.** The goals of periodontal therapy are to alter or eliminate the microbial etiology and contributing risk factors for periodontitis. Scaling and rootplanning procedure is often complemented by local or systemic antimicrobials administration. Due to the fact that antibiotics are prescribed systemically too often, the global problem of antibiotic resistance is becoming one of the major scientific issues of modern times. In local drug delivery, the major goal is to supply therapeutic levels of a drug agent to a periodontal pocket for a prolonged period, reducing possible systemic side effects. Unfortunately, there is limited availability of locally applied periodontal drugs in Poland.

**Objectives.** The aim of the study was the assessment of the efficacy of porous matrix based on gelatin/hydroxyethyl cellulose blend in treatment of deep periodontal pockets resulting from severe chronic periodontitis. The matrix was invented and produced in the Department of Drug Form Technology, Wroclaw Medical University, Poland.

**Material and methods.** To assess the clinical efficacy of the new porous matrix based on gelatin/hydroxyethyl cellulose blend, the matrix was loaded with metronidazole. The porous metronidazole (MTZ) loaded matrix was fabricated by the freeze-drying technique. The morphology, mechanical properties, in vitro degradation, MTZ release from dried sponge were evaluated, and also clinical trials were performed. 15 periodontal pockets >6 mm were examined. To evaluate oral hygiene, API and the gum inflammation indexes – BOP and PBI were used. The clinical examination was performed 3 times. During the 1<sup>st</sup> visit the output indicators were defined. The subgingival scaling was performed and metronidazole on porous matrix was applied to the selected periodontal pockets. Two control tests of periodontal indexes were carried out after 1 week and 1 month after the application. During each visit the probes from periodontal pockets *(Pg.)* DNA.

**Results.** The subgingival application of the assessed matrix did not result in any negative side effects. It was easy to apply and resorbable. No concern was raised by any of the patients in regard to the treatment. The decrease of all tested clinical parameters was observed due to the use of metronidazole. In the samples from 2 pockets, no genetic material of *P.g.* was identified in the PCR test. In the case of all other patients, each collected sample contained the DNA of *P.g.* After a single application of metronidazole, the eradication of bacteria from the periodontal pockets was not successful. However, there was an observable difference noted in the color intensity of the test strap from the 2<sup>nd</sup> test (after 7 days) compared to the 1<sup>st</sup> and 3<sup>rd</sup> test (after 1 month). **Conclusions.** The in-pocket application of the new drug form seems to be safe and effective. It can complement classical periodontitis treatment. The new porous matrix can be loaded with many antibiotics or antiseptics and can become a valuable alternative for the use of systemic antibiotics in periodontal diseases.

**Key words:** local delivery, antimicrobials, chronic periodontitis, microflora

Address for correspondence: malgorzata.szulc@umed.wroc.pl

8.

### Does treatment exclusively in a private dental practice has any effect on selected oral health indicators in patients aged 65–74?

Dariusz Chrzęszczyk<sup>1</sup>, Łukasz Zawada<sup>2</sup>, Joanna Toczewska<sup>1</sup>, Beata Głowacka<sup>1</sup>, Tomasz Konopka<sup>1</sup>

<sup>1</sup>Department of Periodontology, Wrocław Medical University, Poland <sup>2</sup>Private dental practice, Oława, Poland

**Background.** Socio-economical factors have tremendous effect on oral health, especially on the number of remaining teeth, prevalence of dental caries, periodontitis, and dental treatment needs. This influence is even stronger in epidemiological studies among geriatric patients. The form of dental care – treatment financed by the patient in a private dental practice or funded by the government – is one of these factors, and is undeniably related to income and education.

**Objectives.** The aim of the study was to evaluate the effect of the form of dental treatment (only in a private dental practice; only within the procedures funded by the National Health Fund – NFZ) on the number of remaining teeth, dental treatment needs, dental caries and periodontitis prevalence, and selected oral pro-health behaviors among the residents of Wrocław area aged 65-74.

