

# Cumhuriyet Dental Journal



Published By Sivas Cumhuriyet University http://cdj.cumhuriyet.edu.tr

> E-ISSN: 2146-2852 ISSN: 1302-5805

#### **Cumhuriyet Dental Journal**

The Official Journal of the Sivas Cumhuriyet University Faculty of Dentistry. The first issue was published in 1998 and journal's name was changed as Cumhuriyet Dental Journal in 2010. Issues are published quarterly since 2018.

#### Aims and Scope

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CDJ publishes original research papers, reviews, and case reports within clinical dentistry, on all basic science aspects of structure, chemistry, developmental biology, physiology and pathology of relevant tissues, as well as on microbiology, biomaterials and the behavioral sciences as they relate to dentistry.



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## An Evaluation of the Effect on Streptococcus Mutans Adhesion of Surface Roughness in Different Aesthetic Restorative Materials

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Research Article	ABSTRACT
History	<b>Aim:</b> The aim of this study was to evaluate the effect on Streptococcus mutans (S. mutans) adhesion of the surface roughness in different aesthetic restorative materials after the application of different finishing and polishing disc systems to tooth-coloured restorative materials.
Received: 08/04/2024	Material-Method: A total of 126 disc-shaped samples were prepared as 21 for each of the restorative test
Accepted: 09/12/2024	materials used in the study. The samples were separated into 3 groups of 7 for the application of a different finishing and polishing disc set to each group. Pellicle formation on the surface of the materials was obtained by leaving the samples for one hour in artificial saliva containing mucin. The samples were then incubated in solutions containing S. mutans, and after 24 hours, the number of S. mutans showing adhesion were counted. <b>Results:</b> A statistically significant difference was determined between the materials in respect of surface roughness values and bacteria adhesion (p<0.05), and no significant difference was found between the finishing and polishing disc sets (p>0.05). A positive correlation was determined at the rate of 26.2% between the surface roughness of the materials and S. mutans adhesion (p<0.05). <b>Conclusion:</b> Materials show different surface roughness and adhesion values and the chemical content and physical properties of the material have an impact on this. The reasons of bacteria adhesion on material surface is clarified with in-vitro and in-vivo studies.

Keywords: Composite Resin, Glass Ionomer Cement, Streptococcus Mutans.

### Farklı Estetik Restoratif Materyallerde Yüzey Pürüzlülüğünün Streptokok Mutans Adezyonuna Etkisinin Değerlendirilmesi

Research Article	ÖZ
Süreç	Amaç: Bu çalışmanın amacı, diş rengindeki restoratif materyallere farklı bitirme ve polisaj disk sistemlerinin uygulanmasından sonra, farklı estetik restoratif materyallerdeki yüzey pürüzlülüğünün Streptokok mutans (S. mutans) adezyonuna etkisini değerlendirmektir.
Geliş: 08/04/2024 Kabul: 09/12/2024	Gereç-Yöntem: Çalışmada kullanılan restoratif test materyallerinin her biri için 21 adet olmak üzere toplam 126 adet disk şeklinde örnek hazırlandı. Her gruba farklı bitirme ve polisaj diski seti uygulanması için numuneler 7'şerli 3 gruba ayrıldı. Örnekler müsin içeren yapay tükürük içerisinde bir saat bekletilerek malzemelerin yüzeyinde pelikıl oluşumu sağlandı. Örnekler solüsyonlarda inkübe edildi ve 24 saat sonunda adezyon gösteren S. mutans sayısı belirlendi. Bulgular: Malzemeler arasında yüzey pürüzlülük değerleri ve bakteri adezyonu açısından istatistiksel olarak
License E S This work is licensed under Creative Commons Attribution 4.0 International License	anlamlı farklılık tespit edilirken (p<0,05), bitirme ve cilalama disk setleri arasında ise anlamlı bir fark bulunamadı (p>0,05). Malzemelerin yüzey pürüzlülüğü ile S. mutans adezyonu arasında %26,2 oranında pozitif korelasyon belirlendi (p<0,05). <b>Sonuç:</b> Materyallerin farklı pürüzlülük ve adezyon değerleri göstermesinde; materyallerin kimyasal içerik ve fiziksel özelliklerinin etkili olduğu; materyal yüzeyine gerçekleşen bakteri tutulumunun nedenlerinin in-vitro ve in-vivo araştırmalarla aydınlatılabileceği düşünülmektedir. <b>Anahtar Kelimeler:</b> Kompozit rezin, cam iyonomer siman, streptokok mutans
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	5, Bakir EP, Ozcan N. (2024) An Evaluation of the Effect on Streptococcus Mutans Adhesion of Surface Roughness in torative Materials, Cumhuriyet Dental Journal, 27(4):214-221.

#### Introduction

Composite restorative materials were developed both mechanically and physically until today. Also these materials are currently the most preferred aesthetic filling material with its wide are of use. However, it has been shown that microleakage, which develops due to polymerization shrinkage of composite resins, causes secondary decay under the material. Bonding problems of composite restorations are also encountered when they do not be sufficiently isolated from saliva.<sup>1-4</sup> All these reasons led to a preference for glass ionomer cements (GIC), which have the ability to express fluoride, as a restoration material in patients with poor oral hygiene. As GIC achieve chemical bonding through chelation with calcium of the hard dental tissues, express fluoride, and have a low pH, they have an important place in dentistry with these antimicrobial properties.<sup>5</sup>

Even after brushing the teeth, saliva proteins and macromolecules settle on the dental surface, which is covered with a non-cellular, clear film layer, known as the pellicle.<sup>6</sup>Together with the settling of bacteria on the pellicle, the accumulation of food remnants, and saliva glycoproteins, leukocytes, macrophages, and epithelial cells on the pellicle form a soft, semi-transparent, adhesive, microbio dental plaque.<sup>7</sup> Conducted researches show that surface roughness plays a role directly in generation of microbial dental plaque and critical surface roughness value is 0,2  $\mu$ m. According to research reports if this value is exceeded, microbial dental plaque generation also will be rised.<sup>8</sup>

