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– Professor Danuta Waszkiel – Editorial Board member in 2009–2019;

– Professor Wojciech Blonski – Editorial Board member in 2014–2019.

New Members of the Editorial Board

Andreas Wielgosz

Andreas Wielgosz graduated from the University of Toronto, Canada, obtaining a MSc degree in physiology. Then, he obtained the degree of MD at the McMaster University in Hamilton, Canada, and defended his PhD thesis in the field of public health (epidemiology) at the University of North Carolina in USA. He specialized in internal medicine at the University of Ottawa, Canada, and in the Centre of Postgraduate Medical Education in Warsaw, Poland, as well as in cardiology – also at the University of Ottawa, where at present he holds the position of professor in cardiology. He received training in cardiology also within the Robert Wood Johnson Clinical Scholars fellowship at the University of North Carolina, USA, and Heart and Stroke Foundation of Canada research fellowship. In 1985–2005, he was Head of the Department of Cardiology at the Ottawa Hospital, General and Riverside Campuses. For 5 years (1994–1999), he held the position of Director of WHO Collaborating Center for Cardivascular Diseases (CVD) Surveillance. Since 1985, he has been working as Consultant to Transport Canada on Aviation Cardiology. In the years 1998–2004, he worked for the official journal of the World Heart Federation – *CVD Prevention and Control* (currently *Global Heart*) as Editor-in-Chief. In the years 2006–2016, he was also involved in the activities of the Public Health Agency of Canada. At present, he is participating as the site principal investigator in the ongoing Prospective Urban Rural Epidemiologic (PURE) study. Andreas Wielgosz has been a member of various foundations and associations – the Heart and Stroke Foundation of Canada, the World Heart Federation and the InterAmerican Heart Foundation (currently as President). He is a Fellow of the Royal College of Physicians of Canada (FRCPC), a Fellow of American College of Cardiology (FACC) and a Honorary International Fellow of the American Heart Association (FAHA).

Mieszko Więckiewicz

Mieszko Więckiewicz graduated in dentistry in 2009 at Wroclaw Medical University, Poland. In the years 2009–2010, he did a postgraduate internship at the Academic Dental Polyclinic in Wroclaw, Poland. For 5 years (2010–2015), he worked as a research and teaching assistant at the Department of Prosthetic Dentistry at Wroclaw Medical University, and then in the years 2015–2017 – as assistant professor. In 2015, he obtained a specialist degree in the field of prosthodontics. During the course of specialist training, in 2013, he defended his doctoral disseration. Then in 2016, he obtained his PhD in medicine. Since 2012, he has been a guest lecturer and researcher at the Dental School, Faculty of Medicine of Dresden University of Technology in Germany. Since 2017, he has been holding the position of Head of Department of Experimental Dentistry at Wroclaw Medical University, and since 2018, he has been Head of the Outpatient Clinic of Temporomandibular Disorders at Wroclaw Medical University. In 2019, he was appointed as university professor. He has continued his professional development through numerous international courses (Austria, Germany, USA), scholarships (Germany, Slovakia), internships (Israel), and fellowships (USA).

He has participated in a lot of conferences, also as a lecturer, both in Poland and abroad. Mieszko Więckiewicz has been awarded many grants and awards, authored lots of publications. He cooperates as a reviewer and Editorial Board member with recognized specialist journals. He is a member of Polish Dental Association, Polish Association of Temporomandibular Disorders, International Association for Dental Research, International Network for Orofacial Pain and Related Disorders Methodology, and European Academy of Orofacial Pain and Dysfunction.

Assessing Knowledge, Attitudes and Practices of dental practitioners regarding the COVID-19 pandemic: A multinational study

Ocena wiedzy, zachowań i działań praktycznych u lekarzy dentystów w związku z pandemią COVID-19 – wielonarodowe badanie ankietowe

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Abstract

Background. Coronavirus Disease 2019 (COVID-19) has been declared a global public health emergency that is affecting people across the globe.

Objectives. The aim of this study was to assess the Knowledge, Attitudes and Practices (KAP) of dental practitioners regarding the Coronavirus Disease 2019 (COVID-2019) pandemic.

Material and methods. An online questionnaire was distributed among dentists across the globe using a combination of convenience and snowball sampling. The questionnaire was divided into 4 sections: the 1st one contained personal information, whereas the 2nd, 3rd and 4th sections assessed knowledge (11 questions), attitudes (6 questions) and practices (7 questions) of the dentists. The data was subjected to the Shapiro–Wilk test, one-way analysis of variance (ANOVA), multivariate linear regression, and Pearson's correlation; 95% confidence interval (CI) was calculated and odds ratio (OR) was obtained. The analysis was done using IBM SPSS for Windows, v. 21.0.

Results. The total number of the responses received (860) was divided with regard to various continents (Asia, Americas – North and South, Europe, Africa, and other – Australia and Antarctica). The largest number of dentists came from the Asian continent (264; 30.7%). Most dentists had a degree of MDS (Master of Dental Science) (301; 35.0%), followed by BDS (Bachelor of Dental Surgery) (282; 32.8%) and DDS (Doctor of Dental Surgery) (226; 26.3%). High/Good knowledge and practice scores were observed among 92.7% and 79.5 % of the dentists, respectively. Good knowledge scores were significantly associated with qualifications ($p = 0.04$) and years of practice ($p = 0.02$); good practice scores were associated with qualifications only ($p = 0.03$).

Conclusions. The dentists were found to have good knowledge and practice scores, which is important to combat COVID-19. They are advised to follow the Centers of Disease Control and Prevention (CDC) and World Health Organization (WHO) guidelines in their clinics, and sensitize their staff so that no stone is left unturned in defeating this pandemic.

Key words: dentists, COVID-19, pandemic

Słowa kluczowe: dentyści, COVID-19, pandemia

Introduction

At the dawn of a new decade, on 30th January, 2019, the World Health Organization (WHO) declared a global public health emergency against the outbreak of coronavirus disease, which is termed as Coronavirus Disease 2019 (COVID-19), and since then has rapidly achieved a pandemic status. This disease with flu-like symptoms was initially observed among people residing in Wuhan, Hubei Province in China.[1]

The causative organism responsible for this outbreak – the Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) – belongs to the family *Coronaviridae* of the order *Nidovirales.* This virus structurally comprises of a large, single, plus-stranded RNA as its genome.[2,3] A total of 4 genera of coronaviruses have been discovered to date and these are: α- CoV, β-CoV, γ-CoV, and δ-CoV.[4] The α-CoV and β-CoV variants are observed to infect mainly the respiratory, gastrointestinal and central nervous system of humans and/or mammals, whereas γ-CoV and δ-CoV have been reported with reference to the infections limited to bird species.[5]

Humans suffering from this disease clinically present with the primary symptoms of fever, cough, myalgia or fatigue, abnormal chest computed tomography (CT) image, and severe respiratory distress, whereas less common symptoms include sputum production, headache, hemoptysis, and diarrhea.[3,6,7] Initially, it was considered to have a zoonotic route of transmission; however, a new person-to-person route of transmission is causing the disease to spread rapidly across different continents and is more likely to affect elderly males.[3,8]

Environmental contamination has been singled out as the primary factor for the nosocomial spread of the newest strains of viruses.[9,10] With regard to SARS-CoV-2, researchers have confirmed its nosocomial transmission, although very little is known about its mode of transmission and the extent of environmental contamination.[11] In a dental setting, as the dentist and their equipment are in close proximity to the patient, the chance of acquiring infection from the micro-droplets of an infected patient is high and there is a risk of cross-transmission, too.

In developing and tourist-friendly countries, there is a boom of dental tourism due to the availability of quality treatment at affordable prices, which attracts patients from all over the world.[12] In the event of an outbreak, the dentist can be the first person to come in contact with an infected person; they can either unknowingly become a carrier and infect others or by following proper guidelines can prevent the possible spread of the disease and save the entire community from its disastrous consequences. Dentists who specialize in the field of public health dentistry are at an increased risk of contacting such an infection due to the nature of their work, which involves promoting oral health among their communities with limited resources.

To combat an outbreak, dentists should be aware of recent developments, especially those related to public health, and by following apt guidelines (i.e., the WHO guidelines at https://www.who.int/emergencies/diseases/novel-coronavirus-2019/technical-guidance) make efforts to prevent the transmission of such diseases. Hence, the present study was undertaken with the aim to assess the Knowledge, Attitudes and Practices (KAP) of dental practitioners regarding the COVID-2019 pandemic.

Material and methods

Prior to the implementation of this KAP study, the questionnaire design, validation, pilot study, and strategies to enroll dentists for maximal global participation were discussed. This study was approved by the Institutional Ethics Committee of the Institute of Dental Sciences (IDS) in Bareilly, India (No. IEC/57/2019). The main instrument to collect data was an online questionnaire using Google forms and it is available at: https://forms.gle/5sgxWngughJg23K46. Upon clicking on the link, the 1st page assured the confidentiality of data, informed the dentists of the study objectives and stated that the study participation was purely voluntary. The dentists' consent to participate in the study (inclusion criteria) was implied when they clicked on the 'next' button to answer the questionnaire, and they had complete freedom either to decline or answer the questionnaire. Only the principal investigator had access to the data and no personal details (e-mail address, phone number, name, etc.) were required. Responses were sought from only those dentists who at least had completed their graduation in dentistry and a submission was considered only when the 'submit' button was clicked at the end of the questionnaire (inclusion criteria). Among total submissions, if a dentist failed to answer ≥1 question, it was excluded from the analysis (Fig. 1).

The study duration was from 25th December, 2019 to 20th February, 2020, and both convenience sampling (researchers themselves contacted dentists to participate in the study) and snowball sampling (the participating dentists were asked to forward the questionnaire to their colleagues) were used so that maximal participation could be ensured. The questionnaire was distributed personally via a quick response (QR) code as well as posted on various social media platforms like Facebook and WhatsApp. The questionnaire was divided into 4 sections and had a total of 24 questions. The 1st section contained personal information (continent of residence, highest qualification, currently practicing as (an academician, clinician, or both), and years of practice), whereas the 2nd, 3rd and 4th sections assessed the knowledge (11 questions), attitudes (6 questions) and practices (7 questions) of the dentists regarding COVID-19. A pilot study was done on 25 dentists to validate the questionnaire and its Cronbach's alpha (α) was found to be 0.79. The pilot study responses and incomplete responses were excluded from the main analysis.

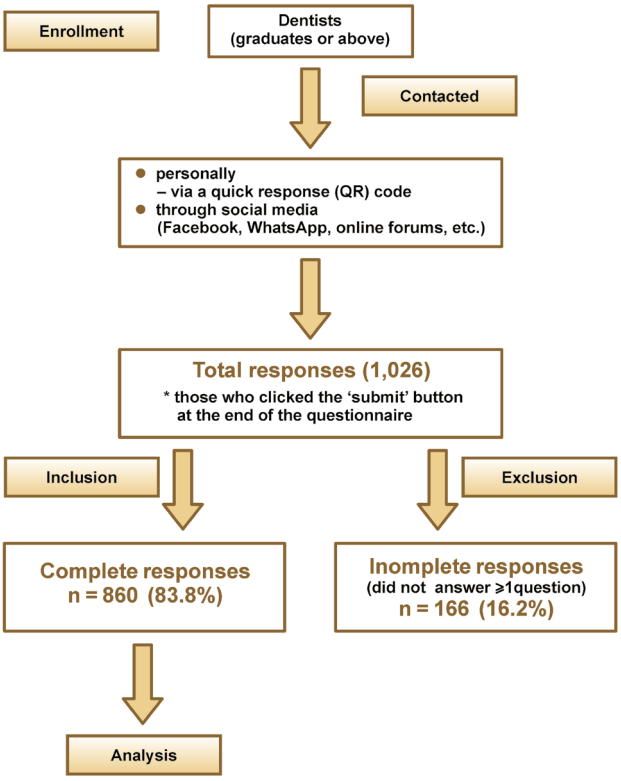

Fig. 1. Study protocol

The data analysis included the Shapiro–Wilk test (to check the data for normality), one-way analysis of variance (ANOVA), multivariate linear regression, and Pearson's correlation coefficient; 95% confidence interval (CI) was calculated and odds ratio (OR) was obtained. The coded data was sent to the statistician so that the confidentially of the data could be maintained. The analysis was done using IBM SPSS for Windows, v. 21.0 (IBM Corp., Armonk, USA).[13]

Results

A total of 1,026 submissions were recorded, among which 860 were complete responses and were included in the analysis (response rate: 83.8%).

Socio-demographic characteristics of the participating dentists

Since the present study adopted a global approach, the total number of the responses received (860) was divided with regard to various continents (Asia, Americas – North and South, Europe, Africa, and other – Australia and Antarctica), with the largest number of dentists coming from the Asian continent (30.7%) and the fewest responses from 'other' continents (5.4%). Most dentists had a degree of MDS (Master of Dental Science) (35.0%), followed by BDS (Bachelor of Dental Surgery) (32.8%) and DDS (Doctor of Dental Surgery) (26.3%). The majority of them were clinicians (65.0%) and an experience period of 2–5 years was mostly reported (34.5%) (Table 1).

Responses to the questionnaire and the source of information regarding COVID-19

The source of information regarding COVID-19 was primarily the Internet (37.7%), followed by social media sites (30.9%), television (20%), newspapers (10%), and other (1.4%) (Table 2).

Knowledge regarding COVID-19

Almost all (99.4%) of the dentists heard about the coronavirus, whereas only 90.9% could name it correctly. A total of 98.4% of the dentists could identify the epicentre; 68.1% and 64.2% of the dentists, respectively, knew about vaccine availability and the method of diagnosing the disease. A total of 95.9% of the dentists believed that COVID-19 was fatal in nature and 99.8% reported that wearing mouth masks could prevent its transmission. Significant differences were observed when the dentists responded to questions regarding the affected system (p = 0.011) and the fatality of COVID-19 (p = 0.023) (Table 2).

Table 1. Sociodemographic characteristics of the participating dentists

Characteristic		n (%)
Continent of residence	Asia	264 (30.7)
	Americas (North and South)	215 (25.0)
	Europe	140 (16.3)
	Africa	194 (22.6)
	other (Australia and Antarctica)	47 (5.4)
Highest qualification	PhD	14 (1.6)
	MDS	301 (35.0)
	DDS	226 (26.3)
	DMD	37 (4.3)
	BDS	282 (32.8)
	other	–
Currently practicing as	an academician	202 (23.5)
	a clinician	559 (65.0)
	both	99 (11.5)
Years of practice	0–2	216 (25.1)
	2–5	297 (34.5)
	5–10	199 (23.1)
	>10	45 (5.3)
	currently not practicing	103 (12.0)

PhD – Doctor of Philosophy; MDS – Master of Dental Science; DDS – Doctor of Dental Surgery; DMD – Doctor of Dental Medicine; BDS – Bachelor of Dental Surgery.

Percentages rounded off to the nearest decimal.

Table 2. Responses to the questionnaire by the participating dentists

Questionnaire item (serial No.)		Overall N = 860	Asia (264)	Americas (215)	Europe (140)	Africa (194)	Other continents (47)	*p*-value
Knowledge	heard about the coronavirus	855 (99.4)	263 (99.6)	215 (100)	140 (100)	190 (97.9)	47 (100)	NS
	could name it correctly	782 (90.9)	232 (87.9)	210 (97.7)	137 (97.9)	163 (84.0)	40 (85.1)	NS
	could correctly identify							
	– place where COVID-19 was first diagnosed	846 (98.4)	260 (98.5)	213 (99.1)	140 (100)	189 (97.4)	44 (93.6)	NS
	– affected system	736 (85.6)	201 (76.1)	205 (95.3)	126 (90.0)	165 (85.0)	39 (83.0)	0.011*
	– transmission route	839 (97.6)	261 (98.9)	211 (98.1)	138 (98.6)	189 (97.4)	40 (85.1)	NS
	– signs and symptoms	850 (98.8)	260 (98.5)	212 (98.6)	139 (99.3)	193 (99.5)	46 (97.9)	NS
	knew about							
	– vaccine availability	586 (68.1)	154 (58.3)	175 (81.4)	101 (72.1)	121 (62.4)	35 (74.5)	NS
	– mode of spreading	829 (96.4)	260 (98.5)	206 (95.8)	136 (97.1)	189 (97.4)	38 (80.9)	NS
	– protection from mouth masks	858 (99.8)	263 (99.6)	215 (100)	140 (100)	194 (100)	46 (97.9)	NS
	– method of diagnosing	552 (64.2)	145 (54.9)	154 (71.6)	126 (90.0)	103 (53.1)	24 (51.1)	NS
	– fatality of the disease	825 (95.9)	257 (97.3)	205 (95.3)	132 (94.3)	191 (98.5)	40 (85.1)	0.023*
Attitudes	possible							
	– to spread awareness regarding COVID-19 by the dentists	860 (100)	264 (100)	215 (100)	140 (100)	194 (100)	47 (100)	NS
	– COVID-19 becoming a pandemic	750 (87.2)	239 (90.5)	211 (98.1)	124 (88.6)	144 (74.2)	32 (68.1)	NS
	– effectiveness of hand hygiene and PPE in preventing infection	860 (100)	264 (100)	215 (100)	140 (100)	194 (100)	47 (100)	NS
	risk							
	– avoiding non-vegetarian food due to COVID-19	310 (36.0)	158 (59.8)	65 (30.2)	29 (20.7)	46 (23.7)	12 (25.5)	0.049*
	– to their health	563 (65.5)	199 (75.4)	101 (47.0)	124 (88.6)	111 (57.2)	28 (59.6)	NS
	– to the patients' health	776 (90.2)	251 (95.1)	189 (87.9)	121 (86.4)	174 (89.7)	41 (87.2)	NS
Practices	– staff sensitized as per the WHO guidelines for the prevention of COVID-19	377 (43.8)	88 (33.3)	126 (58.6)	47 (33.6)	102 (52.6)	14 (29.8)	NS
	– included the travel history while recording the patients' histories	827 (96.2)	261 (99.0)	209 (97.2)	132 (94.3)	183 (94.3)	42 (89.4)	0.025*
	– discussed the risk of COVID-19 with the patients	589 (68.5)	147 (55.7)	212 (98.6)	100 (71.4)	99 (51.0)	31 (66.0)	NS
	– discussed preventive measures against COVID-19 with the patients	679 (79.0)	244 (92.4)	200 (93.0)	83 (59.3)	125 (64.4)	27 (57.4)	NS
	– took action to show the patients the signs and symptoms of COVID-19	275 (32.0)	56 (21.2)	84 (39.1)	26 (18.6)	87 (44.8)	22 (46.8)	NS
	– took preventive measures against COVID-19	853 (99.2)	263 (99.6)	215 (100)	140 (100)	188 (96.9)	47 (100)	NS
	– Has COVID-19 affected your social life?	224 (26.0)	57 (21.6)	16 (7.4)	64 (45.7)	78 (40.2)	9 (19.1)	0.036*
Primary source of information regarding COVID-19	newspapers	86 (10.0)						
	television	172 (20.0)						
	the Internet	325 (37.7)			–			NA
	social media	265 (30.9)						
	other	12 (1.4)						

COVID-19 – Coronavirus Disease 2019; PPE – personal protective equipment; WHO – World Health Organization; NS – nonsignificant; NA – not applicable; * statistically significant ($p < 0.05$).

Data presented as number (percentage); percentages rounded off to the nearest decimal.

Attitudes regarding COVID-19

All dentists (100%) agreed that it was possible for dentists to spread awareness regarding COVID-19, and that hand hygiene and personal protective equipment (PPE) were highly effective in preventing infection. A total of 87.2% of the dentists believed that COVID-19 could achieve a pandemic status; 65.5% responded that it was a risk to their own health, whereas 90.2% believed that it was a risk to their patients' health. Non-vegetarian food was being avoided by 36% of the dentists and the difference was found to be statistically significant ($p = 0.049$) (Table 2).

Practices regarding COVID-19

At the time of responding to the questionnaire, only 43.8% of the dentists had sensitized their staff as per the WHO guidelines for the prevention of COVID-19 in their workplace. Discussion regarding the risk of COVID-19

and preventive measures to be taken among their patients was reported by 68.5% and 79.0% of the dentists, respectively. A total of 32.0% of dentists took some kind of action to show their patients the signs and symptoms of the disease, whereas 99.2% took preventive measures against COVID-19. As many as 96.2% of the dentists admitted to including the travel history while recording the case history of the patient (p = 0.025), whereas 26% of them responded that COVID-19 had an effect on their social life (p = 0.036) (Table 2).

Knowledge and practice scores of the participating dentists

Based on the median of the scores obtained, the cut-off points for the knowledge (maximum score 13) and practice (maximum score 7) scores were 8 and 4 respectively. No dentist achieved a prefect knowledge score (range: 5–12), but perfect scores were observed in the practice section of the questionnaire (range 1–7). High/Good knowledge and practice scores were seen in 92.7% and 79.5% of the participating dentists, respectively (Table 3).

Table 3. Knowledge and practice scores of the participants regarding Coronavirus Disease 2019 (COVID-19)

Characteristic		Value
Knowledge	range of scores achieved (maximum 13)	5–12
	mean ±*SD*	8.1 ±2.5
	median (cut-off point)	8
	high/good knowledge scores n (%)	797 (92.7)
	low/poor knowledge scores n (%)	63 (7.3)
Practice	range of scores achieved (maximum 7)	1–7
	mean ±*SD*	4.1 ±2.5
	median (cut-off point)	4
	high/good practice scores n (%)	684 (79.5)
	low/poor practice scores n (%)	176 (20.5)

SD – standard deviation.

Association between demographic variables and knowledge and practice scores

The multiple linear regression model to analyze the knowledge and practice scores in relation to demographic variables revealed that good knowledge scores were significantly associated with qualifications (p = 0.04) and the years of practice (p = 0.02), whereas good practice scores were associated with qualifications only (p = 0.03) (Table 4).

Relationship between knowledge and practice scores

A positive, linear, great strength of association (r: +0.669) and a significant relationship (p = 0.02) was found between good knowledge and practice scores using Pearson's correlation coefficient (Table 5).

Discussion

The transmission of COVID-19 poses a risk for people who come in close contact with an infected individual, and the risk is greater among those who are in close proximity to or work near the patient, i.e., relatives and healthcare workers. The distance between the working field and the dentist is approx. 35–40 cm, and certain procedures can be very time-consuming, which puts the dentist at a higher risk of contacting COVID-19.[14,15]

Table 4. Association between demographic variables and the participants' knowledge and practice scores using the multivariate linear regression analysis

Predictor		Coefficient	*SD*	*t*	*p*-value
Knowledge	constant	26.42	2.26	40.21	0.00
	qualifications	2.23	3.19	2.60	0.04*
	years of practice	–1.02	0.52	–0.99	0.02*
	location (continent of residence)	2.32	0.43	6.21	0.55
Practice	constant	33.47	1.02	20.06	0.00
	qualifications	1.23	1.14	4.30	0.03*
	years of practice	5.21	2.12	2.11	1.62
	location (continent of residence)	–6.88	0.25	2.66	0.83

* statistically significant ($p < 0.05$).

Table 5. Correlation between knowledge, attitudes and practices using Pearson's correlation test

Relationship between		Pearson's coefficient of correlation	CI	*p*-value
knowledge	practices	+0.669	0.77–26.64	0.02*

CI – confidence interval; * statistically significant ($p < 0.05$).

To assess the preparedness of the healthcare worker to combat any disease outbreak, researchers across the globe try to assess their knowledge of the disease. The present knowledge scores regarding COVID-19 (92.7%) are higher as compared to those presented by Gupta et al. (the Zika virus (ZIKV) pandemic; 38.2% among Indian dentists), Fatiregun et al. (the swine influenza (H1N1) virus; 31% among senior Nigerian healthcare workers), Aung et al. (the Ebola virus; 54.7% nursing students in Myanmar), Shivlingesh et al. (the influenza A (H1N1) outbreak; 52.6% of the Indian population), and Singh et al. (the ZIKV outbreak; 61.7% among the students of a dental institute).[16–20] An important aspect of this study is that responses were collected on a multinational scale, and such high knowledge scores are promising as far as the role of dentists in combating the COVID-19 outbreak is concerned.

In this context, recording properly the travel history of the patient prior to any treatment becomes paramount. In developing countries, purchasing extra PPE (gowns, gloves, etc.) and the cost of the fumigation/sterilizing of the dental clinic can impact the dental clinician financially; hence, incorporating the travel history can help significantly reduce the transmission as well as the burden of the disease. International travelling has sharply increased over the past few years due to declining air fares, easy accessibility, flexible timings, and an increasing number of airports, which in turn is contributing to traveler-associated infections (especially respiratory infections).[21] In the present study, 96.2% of the dentists reported including the travel history while recording the history of the patient and this was important in a timely diagnosis, which could prevent further propagation of infection.

The initial source or information among the dentists was the Internet (37.7%), followed by the social media (30.9%), which is in agreement with the results obtained by Gupta et al., who reported that during the ZIKV pandemic, most of the knowledge gained by the dentists in the Tricity area in India, had its source in the Internet (37.8%).[16] During the development of a new strain of an infectious agent, there might not be enough data available in scholarly journals and/or textbooks, and hence, dentists might access trusted sites like the ones of the Centers of Disease Control and Prevention (CDC), WHO or the websites of health ministries of their respective countries for information. The use of the Internet however, is dependent on various factors like personal preferences, internet availability, type of device, speed, cost, etc. In contrast to this study, Fatiregun et al. reported television as the primary source of data (73.6%) among senior healthcare workers in Nigeria during the influenza A (H1N1) pandemic.[17]

All dentists agreed that they could help spread awareness regarding the disease, and that hand hygiene and PPE were effective in preventing COVID-19. The threat of any epidemic makes all healthcare providers alerted, as they are at a high risk of contracting infection and it is the nature of their work to selflessly treat their patients. As per the GeoSentinel surveillance survey, 11% of the respiratory tract infections were reported among the travelers returning to their country of residence, and PPE can provide protection as well as reduce the risk of any nosocomial infections and cross-transmission in the dental setting.[22,23] Therefore, the risk to the healthcare worker is not only from external travelers, but also from their regular patients who travel on a regular basis, and this threat was acknowledged by the dentists who could correctly (with statistical significance) identify the system affected in COVID-19.

Alarmingly, only 43.8% of the dentists reported that their staff was sensitized as per the WHO guidelines for the prevention of COVID-19. This has to be addressed immediately and care should be taken to sensitize the auxiliary staff as per the current CDC and WHO guidelines to combat the spread of this disease.

A total of 825 (95.9%) dentists responded that COVID-19 was fatal in nature and this could be attributed to the fact that the mortality associated with any new outbreak (although lower in the case of COVID-19) instills a fear of the unknown among people. These figures are on the higher end when compared to the responses of the healthcare providers assessing the fatality of the ZIKV (54.8%) and H1N1 (51%) pandemics.[24,25] Since the responses of the present study were collected when COVID-19 was spreading to other nations, little was known about the characteristics of the virus and there was less information regarding the patients cured of COVID-19, the participating dentists might have assumed that COVID-19 had a high fatality ratio.

The outbreak of COVID-19 has shown a drastic effect on one's social life, since all mass gatherings and social events are being avoided to reduce the transmission rates. Apart from other preventive measures, significant differences are noticed between the continents regarding the number of people avoiding social gatherings.

It was observed that the dentists with higher qualifications (postgraduates) reported better and significant knowledge scores as compared to graduates. Various authors have documented similar findings during the ZIKV and Ebola hemorrhagic fever pandemics.[16,26,27] The possible explanation might be that postgraduate studies involve performing some kind of research (thesis) and updating the dentist's knowledge based on recent guidelines and evidence-based practice. Contrary to our findings, Harapan et al. reported that general practitioners had a higher OR of having a good knowledge as compared to specialist doctors.[28] This can be attributed to global disparities in the dental curriculum and attitudes of the dental faculty authorities toward motivation, encouragement, involvement, and providing assistance to undergraduates in any kind of research projects.

The study is prone to some limitations, one of them being the social desirability bias. In order to eliminate it, we did not ask for any personal information and assured

the participants as to the confidentiality of their data. Secondly, due to the cross-sectional nature of the study and the employed sampling technique, the self-selection bias on the side of the respondents could have occurred. Since this questionnaire was designed to reach the global population of dentists, and due to geographical variations in the way English is spoken and comprehended across the world, inadvertently, there was a slight possibility that the dentists might have experienced the questionnaire bias while answering the questions. However, during the implementation of the pilot study itself, it was ensured that the questions were kept as neutral and simple as possible to avoid such kind of bias.

Conclusions

In the present study, dentists were found to obtain good knowledge and practice scores, which is important to combat COVID-19. Dentists should appropriately use the social media to spread awareness among people, and in their clinical practice, they should screen, isolate and refer the potential cases having the symptoms of COVID-19. They are also advised to follow the CDC and WHO guidelines in their clinics, and sensitize their staff so that no stone is left unturned in defeating this pandemic.

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Effect of smoking on the proliferation capacity and osteogenic potential of human dental pulp stem cells (DPSCs)

Wpływ nikotynizmu na zdolności proliferacyjne oraz potencjał kościotwórczy ludzkich komórek macierzystych miazgi zębowej

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Abstract

Background. Recently, mesenchymal stem cells (MSCs) have proven to have a high potentiality in tissue regeneration. However, genetic diseases or certain environmental risk factors, such as smoking, may compromise the functioning of MSCs, thus leading to a change in the expected clinical outcomes.

Objectives. The aim of this study was to investigate the proliferation capacity and osteogenic potential of dental pulp stem cells (DPSCs) in smokers in comparison with non-smokers.

Material and methods. Mesenchymal stem cells were isolated from the cultured dental pulp tissue from the third molars of 5 smokers and 5 non-smokers. The proliferation capacity of DPSCs derived from both groups was measured using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) proliferation assay. Alizarin red staining and the gene expression analysis of the alkaline phosphatase (*ALP*) and osteocalcin (*OC*) genes were performed in order to assess osteogenic differentiation.

Results. The MTT proliferation assay revealed that the mean absorbance rate of the DPSCs of the non-smokers was significantly higher than that of the group of smokers ($p < 0.0001$). When stained with alizarin red after 21 days of osteogenic induction, fewer calcium deposits were observed among the smokers. Moreover, the *ALP* and *OC* gene expression was significantly higher in the differentiated DPSCs of the non-smokers ($p < 0.05$).

Conclusions. The group of smokers showed a reduced cell viability. The expression of the *ALP* and *OC* genes was lower in the DPSCs of the smokers. Therefore, smoking has a negative impact on the proliferation and regenerative potential of human MSCs.

Key words: smoking, dental pulp, mesenchymal stem cells, osteogenic

Słowa kluczowe: nikotynizm, miazga zębowa, mezenchymalne komórki macierzyste, kościotwórczy

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Introduction

Mesenchymal stem cells (MSCs) have gained worldwide attention in the past few decades due to their great potential in tissue regeneration and repair.[1] Mesenchymal stem cells are capable of synthesizing various cytokines and growth factors that boost local cellular dynamics. Besides their immunomodulatory effect, MSCs release proangiogenic and chemotactic factors, thus playing a major role in wound healing, bone remodeling and overall tissue regeneration.[2,3] However, recent studies have shown that exposure to cigarette smoke significantly inhibits the regenerative capacity of MSCs. Smoke components may reduce the number and quality of stem cells deposited in tissue reservoirs.[4–9]

The unfavorable effect of the components of tobacco smoke on MSCs includes both the direct impact on the cells and their organizational mechanisms, and alterations in the environment of MSCs. Both mechanisms contribute to limiting the regenerative potential of these cells.[10] In addition, the content of nicotine – as one of the main ingredients of cigarette smoke – has been determined to be nearly 87 times higher in the saliva than in the blood plasma.[11] Being subjected to high doses of nicotine may result in harm to the MSC populations in the oral cavity. One such group is dental pulp stem cells (DPSCs), which have similar characteristics to bone marrow mesenchymal stem cells (BMMSCs).[12] It has been established that DPSCs are characterized by being highly proliferative and by their multipotent differentiation capacity. Dental pulp stem cells can differentiate in vitro into odontoblasts, osteoblasts and chondrocytes to produce dentin, bone and cartilage tissues, respectively, for the repair process.[13–17]

Although cigarette smoking inhibits stem cell recruitment to tissues, thus affecting regeneration,[18,19] studies investigating its effect on stem cell proliferation and potentiality are limited. The present study aimed at investigating the deleterious effect of smoking on the proliferation capacity and osteogenic potential of human DPSCs in vitro.

Material and methods

Ethical statement

The study was conducted at the Department of Biochemistry and Molecular Biology, Faculty of Medicine of Cairo University in Egypt. All of the experimental protocols were performed in accordance with the guidelines of the Ethics Committee of the Faculty of Dentistry of Cairo University (approval No. 9/3/16). All eligible patients signed their informed, written consent.

Tooth collection and the culturing of dental pulp stem cells

The MSCs used in this study were derived from the dental pulp (DPSCs) of normal, sound, impacted third molars taken from 5 cigarette smokers and 5 adult, non-smoking donors, aged 25–35 years. The definition of a smoker was determined according to the National Health Interview Survey (NHIS) standard: an adult who has smoked at least 100 cigarettes.[20] All donors were healthy, free from systemic diseases, non-drinkers, and were not on any medications. They were recruited from the outpatient dental clinic at the Faculty of Dentistry of Cairo University. Informed consent was obtained from each donor before the teeth were extracted. Following extraction, each tooth was cut at the cementoenamel junction to reveal the pulp chamber and to obtain the dental pulp tissue.

Dental pulp stem cells from both groups were isolated by the enzymatic dissociation method. The excised tissues were minced under sterile conditions into small pieces, to which a digesting solution of 3 mg/mL of collagenase type II was added (Worthington Biochemical Corp., Lakewood, USA) for 2 h at 37°C. Single-cell suspensions were obtained by passing the cell suspension through a 70-micrometer strainer (BD Biosciences, San Jose, USA). The suspension was incubated in the Roswell Park Memorial Institute (RPMI) medium (Thermo Fisher Scientific, Waltham, USA), which was supplemented with 10% fetal bovine serum (FBS) (Thermo Fisher Scientific), antibiotics (100 units/mL of penicillin G and 100 µg/mL of streptomycin) and an antimycotic agent (Fungizone® – 0.25 µg/mL) (Thermo Fisher Scientific) in a humid atmosphere with 5% CO_2. The culture medium was changed every 3 days until confluence was achieved. The cells were successfully passaged up to the 3rd passage (P3); these were the cells used in the subsequent experiments. The culture medium was replaced every 3 days over a 21-day period.

Flow cytometric analysis

The isolated stem cells were characterized by analyzing the cell surface antigen expression. Adherent cells from dental pulp at P3 were detached using 0.05% trypsin and adjusted to 1×10^5 cells/mL. Then, 1×10^5 cells were incubated with 10 µL of the monoclonal antibodies against CD29, CD34, CD45, CD90, and CD105 (Beckman Coulter, Inc., Miami, USA) for 30 min at 4°C in the dark. Isotopes served as a negative control. The cell analysis was performed using the Cytomics™ FC 500 flow cytometer (Beckman Coulter, Inc.) and the data was analyzed using the CXP software, v. 2.2 (Beckman Coulter, Inc.).

MTT cell proliferation assay

Cells at P3 were seeded in polylysine-coated 96-well plates at 1 × 10^3 cells/well (SPL Life Sciences Co., Ltd., Pyeongtaek, South Korea). The cells were incubated with the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) reagent (Thermo Fisher Scientific), 50 mg/vial. The proliferation was then assessed using the absorbance quantitative enzyme-linked immunosorbent assay (ELISA) at a wavelength of 450 nm.

Osteogenic differentiation

Cells from both groups at P3 were plated into each well of a flat-bottom 24-well plate (SPL Life Sciences Co., Ltd.) at a density of 2 × 10^3/cm^2 with an osteogenic induction medium – human StemXVivo® Osteogenic/Adipogenic Base Media (catalog No. CCM007) and human StemXVivo® Osteogenic Supplement (catalog No. CCM008) (R&D Systems, Minneapolis, USA) for 21 days. The medium was changed twice a week. The cells were observed regularly for morphological changes using an inverted-light microscope (Olympus America, Inc., Center Valley, USA), with a digital camera (Nikon, Tokyo, Japan) for capturing images. Dental pulp stem cells cultured in the basic medium were used as controls.

Alizarin red staining

After 21 days of osteogenic induction, the differentiated DPSCs from both groups were rinsed twice with phosphate-buffered saline (PBS) and fixed in 10% buffered formalin for 10 min at room temperature. The fixative was carefully removed and the cells were gently rinsed 3 times with distilled water, and then stained with 1% alizarin red solution (birefringent end product) (Sigma-Aldrich Co., St. Louis, USA) for 20 min at room temperature. The alizarin red solution was removed and the plates were washed 4 times with 1 mL of distilled water. The stained mineralized nodules were monitored using an inverted-light microscope and digital micrographs were taken.

Real-time quantitative polymerase chain reaction

On day 21 of osteogenic induction, total RNA was isolated from all of the cultured DPSCs of the smoker and non-smoker groups using a cell extraction kit (Qiagen Sciences, Inc., Germantown, USA), and was analyzed for quantity and quality with a Beckman dual spectrophotometer (Beckman Coulter, Inc.). The synthesis of cDNA was performed using 1 µg of the total RNA and a high-capacity cDNA reverse transcription kit (catalog No. K1621; Fermentas, Waltham, USA). The cDNA was subsequently amplified with the SYBR™ Green I PCR Master Mix (Fermentas) using the StepOne™ instrument (Applied Biosystems, Foster City, USA). The primers used for real-time polymerase chain reaction (PCR) were as follows: glyceraldehyde 3-phosphate dehydrogenase (*GAPDH*), forward: 5'-AGGTCGGTGTGAACGGATTTG-3' and reverse: 5'-TGTAGACCATGTAGTTGAGGTCA-3'; alkaline phosphatase (*ALP*), forward: 5'-ACGTGGCTAAGAATGTCATC-3' and reverse: 5'-CTGGTAGGCGATGTCCTTA-3'; osteocalcin (*OC*), forward: 5'-CAAAGGTGCAGCCTTTGTGTC-3' and reverse: 5'-TCACAGTCCGGATTGAGCTCA-3'. The expression levels in the DPSCs of the smoker group were compared to those in the DPSCs of the non-smokers and the controls after normalization to *GAPDH*.

Statistical analysis

The data was analyzed using IBM SPSS Statistics for Windows, v. 21 (IBM Corp., Armonk, USA). Numerical data is presented as means ± standard deviation (*SD*). The independent *t*-test was used to compare the means between the groups of smokers and non-smokers. If any significance was detected, Tukey's post hoc test was used to determine which group was responsible for the significance. Significance was defined as a p-value of <0.05.

Results

Morphological assessment, flow cytometric analysis and proliferation activity assessment

Pulpal stem cells were successfully isolated from both groups, adhered to plastic and proliferated, reaching 70% confluence by day 14. By day 21, all cells acquired a uniform, spindle-shaped morphology, whereby the cells in the non-smoker group appeared to be more numerous (Fig. 1).

The flow cytometric analysis showed that the DPSCs expressed stem cell surface markers CD29, CD90 and CD105 in over 90%. Contrarily, the hematopoietic cell markers CD34 and CD45 were expressed in a very small percentage (<5%) (Fig. 2).

The MTT proliferation assay showed that the mean absorbance rate of the DPSCs of the non-smoker group (4.242 ±0.748; n = 5) was significantly higher than in the case of the smoker group (1.992 ±0.493; n = 5) at $p < 0.0001$ (Fig. 3).

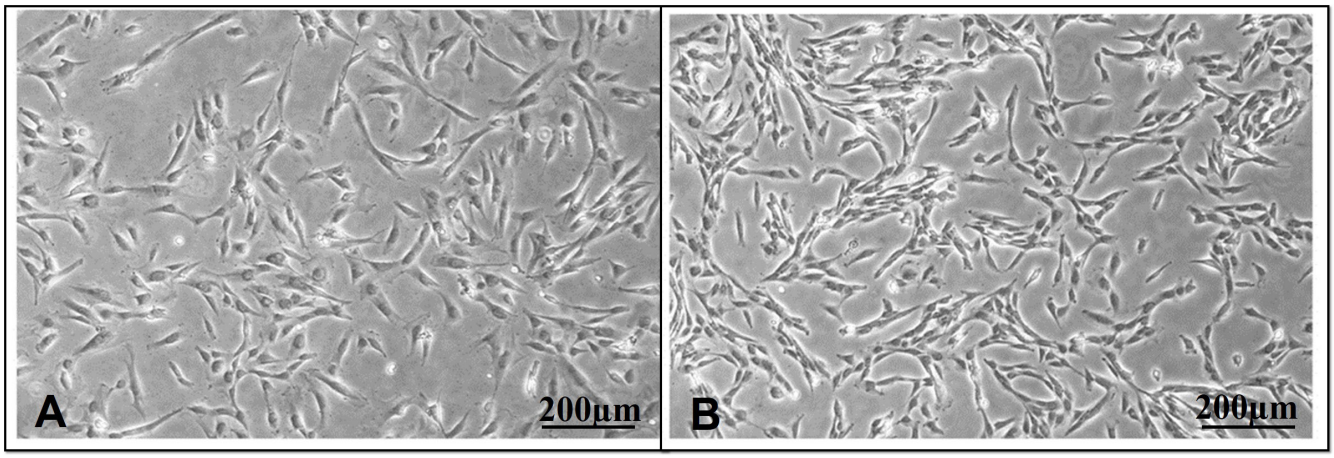

Fig. 1. Morphology of the dental pulp stem cells (DPSCs) from the smoker (A) and non-smoker groups (B) on day 21

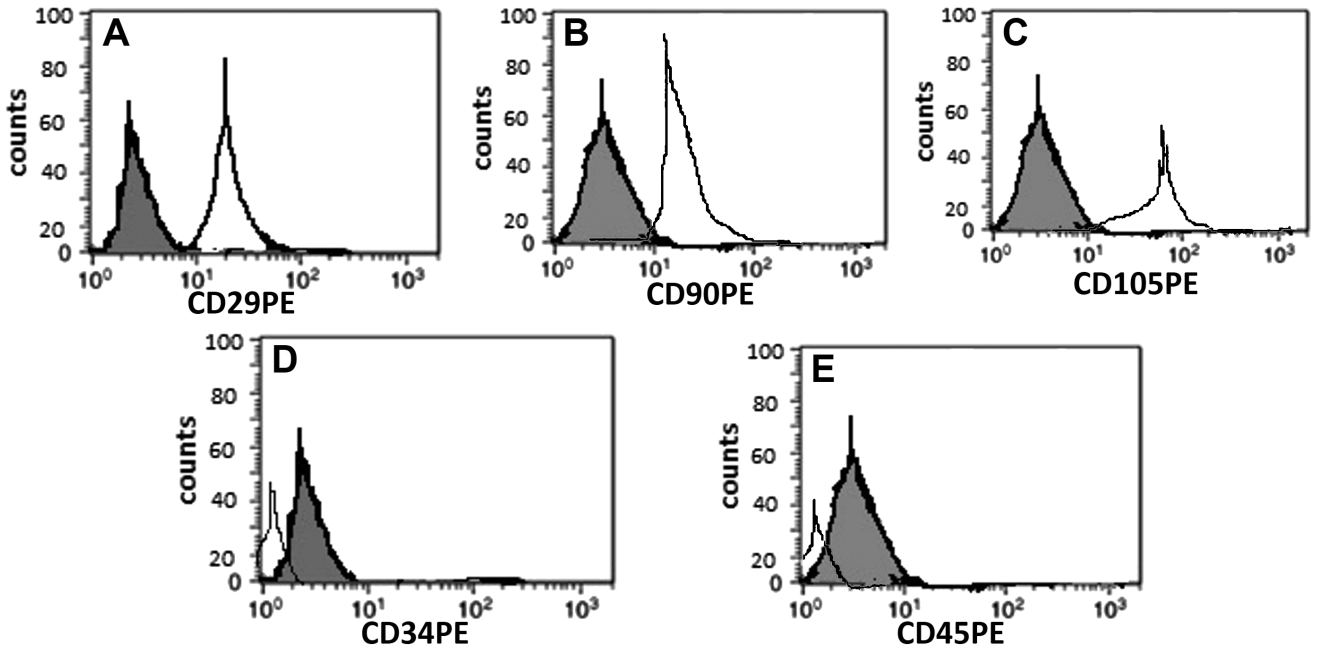

Fig. 2. Representative histograms of the flow cytometric analysis of DPSCs, showing the expression of surface markers: CD29 (A), CD90 (B), CD105 (C), CD34 (D), and CD45 (E)

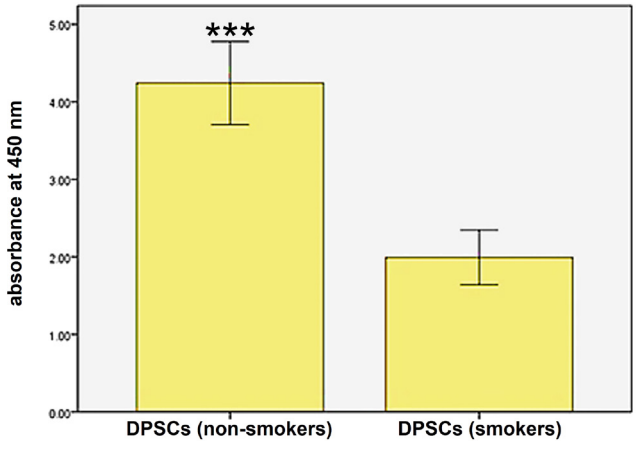

Fig. 3. MTT proliferation assay

MTT – 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide. A significant decrease in proliferation was detected in the DPSCs of smokers as compared to non-smokers. Data expressed as mean ± standard deviation (*SD*); *** $p < 0.0001$.

Osteogenic differentiation assessment

When stained with alizarin red after 21 days of osteogenic induction, a reduced calcium deposition was observed in the DPSCs of the smokers, whereas none was detected in the control cultures (Fig. 4).

When the expression of the *ALP* and *OC* genes was compared, it turned out that the expression of both genes was significantly higher in the differentiated DPSCs of the non-smoker group (*ALP*: 0.854 ±0.439; *OC*: 1.938 ±0.818; n = 5) than in those of the smoker group (*ALP*: 0.480 ±0.211; *OC*: 1.017 ±0.597; n = 5) at $p < 0.05$, whereas the cells in the control cultures did not express the genes at all (Fig. 5,6).

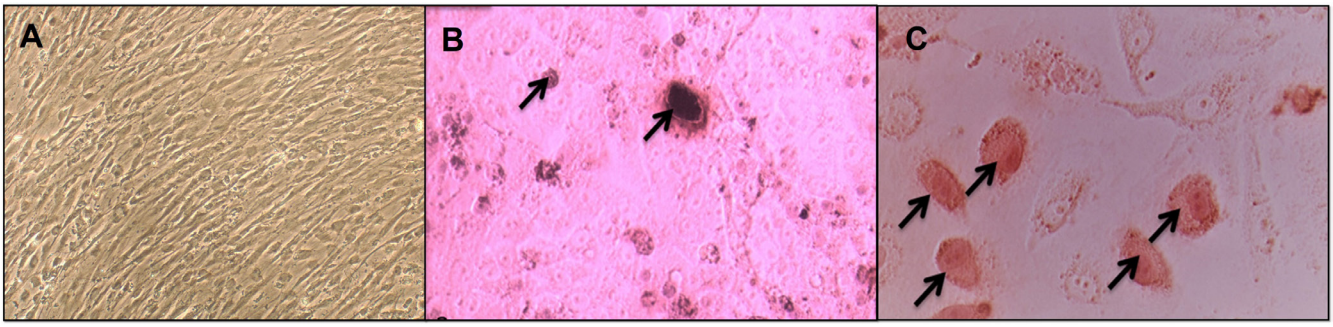

Fig. 4. Alizarin red staining showing calcified areas on day 21 of osteogenic induction, which were more numerous in the non-smoker group as compared to other groups

A – controls; B – DPSCs (smokers); C – DPSCs (non-smokers).

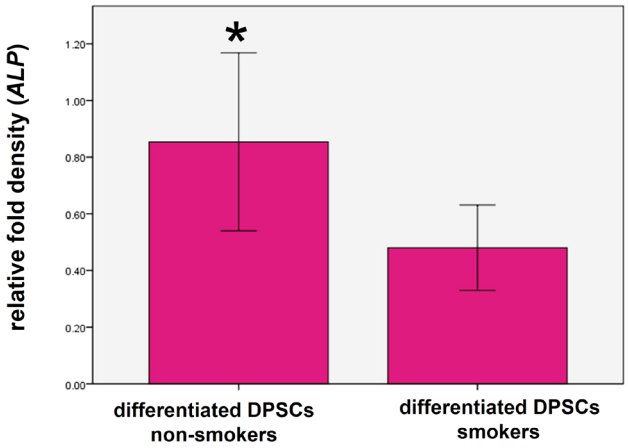

Fig. 5. Quantitative real-time polymerase chain reaction (RT-PCR) for the mRNA levels of the *ALP* gene, showing a significantly higher expression of the gene in the DPSCs of the non-smokers

Data expressed as mean ±*SD*; * $p < 0.05$.

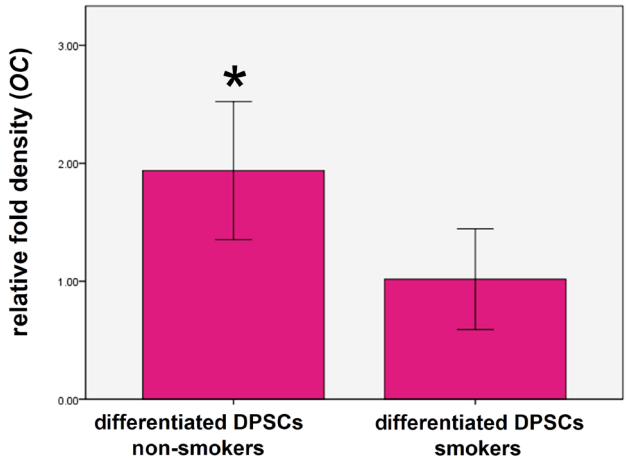

Fig. 6. Quantitative RT-PCR for the mRNA levels of the *OC* gene, showing a significantly higher expression of the gene in the DPSCs of the non-smokers

Data expressed as mean ±*SD*; * $p < 0.05$.

Discussion

Mesenchymal stem cells are frequently considered for various applications in regenerative medicine, being capable of self-renewal in addition to their potentiality to form different cell types.[12] Stem cells are routinely screened prior to transplantation in order to exclude any infectious disease or genetic disturbance that might interfere with the functioning of MSCs. However, the existence of environmental predisposing conditions, such as cigarette smoking, can be missed, and this may affect the ability of MSCs to differentiate and may make the cells unsuitable for transplantation.[11]

Consequently, we investigated the proliferation capacity and osteogenic potential of DPSCs in smokers vs non-smokers. In the present study, MSCs were isolated and expanded from the dental pulp tissue of 10 patients comprising the smoker group (n = 5) and the non-smoker group (n = 5).