Material and methods. The study group consisted of 231 randomly selected people with symmetrical age and sex distribution, treated in dental offices either in Wrocław or Oława: 120 exclusively in private practices, 111 exclusively within the procedures funded by the government. Clinical evaluation of patients included the number of teeth (third molars were excluded), the number of occlusal supporting zones, either on natural teeth or on fixed prosthetics, the presence of dental plaque on buccal, lingual (PI index by O'Leary et al.) and approximal (API index by Lange et al.) surfaces of teeth, and bleeding on probing (BoP index by Ainamo and Bay) at 4 measuring points. Additionally, periodontitis - PD (for each tooth at 4 measuring points, the mean value of PD for all teeth and the number of pockets deeper than 5 mm) and clinical attachment level - CAL (for each tooth at 4 measuring points, the number of approximal surfaces with CAL > 3 mm and > 5 mm) values were recorded. Based on the periodontal examination, community periodontal index (CPI) was assessed and diagnosis was made according to the guidelines of Centers for Disease Control and American Academy of Periodontology (CDC/AAP). Also, the DMF number and its components were recorded.

**Results.** In patients treated only in private dental practices, statistically significant higher number of remaining teeth (17.4 vs 8.3), higher percentage of

people with more than 20 teeth (47% vs 9%) and higher number of fillings (6 vs 2.4) were observed. These patients more often visited dental offices annually, brushed their teeth at least 2 times/day and cleaned interdental spaces. Additionally, there were less edentulous cases (5% vs 35%), less teeth extracted as a result of complications of dental caries (9.4 vs 15.1) and periodontal disease (1.2 vs 4.6), lower values of PI, API (better oral hygiene) and BoP (a smaller extent of gingival inflammation) indicators. On the other hand, there were no differences in recorded periodontal parameters (PD, CAL, CPI, diagnosis according to CDC/AAP).

**Conclusions.** Treatment in a private dental practice was shown to be a protective factor in relation to the number of teeth, dental treatment needs and pro-health dental behaviors. It had no influence on periodontitis prevalence and resulting from this periodontal treatment needs.

Key words: dentition status, older people, epidemiological study, dental care

Address for correspondence: darek.chrzeszczyk@interia.eu

#### 9.

### Efficacy of a coronally advanced flap with a xenogenic collagen matrix in the treatment of gingival recession: Preliminary results of a randomized clinical trial

Rafał Wiench<sup>1</sup>, Anita Ilków<sup>2</sup>, Justyna Kulczycka<sup>1</sup>, Łukasz Gilowski<sup>3</sup>, Patrycja Stelmańska<sup>1</sup>, Natalia Stefanik<sup>1</sup>, Przemysław Jabłoński<sup>4</sup>, Tadeusz F. Krzemiński<sup>5</sup>

<sup>1</sup>Department of Periodontal Disease and Oral Mucosa, School of Medicine with the Division of Dentistry in Zabrze, Medical University of Silesia in Katowice, Poland

<sup>2</sup>Private dental practice, Opole, Poland

<sup>3</sup> Private dental practice, Gliwice, Poland

<sup>4</sup> Private dental practice, Kłobuck, Poland

<sup>5</sup> Department of Pharmacology, School of Medicine with the Division of Dentistry in Zabrze, Medical University of Silesia in Katowice, Poland

**Background.** Gingival recession is an esthetical problem that can lead to hypersensitivity or root caries. Connective tissue graft from the palatine in connection with the advanced flap technique is considered the gold standard for the surgical treatment. Nowadays, a possible alternative to the autologous connective tissue graft (CTG) is the use of a xenogenic collagen matrix.

**Objectives.** The aim of this RCT was to compare the efficacy of root coverage using the coronally advanced flap with connective tissue graft or a collagen matrix in the treatment of multiple recessions Miller Class I and II.

**Material and methods.** Sixteen patients have been evaluated after 3 months so far. The patients in the study group were treated with a xenogenic collagen matrix using the coronally advanced flap. The patients in the control group were treated with the same surgical method and CTG. The clinical parameters of recessions and the percentage of the average recession coverage (ARC) were evaluated at baseline and 3 months after the surgery. Follow-up examinations are planned after 6, 9 and 12 months. **Results.** Significant decreases were recorded in both groups with regard to all the clinical parameters in comparison to the baseline measurements, but no significant differences between the treatment groups were observed. The recession depth decreased from 3.1 mm to 0.4 mm in the control group and from 2.5 mm to 0.7 mm in the study group. The gain in the width of the keratinized tissue was obtained (from 2.2 mm at baseline to 4.0 mm after 3 months in the control group, and adequately 2.5 mm and 2.9 mm in the study group). The mean ARC was 84% in the test group after 3 months and 91% in the control group, but the difference between the groups was insignificant.