Bacteria where are located in oral cavity, necessitate to adhere to surface and have the ability to reproduce for maintaining their presence. At the forefront of microorganisms with this capability is Streptococcus mutans (S. mutans), which is accepted as the most cariogenic and is predominant in enamel decay, and Lactobacilli, predominant in dentin decay, and Actinomyces in root decay. However, these bacteria need to be able to easily adhere to the plaque structure on hard surfaces such as teeth and restorations and to be able to overcome colonization resistance.<sup>6.7</sup> Conducted studies show that bacteria adhesion is directly correlated with materials' surface characteristics and chemical ingredients.  $^{\rm 3}$ 

The surface properties of dental restorations are extremely important in respect of both oral health and restoration life, and aesthetic properties. Finishing and polishing procedures are beneficial in the shaping of the restoration surface and making it as smooth as possible. In this way, plaque retention and discoloration in restorations, and the associated patient complaints overcome, and the formation of gingival irritation and secondary decay are prevented.<sup>9</sup> The materials produced for this purpose are primarily diamond and carbide finishing mills, finishing and polishing discs of different abrasive sizes, polishing pastes, and metal-plastic bands used for the interfaces.<sup>10</sup> Previous studies were shown that finishing and polishing disc sets provide the best results in anterior region restorations, and the form of these discs is more appropriate for use on straight or outward curved surfaces in particular.<sup>9-11</sup>

The main goal of this research was to discuss the effect on S. mutans adhesion of the surface roughness in different aesthetic restorative materials after the application of different finishing and polishing disc systems to toothcoloured restorative materials. The null hypothesis of the study is built that the surface roughness of different aesthetic restorative materials have effect on the adhesion of S. mutans. According to study findings null hypothesis was approved and crucial difference was notice between the materials with respect to bacterial adhesion.

#### Methods

The kind of restorative materials used in this research, the manufacturer's information, the content and LOT numbers are remarked in Table 1.

The finishing and polishing disc materials used in the study, the manufacturer's information, the content, LOT numbers, and application stages are shown in Table 2.

Materials	Туре	Lot No	Content
Filtek <sup>™</sup> ultimate universal restorative (3m espe, St. Pau, MN, USA)	composite (nanofil)	NA16279	Bis-GMA, UDMA, TEGDMA, PEGDMA, Bis-EMA, silica and zirconia. Filler weighted 72.5%, volume weighted 63.3%.
Filtek™ z250 (3m espe, St. Pau, MN, USA)	composite (microhybrid)	NA53191	Bis-GMA, UDMA, Bis-EMA, Zirconia and silica. Filler weighted 82%, volume weighted 60%.
Estelite asteria (Tokuyama, Tokyo, Japan)	composite (nanohybrid)	W128	Bis-GMA, UDMA, Bis-MPEPP, TEGDMA, mequinol, dibutyl hidroxy toluene, UV absorber, glass particles, silicone dioxide, zirconium dioxide. Filler weighted 82%, volume weighted 71%.
Ceram. x duo (Dentsply, Konstanz, Germany)	composite (nanoceramic)	1905000953	Bis-GMA, UDMA, TEGDMA, modified methacrylate polysiloxane, glass particles, iron oxide, titanium oxide. Filler weighted 76%
Photac fil quick aplicap (3m espe, St. Pau, MN, USA)	resin-modified glass ionomer	5725992	Na-Ca-Al fluorosilicate glass, polyacrylic acid, maleic acid, HEMA.

Fuji 2 lc capsule (Gc, Tokyo, Japan)	resin-modified glass ionomer	1910041	Fluoro distilled water, urethane dimet	aluminium polyacrylic acid, nacrylate.	silicate 2-hydroxyethyl	glass, methacrylate,
Polystyrene (SEPAR plastic)	plastic (control group)	-	Styrene monom	er.		

Materials	Content	Lot no	Application stages
Sof-lex (3m espe, St. Pau, USA)	Finishing and polishing discs coated with aluminum oxide urethane	NA54445	Disc set in 4 stages; decreasing height and contouring discs used at 10,000 rpm. Finishing and polishing discs usedat 30,000 rpm.
Optidisc (Kerr, orange, CA, USA)	Contains aluminum oxide, synthetic polymer, polyethylene, epoxy resin adhesive	7053476	A 4-stage application sequence of extra thick (decreasing height and contouring), thick (finishing), fine (polishing) and extra fine (high polishing)
Super-snap (Shofu, Inc, Kyoto, Japan)	Finishing and polishing discs containing aluminum oxide particles	0719020	A 4-stage finishing and polishing system containing no metal parts: leveling, finishing, polishing, and super polishing stages.

#### Preparation of the Restorative Samples

A total of 126 disc-shaped samples were prepared as 21 for each of the restorative test examples used in the research. The 7 samples of polystyrene material used as the control group were purchased from SEPAR Plastik. Using an mouth spatula, all the materials were placed in plastic molds, 8 mm in diameter and 2 mm in depth, and were then compressed between a clear band and two glass plates. After removing the glass plates, polymerization was applied with an LED light device (Woodpecker LED-G, China) in related with the manufacturer's instructions.

## Finishing and Polishing procedures applied to the Prepared Samples

The 21 prepared samples for each material were divided into 3 groups of 7 for the application of the finishing and polishing plans according to the producer's instructions, implementing a different polishing disc set for each group. After completion of the finishing and polishing procedures, the samples were cleaned under running tap water for 20 seconds, and were after rested in distilled water until the bacterial adhesion stage. The polystyrene material used as the control group was not included in these procedures.

#### Dimensions of the Surface Roughness of the Samples

Surface roughness dimensions of the edited samples were made using a profilometer (Veeco Dektak 6M, NY, USA). The measurement was performed by applying 5mg force with the recording end of the profilometer deviceto record a distance of 2 mm at a fixed speed of 10 seconds. The measurements were taken from 3 different regions of each sample and the average of these was taken for the computing of the surface roughness (Ra) values.

After completion of the surface roughness values, the samples were sterilized by washing and then left for 15 minutes in an autoclave at 121°C and 1 atmosphere pressure (Newmed Kronos B, İzmir, Turkiye) before the bacteria adhesion stage.