In addition to the excellent proliferation ability and mineralization potential of DPSCs, they can be obtained easily. Thus, numerous scientists have studied the isolation and osteogenic potential of DPSCs, but nobody has studied the impact of smoking on their function.[21,22]

In this study, the cell proliferation analysis showed that the mean absorbance rate of the DPSCs in the non-smoker group was significantly higher than that in the smoker group ($p < 0.0001$). This finding is in agreement with Ng et al., who found that the proliferation rates of periodontal ligament stem cells (PDLSCs) isolated from smokers showed a 2.53-fold decrease compared with the cells derived from non-smokers.[9] A decreased proliferation was still observed even after sub-culturing the cells 3–5 times, suggesting that exposure to nicotine may have a prolonged or even permanent effect on the cells.[9]

It is well-known that cigarette smoking delays healing, which can be attributed to disturbance in the regenerative ability of MSCs. Previous reports have studied the effect of nicotine exposure on non-smokers' dental stem cells.[7,8,23] However, few studies have investigated the regenerative potential of stem cells in direct relation to cigarette smoking.[9]

In this study, we examined 2 major important processes that regulate the regenerative potential of stem cells: cell proliferation and differentiation.[24]

The cigarette smoke extract was found to inhibit osteoprogenitor cell proliferation.[5] In addition, nicotine also hinders the proliferation of PDLSCs.[8] Our results

further proved that the proliferation rate and/or cell viability of DPSCs from smokers is lower than in the case of non-smokers.

Regarding cell differentiation, several studies have highlighted the capacity of DPSCs to differentiate into functional osteoblasts in vitro,[25,26] but no one has examined the damaging effect of smoking. In the present study, we assessed the expression of both *ALP* as an early bone marker and *OC* as a late bone marker. In addition, we used alizarin red staining after 21 days of osteogenic induction to detect any calcified deposits.

The expression of both the *ALP* and *OC* genes was significantly higher in the differentiated DPSCs of the non-smoker group than in the smoker group ($p < 0.05$). Alizarin red staining showed a reduced calcium deposition in the smokers' DPSCs. Our results are in agreement with those of Ng et al., who found that the PDLSCs of smokers exhibited an overall reduction in calcium deposition and in the production of ALP compared with non-smokers after 14 days of osteogenic differentiation in vitro.[9] Moreover, Zhou et al. reported that nicotine affected the osteogenic differentiation of MSCs in vitro, and found that exposure to 1 mM of nicotine significantly decreased the inherent *RUNX2*, *COL1A1*, *COL1A2*, *ALP*, and *OC* gene expression in both human BMMSCs (hBMMSCs) and human PDLSCs (hPDLSCs) ($p < 0.05$) after just 3 days of exposure.[7]

Conclusions

This study showed that the proliferation and osteogenic differentiation abilities of human DPSCs from smokers were altered. Thus, smoking could have a negative impact on the proliferation and regenerative potential of human MSCs.

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Salivary profile and dental status of patients with multiple sclerosis

Profil ślinowy i stan uzębienia pacjentów ze stwardnieniem rozsianym

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Abstract

Background. Multiple sclerosis (MS) is a chronic autoimmune disease of the central nervous system. The MS patients may display biochemical changes in their cerebrospinal fluid, peripheral blood and saliva. Since the salivary profile plays a critical role in maintaining oral health and function, the analysis of saliva in the MS patients would be beneficial to prevent oral diseases, such as dental caries.

Objectives. The aim of this study was to evaluate the dental status and salivary profile of the MS patients.

Material and methods. The study involved 25 MS patients and 25 healthy controls who were examined with regard to the calcium and phosphorus level, pH and flow rate of saliva as well as the decayed, missing and filled teeth (DMFT) index for permanent first molars. Student's *t*-test, the χ^2 test and the Mann–Whitney test were utilized to compare the study groups.

Results. Significantly lower salivary flow rates were observed in the MS patients as compared to the controls. The salivary calcium and phosphorus levels were significantly higher in the case group during the first 6 years of the disease and 6–11 years after the onset of the disease, respectively, in comparison with the controls; however, there was no significant difference between the groups in terms of pH. The DMFT index for permanent first molars was higher in the MS patients than in the healthy controls, but not significantly. The number of carious and missing permanent first molars was significantly higher in the MS patients.

Conclusions. Multiple sclerosis appears to significantly change the salivary profile and dental status of the patients.

Key words: multiple sclerosis, DMFT, salivary profile

Słowa kluczowe: stwardnienie rozsiane, DMFT, profil ślinowy

Introduction

Multiple sclerosis (MS) is a chronic autoimmune inflammatory demyelinating disease which affects the central nervous system. It is prevalent all around the world, with low-rate prevalence in Eastern Asia and sub-Saharan Africa, and higher levels in North America and Europe.[1] The onset is mostly between the age of 20 and 40 years (range: 10–60 years), with a female predilection (a female-to-male ratio of 1.4).[2] The main environmental-related risk factor for MS is, apart from genetics, the Epstein–Barr virus infection, especially if it occurs during adulthood and is symptomatic. Previous studies have shown that smoking as well as the calcium and vitamin D deficiencies can be identified as risk factors for MS, but the role of stress, traumatic events, vaccines, and allergies have not yet been clarified.[1,3] The MS patients may display biochemical abnormalities in their cerebrospinal fluid, peripheral blood and saliva.[3] The physical and chemical properties of saliva play a key role in maintaining the health and functioning of the oral cavity. Recent studies show that there is a meaningful relationship between oral lesions and the salivary electrolytes, flow rate and pH.[3] There is limited information about the salivary profile and orodental status of patients with MS.[4] In addition to the main symptoms of the disease, spasms, fatigue, tremor, depression, and progressive disability impact the individual's ability to maintain oral health, cope with dental treatment and access dental services. Furthermore, many of the medications used to manage the disease and its related problems, such as psychological disorders, reported in about half of all patients, are potentially capable of causing xerostomia and associated oral abnormalities.[5,6]

The main aim of this study was to evaluate the salivary profile and dental status of the MS patients as compared to the healthy controls. In addition, all salivary and dental variables were compared among the MS patients in 3 subgroups according to disease duration.

Material and methods

Patient selection

In this descriptive study, 25 MS patients (the case group) and 25 age- and sex-matched healthy controls (the control group) with a mean age of 35.74 ±9.45 years (range: 12–54 years), and 18 females (72%) and 7 males (28%) in each of the groups were studied with regard to the salivary calcium, phosphorus, pH, and flow rate as well as the decayed, missing and filled teeth (DMFT) index for permanent first molars. The case group was recruited from among patients at the Neurology Department of Sina Hospital in Tehran, Iran, and the control group was selected from the hospital staff or patient companions from July to December, 2017. The exclusion criteria for the case group were pregnancy, alcohol dependency and oral lesions. Since almost all MS patients use supplemental drugs, this item was not considered an exclusion criterion in this group. The same criteria were applied to the control group, but those with a history of medication intake or use of supplements in the previous 6 months were excluded from this group. In addition, tooth brushing at least once a day was an inclusion criterion for both groups. Participants were informed of the aim of the study, and written consent was obtained from all participants.

This article was extracted from one of the author's (H.M.) general dentistry dissertation, registered in 2017. The reference number confirming the consent of the local bioethics commission (School of Dentistry of Shahid Beheshti University of Medical Sciences in Tehran, Iran) is 0310/303.

Salivary collection, flow rate and pH

Unstimulated whole saliva was collected from both groups according to the method described by Navazesh and Kumar.[7] The participants were asked to refrain from drinking, eating, smoking, and tooth brushing for about 2 h before sampling. One minute after rinsing their mouths with tap water, the participants were asked to swallow all their oral fluid, and then expectorate 5 mL of their whole saliva into a plastic tube. The saliva samples were collected between 10 and 12 am. The samples were centrifuged at 3,800 g for 10 min, and then stored at –70°C for later analysis. The salivary flow rate was measured in mL/min and the salivary pH was determined by means of an electronic pH-meter (pH600; Milwaukee Instruments, Szeged, Hungary).

Calcium and phosphorus detection

The salivary calcium and phosphorus levels were determined using commercially available kits (Darman Faraz Kave Co., Isfahan, Iran and Pars Azmoon Co., Karaj, Iran, respectively) for a spectrophotometric assay. Since Khan demonstrated that the DMFT index for permanent first molars can serve as an indicator of dental caries,[8] we used it to evaluate the dental status of both study groups. In this index, D stands for untreated decayed teeth, M indicates missing teeth and F stands for filled teeth; full-coverage crowns and pontics in dental bridges are considered as F and M, respectively.[9] In addition, the members of the case group were divided into 3 subgroups according to their disease duration as follows: <6 years; 6–11 years; and >11 years, and the study variables were compared among them.

Statistical analysis

The data was analyzed using PASW Statistics for Windows, v. 18 (SPSS Inc., Chicago, USA). Student's *t*-test, the χ^2 test and the Mann–Whitney test were utilized to compare the study groups, and p-values <0.05 were considered statistically significant.

Results

In total, 50 individuals were studied with a mean age of 35.74 ±9.45 years (range: 12–54 years). There was no significant difference in the mean age of the case group (35.80 ±9.58 years) and the control group (35.68 ±9.51 years). Out of 25 individuals in each group, 18 (72%) were females and 7 (28%) males. In the case group, the duration of the disease varied from 21 days to 18 years, with a mean duration of 7.72 ±4.48 years. The saliva analysis showed a significantly lower flow rate in the MS patients than in the controls ($p < 0.001$) (Table 1). There was no significant difference between the 2 groups in terms of pH ($p = 0.138$). The salivary calcium and phosphorus levels were significantly higher in the case group than in the controls ($p = 0.011$ and $p = 0.020$, respectively) (Table 1). With regard to the dental status, the DMFT index scores for permanent first molars were higher among the MS patients than in the healthy controls, but this difference was not statistically significant ($p = 0.452$). However, except for the filled teeth, the number of carious and missing permanent first molars was significantly higher in the case group than in the control group ($p = 0.038$ and $p = 0.019$, respectively) (Table 1). As shown in Table 2, the salivary calcium and phosphorus levels were significantly higher in the MS patients than in the healthy controls during the first 6 years of the disease ($p = 0.016$) and 6–11 years after the onset of the disease ($p = 0.019$), respectively. The salivary flow rate was significantly lower in the MS patients than in the controls during the entire course of the disease (Table 2). In addition, the duration of the disease was not associated with any significant differences in terms of dental status parameters ($p > 0.05$) (Table 3).

Discussion

Multiple sclerosis is the most common cause of neurological disability in young people and it is most frequently observed in the 2nd–4th decades of life.[1–3] In the present study, the mean age of the MS patients was 35.80 ±9.58 years, which is consistent with recent reports by Etemadifar et al.[10] According to Hernán et al., a higher risk of MS has been reported among individuals with a history of oc-currence of measles and other common childhood diseases during adolescence.[11] Multiple sclerosis can be found at any age, but it is referred to as 'early-onset MS' when the first presentation of clinical symptoms occurs before the age of 21, whereas 'late-onset MS' refers to cases in which the first presentation of clinical symptoms occurs after the age of 50 years.[12]

In this study, more than 70% of the MS patients were females, which is comparable to previous studies conducted in Iran and other countries.[1,2,10,13] It is believed that there are genetic mechanisms for sex differences in MS. The epigenetic modifications of DNA may be caused by hormonal or environmental stimuli, which differ between

Table 1. Salivary parameters and the decayed, missing and filled teeth (DMFT) index

Variable	Case group	Control group	*p*-value
Flow rate [mL/min]	0.47 ±0.35	1.17 ±0.74	<0.001
pH	6.66 ±0.60	6.42 ±0.51	0.138
Calcium [mmol/L]	1.46 ±0.90	0.94 ±0.26	0.011
Phosphorus [mmol/L]	3.93 ±1.26	3.01 ±0.55	0.020
DMFT (n)	2.64 ±1.43	2.16 ±1.31	0.452
D (n)	0.56 ±0.76	0.16 ±0.37	0.038
M (n)	1.12 ±1.26	0.20 ±0.50	0.019
F (n)	0.96 ±1.20	1.80 ±1.32	0.147

Data presented as mean ± standard deviation (*SD*).

Table 2. Salivary parameters in the case subgroups with different disease duration and in the controls

Variable		Case subgroups (according to disease duration)			Control group
		<6 years	6–11 years	>11 years	
Flow rate [mL/min]	mean ±*SD*	0.44 ±0.31	0.48 ±0.41	0.48 ±0.38	1.17 ±0.74
	p-value	0.010	0.020	0.010	
pH	mean ±*SD*	6.81 ±0.53	6.43 ±0.77	6.72 ±0.50	6.42 ±0.51
	p-value	0.242	1.000	0.419	
Calcium [mmol/L]	mean ±*SD*	1.73 ±0.85	1.36 ±0.94	1.31 ±0.96	0.94 ±0.26
	p-value	0.016	0.330	0.399	
Phosphorus [mmol/L]	mean ±*SD*	3.85 ±1.25	4.16 ±1.29	3.78 ±1.38	3.01 ±0.55
	p-value	0.110	0.019	0.138	

Table 3. The DMFT index scores in the case subgroups with different disease duration and in the controls

Variable	Group	Mean ±*SD*	Median (P50)	IQR (P25–P75)	*p*-value
DMFT for first molars (n)	control	2.16 ±1.31	2.00	1.00–3.00	–
	<6 years	3.00 ±0.92	3.00	2.00–4.00	0.457
	6–11 years	2.13 ±1.45	2.50	0.50–2.50	
	>11 years	2.44 ±1.59	3.00	1.00–4.00	
D (n)	control	0.16 ±0.37	0.00	0.00–0.00	–
	<6 years	1.00 ±0.92	1.00	0.00–2.00	0.075
	6–11 years	0.25 ±0.46	0.00	0.00–0.75	1.000
	>11 years	0.44 ±0.72	0.00	0.00–1.00	1.000
M (n)	control	0.20 ±0.50	0.00	0.00–0.00	–
	<6 years	1.00 ±1.06	1.00	0.00–2.00	0.294
	6–11 years	1.00 ±1.19	0.50	0.00–2.00	0.321
	>11 years	1.33 ±1.58	1.00	0.00–2.50	0.057
F (n)	control	1.81 ±0.32	2.00	0.50–2.00	–
	<6 years	1.12 ±1.45	0.50	0.00–2.00	0.147
	6–11 years	1.00 ±1.41	0.00	0.00–2.75	
	>11 years	0.77 ±0.83	1.00	0.00–1.50	

IQR – interquartile range.

males and females. Males and females might have different responses to the same environmental stimuli, such as sun exposure and vitamin D supplements. Accordingly, higher levels of vitamin D decrease the incidence of MS more in females than in males. It has also been noted that the X chromosome may play a direct role in autoimmunity; therefore, the presence of 2 X chromosomes increases susceptibility to autoimmune encephalomyelitis. Furthermore, the X chromosome inactivation in females may be skewed, resulting in the over-expression of MS susceptibility genes in women. On the other hand, male sex hormones, such as testosterone, have a protective role in autoimmunity.

Our results show that the MS patients had a significantly lower salivary flow rate than the healthy controls. Similarly, in their 2 recent studies, Cockburn et al. demonstrated that xerostomia was the most common side effect of the medications used to treat MS and related problems, which is followed by dysgeusia, dysphagia, oral ulcers, and sinusitis.[14,15] Further, Sandberg-Wollheim et al. concluded that MS and primary Sjögren's syndrome might coexist in the same individual.[16] In accordance with this hypothesis, Miró et al. and de Seze et al. reported respectively that 3.1% and 16.6% of the MS patients had the clinical evidence of Sjögren's syndrome.[17,18] However, the relationship between MS and primary Sjögren's syndrome is ambiguous, and primary Sjögren's syndrome is not more common among the MS patients than expected in the general population.[16,17]

In the present study, there was no significant difference between the MS patients and the controls in terms of the salivary pH. We did not find a similar study in this regard in the literature to compare our findings with. Normal saliva pH ranges from 6.2 to 7.6, with a mean of 6.7. The resting pH of the mouth does not usually fall below 6.3 and pH in the oral cavity is normally maintained near neutrality (6.7–7.3) by the buffering capacity of saliva.[19]

The salivary calcium and phosphorus levels in our study were found to be significantly higher in the MS patients than in the controls. There is limited information in the literature about the alterations of these factors in the salivary samples of the MS patients in comparison with blood samples. In a case report, Marcus et al. described severe hypercalcemia following vitamin D supplementation in a patient with MS.[20] It is common practice to prescribe high-dose cholecalciferol for the MS patients because of its possible role in immunomodulation and relapse rate reduction. However, cholecalciferol may increase serum calcium, and there seems to be an additive effect in patients who simultaneously use calcium supplements as well. An elevated level of salivary calcium may be related to a greater degree of bone loss and a lower mineral density of bones, as bone loss can cause the release of calcium into blood, and then saliva.[21] Gupta et al. concluded that MS was associated with increased osteoporosis.[22] A reduced mechanical load on the bones (offsetting gravity) and physical inactivity are major factors for osteoporosis in MS. Medications such as glucocorticoids and anticonvulsants may be additional factors leading to a lower bone density in this group of patients. The same findings were also reported by Kampman et al.[23] In addition, chronic secondary hyperparathyroidism in the MS patients stimulates renal calcium reabsorption and bone resorption to elevate the blood calcium levels.[24] According to Kubicka-Baczyk et al., the MS patients had a significantly higher level of serum parathormone than the controls.[25] In their study, daily urine calcium excretion was lower in the patients than in the healthy controls; the serum phosphorus, calcium and alkaline phosphatase levels were higher in the patients than in the controls, but these differences were not statistically significant.[25] In contrast to all the aforementioned studies, Chałas found that the calcium levels in the saliva of the MS patients were significantly lower than in the healthy controls.[3] This difference may be related to different patient characteristics, such as the duration of the disease, the type of medications taken, study sample size, the methods of calcium detection, and different inclusion and exclusion criteria according to the study design. Furthermore, Mohammad Shirazi et al. showed that a daily intake of phosphorus, potassium, manganese, and copper in Iranian MS patients was higher than the standard recommended amounts.[26]

In terms of the dental status, the DMFT index scores did not differ significantly between the patients with MS and the healthy controls. However, the number of decayed and missing teeth was significantly higher in the patients than in the healthy controls. The same findings were also reported by Santa Eulalia-Troisfontaines et al. and Kovac et al.[4,27] As in this study, Kovac et al. found that the number of decayed and missing teeth was higher, but the number of filled teeth was significantly lower in the MS patients than in the controls.[27] In contrast, McGrother et al. found significantly higher DMFT index scores in the MS patients as compared to the healthy controls, and the number of filled and missing teeth was also higher in the patients than in the controls.[28]

In a recent study by Hatipoglu et al., the relationship between the different disability states of the MS patients, as determined by the expanded disability status scale, and dental-periodontal measures was evaluated.[29] Those authors found that patients with greater physical disabilities had higher numbers of missing teeth, higher plaque and gingival indices, and higher periodontal probing depth scores than patients with lesser physical disabilities.[29] According to the aforementioned studies, it seems that patients with MS mostly prefer to extract their decayed teeth rather than to restore them.

Multiple sclerosis is a prime example of systemic diseases requiring multidisciplinary care. The MS care units usually include a variety of medical specialists, such as neurologists, physical therapists, speech therapists, psychologists, social workers, nurses, and – recently – lawyers. However,

there are few reports in the literature on dental care as part of the interdisciplinary therapy for the MS patients. Fragoso et al. found that the MS patients had very poor oral hygiene, and also pointed out that despite recommendations for an interdisciplinary approach for the MS patients, dentists are not usually included in this professional health group.[30] Therefore, oral pathologies in these patients often remain undetected, underassessed or overlooked.

Conclusions

The present study found that due to the quality and quantity of changes in the saliva of patients with MS, the dental caries and loss of permanent first molars were more frequent in the MS patients.

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Color stability of self-adhering composite resins in different solutions

Trwałość barwy samoadhezyjnych żywic kompozytowych w różnych roztworach

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Abstract

Background. The success of composite resin restorations depends to a great extent on their color stability. However, discoloration is still a problem in composite resin restorations.

Objectives. The aim of the study was to evaluate the effect of different staining solutions on the color stability of composite resins.

Material and methods. A total of 96 composite disks, 2 mm in height and 8 mm in diameter, were fabricated of 3 commercially available composite resins. The samples were divided into 4 groups of 8 and were immersed in 4 staining solutions: coffee, tea, soda, and artificial saliva. The color parameters of the samples were measured and recorded before as well as 2, 4 and 8 weeks after immersion by spectrophotometry, using the CIELAB color space. A color change (ΔE) $\leq$3.3 was considered the acceptable threshold for visual perception. The results were analyzed using the one-way analysis of variance (ANOVA) and Tukey's post hoc test ($p < 0.05$).

Results. All the composite resins in the study showed discoloration in all the staining solutions. The ΔE of Vertise™ Flow was the highest in the tea solution. The lowest ΔE occurred in the Filtek™ Z250 composite in artificial saliva.

Conclusions. This in vitro study showed that the color stability of tooth-colored restorations can be influenced by dietary habits.

Key words: composite resins, staining, self-adhesive, VITA Easyshade

Słowa kluczowe: żywice kompozytowe, barwienie, samoadhezyjny, VITA Easyshade

Introduction

Tooth-colored restorative materials are widely used in modern dentistry due to their stability and color match with natural teeth.[1] An increased demand for esthetics has greatly contributed to the popularity of these naturally looking dental materials.[2]

Long-term durability and color stability in the oral environment are common concerns encountered in the use of restorative materials, such as glass ionomers, composite resins and dental ceramics. Glass ionomers are widely used as a restorative material, base and liner. Dental ceramics are also known as esthetic, color-stable and biocompatible dental materials. Composite resins are commonly used esthetic restorative materials in dental clinics.[3] According to the literature, dental ceramics have the highest color stability in the oral environment, followed by composite resins and glass ionomers.[2] Composite resins are more popular among tooth-colored restorative materials due to their optimal strength and esthetics, low cost and strong bonding to the tooth structure.[4]

In order to obtain the best results, composite resin restorations should have the same appearance as natural teeth and must be able to retain this natural appearance over time. Composite resins have been used as direct restorative materials by dentists for over 40 years now, and are considered an acceptable alternative to amalgam and gold restorations. This is due to the development of new composite resins with improved properties.[2,3]

The appearance of a dental restoration should be similar to that of natural teeth. This is directly related to color harmony with the adjacent natural teeth and to the color stability of the material.[4] A thorough understanding of differential colorimetry and color space is required to determine and quantify the color change of restorative materials. Spectrophotometers are currently used to assess the color change (ΔE) of dental materials by measuring the L^*, a^* and b^* color coordinates in the CIELAB color space.[5] The L^* coordinate indicates brightness, the a* coordinate represents the red or green component (positive a^* indicates redness and negative a^* represents greenness), whereas the b^* coordinate is representative of yellowness or blueness (positive b represents yellowness and negative b^* indicates blueness). For instance, after composite polymerization, a change in the b^* coordinate toward the negative indicates that the composite has lost its yellowness after polymerization. The ΔE value is calculated by assessing a change in the 3 color parameters,[6,7] but the L^* coordinate is the most important factor in the esthetic appearance of restorations, as a change in lightness has the greatest impact on color.

However, composite resins applied in the oral cavity often undergo the color change over time.[8] The advantages of composite resins include their color match with the adjacent teeth and their availability in a wide range of shades, their mechanical compatibility and optimal strength against masticatory forces. However, previous studies have shown the discoloration of composite resins in the oral environment.[9–11]

The success of composite resin restorations depends to a great extent on their color stability over time. The discoloration of tooth-colored materials may occur due to internal or external factors. Internal factors include changes in the color of the material itself, i.e., in the resin matrix or at the matrix–filler interface.[12] The color of restorations may change under different physical and chemical conditions, such as changes in temperature and humidity, the staining of resin-based materials due to exposure to solutions such as tea, coffee and other drinks, or color instability may be a result of aging.[13] Lifestyle habits, such as a high consumption of colored beverages or smoking, can result in the color change in composite resin restorations.

Habib et al. reported that chocolate, milk and orange juice are among the children's drinks that have the greatest impact on the color stability of composite resins.[14] Malavasi et al. compared the color stability of 2 self-adhesive composites and a nanofill composite, and showed that the Vertise™ Flow composite had the highest color stability.[15] Reddy et al. found that coffee caused the greatest discoloration in the 3 types of composite resins studied (nanohybrid, microhybrid and hybrid), whereas nanocomposites showed the least color change.[16]

Despite the fact that various studies have evaluated the color stability of various restorative materials, further studies are needed on newly introduced tooth-colored restorative materials. In addition, in order to achieve an acceptable color match, attention has to be paid to the important role of the color stability of restorative materials. Thus, it is imperative to evaluate the color stability of newly released and highly popular products.

Advances in the composition of composite resins have resulted in the recent introduction of self-adhesive composites. Only a few studies are available on various aspects of self-adhesive composites. This study aimed to evaluate the effect of different staining solutions on the color stability of composite resins, including a self-adhesive one.

Material and methods

This was an in vitro experimental study. Table 1 presents the characteristics of the composite resins used in this study: Vertise Flow, Premise™ Flow (both from Kerr Corp., Scafati, Italy) and Filtek™ Z250 (3M, Maplewood, USA). The A2 shade was used, because it is the composite resin shade most widely used in dentistry.

Table 1. Composite resins used in the study and their composition

Composite resin	Composition	Lot No.	Manufacturer
Vertise Flow	Filler: • prepolymerized fillers, containing 0.7 µm barium glass • 1 µm barium glass • 10–40 nm nano-sized colloidal silica • 40 nm nano-sized ytterbium fluoride Resin: • GPDM adhesive monomer; incorporates the Kerr OptiBond™ adhesion technology	3427056	Kerr Corp., Scafati, Italy
Premise Flow	Filler: • 75.5 wt% barium glass, prepolymerized fillers and silica nanoparticles Resin: • Bis-GMA, Bis-EMA, TEGMA, light-cure initiators, and stabilizers	5867548	Kerr Corp., Scafati, Italy
Filtek Z250	Filler: • Zr/Si (60 v%) Resin: • Bis-GMA, Bis-EMA, UDMA	N755574	3M, Maplewood, USA

GPDM – glycerol phosphate dimethacrylate; Bis-GMA – bisphenol A diglycidyl methacrylate; Bis-EMA – ethoxylated bisphenol A diglycidyl methacrylate; TEGMA – triethylene glycol dimethacrylate; UDMA – urethane dimethacrylate.

Sample preparation

Thirty-two samples were prepared of each material using a plexiglass mold, 8 mm in diameter and 2 mm in height. The mold was placed on a glass slide and the composite was applied into the mold. Then, another glass slide was placed over the mold and compressed by placing a 5-kg weight on the top of it for 3 min to ensure the uniformity of the sample thickness and to eliminate voids. The samples were light-cured using the overlapping technique for 40 s on each side (80 s in total). Light-curing was performed using the LED.D light-curing unit (Guilin Woodpecker Medical Instrument Co., Ltd., Guilin, China) at an intensity of 800–850 mW/cm². The light intensity was checked after every 5 samples by the LM-1 radiometer (Guilin Woodpecker Medical Instrument Co., Ltd.). The tip of the light guide was put in contact with the glass slide during the light-curing process. The distance between the light source and the sample was standardized using a standard slide. Each sample was then attached to a thread and immersed in the respective solution. The samples were then placed in distilled water for 24 h to ensure complete polymerization. All samples were polished under a gentle stream of water using 1,000-, 1,500- and 2,000-grit abrasive papers to obtain a homogeneous polished surface and to eliminate possible contamination. This was done to minimize the color change due to the surface roughness of the samples and to ensure that the measured color change was due to the inherent properties of the composites. To ensure the uniformity of the surfaces of the samples at all stages, finishing and polishing were done by the same technician, with uniform pressure and the same number of movements; the final thickness of the disks after the completion of polymerization and polishing was 2 mm. A caliper was used to ensure a uniform thickness throughout the disks.

The color parameters were then evaluated using the VITA Easyshade® spectrophotometer (Vident, Inc., Brea, USA). Using this device, the samples were studied in the CIELAB color space, and the 3 color parameters of L^* (brightness), a^* (red-green) and b^* (yellow-blue) were recorded as the baseline color parameters.

Immersion in staining solutions and the discoloration process

The 32 samples were randomly divided into 4 groups for immersion in 4 solutions (n = 8). Each sample was completely immersed vertically in the respective solution using a piece of thread attached to it. The vertical position minimized the discoloration of the surface of the sample. The samples were immersed in the solutions in such a way so as to prevent them from touching each other or the wall. Thus, the samples were only in contact with the solution.

Preparing staining solutions

Coffee: 5 g of coffee (Farmand Chocolate Co., Tehran, Iran) was added to 250 mL of water, placed on a low-heat flame and removed before boiling (according to the manufacturer's instructions); the solution was then filtered using a paper filter.

Tea: A 2 × 2-inch tea bag (Debsh Tea Co., Tehran, Iran) was immersed in 250 mL of water of a temperature of 100°C for 60 s.

Artificial saliva was prepared in the laboratory of the Dental Research Center of Tehran University of Medical Sciences in Iran, according to the standard procedure.

Soft drink: Coca-Cola® (Khoshgovar Co., Mashhad, Iran).

The samples were immersed in the solutions for 3 h per day for 8 weeks (168 h). To maximize the simulation of clinical conditions, the coffee, tea and artificial saliva

solutions were placed in the Abzar Pezeshki incubator (Kavoosh Mega Co., Tehran, Iran) at 37°C (oral temperature), and the cola was placed in a refrigerator at 4°C for 3 h per day.

The solutions were prepared fresh daily. The samples were gently cleansed each time using water and a soft toothbrush for 30 s to remove any debris due to immersion, and were kept in artificial salvia during the intervals between immersions.

The color parameters of the samples were measured at 2, 4 and 8 weeks after the start of the experiment using the VITA Easyshade spectrophotometer. The total ΔE of the samples was calculated according to the following formula:

$$\Delta E = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2} \quad (1)$$

where:
L^* – lightness;
a^* – red (+)/green (–) color coordinate;
b^* – yellow (+)/blue (–) color coordinate.

Statistical analysis

The data was analyzed using IBM SPSS Statistics for Windows, v. 21 (IBM Corp., Armonk, USA) Descriptive statistics, including mean and standard deviation (*SD*) were reported. The ΔE values following immersion in different beverages were compared using the one-way analysis of variance (ANOVA). Pairwise comparisons were performed using Tukey's test. The level of significance was set at 0.05.

Results

The one-way ANOVA showed that the effects of the type of composite and the type of solution on discoloration were significant at all time points ($p < 0.001$), but the interaction effect of these 2 variables (type of composite and type of solution) on Δa^*, Δb^*, ΔL^*, and ΔE was not significant at 2 weeks ($p = 0.431$), 4 weeks ($p = 0.170$) or 8 weeks ($p = 0.362$).

The results of the study showed that all the composites tested underwent discoloration over time in all solutions, and that staining increased over time.

The lowest mean ΔE at the end of the study (8 weeks) was noted in Premise Flow following immersion in artificial saliva ($\Delta E = 2.25$), whereas the highest mean ΔE was recorded for Vertise Flow following immersion in tea ($\Delta E = 27.24$).

Table 2 presents the mean ΔE of the composite resins after immersion in different solutions for different periods of time.

After 2 weeks of immersion, the two-way ANOVA showed that the interaction effect of the type of composite and the type of solution on color stability was not significant ($p = 0.431$), but the effects of the type of composite and the type of solution were significant ($p < 0.001$). Tukey's test was used for the pairwise comparisons of the composite resins and showed that the difference between Vertise Flow and the 2 other composites was significant ($p < 0.001$). There was no significant difference in the color stability of Filtek Z250 and Premise Flow ($p = 0.949$). The pairwise comparisons of the solutions showed that there was a significant difference between all solutions ($p < 0.001$), except for cola and artificial saliva ($p = 0.799$).

The highest ΔE at the end of the 2nd week was related to the Vertise Flow composite following immersion in coffee and the slightest ΔE was noted in the Filtek Z250 composite following immersion in artificial saliva.

After 4 weeks of immersion, the two-way ANOVA showed that the interaction effect of the type of composite and the type of solution was not significant ($p = 0.170$), but the effects of the type of composite and type of solution on color stability were significant ($p < 0.001$). Tukey's test was used for the pairwise comparisons of the composites and it was found that the difference between Vertise Flow and the other 2 composites was significant ($p < 0.001$). There was no significant difference in this regard between Filtek Z250 and Premise Flow ($p = 0.896$). The results of the pairwise comparisons of the solutions in terms of the discoloration of different composites were similar to those obtained at 2 weeks – there was no significant difference between cola and artificial saliva ($p = 0.991$), but significant differences were noted between the remaining solutions ($p < 0.001$).

At the end of the 4th week, the Vertise Flow composite in tea showed the highest discoloration and, as at the end of the 2nd week, the Filtek Z250 composite in artificial saliva showed the least ΔE.

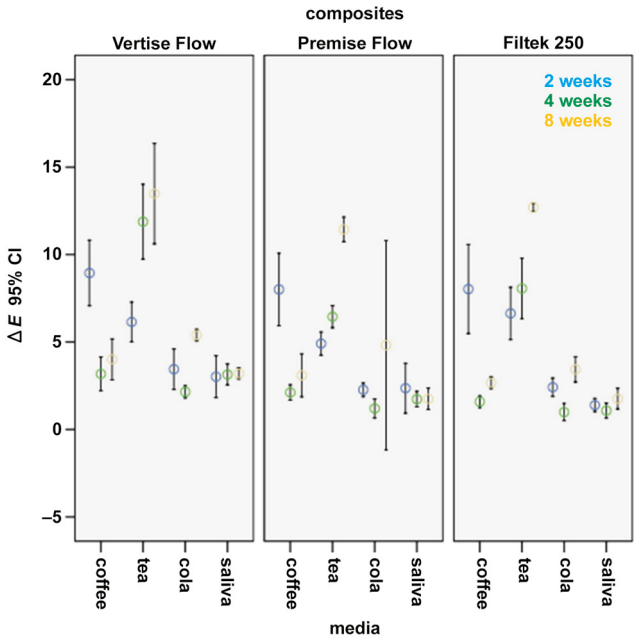

Fig. 1. Composite ΔE following immersion in the solutions for up to 8 weeks
CI – confidence interval.

Table 2. Descriptive information about the color change (Δ*E*) values of the composite resins after immersion in different solutions for different periods of time (n = 8)

Medium	Composite	Δ*E* (compared to baseline)	Minimum	Maximum	Mean	*SD*
Coffee	Vertise Flow	the 2nd week	6.65	13.28	8.9456	2.23728
		the 4th week	6.18	18.00	11.2526	3.66832
		the 8th week	9.53	21.79	14.8293	4.53672
	Premise Flow	the 2nd week	5.60	12.57	8.0065	2.48011
		the 4th week	7.34	15.22	9.8868	2.78348
		the 8th week	8.22	20.97	11.8908	4.51950
	Filtek Z250	the 2nd week	5.04	15.10	8.0277	3.04583
		the 4th week	6.06	16.46	9.4160	3.12565
		the 8th week	7.34	18.57	11.0461	3.37960
Tea	Vertise Flow	the 2nd week	4.64	8.46	6.1452	1.35659
		the 4th week	12.48	19.79	16.3016	2.58172
		the 8th week	21.74	35.42	**27.2485**	5.06403
	Premise Flow	the 2nd week	3.67	6.18	4.9134	0.79045
		the 4th week	9.17	12.62	11.1987	1.12678
		the 8th week	19.92	23.85	22.2165	1.15585
	Filtek Z250	the 2nd week	4.70	9.91	6.6380	1.79172
		the 4th week	8.87	20.06	13.9589	3.23060
		the 8th week	21.46	31.93	26.2044	3.00274
Coca-Cola	Vertise Flow	the 2nd week	0.90	5.39	3.4478	1.38455
		the 4th week	1.56	7.60	4.5157	2.00410
		the 8th week	6.00	11.96	9.0661	2.23966
	Premise Flow	the 2nd week	1.67	2.87	2.2686	0.46026
		the 4th week	1.37	3.20	2.4581	0.57028
		the 8th week	2.14	24.91	5.5623	7.83016
	Filtek Z250	the 2nd week	1.60	3.36	2.4171	0.63374
		the 4th week	1.45	4.38	2.5928	0.82754
		the 8th week	4.10	7.66	5.1984	1.19792
Artificial saliva	Vertise Flow	the 2nd week	0.52	4.50	3.0214	1.42778
		the 4th week	3.14	7.22	5.8412	1.46400
		the 8th week	6.60	10.78	8.6617	1.34524
	Premise Flow	the 2nd week	0.74	6.00	2.3611	1.70622
		the 4th week	1.24	4.73	2.3617	1.18938
		the 8th week	1.34	2.92	**2.2583**	0.56044
	Filtek Z250	the 2nd week	0.82	2.01	1.3959	0.45073
		the 4th week	0.63	3.09	2.1273	0.88576
		the 8th week	1.02	4.40	2.8263	1.01785

SD – standard deviation; values in bold indicate the minimum and maximum mean Δ*E* in all types of dental composite resins.

After 8 weeks of immersion, the two-way ANOVA revealed that the interaction effect of the type of composite and the type of solution on discoloration was not significant ($p = 0.362$), but the effects of the composite type and the type of solution on the discoloration of the composite resins were significant ($p < 0.001$). Tukey's post hoc test was applied for the pairwise comparisons of the composites and it was found that the difference between Vertise Flow and the other 2 composites was significant ($p < 0.001$), whereas there was no significant difference between Filtek Z250 and Premise Flow ($p = 0.926$). The results of the pairwise comparisons of the solutions in terms of the discoloration of different composites were similar to those obtained at 2 weeks. There was no significant difference between cola and artificial saliva ($p = 0.328$), but significant differences were noted between the remaining solutions ($p < 0.001$). As noted earlier, the greatest Δ*E* was recorded for the Vertise Flow composite in tea and the slightest change was seen in the Premise Flow composite in artificial saliva (Fig.1).

Discussion

Dental composite resins are widely used these days as esthetic restorative materials. However, the color change in composite restorations is one of the most common reasons for the replacement of these restorations.[17,18] Color stability is a critical clinical property that affects the esthetic success and longevity of composite restorations. The color change can be considered as an indicator of aging or damage to restorations.[19]

A change in the color of composite restorations over time is a multifactorial process. The color stability of composite resins depends on the resin matrix, filler dimensions, polymerization depth, and color factor, and on chemical differences in the resin components, such as the purity of monomers and oligomers, the type or concentration of the activator, initiator and inhibitor, and the oxidation of the carbon bonding.[20] Studies have shown that physicochemical reactions, such as visible light radiation, ultraviolet radiation, temperature, and heat, can cause internal color variations in composites over time.[21] Composite resin materials are continuously exposed to saliva, foods and drinks. These factors as well as oral hygiene and the smoothness of the surface can all affect the color stability of composite restorations.[22] The aim of the present study was to assess the color stability of a self-adhesive composite resin after immersion in various solutions (coffee, tea, cola, and artificial saliva) as compared with conventional composites.

Natural saliva has a protective effect. By creating a surface barrier, it prevents the staining of the teeth and dilutes staining solutions.[23] In the present study, for the maximal simulation of clinical conditions, coffee, tea and artificial saliva were incubated at 37°C (oral temperature), and the samples immersed in cola were refrigerated at 4°C.

Simplifying the procedures involved in applying dental materials is useful to minimize errors and to save time. Recently, self-adhesive composites have been introduced to the market. Due to their acidic monomer composition, they bond to the dental structure without requiring an adhesive system. In this study, Vertise Flow, the first self-adhesive composite introduced to the market, was compared with other conventional composites. Vertise Flow has a glycerol phosphate dimethacrylate (GPDM) adhesive, which acts as a bonding agent. This acid phosphate group is used for etching and chemical bonding to calcium ions in the tooth structure.[19]

Polishing may affect the quality of the composite surface, which means the polishing technique can also induce the color change of composite resins.[20] In the present study, all samples were polished under the same conditions for the purpose of standardization. This was done to minimize the surface roughness and to ensure that the color change calculated at the end of the study was due to the inherent properties of the composite resins.

Coffee, tea and soft drinks (such as Coca-Cola) are among the commonly consumed drinks that have the potential to cause the staining of restorative materials,[21,22] and were therefore selected for this study.

The CIELAB color scheme was selected for color evaluation in this study, as it is a standard method for measuring color differences based on human perception.[23] The assessment of composite discoloration can be done visually or using instruments. Instrumental techniques have the advantage of eliminating the subjective interpretations of the color change. Therefore, spectrophotometers and colorimeters are widely used tools to detect the color change in dental restorative materials.[24]

According to previous studies, the ΔE values <1 are not recognizable by the human eye, which can detect $\Delta E > 1$.[25–27] The ΔE values $1 < \Delta E < 3.3$ are clinically acceptable, whereas any $\Delta E > 3.3$ is not.[26,27] The results of this study showed that the effects of the type of composite and the type of solution on color stability were significant ($p < 0.05$), but the interaction effect of these 2 factors on Δa^*, Δb^*, ΔL^*, and ΔE was not significant ($p > 0.05$). The ΔE value of a composite resin is especially important for anterior restorations. Composite resins decompose over time due to their polymer nature, which leads to their discoloration.[28]

In the present study, the lowest ΔE value after 8 weeks of immersion was noted in Filtek Z250 in artificial saliva ($\Delta E = 1.02$), whereas the highest ΔE was noted in Vertise Flow in tea ($\Delta E = 35.42$). Also, significant differences were noted in the color stability of the composites in coffee and tea in comparison with other solutions ($p < 0.001$), and ΔE for coffee was significantly higher than that for tea, cola and artificial saliva. At 2 weeks, ΔE for coffee was significantly higher than ΔE for the other 3 staining solutions. At 4 and 8 weeks, tea, coffee, cola, and artificial saliva caused discoloration in a descending order, and there were significant differences between the ΔE values for tea and coffee.

All composites showed discoloration in all staining solutions, which increased over time. Composite discoloration is probably due to the external absorption of stains. Coffee and tea cause yellow stains with different polarization. The release of components of a higher polarity (tea) is greater over time. In the first 2 weeks, coffee caused greater discoloration, but at 4 and 8 weeks, the discoloration caused by tea was greater than that caused by other solutions.[29,30] The discoloration caused by tea is due to the adsorption of polar colorants into the surface of composite resin materials and can be removed by tooth brushing, whereas the discoloration caused by coffee is due to both absorption and adsorption of polar colorants into the surface of the materials. The adsorption and penetration of colorants into the organic phase of the materials are probably due to the compatibility of the polymer phase with the yellow colorants of coffee.[31] Cola, despite having the lowest pH of the staining solutions tested, may possibly cause more degradation, but did not cause as much color change as coffee and tea did, probably due to the lack of yellow colorants in its composition.[30,31]

Arregui et al. assessed the color stability and water sorption of the composites immersed in distilled water, coffee, cola, and orange juice, and showed that the highest degree of Δ*E* was related to tea and coffee.[21] Orange juice caused a moderate Δ*E*; the lowest degree of Δ*E* was caused by distilled water and cola.[21] In a study by Hasani-Tabatabaei et al., the cola and saliva solutions did not cause significant staining in the Filtek™ Supreme, Tetric® EvoCeram, Filtek Z250, or Tetric® Ceram D composites.[27]

Malekipour et al. examined the discoloration of the 3M Filtek™ Z100 composite caused by the tea and coffee solutions, and showed that the highest degrees of staining were related to tea and coffee, whereas water caused the least staining after 14 days.[28] After 1 day of immersion, the least Δ*E* was related to coffee as compared to tea, cola, lemonade, and distilled water.[28]

Arregui et al. evaluated the color stability of the composites immersed in water at 60°C for 30 days. The highest degrees of Δ*E* were noted in self-adhesive Vertise Flow and GF-10, whereas the Premise Flow composite showed the least Δ*E*.[2] These authors also listed a high temperature as a reason for the decomposition of the resin matrix.[2]

Our results showed that at 2 and 4 weeks, the Δ*E* of the Vertise Flow composite was higher than that of the Premise Flow and Filtek Z250 composites ($p = 0.02$); however, the difference between Premise Flow and Filtek Z250 was not statistically significant. At 8 weeks, there was no significant difference in color stability between the composites. A higher degree of Δ*E* for Vertise Flow may be due to its high water sorption.[25] Self-adhesive composites exhibit more hydrophilic properties than other composites, due to carboxylic acids or phosphate groups in their composition.[29,30] The presence of hydroxyl, carboxyl and phosphate groups in monomers confers hydrophilicity, making self-adhesive composites more susceptible to water sorption.[31]

Arregui et al. assessed the color stability and water sorption of the flowable composites immersed in distilled water, coffee, cola, and orange juice, and showed that the lowest degrees of Δ*E* were noted in the Filtek™ Bulk-Fill composite in distilled water, and in Vertise Flow in coffee and orange juice.[21] At the same time, Vertise Flow showed the highest degree of water sorption. There were significant correlations between water sorption and solubility as well as between water sorption and Δ*E*.[21]

The samples immersed in artificial saliva also showed Δ*E* over time due to water sorption by the composites as well as the hydrophilic property of the resin matrix.[12] Water sorption decreases the longevity of composite resins due to the plasticization of the resin and the formation of microcracks, which progress at the matrix–filler interface and lead to the color change.[21,31]

It seems that the degree of staining of a composite depends on the physical properties of the resin matrix, such as its hydrophobicity and hydrophilicity, rather than on the surface properties of the substance and the size of filler particles. It has been observed that hydrophilic materials exhibit a high degree of water sorption, and consequently show a greater color variation than hydrophobic materials. If the composite can absorb water, it can absorb other drinks and stains as well.[27]

Premise Flow is a medium-viscosity nanofill resin with 0.4 nm filler size and 75.5 wt% filler content. The resin matrix of this composite is composed of bisphenol A diglycidyl methacrylate (Bis-GMA), ethoxylated bisphenol A diglycidyl methacrylate (Bis-EMA) and triethylene glycol dimethacrylate (TEGMA). Filtek Z250 is a microhybrid composite. It has silica and zirconia fillers, measuring 0.01–0.50 μm, and containing 78 wt% and 60 v% of mineral fillers. The resin matrix of Filek Z250 consists of Bis-GMA, Bis-EMA and urethane dimethacrylate (UDMA).

Ethoxylated bisphenol A diglycidyl methacrylate is a type of ethoxylated Bis-GMA, which is highly hydrophilic and has no reactive hydroxyl group in its main polymer chains; it should therefore exhibit insignificant water sorption. Yap and Wee also showed that Bis-EMA-based composites were very resistant to the adverse effects of foods on color stability.[12] On the other hand, composites with UDMA or Bis-GMA modified with UDMA were more resistant to the color change and water sorption than those with the Bis-GMA base.[32]

The size and distribution of filler particles are also related to the color change. Composites with larger filler particles are more susceptible to the color change from water storage than those with smaller filler particles, which is due to the hydrolysis at the filler–matrix interface.[24] Ertaş et al. reported that Filtek Z250 had the lowest degree of staining as compared to nanohybrid composites.[23]

Liebermann et al. evaluated the color stability of self-adhesive composites in different solutions for 1 year, and concluded that self-adhesive composites had high water sorption and a high Δ*E*.[30]

In our study, the Δ*E* values were >3.3 for all 3 composite resins, which was clinically unacceptable. Several factors may be responsible for this finding. First of all, not using a toothbrush with toothpaste during the study could have resulted in a reduction in the oral hygiene status in comparison with conditions in the oral cavity. Saliva has a protective and diluting effect in the oral cavity, and consequently prevents the staining of the teeth and dental restorations. In addition, in this in vitro study, all samples were immersed in the solutions for 3 h continuously, which is different from the clinical setting. Thus, it may be concluded that the severity of staining which occurs in vitro is higher than that in the clinical setting.[31,32]

Methodological limitations are inherent to all in vitro studies. In this study, the samples had flat surfaces, whereas resin restorations in the oral environment have irregular, convex or concave surfaces. Moreover, in this study, the samples were dipped in static staining solutions, unlike in the oral cavity, where solutions are in a dynamic state. Last but not least, factors such as thermal alterations or abrasion were not simulated in this study.[33]

Conclusions

Our findings demonstrated that the self-adhesive composite resin tested in the study underwent greater discoloration after immersion in the staining solutions than the conventional composites. Coffee after 2 weeks, and tea after 4 and 8 weeks caused a greater color change in all 3 types of composite resins as compared to other solutions.

Since a number of factors play a role in the oral environment, it is difficult to generalize in vitro findings to the clinical behavior of restorative materials in the oral cavity; therefore, in vivo studies are required to obtain more reliable results.

Further studies are needed to clarify the issue, but it can be concluded that the color stability of tooth-colored restorations in the oral environment depends on dietary habits, and can be preserved by limiting the intake of colored foods and drinks.

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Comparative evaluation of the depth of cure and surface roughness of bulk-fill composites: An in vitro study

Ocena porównawcza głębokości utwardzenia i chropowatości powierzchni materiałów kompozytowych typu bulk-fill – badanie in vitro

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

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Abstract

Background. Composites are in great demand due to the esthetic needs of the patients, which explains a wide variation in the types of available composites. However, the mechanical strength of the materials is questionable. Therefore, the mechanical properties of the newly available bulk-fill composites have been tested.

Objectives. The main objective of the study was to compare the depth of cure (DOC) and surface roughness of 3 different bulk-fill composites: X-tra fil® (XTF), Tetric EvoCeram® Bulk Fill (TEC) and Beautifil® Bulk Restorative (BBR).

Material and methods. Fifty-seven (n = 19 in each group) samples were made using brass molds. All samples were subjected to Vickers hardness testing and profilometry. The one-way analysis of variance (ANOVA) test was used for the data analysis, followed by Tukey's post hoc test.

Results. The differences in the mean surface microhardness values of the materials were statistically significant ($p < 0.001$), with XTF showing the highest value. The TEC composite showed a higher surface roughness as compared to BBR and XTF.

Conclusions. The results of the present study indicate that variations in the filler size and amount significantly influence the DOC and surface roughness of dental composites. Among the tested composites, the multi-hybrid composite exhibited superior DOC (XTF), whereas the nanohybrid composite exhibited superior surface finish (TEC).