**Conclusions.** The preliminary results of multiple recession coverage with the coronally advanced flap using a xenogenic collagen matrix or CTG are similar. Clinically important differences between the groups are statistically insignificant because of the small number of patients examined after 3 months. Larger groups of patients observed for a longer period of time are required to better judge the efficacy of these methods.

**Key words:** gingival recession, coronally advanced flap, collagen matrix, connective tissue graft, mucogingival surgery

Address for correspondence: aplg@wp.pl

### 10. The correlation between clinical and histopathological diagnoses of oral white lesions: A retrospective study of leukoplakia and oral lichen planus

Maciej Kuzio<sup>1</sup>, Karolina Wapniarska<sup>1</sup>, Marian Danilewicz<sup>2</sup>, Natalia Lewkowicz<sup>1</sup>

<sup>1</sup>Department of General Dentistry, Medical University of Lodz, Poland <sup>2</sup>Department of Pathomorphology, Medical University of Lodz, Poland

**Background.** The presence of white lesions in oral cavity can cause diagnostic difficulties even for experienced clinicians. Multitude of diseases and similarities in their clinical manifestation can lead to misdiagnosis and ineffective treatment. Considering potentially malignant character of lesions like leukoplakia and oral lichen planus (OLP), the proper diagnosis is crucial.

**Objectives.** The aim of this study was to assess the concordance of clinical and histopathological diagnoses of biopsied white lesions.

**Material and methods.** The results of 401 biopsies from oral cavity were retrospectively analyzed. A total of 114 results with initial or histopathological diagnosis of leukoplakia (33 cases) or OLP (81 cases) were selected. The initial diagnosis was then compared with the histopathological result found in each patient's records. The diagnosis was accepted for concordance if the histopathological result was decisive. In case of an indecisive histopathological result, the initial diagnosis was considered unlikely. If both diagnoses were different, the possibility of clinical or histopathological error was investigated. The correspondence of both diagnoses was evaluated as follows: 1. clinical initial diagnosis (ID); 2. final histopathological diagnosis (HD); 3. corresponding data (CD) – cases of both diagnoses overlapping. The result was calculated using the formula: final concordance FC = CD × 100%/ID, showing the percentage of corresponding data. **Results.** The correspondence between clinical diagnosis of oral leukoplakia and histopathological results was 15.15%, and for OLP – 49.37%. Most commonly found histopathological diagnosis in clinical suspicion of leukoplakia was frictional keratosis (51.51%). In 16.46% cases of ID of OLP, frictional keratosis was diagnosed histopathologically. In 1 case of initial suspicion of OLP, a squamous cell carcinoma was found (1.27%).

**Conclusions.** White lesions like leukoplakia and OLP can be challenging to diagnose only by their clinical features. Frictional keratosis could clinically resemble both leukoplakia and plaque-like OLP. Histopathological data should be considered an integral part of final diagnosis. Clinical evaluation of white lesions still needs improvement, and histopathological criteria of diagnosis require further standardization. In future, molecular biomarkers may improve definitive diagnosis of both leukoplakia and OPL.

Key words: leukoplakia, lichen planus, diagnosis, biopsy

Address for correspondence: natalia.lewkowicz.nl@gmail.com

### 11. SPARC protein expression in different stages of oral mucous membrane epithelium pathologies

Irena Duś-Ilnicka<sup>1</sup>, Agnieszka Hałoń<sup>2</sup>, Małgorzata Radwan-Oczko<sup>1</sup>

<sup>1</sup>Department of Oral Pathology, Wroclaw Medical University, Poland <sup>2</sup>Department of Pathomorphology and Oncological Cytology, Wroclaw Medical University, Poland

**Background.** There are suggestions that molecular basis for malignant transformations of oral mucous membrane epithelium might be related with strengthened tissue reconstruction and intensified under different stimulus reconstruction processes. A secreted protein, acidic and rich in cysteine (SPARC), is a phosphorylated glycoprotein with a molecular mass of 32–43 kDa. The presence of this protein has been demonstrated in tissues that undergo frequent renewal, such as the gut epithelium. The contribution of SPARC in the progression of carcinogenesis confirms its prevalence in malignant tumors. SPARC is considered to be a protein inducing the angiogenesis. In the presented research we evaluated SPARC production in different histopathological states of oral mucous compared with physiological mucous membrane (negative control) and oral squamous cell carcinoma (SCC).