#### Preparation of the Synthetic Saliva

At the stage of preparation of the synthetic saliva in which the samples were to be placed, a solution was prepared of 2 lt of distilled water with the addition of 2560 mg sodium chloride (NaCl), 332.97 mg calcium chloride (CaCl<sub>2</sub>), 250 mg magnesium chloride hexahidrate (MgCl<sub>2</sub>(6H<sub>2</sub>O)), 189.48 mg potassium chloride (KCl), 3015 mg potassium acetate (CH<sub>3</sub>COOK), 772 mg tripotassium phosphate trihydrate (K<sub>3</sub>PO<sub>4</sub>(3H<sub>2</sub>O)) and 0.1 ml 85% phosphoric acid (H<sub>3</sub>PO<sub>4</sub>). The solution was mixed until it became clear.<sup>10</sup> The synthetic saliva was prepared to have pH of 6.5-7, with the addition of 140 mg Type II mucin (Sigma-Aldrich Chemie GmbH, Deisenhofen, Germany) to every 100 ml of synthetic saliva, sterilized in the autoclave.

## S. mutans Adhesion to the Restorative Material Samples

For the adhesion test in this study, S. mutans ATCC® 25175 (RSHM NO:7038) isolate was used, purchased from the Turkish Public Health Institute culture collection. The isolate was revitalized by adding Tryptic Soy Broth (TSB) to the lyophilised isolate. A 5% sheep blood agar (SBA) subculture was made to a solid medium from the suspension of the isolate in TSB and left for 24 hours incubation at 35-37°C in jars containing 5-10% CO<sub>2</sub>. The bacteria produced from the colonies were activated by again making a subculture of the SBA medium. After sterilization in the autoclave, the restorative material samples were placed in sterile petri dishes 90 mm in diameter. Synthetic saliva containing 20 ml mucin was added to the petri dishes to completely cover each sample, and then after waiting 1 hour for the pellicle formation, the synthetic saliva was removed from the petri dish.

From the activated S. mutans bacteria, a bacteria suspension at 0.5 McFarland turbidity (1 x  $10^8$  CFU/ml) was obtained using a Nephelometer and Vortex device within the TSB, and was added to the petri dishes to cover the samples.

The petri dishes containing the samples and bacteria suspension in jars containing 5-10% CO<sub>2</sub> were incubated for 24 hours at  $35-37^{\circ}$ C. Following the incubation, the

samples were removed from the petri dishes and washed 3 times with sterile phosphate buffer solution (PBS) to remove the bacteria not showing full adhesion to the surface. Each washed sample was placed in a tube containing 1 ml sterile PBS, and these were left for 5 mins in an ultrasonic bath operating at 285 W and 50/60 KHz (Gen-Probe, San Diego, CA, USA). Thus the bacteria showing adhesion were transferred from the sample surface to the PBS.

To not cause a change in the number of bacteria, a sample was taken with 0.01 ml extract without being removed from the restorative material within the PBS and was seeded in 5% SBA and left for incubation for 24 hours. The colonies formed in the medium after 24 hours of incubation were determined with the bacteria count method and recorded as Colony-Forming Units (CFU/ml).

#### **Statistical Analysis**

The data get from this study were analyzed statistically using SPSS 25 software (Statistical Package for the Social Sciences).Identified statistics were indicated as mean  $\pm$ standard deviation values. To investigate the effect on a dependent variable of two different factors formed from multiple groups, Two-Way Variance Analysis was applied, and to decide from which group the discrepancy originated, the Tukey Multiple Comparison test was used. Outcomes were indicated in a 95% confidence interval. When interpreting the results, the level of statistical significance was accepted as 0.05.

#### Results

## Examination of the Surface Roughness Values of the Materials

The mean surface roughness values obtained using 6 different aesthetic filling materials and 3 different finishing and polishing disc sets are shown in Table 3.

The materials showing the greatest surface roughness were Photac Fil Quick Aplicap (3m espe, St. Pau, MN, USA), a resin-modified glass ionomer cement (RMGIC), followed by Fuji 2 LC Capsule (Gc, Tokyo, Japan) another RMGIC material. The material with the least surface roughness, excluding polystyrene used as the control group, was the nano-hybrid-based Estelite Asteria (Tokuyama, Tokyo Japan) composite resin material.

Two-way variance analysis is used for compare the effects of the polishing and finishing disc sets on the surface roughness values. In terms of surface roughness there is a significant difference between materials (p<0.05) However there is no meaningful difference exist between the polishing and finishing materials (p>0.05)

According to the Tukey Multiple Comparison test to determine from which material the difference originated, the nanofil-based Filtek<sup>™</sup> Ultimate Universal Restorative (3m espe, St. Pau, MN, USA) material showed significantly lower roughness compared to only the two RMGIC materials (p<0.05) and the nano-hybrid-based Estelite Asteria (Tokuyama, Tokyo Japan) showed significantly lower roughness compared to the two RMGIC materials and the nano-ceramic-based Ceram.x Duo (Dentsply, Konstanz, Germany) from the composite resins (p<0.05). No statistically curicial difference was observed between the other materials in respect of surface roughness (p>0.05). Tukey Multiple Comparision test results are shown in Table 4.

Materials	Finishing and polishing disc sets	Mean value	Standard deviation
Polistren (control group)	-	0.23739	0.120210
3m filtek™ ultimate universal	3m sof-lex	2.07388	1.113412
restorative	Kerr optidisc	2.44061	0.913932
restorative	Shofu super-snap	2.19690	1.190862
	3m sof-lex	2.34977	0.663118
3m filtek™ ultimate z250	Kerr optidisc	2.77105	0.943048
Sin nicek ultimate 2250	Shofu super-snap	2.68842	1.282877
	3m sof-lex	1.93706	0.924018
Tokuyama estelite asteria	Kerr optidisc	1.82675	0.861827
	Shofu super-snap	1.56323	0.494112
	3m sof-lex	2.86446	0.722355
Dentsply ceram.x duo	Kerr optidisc	2.93628	1.214530
	Shofu super-snap	2.61183	0.756362
	3m sof-lex	3.55144	1.134469
3m photac fil quick aplicap	Kerr optidisc	3.39085	1.587742
	Shofu Super-Snap	3.15430	0.755827
	3m sof-lex	3.21044	0.529651
Gc fuji 2 lc capsule	Kerr optidisc	3.57197	0.838714
	Shofu super-snap	3.24643	0.783736