Key words: hardness, surface roughness, bulk-fill composites, depth of cure

Słowa kluczowe: wytrzymałość, chropowatość powierzchni, materiały kompozytowe typu bulk-fill, głębokość utwardzenia

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Introduction

The growing demand for life-like restorations and the motivation of some dentists to provide mercury-free, tooth-colored restorations have led to an increase in the use of resin-based composite (RBC) materials. Some of the advantages of composite restorations include better esthetics, reduced need for extensive tooth preparation and reinforcement of the remaining tooth structure.[1]

At present, most RBCs are supplied as light-activated materials and their clinical placement requires the incremental layering technique.[2] The material has to be placed in layers to allow light penetration, which would ensure complete polymerization. This process is time-consuming and may lead to the inclusion of voids in the restoration. In addition, non-uniform curing may lead to uncured RBCs at the bottom or in between the increments, resulting in the restoration with inadequate strength and marginal leakage, and thus of a reduced longevity. Furthermore, uncured RBCs may also cause postoperative sensitivity. To overcome the disadvantages of the conventional incremental placement of RBCs, bulk-fill composites were introduced to reduce the chair time and to offer a less technique-sensitive material.[3] Bulk-fill composites are newer restorative materials which are said to present improved cure, controlled polymerization contraction stresses and a reduced cuspal deflection; they are effectively photoactivated in layers up to 4 mm, and as such can be used in deep preparations.[4,5]

A higher translucency observed in bulk-fill composites as compared to conventional resin composites enables superior light transmission, and thus better polymerization. A higher percentage degree of conversion (DC) displayed by bulk-fill composites is due to better light penetration, as the materials exhibit a reduced opacity. Other factors, such as the filler content, size modifications or the use of a monomer of a higher molecular mass in the resin, also significantly influence the translucency of these materials.[6]

Several bulk-fill composites are now available on the market, claimed to exhibit superior depth of cure (DOC), exceeding 4 mm. Among these, X-tra fil® (XTF) (Voco GmbH, Cuxhaven, Germany), a posterior composite based on the multi-hybrid filler technology, is available in a universal shade, which can be cured up to a depth of 4 mm in 10 s. It consists of bisphenol A-glycidyl methacrylate (Bis-GMA), urethane dimethacrylate (UDMA), triethylene glycol dimethacrylate (TEGDMA), and the barium-boron-aluminosilicate (Ba-B-Al-Si) glass filler. By adjusting the filler size, an extremely high filler content (86 wt%) has been achieved in the material, with a consequent increase in wear resistance and a lower polymerization shrinkage. In addition, it also exhibits superior radiopacity.

Tetric EvoCeram® Bulk Fill (TEC) (Ivoclar Vivadent AG, Schaan, Liechtenstein) is a nanohybrid composite containing dimethacrylates, which make up 20–21 wt% of the monomer matrix. Fillers such as ytterbium trifluoride (YbF_3), barium (Ba) glass, mixed oxides, and a prepolymer constitute up to about 79–81 wt%. The particle size of the filler varies between 40 and 3,000 nm, with a mean particle size of 500 nm. It also contains Ivocerin®, a photoinitiator, which allows DOC of 4 mm and makes it possible to minimize the shrinkage stress.

Beautifil® Bulk (Shofu Dental Corporation, San Marcos, USA) is prepared based on the pre-reacted glass (PRG) technology, in which acid-reactive fluoride glass is made to react with polyacid in the presence of water, after which it is milled, silanized and ground to fine powder for use as a filler in the resin matrix. The resin matrix comprises Bis-GMA, UDMA, 2,2-bis[(4-methacryloxy polyethoxy)phenyl]propane (Bis-MPEPP), and TEGDMA. It is a radiopaque light-cured material with a 4-millimeter DOC. Apart from being biocompatible, this material has the ability to reduce tooth demineralization by releasing fluoride, which also imparts anti-plaque activity.[7] It is available in low viscosity (Beautifil Bulk Flowable) as well as in packable consistency (Beautifil Bulk Restorative – BBR).

Though the compositional differences account for significant differences in the properties of the commercially available composites, it is of utmost importance to ensure the complete polymerization and highly polished surfaces of these materials during clinical placement. Surface texture has a great influence on plaque accumulation, and on the discoloration, wear and esthetic appearance of the restoration.[8] The polishability of RBCs is directly affected by the structure of the organic matrix and the characteristics of the fillers.

In addition, the extent of polymerization of RBCs also influences their physical, mechanical and biological characteristics. Incomplete polymerization due to the inadequate curing of RBCs may impair their physical and mechanical properties, and even make them toxic to the pulp. Surface hardness has been widely used in the literature as an indicator of the extent of polymerization of dental composites. An arbitrary minimum value of the bottom-to-top surface hardness ratio is commonly calculated to establish the DOC of RBCs. For a material to be considered as adequately cured, the value of the ratio needs to be in the range of 0.8–0.85.[7]

The various available bulk-fill composites certainly widen the selection of tooth-colored restorative materials and help reduce the chair time during the placement of the restoration due to generally better DOC. However, the performance and longevity of such materials largely depend on the exact DOC and complete polymerization. Hence, the aim of the present study was to evaluate the surface roughness and DOC of commercially available bulk-fill composites, such as XTF, TEC and BBR.

Material and methods

Three packable bulk-fill composites – XTF, TEC and BBR – in universal A shades (IVA) were used in the present study. The IVA shade was selected to minimize the effects of colorants on light polymerization.

Specimen preparation

A total of 57 samples were prepared, 19 samples from each of the 3 selected bulk-fill composite materials (Fig. 1). The specimens were prepared using a custom-made brass mold, consisting of 5 cylindrical slots of a diameter of 10 mm and a height of 4 mm (Fig. 2). The mold was filled using a single increment of the composite material and covered with a Mylar strip. The mold was pressed between 2 transparent glass plates to remove the excess material. Subsequently, the composite was light-cured according to the manufacturer's recommendations, using the Bluephase® G2 light-cure device (Ivoclar Vivadent AG) within the range of 1,200–1,400 Mw/cm^2. The tip of the curing device was kept in direct contact with the glass plate to maintain the standardized distance from the tip of the device to the top surface of the specimen.

After curing, the samples were retrieved and polished using the Super-Snap disks (Shofu Dental Corporation) at coarse, medium, fine, and superfine grits for 30 s. After each step of polishing, the specimens were thoroughly rinsed with water and air-dried before the next step until final polishing. The samples were stored in artificial saliva at 37°C for 24 h before testing. All specimens were observed under a bright light source, and specimens with any voids or cracks were not included in the study (Fig. 3).

Fig. 2. Schematic representation of a brass mold used for specimen preparation

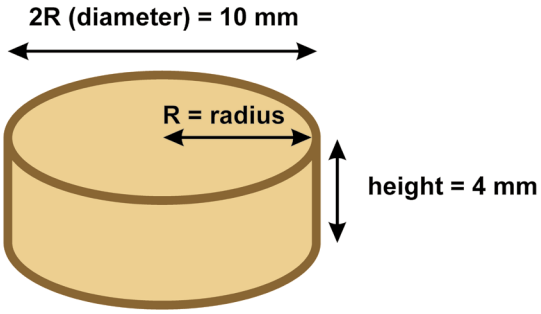

Fig. 3. Schematic representation of a composite specimen

Depth of cure by the Vickers hardness testing method

The surface hardness of the specimens was measured on both the top and bottom surfaces using the Vickers hardness tester (MMT-X7A; Matsuzawa Co. Ltd., Tokyo, Japan). The specimens were secured onto a platform and were subsequently indented with a square-based diamond pyramid indenter with a load of 300 g for 15 s, with an automatic loading and release mechanism. Then, the lengths

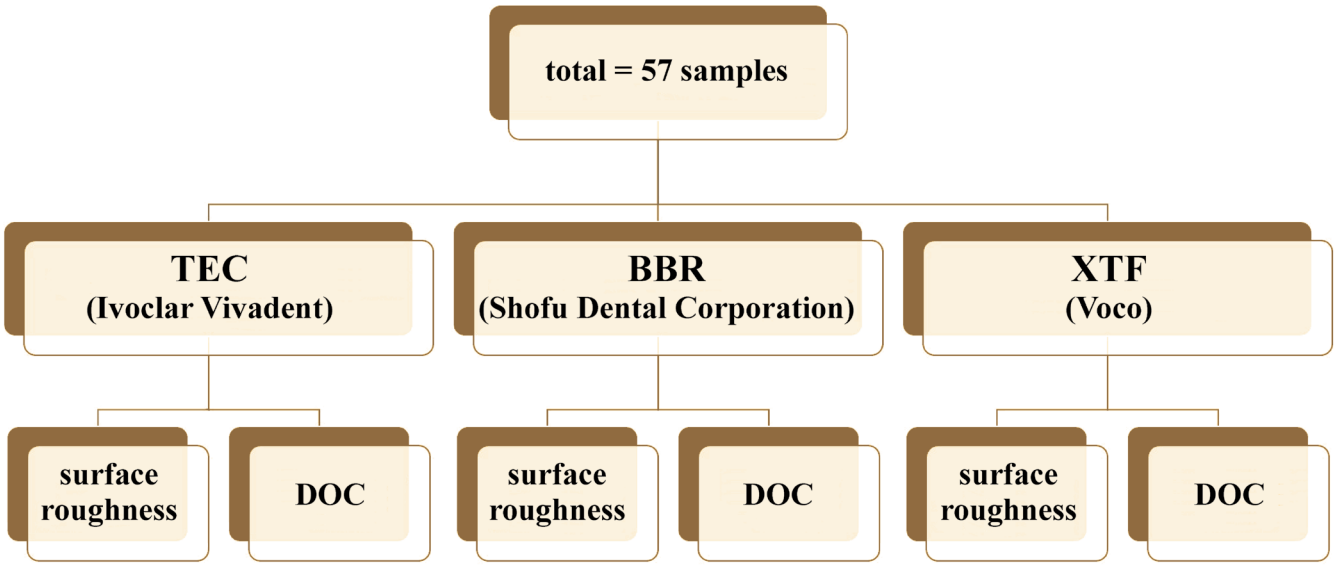

Fig. 1. Study design

XTF – X-tra fil; BBR – Beautifil Bulk Restorative; TEC – Tetric Evoceram Bulk Fill; DOC – depth of cure.

of the diagonals of the indentations were measured using a built-in microscope, and the surface hardness for each indentation was automatically calculated and displayed on the digital readout of the machine. Three different indentations were done on both the top and bottom surfaces of each specimen. The surface hardness values measured on the top of the specimen were considered as 100%, and the values measured at a 4-millimeter distance were expressed as a percentage of the top surface hardness value and were obtained using the following formula:

$$VHN = \frac{\text{bottom VHN}}{\text{top VHN}} \times 100 \quad [\%] \qquad (1)$$

where:
VHN – Vickers hardness number.

Surface roughness measurement

After polishing, the composite surfaces were assessed quantitatively for surface roughness using profilometry (Talysurf®; Taylor-Hobson Ltd., Leicester, UK) with a measurement range of 0.05–10.0 μm and an accuracy of ±0.01 μm. Surface roughness was described with the arithmetic mean of the absolute ordinate values (average roughness Ra, as per ISO 4287). The specimens were secured onto a non-vibrating specimen holder and the stylus of the profilometer was lowered onto the specimen perpendicularly. Surface roughness was measured by moving the stylus along a 0.8-millimeter length of the surface at 3 different locations on each surface.

Statistical analysis

The obtained results were then subjected to the statistical analysis using the PASW Statistics for Windows software, v. 18.0 (SPSS Inc., Chicago, USA). The one-way analysis of variance (ANOVA) with Tukey's post hoc test for intergroup comparison were performed, and a *p*-value of <0.05 was considered statistically significant.

Results

The main aim of the present study was to evaluate the surface roughness and surface hardness of 3 commercially available bulk-fill composites. The mean top surface microhardness values for the XTF, BBR and TEC composite resins were found to be 91.87 ±3.68, 61.92 ±2.22 and 45.44 ±3.05 VHN, respectively (Fig. 4, Table 1). The mean surface microhardness values at a depth of 4 mm (the bottom surface of the specimen) for the XTF, BBR and TEC composite resins were found to be 73.97 ±2.96, 46.78 ±2.34 and 35.40 ±1.81 VHN, respectively (Fig. 4, Table 1). The observed differences between the materials in the surface hardness values on both the top and bottom surfaces were found to be statistically significant ($p < 0.001$). Among the composites, XTF showed a significantly higher surface hardness than other materials on both surfaces ($p < 0.001$). As compared to TEC, a significantly higher surface hardness was observed for the BBR composite ($p < 0.001$).

The calculated DOC at 4 mm for the composite materials used in the present study was found to be 80.62 ±4.20, 75.56 ±2.67 and 78.24 ±6.72 for XTF, BBR and TEC, respectively (Table 1). The DOC of XTF was significantly higher in comparison with other materials ($p < 0.001$).

The mean surface roughness, Ra, is presented in Fig. 5. The mean surface roughness of XTF and BBR was found to be 0.04 ±0.02 and 0.04 ±0.01 μm, respectively, which means a statistically non-significant difference (Table 1). However, the mean surface roughness of TEC was found to be 0.06 ±0.01 μm, which is significantly different from that for BBR ($p < 0.001$).

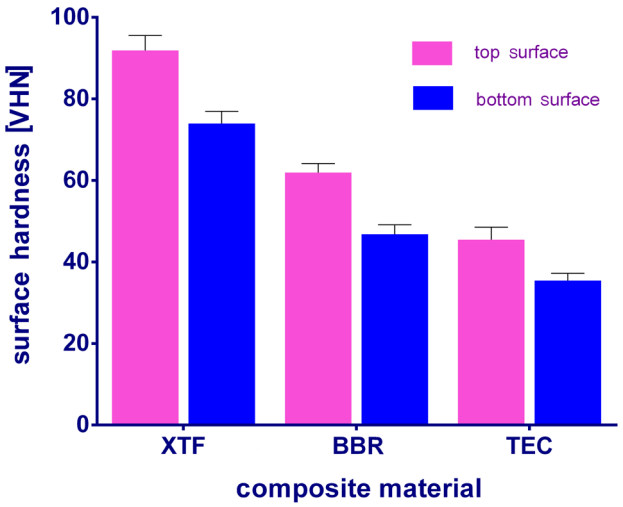

Fig. 4. Top and bottom surface hardness of the composite materials
VHN – Vickers hardness number.
Data presented as mean ± standard deviation (*SD*).

Table 1. Comparison of surface hardness at the top and bottom (a 4-millimeter depth) surfaces, DOC and surface roughness (Ra) of the composite resins

Property	XTF[1]	BBR[2]	TEC[3]	*p*-value	post hoc test
Surface hardness at the top surface [VHN]	91.87 ±3.68	61.92 ±2.22	45.44 ±3.05	<0.001	1 > 2 > 3
Surface hardness at the bottom surface [VHN]	73.97 ±2.96	46.78 ±2.34	35.40 ±1.81	<0.001	1 > 2 > 3
DOC	80.62 ±4.20	75.56 ±2.67	78.24 ±6.72	<0.001	1 > 3 > 2
Surface roughness (Ra) [μm]	0.04 ±0.02	0.04 ±0.01	0.06 ±0.01	<0.001	3 > 2, 1

Data expressed as mean ±*SD*.

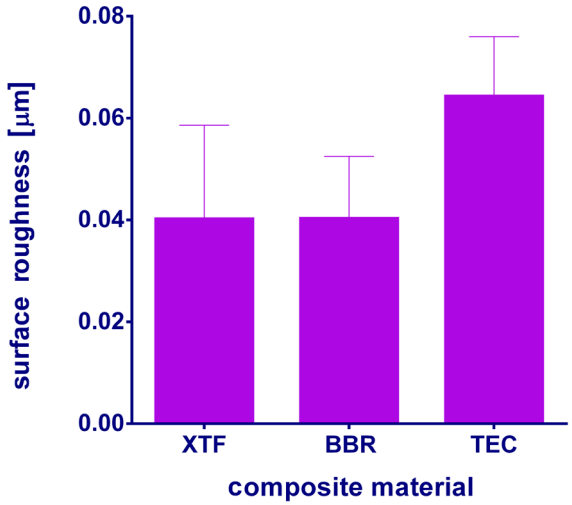

Fig. 5. Surface roughness (Ra) of the composite materials
Data presented as mean ±*SD*.

Discussion

Ultimately, any dental restorative material is to mimic the biological, functional and esthetic properties of a healthy tooth structure. Over the years, there has been an increasing need for better tooth-colored restorative materials to replace the missing tooth structure. The development of resin composite materials for direct restorations with improved physical and mechanical properties, esthetics and durability has been the focus of research in the recent past.[9] The most common strategies to improve the properties of dental composites include modifications in the filler content, and variations in the size, type and morphology of the filler particles and the organic matrix.[10] Together, these changes have resulted in the higher mechanical strength and modulus of elasticity of newer resin composite materials.

To facilitate the reduction of the clinical working time of composite resin placement, bulk-fill composites were developed with a single curing depth of 4 mm. Currently, they are available as low-viscosity bulk-fill composites for use as a base or high-viscosity bulk-fill composites for restorative purposes.[11] The manufacturers of bulk-fill composite materials claim their higher DOC, and over the years, multiple techniques have been investigated to accurately measure the DOC of composite resins. Among these, ISO 4049 or the scraping technique is widely used to determine hardness; still, for bulk-fill RBCs, this technique tends to be overrated. Some authors have demonstrated a good correlation between hardness testing and DC using Raman or Fourier transform infrared (FTIR) spectroscopy for measuring the DOC of composite resins.[12] However, in the present study, the Vickers microhardness tester was used to evaluate the DOC of the experimental resin composites. This method is easy and quick, and requires a minimal area of the specimen surface for testing.[12–14] The size of the Vickers hardness indenter is larger than the size of the filler particles in the material complex; as a result, VHN takes into account not only the filler component, but also the surrounding softer resin matrix. In this regard, VHN indirectly considers the entire matrix network crosslinking.[15,16] A low surface hardness value is largely related to inadequate wear resistance and susceptibility to scratch damage, which can compromise the fatigue strength of the restoration and lead to its failure.[17,18]

In this study, XTF showed the highest DOC in comparison with the other 2 materials. A high DOC shown by XTF can be attributed to the presence of macrofillers (>20 μm) in XTF, increasing its translucency, which in turn may have led to an increased DOC. However, the difference between the values for XTF and TEC proved to be statistically non-significant. Both TEC and BBR showed lower DOC values in a descending order, though the difference was not statistically significant. X-tra fil is a multi-hybrid composite with 86 wt% of the Ba-B-Al-Si glass filler. Tetric EvoCeram Bulk Fill contains 79–81 wt% of YbF_3, Ba glass, mixed oxides, and a prepolymer. On the other hand, BBR contains 87 wt% or 74.5 vol% of fluorine-boron-aluminosilicate (F-B-Al-Si) glass as a filler. Light reflection from RBCs, deflection of light from the filler particles and light absorption by the photoinitiators lead to a reduction in light penetration into the depth of conventional composites, thus limiting their DOC to 2 mm. The filler composition as well as the filler size play an important role in light diffusion in RBCs, determining their DOC. Bulk-fill RBCs have an increased filler size (macrofillers). Light penetration is higher due to a reduced surface area of the macrofillers with a reduced resin–filler interface, and hence DOC is increased. Similarly, some of the low-viscosity bulk-fill composites with a lower filler content also exhibit a higher DOC.[19]

A decreased DOC of TEC could be due to the difference in the photoinitiator system. This composite contains Ivocerin and (2,4,6-trimethylbenzoyl)diphenylphosphine oxide (TPO) as photoinitiators, which are suggested to increase DC as compared to camphorquinone (CQ). However, in this study TEC showed a lower DOC than XTF. This may be explained by the filler size in TEC, which is smaller than in the case of XTF, leading to a higher light reflection and a decreased translucency, which in turn reduces DOC.[20] The BBR composite showed the lowest DOC among the tested materials. This could be attributed to an increased filler content (87 wt% or 74.5 vol%), which may reduce the translucency of the material.[21] These results are in agreement with previous studies suggesting that filler loading and the filler particle size influence the DOC of composite resins.[19,21,22]

The longevity of the RBC restoration depends on its resistance to wear in the oral cavity, which is influenced by a variety of factors, such as the magnitude of forces of mastication, the patient's diet, temperature variations, bacterial products, enzymes, etc.[23–25] Mylar strips enhance surface smoothness, but, clinically, restorations need contouring in order to eliminate the excess material. Various polishing systems are available on the market. However, in this study, the single polishing system with multi-step polishing SuperSnap was used as a standardized method.[26] Profilometry

and atomic force microscopy (AFM) have been commonly used to assess surface roughness. A well-established method for checking surface roughness is stylus profilometry, which is simple and easy to perform.[27]

In this study, TEC showed the highest surface roughness as compared to XTF and BBR. The filler content in TEC is lower (79–81 wt%) than in other composites. Conventionally, it has been observed that RBCs with bigger filler particles show a greater surface roughness following polishing. In this study, the Super-Snap disks were used for polishing and the scope for polishing in the different materials could have varied.[28] The similar surface roughness of BBR and XTF can be attributed to the similar filler size and content (by weight). The results of the present study indicate that the surface roughness of RBCs is significantly influenced by the filler size and content.[29] However, the polishability and the resultant surface roughness are also affected by other factors, such as the abrasive agent used, the amount of time each abrasive agent is used, pressure, the number of uses, and the direction in which the abrasive agent is used.[30]

Conclusions

Within the limitations of the present study, it can be concluded that the DOC and surface roughness of the tested materials were significantly different. Among the materials evaluated in the present study, XTF exhibited superior DOC as compared to TEC and BBR. The TEC composite exhibited a higher surface roughness than XTF and BBR.

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Assessment of a single versus double application of low-level laser therapy in pain reduction following orthodontic elastomeric separation: A randomized controlled trial

Ocena pojedynczego lub podwójnego użycia lasera małej mocy w redukowaniu bólu związanego z separacją elastomerową – randomizowane badanie kontrolowane

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Abstract

Background. Periodontal ligament (PDL) pain, associated with the insertion of elastomeric separators, is one of the most annoying experiences at the beginning of orthodontic treatment. Low-level laser therapy (LLLT) has recently been suggested as a method of controlling this pain.

Objectives. The aim of this study was to evaluate the effectiveness of LLLT on reducing the pain associated with elastomeric separation and to compare a single dose of LLLT (1 h before the insertion of elastomeric separators) vs 2 doses of LLLT (1 h before and immediately after the insertion).

Material and methods. This randomized controlled trial was conducted at the Department of Orthodontics, Faculty of Dentistry of University of Damascus and at the Department of Orthodontics of Adib-Allaham Center in Damascus, Syria. The sample population was comprised of 36 patients, aged between 12 and 25 years (mean age: 17 ±3.7 years), and the patients were randomly divided into 2 groups. A split-mouth design was employed. One group received 1 dose of laser therapy 1 h before the insertion of elastomeric separators, whereas the other one received 2 doses of laser therapy – 1 h before and immediately after the insertion of elastomeric separators. A GaAlAs laser with a wavelength of 830 nm and a power of 100 mW was used. The pain levels were recorded using the Visual Analog Scale (VAS) after 1, 6, 24, 48, and 96 h. Student's *t*-test and the repeated measures analysis of variance (ANOVA) were employed to detect significant differences.

Results. Low-level laser therapy significantly reduced post-separation pain when the experimental side was compared to the placebo side in the single-irradiation group ($p < 0.05$). Low-level laser therapy significantly reduced separation pain with no statistical difference between single and double irradiation protocols ($p > 0.05$).

Conclusions. Low-level laser therapy had a positive effect on reducing the pain associated with elastomeric separation, whether it was applied as a single dose before elastomeric separation or as a double dose before and after this procedure.

Key words: pain control, low-level laser therapy, elastomeric separators, periodontal ligament pain

Słowa kluczowe: kontrola bólu, terapia laserem małej mocy, separatory elastomeryczne, ból więzadeł przyzębnych

Introduction

The periodontal ligament (PDL) is a slightly elastic, fibrous structure connecting the tooth to the alveolar bone.[1] It plays a key role in the physiological and orthodontic movements of the tooth.[2] When the tooth moves, tensile strain occurs in PDL, which is then transferred to and received by the receptors in the alveolar bone.[1] Orthodontic forces stimulate the production of pro-inflammatory mediators, such as histamine, dopamine, serotonin, glycine, prostaglandins, and cytokines.[3] This may cause severe pain due to the inflammatory process occurring in PDL, which may hamper routine oral hygiene.[4] Many surveys have considered pain to be the main cause of the interrupted orthodontic treatment.[5] Other studies have revealed that 70–90% of patients complain about pain during orthodontic treatment,[6] and that 30% of patients discontinue the treatment.[7] Dental separation is used during fixed orthodontic treatment,[8] aiding in the case of impacted teeth – in particular the second molars[9] – prostheses and interproximal dental restorations.[10]

Elastomeric separators are widely used due to their durability and ease of use with children and adolescents.[11,12] The application of elastomeric separators is usually accompanied by pain.[7,13] This pain typically starts within 24 h of insertion,[7,13,14] increases over the next 24–48 h[7,15] and subsides within 5–7 days.[8,14,15] The patient may need an analgesic to relieve it.[16] Non-steroidal anti-inflammatory drugs (NSAIDs) are widely used in dentistry to control the pain which accompanies various dental procedures.[17] A prophylactic dose seems to be more effective in controlling pain than the standard method of administering analgesics after treatment.[18,19] It seems that taking a prophylactic NSAID allows enough time for it to be metabolized and begin its analgesic effect before pain sets in.[13] Non-steroidal anti-inflammatory drugs should be avoided in patients suffering from allergic reactions (especially to aspirin), gastric ulcers, respiratory ailments (asthma), kidney or liver injuries, bleeding from an unknown source, hypertension, bone marrow disorders, or epilepsy.[20]

Analgesics should be avoided during orthodontic treatment, as they interfere with it and may prolong the duration of the treatment.[21] These precautions have encouraged researchers to seek other analgesic methods that would be safer and possibly produce no side effects.[16] Some researchers have suggested non-medicinal methods of controlling pain during orthodontic treatment.[22] These methods include biting on a plastic rod, an acrylic stent or cotton rolls[22,23]; swishing warm water around the mouth also has some analgesic effect if done at the onset of pain.[23] These aforementioned methods increase the supply of blood to the periodontium, and therefore may accelerate pain relief.[22]

Low-level laser therapy (LLLT) has been suggested to reduce pain, and is globally considered to be a safe and effective method for pain management.[17] Low-level laser therapy stimulates cells to produce enkephalins and endorphins,[24] and blocks nervous impulses in the slow-conduction velocity peripheral nerves.[25] It can obstruct the Na^+/K^+ pump in the cell membrane, thus blocking pain impulses and causing an analgesic effect.[26] Low-level laser therapy might induce the production of bradykinin, reducing the efficacy of C-nerve fibers and changing the pain threshold.[27] It reduces edema and decreases pain by reducing the production of prostoglandin E2 (PGE2) and cyclooxygenase-2 (COX-2).[7] It may also reduce the production of tumor necrosis factor alpha (TNF-α) during acute inflammation.[28] The therapy is used in dentistry to control the pain that accompanies extraction and endodontic treatment[29,30] as well as the pain induced by elastomeric orthodontic separation.[31–33]

Several recent studies have found LLLT to be effective in pain reduction when used in conjunction with the insertion of elastomeric separators.[31–33] Eslamian et al. evaluated the effect of double LLLT irradiation on pain reduction and found it to be of great benefit,[7] but no previous studies have tried to compare different protocols of LLLT application to arrive at the best choice for pain control. Therefore, the objectives of this randomized controlled trial were to evaluate the efficacy of LLLT in controlling the pain caused by elastomeric separators and to compare a single irradiation dose 1 h before the insertion of elastomeric separators vs double irradiation, conducted 1 h before and 1 h after the insertion, in terms of pain reduction.

Material and methods

This was a randomized, compound controlled clinical trial, i.e., a two-arm, parallel-group trial and a split-mouth design in each group. The subjects were selected from the patients of the Department of Orthodontics, Faculty of Dentistry of University of Damascus and the Department of Orthodontics of Adib-Allaham Center (Ministry of Health) in Damascus, Syria, between November 2014 and June 2015. This study was approved by the Faculty of Dentistry's Local Research Ethics Committee (reference No. UDDS-380-30032014/SRC-2209) and was registered at https://clinicaltrials.gov (identifier: NCT02209818) before the commencement of data collection. The trial was funded by the University of Damascus Postgraduate Research Budget (Ref. No: 83324208070DEN). The candidates for inclusion were given information sheets and their consent was obtained.

Sample size calculation

The calculation of the sample size was performed using Minitab®, v. 17 (Minitab LLC, State College, USA) with an alpha level of 0.05 and a power of 90%, assuming that the smallest difference requiring detection in the pain level was 10 mm on the Visual Analog Scale (VAS) with a variation (standard deviation – *SD*) of 8.1, based on a previous study.

A sample of 30 patients (15 in each group) was required. The study included 36 patients to compensate for any potential withdrawals.

Eligibility criteria

The inclusion criteria were patients undergoing orthodontic treatment with the need of molar separation before banding and in the age range of 12–25 years. The exclusion criteria comprised chronic disease, chronic or neural pain, periodontitis, treated or untreated apical lesions on the first molar, loose mesial and distal contact points on the first molar, and the presence of gingival pigmentation in the area to be irradiated.

Study groups

The study groups were selected randomly using a list of random numbers generated by a computer program (Minitab, v. 17), which allocated patients to one of 2 equal groups (Fig. 1). The 1st group received a single irradiation dose and the 2nd group received double irradiation. Either the upper or lower jaw was irradiated for each patient, never both. The treatment and placebo sides in each patient were also selected randomly using the list of different allocations generated in Minitab, v. 17.

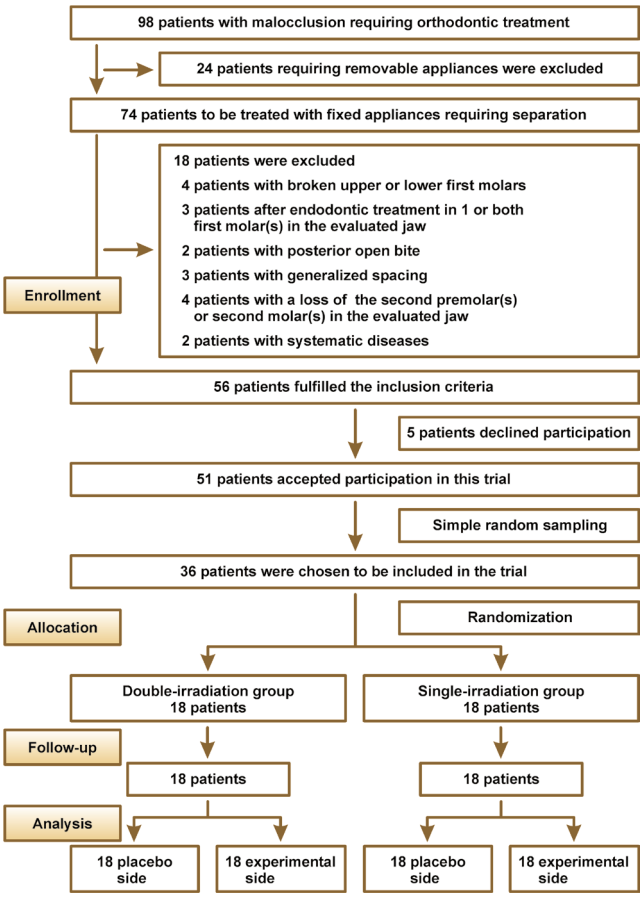

Fig. 1. CONSORT (Consolidated Standards of Reporting Trials) flow diagram of patient recruitment and follow-up

Laser irradiation technique

After isolating the field using cotton rolls, a laser was applied on the experimental side 1 h before the insertion of elastomeric separators in the single-irradiation group, and 1 h before and immediately after the insertion of elastomeric separators in the double-irradiation group. Eight points were irradiated: mesial and distal of the first molar, distal of the second premolar, and mesial of the second molar, on both the buccal and lingual sides (Fig. 2). A GaAlAs laser with a wavelength of 830 nm, an energy density of 4 J/cm^2 and a power output of 100 mW was used for an automated duration of 28 s per point and a laser spot diameter of 7 mm (DioBeam®; CMS Dental, Copenhagen, Denmark) (Fig. 3).

On the placebo side, the same laser device was used after the safety cover was removed so that the device produced the same sounds, but no irradiation occurred. The tip was applied to the same points and for the same duration. The dentist and the patient wore laser protective goggles to prevent any harm to the eyes. To prevent any deviations due to gender, age or personal pain threshold,[34] the patients were not aware of which side represented the placebo (double-blinded study design).

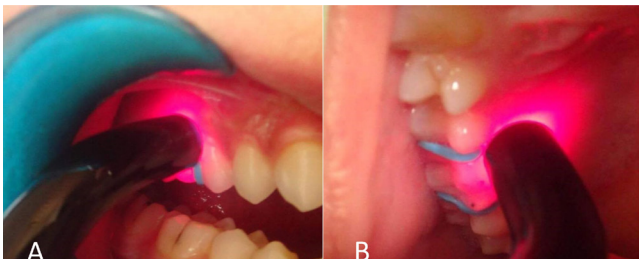

Fig. 2. A – laser irradiation on the buccal side; B – laser irradiation on the lingual side

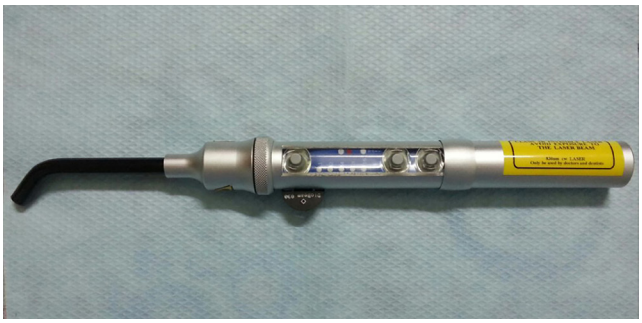

Fig. 3. Laser device used in the current trial

Pain assessment

The patients were given questionnaires to evaluate their perception of pain at the following assessment time points: 1, 6, 24, 48, and 96 h after separation. Every patient was instructed to rate the level of their pain on VAS.

A 100-mm line was used, with the left side representing no pain (i.e., score 0) and the right side representing the worst pain (i.e., score 100). The patients were asked to put a vertical mark on the line at the point which best represented the level of pain they felt. The pain assessment was done by measuring the distance from the left side to the vertical mark in millimeters. The patients were instructed not to take any analgesics during the pain assessment period. In case of severe pain, they were allowed to take 1 or 2 500-mg tablets of paracetamol (acetaminophen), but they were asked to fill in the questionnaire before taking any analgesics.

Statistical analysis

The Microsoft® Excel files (Office Excel 2013; Microsoft Corporation, Redmond, USA) were used for data entry. The statistical analysis was done using Minitab, v. 17 and IBM SPSS Statistics for Windows, v. 21 (IBM Corp., Armonk, USA). Two-sample *t*-tests were employed to evaluate the efficacy of LLLT in reducing pain by comparing the single- and double-irradiation groups. The repeated measures analysis of variance (ANOVA) was employed to evaluate changes in pain perception over time in each group, and the least significant difference (LSD) method was used for the post-hoc tests.

Results

This study was conducted on 36 patients (12 males and 24 females) at an average age of 17.44 years (range: 12–25 years) (Table 1). Pain perception on 1 or both sides started after 1 h in 63.89% of the sample, after 6 h in 13.89% of the sample and after 24 h in 11.11% of the sample. Surprisingly, 4 patients (11.11%) never felt pain at any assessment time point. The maximum pain perception was recorded at 24 h after separation.

In the single-irradiation group, pain increased over time, then decreased, with no significant differences in the pain levels between the assessment time points, whether the comparisons were made on the experimental side or the placebo side (Table 2). In the double-irradiation group, pain increased, then decreased, with statistically significant changes in the pain levels for both the experimental and the placebo sides (Table 2). The post-hoc tests in this group revealed that there were significant differences between the pain levels 1 h after separation as compared to 24 and 48 h, and between the pain levels 24 and 48 h after separation as compared to 96 h (Table 3). On the placebo side, significant differences were found between the pain levels 1 h after separation as compared to 6, 24 and 48 h, and at 24 and 48 h after separation as compared to 96 h (Table 4).

Significant differences in the pain levels were found between the placebo and experimental sides at 6, 24 and 48 h after separation in the single-irradiation group (Table 5). Significant differences in the pain levels were also found between the 2 sides at all assessment time points (at 1, 6, 24, 48, and 96 h) in the double-irradiation group (Table 5). Regarding the experimental sides, when single irradiation was compared to double irradiation, no significant differences between them were observed at any assessment time point (Table 6).

Table 1. Baseline characteristics of the study population

Variable	Single-irradiation group		Double-irradiation group		Total
	male	female	male	female	
Gender n (%)	6 (16.67)	12 (33.33)	6 (16.67)	12 (33.33)	36 (100)
Maximal age [years]	25	21	23	22	25
Minimal age [years]	14	12	20	12	12
Age mean ±*SD*	18.67 ±4.13	16.42 ±3.50	20.83 ±1.17	16.17 ±3.69	17.44 ±3.73
Separation in the maxilla n (%)	5 (13.89)	10 (27.78)	6 (16.67)	9 (25.00)	30 (83.33)
Separation in the mandible n (%)	1 (2.78)	2 (5.56)	0 (0)	3 (8.33)	6 (16.67)

SD – standard deviation.

Table 2. Descriptive statistics of the pain levels in the study groups at the different assessment time points, with the *p*-values of the repeated measures analysis of variance (ANOVA) test

Group	Side	T1	T2	T3	T4	T5	*F*-value	*p*-value
Single-irradiation	experimental	8.50 ±14.29	13.44 ±19.39	22.61 ±28.16	19.28 ±24.59	17.06 ±25.40	1.772	0.171
	placebo	14.94 ±23.99	27.17 ±26.99	33.28 ±31.70	31.22 ±29.60	23.39 ±29.77	2.618	0.071
Double-irradiation	experimental	9.56 ±15.77	19.94 ±27.21	30.28 ±28.22	22.61 ±25.74	15.17 ±23.83	3.499	0.027*
	placebo	20.17 ±18.10	36.95 ±28.69	42.06 ±26.57	37.61 ±28.66	21.50 ±23.44	4.602	0.006**

T1 – 1 h after separation; T2 – 6 h after separation; T3 – 24 h after separation; T4 – 48 h after separation; T5 – 96 h after separation;
* significant difference at $p < 0.05$; ** significant difference at $p < 0.01$. Data for T1–T5 presented as mean ±*SD*.

Table 3. Descriptive statistics of the changes observed between the different assessment time points (pairwise comparisons) in the double-irradiation group on the experimental side

Time point	Comparison	Mean ±*SD*	*T*-value	*p*-value	95% CI	
					lower limit	upper limit
T1	T1 vs T2	−10.39 ±23.04	−1.91	0.073	−21.85	1.07
	T1 vs T3	− 20.72 ±29.35	−3.00	0.008**	−35.32	−6.13
	T1 vs T4	−13.06 ±19.78	−2.80	0.012*	−22.89	−3.22
	T1 vs T5	−5.61 ±19.26	−1.24	0.233	−15.19	3.97
T2	T2 vs T3	−10.33 ±29.73	−1.47	0.159	−25.12	4.45
	T2 vs T4	−2.67 ±29.24	−0.39	0.704	−17.21	11.88
	T2 vs T5	4.78 ±31.82	0.64	0.533	−11.05	20.60
T3	T3 vs T4	7.67 ±25.39	1.28	0.217	−4.96	20.29
	T3 vs T5	15.11 ±22.40	2.86	0.011*	3.97	26.25
T4	T4 vs T5	7.44 ±14.71	2.15	0.046*	0.13	14.76

CI – confidence interval; * significant difference at $p < 0.05$; ** significant difference at $p < 0.01$. The post-hoc tests were performed using the least significant difference (LSD) method and the *p*-values of the significance tests are given.

Table 4. Descriptive statistics of the changes observed between the different assessment time points (pairwise comparisons) in the double-irradiation group on the placebo side

Time point	Comparison	Mean ±*SD*	*T*-value	*p*-value	95% CI	
					lower limit	upper limit
T1	T1 vs T2	−16.78 ±26.45	−2.69	0.015*	−29.93	−3.63
	T1 vs T3	−21.89 ±29.14	−3.19	0.005**	−36.38	−7.40
	T1 vs T4	−17.44 ±23.08	−3.21	0.005**	−28.92	−5.97
	T1 vs T5	−1.33 ±26.54	−0.21	0.834	−14.53	11.86
T2	T2 vs T3	−5.11 ±27.58	−0.79	0.443	−18.83	8.60
	T2 vs T4	−0.67 ±29.08	−0.10	0.924	−15.13	13.79
	T2 vs T5	15.44 ±38.03	1.72	0.103	−3.47	34.36
T3	T3 vs T4	4.44 ±28.15	0.67	0.512	−9.55	18.44
	T3 vs T5	20.56 ±26.84	3.25	0.005**	7.21	33.90
T4	T4 vs T5	16.11 ±24.56	2.78	0.013*	3.90	28.33

* significant difference at $p < 0.05$; ** significant difference at $p < 0.01$. The post-hoc tests were performed using the LSD method and the *p*-values of the significance tests are given.

Table 5. Descriptive statistics of the differences between the experimental and placebo sides in each group at each assessment time point, with the *p*-values of the paired *t*-test

Group	Time point	Difference between the experimental and placebo sides mean ±*SD*	*T*-value	*p*-value	95% CI	
					lower limit	upper limit
Single-irradiation	T1	6.44 ±15.88	0.103	1.72	14.34	−1.45
	T2	13.72 ±18.21	0.005**	3.20	22.78	4.67
	T3	10.67 ±20.80	0.044*	2.18	21.01	0.32
	T4	11.94 ±21.19	0.029*	2.39	22.48	1.41
	T5	6.33 ±15.31	0.097	1.75	13.95	−1.28
Double-irradiation	T1	10.61 ±16.10	0.012*	2.80	18.62	2.61
	T2	17.00 ±21.84	0.004**	3.30	27.86	6.14
	T3	11.78 ±17.69	0.012*	2.82	20.58	2.98
	T4	15.00 ±17.14	0.002**	3.71	23.52	6.48
	T5	6.33 ±10.36	0.019*	2.59	11.48	1.18

* significant difference at $p < 0.05$; ** significant difference at $p < 0.01$.

Table 6. Descriptive statistics of the differences between the single-irradiation and double-irradiation groups on the experimental side at each assessment time point, with the *p*-values of the paired *t*-test

Time point	Single-irradiation group mean ±*SD*	Double-irradiation group mean ±*SD*	Mean difference	*T*-value	*p*-value	95% CI	
						lower limit	upper limit
T1	8.50 ±14.29	9.56 ±15.77	9.15	–11.26	0.835	–0.21	–1.06
T2	13.44 ±19.39	19.94 ±27.21	9.58	–22.58	0.416	–0.83	–6.50
T3	22.61 ±28.16	30.28 ±28.22	11.45	–26.78	0.420	–0.82	–7.67
T4	19.28 ±19.28	22.61 ±25.74	13.74	–20.40	0.694	–0.40	–3.33
T5	17.06 ±17.06	15.17 ±23.83	18.59	–14.81	0.819	0.23	1.89

Discussion

Many studies have used LLLT to reduce the pain caused by elastomeric separation and most of these studies applied single irradiation immediately after inserting elastomeric separators. The results have differed greatly between these studies, ranging from a significant reduction in pain with the use of LLLT to no difference at all.[16,32–36] Reviewing the literature reveals that no previous study has evaluated the use of LLLT before the insertion of elastomeric separators; therefore, no papers are available to compare the current results with.

The pain caused by elastomeric separation is inflammatory in nature. That is why it is important to use a laser that has an anti-inflammatory effect, one which does not cause any thermal changes in the irradiated area. This feature can be found in lasers with a wavelength of 600–1,000 nm.[37] In this study, a GaAlAs laser with an 830-nm wavelength was used. It has been shown to have excellent tissue penetration and to be highly effective in reducing pain in comparison with other lasers.[35,36] Kim et al. used an AlGaInP laser with a wavelength of 635 nm.[34] They used multiple irradiations and found significant differences in pain reduction only on the 1st day, not on the following days.[34] Different outcomes might be due to the use of different types of lasers with different wavelengths. As mentioned previously, a GaAlAs laser with a wavelength of 830 nm is more effective in reducing pain according to previous studies.[35,36]

In the present study, a GaAlAs laser was applied to 8 points: mesial and distal of the first molar, mesial of the second premolar and distal of the second molar, both buccally and lingually. This was similar to what Eslamian et al. used in their study.[7] It has also been documented that low doses of laser irradiation would achieve the desired effect, whereas higher doses (exceeding 5 J/cm^2 per point and 20 J/cm^2 per tooth) could eliminate the analgesic and anti-inflammatory effect.[7,37,38] In the present study, a dose of 4 J/cm^2 was used at each irradiated point, totaling 16 J/cm^2 per tooth; thus, the recommended 20 J/cm^2 limit was not exceeded. Furquim et al. used a high dose of 80 J/cm^2 and found no significant differences between the treatment and placebo sides.[33] This might be due to exceeding the suggested dosage of 0.3–19 J/cm^2.[38]

The current study applied a laser to only 1 jaw in an attempt to prevent the radiation of pain from one jaw to the other.[39] No jaw was preferred over the other, as pain perception in both jaws is similar.[31,37] This was similar to what Eslamian et al. and Marini et al. did in their studies.[7,31]

Eslamian et al. used a double-irradiation method, immediately and 24 h after separation, and found positive results on the side of double irradiation.[7] Few studies have tried to employ multiple-irradiation methods.[19,34] A quick comparison between the studies of Abtahi et al. and Kim et al. reveals no difference in the final outcomes in terms of pain control despite the fact that the 2 studies used different kinds of laser devices, application protocols, laser parameters, and sample sizes.[19,34] Abtahi et al. used a GaAr laser with a wavelength of 904 nm, 6 J of energy and a 7-mm tip diameter,[19] whereas Kim et al. used an AlGaInP laser with a wavelength of 635 nm, 10 J of energy and a 5.6-mm tip diameter.[34] The sample size in the former study was 29 patients divided into 2 groups (the laser group and the control group), with the laser group receiving laser irradiation once per day for 5 days[19]; the sample size of the latter study included 88 patients divided into 3 groups (the laser group, the placebo group and the control group), with laser irradiation being applied every 12 h for 7 days in the laser group.[34] Both studies found that laser application was useful in reducing the peak of pain perception, which generally occurs 24 h after separation, but at the same time they found no statistically significant differences between the laser and control groups at other observation time points.[19,34]

Lim et al. assessed pain in 39 patients after applying different laser doses on each quadrant of both the upper and lower jaws, and found no significant differences between the 4 quadrants regarding pain perception.[40] This might be attributed to the presence of multiple pain-stimulating points (i.e., 4 quadrants), thereby

affecting the patient's ability to point out or distinguish the source of pain, or it might be due to pain radiating from one jaw to the other.

The time of pain perception differed between the patients studied, starting after 1 h in 63.89% of the sample, after 6 h in 13.89% of the sample and after 24 h in 11.11% of the study population. Thus, the majority of patients perceived pain within 1–24 h after dental separation. These results were similar to what Shetty et al. and Artés-Rebas et al. found in their research.[13,35] They found that pain started within 2–24 h of the insertion of elastomeric separators.[13,35] Eslamian et al. found that pain appeared immediately and up to 24 h after separation, and Ngan et al. reported that pain perception began 4–24 h after separation.[7,14]

In the current study, the maximum pain perception was recorded 24 h after the insertion of elastomeric separators, which is in agreement with the findings of Eslamian et al. and Fujiyama et al.,[7,16] whereas Abtahi et al. recorded the maximum pain perception 48 h after separation.[19] This delay might be attributed to the different number of laser applications. Abtahi et al. applied the laser daily for 5 days,[19] whereas Eslamian et al. performed double irradiation[7] and Fujiyama et al. administered a single irradiating dose.[16]

In the current study, no significant differences were found between the experimental sides in the single- and double-irradiation groups, indicating that double irradiation had a similar action to a single application of a laser. However, within each group in this trial, double irradiation appeared to have a superior effect on pain reduction, since all of the comparisons made between the experimental and placebo sides revealed statistically significant differences, whereas in the single-irradiation group significant differences were observed only at 3 assessment time points.

Previously published work has shown that elastomeric separation pain usually decreases within 48–72 h[36] and subsides after 5–7 days.[8,14] Therefore, any assessment after 96 h (i.e., 4 days) would be of great benefit in evaluating the pain levels at later stages, before pain disappears. However, it should be noted that one of the shortcomings of the current study is that the last assessment time was 4 days after separation, and the assessment of the pain levels should have been taken 5, 6 or 7days after the insertion of separators.

The present study found LLLT to be of significant benefit in pain reduction. This effect was similar to the ones reported in different studies when pharmaceutical analgesics were used at the same time points as those used in the current study for laser irradiation.[18,41,42] Low et al. and Minor et al. used an analgesic 1 h before separation.[18,41] Bernhardt et al. administered an analgesic 1 h before separation and 5 h after separation.[42] The results of these studies were similar to those of the current study, indicating the possibility of replacing medication with LLLT when the use of separators is planned.

Limitations

Pain control in the early phase of orthodontic treatment was not studied in this trial, though light should be shed on this very important stage of orthodontic treatment. Comparisons should be made between analgesics and LLLT to find out which is more effective in controlling pain in the daily practice of orthodontists. Future work should also focus on the best laser parameters to yield the most effective pain control during orthodontic treatment.

Conclusions

The application of LLLT significantly reduced the pain induced by elastomeric separation. Low-level laser therapy was beneficial in pain reduction, whether applied in a single dose or a double dose, with no significant differences between the 2 methods.

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Effects of low-intensity pulsed ultrasound (LIPUS) applied on the temporomandibular joint (TMJ) region on the functional treatment of class II malocclusion: A randomized controlled trial

Oddziaływania ultradźwięków o niskiej intensywności stosowanych na okolicę stawu skroniowo-żuchwowego w leczeniu czynnościowym wad zgryzu klasy II – randomizowane badanie kontrolowane

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Abstract

Background. Low-intensity pulsed ultrasound (LIPUS) is considered one of the techniques used to improve the mandibular growth. Many animal studies have reported that significant results can be obtained using LIPUS therapy with functional appliances.

Objectives. This research aimed to evaluate the dentoskeletal changes produced by the combination of LIPUS therapy and functional treatment during the correction of skeletal class II malocclusion.

Material and methods. Forty-five patients aged 10.5–14 years with skeletal class II division 1 malocclusion were randomly divided into 3 equal groups: the LIPUS group, treated with a Twin-Block appliance in combination with LIPUS therapy; the TB group, treated with a Twin-Block appliance only; and the control group, which was observational and received no treatment. Cephalometric changes were compared between the 3 groups using the analysis of variance (ANOVA) and Tukey's post hoc tests at $p < 0.05$.

Results. A greater significant decrease in the ANB (A point, nasion, B point) angle was observed in the treated groups (–2.67° for the LIPUS group and –2.11° for the TB group) as compared to the control group ($p < 0.001$). A greater improvement in the mandibular length and position was observed in the LIPUS group than in the TB group ($p < 0.001$). The changes in the control group as a result of continuing growth were minimal and clinically non-significant.

Conclusions. The application of LIPUS therapy in combination with functional treatment can have a great effect on growth stimulation during the correction of class II malocclusion. In addition, LIPUS was effective in reducing the duration of functional treatment.