**Objectives.** The aim of this study was to evaluate the immunohistochemical SPARC protein expression in different stages of clinically diagnosed leukoplakia.

**Material and methods.** The evaluated material consisted of 2 groups of patients' oral mucous membrane tissue biopsies, formalin-fixed, paraffin-embedded (FFPE) samples: group of 37 samples with clinically diagnosed leukoplakia, and the group of 29 samples with oral SCC diagnosis. Archival FFPE blocks were utilized from the archives of the Department of Pathomorphology and Oncological Cytology, Wroclaw Medical University. For immuno-histochemical research SPARC Mouse Monoclonal Antibody (AON-5031, Santa Cruz Biotechnology, US) was applied. The samples were incubated in an automated system IHC Dako Link48 Autostainer®. EnVison FLEX® (DakoCytomation, Glostrup, Germany) was used to visualize the color reaction reagent.

**Results.** The expression of the SPARC protein in the group of clinically diagnosed oral mucosa leukoplakia was determined in 30 cases. Score 0 was detected in 66.67%, score 1 in 30% of cases, score 3 and 4 were not observed in the tested samples. The expression of the SPARC protein in the group of oral mucosa SCC was assessed in 26 cases. Score 0 was detected in 11.54%, score 1 in 34.62%, score 2 was expressed in 30.77%, and score 3 in 15.15%. The highest score – 4, was shown in 7.69% of the assessed samples.

**Conclusions.** An increased SPARC production was observed during the progression of pathological changes of oral mucous membrane, especially from the stage of cell dysplasia. The production of SPARC might be induced by dysplastic cells proliferation in the first stage of pathological development. However, its expression is not a proper marker of metastasis in oral SCC, but might be a marker for malignancy of the pathomorphological diagnosis of leukoplakia for which meta-analysis should be provided.

Key words: SPARC, oral leukoplakia, oral squamous cell carcinoma

Address for correspondence: malgorzata.radwan-oczko@umed.wroc.pl

#### 12.

### Prediction of the level of cancer risk of oral mucosa based on the results of blood and urine tests and selected elements of medical history: Preliminary results

Zbigniew Suszyński<sup>1</sup>, Joanna Łoś<sup>2</sup>, Robert Świta<sup>1</sup>, Michał Dojs<sup>3</sup>, Anna Dojs<sup>3</sup>

- <sup>1</sup>Chair of Multimedia Systems and Artificial Intelligence, Faculty of Electronics and Computer Sciences, Koszalin University of Technology, Poland
- <sup>2</sup>Department of Medical Emergency, Institute of Humanities, State Higher Vocational School in Koszalin, Poland
- <sup>3</sup> Specialist Periodontal Practice, Gorzów Wielkopolski, Poland

**Background.** Autofluorescence devices help detect abnormalities but they are not able to evaluate the predicted risk of oral mucosa cancer.

**Objectives.** The aim of this investigation was to assess formal statistical analysis of the relationship between the threat of oral mucosa cancer, the results of laboratory tests such as blood and urine, and smoking, age and patrient's sex.

**Material and methods.** The above analysis was performed in a group of 62 patients, including 6 reference ones. The analysis included the results of tests performed several years before, which allowed us to compare their results with the current state of patients' health. Forty-two characteristics were assigned to each patient. The test method used the principal component analysis (PCA) for a standardized observation matrix. The direction of hazard level diversification was determined as a resultant between the 2<sup>nd</sup> and the 3<sup>rd</sup> major direction.

**Results.** The most important elements increasing the risk of cancer were, among others: patient's age, smoking, and the increased levels of MCV, MCH, % of lymphocytes, % of basocytes, a number of basocytes, ESR, glucose, tri-glycerides, and CRP. At the same time, the risk of neoplastic disease decreased with the increased RBC, hemoglobin, MCHC, and prothrombin time. Relatively

neutral characteristics, i.e., those for which no strong relationship was found with the positioning of the patient in the cancer threat scale included: WBC, platelets, and all forms of cholesterol. The impact of individual elements, like the scale of cancer risk, is quantitative, which allows individual positioning of patients in the risk group. The described method of patients' qualification has been verified with the knowledge of the attending physician, the history of the disease and the results of other tests.