#### Table 3. Surface roughness values (Ra)

est materials		р
Polistren (control group)	3m filtek™ ultimate universal	0.0*
	3m filtek™ ultimate z250	0.0*
	Tokuyama estelite asteria	0.003*
	Dentsply ceram.x duo	0.0*
	3m photac fil quick aplicap	0.0*
	Gc fuji 2 lc capsule	0.0*
8m filtek™ ultimate universal	3m filtek™ ultimate z250	1.0
	Tokuyama estelite asteria	1.0
	Dentsply ceram.x duo	0.774165
	3m photac fil quick aplicap	0.002295*
	Gc fuji 2 lc capsule	0.003*
8m filtek™ ultimate z250	Tokuyama estelite asteria	0.073197
	Dentsply ceram.x duo	1.0
	3M Photac fil quick aplicap	0.139681
	Gc fuji 2 lc capsule	0.173326
okuyama estelite asteria	Dentsply ceram.x duo	0.007859*
	3m photac fil quick aplicap	0.0*
	Gc fuji 2 lc capsule	0.0*
Dentsply ceram.x duo	3m photac fil quick aplicap	0.809669
	Gc fuji 2 lc capsule	0.963323
8m photac fil quick aplicap	Gc fuji 2 lc capsule	1.0

### \* p<0.05

## Examination of the S. mutans Adhesion Values of the Materials

The mean S. mutans adhesion values determined on the surface of the restorative materials are shown in Table 5.

The material with the greatest S. mutans adhesion was the nano-ceramic-based Ceram.x Duo (Dentsply, Konstanz, Germany), followed by the RMGIC (Fuji 2 LC Capsule, Photac Fil Quick Aplicap), and the nanofil-based Filtek<sup>™</sup> Ultimate Universal Restorative (3m espe, St. Pau, MN, USA) material. The restorative material showing the least bacteria involvement, excluding polystyrene used as the control group, was the nano-hybrid-based Estelite Asteria (Tokuyama, Tokyo Japan) composite resin material.

As a result of the Two-Way Variance Analysis used to compare the effects of the finishing and polishing disc sets on the S. mutans adhesion values, a statistically crucial difference was observed between the materials (p<0.05) and no significant difference was get between the finishing and polishing disc sets (p>0.05).

According to the Tukey Multiple Comparison test to determine from which material the difference originated, there was determined to be no statistically crucial difference between the nano-hybrid-based Estelite Asteria (Tokuyama, Tokyo, Japan) and the micro-hybrid-based Filtek<sup>TM</sup> Ultimate Z250 (3m espe, St. Pau, MN, USA), which showed the lowest S. mutans adhesion values (p>0.05) and the difference between these two composite resins and all the other materials was found to be statistically significant (p<0.05). No statistically crucial difference was determined between the other materials in respect of bacteria adhesion (p>0.05). The values of bacteria adhesion's Tukey Multiple Comparision test results are shown in Table 6.

When the relationship between surface roughness and bacteria adhesion was evaluated, a positive linear correlation was determined at the rate of 26.2% between the surface roughness of the materials and the amount of S. mutans adhesion to the restoration surface (p<0.05). As the surface roughness increased, so there was an increase in bacteria involvement.

Materials	Finishing and polishing disc sets	Average value	Standard deviation
Polistren (control group)	-	5971.43	2114.01
3m filtek™ ultimate universal	3m sof-lex	54714.28	30210.78
	Kerr optidisc	68042.86	24421.15
restorative	Shofu super-snap	67428.57	14125.23
	3m sof-lex	39571.43	9182.72
3m filtek™ ultimate z250	Kerr optidisc	50814.29	11211.07
	Shofu super-snap	46714.28	18693.53
	3m sof-lex	35685.71	11809.66
Tokuyama estelite asteria	Kerr optidisc	29257.14	13218.15
	Shofu super-snap	32142.86	5365.27
	3m sof-lex	75742.86	14810.79
Dentsply ceram.x duo	Kerr optidisc	79428.57	13813.60
	Shofu super-snap	78600.00	15275.68

Table 5. The mean S. mutans adhesion values determined on the surface of the restorative materials (cfu/ml)

	3m sof-lex	66385.71	17157.35
3m photac fil quick aplicap	Kerr optidisc	67985.71	16977.47
	Shofu super-snap	69414.29	16926.35
	3m sof-lex	71828.57	16911.01
Gc fuji 2 lc capsule	Kerr optidisc	70371.43	17975.79
	Shofu super-snap	68957.13	10489.97

*Table 6.* The values of bacteria adhesion's tukey multiple comparision test results

Test materials		р	
Polistren (control group)	3m filtek™ ultimate universal	0.0*	
	3m filtek™ ultimate z250	0.0*	
	Tokuyama estelite asteria	0.0*	
	Dentsply ceram.x duo	0.0*	
	3m photac fil quick aplicap	0.0*	
	Gc fuji 2 lc capsule	0.0*	
3m filtek™ ultimate universal	3m filtek™ ultimate z250	0.007*	
	Tokuyama estelite asteria	0.0*	
	Dentsply ceram.x duo	0.059	
	3m photac fil quick aplicap	1.0	
	Gc fuji 2 lc capsule	1.0	
3m filtek™ ultimate z250	Tokuyama estelite asteria	0.119	
	Dentsply ceram.x duo	0.0*	
	3m photac fil quick aplicap	0.0*	
	Gc fuji 2 lc capsule	0.0*	
Tokuyama estelite asteria	Dentsply ceram.x duo	0.0*	
	3m photac fil quick aplicap	0.0*	
	Gc fuji 2 lc capsule	0.0*	
Dentsply ceram.x duo	3m photac fil quick aplicap	0.679	
	Gc fuji 2 lc capsule	1.0	
3m photac fil quick aplicap	Gc fuji 2 lc capsule	1.0	
* n=0.0E			

\* p<0.05

#### Discussion

New materials are continuously being introduced in restorative dentistry and studies are ongoing for the development of the ideal material. Of these materials, composite resins take first place.<sup>12</sup> Although there is currently great use of composite resins, because of negative properties such as the fact that they are not be used in cavities that are not be isolated, microleakage seen as a result of polymerization shrinkage and associated secondary decay, the use of GIC, which express fluoride, have antibacterial properties and chemically bonds to dental hard tissues, has come to the fore especially for patients with poor oral hygiene.<sup>3,4</sup>

To prolong the clinical life of restorations and to be able to obtain a more aesthetic appearance, finishing and polishing procedures are required. In restorations where the finishing and polishing procedures are unapplied correctly, surface discolouration associated with plaque accumulation and gingival irritation is occur.<sup>13,14</sup>