Key words: Twin-Block appliance, low-intensity pulsed ultrasound (LIPUS), functional orthodontic treatment

Słowa kluczowe: aparat Twin-Block, ultradźwięki o niskiej intensywności (LIPUS), ortodontyczne leczenie czynnościowe

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Introduction

Class II malocclusion is one of the most frequent orthodontic problems, as its occurrence ranges from 18% to approx. 32% of the population,[1] and most class II malocclusion cases are the result of mandibular deficiency and not of maxillary excess.[2,3] The treatment of class II malocclusion varies widely; growth modification treatment shows the best results in patients with growth potential, but it depends on the phase of skeletal growth.[4,5]

A wide range of functional appliances have been used to stimulate the mandibular growth by forwarding the mandible; they can cause a significant improvement in the facial esthetics and occlusal relationships.[6] Several techniques have been suggested to improve the mandibular growth, such as low-level laser therapy (LLLT), growth hormones and low-intensity pulsed ultrasound (LIPUS). Moreover, these techniques can also reduce the duration of functional treatment.[7]

Recently, LIPUS has been applied to accelerate bone fracture healing.[8,9] In addition, it can stimulate the mandibular growth by effectively increasing the cartilaginous growth potential in the mandibular condylar cartilage. The U.S. Food and Drug Administration (FDA) has tested and approved the following parameters: a 1.5-MHz sine wave, repeated at 1 kHz at an intensity of 30 mw/cm^2, with a pulse width of 200 μs, delivered for 20 min a day.

Many animal studies have reported that significant results can be obtained using LIPUS therapy combined with functional appliances.[10,11] El-Bialy et al. evaluated the effectiveness of LIPUS therapy with functional appliances in treating patients with hemifacial microsomia (HFM).[12] They concluded that a daily application of LIPUS with hybrid bite-jumping appliances had significantly improved growth in the affected side of the patients' mandible.[12] Maurya et al. also studied the effects of LIPUS therapy with fixed functional appliances.[13] They concluded that LIPUS therapy positively affects the size of the joint space, improving the outcomes of functional treatment in growing patients with skeletal class II malocclusion.[13]

The purpose of this randomized controlled clinical trial (RCT) was to evaluate the changes resulting from the combined treatment with LIPUS and functional appliances (Twin-Block) in the correction of skeletal class II malocclusion.

Material and methods

Trial design

This study was designed as a three-pronged RCT with 1:1:1 allocation ratio. It was conducted at the Department of Orthodontics, Faculty of Dentistry of the University of Damascus in Syria, between March 2017 and November 2018. This trial was registered at the Clinical Trials.gov website on August 16, 2017 (identifier: NCT03251807). The local Research Ethics Committee approval was obtained. No serious harm to the patients was noticed.

Sample size calculation

The G*Power software, v. 3.1.3 (the Heinrich Heine University, Düsseldorf, Germany), available at http://www.psychologie.hhu.de/fileadmin/redaktion/Fakultaeten/Mathematisch-Naturwissenschaftliche_Fakultaet/Psychologie/AAP/gpower/GPowerWin_3.1.9.7.zip, was used to determine the sample size. A statistical power of 95% and α-error of 5% were assumed. The mean differences in the ANB angular measurements in the 3 groups were used based on the results of Baysal and Uysal,[14] and the one-way analysis of variance (ANOVA) test was applied. This indicated a sample size of 14 patients in each group. This study involved 15 patients in each group to compensate for the potential dropouts.

Eligibility criteria for participants, setting and location of data collection

The participants in the trial were selected randomly from the patients who arrived for consultation at the Department of Orthodontics, according to the following eligibility criteria: skeletal class II division 1 malocclusion with normal maxilla and retrognathic mandible (SNB < 78°), with the ANB values from 4° to 8°, and an overjet of 6–10 mm; patients at the pubertal growth spurt peak, which was assessed using hand-wrist radiographs according to the Fishman method of skeletal maturation.[15]

The exclusion criteria were the following: previous orthodontic treatment; systemic diseases that may affect the orthodontic treatment results; severe facial asymmetry; and poor oral hygiene.

The parents of the patients who met the inclusion criteria and were invited to participate were informed about the study; then, informed consent for each patient was obtained.

Randomization

The 45 enrolled patients were listed and a computer-generated randomization was applied to divide the patients into 3 equal groups. The distribution was concealed from the patient and the researcher until the time of intervention.

Blinding

Blinding of the patient or of the operator during intervention was not applied throughout the trial.

However, blinding during assessment was performed. The pre- and post-treatment cephalograms were saved and coded by an assistant (not involved in the study), so the researcher was unaware which group each radiograph belonged to when the records were evaluated.

Interventions

Forty-five patients aged 10.5–14 years were involved in the trial and randomly divided into 3 equal groups (2 treated and 1 control). The 1st group was the LIPUS group, receiving the combined treatment with LIPUS and a functional Twin-Block appliance. The 2nd group was the TB group; it received functional treatment with Twin-Block only. The 3rd group was an observational group (the control group), receiving no intervention.

Each patient in both treated groups had a single-step mandibular advancement and an edge-to-edge incisal relationship with a 2–3-mm bite opening between the central incisors. Twin-Block appliances with the conventional design according to Clark[16] consisted of 2 plates with a midline screw in the maxillary plate, which was turned once per week (0.2 mm) by the patient.

In the LIPUS group, gel was applied on the right and left temporomandibular joint (TMJ) regions to ensure LIPUS wave propagation, and the UltraCure PRO Plus™ device (EZUltrasound, Mobridge, USA) was used. The device generated 200-μs bursts of a 1-MHz sine wave with a repetition rate of 1 kHz and a temporal averaged intensity of 30 mw/cm^2. These parameters have been approved by FDA.[17]

The LIPUS stimulation was applied for 20 min daily for the first 21 days of the treatment and every 3 weeks during the active phase of functional treatment (Fig. 1). This treatment protocol has been used in a variety of human and animal models to stimulate bone fracture healing by inducing new vascularization as well as to promote growth and healing after distraction ostogenesis.[18,19] In addition, El-Bialy et al. in 2010 applied a similar LIPUS treatment protocol in their study on patients with HFM.[12]

All participants in the treated groups were instructed to wear their appliances at all times except at meal time, and were checked every 3 weeks until the end of the active phase of the functional therapy. The active phase was ended when the overjet was 0–1.5 mm and the occlusion settled into a class I or superclass I molar relationship.[14] The control group was monitored for 8 months.

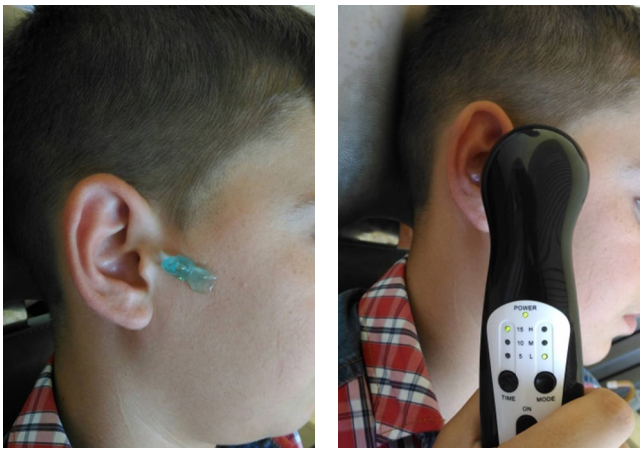

Fig. 1. Technique of applying the low-intensity pulsed ultrasound (LIPUS) device on the temporomandibular joint (TMJ) region

Cephalometric analysis

Lateral cephalometric radiographs were obtained before and at the end of the active phase of the treatment for the treated groups, and at the beginning and end of the observation period for the control group. All cephalometric radiographs were taken with the same machine, i.e., PaX-i3D (VATEH Co., Ltd., Hwaseong, Korea), with the same settings. The cephalograms were traced and analyzed using cephalometric software (Viewbox, v. 4.0.1.6; dHAL Software, Kifissia, Greece) by the same researcher, who was blinded to the patients' names during the measurement.

Outcomes

The skeletal and dentoalveolar changes after the treatment or observation were evaluated by measuring 11 angular variables and 5 linear variables (measured in millimeters) on the lateral cephalometric radiographs (Fig. 2). Ten randomly selected cephalograms were retraced and analyzed after 1 month to determine the method error. Reliability was evaluated using the intraclass correlation coefficient (ICC), which revealed a strong intra-examiner reliability (ICC = 0.992). The paired-sample *t*-test was applied to all cephalometric measurements to detect any systematic error. No statistically significant difference was found between the 2 measurements ($p > 0.05$).

Statistical analysis

The data was gathered and analyzed using the IBM SPSS Statistics for Windows, v. 20.0 software (IBM Corp., Armonk, USA). The χ^2 tests were used to assess differences in gender distribution between the groups. The Shapiro–Wilk normality test was used to ensure the normal distribution of data. The paired sample *t*-test was used to study the significance of differences between the pre- and post-treatment variables in each group, and to detect the intra-group changes after the treatment/observation; the level of significance was set at $p < 0.05$.

The one-way analysis of variance (ANOVA) and Tukey's multiple comparison post hoc test were used to assess the significance of differences between the 3 groups after the treatment/observation, and the level of significance was set at $p < 0.05$.

Results

Pre-treatment equivalence

Forty-five patients were randomized with a 1:1:1 allocation ratio into 3 groups (15 patients in each group). No patients were lost during the follow-up and a total of 45 patients were available for the statistical analysis (Fig. 3). There was no significant difference between the groups

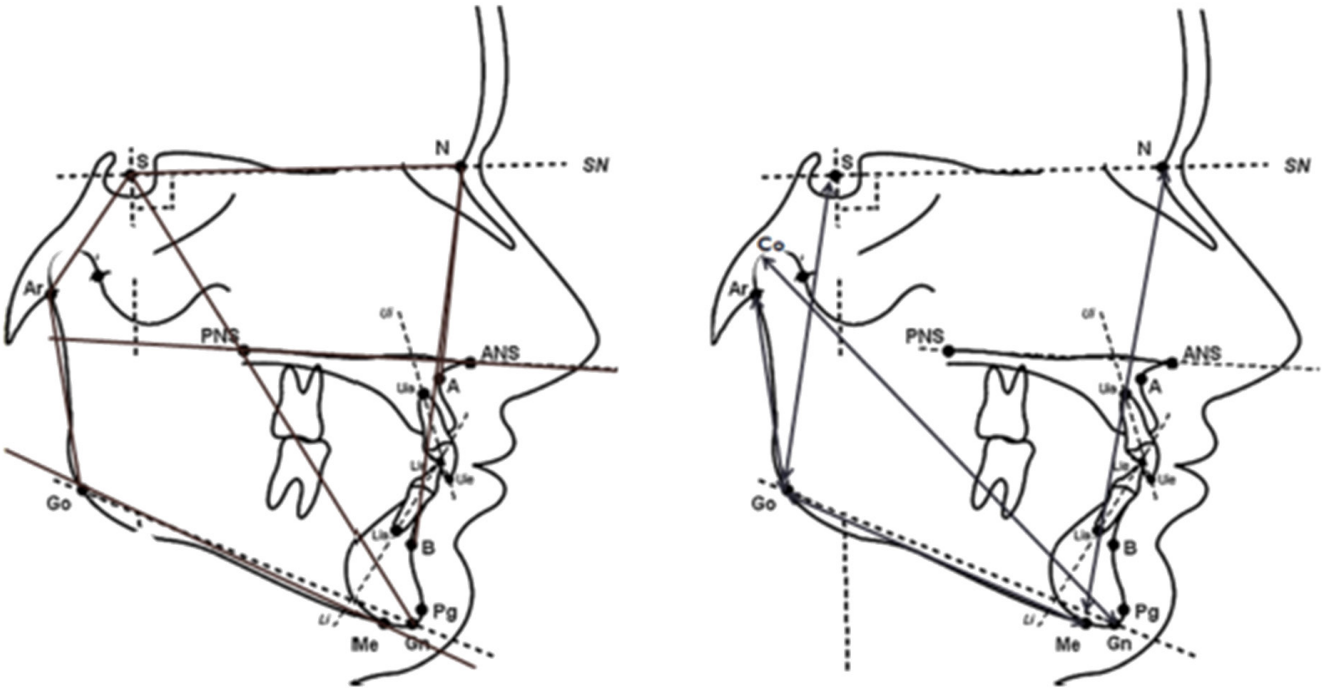

Fig. 2. Cephalometric measurements on the radiograms

A – A point; ANS – anterior nasal spine; Ar – articular; B – B point; Gn – gnathion; Go – gonion; Me – menton; N – nasion; Pg – pogonion; PNS – posterior nasal spine; S – sella.

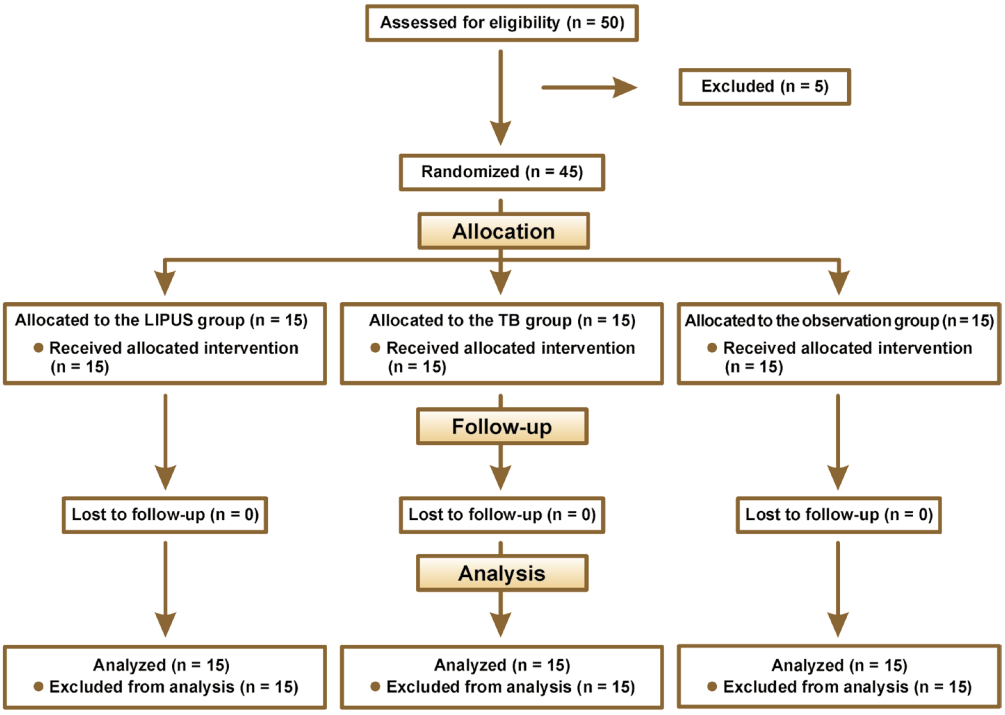

Fig. 3. CONSORT (Consolidated Standards of Reporting Trials) flow diagram of the patients' recruitment and follow-up

LIPUS – low-intensity pulsed ultrasound; TB – Twin-Block.

regarding gender distribution at the start of the study (the χ^2 tests with Yates's correction: $\chi^2 = 0.556$; $p = 0.757$) (Table 1). The ANOVA test indicated no statistically significant differences between the 3 groups for all the studied variables before the treatment ($p > 0.05$), with the exception of the Y-axis and the inclination of the maxillary incisors to the maxilla plane, the former being significantly larger in the control group than in the LIPUS and TB groups (Table 2).

Intra-group comparison

The changes after the treatment or observation period in each group are presented in Table 3.

Table 1. Gender distribution between the groups

Sex	LIPUS group	TB group	Control group	Total	χ^2	*p*-value
Males (n)	5	6	8	19		
Females (n)	10	9	7	26	0.556	0.757
Total (N)	15	15	15	45		

Table 2. Comparison of pre-treatment cephalometric variables

	Variables	LIPUS group		TB group		Control group		*p*-value	Multiple comparison		
		mean	*SD*	mean	*SD*	mean	*SD*		LIPUS/TB	LIPUS/control	TB/control
Angular variables [°]	SNA	81.40	2.23	81.78	1.98	80.92	2.66	0.924	NS	NS	NS
	SNB	74.98	2.08	74.97	1.52	75.01	2.02	0.644	NS	NS	NS
	ANB	6.47	0.69	6.81	0.96	5.93	0.85	0.074	NS	NS	NS
	NSAr	125.42	5.85	123.86	5.80	126.00	3.98	0.395	NS	NS	NS
	SArGo	140.88	4.30	143.13	3.30	142.05	3.70	0.620	NS	NS	NS
	ArGoMe	130.40	7.37	125.74	4.61	128.39	6.72	0.166	NS	NS	NS
	Total sum of Björk	396.70	5.05	395.97	3.53	398.55	5.22	0.627	NS	NS	NS
	Y-axis	60.49	3.18	61.06	2.82	62.47	2.18	0.008	NS	0.009	0.040
	MM	30.09	4.73	30.72	3.02	32.58	4.09	0.621	NS	NS	NS
	U1:Spp	111.87	4.41	111.32	4.98	114.26	1.80	0.020	NS	NS	0.020
	L1:GoMe	98.56	5.45	97.71	8.05	95.69	7.69	0.736	NS	NS	NS
Linear variables [mm]	Ramus height (Ar-Go)	48.10	6.42	50.42	5.48	50.73	5.38	0.763	NS	NS	NS
	Mandibular body (Go-Me)	85.69	8.50	82.33	5.98	79.86	6.70	0.776	NS	NS	NS
	Mandibular length (Co-Gn)	135.25	8.23	134.77	6.88	134.65	5.87	0.938	NS	NS	NS
	Wits	5.58	1.98	6.50	1.38	5.98	2.20	0.789	NS	NS	NS
	Overjet	7.23	0.96	7.33	1.57	7.60	1.57	0.395	NS	NS	NS

SD – standard deviation; MM – intermaxillary angle; U1 – upper central incisor; Spp – palatal plane; L1 – lower central incisor; Co – condylion; NS – non-significant.

Table 3. Comparison of the changes in the angular and linear variables between the 3 groups

	Variables	LIPUS group			TB group			Control group			Multiple comparison		
		mean	*SD*	*p*-value	mean	*SD*	*p*-value	mean	*SD*	*p*-value	LIPUS/TB	LIPUS/control	TB/control
Angular variables [°]	SNA	0.47	0.48	0.002	0.42	0.26	<0.001	0.12	0.09	NS	NS	0.004	0.021
	SNB	3.23	0.77	<0.001	3.09	0.61	<0.001	0.30	0.10	<0.001	NS	<0.001	<0.001
	ANB	−2.69	0.72	<0.001	−2.67	0.66	<0.001	−0.21	0.17	<0.001	NS	<0.001	<0.001
	NSAr	−1.96	0.58	<0.001	−1.66	0.88	<0.001	0.33	0.16	0.035	NS	<0.001	0.012
	SArGo	2.05	0.87	<0.001	1.91	1.00	<0.001	−0.20	0.47	NS	NS	<0.001	<0.001
	ArGoMe	1.18	1.28	NS	0.81	1.04	0.022	−0.15	0.46	0.037	0.021	NS	<0.001
	Total sum of Björk	1.27	1.99	0.014	−0.22	1.00	NS	0.02	0.09	NS	0.009	0.027	NS
	Y-axis	0.01	0.88	NS	−0.29	0.79	NS	0.43	0.32	NS	NS	NS	NS
	MM	0.53	1.60	0.044	0.21	0.54	NS	0.39	0.38	NS	0.010	0.020	NS
	U1:Spp	−2.60	1.68	<0.001	−3.22	0.95	<0.001	−0.27	0.22	<0.001	0.001	<0.001	<0.001
	L1:GoMe	2.74	1.23	0.033	3.30	1.19	<0.001	0.25	0.35	NS	NS	<0.001	<0.001
Linear variables [mm]	Ramus height (Ar-Go)	2.37	1.19	<0.001	2.25	0.85	<0.001	0.84	0.28	0.020	0.021	<0.001	<0.001
	Mandibular body (Go-Me)	1.55	1.62	<0.001	2.21	0.77	<0.001	1.03	0.45	NS	NS	0.0130	<0.001
	Mandibular length (Co-Gn)	4.09	1.21	0.001	3.54	0.79	<0.001	0.77	0.23	NS	0.002	<0.001	<0.001
	Wits	−4.83	1.21	<0.001	−5.43	1.22	<0.001	0.07	0.19	NS	NS	<0.001	<0.001
	Overjet	−5.94	0.74	<0.001	−5.27	1.33	<0.001	−0.06	0.40	0.001	NS	<0.001	<0.001

Treated groups (LIPUS and TB)

Similar, statistically significant changes were observed in the LIPUS and TB groups after the treatment.

There was a significant decrease in the value of ANB ($p < 0.001$), which was caused by a significant increase in SNB and the anterior displacement of TMJ, which was demonstrated by a significant decrease in NSAr ($p < 0.001$). Also, there was a significant increase in the mandibular length (Co-Gn; 4.09 mm for the LIPUS group and 3.54 mm for the TB group), whereas SNA showed a minimal increase in both groups.

There were minimal increases in the vertical skeletal relationships, as the vertical angles (total sum of Björk and MM) were increased in the LIPUS group, whereas no statistically significant changes were observed in the TB group.

As for the dentoalveolar changes, the lower incisors were significantly proclined ($p < 0.001$). Also, the upper incisors were significantly retruded (–2.60° in the LIPUS group and –3.22° in the TB group). The overjets were significantly decreased ($p < 0.001$).

Control group

There were minimal changes in the sagittal angular variables (0.30° for SNB and –0.21° for ANB). The posterior displacement of TMJ was observed through an increase in NSAr ($p = 0.035$). Changes in the vertical measurements and all linear measurements were not statistically significant, except the Ar-Go measurement, which showed a significant increase ($p = 0.020$). The lower incisor angles were not changed, whereas the upper incisors were minimally retruded (–0.27°) and the overjets were statistically significantly reduced (–0.06 mm) ($p < 0.001$).

Inter-group comparison

Multiple comparisons between the groups after the treatment or observation period are presented in Table 3.

Both treated groups showed a greater statistically significant increase in SNA, SNB and ANB as compared to the control group ($p < 0.05$), without any significant difference between the treated groups. A similar, statistically significant decrease was observed in NSAr ($p < 0.001$) in each treated group, whereas NSAr was significantly increased in the control group. Greater increases in the ramus height (Ar-Go) and mandibular length (Co-Gn) were observed in the LIPUS group as compared to the TB group ($p = 0.021$ and $p = 0.002$, respectively), and in both treated groups in comparison with the control group ($p < 0.001$). Changes in the mandibular body showed a greater significant increase in the treated groups as compared to the control group.

There were minimal increases in the vertical angles (total sum of Björk and MM) in the LIPUS group only. The differences in the Y-axis were not significant between the 3 groups. Upper incisor retroclination was significant in the TB group as compared to the LIPUS group and the control group ($p < 0.001$), whereas the lower incisors were more protruded in the treated groups than in the control group. Also, the overjets were significantly more decreased in the treated groups compared to the control group ($p < 0.001$), without any significant difference between the treated groups.

Discussion

Low-intensity pulsed ultrasound is one of the non-invasive approaches that is used to enhance bone growth and formation during functional treatment. This RCT was the first study to evaluate the effects of LIPUS combined with functional appliances on the stimulation of the mandibular growth in patients with class II malocclusion. The LIPUS device produces mechanical waves at a pulse frequency of 1 MHz with a pulse repetition frequency of 1 kHz. Daily treatment with LIPUS for 20 min at an intensity of 30 mw/cm^2 (according to FDA) has been found to stimulate bone healing. A daily direct application of LIPUS for 21 days has also been found to stimulate the mandibular bone growth in rats and in humans, especially when combined with functional appliances.[12,13]

In the current study, all patients were at the peak of the pubertal growth spurt to ensure the best effects of the treatment. The cephalometric changes were evaluated at the end of the active phase of functional treatment.

Sagittal skeletal changes

In both treated groups, SNA was minimally increased, but this increase was not clinically significant. It might be due to the fact that the upper incisor apex was forwarded in both groups and point A moved anteriorly as the result of alveolar bone reshaping; the sagittal growth of the maxilla did not occur. In their study, O'Brien et al. reported the restriction of the maxillary growth,[20] whereas other researchers did not.[21] In the control group, no significant changes were observed in SNA ($p = 0.452$).

A greater mandibular length increase in a shorter time was observed in the LIPUS group as compared to the TB group and the control group ($p < 0.001$). After 166 ±18.9 days of active functional treatment, an increase in the mandibular length (Co-Gn) was 4.09 mm in the LIPUS group, whereas it was 3.54 mm after 245 ±27.2 days in the TB group. The independent samples *t*-test showed a significant difference between the 2 groups ($p = 0.002$), which means that LIPUS significantly shortened the duration of active functional treatment. This result agrees with the study by El-Bialy et al. on the affected mandibles.[12]

The treated groups showed a significant increase in SNB as the result of point B moving to a more anterior

position (3.23° in the LIPUS group and 3.09° in the TB group), whereas the changes in the control group were not clinically significant. This was reported in several studies,[14,22] although the LIPUS group in the current study has shown a greater increase than in the case of Tümer and Gültan's study after 12–16 months of active functional treatment.[23]

The anterior movement of TMJ was obvious from a significant decrease in NSAr in both treated groups, and there was no significant difference between the treated groups. Although many previous studies have reported the anterior movement of TMJ as a significant finding after functional treatment,[24] many studies have neglected this variable.[14,25]

In both treated groups, ANB was decreased. This decrease had an effect on the skeletal discrepancy between the jaws. This result agreed with previous studies, which demonstrated the great role of functional appliances in improving the relationship between the maxilla and the mandible.[5,26]

Vertical skeletal changes

Minimal, statistically significant increases in the vertical measurements (total sum of Björk and MM) were observed in the LIPUS group only, which may possibly be related to the effects of the application of LIPUS. However, the TB group showed no significant changes in the vertical measurements. It appears that the posterior acrylic bite blocks of the Twin-Block appliance might be able to prevent any increases in the vertical dimension. Burhan and Nawaya reported a similar result.[22] In contrast, this result disagrees with the results provided by Giuntini et al. – the posterior acrylic bite blocks of the Twin-Block appliance in the latter study were trimmed only in patients with a deep bite.[27]

Conversely, the treated groups showed a statistically significantly greater elongation of the ramus height ($p < 0.001$) as compared to the control group. This result closely resembles that of Mills and McCulloch's study, where a 2.9-mm increase in the ramus height was reported.[21]

Dental changes

The dentoalveolar components demonstrated a significant inclination of the anterior teeth in both treated groups, whereas the dentoalveolar changes in the control group were not clinically significant. The upper incisors were also more retruded in the TB group (–3.22°) than in the LIPUS group (–2.60°). This result proved that the correction of class II malocclusion depending on dentoalveolar changes was greater in the LIPUS group than in the TB group.

Many studies have mentioned a significant lower incisor proclination during functional treatment.[22,27] The retrusion of the upper incisors is also a consistent finding in many previous studies.[21,28]

These dentoalveolar changes significantly contributed to the correction of the overjet. The overjet showed a greater correction in both treated groups as compared to the control group. These results represent a desirable consequence of the treatment of skeletal class II malocclusion, and were achieved by the combination of dentoalveolar and skeletal changes which occurred in the treated groups.[22,29]

Conclusions

Based on the results of the current study, it can be concluded that the combined treatment with LIPUS and functional appliances is of great effectiveness in correcting class II malocclusion in growing individuals. It accelerates the growth of the mandible and improves the skeletal discrepancy. Therefore, LIPUS could shorten the duration of functional treatment.

A limitation of this study is a lack of cone-beam computed tomography (CBCT) imaging, which can help in measuring the TMJ component dimensions and mandibular dimensions more accurately. However, it is not ethical to order CBCT to patients in an observation group, as no treatment is given to them.

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Effects of different light sources on tooth shade selection

Wpływ różnych źródeł światła na wybór odcienia koloru zęba

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

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Abstract

Background. Differences in lighting conditions can make matching the shade of a single porcelain tooth to the natural adjacent teeth very challenging.

Objectives. The purpose of the study was to examine visual shade selection under 3 different light sources – natural daylight, a dental operating light and a color-corrected light.

Material and methods. The visual assessment was based on a comparison between a shade guide and the target tooth. One observer with normal color vision was asked to visually match the color of the maxillary central right incisor and the maxillary right canine in a group of 100 subjects, aged 22–40 years. Two shade guides were used – VITA Vitapan Classical® and VITA 3D Master® – in natural daylight as well as under an operating light and a handheld light (Demetron Shade Light®). The VITA Easyshade® spectrophotometer was used to evaluate the results of the visual shade selection method.

Results. Significant differences were found between the effects of using the shade guide systems ($p < 0.05$) and light sources ($p < 0.05$). Overall, the use of the Vitapan Classical shade guide significantly improved the correlation between visual matching and the spectrophotometer readings.

Conclusions. Within the limitations of this study, the Vitapan Classical shade guide demonstrated superior agreement in shade selection as compared to the 3D Master shade guide. The Demetron Shade Light was proven to be a useful device for the color matching of artificial teeth in prosthodontic treatment. Visual tooth color matching is a subjective method and it produces the best results under a color-matching light.

Key words: color perception, light sources, shade guides, spectrophotometer, tooth color

Słowa kluczowe: postrzeganie kolorów, źródła światła, wzorniki kolorów, spektrofotometr, kolor zęba

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Introduction

Correct color evaluation and shade matching in prosthetic restorations is an important aspect of prosthetic treatment, which largely affects patient satisfaction.[1] Tooth color is most often assessed visually with ready-made shade guides provided by the manufacturers of dental products.[2] However, the method is subjective, as color perception is affected by several parameters, e.g., shade, saturation, brightness, translucency, opalescence, reflectance, and fluorescence,[3] as well as by the individual differences of the human eye and brain.[4] Color perceived by the human eye may be distorted by lighting conditions, gingiva color and the colors of the surrounding environment.[5] In order to improve the esthetic outcome of the treatment and choose the best tooth shade for the patient, a growing number of devices have been made available to assist dentists in optimizing the process of color evaluation and shade selection. Such aids for qualitative and quantitative color evaluation include digital cameras, colorimeters and spectrophotometers. Other noteworthy products include lamps specially designed for color matching.

Factors affecting artificial tooth shade matching include the source of light, the object observed and the observer.[4] Three sources of light are available in a dental surgery: natural daylight, which is highly variable; the operating light of a dental unit, with a bias toward the red region of the visible spectrum as compared with natural sunlight; and finally, fluorescent ceiling lights, which – unlike incandescent lights – have various color-rendering properties depending on the specified color temperature.[6] The optimal conditions for tooth shade matching are provided by a light having a color temperature between 5,500 K and 6,500 K, and the Color Rendering Index (CRI) greater than 90. When matching the shade of artificial teeth in prosthetics, one should perform the assessment in the presence of only 1 light source, as overlapping illumination from different sources may promote metamerism.[6]

The color of the natural teeth depends on the optical properties of the enamel and dentin. When light reaches the tooth surface, the following phenomena may be observed: light transmission; reflection; dispersion; and absorption.[2] Translucency, opacity, opalescence, surface gloss, and fluorescence are secondary features to shade, saturation and brightness affecting the appearance of the teeth.[3] The first 2 features are considered the most relevant, and they depend on the spectral distribution and quantity of the reflected light.[7] The teeth are most translucent at the incisal edges, decreasing toward the center, with the lowest translucency in the cervical area.[8] The enamel of the natural tooth is responsible for opalescence and its translucency causes the scintillation of short-wavelength light, resulting in blue-grey reflexes, easily noticeable at the incisal edge.[9] Opalescence promotes the rainbow effect, and depends on the direction and location of the lighting around the tooth as well as on the dispersion, diffraction and interference of light waves.[3] The shade of the teeth is mainly due to the color of the dentin.[10] After penetrating the enamel, UV rays reach the dentin and cause fluorescence, ranging from intense white to light blue color.[9] When surrounded by a cofferdam for 20 min, the dry teeth appear whiter, but regain their natural color 15 min after the removal of the cofferdam.[11] Similarly, the teeth appear whiter for half an hour after impressions are taken with polyvinyl siloxane (PVS).[11] Therefore, it is very important to match the shade of the teeth prior to performing any procedures that would dry them.[12] Furthermore, the morphology of the tooth surface affects the amount and color of the reflected light; an uneven or rough surface results in a greater dispersion of light as compared to a smooth surface. Thus, the amount of the light reflected from the surface of the enamel increases immediately after the teeth are brushed.[13] Moreover, the effect of contrast creates optical illusions and affects the perceived brightness of the teeth. The surrounding colors, such as the skin tone, the color of the eyes, adjacent teeth, lips, and gingivae, will also change the perception of tooth color.[5] Darker surroundings will cause the teeth to look brighter. When assessing the shade, a bright background should not be placed behind the observed teeth, as the dark oral cavity absorbs light.[14] It is also important to remember that the teeth observed in close proximity appear bigger, and therefore brighter. Prosthodontists color match the teeth in a more precise manner than general dental practitioners. Education and training in color matching affect the selection of the optimal shade.[15]

The purpose of this study was to examine visual shade selection under 3 different light sources – natural daylight, operating light and corrected light.

Material and methods

One hundred participants (22 males and 78 females) were recruited for this study. The age of the patients ranged from 22 to 40 years (mean age: 25.11 ±3.24 years). Each participant was provided with a full explanation of the study and the procedures to be followed. The participants' written informed consent was obtained before they were enrolled in the study.

The study was approved by the Ethics Committee of Poznan University of Medical Sciences in Poland.

The visual assessment was based on a comparison between 2 shade guides and the target tooth. One observer with normal color vision was asked to visually match the color of the maxillary central right incisor and the maxillary right canine in a group of 100 subjects.

Two shade guides were used – VITA Vitapan Classical® and VITA 3D Master® (VITA Zahnfabrik H. Rauter GmbH & Co. KG, Bad Säckingen, Germany). The assessment was performed under 3 lighting conditions: in natural daylight – all measurements were carried out on 1 dental chair located by the window facing north, on a partly cloudy day before noon; using the operating light KAVOLUX® 1410C (Kaltenbach & Voigt GmbH & Co. KG, Biberach an der Riß, Germany); and using a handheld light specially designed for color matching in dentistry – Demetron Shade Light® (Kerr Corporation, Orange, USA). The results were then re-evaluated with the use of the VITA Easyshade® spectrophotometer (VITA Zahnfabrik H. Rauter GmbH & Co. KG).

The inclusion criteria required: live, healthy maxillary central right incisors and maxillary right canines with no cracks, fillings and/or discolorations. The exclusion criteria comprised the following: dead teeth with discolorations, fillings and after whitening procedures.

The statistical analysis was performed with Statistica PL, v. 12.0 (StatSoft Polska Sp. z o.o., Cracow, Poland). All comparisons between daylight, the operating light, the Demetron light, the Vitapan Classical shade guide, and the 3D Master guide were carried out with a test for differences between 2 structure indicators. The results were deemed statistically significant at $p < 0.05$.

Results

The agreement differed significantly between the types of lighting conditions. Under the Demetron light, an agreement of 69% was obtained for the maxillary right central incisors with respect to the Vitapan Classical guide and 33% when using the 3D Master guide; these scores were significantly higher ($p < 0.05$) than those obtained with daylight and the operating light for both shade guides. The use of the Demetron light also resulted in the highest agreement for the canines – 73% in the case of the Vitapan Classical guide and 33% for the 3D Master guide. These results are shown in Fig. 1.

A significant difference was evident between the Vitapan Classical and 3D Master shade guides in incisor color matching compatibility according to the dentist's assessment and the Easyshade spectrophotometer measurements ($p < 0.05$), with the 3D Master shade guide showing a statistically significantly lower agreement. The results of incisor color matching compatibility for the assessments made by the dentist under 3 different lighting conditions and the selection based on the Easyshade spectrophotometer for the Vitapan Classical and 3D Master shade guides are presented in Table 1.

A comparative analysis demonstrated a significant difference between the Vitapan Classical and 3D Master shade guides in canine color matching compatibility when the measurements were performed by the dentist in daylight and with the Easyshade spectrophotometer ($p < 0.05$). A significant difference was found between the Vitapan Classical and 3D Master shade guides in canine color matching compatibility when the measurements were performed by the dentist under the operating light and with the Easyshade spectrophotometer ($p < 0.05$). The 3D Master guide showed a poorer agreement in canine color measurements carried out by the dentist with the Demetron light and by means of the Easyshade spectrophotometer ($p < 0.05$). These results are summarized in Table 2.

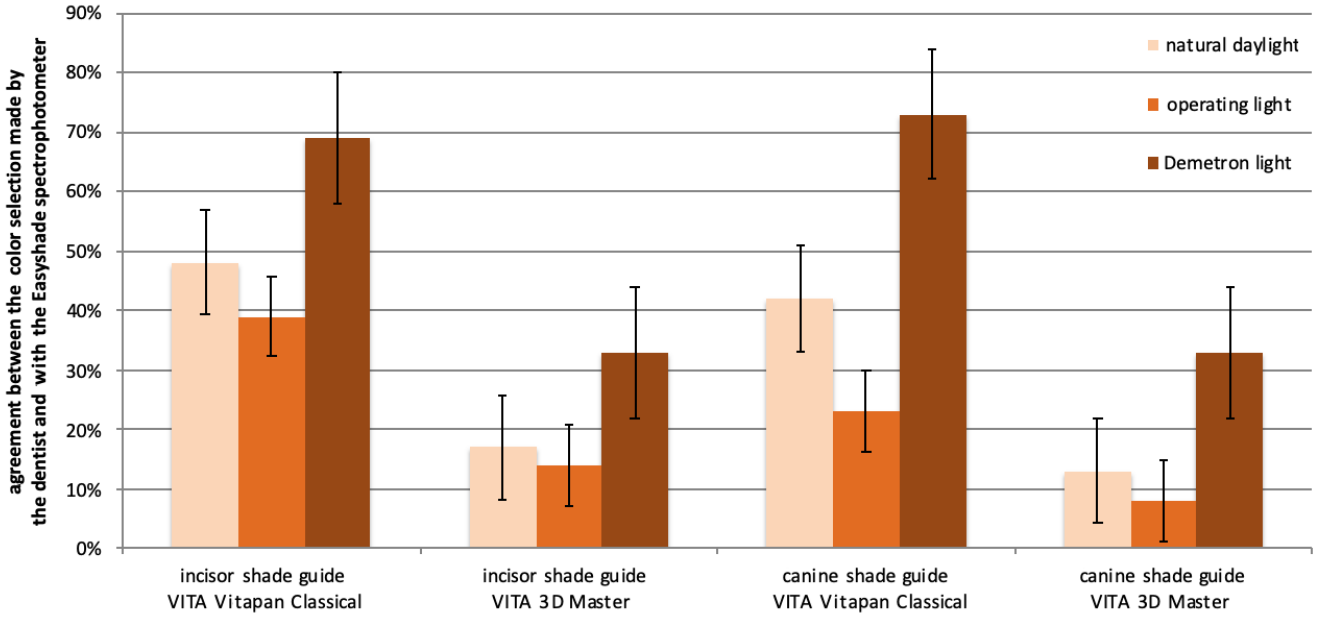

Fig. 1. Percentage of the compatibility of the colors chosen by the dentist and with the Easyshade device under different light sources
Data presented as mean ± standard deviation (*SD*).

Table 1. Compatibility of the Easyshade measurements with the dentist's selection under different lighting conditions for the incisors

Compatibility	Daylight	Operating light	Demetron	*p*-value		
				daylight vs operating light	daylight vs Demetron	operating light vs Demetron
Agreement of Easyshade Vitapan Classical with Vitapan Classical [%] (N = 100)	48	39	69	0.1993	0.0002	0.0001
Agreement of Easyshade 3D Master with 3D Master [%] (N = 100)	14	17	33	0.5578	0.0015	0.0090
p-value	0.0001	0.0005	0.0001	–	–	–

Table 2. Compatibility of the Easyshade measurements with the dentist's selection under different lighting conditions for the canines

Compatibility	Daylight	Operating light	Demetron	*p*-value		
				daylight vs operating light	daylight vs Demetron	operating light vs Demetron
Agreement of Easyshade Vitapan Classical with Vitapan Classical [%] (N = 100)	42	23	73	0.0041	0.0001	0.0001
Agreement of Easyshade 3D Master with 3D Master [%] (N = 100)	13	8	33	0.2488	0.0003	0.0001
p-value	0.0001	0.0005	0.0001	–	–	–

Discussion

The visual method of artificial tooth shade matching using a standard shade guide is the most commonly used method in dentistry.[16] The human eye can successfully distinguish even slight differences in color.[17] However, the visual assessment of tooth color is considered imperfect.[18] Variations have been observed in the shades selected by different observers as well as in the case of a single observer matching the shade for the same tooth on different occasions.[19]

Shade matching should be performed in adequate lighting conditions; however, it is difficult to ensure them in everyday dental practice. Our research demonstrated inconsistencies in the tooth shade selection made by a single dentist. The same incisal shade according to the Vitapan Classical shade guide examined under 3 different light sources was selected in only 29 cases in a sample of 100 patients and in nearly 50 cases (47% of the study population) when the 3D Master shade guide was used. Canine shade matching compatibility was also low, accounting for only 30% for both shade guides. The research confirmed the thesis that the visual method of artificial tooth color matching is inconsistent. The results demonstrated a significant influence of illumination on color perception. The color temperature of natural daylight ranges from 1,000 K to 20,000 K[20]; therefore, such a light source may not be adequate for tooth color matching.[4] A surgery operating light may also affect color perception.[6] However, lighting systems dedicated to tooth color evaluation promote accurate shade selection when using a guide.[21] The Demetron Shade Light used to illuminate the teeth emits white light, which simulates the northern hemisphere daylight of 6,500 K. This color temperature is the most suitable for color evaluation according to a previous study.[22] The Demetron light is held at a distance of 5–7.5 cm from the inspected teeth, with the shade guide nearby. Observations are made through a specially designed, small window. Our research determined that during subjective evaluation, the highest compatibility of the color assessment for incisors was obtained in daylight with the Demetron light, whereas the lowest was found when daylight was combined with the operating light using the Vitapan Classical shade guide. The 3D Master shade guide also demonstrated good compatibility in daylight with the Demetron light. However, no statistically significant difference was found in the case of other compatibility measurements with respect to color matching dependence on the light source used. Similar results were obtained when both shade guides were compared in daylight and using the Demetron light, with 59% and 63% compatibility for the Vitapan Classical guide and the 3D Master guide, respectively. Observations made in the current study suggest that dentists tend to choose brighter samples from a guide when using an operating light. When the subjective and objective tooth color matching methods were compared, the colors chosen by the dentist using the Demetron light showed the highest compatibility. In the case of the incisors, color matching compatibility occurred in

69% of cases using the Vitapan Classical shade guide and in 33% of cases using the 3D Master guide. Likewise, in the case of the canines, it accounted for 73% of the measurements made with the Vitaplan Classical guide and 33% when using the 3D Master guide. A study which compared the effects of 2 light sources – daylight and the color-corrected Demetron light – on tooth color matching compatibility, performed by dentistry students, found a greater compatibility using the Demetron light as compared to daylight only.[23] The advantage of using this light source in tooth color matching carried out by students was also observed in later studies.[24] The device, emitting light of 5,500–6,500 K and 1,000 Lx, and CRI exceeding 90, has optimized lighting conditions; however, the task of selecting tooth color continues to be the responsibility of the prosthodontist and depends on factors like the observer's age, gender, vision defects, experience, fatigue, and emotional state.[15] The dentist's knowledge of shade selection plays an important role in ensuring accurate measurements. Research by other authors indicates a need to improve the qualifications of the dental care team through courses and training covering this topic.[21,23] For a number of years studies have been underway to develop an ideal, objective measuring device, which would eliminate all the abovementioned flaws of shade selection. Our research showed a greater compatibility in the color assessment of the incisors and canines when using the Vitapan Classical shade guide as compared to the 3D Master shade guide. Most reported studies indicated a higher compatibility of the measurements carried out with the Vitapan Classical shade guide with the use of different spectrophotometric and colorimetric devices as compared to the 3D Master shade guide.[19,25]

The 3D Master shade guide was introduced into the market in the 1990s and was supposed to cover all of the natural tooth shades. The system is recommended by the American Dental Association and considered by some authors as the optimal shade guide for assessing tooth color, with fewer errors in the spatial distribution of colors as compared to the Vitapan Classical shade guide.[26,27] Lee et al. decided to verify the differences in the distribution of brightness and saturation between the adjacent samples of the 3D Master shade guide with a spectroradiometer.[28] The authors believed that the differences between the individual guide colors were irregular.[28] The manufacturer of the 3D Master guide recommends a 3-step methodology when matching tooth color. First, a sample is chosen from one of the 5 brightness groups. Next, saturation is selected, and finally a shade is chosen. However, previous studies demonstrated that samples of a single group did not belong to the same brightness group at all.[29] Our research indicated that it was often the case that during color matching, 2 samples from 2 different brightness groups were most similar to the natural tooth shade, which may have resulted in low compatibility between the selected colors and the spectrophotometer readings. Additionally, a lower compatibility of the dentist's choice and the selection made with the Easyshade spectrophotometer for the 3D Master shade guide may be due to the intermediate tones, which are not included among the 26 guide samples, whereas the spectrophotometer does not encounter this problem.[28] Apart from that, the compatibility rate may significantly differ with respect to the color-measuring devices and the type of teeth evaluated.[30] Regardless of lighting conditions, the compatibility of the Easyshade device measurements with the selection made by the dentist was always higher in the case of the Vitapan Classical shade guide for both the incisors and canines.

Conclusions

This study showed that the VITA Vitapan Classical system demonstrated superior compatibility in shade selection as compared to the VITA 3D Master system. Visual tooth color matching is a subjective method, dependent on lighting conditions. The Demetron Shade Light was found to be a useful device for the color matching of artificial teeth in prosthodontic treatment.

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Evaluation of osteoarthritic changes in the temporomandibular joint and their correlations with age: A retrospective CBCT study

Ocena zmian kostno-stawowych w stawie skroniowo-żuchwowym i ich związków z wiekiem – retrospektywne badanie CBCT

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

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Abstract

Background. Cone-beam computed tomography (CBCT) enables the radiographic examination and evaluation of osseous changes in the temporomandibular joint (TMJ).

Objectives. The aim of this study was to determine the prevalence of various bony changes in TMJ in patients from a wide age range as well as to evaluate the CBCT findings of TMJ osteoarthritis (OA) and correlate them with age.

Material and methods. The study included the CBCT images of 150 patients (43 males and 107 females) who were refferred to the Department of Dentomaxillofacial Radiology for the evaluation of TMJs. Each TMJ was evaluated separately for the presence of any osseous changes in the condylar head or articular fossa/eminence, and for joint space narrowing.

Results. The mean age of the sample was 37.26 years (range: 10–90 years). A total of 101 (67.3%) patients presented 1 or more osseous changes. No significant differences were found between the right and left TMJs concerning the prevalence rates of osseous changes. Significant differences were found in the mean ages with regard to the absence and presence of the following findings: condylar erosion, osteophytes, loose bodies, erosion in the articular fossa, and joint space narrowing.

Conclusions. Degenerative changes in TMJ may reflect an age-related bone remodeling process. Older patients may have more common findings of OA, such as condylar and articular erosion, osteophytes, loose joint bodies, and joint space narrowing.

Key words: osteoarthritis, cone-beam computed tomography, temporomandibular joint

Słowa kluczowe: choroba zwyrodnieniowa stawów, komputerowa tomografia stożkowa, staw skroniowo-żuchwowy

Introduction

Osteoarthritis (OA) is a term that describes an age-related non-inflammatory disease starting with changes within the bone and spreading over the tissues of the joint to the subsequent degeneration of articular surfaces.[1] This condition is sometimes called degenerative joint disease and involves a remodeling process that aims to adapt the structure of the temporomandibular joint (TMJ) to mechanical forces to ensure normal functioning.[2] Excessive or continuing forces lead to the progressive degeneration of the bony articular surfaces of TMJ, which eventually results in the radiographic findings of OA, such as flattening, the formation of osteophytes and loose bodies, erosion, a reduced joint space, subcortical sclerosis, and cysts.[3] It should be noted that the remodeling of TMJ may be detected in the absence of symptoms and clinical signs. Severe osseous changes and the presence of clinical symptoms, such as pain or dysfunction, indicate the advanced stages of TMJ OA.[4]

Cone-beam computed tomography (CBCT) provides detailed 3-dimensional (3D) images for the diagnosis of degenerative changes in TMJ, at the same time ensuring a lower radiation dose, a shorter exposure time, a higher spatial resolution, and a lower cost as compared to computed tomography (CT).[5,6] Although it is well-acknowledged that CBCT provides information about the osseous components of TMJ, it also images in detail the structures of TMJ, thereby contributing to the early detection and staging of OA as well as to the monitoring of changes in the disease over time.[7] The Research Diagnostic Criteria for Temporomandibular Disorders (RDC/TMD) – a standardized protocol to diagnose TMD – was proposed in 1992 and has been developed to provide the image analysis for OA.[2,8]

The aim of this study was to determine the prevalence of various bony changes in TMJ in patients from a wide range of age as well as to evaluate the CBCT findings of TMJ OA and correlate them with age.

Material and methods

This retrospective study was approved by the Research Ethics Board of Hacettepe University in Ankara, Turkey (GO 19/02). The CBCT images of 150 patients (43 males and 107 females) referred to the Department of Dentomaxillofacial Radiology, Faculty of Dentistry of Hacettepe University for the evaluation of TMJs between June 2017 and May 2018 were included in the study.

The CBCT images were obtained using the i-CAT™ Next Generation device (Imaging Sciences International, Hatfield, USA) with a tube voltage of 120 kV, a tube current of 5 mA and a voxel size of 0.2 mm. All examinations were performed in a maximum intercuspation position. A series of axial views of 1-millimeter thickness was automatically produced following the reconstruction of the raw data. The TMJs were evaluated on the TMJ screen of the CBCT software from the lateral images perpendicular to the long axis of the condyle and coronal images parallel to the long axis of the condyle.[9] The slice thickness and the distance between slices were 1 mm. The presence of an osseous change was confirmed when it was detected in at least 2 consecutive slices.[10]

The mean age of the sample was 37.26 years (range: 10–90 years). Table 1 shows the distribution of the sample according to age and gender. A total number of 300 TMJs were evaluated. Age, gender and osseous changes were recorded for each patient. After excluding CBCT scans obtained for the imaging of initial injuries or the post-treatment evaluation of TMJ and inflammatory arthritis, the presence or absence of the following characteristics were evaluated based on RDC/TMD[2]:

– bony changes in the condylar head:
  - flattening – the loss of the convex form of the articular surface,
  - erosion – the loss of continuity in the cortical bone margin,
  - osteophytes – marginal hypertrophy with sclerotic borders and the exophytic angular formation of the osseous tissue arising from the surface,
  - loose joint bodies – calcified structures that are not continuous with the disk or osseous structures of the joint,
  - subcortical sclerosis – an increase in the thickness of the cortical plate,
  - a subchondral cyst – a cavity below the articular surface that deviates from the normal marrow pattern,
  - condylar hypoplasia or hyperplasia – the size of the condyle is small or large from all dimensions, but its morphology is normal, and
  - condylar bifidity[2,11];
– bony changes in the articular fossa:
  - erosion,
  - sclerosis,
  - resorption, and
  - the flattening of the articular eminence[9,11];
– joint space narrowing – a reduction in space (<1.5 mm) in all directions (anterior, superior and posterior).[10]

Table 1. Distribution of the sample according to age and gender

Gender	Age [years]						Total (n)	Percentage [%]
	10–19	20–29	30–39	40–49	50–59	60+		
Male (n)	8	11	6	3	9	6	43	28.7
Female (n)	19	27	19	17	16	9	107	71.3
Total (n)	27	38	25	20	25	15	150	100.0

Data analysis

The IBM SPSS Statistics for Windows software, v. 23.0 (IBM Corp., Armonk, USA) was used for the data analysis and statistical procedures. The descriptive statistical data was recorded, and the independent samples *t*-test was conducted to assess the relationship between age and the percentages of degenerative bony changes. The prevalence rates of bony changes in the right and left sides were compared by means of McNemar's test. A *p*-value <0.05 was considered statistically significant.