**Conclusions.** The results of this analysis are preliminary. They can be affected by, among others, missing data (about 15%), the predominance of women in the study group, and the low representativeness of the reference group. A good correlation was obtained between the cancer risk factors determined based on the results of blood and urine tests and its actual occurrence – all cases of cancer detected within 2–3 years after the tests were associated with a high-risk ratio, but no cases of neoplastic disease were found in patients with low or medium risk factor. It can be expected that this correlation will be better if additional patient characteristics are taken into account (pH of the oral cavity, bacterial flora, presence of viruses, etc.) along with a wider range of risk factors. The presented method is universal and may be extended in the future to predict other threats of diseases.

Key words: cancer risk, oral mucosa, blood and urine tests

Address for correspondence: zbigniew.suszynski@tu.koszalin.pl

### 13. Modified minimally invasive surgical technique (m-mist) in regenerative periodontal therapy: A 6 months follow-up case report

#### Muammer Gozlu, Halis Ozdemir, Mustafa Inan

#### Private Practice Dentestetik Dental Center, Konya, Turkey

There are recent advances in periodontal surgical techniques, the focus of surgical access has shifted to the regeneration of lost tissues. With the popularity of minimally invasive surgery (MIS) in periodontics, numerous publications have evaluated the benefits of MIS with or without various regenerative biomaterials in the treatment of periodontal intra-bony defects. To achieve goals of the periodontal surgical therapy, MIS can be considered as a potential future for the treatment of periodontal disease. Cortellini and Tonetti proposed the minimally invasive surgical technique (MIST) in 2007. Later, they introduced the concept of providing the space for the regeneration with the modified MIST (M-MIST). The M-MIST surgical approach consists of a tiny interdental access in which only buccal intrasulcular incisions are performed and connected with a buccal horizontal incision of the papilla performed as close as possible to the papilla tip.

A 52-year-old female patient, non-smoker, systematically healthy, was admitted to our clinic with complaining pain and swelling. The clinical parameters were measured: pocket depth, recession, and clinical attachment level. The patient was surgically accessed with the M-MIST. The M-MIST consisted of a buccal incision of the defect-associated papilla, according to the principles of the papilla preservation techniques. Only a buccal flap was

raised while the interdental papilla was left in site. The granulation tissue filling the defect was removed, leaving the interdental and palatal tissues untouched. Root and defect debridement were performed using Gracey curettes. Then, we applied the regenerative material with collagen membrane and with xenograft material. The primary closure of the flaps was attained with a single internal mattress suture.

Early wound healing was uneventful: primary wound closure was attained and maintained in this defect. Post-operative evaluations revealed significant reduction in pocket probing depth (PPD). Clinical attachment level (CAL) gain for #35 was 6 mm at the end of 6 months. The results of this case study indicated that treatment with a combination of the guided tissue regeneration and demineralized bone graft led to a significantly favorable clinical improvement in periodontal intrabony defect 6 months after the surgery.

**Key words:** minimally invasive surgery, intrabony defect, periodontal regeneration

Address for correspondence: mgozlu@gmail.com

### 14. Gingival recession coverage and biotype augmentation prior to orthodontic treatment in a patient with class III open-bite malocclusion

#### Bartłomiej Górski, Tomasz Kaczyński

## Department of Periodontology and Oral Diseases, Medical University of Warsaw, Poland

A 28-year old woman with class III open-bite malocclusion and multiple gingival recessions was referred for periodontal assessment prior to orthodontic and orthognathic treatment. Clinical periodontal examination revealed multiple gingival recessions (Miller's class I, II and III) due to brushing trauma and thin gingival biotype in both arches. Probing pocket depths were not exceeding 3 mm. Recession coverage and biotype augmentation procedures were planned to prevent new recession formation and the aggravation of the present recession. Firstly, gingival augmentation was performed with tunneling technique and 2 connective tissue grafts (CTG) in teeth 33–43. After 3 months, recession coverage with tunneling technique and CTG was performed in teeth 36–33 and 43–46. Following next 3 months, recession coverage was performed in teeth 15-12 and 22-24 with coronally advanced flap technique and CTG. After 8 months of healing time, clinical examination revealed thick gingival biotype in augmented areas, as well as significant improvements in roots coverage. As the periodontal conditions were more favorable for orthodontic teeth movement, orthodontic treatment was initiated.