Although an anatomic form of the restoration is obtained with finishing procedures, a scratched and rough area is formed on the surface of the material, sopolishing of the restorative material surface is recommended to prevent this.<sup>14</sup> Well-applied finishing and polishing procedures increase the surface hardness of the restoration, increase colour stabilization, and prolong clinical life.<sup>15</sup>

Gauthier *et al.* (2005) examined the surface properties of composite resins and reported that the oxygen

inhibition layer in the outermost layer formed during polymerization was very important for the surface property of the material. Incomplete polymerization in this layer causes a decrease in the restoration surface hardness. The idea has gained weight that because of the finishing and polishing processes the oxygen inhibition layer are removed, thereby obtaining a smoother surface that prevents bacterial adhesion to the material surface.<sup>16</sup>

Different filler particle dimensions and organic matrix hardness of the material shows an significant role in the degree of the effect of the finishing and polishing procedures on the material surface. The harder ones are preferred in abrasives used for the finishing and polishing systems with respect to other filling particles. When this is not the case, there is separation of the organic matrix, filler particles not abraded remain above, and thus there is the possibility of a surface of increased roughness. Moreover, the type, shape, size, amount, and distribution of the filler particles contained in the material are wise to have an effect on surface roughness. Following finishing and polishing procedures, greater roughnessis encountered on the surfaces of resin materials with large particles.17,18

In a study by Koh *et al.* (2008), comparisons were made of the effect of single-stage and multi-stage polishing systems on the surface roughness of microhybrid and nanofil composites. The results showed that the nano filler composites showed less surface roughness than the hybrid composites and the Optidisc and Sof-Lex systems were helpful in obtaining a better surface.<sup>19</sup> The surface roughness values of GIC and composite resins were investigated in several studies, and GIC was reported to show greater surface roughness.<sup>20,21</sup> In a study by Eick *et al.* (2004), the surface roughness of composite, RMGIC, traditional GIC, compomer, ceramic, and amalgam materials was examined, and it was reported that traditional GIC showed the highest roughness value, followed by RMGIC.<sup>22</sup>

In the this study, when compared with polystyrene material used as a positive control group, the surface roughness of the other materials was established to be statistically meaningfully low (p<0.05). The nanohybrid-based Tokuyama Estelite Asteria (Tokuyama, Tokyo, Japan) composite material showed less surface roughness in all the finishing and polishing systems compared with the other restorative materials, and the two RMGIC (Photac Fil Quick Aplicap and Fuji 2 LC Capsule) were the restorative materials showing the worst surface roughness.

Previous studies demonstrated that the material's critical surface rougness value is 0,2  $\mu$ m for bacteria that responsible for decay to shows adhesion. Moreover it is also reported that in the situation of exceeding 0,2  $\mu$ m value, the microbiodental plaque formation was increased.<sup>8</sup> In this study, all the restorative materials used, including the control group, were seen to have surface roughness above the critical surface roughness value.

According to several studies, there is a positive relationship between surface roughness of the material and the number of bacteria showing adhesion to the surface, that plaque formation is increased on rough surfaces, and that bacterial colonization starts from rough areas such as a groove, crack, or wear defect in the restorative material.<sup>23</sup>

Tanner *et al.* (2003) reported that surface roughness affected the adhesion of S. mutans, and rough surfaces formed a retention area for bacterial involvement.<sup>24</sup>

In a study by Brambilla *et al.* (2005) using composite, compomer and GIC, it was concluded that the material with the highest S. mutans adhesion was compomer and the material with the lowest value was GIC.<sup>25</sup>

In different studies using traditional GIC and composite resin, Carlen *et al.* (2001) reported a lower number of S. mutans showing adhesion to the composite resin surface compared to GIC.<sup>26</sup> Eick *et al.* (2004) examined the relationship between bacteria adhesion and the surface roughness of amalgam, composite, compomer, ceramic, traditional GIC and RMGIC materials, and stated that the highest level of bacteria adhesion was to the traditional GIC material which had the highest roughness value.

It has also been claimed that fluoride, which has an antibacterial property, in the content of GICs does not prevent the adhesion of S. mutans to the material surface.<sup>22</sup> Montanaro *et al.* (2004) showed that fluoride inhibited the proliferation and metabolism of bacteria, but remained insufficient in preventing bacteria adhesion.<sup>27</sup> Similar studies confirmed that although fluoride strengthens the enamel surface against external

factors and raises the plaque pH, it does not decrease S. mutans adhesion to the material surface.<sup>28-31</sup>

The results of the this study was confirmed the findings of other studies. When the polystyrene used as the control group was excluded, the restorative material showing the least S. mutans adhesion was determined to be the nanohybrid-based Estelite Asteria (Tokuyama, Tokyo, Japan) composite material, which had the best surface smoothness after polishing. The material with the second least bacteria adhesion was the microhybridbased Filtek<sup>™</sup> Ultimate Z250 (3m espe, St. Pau, MN, USA) composite material, and the difference between the two materials was not statistically significant (p>0.05). However, a statistically significant difference was determined between these materials and the others (p<0.05). The material showing the most S. mutans adhesion after polishing was the nanoceramic-based Ceram.x Duo(Dentsply, Konstanz, Germany), which is also a composite resin. This was followed by the RMGIC, Fuji 2 LC Capsule (Gc, Tokyo, Japan) and Photac Fil Quick Aplicap (3m espe, St. Pau, MN, USA), and then Filtek<sup>™</sup> Ultimate Universal Restorative (3m espe, St. Pau, MN, USA) composite resin. However, no statistically significant difference was seen between these four restorative materials (p>0.05).

The possibility of a clinical follow-up study of patients followed up related to the study hypothesis was severely restricted because of the COVID-19 pandemic. A further limitation was that different bacteria species showing aerobic and anaerobic properties which are seen in plaque were not included in the study due to the need for specific environments and techniques. There is a need for further in-vivo and in-vitro studies on this subject.

#### Conclusions

Within the limitations of this study, it was concluded that there is a positive correlation between surface roughness and bacteria adhesion, as the surface roughness increases, the bacteria adhesion also increases. According to this study's findings  $H_{(0)}$  hypothesis is supported and meaningful differences are observed in materials between in the context of bacterial adhesion.

In showing the different roughness and adhesion values of composite resin materials, the degree of the effect of the chemical content and physical properties of materials and the causes of bacteria adhesion to the material surface is needed to explain with further in-vitro and in-vivo studies. In the light of the findings in this study, bacteria adhesion is considered to increase in direct relation to the increase in surface roughness as a result of fluoride expression in RMGIC.