Results

Out of the 150 patients included in this study, 49 patients (32.7%) had no radiographic changes in the condyle, articular fossa/eminence or joint space, whereas 101 patients (67.3%) did. The examplary images of osseous changes in the TMJs are shown in Fig. 1.

Table 2 shows the comparison of osseous changes with regard to the right and left TMJs. The results of McNemar's test showed that the prevalence rates among the 2 sides were not significantly different ($p = 0.442$).

Table 3 shows the frequency distribution of osseous changes according to patients and TMJs. Of the 300 TMJs, 108 (36%) showed condylar flattening, 39 (13%) showed condylar erosion, 30 (10%) showed condylar osteophytes, 28 (9.3%) showed condylar sclerosis, 8 (2.7%) showed subchondral cysts, 25 (8.3%) showed condylar hypoplasia or hyperplasia, and 3 (1%) showed condylar bifidity.

Table 2. Comparison of osseous changes with regard to the right and left TMJs

McNemar's test		Left TMJs		
		with bone changes	without bone changes	total
Right TMJs	with bone changes	74	11	85
	without bone changes	16	49	65
	total	90	60	150

Table 3. Frequency distribution of osseous changes according to patients and TMJs

Osseous changes	Number of patients (n)*	Percentage of patients [%]	Number of TMJs (n)**	Percentage of TMJs [%]
Condylar flattening	71	47.3	108	36.0
Condylar erosion	30	20.0	39	13.0
Condylar osteophytes	26	17.3	30	10.0
Condylar sclerosis	24	16.0	28	9.3
Subchondral cyst	7	4.7	8	2.7
Condylar hypoplasia/ hyperplasia	18	12.0	25	8.3
Bifid condyle	3	2.0	3	1.0
Flattening of the articular eminence	14	9.3	16	5.3
Sclerosis of the articular fossa	3	2.0	3	1.0
Erosion in the articular fossa	13	8.7	17	5.7
Joint space narrowing	25	16.7	35	11.7
Loose bodies	6	4.0	7	2.3

* out of N = 150; ** out of N = 300.

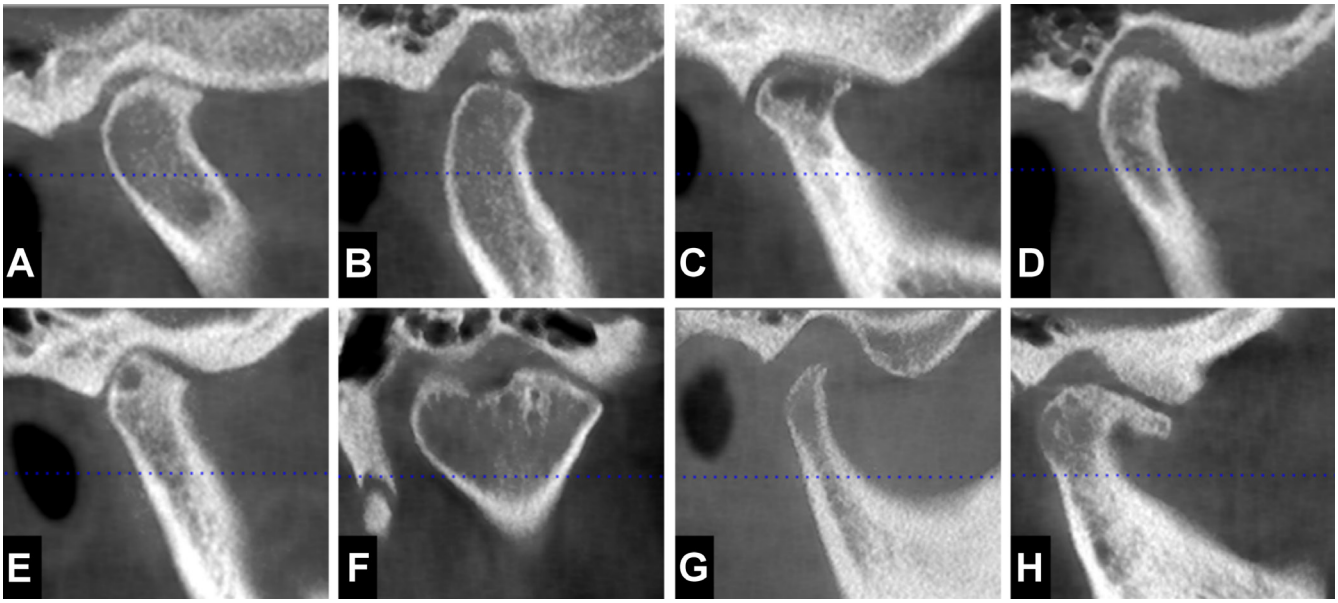

Fig. 1. Examples of the CBCT images of osseous changes in the temporomandibular joints (TMJs)

A – condylar flattening, subcortical sclerosis and joint space narrowing in a female aged 53 years; B – loose calcified body in a male aged 62 years; C – erosion in the condylar head and the sclerosis of the articular fossa in a female aged 58 years; D – osteophyte formation of the condyle in a female aged 45 years; E – subchondral cyst and the flattening of articular surfaces in a female aged 50 years; F – bifid condyle in a male aged 48 years; G – condylar hypoplasia in a female aged 20 years; H – osteophyte formation of the condyle, the sclerosis of the articular fossa and the flattening of the articular eminence in a male aged 69 years.

The flattening of the articular eminence was observed in 16 (5.3%) TMJs. Three (1%) joints showed the sclerosis of the articular fossa and 17 (5.7%) showed erosion in the articular fossa. Joint space narrowing was observed in 35 (11.7%) TMJs and loose bodies were observed in 7 (2.3%) joints.

Table 4 shows the frequency of osseous changes in the condyle and the respective mean ages. There were significant differences in the mean ages with regard to the absence and presence of condylar erosion ($p = 0.001$), osteophytes ($p = 0.001$) and loose bodies ($p = 0.005$). No differences were found with respect to condylar flattening, condylar sclerosis or subchondral cysts.

Table 5 shows the frequency of changes related to the articular fossa, the articular eminence and joint space, and the respective mean ages. Significant differences were found in the mean ages with regard to the absence and presence of erosion in the articular fossa ($p = 0.006$) and joint space narrowing ($p = 0.017$). No differences were found with respect to the flattening of the articular eminence or the sclerosis of the articular fossa.

Table 4. Frequency of osseous changes in the condyle and the respective mean ages

Osseous changes	Number of TMJs (n)	Percentage of TMJs [%]	Age [years]	*p*-value
Condylar flattening				
present	108	36.0	38.95 ±17.09	0.212
absent	192	64.0	36.27 ±18.21	
Condylar erosion				
present	39	13.0	45.87 ±18.81	0.001
absent	261	87.0	35.95 ±17.35	
Condylar osteophytes				
present	30	10.0	49.00 ±16.53	0.001
absent	270	90.0	35.93 ±17.51	
Condylar sclerosis				
present	28	9.3	41.28 ±18.70	0.157
absent	272	90.7	36.36 ±17.37	
Subchondral cyst				
present	8	2.7	45.00 ±7.40	0.212
absent	292	97.3	37.02 ±17.99	
Loose bodies				
present	7	2.3	55.57 ±20.63	0.005
absent	293	97.7	36.80 ±17.56	

Data concerning age presented as mean ± standard deviation (*SD*).

Table 5. Frequency of changes in the articular eminence, articular fossa and joint space, and the respective mean ages

Osseous changes	Number of TMJs (n)	Percentage of TMJs [%]	Age [years]	*p*-value
Flattening of the articular eminence				
present	16	5.3	45.31 ±17.46	0.062
absent	284	94.7	36.78 ±17.77	
Sclerosis of the articular fossa				
present	3	1.0	45.33 ±10.59	0.430
absent	297	99.0	37.15 ±17.88	
Erosion in the articular fossa				
present	17	5.7	49.38 ±15.88	0.006
absent	283	94.3	37.07 ±17.96	
Space narrowing				
present	35	11.7	43.97 ±16.12	0.017
absent	265	88.3	36.35 ±17.88	

Data concerning age presented as mean ±*SD*.

Discussion

Cone-beam computed tomography imaging has been widely applied in the temporomandibular region, and has been proven to have superior diagnostic reliability in the detection of osseous abnormalities in TMJ as compared to plain radiography and conventional tomography.[12,13] Cone-beam computed tomography is found to be comparable to multi-slice CT in the detection of morphological changes in TMJ, and consequently is preferable to CT, as it comes with a lower radiation dose and high diagnostic reliability in evaluating the bone structures of TMJ. However, CBCT does not display Hounsfield units (HU) as in medical CT,[14] and is more sensitive to artefacts caused by motion and metal objects.[5,6] Despite the superior features of CBCT imaging in the detection of bone alterations, diagnostic information is limited to osseous morphology, cortical bone integrity and subcortical osseous changes. Magnetic resonance imaging (MRI) is indicated when soft-tissue pathology or an inflammatory condition is suspected.[4,7]

Prior studies on CBCT evaluating the effect of the voxel size on the detection of osseous defects in TMJ have demonstrated that images reconstructed with a smaller voxel size (≤0.2 mm) have a higher spatial resolution and adequate diagnostic accuracy.[15,16]

The detection of degenerative changes in TMJ is substantial in both symptomatic and asymptomatic patients. Radiographic findings give important information regarding the presence and severity of bone alterations in TMJ. Since some of these alterations may be part of an age-related remodeling process or may comprise a physiological response, they can occur in asymptomatic patients. Hence, it is not always possible to correlate the radiographic data with the clinical status.[11,17] The CBCT scans included in this study were taken with the aim of imaging TMJs and were retrieved from the computer database. Consequently, the CBCT assessments did not take into account the patients' clinical status.

The present study was designed to evaluate the prevalence and imaging characteristics of bony changes in TMJ, and intended to identify the relationship between each bony change of OA and age. The radiographic evaluation of osseous changes was made based on the RDC/TMD image analysis criteria.[2] Of the 150 patients who were referred for the evaluation of TMJs, 101 (26 males and 75 females) had the findings of TMJ OA. Some studies have reported that women are more likely to develop OA, which is mainly

attributed to the hormonal differences between men and women.[9,18,19] The potential role of sex hormones has been suggested in articular bone resorption and degeneration.[20]

The prevalence of osseous changes in TMJ detected in this study (67.3%) was similar to the results reported by dos Anjos Pontual et al. (71%) and Chang et al. (73%), which may be due to the fact that all the patients included in the sample were referred for the CBCT examination with the initial diagnosis of a TMJ disease.[18,21]

The patients with changes of TMJ OA ranged in age from 10 to 82 years, with a mean age of 38.9 years, which was significantly higher as compared to those without any findings (mean age: 33.8 years; range: 10–90 years; $p = 0.02$). This result indicates that OA is common even in children and young adults, and is in agreement with the findings of other authors.[18,22,23] Some researchers have reported an association between an increasing age and the prevalence of TMJ OA,[9,18] whereas others have found no such correlation.[24,25]

In the present study, bony changes in the condyle were more frequently detected than changes in the articular eminence and articular fossa. The most frequent finding in this study was condylar flattening, which is mostly consistent with previous studies.[9,18,19,24] However, this finding is incompatible with the results reported by Massilla Mani and Sivasubramanian, who found erosion as the predominant finding,[26] and by Nah, who found sclerosis to be the most common finding.[27] The discrepancy between these results may be attributed to gender and age differences, racial/ethnic disparity and the diagnostic criteria for OA.

The current findings, showing no significant mean age differences with regard to the absence and presence of condylar flattening, may suggest that this bony change might be a physiological response rather than a sign of age-related degeneration.

As degeneration progresses into the bony structures of TMJ, changes in bone surfaces occur, and the progress eventually results in bone erosion. The present results demonstrated that condylar erosion is more prevalent in older patients.[10]

In this study, the mean age of the patients with TMJ osteophytes was significantly higher as compared to the patients without osteophytes, which is in accordance with previous studies, reporting a higher prevalence of TMJ osteophytes at older age.[9,28]

At the advanced stage of TMJ OA, the structural deterioration of the subchondral bone leads to the thickening of the subchondral bone plate and an increase in bone density becomes evident. This elevated bone density is called subcortical sclerosis and has been strongly associated with the formation of subchondral cysts.[29] No significant differences were found in the mean ages with regard to either the absence and presence of subcortical sclerosis or the absence and presence of subchondral cysts in this study.

Although loose joint bodies were amongst the less frequent condylar findings in this study, a significant difference was noted between the mean ages of the patients with and without loose bodies. Additionally, all cases with loose bodies were associated with other findings of condylar OA, such as erosion, sclerosis, osteophytes, and cyst formation. According to Ahmad et al., the presence of loose bodies may be considered as an indicator of OA in cases of the concomitant presence of other features of TMJ OA.[2]

The flattening of the articular eminence and erosion in the articular fossa were the most common temporal bone changes of OA in this study. The mean age of the patients with erosion in the articular fossa was significantly higher as compared to the patients with a normal articular fossa. However, no difference was found for the flattening of the articular eminence, although the p-value was close to being significant ($p = 0.062$).

The sclerosis of the articular fossa was the least common radiographic finding in this study (1%). The present results are consistent with the studies in which the mean ages of the patients were similar with regard to the absence and presence of the sclerosis of the articular fossa.[24,25]

The RDC/TMD criteria suggest the evaluation of the joint space in a maximum intercuspation position of the condyle, because the joint space can change during mastication or mouth opening.[2] The narrowing of the joint space was one of the common alterations in this study, which coexisted with other signs of OA in all cases. The current findings suggest that joint space narrowing in all directions may be a feature of OA.

Conclusions

In conclusion, degenerative changes in TMJ may reflect an age-related bone remodeling process. Older patients may have more common findings of OA, such as condylar and articular erosion, osteophytes, loose joint bodies, and joint space narrowing.

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Effectiveness of low-level laser therapy in accelerating the orthodontic tooth movement: A systematic review and meta-analysis

Skuteczność terapii laserem małej mocy w przyspieszaniu ortodontycznych przesunięć zębowych – systematyczny przegląd piśmiennictwa i metaanaliza

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Abstract

Objectives. The aim of the paper was to appraise the current evidence of the effectiveness of low-level laser therapy (LLLT) in accelerating the tooth movement.

Methods. A comprehensive search was performed in 9 databases up to June 2019. Only randomized controlled trials (RCTs) were included. The risk of bias was assessed using the Cochrane Collaboration tool. The quantitative data synthesis was attainable only for the studies evaluating the effect of laser on canine retraction; the qualitative description was used for the rest of the studies. The overall quality of evidence was rated using the Grading of Recommendations Assessment, Development and Evaluation (GRADE) criteria.

Results. A total of 25 RCTs were included in this review. The radiated upper canines showed a greater retraction – 0.50 mm and 0.49 mm at months 2 and 3, respectively. The radiated lower canines showed a greater retraction – 0.28 mm and 0.52 mm at months 2 and 3, respectively. No statistically significant differences were observed among the upper and lower canines after the 1st month of retraction. When the GRADE approach was utilized, the overall quality of evidence limited confidence in the estimates. The qualitative description revealed enhanced tooth movement when LLLT was applied. The attrition bias was the main risk factor affecting the methodology of the studies.

Conclusions. Low-level laser therapy can speed up the rate of the tooth movement. However, the overall quality of evidence ranged from low to very low and the clinical significance of the obtained statistically significant differences is questionable. Hence, more precise studies are needed. As discussed in this review, it is highly recommended to express and compare the laser dosage with the total number of joules applied per month rather than the previously used J/cm^2. Moreover, the previous recommendation indicating that lower energy densities (2.5, 5 and 8 J/cm^2) are more effective than 20 and 25 J/cm^2 is misleading.

Key words: orthodontics, acceleration, lasers, low-level laser therapy

Słowa kluczowe: ortodoncja, przyspieszenie, lasery, terapia laserem małej mocy

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Introduction

The primary objective of orthodontic treatment is usually to achieve an optimal occlusion with minimal complications within a relatively short period of time.[1] According to a recent systematic review of 22 studies involving 1,089 participants, the mean duration of comprehensive orthodontic treatment with a fixed appliance is 19.9 months.[2] This long treatment duration is associated with an increased risk of developing white spots, caries, gingivitis, and root resorption.[3] Therefore, accelerating the tooth movement, which leads to a reduction in the duration and complications of the treatment, is desirable for both patients and orthodontists.

Over the last decade, numerous studies have been conducted to investigate the efficacy of different interventions in speeding up the tooth movement. One of these interventions is low-level laser therapy (LLLT). Initially, histological research showed that LLLT contributes to inducing remodeling processes in the alveolar bone by increasing the numbers of osteoblasts and osteoclasts.[4,5] In consequence, an exponential growth in the number of studies conducted to investigate the effectiveness of laser treatment in accelerating the tooth movement has become apparent. Interestingly, LLLT enjoys high patient acceptability and can be easily utilized, especially with the availability of small portable devices.[6]

Electronic literature searches in the PubMed and Scopus databases have yielded some systematic reviews that also concern this issue. Ge et al. demonstrated that LLLT might speed up the tooth movement, adding that relatively low energy densities (2.5, 5 and 8 J/cm^2) are more effective than 20 or 25 J/cm^2 and higher.[7] de Almeida et al. concluded that there was no evidence showing that the use of laser therapy can accelerate the induced tooth movement.[8] Imani et al. found that LLLT could increase the rate of the orthodontic movement.[9] Those differing conclusions in previous publications could be ascribed to variations in review methodology, the number of studies included and the publication dates. Hence, the objective of this review was to provide an updated assessment of the current scientific evidence concerning the efficacy of utilizing LLLT in the acceleration of the tooth movement.

Methods

Research question

The research question of whether or not LLLT accelerates the tooth movement was defined according to the PICOS format:

- Participants: Healthy male and female patients (at least 10 patients in each study) at any age and of any ethnic group who underwent orthodontic treatment;
- Interventions: Orthodontic treatment assisted by LLLT in order to accelerate the tooth movement;
- Comparisons: Orthodontic treatment without laser therapy;
- Outcomes: The rate of tooth movement (millimeters of tooth movement per time period) or any equivalent measurement indicating the efficacy of the intervention used;
- Study design: Only randomized controlled trials (RCTs) were included in this review in order to minimize confounding factors and to reduce bias.

Search strategy

A comprehensive electronic search was performed independently and in duplicate by the authors, with no limitations on language, year or publication status, from the inception to June 2019 in the following databases: the Cochrane Central Register of Controlled Trials (CENTRAL), Scopus, Trip, CINAHL via EBSCO, PubMed, OpenGrey (for grey literature), and ProQuest (for dissertations and theses). The bibliographies of the included studies and relevant reviews were screened for possible further studies. Ongoing trials were also checked through the World Health Organization (WHO) International Clinical Trials Registry Platform (ICTRP) Search Portal and ClinicalTrials.gov. More details on the electronic search strategy can be found in Supplementary Table 1.

Study selection and data extraction

The 2 authors independently assessed the studies identified during the search. Initially, a screening process was carried out by assessing titles and abstracts to identify potentially relevant articles. Then, the full-text copies of potentially relevant studies were assessed and subjected to the eligibility criteria. At this stage, to avoid any conflict of interest or any possible bias, a blinding was performed by masking the authors' and the journals' names. Any disagreements between the 2 reviewers were resolved through discussion and consensus.

Finally, information was extracted from the studies, including the authors' names, setting, PICOS data, follow-up period, and main findings. When doubtful information was found, the corresponding authors of the studies were contacted for clarification.

Assessing the risk of bias of the included studies

The risk of bias of the included studies was assessed independently by the 2 authors using the Cochrane Collaboration tool for assessing the risk of bias. The following fields were described as having a high, low or unclear risk of bias: random sequence generation (selection bias); allocation concealment (selection bias); blinding of participants and personnel (performance bias); blinding of outcome assessment (detection bias); incomplete outcome

Supplementary Table 1. Electronic search strategy

No.	Database	Search strategy	Results
1	**Cochrane Central Register of Controlled Trials (CENTRAL)** from the inception up to June 2019, with no limits	#1 orthodontic* OR 'dental movement' OR 'tooth movement' OR 'orthodontic tooth movement' #2 accelerat* OR expedit* OR speed* OR fast* OR rapid* OR velocity OR duration OR rate OR time #3 laser OR 'laser therapy' OR 'low level laser' OR 'low level laser therapy' OR LLL OR LLLT OR 'low powered laser' OR 'low intensity laser' OR 'low level light therapy' OR photobiomodulation OR photobiostimulation OR biomodulation OR biostimulation OR irradiat* OR phototherapy OR 'light emitting diode' OR LED OR diode #4 #1 AND #2 AND #3	#4 = 261
2	**Scopus** from the inception up to June 2019, with no limits	#1 TITLE-ABS-KEY (orthodontic* OR 'dental movement' OR 'tooth movement' OR 'orthodontic tooth movement') #2 TITLE-ABS-KEY (accelerat* OR expedit* OR speed* OR fast* OR rapid* OR velocity OR duration OR rate OR time) #3 TITLE-ABS-KEY (laser OR 'laser therapy' OR 'low level laser' OR 'low level laser therapy' OR LLL OR LLLT OR 'low powered laser' OR 'low intensity laser' OR 'low level light therapy' OR photobiomodulation OR photobiostimulation OR biomodulation OR biostimulation OR irradiat* OR phototherapy OR 'light emitting diode' OR LED OR diode) #4 #1 AND #2 AND #3	#4 = 923
3	**Trip** from the inception up to June 2019, with no limits	(orthodontic* OR 'dental movement' OR 'tooth movement' OR 'orthodontic tooth movement') AND (accelerat* OR expedit* OR speed* OR fast* OR rapid* OR velocity OR duration OR rate OR time) AND (laser OR 'laser therapy' OR 'low level laser' OR 'low level laser therapy' OR LLL OR LLLT OR 'low powered laser' OR 'low intensity laser' OR 'low level light therapy' OR photobiomodulation OR photobiostimulation OR biomodulation OR biostimulation OR irradiat* OR phototherapy OR 'light emitting diode' OR LED OR diode)	76
4	**CINAHL via EBSCO** TX: All Text; from the inception up to June 2019, with no limits	TX (orthodontic* OR 'dental movement' OR 'tooth movement' OR 'orthodontic tooth movement') AND TX (accelerat* OR expedit* OR speed* OR fast* OR rapid* OR velocity OR duration OR rate OR time) AND TX (laser OR 'laser therapy' OR 'low level laser' OR 'low level laser therapy' OR LLL OR LLLT OR 'low powered laser' OR 'low intensity laser' OR 'low level light therapy' OR photobiomodulation OR photobiostimulation OR biomodulation OR biostimulation OR irradiat* OR phototherapy OR 'light emitting diode' OR LED OR diode)	214
5	**PubMed** All Fields; from the inception up to June 2019, with no limits	#1 orthodontic* OR 'dental movement' OR 'tooth movement' OR 'orthodontic tooth movement' #2 accelerat* OR expedit* OR speed* OR fast* OR rapid* OR velocity OR duration OR rate OR time #3 laser OR 'laser therapy' OR 'low level laser' OR 'low level laser therapy' OR LLL OR LLLT OR 'low powered laser' OR 'low intensity laser' OR 'low level light therapy' OR photobiomodulation OR photobiostimulation OR biomodulation OR biostimulation OR irradiat* OR phototherapy OR 'light emitting diode' OR LED OR diode #4 #1 AND #2 AND #3	#4 = 492
6	**OpenGrey** up to June 2019	#1 (acceleration OR accelerating OR accelerated OR rapid OR fast) AND (orthodontic OR tooth movement) #2 (orthodontic OR tooth movement) AND (laser OR phototherapy OR photobiomodulation)	8 #1 = 4 #2 = 4
7	**ProQuest** up to June 2019	ab(orthodontic OR 'dental movement' OR 'tooth movement' OR 'orthodontic tooth movement') AND ab(accelerate OR acceleration OR accelerating OR accelerated OR speed OR fast OR rapid OR velocity OR duration OR rate OR time) AND ab(laser OR 'laser therapy' OR 'low level laser' OR 'low level laser therapy' OR LLL OR LLLT OR 'low powered laser' OR 'low intensity laser' OR 'low level light therapy' OR photobiomodulation OR photobiostimulation OR biomodulation OR biostimulation OR phototherapy OR 'light emitting diode' OR LED OR diode)	49
8	**World Health Organization (WHO) International Clinical Trials Registry Platform (ICTRP) Search Portal** advanced search (Title) up to June 2019	(orthodontic OR 'dental movement' OR 'tooth movement' OR 'orthodontic tooth movement') AND (laser OR photobiomodulation)	12
9	**ClinicalTrials.gov** up to June 2019	(orthodontic OR 'dental movement' OR 'tooth movement' OR 'orthodontic tooth movement') AND (laser OR photobiomodulation)	22

data (attrition bias); selective outcome reporting (reporting bias); and other sources of bias. Then, an overall risk of bias for each trial included was reported according to the following criteria:

– when all fields were assessed as having a low risk of bias, a low risk of bias was reported;
– when 1 or more fields were assessed as having an unclear risk of bias, a moderate risk of bias was reported;
– when 1 or more fields were assessed as being at high risk of bias, a high risk of bias was reported.

The judgments of both reviewers were compared and any disagreements were discussed until a consensus was reached.

Data synthesis

The data was pooled using the Review Manager (RevMan) v. 5.3 software (the Nordic Cochrane Centre, Copenhagen, Denmark). The inverse variance method with the random effect analysis and mean differences (MDs) with associated 95% confidence intervals (CIs) were chosen as the analysis methods. The amount of variability among the pooled studies was evaluated by applying the χ^2 test and calculating the I^2 index for heterogeneity. The publication bias was assessed visually using funnel plots for outcomes that were evaluated by 10 trials or more. Finally, the Grading of Recommendations Assessment, Development and Evaluation (GRADE) guidelines were used to rate the overall quality of evidence.

Results

Literature flow

A total number of 2,057 references were identified in the electronic search. Duplicates and articles that were beyond the scope of the defined question and PICOS were eliminated. As a result, 57 references were potentially relevant, and were therefore checked in depth. The final results included 25 completed RCTs and 10 ongoing RCTs. Figure 1 shows the detailed search process (PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) flow chart). The list of the studies excluded after the full-text assessment, with the reasons for exclusion, is provided in Supplementary Table 2.

Description of the studies

Twenty-five completed RCTs, including a total of 570 patients, were conducted to investigate the effects of LLLT on the tooth movement. Of the completed studies included, 2 were theses and 3 were reported in a language other than English (2 in Portuguese and 1 in Persian). Different types of tooth movement were described, including leveling and alignment, canine retraction, and en-masse retraction. Table 1 summarizes the characteristics of the completed studies included, whereas Supplementary Table 3 summarizes the characteristics of the ongoing studies.

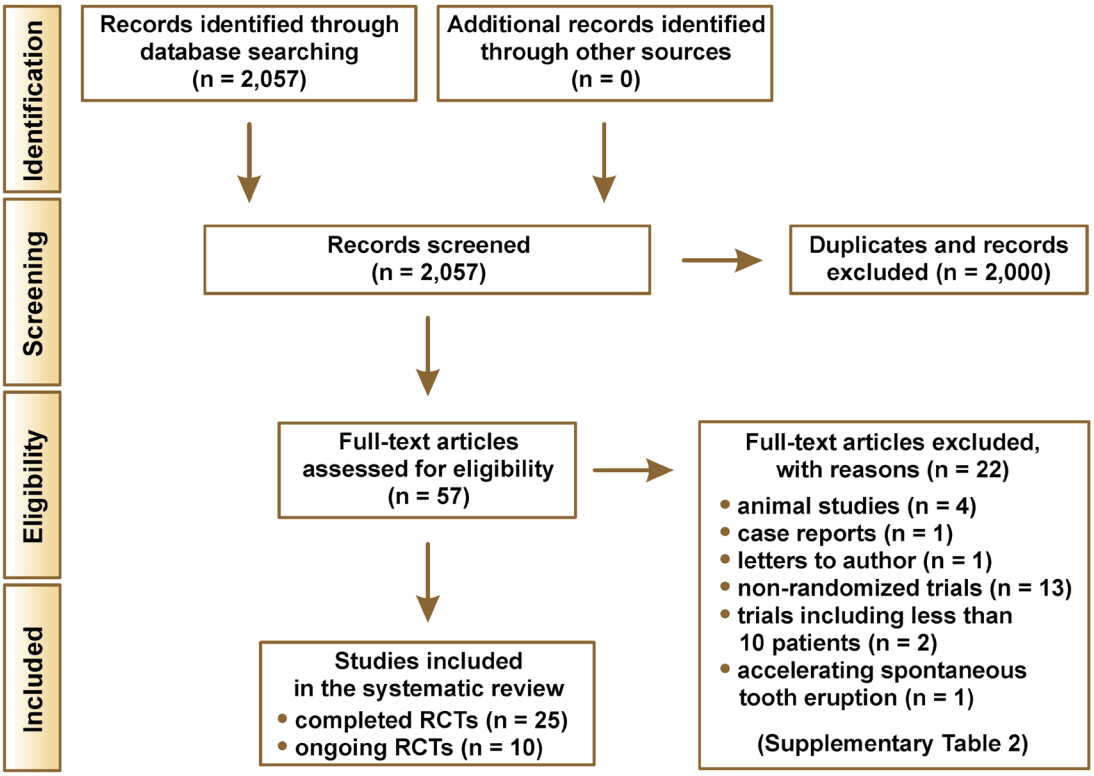

Fig. 1. PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) flow chart
RCT – randomized controlled trial.

Supplementary Table 2. Studies excluded and the reasons for exclusion

No.	Study	Reason for exclusion
1	Genc G, Kocadereli I, Tasar F, Kilinc K, El S, Sarkarati B. Effect of low-level laser therapy (LLLT) on orthodontic tooth movement. *Lasers Med Sci.* 2013;28(1):41–47.	non-randomized controlled trial
2	Domínguez A, Gómez C, Palma JC. Effects of low-level laser therapy on orthodontics: Rate of tooth movement, pain, and release of RANKL and OPG in GCF. *Lasers Med Sci.* 2015;30(2):915–923.	non-randomized controlled trial
3	Camacho AD, Cujar SAV. Acceleration effect of orthodontic movement by application of low-intensity laser. *J Oral Laser Appl.* 2010;10:99–105.	non-randomized controlled trial
4	Youssef M, Ashkar S, Hamade E, Gutknecht N, Lampert F, Mir M. The effect of low-level laser therapy during orthodontic movement: A preliminary study. *Lasers Med Sci.* 2008;23(1):27–33.	non-randomized controlled trial
5	Xu CW, Zhang ZJ, Zhao J, Cao G. The effect of low energy laser on accelerating orthodontic tooth movement. *Med J Qilu.* 2006;1:45–46.	non-randomized controlled trial
6	Shaughnessy T, Kantarci A, Kau CH, Skrenes D, Skrenes S, Ma D. Intraoral photobiomodulation-induced orthodontic tooth alignment: A preliminary study. *BMC Oral Health.* 2016;16:3.	non-randomized controlled trial
7	Gui L, Qu H. Clinical application of low energy laser in acceleration of orthodontic tooth movement. *Journal of Dalian Medical University*. 2008;30:155–156.	non-randomized controlled trial
8	Kuznetsova M, Zueva SM, Gunenkova IV, Ezhova EE, Ozerova EM. The use of the Optodan laser physiotherapeutic apparatus for the prevention of complications and the acceleration of the time in treating anomalies in the position of individual teeth with fixed orthodontic appliances [in Russian]. *Stomatologiia (Mosk).* 1998;77(3):56–60.	accelerating tooth eruption
9	Altan BA, Sokucu O, Toker H, Sumer Z. The effects of low-level laser therapy on orthodontic tooth movement: Metrical and immunological investigation. *JSM Dent.* 2014;2(4):1040.	non-randomized controlled trial
10	Matarese G, Matarese M, Picciolo G, Fiorillo L, Isola G. Evaluation of low-level laser therapy with diode laser for the enhancement of the orthodontic tooth movement: A split-mouth study. *Preprints.* 2018:2018090273.	non-randomized controlled trial
11	Monea A, Mo M, Pop D, Bersescu G. The effect of low level laser therapy on orthodontic tooth movement. *J Optoelectron Adv M.* 2015;9(1–2):286–289.	non-randomized controlled trial
12	Mc Quattie Pimentel IC. The effect of light accelerated therapy for orthodontic tooth movement: A prospective split-mouth clinical trial. 2017. (Order No. 10259785). Available from ProQuest Dissertations & Theses Global (1894849499).	only 5 patients included
13	Chung SE, Tompson B, Gong SG. The effect of light emitting diode phototherapy on rate of orthodontic tooth movement: A split mouth, controlled clinical trial. *J Orthod.* 2015;42(4):274–283.	less than 10 patients included
14	NCT03202355	non-randomized controlled trial
15	ACTRN12610001067066	non-randomized controlled trial
16	Fernandes MRU, Suzuki SS, Suzuki H, Martinez E, Garcez AS. Photobiomodulation increases intrusion tooth movement and modulates IL-6, IL-8 and IL-1β expression during orthodontically bone remodeling. *J Biophotonics.* 2019;12(10):e201800311.	non-randomized controlled trial
17	Isola G, Ferlito S, Rapisarda E. Low-level laser therapy increases interleukin-1β in gingival crevicular fluid and enhances the rate of orthodontic tooth movement. *Am J Orthod Dentofacial Orthop.* 2019;155(4):456–457.	letter to author
18	Yang H, Liu J, Yang K. Comparative study of 660 and 830 nm photobiomodulation in promoting orthodontic tooth movement. *Photobiomodul Photomed Laser Surg.* 2019;37(6):349–355.	animal study
19	Cordeiro JM, Sahad MG, Cavalcanti MFXB, et al. Laser photobiomodulation over teeth subjected to orthodontic movement. *Photomed Laser Surg.* 2018;36(12):647–652.	animal study
20	Ojima K, Dan C, Watanabe H, Kumagai Y. Upper molar distalization with Invisalign treatment accelerated by photobiomodulation. *J Clin Orthod.* 2018;52(12):675–683.	case report
21	Hsu LF, Tsai MH, Shih AH,et al. 970 nm low-level laser affects bone metabolism in orthodontic tooth movement. *J Photochem Photobiol B.* 2018;186:41–50.	animal study
22	Narmada IB, Rubianto M, Putra ST. The role of low-intensity biostimulation laser therapy in transforming growth factor β1, bone alkaline phosphatase and osteocalcin expression during orthodontic tooth movement in *Cavia porcellus*. *Eur J Dent.* 2019;13(1):102–107.	animal study

Effects of interventions

The included studies were grouped according to the type of tooth movement achieved and the region of LLLT application (i.e., intra-orally or extra-orally). The vast majority of the included studies used J/cm^2 to express the dosage used. However, the current recommendations of the World Association for photobiomoduLation Therapy (WALT) suggest reporting the low-level laser (LLL) dosage in terms of the total number of joules applied (J, total energy). Accordingly, we calculated the total number of joules applied per month for each study, using the formula:

$$\text{total number of joules} = \text{power (total watts applied)} \times \text{time (total seconds of application)}$$

per month (Table 1). However, it was unattainable to subcategorize the studies according to the total number of joules applied per month due to a wide variety of doses used.

Table 1. Characteristics of the studies included: PICOS, follow-up period and main findings

No.	Study/Setting	Participants	Intervention	Laser application schedule	Total energy dose per month	Comparison	Outcomes	Study design	Follow-up	Main findings
1	Varella et al. 2018[13] India	10 (M + F) patients U 1st premolars extraction mean age: 17.7 years	– U canine retraction – conventional brackets + NiTi closed-coil springs – GaAlAs laser 940 nm, 100 mW, 8 J/cm^2	5 pts B, 5 pts Ln 10 s each pt for 3 consecutive days at the beginning of canine retraction, 4 weeks later and 8 weeks later	30 J	OT + LLLT vs OT + sham	– RTM – IL-1ß secretion	RCT SMD	8 weeks	– LLLT accelerates OTM – an increased level of IL-1ß in the LLLT group
2	Samara et al. 2018[28] UAE	60 patients U 1st premolars extraction mean age: 20.4 years	– en-masse retraction – conventional brackets + NiTi closed-coil springs – OrthoPulse® device 850 nm, 33 mW/cm^2, 6 J/cm^2	3 min per arch daily	178.2 J per arch	OT + LLLT vs OT	RTM	RCT parallel	till complete space closure	LLLT accelerates OTM
3	Arumughan et al. 2018[29] India	12 patients 1st premolars extraction age:17–35 years	– U canine retraction and en-masse retraction distributed equally – conventional brackets + NiTi closed-coil springs – GaAlAs laser 810 nm, 100 mW	5 pts B, 5 pts Ln 10 s each pt every 3 weeks	10 J/ 3 weeks	OT + LLLT vs OT	RTM	RCT SMD	84 days	LLLT accelerates OTM
4	Al-Okla et al. 2018[12] UAE	38 (M + F) patients U crowding >4 mm without extraction age: 12–40 years	– leveling and alignment – conventional brackets – OrthoPulse device, 850 nm, 65 mW, 0.065 J/cm^2	5 min per arch daily	585 J per arch	OT + LLLT vs OT + sham	– RTM – root resorption	RCT parallel	6 months	– LLLT accelerates OTM – root length at a 6-month interval was shorter in the LLLT group
5	Guram et al. 2018[14] India	20 (M + F) patients 1st premolars extraction mean age: 20.5 years	– U + L canine retraction – conventional brackets + sectional closing loops – GaAlAs laser 810 nm, 0.2 W, 5 J/cm^2, 2 Hz, continuous mode	4 pts B, 4 pts Ln on the canine 10 s each pt weekly for 21 days	48 J/ 3 weeks	OT + LLLT vs OT + sham	– RTM – pain experience	RCT SMD	till complete canine retraction	LLLT accelerates OTM and reduces pain experience
6	Qamruddin et al. 2017[15] Pakistan	22 (M + F) patients 1st premolars extraction mean age: 19.8 ±3.1 years	– U canine retraction – self-ligating brackets + NiTi closed-coil springs – GaAlAs laser 940 nm, 100 mW, 7.5 J/cm2, continuous mode	5 pts B, 5 pts Ln on the U canine 3 s each pt on days 0, 21 and 42	3 J/ 3 weeks	OT + LLLT vs OT + sham	– RTM – pain experience	RCT SMD	9 weeks	LLLT accelerates OTM and reduces pain experience
7	Caccianiga et al. 2017[11] Italy	36 (M + F) patients L mild crowding mean age: 16.9 years	– alignment – self-ligating brackets – diode laser 980 nm, 1 W, 150 J/cm^2 per session, continuous mode	the mandible divided into 6 dental segments, each segment containing 2 teeth: (36,35) (46,45) irradiated for 9 s, the remaining segments for 8 s; it was repeated 3 times at intervals of 2 min between the sessions a single monthly administration	150 J	OT + LLLT vs OT	treatment duration	RCT parallel	till complete crowding resolution	treatment duration was shorter in the LLLT group

No.	Study/Setting	Participants	Intervention	Laser application schedule	Total energy dose per month	Comparison	Outcomes	Study design	Follow-up	Main findings
8	AlSayed Hasan et al. 2017[10] Syria	26 (M + F) patients U mild crowding with extraction mean age: 20 years	– leveling and alignment, – conventional brackets, – GaAlAs laser 830 nm, 150 mW, 2.25 J/cm^2, continuous mode	2 pts B, 2 pts Ln on each root of the 6 maxillary anterior teeth 30 s each pt the 1st month: on days 0, 3, 7, and 14, the 2nd month: every 15 days	72 J/tooth for the 1st month, then 36 J/tooth	OT + LLLT vs OT	– time for alignment – alignment improvement percentage	RCT parallel	till complete crowding resolution	LLLT accelerates OTM
9	Kochar et al. 2017[16] India	20 (M + F) patients 1st premolars extraction mean age: 20 years	– U + L canine retraction – conventional brackets + NiTi closed-coil springs – GaAlAs laser 810 nm, 100 mW, 5 J/cm^2, continuous mode	5 pts B, 5 pts Ln on the canine 10 s each pt days: 0, 3 and 7 of force application	30 J	OT + LLLT vs OT + sham	– RTM – pain experience	RCT SMD	till complete canine retraction	LLLT accelerates OTM and reduces pain experience
10	Üretürk et al. 2017[17] Turkey	15 (M + F) patients U 1st premolars extraction mean age: 16.20 ±1.32 years	– U canine retraction – self-ligating brackets + NiTi closed-coil springs – GaAlAs laser 820 nm,20 mW, 5 J/cm^2, continuous mode	5 pts B, 5 pts Ln on the U canine 10 s each pt, 0.2 J/pt on days 0, 3 ,7, 14, 21, 30, 33, 37, 44, 51, 60, 63, 67, 74, 81, 84, and 90	10 J	OT + LLLT vs OT + sham	– RTM – the IL-1ß and TGF-ß1 levels in GCF – periodontal indices	RCT SMD	90 days	LLLT accelerates OTM, increases the IL-1ß and TGF-ß1 levels with no sign of gingival inflammation
11	Nahas et al. 2017[30] UAE	40 (M + F) patients L anterior crowding mean age: 21.8 years	– leveling and alignment – self-ligating brackets – extra-oral LED device (OrthoPulse) 850 nm, 90 mW/cm^2, 108 J/cm^2 on the surface of the cheek	20 min daily	3,240 J	OT + LLLT vs OT	RTM	RCT parallel	till complete alignment	LLLT accelerates OTM
12	Ekizer et al. 2016[32] Turkey	20 (M + F) patients U 1st premolars extraction mean age: 16.77 ±1.41 years	– canine retraction – conventional brackets + NiTi closed-coil springs + mini-screw application – extra-oral LED device 618 nm, 20 mW/cm^2	20 min daily for 21 days	508 J	OT + LLLT vs OT + sham	– RTM – mini-screw stability – the IL-1ß level	RCT SMD	3 months	LLLT accelerates OTM and increases the stability of mini-screws, but has no effect on the IL-1ß level
13	Yassaei et al. 2016[18] Iran	11 (F) patients 1st premolars extraction mean age: 19.00 ±4.21 years	– U canine retraction – conventional brackets + NiTi closed-coil springs – GaAlAs laser 980 nm, 100 mW, 5.6 J/cm^2, continuous mode	3 pts B, 3 pts Ln on the U canine the apical third for 8 s, the cervical and middle ones for 10 s on days 0, 7, 14, 21, and 28 of each month	28 J	OT + LLLT vs OT + sham	– RTM – the IL-6 level in GCF	RCT SMD	11 months	no significant results for accelerating OTM and the IL-6 level
14	Caccianiga et al. 2016[33] Italy	21 (M + F) patients moderate crowding in the mandibular arch mean age: 26.0 ±5.4 years	– alignment – aligners applied 12 h/day – extra-oral diode laser 980 nm, 1 W, 150 J/cm^2, continuous mode	laser applied on the maxillary and mandible arch 3 applications for each arch each application for 50 s, a total of 150 s for each arch laser applied every other week	300 J	OT + LLLT vs OT	No. of aligners fitted correctly and No. of treatments finished successfully	RCT parallel	42 weeks	patients have to wear aligners for fewer hours when laser is applied

No.	Study/Setting	Participants	Intervention	Laser application schedule	Total energy dose per month	Comparison	Outcomes	Study design	Follow-up	Main findings
15	Dalaie et al. 2015[19] Iran	12 (M + F) patients U + L 1st premolars extraction mean age: 20.1 years	– U + L canine retraction – conventional brackets + sectional closing loops – GaAlAs laser 880 nm, 100 mW, 5 J/cm^2, continuous mode	4 pts B, 4 pts Ln on the canines 10 s each pt	*	OT + LLLT vs OT	– RTM – pain experience	RCT SMD	67 days	no significant results for accelerating OTM or reducing pain
16	Kansal et al. 2014[20] India	10 (M + F) patients U 1st premolar extraction	– U canine retraction – conventional brackets – GaAs diode laser 904 nm, 12 mW, 4.2 J/cm^2	5 pts B, 5 pts Ln on the U canines 10 s each pt on days 1, 3, 7, 14, 21, 28, 35, 42, 49, and 56 during the canine retraction phase	7.2 J for the 1st month	OT + LLLT vs OT	RTM	RCT SMD	63 days	no significant results for accelerating OTM
17	Pereira 2014[22] Brazil (thesis)	11 patients U + L 1st premolars extraction mean age: 14.04 years	– U + L canine retraction – conventional brackets + closing coil springs – 780 nm wavelength	5 pts B, 5 pts Ln on the canines the L canine B and Ln, and the U canine B – 40 mW power, 10 J/cm^2 energy density, 10 s each pt, 0.4 J/pt; the U canine Ln – 70 mW power, 35 J/cm^2 energy density, 20 s each point, 1.4 J/pt application each month	U: 9 J L: 4 J	OT + LLLT vs OT	– RTM – root and bone resorption	RCT SMD	3 months	no significant results for accelerating OTM
18	Heravi et al. 2014[21] Iran	20 (M + F) patients U 1st premolars extraction mean age: 22.1 ±5.3 years	– U canine retraction – conventional brackets + vertical loop – GaAlAs laser 810 nm, 200 mW, 21.4 J/cm^2/pt, continuous mode	5 pts B, 5 pts Ln on the U canines 30 s each pt days: 0, 3, 7, 11, and 15 of force application; on day 28, force adjusted and the same protocol repeated	300 J	OT + LLLT vs OT + sham	– RTM – pain experience	RCT SMD	56 days	no significant results for accelerating OTM or reducing pain
19	Souza 2014[23] Brazil (thesis)	11 patients U + L 1st premolars extraction age: 12–17 years	– U + L canine retraction – conventional brackets + closed-coil springs – 780 nm wavelength, 40 mW power, 10 J/cm^2 energy density	5 pts B, 5 pts Ln on the canines the L canine B and Ln, and the U canine B – 40 mW power, 10 J/cm^2 energy density, 10 s each point, 0.4 J/pt; the U canine Ln – 70 mW, 35 J/cm^2 energy density, 20 s each pt, 1.4 J/pt	U: 9 J L: 4 J	OT + LLLT vs OT	– RTM – pain experience	RCT SMD	3 months	LLLT was effective only in accelerating mandibular canine retraction and in the 1st month only
20	Kau et al. 2013[31] USA	90 (M + F) patients irregularity index >2 mm mean age: 18 ±7 years	– alignment – conventional brackets – extra-oral OrthoPulse device, 850 nm, 60 mW/cm2	a single exposure of 20 min/day = 72 J/cm^2 a single exposure of 30 min/day = 108 J/cm^2 a single exposure of 60 min/week = 216 J/cm^2	72 J/day, 2,160 J/month 108 J/day, 3,240 J/month 216 J/day, 6,480 J/month	OT vs OT + LLLT 20 min/day or 30 min/day or 60 min/week	RTM	RCT parallel	till complete alignment	LLLT accelerates OTM

No.	Study/Setting	Participants	Intervention	Laser application schedule	Total energy dose per month	Comparison	Outcomes	Study design	Follow-up	Main findings
21	Doshi-Mehta and Bhad-Patil 2012[6] India	20 (M + F) patients U 1st premolars extraction age: 12–23 years	– U canine retraction – conventional brackets + NiTi closed-coil springs – GaAlAs laser 810 nm, 0.25 mW, continuous mode	5 pts B, 5 pts Ln on the U canines 10 s each pt the 1st month: on days 0, 3, 7, and 14 every 15 days till complete retraction	**	OT + LLLT vs OT + sham	– RTM – pain experience	RCT SMD	till complete canine retraction	LLLT accelerates OTM and reduces pain
22	Sousa et al. 2011[24] Brazil	10 (M + F) patients 1st U or L premolars extraction mean age: 13.1 years	– canine retraction – conventional brackets + closed-coil springs – diode laser 780 nm, 20 mW, 5 J/cm^2, continuous mode	5 pts B, 5 pts Ln on the canines 10 s each pt on days 0, 3 and 7 after each activation (every month)	6 J	OT + LLLT vs OT + sham	RTM	RCT SMD	90 days	LLLT accelerates OTM
23	Hosseini et al. 2011[25] Iran	12 (M + F) patients 1st U or L premolars extraction mean age: 16.9 ±3.4 years	– U canine retraction – closed-coil springs – GaAlAs laser 890 nm, pulsed mode	on the buccal and palatal mucosa by a slow movement of the probe at the beginning of the 1st month (6 J every 48 h) laser applied in the 1st month only	72 J	OT + LLLT vs OT	RTM	RCT SMD	2 months	no significant results for accelerating OTM
24	Limpanichkul et al. 2006[26] Thailand	12 (M + F) patients U 1st premolars extraction mean age: 20.11 ±3.40 years	– U canine retraction – self-ligating brackets on U canines – GaAlAs laser 860 nm, 100 mW, 25 J/cm^2, 2.3 J/pt, continuous mode	4 pts B, 4 pts Ln on the canines 23 s each pt on days 0, 1 and 2 after each activation (repeated at the end of the 1st, 2nd and 3rd month)	55.2 J	OT + LLLT vs OT + sham	RTM	RCT SMD	3 months	no significant results for accelerating OTM
25	Cruz et al. 2004[27] Brazil	11 (M + F) patients U 1st premolars extraction age: 12–18 years	– U canine retraction – conventional brackets + closed-coil springs – GaAlAs laser 780 nm, 20 mW, 5 J/cm2, continuous mode	5 pts B, 5 pts Ln on the canines 10 s each pt 4 days of each month	8 J	OT + LLLT vs OT	RTM	RCT SMD	2 months	LLLT accelerates OTM

M – males; F – females; U – upper; L – lower; NiTi – nickel-titanium; GaAlAs – gallium-aluminum-arsenide; LED – light-emitting diode; GaAs – gallium-arsenide; pt(s) – point(s); B – buccal; Ln – lingual; OT – orthodontic treatment; LLLT – low-level laser therapy; RTM – rate of tooth movement; IL-1ß – interleukin 1 beta; TNF-ß1 – tumor necrosis factor beta 1; GCF – gingival crevicular fluid; IL-6 – interleukin 6; SMD – split-mouth design; OTM – orthodontic tooth movement.* Days of application were not mentioned, and no response to our email inquiry was received. ** On page 291, it is stated that for the biostimulation effect, the output power was 0.25 mW, a total of 10 pts (5 B and 5 Ln) were subjected to irradiation with an exposure time of 10 s/pt, and the result was 2 × 50 s × 0.25 mW = 0.025 J/session; however, it is also stated on the same page that the total amount of energy applied per session was 8 J (2 × 40 s × 100 mW). We asked for clarifications via email, but received no response.