Presented gingival augmentation and recession coverage procedures with CTG were effective in a patient with severe class III open-bite malocclusion.

Key words: gingival recession, malocclusion, surgical treatment

Address for correspondence: t.kaczynski@me.com

### 15. Tunneling method as gingival recession treatment option: Case report

Andrzej Żegleń, Anna Jakubik, Jolanta Pytko-Polończyk

## Department of Integrated Dentistry, Jagiellonian University Medical College, Kraków, Poland

Gingival recession is becoming a more common clinical problem. It is the apical displacement of gingival margin relative to the cementoenamel junction (CEJ), leading to the exposure of roots' surface. It may cause tooth sensitivity, root caries and aesthetic disorders, especially in the anterior region. A growing number of patients visit dental practices to treat gingival recession. Achieving the expected treatment effect depends, among others, on the surgical method. One of established treatments of gingival recession is the coronally advanced flap (CAF) using the tunneling method, optionally with a connective tissue graft (CTG). Presented case illustrates the course of recession treatment using the coronally advanced flap, locally supported with a connective tissue graft.

A 58-year-old patient with multiple recessions, complicated by cervical lesions covered with composite fillings, was referred to periodontal practice. The patient reported significant aesthetic discomfort and sensitivity of upper front teeth. Teeth 15–23 had undergone a recession coverage treatment with tunneling method. Check-ups performed 6, 12 and 30 months after the surgery showed a stable clinical condition and satisfactory coverage degree, with an aesthetic effect fully satisfactory to the patient. The results indicated that CAF and CAF + CTG is the method of choice for multiple recessions treatment. It is essential to avoid iatrogenic complications by careful diagnostics and treatment planning. Before performing any surgical procedure it is important to properly prepare a patient by providing preventive and conservative treatment, as well as to analyze possible surgical techniques so that the treatment effect is predictable, satisfactory and stable.

Key words: gingival recession, surgical treatment, tunneling technique

Address for correspondence: amjakubik@gmail.com

### 16. The periodontal approach in the management of labially impacted or erupted canines in maxilla: Case reports

Aneta Furtak<sup>1</sup>, Wojciech Bednarz<sup>1,2</sup>

<sup>1</sup>Specialist Outpatient Medical Clinic MEDIDENT, Gorlice, Poland <sup>2</sup>Department of Periodontology, Wroclaw Medical University, Poland

Maxillary canines are second most frequently occurring impacted teeth after third molars. The population prevalence of permanent canine impaction or ectopic eruption is estimated at 1-2%. Untreated maxillary canines may cause malocclusion, resorption of adjacent teeth, cystic lesions or infections.

Decision of choosing proper surgical technique in both clinical situations is crucial to achieve a good esthetic healing outcome. This case report presents 2 different periodontal techniques of managing the soft tissue: uncovering the impacted maxillary canine with apically positioned flap (APF), and creating the keratinized tissue in the ectopic erupted maxillary canine with laterally positioned flap (LPF).

Two different techniques of the soft tissue management in the labially impacted and erupted canines in maxilla are presented. First, a 16-year-old female was referred to the periodontist before the orthodontic treatment. The deciduous tooth (63) was present, left upper canine was labially impacted within the keratinized gingiva. After the extraction of the tooth 63, the apically positioned flap periodontal procedure was performed to preserve the keratinized tissue width and the tooth 23 was orthodontically extruded. In the second case, a 10-year-old female was referred to the periodontist during the orthodontic treatment. Tooth 23 erupted ectopically within the lining mucosa, the laterally positioned flap procedure was performed to create keratinized tissue and the tooth was positioned in the arch. Final results demonstrated that treatment objectives were achieved. The treatment plan for the patients with the impacted and ectopic erupted canines should be performed interdisciplinary by the orthodontist and periodontist. The position of the canine in the alveolar ridge of the maxilla, location relative to the mucogingival line and the presence of the keratinized tissue are the main factors in choosing the proper surgical technique.

Presented periodontal approach may help achieving harmonious gingival contour after the orthodontic treatment in the esthetic zone.

Key words: impacted canines, ectopic eruption, gingival surgery, keratinized gingiva

Address for correspondence: anetkafurtak@gmail.com

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