#### Acknowledgments

This study was designed with reference to the dentistry specialism thesis of Faculty Member Dr Şeyhmus Bakır, entitled, "An evaluation of the effect on Streptococcus Mutans adhesion of the surface roughness of different aesthetic restorative materials". The thesis

study as the source of this article was supported by the Scientific Research Projects Commission Directorate of Dicle University (project number: DİŞ 20.001).

#### **Conflict of Interest**

The authors indicate no financial support or financial conflict of interest. The authors have indicated they have no financial relationships with any company and no external funding.

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### Crista Galli Morphometry and Morphology in Sagittal Skeletal Malocclusions: A Retrospective Cross-Sectional Study

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Research Article	ABSTRACT
	Objective: This study aimed that examine the relationship between crista galli (CG) morphometry and
History	morphology in different types of sagittal skeletal malocclusion.
	Material-Methods: A total of 45 individuals included in the study were examined by dividing them into three
Received: 09/04/2024	subgroups Class I, Class II, and Class III, which included an equal number of samples according to the ANB°. In
Accepted: 16/10/2024	this study, different morphologies (teardrop, tubular, ossified) and morphometric (length, width, height)
	measurements of the CG were analyzed in adult patients with different sagittal malocclusions.
	Results: When linear measurements of the CG were evaluated, no difference was found between skeletal
	malocclusion classes in terms of width and height. Crista galli length is significantly larger in Class I individuals
	(13.13±1.93 mm) than in Class III individuals (11.56±1.26 mm). On the other hand, the CG length of Class I and
	Class II individuals (11.67±2.28 mm) is similar. There is no significant relationship between crista galli
	morphology, Keros classification and CG pneumatization and skeletal malocclusion groups.
	Conclusion: The anteroposterior CG length of skeletal class III individuals is less than that of skeletal class I
	individuals. Crista galli morphology does not differ according to sagittal skeletal malocclusions.

Keywords: Cone-Beam Computed Tomography, Malocclusion, Skull Base.

## Sagittal İskeletsel Maloklüzyonlarda Crista Galli Morfometri ve Morfolojisi: Bir Retrospektif Kesitsel Çalışma

Research Article	
Süreç	Amaç: Bu çalışmada farklı sagittal iskelet maloklüzyonu tiplerinde crista galli (CG) morfometrisi ve morfolojisi arasındaki ilişkinin incelenmesi amaçlandı.
	Gerec-Yöntem: Calışmaya dahil edilen toplam 45 birey, ANB" ye göre eşit sayıda örnek içeren Sınıf I, Sınıf II ve
Geliş: 09/04/2024	Sınıf III olmak üzere üç alt gruba ayrılarak incelendi. Bu calışmada, farklı sagittal iskeletsel maloklüzyona sahip
Kabul: 16/10/2024	erişkin hastalarda crista galli'nin farklı morfolojileri (gözyaşı, tübüler, kemikleşmiş) ve morfometrik (uzunluk,
	genişlik, yükseklik) ölçümleri analiz edildi.
	Bulgular: Crista galli'nin lineer ölçümleri değerlendirildiğinde, iskeletsel maloklüzyon sınıfları arasında genişlik ve
	yükseklik açısından fark bulunamadı. Crista galli uzunluğu Sınıf I bireylerde (13,13±1,93 mm), Sınıf III bireylere
	(11,56±1,26 mm) göre önemli ölçüde daha büyüktür. Buna karşın Sınıf I ile Sınıf II bireylerin (11,67±2,28 mm) CG
	uzunluğu benzer büyüklüktedir. Crista galli morfolojisi, Keros sınıflandırması ve crista galli pnömatizasyonu ile
	iskeletsel maloklüzyon grupları arasında anlamlı bir ilişki bulunmadı.
License	<b>Sonuç:</b> İskeletsel III bireylerin ön-arka crista galli uzunluğu iskeletsel sınıf I bireylere göre daha azdır. Crista galli morfolojisi sagittal iskeletsel maloklüzyona göre farklılık göstermez.
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How to Cite: Ozturk T, Ucaker YE, Cukurluoglu A. (2024) Crista Galli Morphometry and Morphology in Sagittal Skeletal Malocclusions: A Retrospective Cross-Sectional Study, Cumhuriyet Dental Journal, 27(4):222-229.

#### Introduction

The crista galli (CG) is a smooth, thick, pyramidal bone protrusion that extends sagittally in the shape of a rooster's crest in the front and middle of the lamina cribrosa of the ethmoid bone, one of the bones in the neurocranium.<sup>1</sup> The falx cerebri is attached to this anatomical formation, which is an important landmark in the anterior cranial fossa.<sup>2</sup> The location and features of the CG mean it has an important place in anterior skull base (e.g., hypophysis gland) surgical procedures and endoscopic sinus (e.g., ethmoid, frontal sinus) surgery.<sup>3-5</sup> It can also show pneumatization because it is in the pneumatized ethmoid bone structure. It has been examined in studies on sex determination.<sup>6-8</sup> Sagittal skeletal malocclusions have an important place in orthodontic practice. They provide classification by indicating the positions of the maxilla and mandible relative to the skull base and each other.<sup>9</sup> Cranial anatomical structures, which were previously evaluated with two-dimensional images, are now more frequently examined with three-dimensional methods.<sup>2,9-11</sup> In addition, with the development of multidisciplinary relationships, opportunities to examine normal anatomy or problems from a broad perspective rather than examining a small focal point have also improved.<sup>11-13</sup>One of the three-dimensional imaging methods used to examine the anatomy, structure, pathology, and variations of the craniofacial region is cone-beam computed tomography (CBCT).12 It offers the advantages of lower radiation exposure, reduced artifacts, cost-effectiveness, and ease of use compared to computed tomography (CT) imaging, which is known as the gold standard and is more accessible.<sup>14</sup> The relationship between sagittal skeletal anomalies and craniofacial region structures and anomalies has long been studied.<sup>9,12,15</sup> The relationship between skeletal malocclusions and paranasal sinuses has also been examined previously.<sup>12,16</sup> However, the relationship between malocclusions and the CG, a structure very close to the paranasal sinuses, has not yet been examined. Therefore, this study aimed to examine the relationship between CG morphometry and morphology in various types of sagittal skeletal malocclusion by examining CBCT images. The null hypothesis was that CG morphometry and morphology would not differ across different sagittal skeletal malocclusion types.