Supplementary Table 3. Characteristics of the ongoing studies on the tooth movement facilitated by low-level-laser therapy (LLLT): Study design and intervention

No.	Study/Setting	Study design	Intervention
1	NCT03646942, 2017, Syria (recruiting)	RCT	lingual fixed orthodontic appliance
2	CTRI/2018/05/014328, 2017, India	RCT	fixed orthodontic appliance
3	CTRI/2018/04/013156, 2017, India	RCT	fixed orthodontic appliance
4	CTRI/2018/03/012316, 2018, India	RCT	fixed orthodontic appliance
5	CTRI/2017/07/009153, 2016, India	RCT	fixed orthodontic appliance
6	NCT02181439, 2014, France (completed)	RCT	intermaxillary elastics II
7	NCT02954133, 2016, USA (active, not recruiting)	RCT	aligners
8	NCT02267850, 2014, USA (completed)	RCT	fixed orthodontic appliance
9	NCT02267824, 2014, USA (completed)	RCT	fixed orthodontic appliance
10	DRKS00014964, 2018, Germany (pending)	RCT	fixed orthodontic appliance

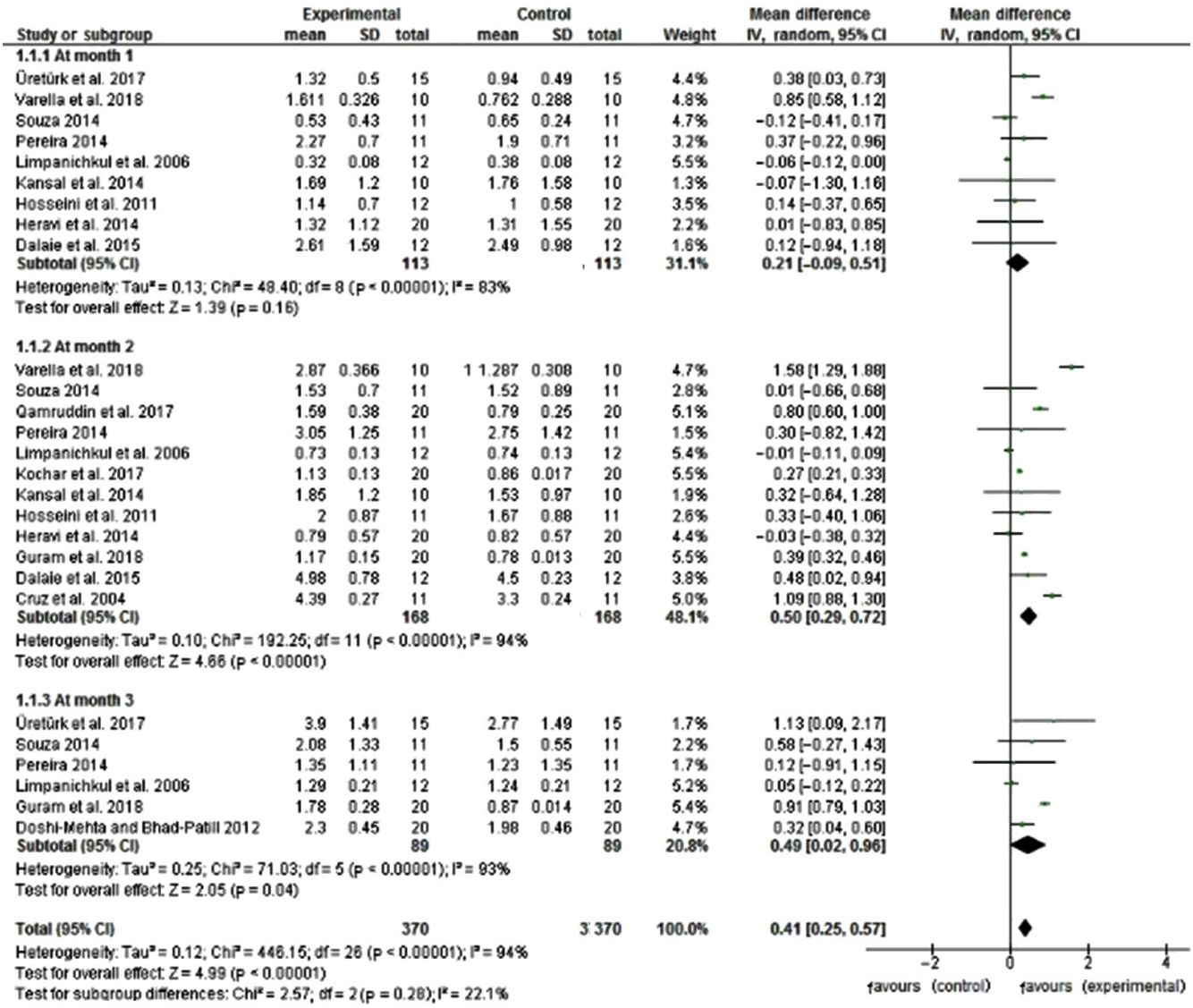

Fig. 2. Forest plot of the comparison between the LLLT and control groups at months 1, 2 and 3 of upper canine retraction

SD – standard deviation; CI – confidence interval; df – degrees of freedom.

Effect of intra-oral LLLT in accelerating leveling and alignment

Three parallel-design RCTs assessed the efficacy of intra-oral LLLT in accelerating leveling and alignment, either by assessing the overall treatment time needed for crowding resolution[10,11] or by assessing the rate of tooth movement.[12] All 3 studies indicated that LLLT is effective in accelerating the tooth movement. However, the data could not be pooled due to the differences in the treatment scenarios: upper alignment with extraction, lower alignment without extraction and upper alignment without extraction, respectively.

Effect of intra-oral LLLT in accelerating upper canine retraction

Sixteen split-mouth-design RCTs assessed the efficacy of intra-oral LLLT in accelerating upper canine retraction.[6,13–27] Noteworthy, the studies by Yassaei et al. and Sousa et al. were not included in data pooling.[18,24] In Yassaei et al.'s study, the analyzed sample comprised fewer than 10 patients due to the early closed extraction spaces at the beginning of the retraction phase.[18] In the case of the trial by Sousa et al., the data was obtained from a mixture of the upper and lower canines.[24] Therefore, these 2 studies were omitted to provide an accurate comparison. The amount of retraction at month 1 was assessed by 9 trials, comprising 226 canines.[13,17,19–23,25,26] The pooled estimate showed no statistically significant differences between the radiated and non-radiated groups (Fig. 2, 1.1.1: WMD (weighted mean difference) = 0.21; 95% CI (–0.09, 0.51); $p = 0.16$). Heterogeneity was very significant ($\chi^2 = 48.40$; $p < 0.001$; $I^2 = 83\%$). According to GRADE, the overall quality of evidence supporting this outcome is very low (Table 2).

The degree of retraction at month 2 was assessed by 12 trials, comprising 336 canines.[13–16,19–23,25–27] The pooled estimate showed a greater canine retraction (0.50 mm) in the radiated group (Fig. 2, 1.1.2: WMD = 0.50; 95% CI (0.29, 0.72); $p < 0.001$). Heterogeneity was very significant ($\chi^2 = 192.25$; $p < 0.001$; $I^2 = 94\%$). According to GRADE, the overall quality of evidence supporting this outcome is very low (Table 2).

The degree of retraction at month 3 was assessed by 6 trials, comprising 204 canines.[6,14,17,22,23,26] The pooled estimate showed a greater canine retraction (0.49 mm) in the radiated group (Fig. 2, 1.1.3: WMD = 0.49; 95% CI (0.02, 0.96); $p = 0.04$). Heterogeneity was very significant ($\chi^2 = 71.03$; $p < 0.001$; $I^2 = 93\%$). According to GRADE, the overall quality of evidence supporting this outcome is low (Table 2).

Effect of intra-oral LLLT in accelerating lower canine retraction

Five split-mouth-design RCTs assessed this outcome.[14,16,19,22,23] Three trials including 68 canines assessed the degree of retraction at month 1.[19,22,23] The pooled estimate showed no significant differences between the radiated

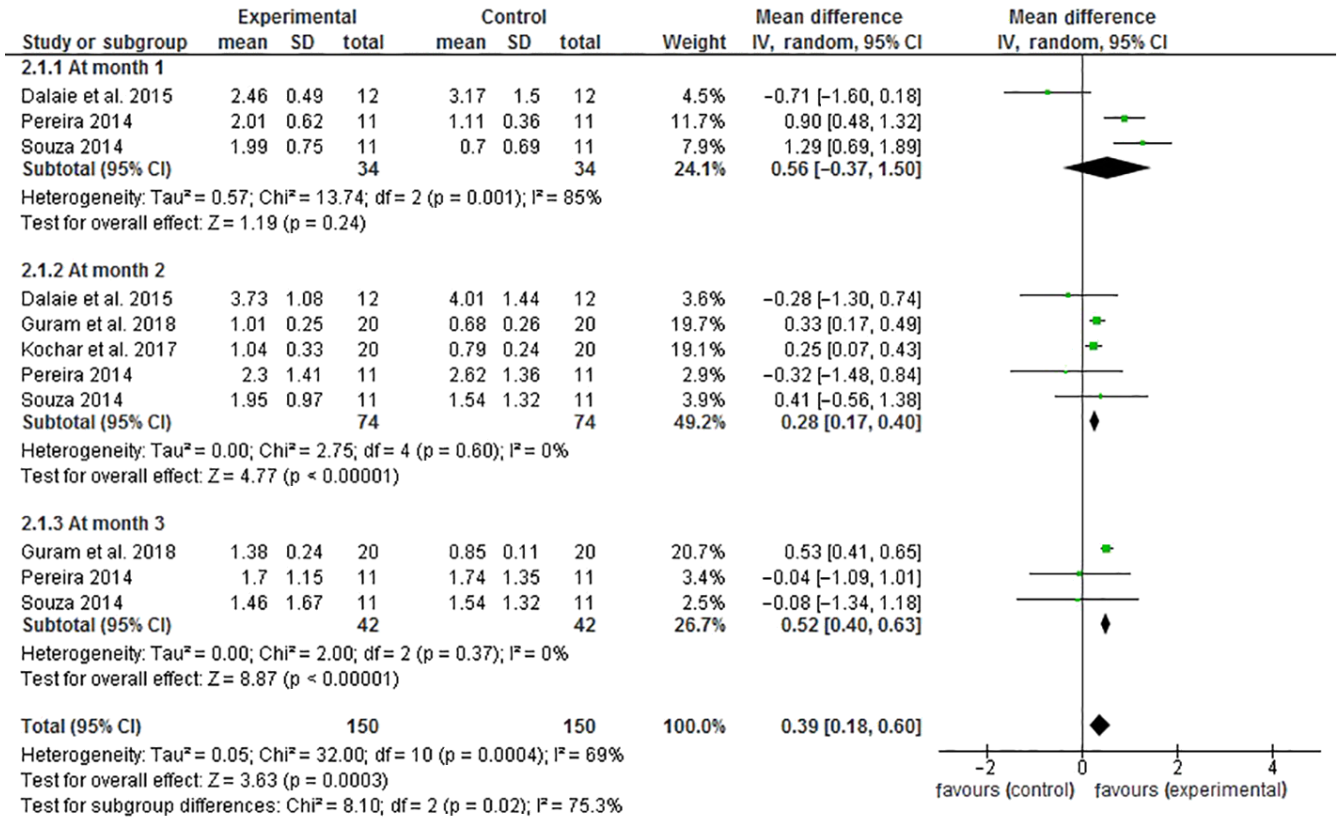

Fig. 3. Forest plot of the comparison between the LLLT and control groups at months 1, 2 and 3 of lower canine retraction

Table 2. Summary of findings according to the Grading of Recommendations Assessment, Development and Evaluation (GRADE) guidelines

Certainty assessment				Summary of findings						Comments
No. of studies	risk of bias	inconsistency	indirectness	imprecision	other considerations	No. of patients	effects		certainty	
							relative (95% CI)	absolute (95% CI)		
upper canine retraction facilitated by LLLT (month 2)										
12 RCTs	serious	not serious	serious	not serious	publication bias strongly suspected	171 SMD	–	MD 0.50 higher (0.29 lower to 0.72 higher)	⊕○○○ very low[a]	This outcome was also measured at month 1 in 9 RCTs (113 patients, SMD); the difference was not significant between both groups (–0.09 lower to 0.51 higher) with evidence quality very low[b] ⊕○○○. Also measured at month 3 in 6 RCTs (89 patients, SMD); the mean retraction in the intervention group was 0.49 higher (0.02 lower to 0.96 higher) with evidence quality low[c] ⊕⊕○○.
lower canine retraction facilitated by LLLT (month 2)										
5 RCTs	serious	not serious	serious	not serious	none	74 SMD	–	MD 0.28 higher (0.17 lower to 0.40 higher)	⊕⊕○○ low[d]	This outcome was also measured at month 1 in 3 RCTs (34 patients, SMD); the difference was not significant between both groups (–0.37 lower to 1.50 higher) with evidence quality low[e] ⊕⊕○○. Also measured at month 3 in 3 RCTs (42 patients, SMD); the mean retraction in the intervention group was 0.52 higher (0.40 lower to 0.63 higher) with evidence quality low[f] ⊕⊕○○.

High quality: We are very confident that the true effect lies close to that of the estimate of the effect. Moderate quality: We are moderately confident in the effect estimate – the true effect is likely to be close to the estimate of the effect, but there is a possibility that it is substantially different. Low quality: Our confidence in the effect estimate is limited – the true effect may be substantially different from the estimate of the effect. Very low quality: We have very little confidence in the effect estimate – the true effect is likely to be substantially different from the estimate of effect.

[a] Decline 1 level for the risk of bias: allocation concealment was high in [15] and unclear in [14, 16, 19, 21, 22, 23, 26, and 27]; blinding of outcome assessment was unclear in [21, 26 and 27]; outcome data attrition was unclear in [14, 16, 19, 21, 26, and 27]; 1 level for indirectness (differences in the population age); and 1 level for strongly suspected publication bias.

[b] Decline 1 level for the risk of bias: allocation concealment was unclear in [17, 19, 21, 22, 23, and 26]; blinding of outcome assessment was unclear in [17, 21 and 26]; outcome data attrition was unclear in [17, 19, 21, and 26]; 1 level for indirectness (differences in the population age); and 1 level for strongly suspected publication bias.

[c] Decline 1 level for the risk of bias: allocation concealment was unclear in [6, 14, 17, 22, 23, and 26]; blinding of outcome assessment was unclear in [17 and 26]; outcome data attrition was unclear in [6,14, 17, and 26]; and 1 level for indirectness (differences in the population age).

[d] Decline 1 level for the risk of bias: allocation concealment was unclear in [14, 16, 19, 22, and 23]; outcome data attrition was unclear in [14, 16 and 19]; and 1 level for indirectness (differences in the population age).

[e] Decline 1 level for the risk of bias: allocation concealment was unclear in [19, 22 and 23]; outcome data attrition was unclear in [19]; and 1 level for indirectness (differences in the population age).

[f] Decline 1 level for the risk of bias: allocation concealment was unclear in [14, 22 and 23]; outcome data attrition was unclear in [14]; and 1 level for indirectness (differences in the population age).

MD – mean difference.

and non-radiated groups after the 1st month of retraction (Fig. 3, 2.1.1: WMD = 0.56; 95% CI (–0.37, 1.50); $p = 0.24$). Heterogeneity was very significant ($\chi^2 = 13.74$; $p = 0.001$; $I^2 = 85\%$). According to GRADE, the overall quality of evidence supporting this outcome is low (Table 2).

Five trials including 148 canines assessed the degree of retraction at month 2.[14,16,19,22,23] The pooled estimate showed a greater tooth movement (0.28 mm) in the radiated group (Fig. 3, 2.1.2: WMD = 0.28; 95% CI (0.17, 0.40); $p < 0.001$). Heterogeneity was low ($\chi^2 = 2.75$; $p = 0.60$; $I^2 = 0\%$). According to GRADE, the overall quality of evidence supporting this outcome is low (Table 2).

Three trials including 84 canines assessed the degree of retraction at month 3.[14,22,23] The pooled estimate showed a greater tooth movement (0.52 mm) in the radiated group (Fig. 3, 2.1.3: WMD = 0.52; 95% CI (0.40, 0.63); $p < 0.001$). Heterogeneity was low ($\chi^2 = 2.00$; $p = 0.37$; $I^2 = 0\%$). According to GRADE, the overall quality of evidence supporting this outcome is low (Table 2).

Effect of intra-oral LLLT in accelerating anterior en-masse retraction

Two RCTs evaluated this outcome.[28,29] According to Samara et al., the patients treated with LLLT exhibited a significantly higher velocity of space closure by 0.22 mm/month than the non-radiated patients.[28] According to Arumughan et al., each round of laser application (21 days) accelerated the orthodontic tooth movement by 12.55% as compared to the conventional retraction technique.[29] However, the data from the 2 studies could not be pooled, because Arumughan et al.'s sample consisted of en-masse retraction and canine retraction distributed equally between the experimental and control groups.

Effect of extra-oral LLLT in accelerating leveling and alignment

Two RCTs assessed the efficacy of extra-oral LLLT in accelerating leveling and alignment.[30,31] According to Nahas et al., the time was significantly reduced (by 22%) in the test group as compared to the control group (68.3 vs 87.8 days, respectively; $p < 0.043$).[30] According to Kau et al., the mean rates of change in Little's Irregularity Index were 0.49 and 1.12 mm/week for the control and experimental groups, respectively.[31] However, the data from the 2 studies could not be pooled, because Nahas et al. used self-ligating brackets, whereas Kau et al. used conventional brackets.

Effect of extra-oral LLLT in accelerating upper canine retraction

One RCT comprising 40 upper canines from 20 patients assessed the efficacy of extra-oral LLLT in accelerating canine retraction using conventional brackets by evaluating the rate of tooth movement (mm/day).[32] The results showed a greater tooth movement in the radiated group, by 0.54 mm in the 1st month, 0.24 mm in the 2nd month and 0.22 mm in the 3rd month of retraction.

Effect of extra-oral LLLT in accelerating leveling and alignment using aligners

One RCT assessed the efficacy of extra-oral LLLT in accelerating leveling and alignment using aligners.[33] Caccianiga et al. allocated 21 patients to either the radiation group or the control group. All the patients were instructed to wear each aligner for 12 h per day for 2 weeks. In the control group, this 12-h protocol failed and was replaced by wearing aligners for 22 h per day. On the other hand, the 12-hour protocol was suitable for the radiated group, meaning the patients had to wear aligners for fewer hours when laser was applied.

Risk of bias in the included studies

Our analysis found that 4 studies were at low risk of bias, 17 studies were at moderate risk of bias and 4 studies were at high risk of bias. The principle risk factor affecting the methodology of laser studies was incomplete outcome data (attrition bias): 8% were at high risk of attrition bias, with more than 20% of the total sample size missing, and 48% were at unclear risk of bias. Figures 4 and 5 show the summary and graph of the risk of bias of the studies. More details on the assessment of the risk of bias with the supporting reasons for each assessment can be found in Supplementary Table 4.

Publication bias

The publication bias was assessed visually using standard funnel plots for the outcomes evaluated by 10 studies or more (i.e., the 1st and 2nd month of upper canine retraction). Figure 6 shows the funnel plots of the effect estimate against standard error (*SE*) for the outcomes. The shapes of the 2 funnel plots were deemed to be asymmetrical, which revealed the existence of the publication bias.

Discussion

The present systematic review comprised 570 patients from 25 completed RCTs, which reflects the interests of orthodontists in utilizing LLLT for accelerating tooth movement.

The pooled results of canine retraction facilitated by intra-oral LLLT showed no differences in the tooth movement at end of the 1st month, but significant differences after the 2nd and 3rd month. Since radiation has a cumulative effect, it seems that a 1-month period might be needed to achieve the biostimulation effects necessary to stimulate acceleration.

Supplementary Table 4. Assessment of the risk of bias with the supporting reasons for each assessment

No.	Study/Setting	Selection bias (randomization)	Selection bias (allocation concealment)	Performance bias
1	Varella et al. 2018 India	Low Risk "The experimental side was assigned by means of a lottery method with a sealed envelope."	Low Risk "The experimental side was assigned by means of a lottery method with a sealed envelope."	Low Risk The participants only were blinded (it was confirmed by contacting the corresponding author). According to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
2	Samara et al. 2018 UAE	Low Risk "Randomization and allocation concealment to the patient were achieved by asking each patient to draw a sealed envelope containing the allocation."	Low Risk "Randomization and allocation concealment to the patient were achieved by asking each patient to draw a sealed envelope containing the allocation."	Low Risk "It was not possible to conceal the treatment from both the patients and clinicians." However, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
3	Arumughan et al. 2018 India	Low Risk "The experimental side and the control side were randomly selected by an individual who was not part of the study." The method of randomization was not mentioned in the paper. An email was sent to the corresponding author, but there was no response.	Unclear Risk Concealment was not described in the article. An email was sent to the corresponding author, but there was no response.	Low Risk The study did not address this outcome. However, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
4	Al-Okla et al. 2018 UAE	Low Risk "The subjects were randomly divided into 2 groups." The method of randomization was not mentioned in the paper. An email was sent to the corresponding author, but there was no response.	Unclear Risk Concealment was not described in the article. An email was sent to the corresponding author, but there was no response.	Low Risk It was a double-blind clinical trial. Moreover, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
5	Guram et al. 2018 India	Low Risk "The 4 quadrants were randomly divided into the laser and control groups." The method of randomization was not mentioned in the paper. An email was sent to the corresponding author, but there was no response.	Unclear Risk Concealment was not described in the article. An email was sent to the corresponding author, but there was no response.	Low Risk "Neither the participant nor the 1st evaluator knows the grouping." Moreover, according to the criteria of judging the of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
6	Qamruddin et al. 2017 Pakistan	Low Risk "The maxillary arch was divided into the experimental and placebo groups by flipping a coin."	High Risk It was confirmed by an email, since the author stated that there was no allocation concealment.	Low Risk Although the participants only were blinded, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
7	Caccianiga et al. 2017 Italy	Low Risk "The subjects were randomly allocated to receive orthodontic treatment with a fixed appliance plus the administration of LLLT." The SPSS Statistics software was used to generate an allocation sequence.	Low Risk "Each subject was assigned a study number that was concealed until the date of bonding a fixed appliance."	Low Risk The study did not address this outcome. However, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
8	AlSayed Hasan et al. 2017 Syria	Low Risk A simple randomization technique was used. "Each patient was asked to select a folded piece of paper from a box containing 26 pieces of paper."	Low Risk Allocation concealment was done (it was confirmed by contacting the corresponding author).	Low Risk Although there was no blinding, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
9	Kochar et al. 2017 India	Low Risk "The randomly selected split-mouth design was used." The method of randomization was not described in the paper. An email was sent to the corresponding author, but there was no response.	Unclear Risk Concealment was not described in the article. An email was sent to the corresponding author, but there was no response.	Low Risk Although the participants only were blinded, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.

Detection bias	Attrition bias	Reporting bias	Other bias
Low Risk The outcome assessor was blinded (it was confirmed by contacting the corresponding author).	Low Risk No dropouts, as mentioned in the corresponding author's email.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Low Risk "The study models were pooled and coded. All measurements were obtained by a single investigator who was blinded to the group allocation of the study models."	High Risk More than 20% (25%) of the sample were dropped. Moreover, the missing data was not balanced in the numbers and reasons across the groups.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk "The photobiomodulation devices were provided by Biolux Research, which had no role in the design or execution of this study."
Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response. It is mentioned that the distance was measured 3 times, and the mean value is used for the data.	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	High Risk More than 20% (31%) of the sample were dropped. Moreover, the missing data was not balanced in the numbers across the groups.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk "This study was self-funded by the authors and their institution."
Low Risk The outcome was instrumentally measured and rechecked by another investigator for verification.	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Low Risk It was confirmed by an email, since the author stated that the outcome was instrumentally measured and rechecked by other assessors without any knowledge of the grouping.	Low Risk The missing outcome data was less than 20% (18%). Moreover, the missing data was balanced in the numbers, with similar reasons for the missing data across the groups.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk As illustrated on the CONSORT flow chart, there was no missing data.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
High Risk The outcome assessor was not blinded (it was confirmed by contacting the corresponding author).	Low Risk As illustrated on the CONSORT flow chart, there was no missing data.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Low Risk "All measurements were recorded by the same person. He/she was blinded to the control and lased sides."	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.

No.	Study/Setting	Selection bias (randomization)	Selection bias (allocation concealment)	Performance bias
10	Üretürk et al. 2017 Turkey	Low Risk "The canines were randomly separated into 2 groups consisting of 30 teeth." The method of randomization was not described in the paper. An email was sent to the corresponding author, but there was no response.	Unclear Risk Concealment was not described in the article. An email was sent to the corresponding author, but there was no response.	Low Risk The study did not address this outcome. However, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
11	Nahas et al. 2016 UAE	Low Risk "The patients were randomly assigned to one of the 2 groups utilizing simple randomization by asking them to draw a sealed envelope."	Low Risk They used a sealed envelope to conceal allocation.	Low Risk Although there was no blinding, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
12	Ekizer et al. 2016 Turkey	Low Risk "Randomization was accomplished with random LPT application side selection by coin tossing."	Low Risk "The treatment allocation was concealed in envelopes labeled with the study identification number."	Low Risk Although the participants only were blinded, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
13	Yassaei et al. 2016 Iran	Low Risk "All irradiations were performed by 1 operator in 1 maxillary quadrant which was randomly selected." The method of randomization was not described in the paper. An email was sent to the corresponding author, but there was no response.	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk The participants and the clinicians responsible for the treatment stages were blinded. Moreover, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
14	Caccianiga et al. 2016 Italy	Low Risk "The block randomization method was used. Each block contained 4 patients."	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk Although there was no blinding, according to the criteria of judging risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
15	Dalaie et al. 2015 Iran	Low Risk "The 4 quadrants were randomly selected in a complete block randomization manner using the Microsoft Excel software."	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk Although the participants only were blinded, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
16	Kansal et al. 2014 India	Low Risk "The right and left quadrants were assigned to either of the groups randomly at a coin toss to eliminate the left and right side bias, if any."	Low Risk "Random selection was done by 1 operator, who conveyed to the person doing the laser application as to which quadrant of the patient belongs to the laser group."	Low Risk According to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
17	Pereira 2014 Brazil (thesis)	Low Risk Laser was applied randomly only in 1 of the canines in the maxilla and in 1 of the canines in the mandible.	Unclear Risk The study did not address this outcome.	Low Risk According to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
18	Heravi et al. 2014 Iran	Low Risk "In each patient, one side was randomly allocated to laser treatment and another side to the placebo application." The method of randomization was not described in the paper. An email was sent to the corresponding author, but there was no response.	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk Although the participants only were blinded, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.

Detection bias	Attrition bias	Reporting bias	Other bias
Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response. Noteworthy, the movements were measured using the Ortho Analyzer™ software, whereas the reference points were selected manually.	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Low Risk "The alignment of the 6 lower anterior teeth was evaluated by a single investigator who was blinded regarding the patients' group allocation."	Unclear Risk Although the missing data was less than 20% (15%), it was not equally distributed between the 2 groups. Furthermore, the reasons for dropping out were different.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk It is mentioned that the devices were provided by Biolux Research and American Orthodontics. None of the sponsors (listed companies) had any role in the design or execution of this study.
Low Risk "The measurements of the data were done by a clinician blinded to the assignment."	Low Risk As illustrated on the CONSORT flow chart, there was no missing data.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk As illustrated on the CONSORT flow chart, there was no missing data.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was had no response.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Low Risk "... were aware of the irradiation side and the results were recorded by a third party."	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Low Risk "The distance measurement was done by the 3rd operator, who had no knowledge about which quadrant had been considered in the laser group."	Low Risk No dropouts occurred. The same sample number as mentioned in the methods part was mentioned again in the results part.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Low Risk The measurements were performed by an examiner other than the operator who applied the laser, which allowed a double-blind study, as the examiner did not know which side was the irradiated one when performing the clinical measurements.	Low Risk No dropouts, as shown in the results tables.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response. Noteworthy, the movements were measured using the Smile Analyzer™ software; the reference points were selected by one investigator and confirmed by another one.	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.

No.	Study/Setting	Selection bias (randomization)	Selection bias (allocation concealment)	Performance bias
19	Souza 2014 Brazil (thesis)	Low Risk Laser was applied randomly only in 1 of the canines in the maxilla and in 1 of the canines in the mandible.	Unclear Risk The study did not address this outcome.	Low Risk According to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
20	Kau et al. 2013 USA	Low Risk "The subjects were randomized into groups with varying treatment exposure times."	Low Risk It was confirmed by contacting the corresponding author.	Low Risk According to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
21	Doshi-Mehta and Bhad-Patill 2012 India	Low Risk "A randomly assigned incomplete block was used."	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk Although the participants only were blinded, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
22	Sousa et al. 2011 Brazil	Low Risk "The laser application was performed by 1 operator at the predetermined point areas, in only 1 of the canines randomly chosen."	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk Although the participants only were blinded, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
23	Hosseini et al. 2011 Iran	Low Risk "In this randomized clinical trial ... ." The method of randomization was not mentioned.	Low Risk It was confirmed by contacting the corresponding author.	Low Risk According to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
24	Limpanichkul et al. 2006 Thailand	Low Risk "Block randomization was used to allocate the side of the maxillary teeth (the left and right sides) to be subjected to LLLT and the placebo sides."	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk The participants and the clinicians responsible for the treatment stages were blinded. Moreover, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.
25	Cruz et al. 2004 Brazil	Low Risk "The left and right halves of the upper arcades were randomly divided into the described groups. The method of randomization was not described in the paper. An email was sent to the corresponding author, but there was no response.	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author; however, the corresponding author could not give definite answers, as the research had been done a long time ago.	Low Risk The study did not address this outcome. However, according to the criteria of judging the risk of bias, a low risk of bias is considered when the outcome is unlikely to be influenced by a lack of blinding.

CONSORT – Consolidated Standards of Reporting Trials; LPT – laser phototherapy.
Note: The attrition bias was considered as at: low risk if the missing data was less than 10% or less than 20%,
but with a balanced number and similar reasons for dropping out across the groups;
unclear risk if the missing data was between 10% and 20% with an unequal number and different reasons for dropping out across the groups;
and high risk if the missing data was more than 20%.

Detection bias	Attrition bias	Reporting bias	Other bias
Low Risk The analysis was performed at least 3 months after the end of the laser application with the objective of the examiner not to remember which was the irradiated and non-irradiated side of each patient, giving the study a double-blind character.	Low Risk No dropouts, as shown in the results tables.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Low Risk Assessor blinding was achieved, as confirmed by the corresponding author.	Low Risk No dropouts, as confirmed by the corresponding author.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk The trial was sponsored by Biolux Research. This company manufactures the devices used in this clinical trial. The authors did not state clearly that sponsors had no role in the design or execution of this study.
Low Risk "The measurement recorder was blinded to the experimental and control sides."	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Low Risk "Both the patient and the evaluators were not informed which tooth was irradiated."	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Low Risk It was confirmed by contacting the corresponding author.	Low Risk The missing data was less than 20% (8%).	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author, but there was no response.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.
Unclear Risk The study did not address this outcome. An email was sent to the corresponding author; however, the corresponding author could not give definite answers, as the research had been done a long time ago.	Unclear Risk The study did not address this outcome. An email was sent to the corresponding author; however, the corresponding author could not give definite answers, as the research had been done a long time ago.	Low Risk All the outcomes defined in the methods section were measured and reported.	Low Risk None.

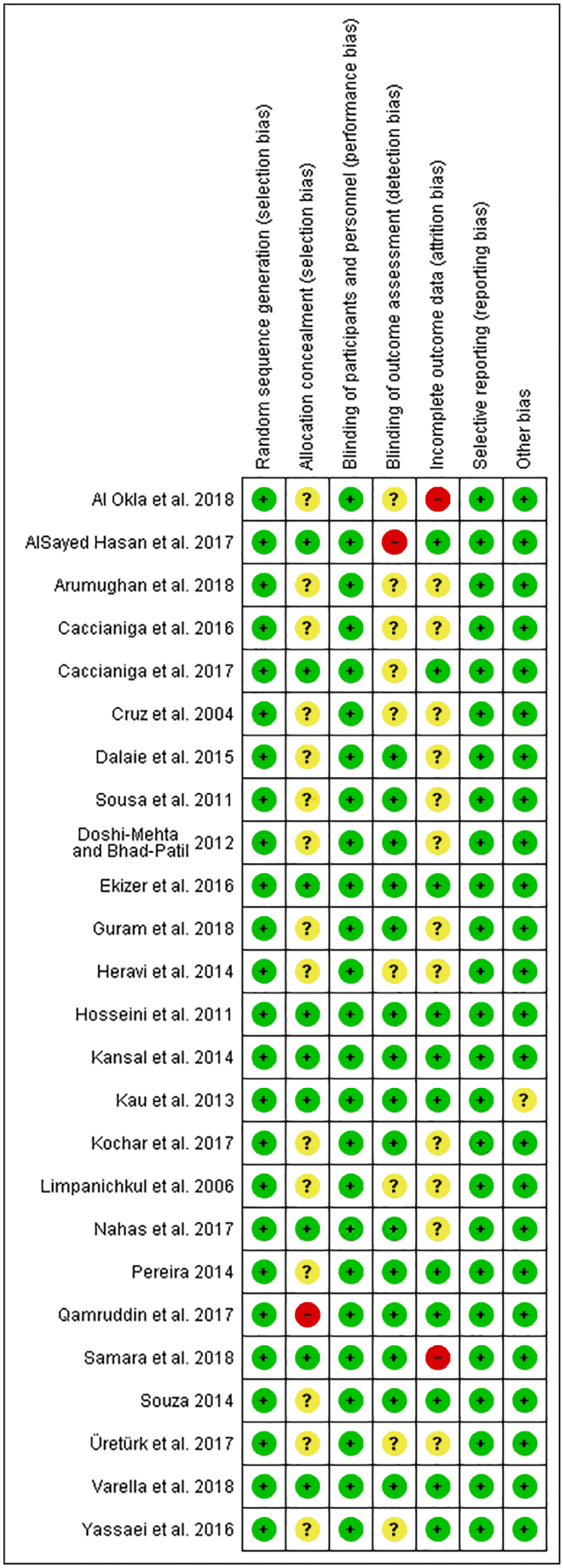

Fig. 4. Risk of bias summary: The review authors' judgments about each item of the risk of bias for the studies included

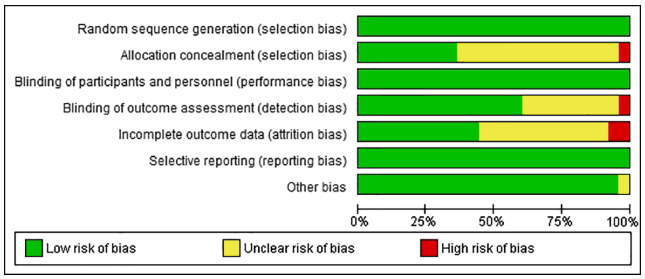

Fig. 5. Risk of bias graph: The review authors' judgments about each item of the risk of bias, presented as percentages across all the studies included

Although relevant grey literature, dissertations and non-English RCTs were sought in order to reduce the potential for the publication bias, the resultant asymmetrical funnel plots might be affected by the significant heterogeneity presented among the included studies.

It is worth highlighting that reporting the total number of joules applied per time period rather than J/cm^2 is recommended for expressing the laser dosage in future trials. This recommendation is clinically important, as it allows a precise comparison between different protocols, and thus allows avoiding confusion. To illustrate this, Kochar et al. and Cruz et al. used the same energy density (5 J/cm^2), but the total amount of energy applied was 30 J/month and 8 J/month, respectively.[16,27] Adding to the confusion, Guram et al. used 5 J/cm^2, whereas Limpanichkul et al. used 25 J/cm^2, but calculating the total amount of energy applied resulted in 48 J/3 weeks and 55.2 J/month, respectively.[14,26] This means the systematic review results of Ge et al., where 5 and 8 J/cm^2 were reported to be more effective than 20 and 25J/cm^2, constitute a misleading way of comparing different study protocols.[7]

Finally, the methodology of the studies utilizing physical stimuli was mostly affected by incomplete outcome data (attrition bias). Applying physical stimulation requires a highly compliant patient, who is actively interested in a shorter orthodontic treatment. Laser protocols often involve laser applications at several daily appointments, which requires high compliance to ensure efficacy.

As with all reviews, ours has strengths and limitations. With regard to strengths, we applied an extensive electronic search, sought grey literature, dissertations and non-English references, checked ongoing registered trials, used the Cochrane Collaboration tool for the assessment of the risk of bias, assessed the publication bias, and finally rated the overall evidence quality using the GRADE criteria. We tried to group LLLT studies according to the total number of joules applied per month, but there was no similarity among the studies in terms of the dosage used. Therefore, a wide variety of LLLT dosimetry in the included studies is considered a limitation of this systematic review.

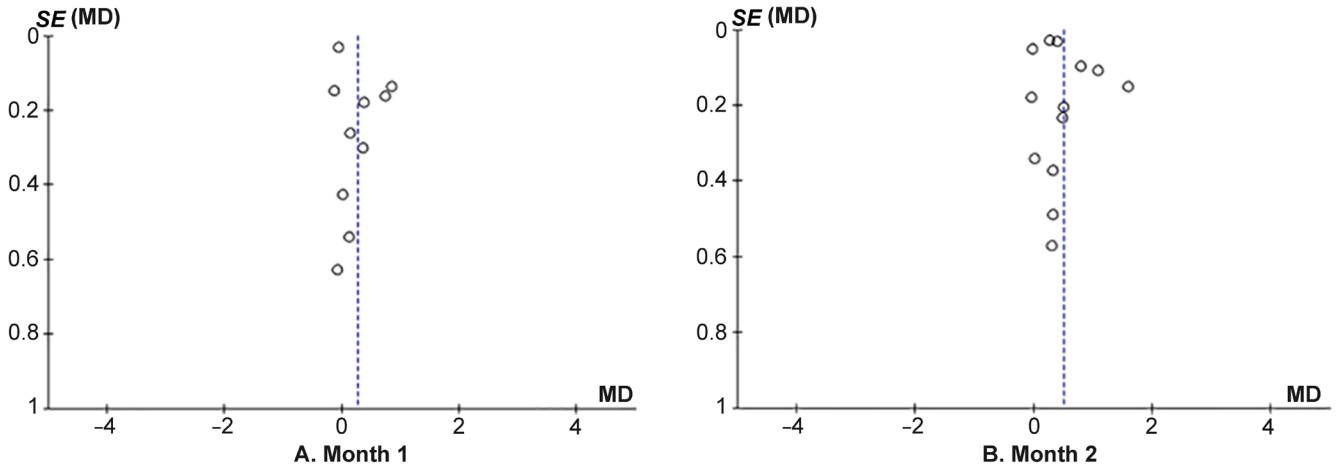

Fig. 6. Funnel plots of the effect estimate against standard error (*SE*)

A – month 1 of upper canine retraction facilitated by LLLT; B – month 2 of upper canine retraction facilitated by LLLT.

Conclusions

Low-level laser therapy can speed up the rate of the orthodontic tooth movement, and consequently decrease the treatment time. However, the quality of evidence ranged from low to very low and the clinical significance is questionable, so more precise studies are needed. It is highly recommended to express and compare the laser dosage in future trials by the total number of joules applied per time period rather than the previously used J/cm^2.

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Current perspectives and the future of *Candida albicans*-associated denture stomatitis treatment

Stan obecny i perspektywy leczenia stomatopatii protetycznej związanej z zakażeniem *Candida albicans*

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Abstract

Denture stomatitis (DS) is a multifactorial disease, but the proliferation of *Candida albicans* (*C. albicans*) is the main causative factor. Different modalities have been suggested for the prevention and treatment of DS. Among the different approaches that have been implemented to inhibit and control DS there are the topical application of antifungal agents, the surface modification of the denture base and the incorporation of antimicrobial agents into the denture base material. Antifungal agents can effectively control DS, but the recurrence of the disease is common. Accordingly, it has been suggested that coating the surface of the acrylic denture base may result in a decreased fungal adhesion. In recent years, nanotechnology has dominated the research, and several nanoparticles have demonstrated antifungal effects.

Therefore, the aim of this article was to review the antifungal effects of the different methods that have been suggested for the prevention and/or control of DS as well as the antimicrobial activity of denture base acrylic resin additives, including nanoparticles. Studies reporting the incorporation of antifungal/antimicrobial agents into the polymethyl methacrylate (PMMA) denture base were included in this review. The PubMed, Web of Science, Google Scholar, and Scopus databases were searched for the articles published between January 2000 and December 2018 using the following key words: dental prosthesis, denture stomatitis, candidiasis, antifungal agents, biofilm formation, polymethyl methacrylate, and PMMA. The antimicrobial material incorporated into the resin may have a superior effect in preventing DS over simply coating the surface of the denture base. However, some antimicrobial fillers can have adverse effects on the physical and mechanical properties of the denture base resin.

Key words: polymethyl methacrylate, denture stomatitis, candidiasis, antifungal agents, dental prosthesis

Słowa kluczowe: polimetakrylan metylu, stomatopatia protetyczna, kandydoza, czynniki przeciwgrzybicze, proteza dentystyczna

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Introduction

Denture stomatitis (DS) is an inflammatory reaction that mainly occurs due to a fungal infection by *Candida albicans* (*C. albicans*).[1] According to Newton and Silva Pinto et al., DS is classified into 3 types that appear in the denture-bearing area: dispersed petechiae in the palatal mucosa (type I); diffused erythema (type II); and papillary hyperplasia with diffused erythema (type III).[2,3] Many factors contribute to its occurrence, including ill-fitting dentures, continuous wear, poor oral hygiene, and the type of denture base material.[1] Additionally, any systemic conditions of the patient, such as xerostomia or diabetes, can be predominant factors in the pathology of DS.[4]

The surface properties of the denture base material, such as surface roughness and hardness, can have a significant influence on the incidence of DS.[5] For example, a denture base material of high hydrophobicity and surface roughness enhances *C. albicans* adhesion and proliferation.[1,5] In addition, the adequate hardness of the denture base resin is important in order to resist scratches which may occur when using or cleaning the denture.[1] These properties are adversely affected by denture cleansers, which impacts the long-term success of the prosthesis.[5] The acrylic denture base is a harbor for microbial colonization; therefore, a denture base material that resists *C. albicans* adhesion and proliferation could reduce the incidence of DS.[1,5] Different approaches have been implemented to inhibit and control DS, such as topically applying antifungal agents, modifying the denture base surface and incorporating antimicrobial agents into the denture base resin.

Study design

Search strategy

This review was designed to address the different methods that have been implemented in the control or prevention of DS, including the latest approaches of incorporating antimicrobial agents and nanofillers into the heat-polymerized polymethyl methacrylate (PMMA). The question posed was as follows: Is coating or incorporating antifungal agents into the PMMA denture base resins more effective than conventional DS treatment? This review was performed according to the PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) guidelines (Fig. 1).

The authors performed an extensive bibliographic search using the PubMed, Web of Science (the core collection), Google Scholar, and Scopus databases to identify relevant full-text articles in English which were published between January 2000 and December 2018. The manual search of the reference sections of individual studies did not yield additional articles. The following key words were employed in the search of all the selected databases: dental prosthesis, denture stomatitis, candidiasis, antifungal agents, biofilm formation, polymethyl methacrylate, and PMMA.

Eligibility criteria

The inclusion criteria comprised in vitro, in vivo and clinical randomized control trials that investigated the different modalities of DS treatment, including the modification of the heat-polymerized PMMA denture base material. Articles which were not related to the field of dentistry were excluded to eliminate any bias in the testing and reporting methods. Review articles and research papers in languages other than English were excluded from the study. Letters to the editor, personal communications, abstracts, and published theses were excluded as well as any unpublished data. Likewise, studies in which antifungal agents were added to tissue conditioners, soft liners and cold-cured acrylic denture base materials were excluded as well as studies reporting the effects of these antifungal agents on the surface and mechanical properties of PMMA.

Data management

The authors (M.M.G. and S.M.F.) independently screened the titles and abstracts only. After duplicates and irrelevant articles were excluded, the full-length articles

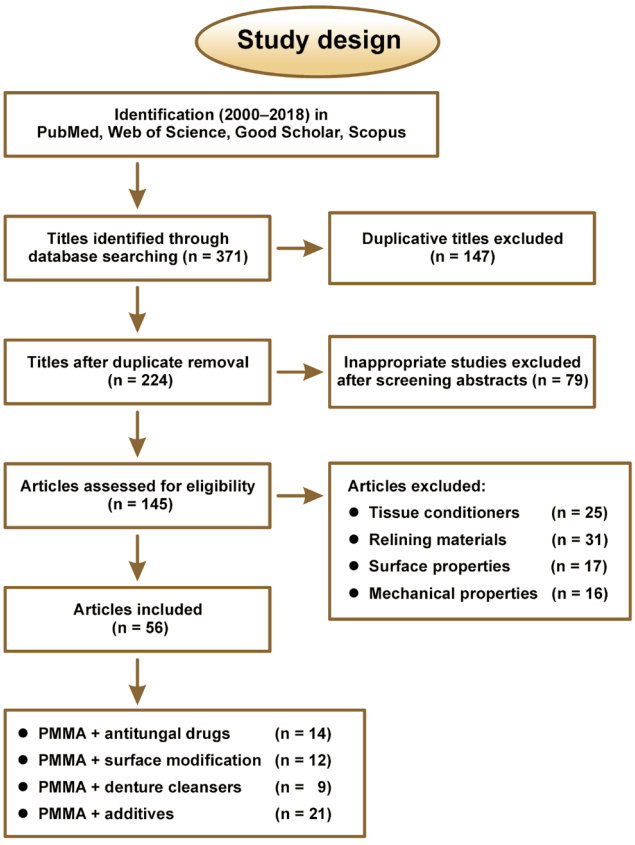

Fig. 1. Study design

PMMA – polymethyl methacrylate.

were carefully reviewed and the 2 authors decided together which articles to include in the review. The articles were then classified according to the mode of treatment: oral antifungal agents, natural extract-based antifungal agents, cleansing agents, surface modifications of the denture base resin, and antimicrobial resins. The data obtained from the articles categorized under the inclusion criteria were tabulated in the prepared Microsoft® Excel sheet (Microsoft Corporation, Redmond, USA). The authors categorized the information based on author/year, type of study, type of resin, antifungal agents (type, methods of use and applications, type of additive, modifications/treatment, and mechanism of action), results, and conclusions/outcomes. The effects of different treatment modalities were categorized for a descriptive review. Due to significant variations between the included studies, the data was analyzed descriptively, as the statistical meta-analysis was not applicable.

Results and discussion

Oral antifungal agents

Several oral antifungal agents have been suggested for the treatment of DS, such as fluconazole, nystatin, amphotericin B, miconazole, ketoconazole, itraconazole, and clotrimazole (Table 1).[6–9] They are effective in the treatment of DS, but their drawbacks include toxic side effects and the development of resistant strains.[6,9] Moreover, DS commonly recurs. Therefore, after the completion of the antifungal therapy, dentures must be disinfected and kept clean in order to avoid the recurrence of DS.[10] The topical application of antifungal agents is effective only for a short period of time. Besides, these agents are usually negatively affected by the constant flushing of saliva.[6,10]

Natural extract-based antifungal agents

Several natural products have antifungal and antimicrobial effects, which can be used to combat DS. Natural products are cheaper, less toxic and less likely to induce microbial resistance in comparison with conventional drugs.[11] However, some of these natural products can show toxicity or cause intolerance; therefore, they must be used cautiously.[12]

It has been found that the *Streblus asper* leaf ethanolic extract (SAE), when used at a dose of 62.5 mg/mL, reduces the adherence of *C. albicans*.[4] It has been suggested that SAE affects the cell walls of *C. albicans* and/or changes the chemical structure of the resin surface.[4] Pinelli et al. reported that *Ricinus communis*, when used as a mouthwash 4 times a day, could combat DS by decreasing biofilm formation.[13] In addition, propolis, when added at 2% to an oral gel, demonstrated anti-inflammatory effects, curing the lesions caused by *C. albicans* infections.[14] It has been suggested that propolis may not reduce the quantity of *C. albicans*, but rather it changes the fungus phenotype and acts on the dimorphism of the yeast.[14] Moreover, the *Equisetum giganteum* extract contains phenolic compounds and flavonoid heterosides, which have antimicrobial properties.[15] They showed their antimicrobial effects when brushed on the acrylic resin surface at concentrations of 4 or 8 mg/mL.[11,15]

Table 1. Antifungal agents used in denture stomatitis (DS) treatment

Antifungal agent	Formulation	Dose	Effectiveness	Adverse effects
Fluconazole	tablets suspension	50–100 mg/day 100 mg/mL per day	effective; relapse after 4 weeks	nausea, vomiting, diarrhea, abdominal pain
Nystatin	suspension, 60 mL ointment, 30 g tablets	4–6 mL/6 h 2–4 applications/day 2/8 h	effective; clinical cure – 87.5% mycological cure – 66%	well-tolerated uncommon nausea, vomiting, gastrointestinal effects
Amphotericin B	topical infusion	3 times/day 100–200 mg/6 h	effective; relapse after 12 weeks	renal, cardiovascular, spinal, and neurological
Miconazole	gel, 2%	100 mg/6 h	effective	uncommon; burning sensation, irritation, nausea, diarrhea
Ketonazole	gel, 2% tablets suspension, 30 or 10 cc	3 times/day 200 mg 1–2 times/day 200 mg (20 mL) 4 times/day	effective	nausea, vomiting, abdominal pain
Itraconazole	capsules	100–200 mg/day	effective; relapse in patients with poor oral hygiene	nausea, vomiting, diarrhea, abdominal pain
Clotrimazole	gel, 1% tablets, 10 mg	3 times/day 5 times/day	effective	occasional skin irritation, burning sensation

The antimicrobial activity of the *Equisetum giganteum* extract results from inactivating the adhesion proteins and disrupting the microbial cell membrane.[11] The extract may also increase the hydrophilicity of the acrylic resin surface and reduce its surface energy, thus decreasing fungal adhesion.[15]

The *Plantago major* extract has shown an antifungal effect against *C. albicans* through the active components aucubin and baicalein.[16] They strongly inhibit *C. albicans* adhesion and growth by preventing the cell surface hydrophobicity pathway.[16] *Terminalia catappa* and the lemon grass extract have been proven to cause a reduction in *C. albicans* adhesion when used for the immersion of the denture base.[17,18] Accordingly, they can be used as effective disinfectants against the formation of the *C. albicans* biofilms.[18] In addition, the immersion of dentures overnight in a 10% vinegar solution for 45 days has been found to exert a disinfectant effect against *C. albicans*.[3,19]

Cleansing agents

Oral mouth rinses are more conservative in the management of DS as compared to systemic drugs.[20] Several cleansing agents are available for denture immersion, such as sodium perborate, sodium hypochlorite, coconut soap, phosphoric and benzoic acids, chlorhexidine digluconate, and glutaraldehyde.[20–23] Gornitsky et al. confirmed the antimicrobial effects of sodium perborate-based tablets and suggested the controlled use of these cleansers.[20] Sodium perborate is an effective denture cleanser when used at a concentration of 3.8% for 10 min daily.[20] In addition, sodium hypochlorite (0.5% concentration for 10 min/day) eliminates denture plaque, but it also causes metal corrosion and skin irritation.[21] The antifungal effects of sodium bicarbonate have been reported at a concentration of 5%; therefore, it can be used for the disinfection of removable prostheses.[22] Chlorhexidine has excellent antimicrobial effects at a low concentration (0.12%) and it is practical for everyday use.[23]

Denture cleansers are effective antimicrobial agents; however, their effects depend mainly on their continuous and proper use, according to the guidelines on preparation and immersion time.[24] Unfortunately, denture cleansers adversely affect the physical properties of the denture base resin.[24,25] Several studies have reported an increase in the surface roughness of the denture base resin following the use of denture cleansers, which consequently increased the accumulation of plaque.[25] Denture cleansers may also cause color changes to the denture base resin and a reduction in the flexural strength.[24,25] Soft tissue irritation and mild cytotoxic effects have been reported when using glutaraldehyde and sodium hypochlorite, respectively. Additionally, sodium hypochlorite causes the bleaching of the denture base resin.[25] Furthermore, chlorhexidine causes a burning sensation to the oral mucosa, tooth staining and denture discoloration.[23]

Surface modifications (resin coating)

The surface roughness of the inner surface of a dental prosthesis is one of the factors that affect the adhesion of *C. albicans* and biofilm formation.[26] The adherence of *C. albicans* to the denture base resin followed by the colonization of microorganisms occurs mainly due to hydrophobic interactions and electrostatic forces.[27] In addition, an increased surface roughness of the denture base resin enhances microbial adhesion[28] and makes it difficult to detach microorganisms, even with the use of antimicrobial agents.[27] Therefore, it has been suggested that coating the surface of the denture base resin may decrease microbial adhesion.[1] Several materials have been used for coating the surface of the denture base resin and reducing *C. albicans* adhesion, such as 2-octyl cyanoacrylate, silane-silicon dioxide (SiO_2) nanocomposite films, and coatings containing zwitterion or hydrophilic monomers.[29–31] Ali et al. suggested that coating the denture base resin with 2-octyl cyanoacrylate could reduce *C. albicans* adhesion by increasing surface hydrophilicity and providing a smoother surface.[29] Similarly, silane-SiO_2 nanocomposite films reduced the surface energy of the coated acrylic resin and increased its hydrophilicity, thus decreasing *C. albicans* adhesion.[30]

Lazarin et al. found that photo-polymerized coatings containing zwitterion or hydrophilic monomers could reduce the adhesion of *C. albicans* to the acrylic resin, particularly at a high concentration (35%).[31,32] They concluded that these coatings changed the chemical composition, but did not alter the hydrophobicity of the surface. A significant decrease in *C. albicans* adhesion was reported when the surface was coated with sulfobetaine or hydrophilic monomers.[31–33] They caused changes to the carbon and oxygen content of the acrylic surface, and so did the presence of sulfur.[34] Applying a zwitterionic coating to the surface of the substrate reduced microbial adhesion, including that of *C. albicans*, and this antibioadherent property was associated with the sulfobetaine component.[34]

A reduction in *C. albicans* adhesion was detected in the case of certain polymer coatings, such as parylene, which also improved surface wettability.[35] Accordingly, increasing the hydrophilicity of the denture surface could reduce *C. albicans* adhesion and microbial colonization.[32] Park et al. found a significant reduction in the amount of *C. albicans* adhering to the resin surface modified with surface charges and self-bonding polymers.[36] They suggested that these surface modifications created an electrostatic repulsion between *C. albicans* and the acrylic resin, which prevented the initial attachment caused by hydrophobic interactions and electrostatic forces.[36] Similarly, Hirasawa et al. reported a significant decrease in the initial adhesion of *C. albicans* to the denture base coated with a crosslinkable copolymer containing sulfobetaine methacrylamide (SBMAm).[34] Sulfobetaine methacrylamide increases the hydrophilicity of the surface it coats, thus decreasing

C. albicans adhesion by reducing the hydrophobic interactions between *C. albicans* and the denture base resin.[37,38]

Titanium dioxide (TiO_2) has been used to coat a number of medical products. It is biocompatible, and it has photocatalytic effects, which allows it to be used to coat the denture base resin.[39] The TiO_2 coating of the denture base resin has shown an inhibitory effect against the adhesion of *Streptococcus sanguinis* and *C. albicans*.[31,39,40]

Based on the review of coatings, it can be stated that coating the denture base modifies the surface, changing surface characteristics from rough hydrophobic to smooth hydrophilic, thus decreasing biofilm formation and *C. albicans* adhesion. However, further studies are required to test the biocompatibility of these coatings as well as their durability and resistance to the chemical and mechanical procedures of denture cleaning.

Antimicrobial resin (resin with incorporated filler and nanofiller antimicrobial agents)

The conventional chemical and mechanical methods used to clean dentures are satisfactory in many cases. However, these methods present challenges for some geriatric patients and patients with physical disabilities.[19] Incorporating antimicrobial and antifungal agents into the denture base resin could be an effective way to eliminate biofilm formation and microbial adhesion. These effects have been investigated in many studies, but biofilms are difficult to eliminate because of their inherent resistance to the incorporated antimicrobial and antifungal agents, in addition to altering the physical and mechanical properties of the resin.[41]

The addition of a fluoridated glass filler or surface pre-reacted glass-ionomer (S-PRG) filler to the acrylic denture base significantly reduces *C. albicans* adhesion without negatively altering the physical properties of the resin. The modified resin releases fluoride ions and acts as a fluoride reservoir (Table 2).[9,42,43]

Microorganisms tend to adhere more to hydrophobic surfaces than to hydrophilic ones through the formation of strong hydrophobic bonds.[3] Therefore, microbial adhesion is enhanced by the hydrophobic nature of the denture base resin and electrostatic interactions.[2,44] The addition of methacrylic acid to PMMA creates a negative charge, which produces a repulsive force toward the negatively charged surfaces of most bacteria, thus reducing adherence.[45] There have been reports on the significant antimicrobial activity of the denture base acrylic resin containing antimicrobial polymers (2-tertbutylaminoethyl methacrylate). While its antimicrobial activity was significant against the *Staphylococcus aureus* and

Table 2. Types and mechanisms of action of the fillers and nanofillers incorporated into PMMA

Additive	Type	Mechanism of action
Glass filler	S-PRG filler[9]	A S-PRG filler is capable of releasing 6 types of ions (Na^+, Sr^{2+}, SiO_3^{2-}, Al^{3+}, BO_3^{3-}, and F^-) and exhibits fungistatic effects against *C. albicans*. Boric acid can destabilize the membranes of *C. albicans* by decreasing the relative ergosterol content and it causes the disintegration of the cytoskeletal elements of *C. albicans* at the hyphal tip, thus inhibiting the growth of this fungus. Sodium fluoride (NaF) exhibits fungicidal activity; its antifungal effects are ascribed to the interaction of the stannous fluoride component with the plasma membrane of the yeast cells. Sodium (N) can create high osmotic stress and kill *C. albicans* cells, while increasing the doubling time for *C. albicans*.
	fluoridated glass filler[42]	An acrylic resin modified with fluoridated glass fillers acts as a fluoride-releasing device.
Nanofiller	AgNPs[52–54]	AgNPs wrap around *C. albicans* cells and cause the disruption of the fungal membrane and the inhibition of the normal budding process. Silver (Ag) ions or AgNPs are released into the aqueous medium from the acrylic resin to exert their antifungal influence. AgNPs, with their rapid and broad-spectrum efficacy and the sustained silver cation (Ag^+) release, appear to be more effective antimicrobial agents than micro-sized silver powder [µm], which shows lower antimicrobial activity owing to its limited surface area. The cation Ag^+ interacts with cytoplasmic components and nucleic acids to inhibit the respiratory chain enzymes and to interfere with membrane permeability. AgNPs are sensitive to oxygen and convert oxygen into active oxygen through catalytic action. This active oxygen causes structural damage to microorganisms, which is called the oligodynamic action of Ag. Silver ions are positively charged, so they interact with the negatively charged cell membranes of bacteria, causing their death due to an increased cell wall permeability. In addition, Ag adheres to bacterial DNA, RNA, proteins, and enzymes, preventing cell division and damaging the cellular content of bacteria.
	TiO_2NPs[41,44]	The antibacterial effect of titanium dioxide (TiO_2) occurs due to the deactivation of the cellular enzymes by coordinating the electron-donating groups, which results in gaps arising in the bacterial cell walls, leading to their higher permeability, and finally to cell death. The cell components of microorganisms degrade with no release of potentially toxic compounds. TiO_2 demonstrates photocatalytic properties in the presence of photons with wavelengths lower than 388 nm, by which electrons get excited (the prominent catalytic effect). Free radicals are subsequently formed with such a high level of energy that they can react with various organic materials and enable their degradation.
	ZnO_2NPs[60]	ZnO_2NPs are responsible for the production of reactive oxygen species as well as for the accumulation of the nanoparticles in the cytoplasm or on the outer cell membranes of bacteria. The antifungal effect might also be attributed to the dissociation of zinc (Zn) ions from ZnO_2NPs.

S-PRG – surface reaction-type pre-reacted glass-ionomer; AgNPs – silver nanoparticles; TiO_2NPs – titanium dioxide nanoparticles; ZnO_2NPs – zinc peroxide nanoparticles; *C. albicans* – *Candida albicans*.

Streptococcus mutans biofilms, it was not significantly effective against *C. albicans*.[46] Moreover, the surface roughness and wettability of the acrylic resin surface increased, and color changes have also been observed.[46,47]

Adding natural extracts such as henna (*Lawsonia inermis*) and the seeds of *Nigella sativa* as fillers in the PMMA denture bases can play a significant role in the prevention of microbial or fungal adhesion and proliferation.[48,49] Nawasrah et al. found that a natural antimicrobial agent (henna) may control *C. albicans* proliferation on the denture surface, and that it might serve as a possible method of prevention and treatment of DS.[48] Thymoquinone (TQ) is the major ingredient of the essential oil obtained from the *Nigella sativa* seeds, and it has promising therapeutic potential in medicine and dentistry. Al-Thobity et al. reported that adding TQ to the denture base resin could effectively prevent *C. albicans* adhesion and proliferation.[49] However, even with the significant antifungal properties of natural extracts, their adverse effects on the physical and mechanical properties of the denture base resin may restrict their usefulness.[50]

Antimicrobial and antifungal effects have been reported for nanofillers when they are added to the PMMA denture base materials, forming nanocomposites with superior properties.[51] Nanocomposites have been investigated in many studies in an attempt to produce a denture base material that is resistant to fungal and microbial adhesion, and subsequently to DS.[51,52] Several studies have reported the antimicrobial effects of nano-sized particles, such as silver nanoparticles (AgNPs), titanium dioxide nanoparticles (TiO_2NPs), zinc oxide nanoparticles (ZnONPs), and zirconium dioxide nanoparticles (ZrO_2NPs) (Table 2).

Silver nanoparticles are biocompatible and have shown strong antimicrobial effects against a wide range of bacteria, viruses and fungi.[53,54] Consequently, their incorporation into the denture acrylic resin may prevent DS.[54] They have a smaller tendency to induce microbial resistance as compared to conventional antibiotics.[55] Although several studies have reported the antifungal properties of the denture base resin containing AgNPs – especially at high concentrations – the nanoparticles caused resin discoloration and did not improve their mechanical properties.[54,56] The recent studies using silver-containing nanomaterials have suggested that the bacterial toxicity of these materials originates partially from membrane damage and the disruption of ion homeostasis, with unpredictable effects on human health.[53,54,57–59]

The incorporation of ZnONPs into the acrylic denture resin has also been tested and it has been found that they might prevent fungal infections.[57,60] The results indicate that filling denture base resin materials with commercially available ZnO, calcium oxide (CaO) or TiO_2 nanopowders inhibits biofilm formation on their surfaces.[51]

Titanium dioxide nanoparticles are considered to be an excellent filler for several reasons.[61] Most importantly, titanium-polymer nanocomposites are biocompatible and they exert a non-contact biocidal impact.[44] In addition to their antibacterial and antifungal properties, TiO_2NPs are inexpensive and they enhance the mechanical properties of the denture base resin[61]; they also provide the resin with photocatalytic effects. However, their antimicrobial and photocatalytic properties are significant only at a relatively high concentration of TiO_2NPs (5 wt%), which may weaken the resin.[39] When the TiO_2 photocatalyst is exposed to ultraviolet light, it generates oxidative species, which kill microorganisms by damaging their cell walls. Additionally, it has excellent hydrophilicity.[39,61]

Oral hygiene and denture care are important factors in the prevention and treatment of DS. Based on the aforementioned factors, the question raised now is: Which is better – coating or incorporating the antimicrobial agent into the denture base resin? Coatings provide smooth surfaces, which prevents *C. albicans* adhesion, but they may affect denture retention by obliterating surface details as well as tissue adaptability. Moreover, the mechanical and chemical cleaning of dentures may disturb the coating, leaving a rougher surface with an increased tendency toward microbial adhesion, staining and poor esthetics. Incorporating antimicrobial agents into the PMMA denture base may eliminate the drawbacks of coating by imbedding the agents within the resin matrix.

Good esthetics is highly desirable in the case of removable dental prostheses; therefore, the use of antimicrobial agents, whether they are coated or incorporated, should not compromise the esthetics of the final composites. It has been found that coating does not alter the optical properties of the acrylic resin.[37] However, over time, the optical properties are affected by the scratches resulting from mechanical cleaning, which makes the surface rougher and increases the susceptibility of the denture to microbial adhesion and staining. Antimicrobial monomers and glass fillers result in minimal color changes to the denture base resin, followed by the TiO_2 nanofiller, which causes white discoloration, whereas AgNPs displayed the worst effects on the esthetics, with a grey-colored acrylic resin. Silver nanoparticles have been reported to have antifungal properties, but their adverse effects on the esthetics of the acrylic base may limit their clinical use. Therefore, it has been recommended to add them to less visible areas. The same method may be applied to natural extracts, such as henna and TQ, in order to avoid their deteriorating effects on the mechanical and physical properties of PMMA.

In the future, nanoparticle antimicrobial agents may become more common due to their potential antimicrobial activities. The use of nanomaterials with larger active surfaces provides antimicrobial effects at much lower doses, without affecting the PMMA structure. Moreover, some antimicrobial fillers are promising in terms of having the required physical and mechanical properties for denture fabrication. Therefore, further investigations are required to evaluate the correlations between the effects

of antimicrobial agents and the surface roughness of the denture base resin. Are the resultant antimicrobial effects due to their antimicrobial activity or their action on the surface properties of the acrylic resin? The answer is very important in order to develop an antimicrobial resin that is satisfactory for dental prosthesis fabrication, with longevity and reasonable clinical applications. It is also important to test the resistance of denture coatings to cleaning methods, including brushing and immersion in denture cleansers, before they are used clinically.[13]

The addition of an antimicrobial material to PMMA has been suggested in order to obtain an antimicrobial denture base resin. However, the percentage of the additive which would be effective without altering the physical properties and biocompatibility of the acrylic resin must be considered. Overall, preventing DS is better than managing it after it occurs. In addition, an antimicrobial material incorporated into the resin may be superior to a coating on the surface of the denture base in the prevention of DS. Further studies are required to test the correlations between the antimicrobial effects of nanoparticles and the surface roughness of the acrylic resin, in addition to its durability and aging effects, under simulated oral conditions.

Conclusions

The present review included studies on the treatment of DS using different approaches. Based on the literature review, it can be concluded that the incorporation of different antifungal agents into the PMMA denture base material can control DS. In addition, coating the denture base resin with antifungal agents and topically applying cleaning agents are both effective in the treatment of DS in vitro. However, most of the published articles were based on in vitro studies, with or without simulating the clinical situations. There is also a lack of studies investigating the long-term effects of these treatment methods, and the relationship between surface properties and nanofiller additives. Therefore, further research is required to answer the remaining questions, such as those concerning the optimal dosage and the controlled release of drugs.

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Complex orthodontic and surgical management of an adult patient with transverse maxillary deficiency and skeletal class III malocclusion: A case report

Kompleksowe leczenie ortodontyczno-chirurgiczne pacjentki dorosłej z niedorozwojem poprzecznym szczęki i III klasą szkieletową – opis przypadku

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Abstract

Skeletal deformities constitute a relatively common structural and functional craniofacial abnormality. The chief complaints reported by patients include a lack of satisfaction with facial appearance, difficulty with breathing or eating, and altered speech. The management of skeletal malocclusion requires a complex orthodontic and surgical approach.

The paper presents the case of a 28-year-old woman with maxillary constriction and skeletal class III malocclusion. Transpalatal distraction (TPD), based on the distraction osteogenesis phenomenon, was used for correcting transverse maxillary deficiency by increasing the maxillary bone base, and therefore the transverse maxillary dimension. The next stage was orthodontic treatment, involving dentoalveolar decompensation, as part of preparation for bimaxillary osteotomy (BIMAX). The last stage of the complex treatment was BIMAX, which ultimately eliminated skeletal defects in both the sagittal and frontal planes. This complex multidisciplinary management significantly improved facial harmony, increased nasal volume, caused a shift from mouth breathing to nasal breathing, and restored normal occlusal relationships.

Key words: orthognathic surgery, orthodontics, skeletal maxillary constriction, skeletal deformities

Słowa kluczowe: chirurgia ortognatyczna, ortodoncja, zwężenie szczęki, wady gnatyczne

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Introduction

Skeletal malocclusion is commonly encountered in orthodontics. These are craniofacial structural abnormalities involving 1 or more planes. As in the case of all structural abnormalities, skeletal defects always cause some degree of dysfunction. The chief complaints reported by patients include a lack of satisfaction with facial appearance, difficulty in breathing or eating, and altered speech.[1–3] A multidisciplinary approach is becoming the treatment of choice due to the orthodontists' growing knowledge of available treatment options for skeletal malocclusion on one hand, and the patients' expectations of improved facial esthetics and function on the other hand.

Orthognathic surgery is a subdomain of craniomaxillofacial surgery, which deals specifically with correcting skeletal malocclusion. Surgical treatment involves typical craniofacial osteotomy procedures and determining proper spatial relationships, followed by the fixation of the osteotomized fragments in the predetermined position by means of osteosynthesis.[4–6]

Another important element in the management of skeletal malocclusion is distraction osteogenesis, used for increasing the bone base in order to lengthen a particular facial bone element. It is achieved by using different extra- and intraoral distractors.[7]

Case report

A 28-year-old healthy female was referred to the Department of Maxillofacial Surgery of Wroclaw Medical University in Poland due to significant malocclusion. The patient brought her current plaster models, orthopantomogram (OPG), lateral cephalometric radiograph (Fig. 1), posteroanterior (PA) cephalogram, and computed tomography (CT) scans for the appointment.

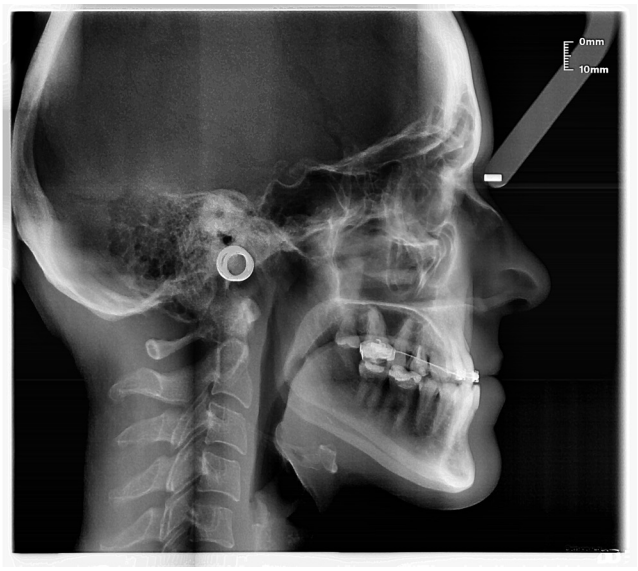

Fig. 1. Lateral cephalogram before treatment

After the clinical assessment and evaluation of the available plaster models, extra- (Fig. 2) and intraoral photographs (Fig. 3), radiographs, and CT scans with three-dimensional (3D) image rendering as well as the cephalometric Segner–Hasund analysis, the patient was diagnosed with:

– class III skeletal malocclusion – maxillary retrognathia and mandibular prognathia;
– vertical and transverse maxillary deficiency – vertical maxillary hypoplasia;
– right mandibular laterognathism;
– dental malocclusion.

The patient was offered a multidisciplinary management plan, involving orthodontic treatment and 2 surgical procedures.

The 1st procedure, after the preliminary orthodontic treatment, was transpalatal distraction (TPD), aiming at increasing the transverse maxillary dimension. The next step, after the decompensation of dentoalveolar malocclusion, was bimaxillary osteotomy (BIMAX), performed in order to achieve the optimal spatial and occlusal relationships of the craniofacial structures, and thus to improve the facial profile. Additionally, the patient was advised on the intervals between the 2 procedures, and educated about the potential complications and difficulties as well as the anticipated treatment outcomes.

After the patient gave her informed written consent, the 1st stage of treatment was commenced.

Due to the skeletal nature of transverse maxillary deficiency and the patient's age, we decided to abandon orthodontic treatment aiming at increasing the transverse maxillary dimension.

Maxillary constriction is clinically manifested as a complete unilateral or bilateral crossbite, a narrow palate and a V-shaped high palatal vault, dark buccal corridors, the compensatory buccal inclination of the lateral teeth, and mouth breathing.[8,9] The presence of at least 2 of the above features indicates the skeletal nature of malocclusion, which determines the subsequent management. Transverse maxillary deficiency jeopardizes the stability of surgical treatment outcomes in patients with skeletal malocclusion.

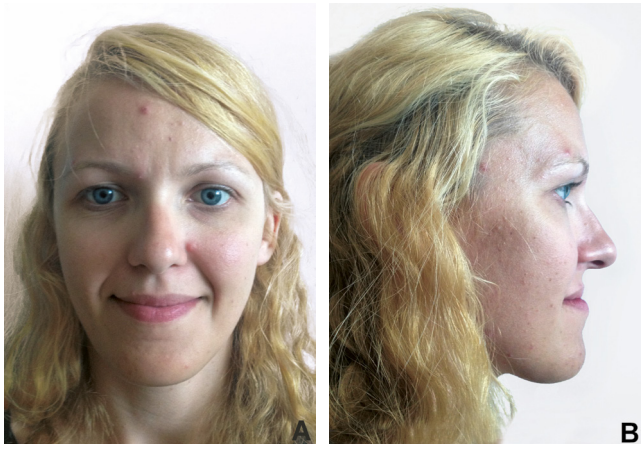

Fig. 2. Facial views before surgery
A – frontal view; B – profile view.

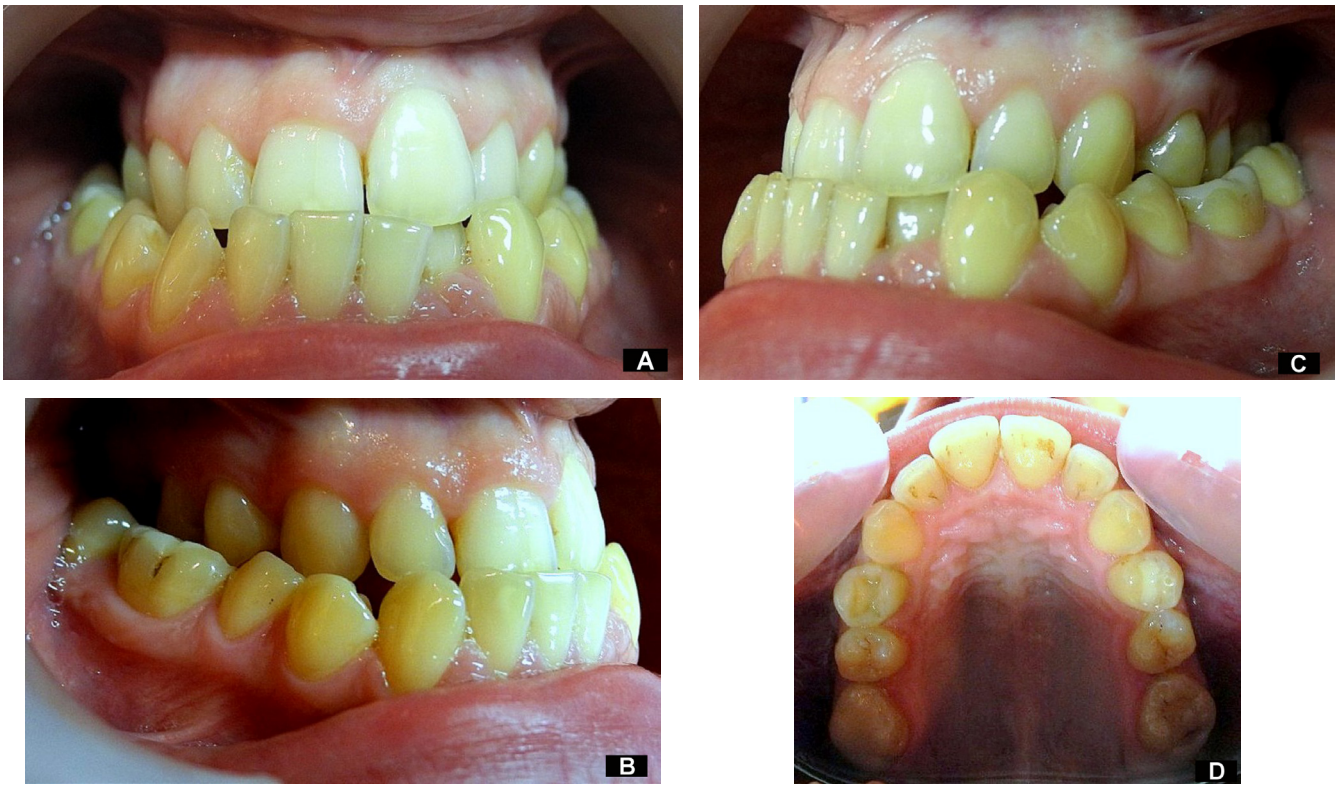

Fig. 3. A–C – occlusal views before treatment; D – maxillary constriction

Transpalatal distraction is one of the approaches used for increasing the transverse maxillary dimension. Introduced by Maurice Y. Mommaerts in 1999, TPD works on a principle of distraction osteogenesis, increasing the maxillary base width and its transverse dimension. What is innovative in comparison with surgically assisted rapid maxillary expansion (SARME) is using a bone-anchored transpalatal distractor as the force-generating device. Placed within the palatal bone, the distractor exerts force on the palatine process of the maxilla only, without any adverse effect on the lateral teeth.[10,11]

As part of preparation for TPD, an upper fixed appliance was used (the Roth system with the 0.018 slot size) and the 0.016 × 0.022 passive stainless steel (SS) archwire (Natural Arch Form III; American Orthodontics, Sheboygan, USA) was engaged.

Next, the surgery was scheduled and performed under general anesthesia. The Le Fort I osteotomy was performed from the symmetrical approaches in the oral vestibule and maxillary labial frenulum, including the bilateral separation of the maxilla from the pterygoid processes of the sphenoid bone and the surgical mobilization of the midpalatal suture. After checking the mobility of the maxillary bones, the size 16 UNI-Smile® transpalatal distractor (Titamed, Kontich, Belgium) was fixed on the hard palate at the level of the second premolars. The abutment plates were located horizontally at 1.0 cm from the gingival margin, perpendicular to the skeletal line of the midpalatal suture. The activation of the intraoperative distractor yielded a 1.0-millimeter-wide diastema. The wound was closed using 4-0 absorbable sutures (Safil®; B. Braun Austria GmbH, Maria Enzersdorf, Austria). Medical treatment involved the administration of peri- and postoperative preventive antibiotic therapy, analgesics, antiedema agents, and anticongestant nasal drops.

The distraction protocol involved the 3 standard stages of latency, distraction and retention. The surgery was followed by a 6-day latency phase. The distractor did not expand spontaneously owing to the blocking screw, placed and tightened intraoperatively. As the blocking screw was removed, the patient was instructed to activate the device herself by performing 2 rotations of the screw (each being a quarter turn) daily, which translates into an expansion of 0.5 mm a day. The duration of the active treatment phase depends on transverse maxillary deficiency. In this particular case, the distraction phase lasted for 15 days. The total number of activations was 29, which corresponded to the overall expansion of 7.25 mm. After the completion of the active treatment, the blocking screw was placed and tightened again. The follow-up X-ray, intraoral photographs and craniofacial CT scan with 3D image rendering were done. Six weeks after the completion of distraction, orthodontic treatment was commenced.

The aim of orthodontic treatment was to reshape the upper arch, close the diastema caused by palatal distraction and reposition the maxillary teeth. Additionally, a fixed appliance was used on the lower arch to achieve the appropriate inclination of the lower incisors and torque of the lateral teeth. As a result, the complete decompensation of skeletal class III malocclusion was achieved.

The distractor was evacuated during an outpatient procedure under local anesthesia. As the retention period ended, we did the follow-up imaging, including OPG, a cranial X-ray PA 0°, cephalometric radiographs, and intraoral photographs (Fig. 4) as well as a craniofacial CT scan with 3D image rendering (Fig. 5).

The next stage of the complex orthodontic and surgical treatment was bimaxillary osteotomy (BIMAX). Historically, the first maxillo-mandibular surgery was performed by Hugo L. Obwegeser back in 1970.[12] Trauner and Obwegeser also developed the commonly used bilateral sagittal split osteotomy (BSSO) in 1955, which was later modified by the Italian surgeon Dal Pont in 1961.[13–15] Further modifications to the BSSO technique were introduced by Hunsuck (1968) and Epker (1977). Nevertheless, the main principle postulated by Obwegeser and Dal Pont has remained unchanged.[16]

Having obtained the extra- (Fig. 6) and intraoral photographs (Fig. 7), OPG (Fig. 8), cephalometric radiographs (Fig. 9), plaster models, and occlusal records from an articulator, the surgery was planned.

Maxillary and mandibular dental casts mounted on an articulator were used as a surgery phantom. During this procedure, interocclusal relationships were recorded and used for making surgical templates – an intermediate occlusal splint, which determined maxillary repositioning, and a final occlusal splint, which determined mandibular repositioning. An anterior maxillary repositioning of 5 mm was planned, with the slight impaction and clockwise rotation (3 mm) of the posterior maxillary segment. A posterior mandibular repositioning of 3 mm was planned, with the slight rotation of the distal segment to the left for midline correction and skeletal symmetry.

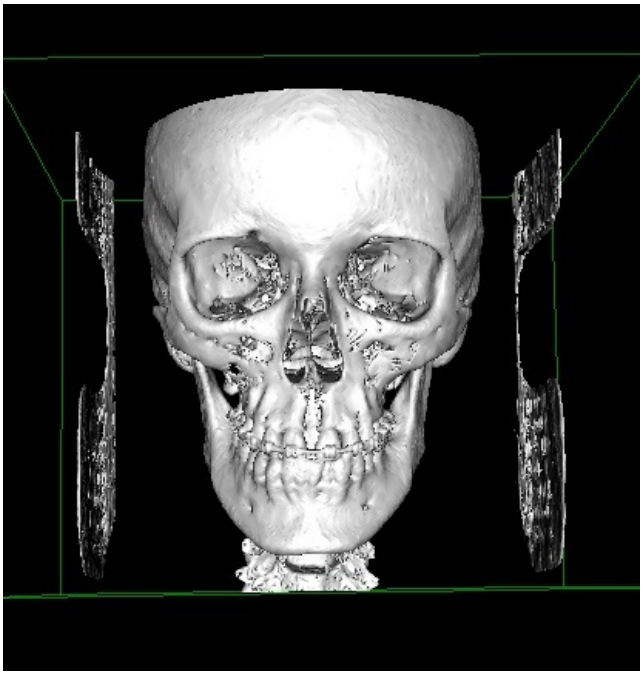

Fig. 5. Three-dimensional (3D) reconstruction of the facial skeleton before the removal of the transpalatal distractor

Prior to the surgery, surgical archwires, i.e., the 0.017 × 0.025 full-sized SS archwires (Natural Arch Form III; American Orthodontics) with additional hooks to attach intermaxillary elastics were applied.

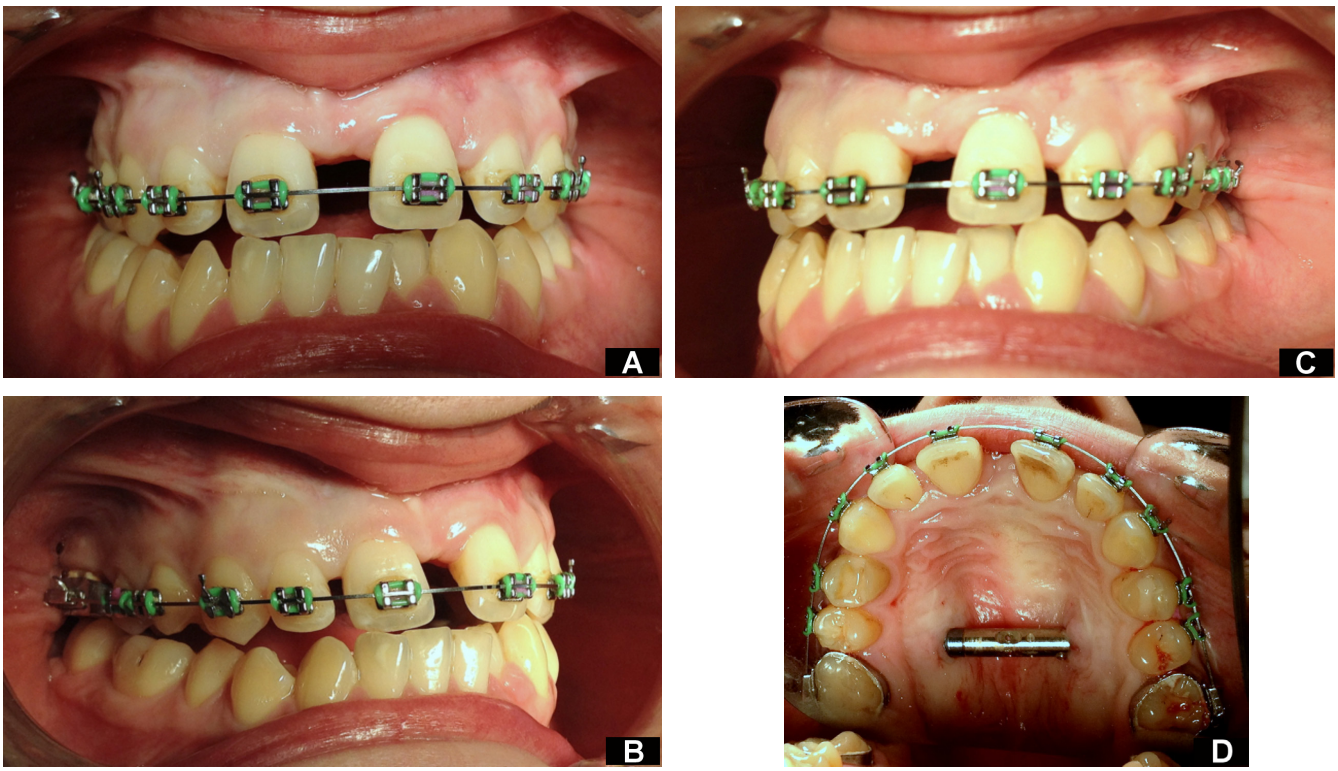

Fig. 4. A–C – occlusion changes after maxillary distraction; D – view of the maxilla after distraction

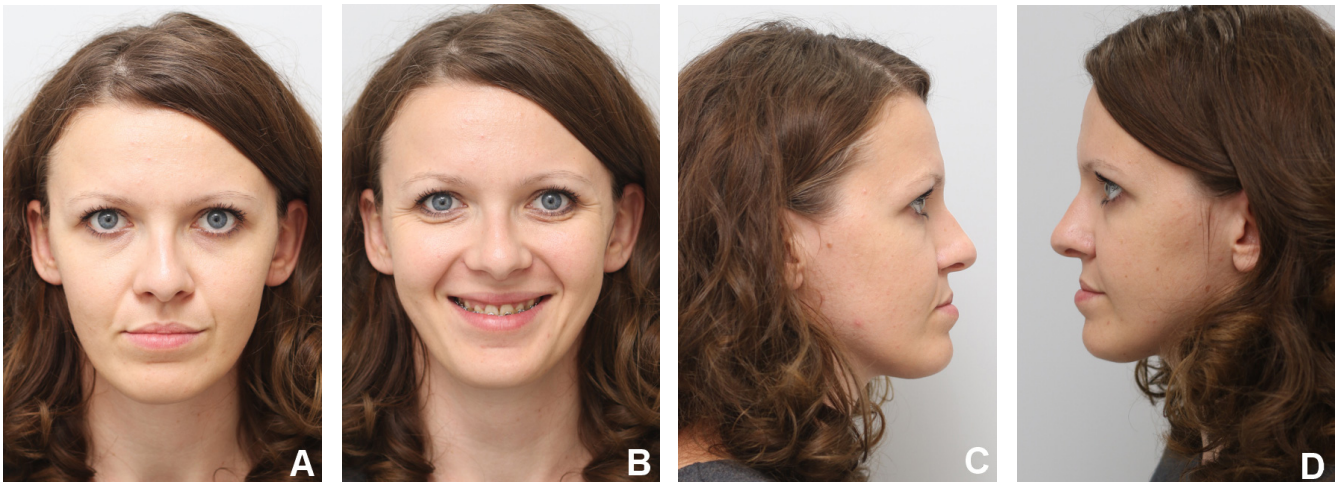

Fig. 6. Facial views before bimaxillary osteotomy (BIMAX)
A – frontal view; B – smile; C, D – profile views.

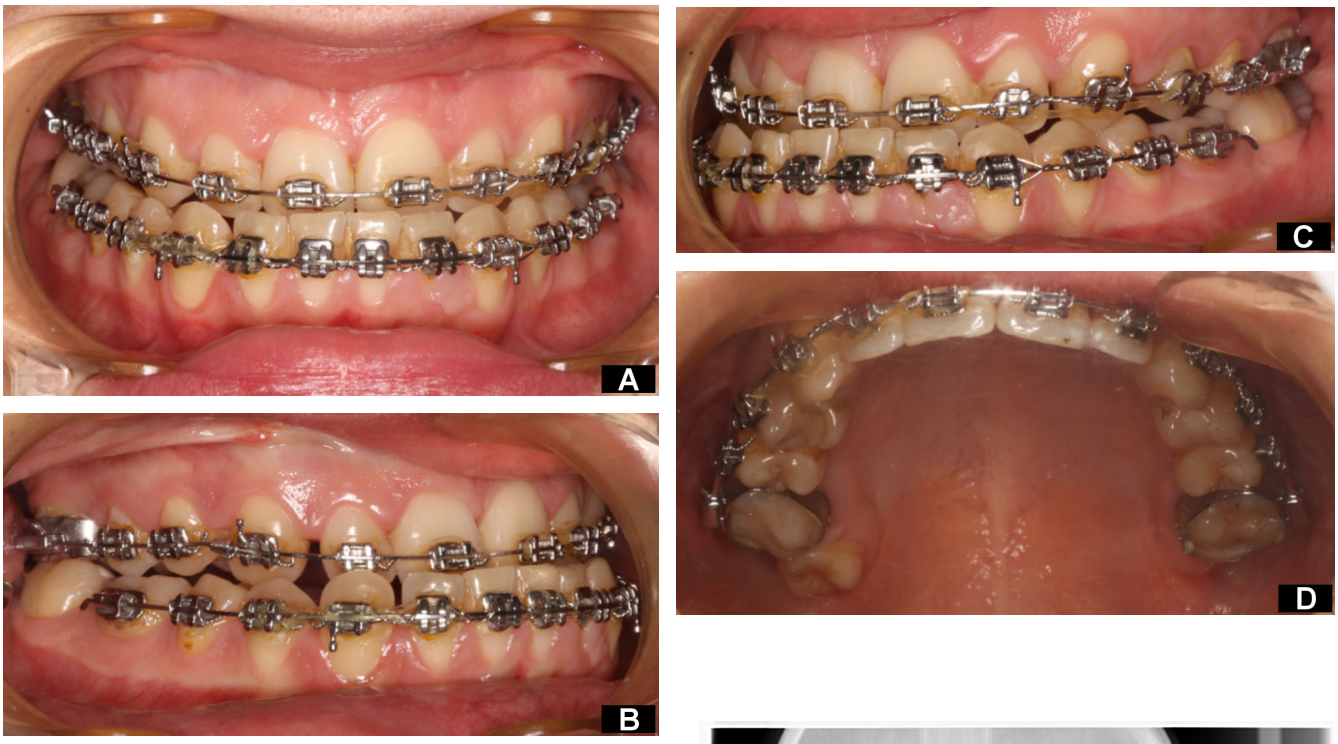

Fig. 7. A–C – occlusion before BIMAX; D – maxilla before BIMAX

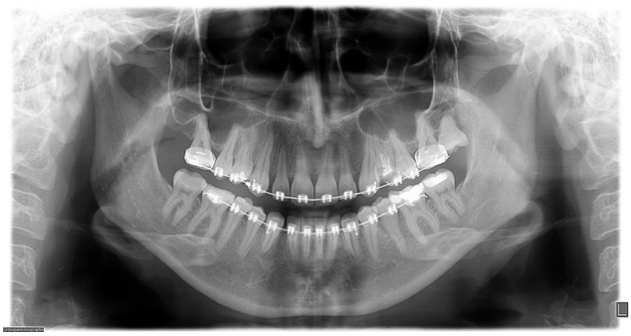

Fig. 8. Orthopantomogram (OPG) before BIMAX

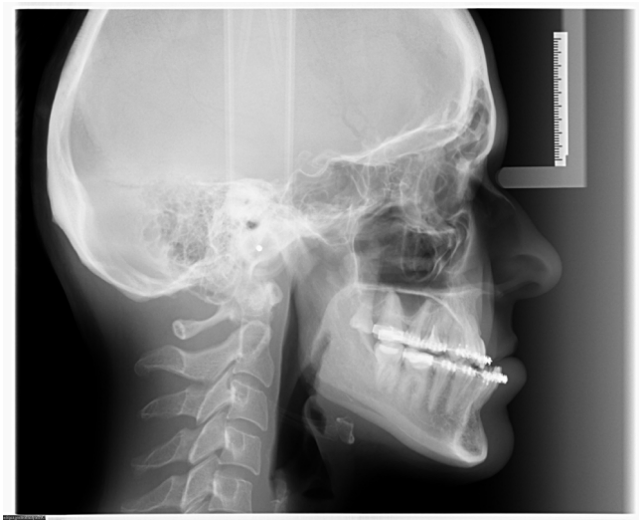

Fig. 9. Lateral cephalogram before BIMAX

The surgery was performed under general anesthesia. The anterior and lateral maxillary surface was exposed using the maxillary vestibular approach. Next, the Le Fort I osteotomy was performed, followed by lateral and septal osteotomy and the separation of the pterygomaxillary junction (PMJ) as well as the down-fracture and mobilization of the maxilla. The osteosynthesis of the bone fragments in the predetermined position was performed using miniplates and screws 2.3 (Titamed). Subsequently, BSSO (Obwegeser–Dal Pont) was performed and the distal segment was positioned as planned. The mandibular fragments were fixed using bicortical screws 2.3 (Titamed), 3 screws on each side. Medical peri- and postoperative treatment involved the administration of antibiotics, antiedema agents and nasal sprays, which lubricated the nasal passages and maintained their patency. In order to provide additional support, class III intermaxillary elastics were used from the 1st day postoperatively. The follow-up OPG (Fig. 10) and cephalometric radiographs (Fig. 11) were also performed.

In the 1st week postoperatively, the patient started a masticatory rehabilitation program, involving the orbicularis oris muscle exercises and passive mouth-opening exercises. In the 2nd week, the patient started mild, pain-free active mouth-opening exercises. In subsequent weeks, the physical therapy program was modified by increasing the exercise frequency as well as the muscular force and the range of motion (ROM) of the temporomandibular joint (TMJ), and simultaneously shortening the wearing time of the intermaxillary elastics.

In the 6th week postoperatively, the surgical archwires were removed and replaced with the archwires enabling the occlusion to settle – the 0.017 × 0.025 nickel-titanium (NiTi) archwire (Form III NT3™ SE NiTi; American Orthodontics) and the 0.017 × 0.025 Turbo Arch NiTi archwire (Ormco™, Chisinau, Moldova). The patient was advised to wear the intermaxillary elastics at night. While settling the occlusion, the premature occlusal contacts and occlusal barriers were corrected so as to ensure proper resting and functional occlusal relationships (Fig. 12). The complex surgical-orthodontic treatment led to the improvement of facial features (Fig. 13).

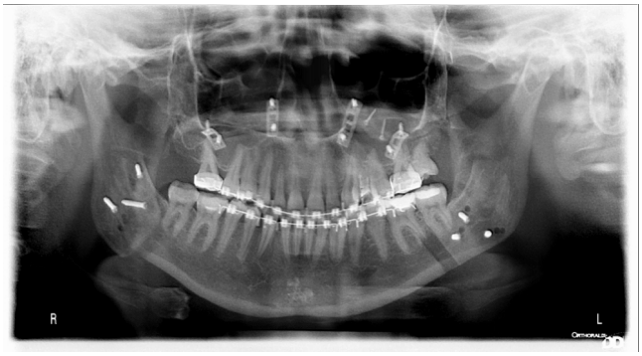

Fig. 10. OPG after BIMAX

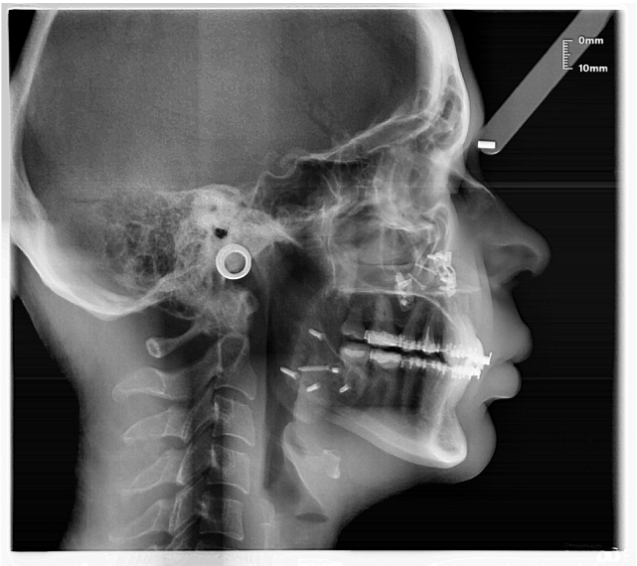

Fig. 11. Lateral cephalogram after BIMAX

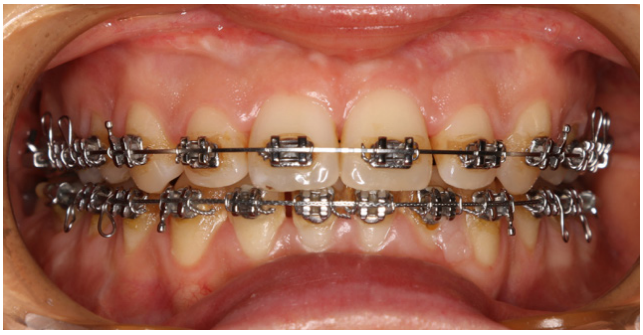

Fig. 12. Occlusion after treatment

Fig. 13. Facial views after treatment
A – frontal view; B,C – profile views.

Conclusions

A complex orthodontic and surgical approach is a key to the successful correction of skeletal malocclusion. It is crucial to take time to educate the patient on the assumptions of this multidisciplinary approach and provide all necessary explanations. This complex orthodontic and surgical management significantly improved facial harmony, increased nasal volume, caused a shift from mouth breathing to nasal breathing, and restored normal occlusal relationships. Despite its complexity and duration, in the case of severe skeletal malocclusion, multidisciplinary treatment is a method of choice, aiming at determining and correcting the underlying cause of the abnormality, which in turn offers a better chance of achieving a stable effect and patient satisfaction.

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Sagittal mandibular osteotomy in a patient with Eisenmenger's syndrome: A case report

Strzałkowa osteotomia żuchwy u pacjenta z zespołem Eisenmengera – opis przypadku

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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

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Abstract

The various abnormalities of occlusion cause significant discomfort to the patient suffering from them. Currently, the surgical treatment of malocclusion in healthy patients is a routine process. The situation is completely different when the patient has a serious disease of the respiratory or cardiovascular system – a condition which may contraindicate such treatment.

A 30-year-old female patient, with a class III skeletal defect (open bite and progeny) and Eisenmenger's syndrome was chosen as a clinical case. The DDS-Pro software was selected to plan the operation. The bilateral sagittal split osteotomy of the mandible was selected as the method of surgery. At the time of the initial examination, the patient had been denied surgical treatment several times in several other clinics. Before the treatment began, the patient underwent intensive preparation in the cardiac surgery unit for 2.5 months. Using the software, a surgical intervention was planned with the production of a surgical template. The operation was then performed; the treatment period was unremarkable. The resulting occlusion and changes in the shape of the face fully met the patient's psychological and esthetic expectations.

In conclusion, a complicated cardiovascular pathology does not always deprive patients with malocclusion of the possibility to undergo surgical treatment.

Key words: orthognathic surgery, malocclusion, progeny, Eisenmenger's syndrome, osteotomy of the mandible

Słowa kluczowe: chirurgia ortognatyczna, wada zgryzu, progenia, zespół Eisenmengera, osteotomia żuchwy

Introduction

The human face has always received special consideration as a social signal which performs both informative and communicative functions. Unfortunately, the human face may be distorted by the presence of various congenital skeletal deformities, such as bite anomalies. The appearance of congenital abnormalities, especially in the maxillofacial area, is caused by different risk factors or genetic disorders during prenatal development.[1] These disorders may be the source of esthetic and psychological discomfort.