#### **Materials and Methods**

This was a retrospective cross-sectional study. Before starting the study, permission for the protocol was obtained from the Clinical Research Ethics Committee of Ercives University, dated 10<sup>th</sup> May 2023 with decision number 2023/338. All examinations were performed following the Declaration of Helsinki protocol, and informed consent forms were obtained from all patients before the study. This study examined the morphometry and classified the morphology of the CG in adult patients with various sagittal malocclusions. A power analysis (for 95% power, d = 1.67 and alpha = 0.05 margin of error) performed with G\*Power software (ver. 3.0.10, Franz Faul, Universität Kiel, Germany) determined that 11 patients were required in each group.<sup>7</sup> The inclusion criteria were patients with CG formation on CBCT recordings; patients craniofacial deformity, trauma, without syndrome, orthodontic treatment, or surgical history; high-quality image content; and patients in whom the nasion, maxillary, and mandibular regions were clearly observed to determine the sagittal skeletal malocclusion.

#### Classification of Orthodontic Malocclusion

For individuals with cephalometric radiography, cephalometric evaluation was performed. For individuals without cephalometric radiography, cephalometric evaluation was carried out from the middle point of the sagittal plane sections of images taken with CBCT, which included the mandible (Point B) and maxilla (Point A) and the nasion structure (nasion point). The classification of malocclusion in the sagittal dimension was performed using the ANB°.<sup>17</sup> A total of 45 individuals were divided into three subgroups: Class I (mean ANB:  $1.83\pm1.29^\circ$ ; 7 males, 8 females), Class II (mean ANB:  $7.35\pm2.07^\circ$ ; 5 males, 10 females), and Class III (mean ANB:  $-4.13\pm2.03^\circ$ ; 5 males, 10 females), which included equal numbers of samples according to the ANB° (Table 1).

Table 1. Comparisons of crista galli morphometry and morphology of different skeletal malocclusion classes.

		Skeletal Class I (n=15)	Skeletal Class II (n=15)	Skeletal Class III (n=15)	P values
Ag	e	18.7±8.6 ª	24.7±8.0 <sup>b</sup>	22.7±7.6 <sup>a,b</sup>	0.020 * <sup>KW</sup>
AN	В	1.83±1.29 ª	7.35±2.07 <sup>b</sup>	-4.13±2.03 °	<0.001 *** OWA
SN/Go	oGn	33.89±5.35 ª	44.49±9.18 <sup>b</sup>	35.24±9.64 <sup>a</sup>	0.003 ** <sup>OWA</sup>
Gender	Male	7 (46.7)	5 (33.3)	3 (20.0)	0.031 * <sup>p</sup>
Genuer	Female	8 (53.3)	10 (66.7)	12 (80.0)	
CC Linear	Length	13.13±1.93ª	11.67±2.28 <sup>a,b</sup>	11.56±1.26 <sup>b</sup>	0.048 * <sup>KW</sup>
CG Linear	Width	4.68±1.52	4.71±1.45	5.05±1.65	0.782 <sup>OWA</sup>
Measurements	Height	14.25±2.21	13.53±2.68	13.39±2.86	0.595 OWA
	Tear Drop	5 (33.3)	10 (66.7)	10 (66.7)	
CG Morphology	Tubular	8 (53.3)	4 (26.7)	4 (26.7)	0.343 <sup>p</sup>
	Ossified	2 (13.3)	1 (6.7)	1 (6.7)	
	Туре І	3 (20.0)	2 (13.3)	3 (20.0)	
Keros Classification	Type II	10 (66.7)	12 (80.0)	10 (66.7)	0.925 <sup>p</sup>
	Type III	2 (13.3)	1 (6.7)	2 (13.3)	
Pneumatization	Present	0 (0.0)	1 (6.7)	1 (6.7)	0.594 <sup>p</sup>
Pheumatization	Absent	15 (100.0)	14 (93.3)	14 (93.3)	0.334

Numeric data was given Mean ± Standard Deviation. Categorical data was given Count (Percentage). <sup>P</sup>: Pearson Chi-Square test. <sup>KW</sup>: Kruskal-Wallis H test. <sup>OWA</sup>: One-Way ANOVA test. Statistically significant degree was given p<0.05. \*P < 0.05; \*\*P < 0.01; \*\*\*P < 0.001.

#### **Cephalometric Analysis Parameters**

In performing cephalometric analyses, hard tissue points nasion (N), anterior nasal spine (ANS), sella (S), gonion (G), condylion (Co), gnathion (Gn), A point (A) and B point (B) were used (Figure 1A). Maxillomandibular

relationship (ANB), Upper Face Height (N-ANS), Posterior Face Height (S-Go), SN/GoGn and Mandibular Length (Co-Gn) measurements were made using these points (Figure 1B).



Figure 1: A) Cephalometric points used for analyses. B) Angular and linear measurements used in analysis.

#### **Cone Beam Computed Tomography Parameters**

All CBCT images to be scanned were obtained with the New-Tom 5G device (QR, Verona, Italy), at 110 kV and 3–5 mA, with a voxel size of 0.16 mm, a size of  $18 \times 16$  cm, and a typical time of 5.4 s. Examinations and measurements of axial and coronal sections were made with the built-in NNT (New-Tom Image Viewer, QR, Verona, Italy) software.

#### Crista Galli Measurements

*Linear Measurements:* By examining the coronal and axial sections of the CBCT images, the height of the CG (linear distance between the cribriform plate in the supero-inferior direction and the top point of the CG) was measured on the coronal sections (Figure 2a). The width in the mediolateral direction (linear distance between the CG cortical outer borders) was measured (Figure 2b). The length (between the frontal bone inner cortex tip and the most posterior point of the CG) was measured by examining the axial section measurements (Figure 2c).<sup>8</sup>



*Figure 2:* CG measurements. a) Measurement of the height of the CG on coronal CBCT slice. b) Measurement of the width of the CG on axial CBCT slice. c) Measurement of the length of the CG on axial CBCT slice.