However, the presence of concomitant – most often congenital – somatic pathologies in orthognathic patients greatly reduces their chances of receiving any necessary surgical treatment in general, and the correction of facial defects in particular. These are mostly the diseases of the respiratory and cardiovascular systems. The presence of one of these pathologies increases to a great extent the risk of nonsurgical bleeding and complications during general anesthesia.[2] Moreover, these are high-risk patients for small clinics and they require treatment in large hospitals, equipped with full-sized resuscitation departments. The situation is further complicated by the fact that orthognathic surgery is considered a medical procedure, and not urgent care. It therefore depends on the implementation of the scheduled interventions and may be delayed indefinitely.

Eisenmenger's syndrome is a life-threatening condition, requiring careful medical monitoring. It is a congenital cardiac defect, commonly caused by a shunt between the 2 ventricles of the patient's heart. Eisenmenger's syndrome occurs when the increased pressure of the blood flow in the lung becomes so great that the direction of the blood flow through the shunt reverses. Oxygen-poor blood from the right side of the heart flows into the left side of the heart and is pumped throughout the patient's body, causing insufficient oxygen supply to all organs and tissues.[3]

Today, thanks to the development of medical science, surgical treatment to correct the facial shape and size has become a routine process. Modern orthognathic surgery enables complex interventions, consisting of diagnostic and planning measures, surgery, and patient rehabilitation. Currently, the standard interventions for bite correction include the Le Fort I osteotomy of the maxilla, the bilateral sagittal osteotomy of the mandible and the compression-distraction techniques.[4] It should also be noted that osteotomy is less costly for the patient than the compression-distraction techniques, being at the same time equally effective; due to the intraoral surgical approach, it does not distort the patient's face and is less time-consuming than the compression-distraction techniques. The bilateral osteotomy of the mandible has been recognized as an excellent technique for the correction of severe open-bite cases when the patient has a normally developed mandible and no indication for bimaxillary osteotomy.[4,5]

Planning is an important and precise part of such interventions. After this stage, the surgeon knows exactly which bones to remodel and how to proceed in order to achieve the best results.[6,7]

Taking into account all of the above, this article presents our experience using a multidisciplinary approach to the orthognathic treatment of patients with maxillofacial malformations in combination with a severe concomitant pathology.

Case report

A young woman, aged 30, visited our clinic in October 2017. She had been diagnosed with congenital open bite (a class III skeletal facial deformity, characterized by a significant esthetic and functional defect) as well as Eisenmenger's syndrome with functional class II–III pulmonary hypertension. The patient had been denied surgical treatment several times in other clinics due to the complexity of the deformity and the presence of severe concomitant diseases. A thorough examination by our team revealed that it was possible to perform an orthognathic operation after proper preparation and planning. Written consent was given. The patient was examined by a psychologist and her psychological state was defined as satisfactory. Throughout the preparation phase, she remained under the supervision of a cardiologist and a cardiac surgeon. She underwent oxygen therapy and was treated with sildenafil (Strondex®; Microkhim Pharmaceuticals Ltd., Rubizhne, Ukraine). Warfarin, which she ingested daily, was discontinued in order to increase blood coagulation 3 weeks before surgery and 5 days after. At the beginning of the preparation phase, the thromboplastin sensitivity, measured as the International Normalized Ratio (INR), was 4.1. A re-examination was carried out 2.5 months later. As INR was found to be 1.8, we decided that the patient was ready for surgery (Fig. 1,2).

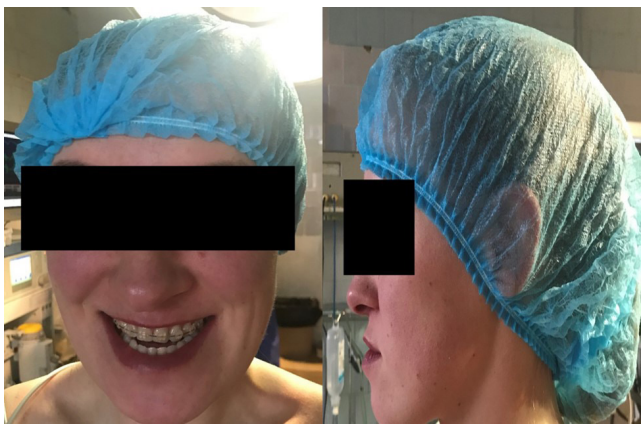

Fig. 1. Appearance of the patient's face before osteotomy

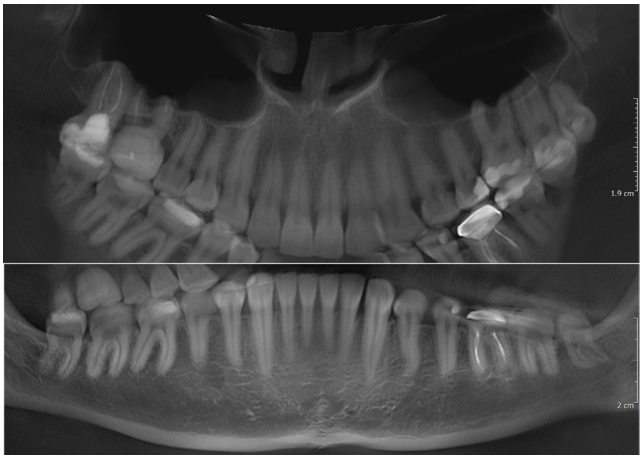

Fig. 2. The patient's orthopantomogram (OPG) before osteotomy

For virtual intervention planning, we used the DDS-Pro software from Digital Dental Services® (London, UK), which simulates operations with a high degree of accuracy and creates surgical templates that can be printed on a three-dimensional (3D) printer. The choice of the planning software was not random. Our team has successfully used the DDS-Pro software for 3 years.[8] The 1st step consisted of scanning the cast models of the patient's upper and lower jaws with a 3D scanner. These files, along with pre-operation spiral computed tomography (CT) with a slice thickness of 0.5 mm, were loaded into the DDS-Pro software, where a 3D skull model was created (Fig. 3). The simulation of bilateral sagittal split osteotomy was performed according to the above-mentioned technique, moving the mandible backward – 7 mm to the right, 4 mm to the left – with a simultaneous left rotation of 5° and 2° upward. The resulting occlusion was verified (Fig. 4). All of the fragments were fixed together and a virtual 3D bite template was created (Fig. 5). A new preoperative 3D skull model was created for the patient and the surgical template was checked virtually (Fig. 6). The procedure was printed out, and the template was tested on the cast models (Fig. 7) and in the patient's oral cavity, on both the maxilla and mandible.

Subsequently, the operation – the bilateral sagittal split osteotomy of the mandible (using the Obwegeser II technique) with the single-stage removal of teeth 38 and 48 – was performed in the cardiac surgery unit under endotracheal anesthesia using sevoflurane gas. After the mandible was incised, the tooth row in each jaw was reduced to the expected occlusion position using

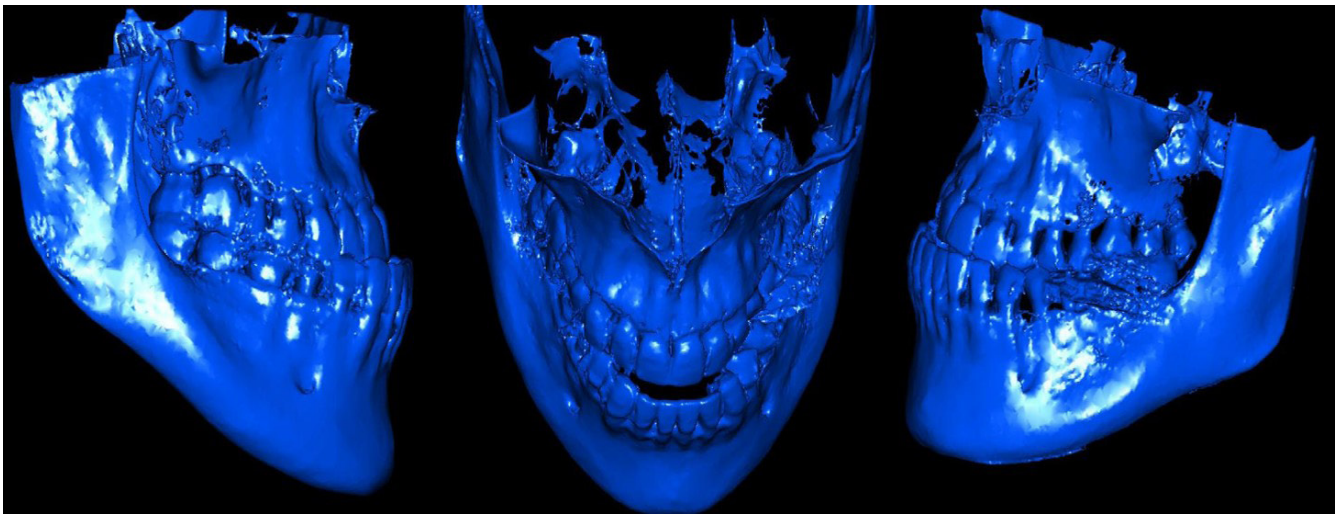

Fig. 3. The patient's initial three-dimensional (3D) skull model (right, top and left views)

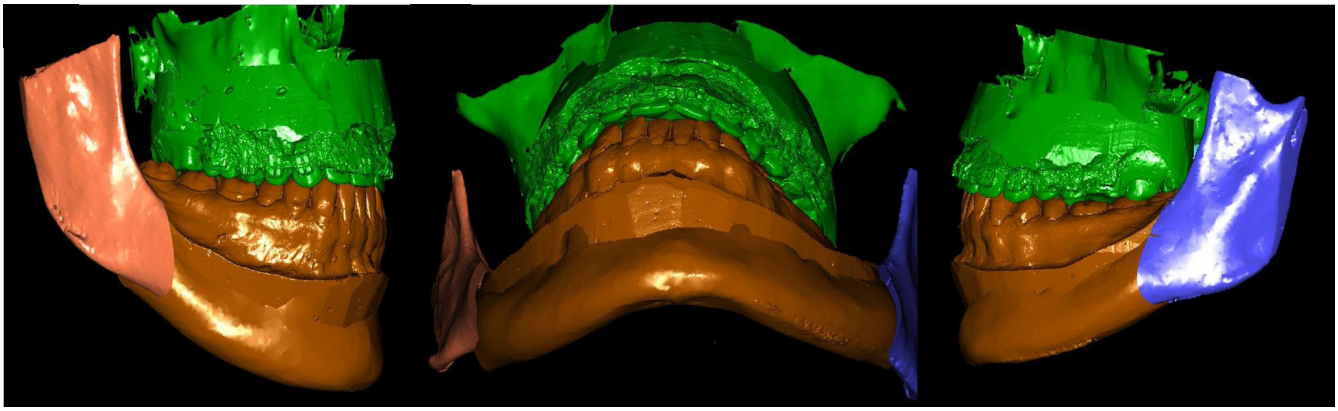

Fig. 4. Simulation of the surgical intervention (right, top and left views)

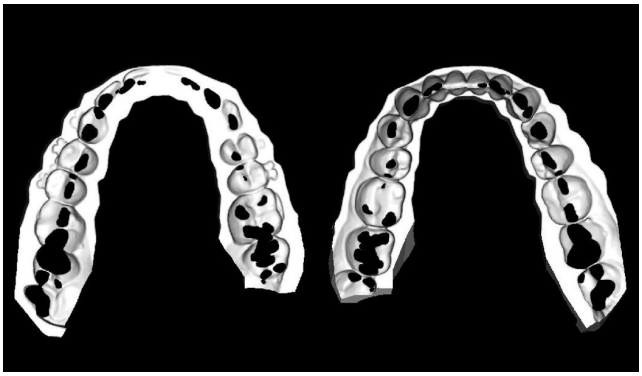

Fig. 5. Surgical template

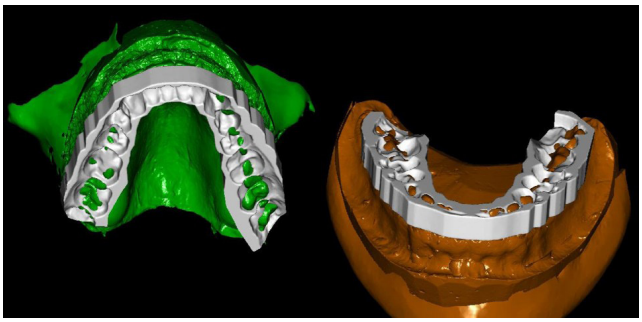

Fig. 6. Overlaying the surgical template on the 3D model

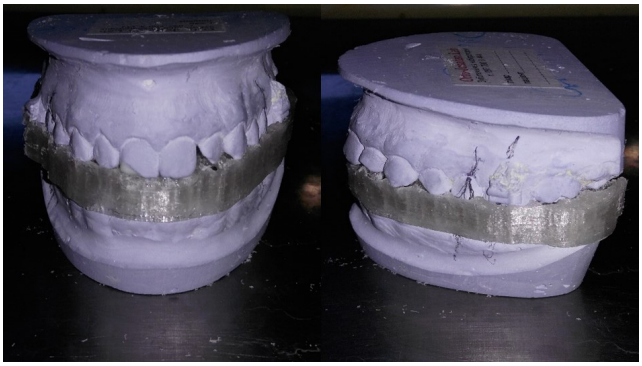

Fig. 7. Surgical template on preoperative models

the prepared template. In our case, mandibular fixation was performed according to Champy et al., using 2.0-mm miniplates and screws produced within the IRENE® OsteoMed system (Tianjin Zhengtian Medical Instruments Co., Ltd., Tianjin, China).[9] The IRENE intermaxillary fixation screws with orthodontic chain elastics were used for maxillo-mandibular fixation in the postoperative period.

The postoperative treatment period was unremarkable. The patient was under strict observation in the intensive care unit for the first 10 days following the surgery. On day 20, the patient was discharged and assigned post-hospital follow-up. The postoperative orthopantomogram (OPG) and photos of the patient's face were taken (Fig. 8,9). The resulting occlusion and changes in the facial shape fully met the patient's psychological and esthetic expectations.

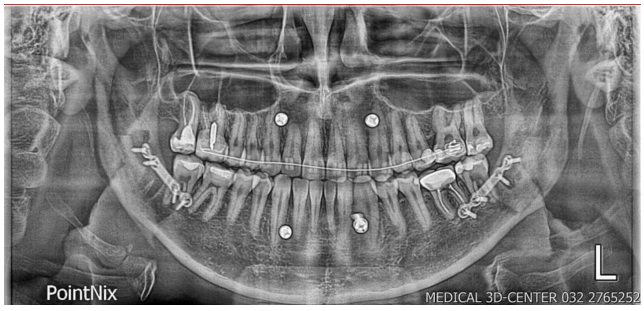

Fig. 8. The patient's postoperative OPG

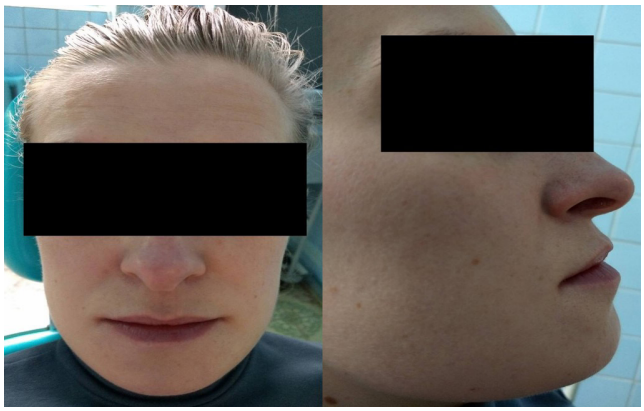

Fig. 9. Appearance of the patien't face (frontal and profile views) 3 weeks after surgery

The 1st follow-up examination of the results was carried out 4 months after the orthognathic surgery. No deviations or complications were detected (Fig. 10). The final follow-up was conducted in May 2019 (15 months after the surgery). The patient did not notice any changes in bite. She was satisfied with the esthetic and functional outcome (Fig. 11). The clinical examination showed stable fixed occlusion throughout the dental arch. No pathological mobility was observed in the osteotomy area. The subsequent OPG showed healing at the osteotomy sites; miniplates and screws were fixed and stable (Fig. 12). The patient's follow-up continues.

Our study was conducted in accordance with the principles of the Declaration of Helsinki and the relevant laws of Ukraine, and it was approved by the Commission of Ethics of Danylo Halytsky Lviv National Medical University in Ukraine (Protocol No. 6/2019).

Discussion

Before the examination, we conducted a literature search for reports on the combined effects of maxillofacial and cardiovascular pathologies. As expected, there was not much information. We found only a few articles, but none were devoted to orthognathic patients with Eisenmenger's syndrome. Hupp presented a report on the successful treatment of a maxillary hemangioma using embolization in a patient with Eisenmenger's syndrome.[10]

Fig. 10. The patient's face (frontal and profile views) and bite 4 months after surgery

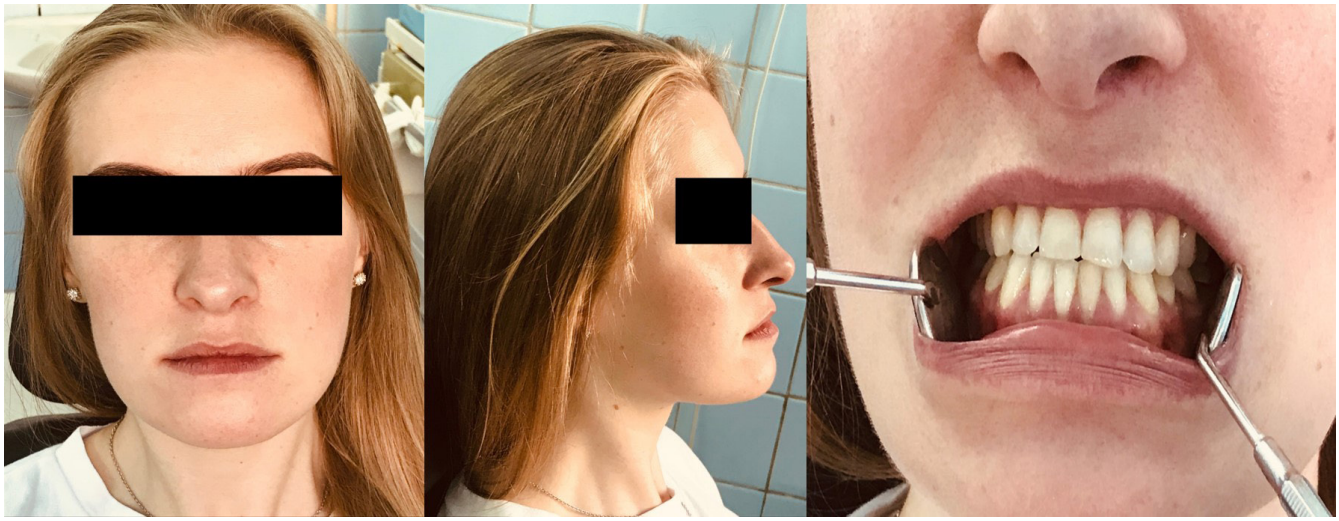

Fig. 11. The patient's face (frontal and profile views) and bite 15 months after surgery

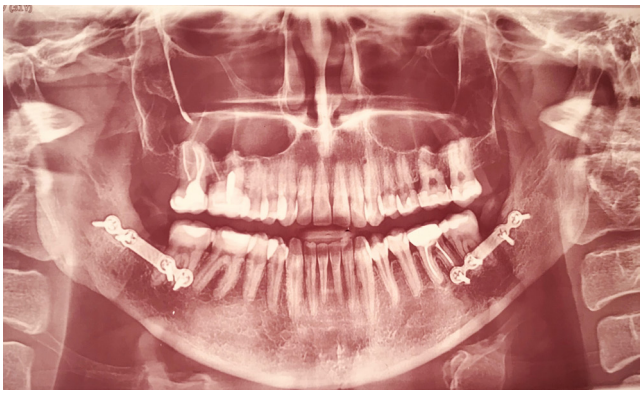

Fig. 12. The patient's OPG on the 15th-month follow-up visit

The author mentioned that in this particular case, the presence of Eisenmenger's syndrome added an challenge to the administration of anesthesia, but the operation and anesthesia were performed without complications.[10] Raines et al. presented a report on the successful outcomes of various non-cardiac surgeries performed on 12 patients with Eisenmenger's syndrome using different types of anesthesia.[11] The authors emphasize that anesthesia can be safe when different types of anesthesia and medication are applied. In their opinion, the expected mortality rate for patients undergoing non-cardiac surgery may be up to 10%, which is lower than the mortality rate for parturient women after birth or cesarean section.[11] Another group of surgeons presented the case of a 38-year-old female who was admitted with cardiac failure and Eisenmenger's syndrome, and underwent total abdominal hysterectomy.[12] She was stabilized preoperatively with digoxin and diuretics. She was successfully managed with general anesthesia, the neuraxial blockade for pain relief, and perioperative bronchodilators and oxygen therapy.[12] This information was good news for our team. After the careful analysis of the abovementioned studies, we reached a consensus on surgical planning and organization.

The issue of which surgical technique to use was easier to resolve: we decided on the standard technique. The advantages of the selected method are described below.

For example, in their study of 2 groups of patients treated with osteotomy and distraction, Van Strijen et al. convincingly proved that distraction osteogenesis required a longer operation time (by 37% on average), but 1 day less of hospital stay than bilateral sagittal split osteotomy.[13] The surgical cost of distraction osteogenesis was 36% higher than in the case of conventional sagittal split osteotomy.[13] Baas et al. mentioned that patients experienced more pain after distraction osteogenesis, required more analgesics in the postoperative period and had more infections than the bilateral sagittal split osteotomy group.[14] Moreover, Vos et al. reported a lack of significant differences in mandibular stability a year after treatment between 2 groups of patients where remodeling was performed – with bilateral sagittal osteotomy in one group and using the distraction technique in the other.[15]

It is clear that osteotomy requires surgeons to damage jaw integrity, i.e., to create an artificial fracture; it is extremely important to choose the appropriate technique and tool for fixation. This aspect becomes even more important when bilateral osteotomy is performed; 2 fractures require equally reliable fixation. In their study, Collins et al. compared standard 2.0-mm monocortical plates with 2.0-mm locking plates in the treatment of mandible fractures.[16] They concluded that the mandible fractures treated with 2.0-mm locking plates and standard 2.0-mm plates presented similar short-term complication rates. Accordingly, our decision on the choice of the surgical technique was simple and definite.

Summarizing all of the above, we can say that the presence of complicated cardiovascular pathologies should not deprive patients with malocclusion of the possibility of undergoing surgical treatment. A multidisciplinary approach to the preoperative preparation of patients with cardiovascular diseases and dentoalveolar anomalies is effective, and can reduce the number of patients who are refused for orthognathic surgery.

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Treatment of an oronasal fistula in a patient on bisphosphonate therapy: A case study

Leczenie przetoki ustno-nosowej u pacjenta przyjmującego leki z grupy bisfosfonianów – opis przypadku

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Abstract

Bisphophonates (BPs) are a group of drugs used in treating bone diseases, which may lead to the development of the osteonecrosis of the jaw (ONJ). The negative impact of BPs on angiogenesis is among the causes of ONJ. The specific mechanisms of complications are unknown. What is taken into consideration is the trauma background, which, in combination with the implemented BP treatment, can induce bone necrosis. One of the possible consequences of necrotic change progression is the development of an oronasal fistula. Treatment generally requires a surgical intervention.

The paper describes the course of treatment of an oronasal fistula in a patient with BP osteitis, currently using an upper denture. The fistula arose a year after the removal of a protruding sequestrum in the region of the hard palate. An attempt was made to treat the fistula by the mobilization of soft tissues from the palate and the bilayered closure of the fistula with the use of a pedicled connective tissue graft on the greater palatine artery, along with a Tinti–Parma-Benfenati (TPB) flap. The patient was subjected to appropriate post-procedural measures. Regular follow-ups did not reveal any abnormalities in the course of healing.

The use of the abovementioned procedure proved to be an effective method of treatment of an oronasal fistula. The use of a pedicled connective tissue graft for the closure of the oronasal fistula caused by BP therapy had a significant effect on the treatment outcome.

Key words: bisphosphonates, oronasal fistula, Tinti–Parma-Benfenati flap

Słowa kluczowe: bisfosfoniany, przetoka ustno-nosowa, płat Tinti–Parma-Benfenati

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Introduction

Drugs from the bisphosphonate (BP) group are nowadays used as the important complementary treatment of bone diseases,[1] especially in oncology – as the supportive therapy of the main disease. Usually BPs are used as supplemental therapy for breast, prostate and kidney cancer and their metastases, osteoporosis, Paget's disease, hypercalcemia of various etiologies, osteogenesis imperfecta (brittle bone disease) as well as for congenital bone fragility, caused by the mutations of the collagen type I gene.[2–8] The advantages of BPs are widely known and the drugs are continuously used despite their side effects. According to the Cancer Research UK charity organization, benefits from the use of BPs constitute an overwhelming majority as compared to side effects, described as moderate, and rarely severe.[9] Therefore, the application of BPs seems to be certain and unwavering.

In addition, BPs are being increasingly used in the treatment of proliferative diseases (e.g., multiple myeloma) and cancerous diseases with metastases to bone tissue (including lung, breast and prostate cancer).[10]

However, the complications caused by chronic BP consumption are becoming a growing problem, the most common being necrosis within the jaw bones and the formation of bone sequestra.[10]

Bisphosphonates, as a group, can be categorized based on multiple criteria, including the presence of a nitrogen atom in the molecule.[1,5,6,8]

Non-nitrogen-containing bisphosphonates (non-N-BPs), such as tiludronate, etidronate and clodronate, are ingested by osteoclasts and block cellular metabolic pathways by releasing a toxic methylene-containing adenosyne triphosphate (ATP) analog; for example, clodronic acid (dichloromethylene bisphosphonate) may be metabolized to a non-hydrolyzable ATP analog – adenosine 5'-(beta, gamma-dichloromethylene) triphosphate – which is likely to block the ATP-using enzymes and induce apoptosis.[1,5,7,8]

Nitrogen-containing bisphosphonates (N-BPs), such as zoledronate, act through a more complex mechanism and block the mevalonate pathway by inhibiting farnesyl pyrophosphate synthetase, leading to the prenylation of small guanosine triphosphate (GTP)-dependent protein molecules (most likely GTPase-activating protein), which are important for the activity and survival of osteoclasts. Consequently, bone resorption ceases, specifically of its mineral matrix, and the production of bone morphogenic proteins (BMPs) and insulin-like growth factor 1 (IGF-1) is decreased, along with the transformation of stem cells into osteoblasts.

The literature on the subject is rich in reports on BP-dependent bone necrosis in the craniofacial region. This leads to the isolation of a new medical condition called bisphosphonate-related osteonecrosis of the jaw (BRONJ).[11]

In 2007, bisphosphonate osteonecrosis was defined as an exposed bone fragment that does not heal spontaneously during a time period of 8 weeks and is not the result of radiotherapy.[6,12]

The mechanisms of dead tissue formation in the absence of blood supply are known. The bone is supplied with nutrients by the periosteum. In cases of soft tissue injury (e.g., the use of removable dentures, a surgical intervention involving the dissection or elevation of the periosteum from the compact bone), blood circulation is disturbed. In a healthy organism, repair is quick; local necrosis is a direct stimulator – on the cellular level – for the formation of new vessels and connections on the periosteum–bone border, which leads to tissue regeneration and the restitution of the proper blood supply. Due to the negative influence BPs exert on vasculogenesis, rapid repair and the restoration of vascularity are not observed. For the same reason, simple procedures like tooth extraction can also lead to BRONJ. However, our observations regarding the antiangiogenic action of BPs did not explain the necrosis of the jaws. We would like to emphasize that other antiangiogenic drugs (i.e., thalidomide) did not cause such a complication.[13]

An extremely important factor affecting the effectiveness of BP therapy is the route of administration of the drug. In the case of oral administration, the absorption of the preparation is about 10%, with the remainder being excreted in an unchanged form by the kidneys. If BPs are administered parenterally, their absorption increases to 50%, which significantly affects the process of drug accumulation in bone tissue.[8]

However, the exact mechanism of BRONJ formation is not fully known. The described research methods referring to the evaluation of pharmacological effects on osteoclasts, despite the involvement of many facilities with different degrees of reference, have not elucidated the mechanism of BRONJ yet. The accepted trauma hypothesis is insufficient, as sequestra are also identified beyond the place directly exposed to injury.[6,7]

The treatment of BRONJ within the oral cavity, a site constantly subjected to mechanical injury (mastication, the use of prosthetics) and also of diverse physiological flora, is exceptionally demanding.[5–7,11]

The progression of BRONJ is similar to that of typical osteitis, usually appearing in the mandible, primarily due to poor blood supply as compared to the maxilla.[14,15]

Initially, in the acute inflammation phase, no bone meshwork abnormalities and no X-ray changes are observed. The abovementioned features appear after 10–12 days of the inflammatory process.[14]

X-ray imaging shows an increase in bone permeability to X-rays, with the uniform disappearance of the bone or a meshwork resembling mole holes. There may also be areas of denser bone structure within the lesions with an increased permeability to X-rays. They comprise the so-called bone islands that have not undergone resorption and are termed bone sequestra. In the case of chronic bone inflammation, areas of density typical for bone tissue are observed as the surroundings of sites with a decreased tissue density. They arise as the result of the reaction of the

bone to the inflammatory process, which is characterized by an increased growth of the trabecular meshwork.[14,15]

Surgical treatment includes the removal of dead tissues, while retaining the regions of the bone that exhibit correct clinical appearance. The scope of the procedure determines the appearance of bleeding on the entire surface of the exposed bone surrounding the sequestrum. Often, the outcome is the formation of an oroantral or oronasal fistula.[5,7,11,14,15]

Case report

The patient, aged 77, with oncological history (prostate cancer treated since 2005, BP therapy from 2005 to 2009, zoledronic acid administered as supplemental therapy), was subjected to the removal of a sequestrum from the region of the hard palate. The patient used a full denture for the upper and lower jaws. The surgery was performed in hospital conditions at the maxillofacial surgery ward in September 2017. After hospitalization, the patient was monitored for 2 months in the clinic, with the purpose of regularly replacing the surgical dressing and wound care. Correct healing, without complaints of pain, was observed during follow-up visits. In February 2018, a clinical examination did not reveal any features of fistulation and the patient did not report oronasal leakage. The patient requested a checkup in September 2018 due to the transfer of fluids from the oral cavity to the nasal cavity. A clinical examination confirmed the separation of the wound margins and the formation of an oronasal fistula (Fig. 1).

The patient was qualified for surgical treatment. Antibiotic protection was implemented for 1 week before the planned procedure (Clindamycin (MIP Pharma Polska Sp. z o.o., Gdańsk, Poland) 600 mg tablet, 1 tablet every 12 h), which was continued for 2 weeks after the procedure.

The surgical procedure was performed under local anesthesia using a 2% solution of lignocaine with norepinephrine in clinic conditions. The described procedure was carried out using an optical magnifying device – a binocular loupe (×3.3 magnification). The surgery and healing proceeded without general or local complications.

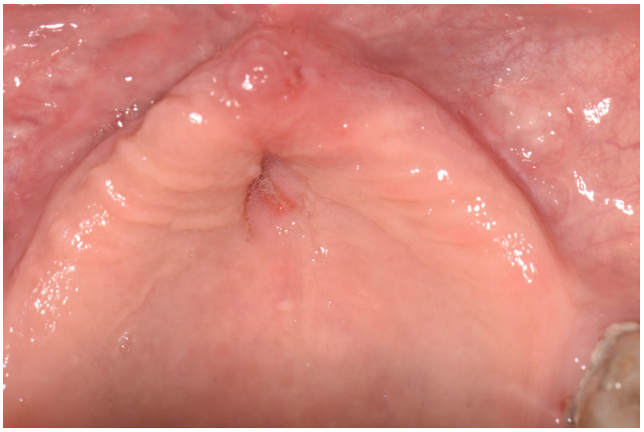

Fig. 1. Status before surgery showing the initial stage

Dissection of the fistula

After local anesthesia was administered, the preparation of the borders of the oronasal fistula was carried out using the generally accepted methods. The revealed bone tissue was unchanged, vividly bleeding. It was, therefore, decided to limit the preparation of the wound edges to the borders of the epithelial layer of the mucous membrane of the hard palate (Fig. 2).

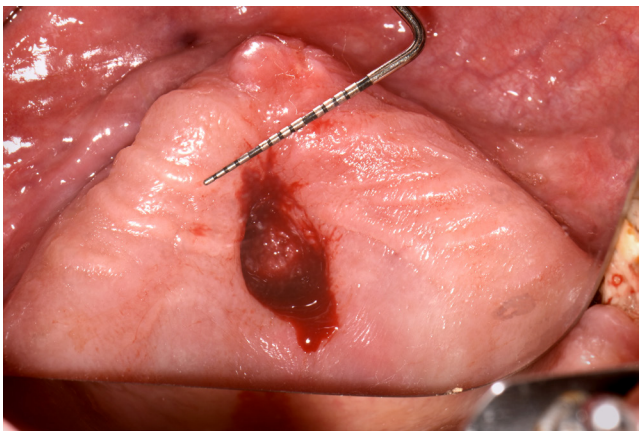

Fig. 2. Excision of the fistula

Pedicled graft

Two parallel releasing incisions were made from the area of the maxillary tuber to the edge of the fistula. The width of the flap amounted to approx. 1 cm (Fig. 3). During the preparation of the divided flap, particular attention was paid to the location of the greater palatine foramen and greater palatine artery. The incisions in the distal part of the flap were made at a distance of about 5 mm from the visible structures (Fig. 4). Then, the greater palatine artery was dissected and the separated connective tissue flap was released, as it was held on the pedicle of this vessel (Fig. 5).

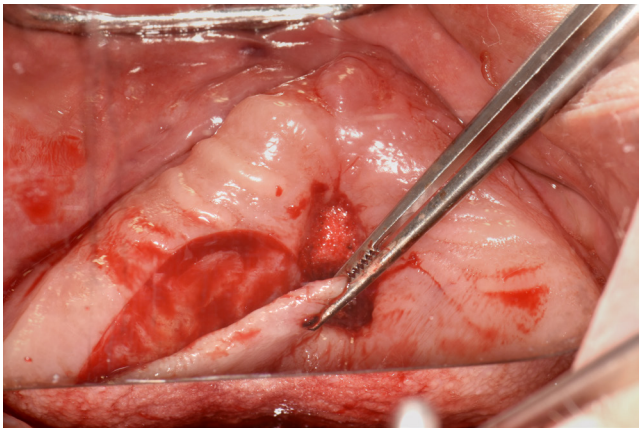

Fig. 3. Preparation of the split flap on the right side

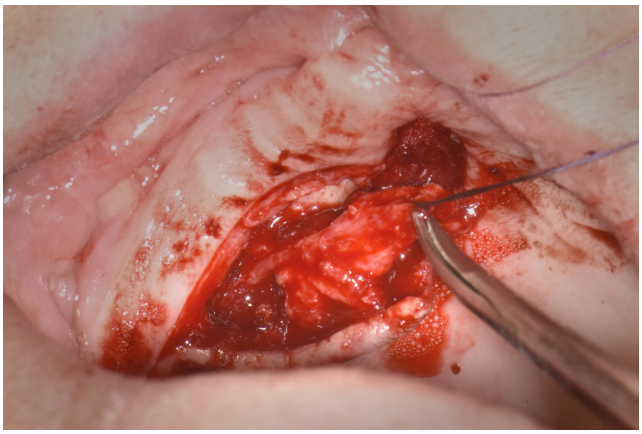

Fig. 4. Preparation of the split flap (visualization of the connective tissue)

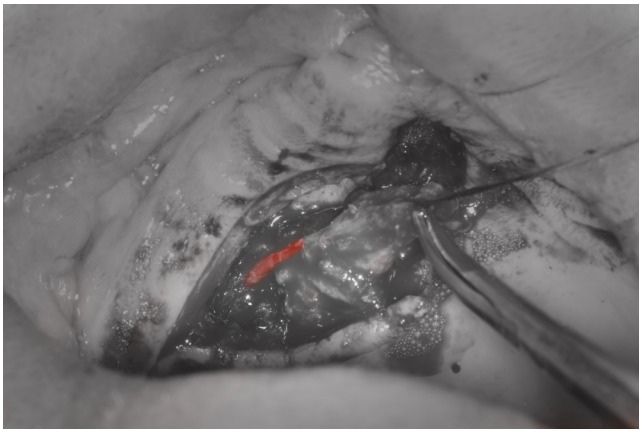

Fig. 5. Visualization of the pedicle made of the greater palatine artery (red)

Tinti–Parma-Benfenati graft

In the 90s, Tinti and Parma-Benfenati proposed a new technique for the implementation of the split graft. The procedure is based on shifting the tissue flap toward the lesion by performing 2 horizontal incisions, parallel to each other, carried out on 2 levels (on the cross-section resembling the letter Z) (Fig. 6). The more superficial incision is done just below the epithelial layer, from the side of the defect. The deeper incision runs under the connective tissue at the base of the graft. Perpendicular releasing incisions should be executed according to the applicable rules of periodontal surgery. It should be noted that a microsurgical scalpel blade was used for the horizontal releasing incisions (dental spoon blade; MJK Instruments, Marseilles, France), an invaluable advantage of which is the ability to shape the bend (i.e., the blade-bend angle) using pliers, according to the demands of the procedure.

Wound suturing

The wound was sutured in layers. The pedicle connective tissue flap supplied by the greater palatine artery was moved above the fistula and was stabilized using an absorbable braided suture (3-0 PGLA LACTIC®; Yavo Sp. z o.o., Bełchatów, Poland). The connective tissue fragment was covered with the Tinti–Parma-Benfenati (TPB) graft shifted from the opposite side. Tight wound closure was achieved using a non-absorbable monofilament suture (5-0 Monosof®; Covidien, Dublin, Ireland). The tissue loss resulting from the displaced TPB flap was supplied tightly with non-absorbable monofilament sutures (5-0 Monosof; Covidien). The remaining wound edges were left for open healing by granulation (Fig. 7).

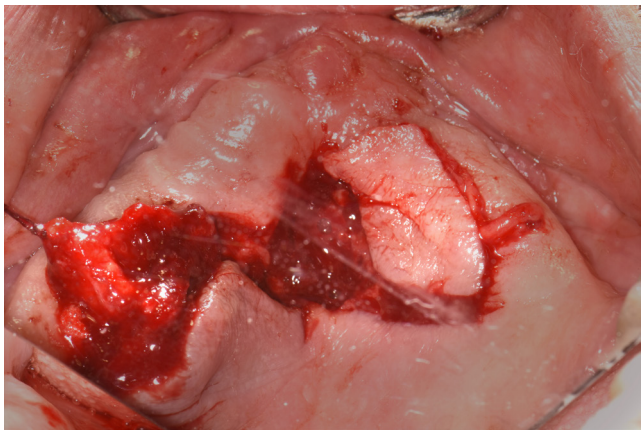

Fig. 6. Preparation of the Tinti–Parma-Benfenati (TPB) flap (right) and the pedicled flap (left)

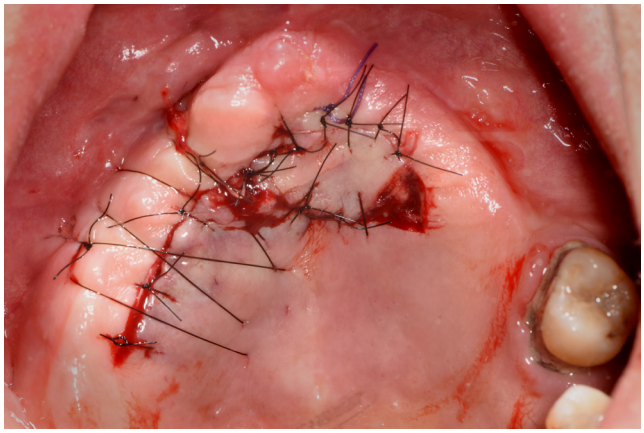

Fig. 7. Wound closure

Healing process

Complaints of pain subsided 3 days after surgery. The sutures were removed after 2 weeks (Fig. 8).

The use of mouth rinse and gel with chlorhexidine (0.1% and 0.2%, respectively) are recommended throughout the healing period according to the manufacturer's instructions. With regard to the tissues not involved in the surgical procedure, the venous return did not show differences noticeable during a clinical examination. The patient did not report any leakage from the nasal passages throughout the follow-up period. In the postoperative period, the discontinuation of the use of the denture was recommended until soft tissues heal and the prosthesis is corrected (Fig. 9,10).

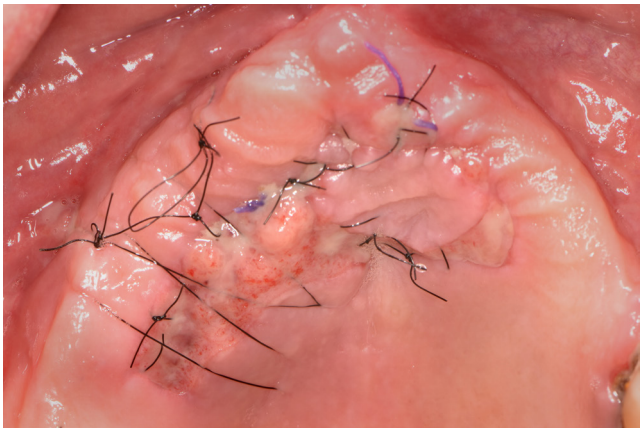

Fig. 8. Status after 2 weeks

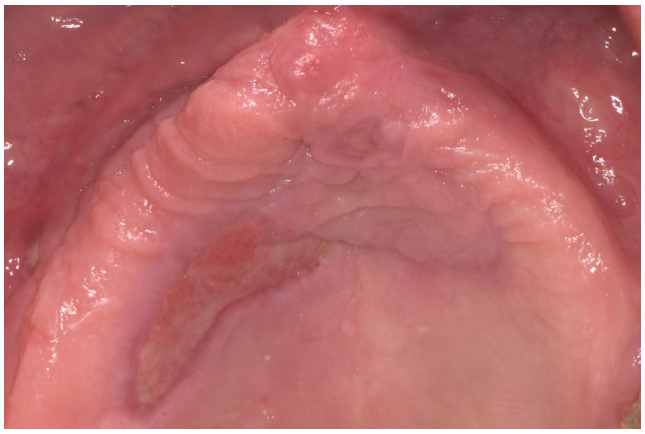

Fig. 9. Status after 3 weeks

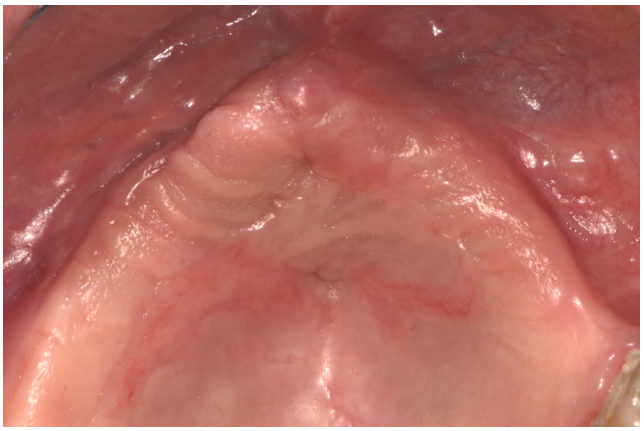

Fig. 10. Wound healing after 6 weeks

The patient was referred to the prosthodontist after surgical treatment. It was necessary to rebase the denture. A follow-up was recommended in June 2019. There were no local complications observed during this period.

The whole procedure is presented in a diagram in Fig. 11. The numbers present the stages of preparation of each layer, respectively. Number 1 presents the 1st preparation of the connective flap, which was moved medially and forward (yellow arrow), whereas number 2 presents the

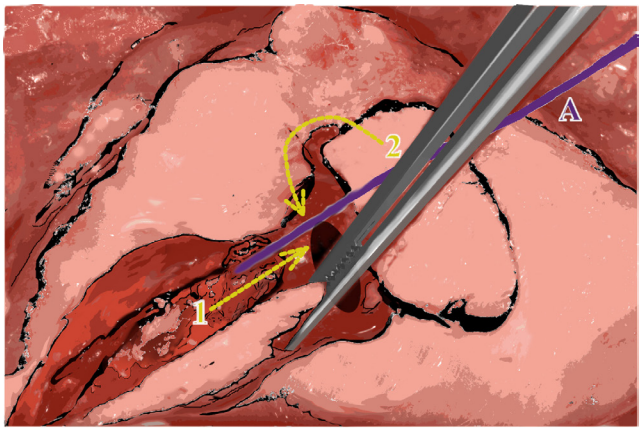

Fig. 11. Scheme of the whole procedure

TPB flap rotated and moved medially, slightly distally (yellow arrow). The violet thread, presented by the letter A, shows the stitch laces used for holding the tissue flap.

Discussion

Treatment in patients with BRONJ is a complex problem and is focused on covering the denuded bone with soft tissue, simultaneously providing good blood supply. The problem emerges in the literature and is still unsolved.[5–8,11] An essential aspect of treatment is the proper care of soft tissues.[5,6,11] Currently, there is a lack of developed methods when it comes to dealing with the complications associated with BP therapy. It has to be emphasized that medication, on the one hand, impairs disease development, but, on the other hand, weakens general health, involving the healing of soft tissues, e.g., the formation of new vessels. A well-known fact is that N-BPs may cause BRONJ development. A good solution seems to be to change the medication and reduce BRONJ expansion. Such treatment is proposed by Oizumi et al., who tried to exchange N-BPs with non-N-BPs.[1] They proposed a simultaneous surgical intervention to diminish infection and promote healing mechanisms.[1] What needs to be mentioned is that this kind of treatment is still perspective and needs further evaluation. Minimally invasive therapy also appears in the literature, especially when reconstructive surgery is taken into consideration. Reconstructive surgery is a medical term used to describe the surgical treatment of patients with sophisticated injuries involving various tissues, such as skin, mucosa, muscle, bone, and nerves. Various types of flaps are mentioned in the literature (nasolabial flap, mylohyoid flap, free tissue transfer). Mücke et al. proposed reconstructive surgery by means of a mylohyoid flap.[16] Despite the fact that this method is rarely used in contemporary reconstructive surgery, it has its advantages, such as the simplicity and predictability of the technique; however, the disadvantages should also be mentioned.

Neurological damage (to the lingual or alveolar nerve), salivary duct damage, and the shallowing or altering of the muscle attachment can lead to severe problems. Usually, patients with BRONJ are edentulous, and thus a lack of good stabilization of a lower denture (observable quite often) and a lack of sensitivity in the lingual nerve seem to be considerable drawbacks. The choice of this kind of treatment should be thoroughly considered.[17]

The authors of this paper focused on the proper and careful preparation of the soft tissues of the hard palate. One of the well-known methods, and the oldest mentioned in the literature, is reconstruction with a nasolabial flap. This kind of reconstruction is very reliable – flaps are thick enough to provide the proper blood supply and the pedicle is longer, which helps to extend the flap. The thickness of the flap is determined by the reconstruction requirements used for covering small oral defects after tumor excision, resulting in good esthetic and functional effects.[13,18] Flaps can be used for the regeneration of the intraoral defects of the hard and soft tissues of the maxilla, palate, gingivae, and buccal mucosa, and for the restoration of different parts of the mouth, such as the anterior mouth floor.[19] A nasolabial flap also has very good blood supply from the adjacent tissues (i.e., dermal and subdermal plexuses as well as facial and transverse facial arteries).[19] Despite indisputable proneness to using this method, it should be approached with caution. It may lead to unwanted inclusion cysts, originating from hair follicles, which may leave unesthetic scars and may expose patients to unnecessary suffering.[13,18–20] Free tissue transfer – a procedure described by Engroff et al. – seems to be a good alternative.[21] It has to be mentioned that harvesting the proper soft tissue for the graft results in more wounds, which may lead to scars, and esthetic or various functional problems.[21] The patient should be carefully examined before any decisions on the specific method are made and the whole case should be thoroughly considered, weighing the pros and cons of each method.

The authors of various publications on the surgical methods of closing the defects with surrounding soft tissue grafts, or with the use of synthetic materials and materials of animal origin are much too critical of exact and tight wound closure.[5,6,11,22] Due to the unpredictability of the vascular blood supply, the proposed methods do not guarantee repeatable satisfactory outcomes. The above procedure is one of the possible methods that can be used while retaining the generally accepted principles of surgical treatment. It should be emphasized that the occurrence of sequestra often eliminates the possibility of using surrounding soft tissue flaps, given the difficulties in covering the developed bone defect.[11]

It is vital that the patient diligently follows the instructions given by the medical personnel, including ceasing BP therapy after consulting the attending physician. In the present study, throughout the entire convalescence period, the patient abided by the recommendations, being aware of his participation in the healing process.

Despite a complication in the initial procedure (i.e., the removal of the sequestrum), the described surgical technique allows the lasting closure of an oronasal fistula and can be recommended in the treatment of similar cases. In the context of blood circulation disorders caused by BP therapy, the maintenance of the vascular pedicle of the graft contributed to proper healing. Thus, the protection of the site against further trauma, the usage of topical chlorhexidine and antibiotic therapy play a crucial supportive role in recovery. Laser therapy and extracorporeal shockwave therapy (ESWT) are mentioned as intensifying healing.[7,17,23] Low-level laser therapy (LLLT) is a widely known method of improving and accelerating the healing of various types of tissues. This method uses low-energy light to increase the production of mitochondrial enzymes, porphyrins, flavins, and cytochromes. Adequately to the region of the body and the type of laser used, the light is absorbed by cells and – by means of a still unclear mechanism – leads to good tissue healing.[17] While LLLT is considered to be non-invasive, ESWT is based on causing damage by means of an externally applied, focused, high-intensity acoustic pulse. A shockwave is an acoustic wave which carries high energy to painful spots and myoskeletal tissues. It is characterized by a jump change in pressure, high amplitude and non-periodicity. This energy is produced by compressed air and is transferred to the transmitter at the end of the applicator, and further into the tissue. Before commencing the procedure, patients are usually sedated or anesthetized in order to help them remain still and to reduce possible discomfort. The exact mechanism of ESWT remains unknown.[23] It has been proven that acoustic waves create micro-ruptures in the bone, which significantly increase the expression of such growth factors as BMPs and vascular endothelial growth factor (VEGF).[24] There is another method successfully used by many doctors to increase the expression of growth factors (e.g., platelet-derived growth factor – PDGF). It uses peripheral blood for preparing a clot (here named platelet-rich fibrin – PRF) by centrifugation; thus, the platelets and their crucial factors are concentrated for healing acceleration. The procedure was invented and introduced by Joseph Choukroun.[acc.24] Such prepared PRF may be used for closing wounds and accelerating healing – a layer is placed directly on the denuded bone, and is then covered by the tissue sutured without pressure.[25] We can find various techniques used for regeneration that utilize PRF. The techniques of surgery, tissue engineering and bone biology are merged to offer promising solutions.[24,26]

All the procedures performed in the present study were carried out in accordance with the ethical standards of the institutional and national research committee, and with the 1964 Declaration of Helsinki and its later amendments, or comparable ethical standards.

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