*Morphology of the Crista Galli:* This assessment utilized the Crista Galli (CG) measurements outlined by Komut et al. in 2022, alongside the classification system based on the presence of the cavitary component (see Figure 3).<sup>7</sup> This analysis was conducted utilizing axial and coronal

sections from Cone Beam Computed Tomography (CBCT) scans. According to this classification system: Type 1 (teardrop type) denotes a CG width exceeding one-third of its height, characterized by a prominent cavitary component (refer to Figure 3a). Type 2 (tubular type)

indicates a width less than one-third of the height, with a cavitary component extending from the base to the top (see Figure 2b). Type 3 (ossified type) features a width less than one-third of its height, lacking a discernible cavitary component (depicted in Figure 3c).<sup>7,8</sup> Additionally, the Keros classification, which assesses the depth of the olfactory fossa relative to the height of the lateral lamella

of the cribriform plate (CP), delineates: 1-3 mm for type 1 (where CP and ethmoid roof (ER) are aligned at the same level, as illustrated in Figure 4a), 4-7 mm for type 2 (as depicted in Figure 4b), and 8-12 mm for type 3 (with CP notably situated downstream of ER, as shown in Figure 4c). Furthermore, the presence of pneumatization within the CG was also scrutinized (refer to Figure 5).<sup>7</sup>



Figure 3: Morphological classification of CG on coronal CBCT slices. a) Teardrop type. b) Tubular type. c) Ossified type.



Figure 4: Keros classification types of the CG on coronal CBCT slices. a) Keros type 1. b) Keros type 2. c) Keros type 3.



Figure 5: Pneumatized crista galli on coronal CBCT slice.

#### **Statistical Analysis**

The Jamovi (version 2.4.8.0, Sydney, Australia) statistical analysis software was used for statistical evaluation of the data.<sup>18</sup> The Pearson chi-square was used

to evaluate categorical data. The Shapiro–Wilk test was used in the normality analysis of the numerical data. The Kruskal–Wallis test (Mann–Whitney U test for post hoc pairwise comparisons) was used in the comparison between groups of non-parametric variables. One-way ANOVA was used for parametric variables. Spearman correlation coefficients were used to examine the correlations between variables. Linear regression analysis was used to determine the factors affecting CG morphometry. Ordinal logistic regression analysis was used to describe the relationships between skeletal malocclusion classes and CG morphometry and morphology. The statistical significance value was taken as p<0.05.

#### Results

The comparisons of CG morphometry and morphology of the skeletal malocclusion classes are given in Table 1. When the linear measurements of the CG were evaluated, no difference was found between skeletal malocclusion classes in terms of width (p=0.782) or height (p=0.595).

In contrast, the CG length was significantly (p=0.048) greater in Class I individuals. On the other hand, the CG length of Class I and Class II individuals ( $11.67\pm2.28$  mm) is similar. No significant relationships existed of CG morphology (p= 0.160), Keros classification (p=0.730), and CG pneumatization (p=0.594) with skeletal malocclusion groups.

The significant relationships between CG linear measurements and cephalometric parameters, controlling for skeletal class, are given in Table 2. Weakly positive significant correlations existed between CG length and Co-Gn (Rho=0.475, p<0.01) and S-Go (Rho=

0.233, p<0.05) lengths. Weakly positive significant correlations were found between CG width and S-Go length (Rho=0.301, p<0.05). A weakly positive significant correlation existed between CG height and N-ANS (Rho=0.314, p<0.05). Although significant positive correlations were found between CG width and height (Rho=0.359, p<0.05), no significant correlation existed between these measurements and CG length.

The linear regression analysis results of the CG linear measurements are given in Table 3. When the CG morphology type was ossified, the CG length decreased by 3.32 times (p=0.016). When the CG length increased by 1 unit, the SN/GoGn angle decreased by 0.20 times (p=0.046), and the CG height increased by 0.37 times (p=0.015). When the CG width was examined according to CG morphology, it decreased 1.82 times (p=0.003) when it was tubular type and 3.27 times (p=0.006). When the CG width increased by 1 unit, the CG height increased by 0.32 units (p=0.002) when it was ossified type. When the CG width increased by 1 unit, the CG height increased by 0.32 units (p=0.006). When the CG height is examined according to CG morphology, it increased by 2.95 times (p=0.009) when it was tubular type and 4.86 times (p=0.013) when it was ossified type.

The results of the ordinal logistic regression analysis examining the relationships between skeletal malocclusion category and CG morphometry and morphology measurements are presented in Table 4 (overall model test, p=0.023). When examined by skeletal class, CG length (p=0.008) and gender (p=0.009) were important risk factors for malocclusion.

#### Table 2. Correlation matrix for crista galli linear measurements.

		CG Length	CG Width	CG Height
CG Width	Pearson's r	0.201	-	
	Spearman's rho	0.252	-	
CC Hoight	Pearson's r	0.272	0.332*	-
CG Height	Spearman's rho	0.171	0.359*	_
6. 6.	Pearson's r	0.403**	0.187	-0.01
Co-Gn	Spearman's rho	0.475**	0.18	-0.053
S-Go	Pearson's r	0.310*	0.251	-0.007
5-60	Spearman's rho	0.233*	0.301*	-0.051
N-ANS	Pearson's r	0.042	0.175	0.293
	Spearman's rho	0.107	0.175	0.314*

Note: Checked for skeletal class. \* p<0.05. \*\* p<0.01.

Table 3. Linear regression model results for crista galli linear measurements.

	Variables		Estimate	%95 Cl Lower	%95 Cl Upper	P values	R <sup>2</sup>	Adjusted R <sup>2</sup>	Overall Model Test	
				LOWCI	opper	values		IX III	F	р
	CG	2-1	-0.67	-2.43	1.08	0.432				
CC Longth	Morphology	3-1	-3.32	-5.95	-0.70	0.016	0.792	0.518	2.89	0.010*
CG Length	SN/GoGn		-0.20	-0.39	-0.01	0.046	0.792	0.518	2.89	0.010
	CG Height		0.37	0.08	0.65	0.015				
	CG	2-1	-1.82	-2.95	-0.70	0.003				
CG Width	Morphology	3-1	-3.27	-5.15	-1.38	0.002	0.769	0.466	2.53	0.021*
	CG Height		0.32	0.10	0.54	0.006				
	CG	2-1	2.95	0.83	5.07	0.009				
CC Upight	Morphology	3-1	4.86	1.15	8.57	0.013	0.751	0 422	2 20	0.024*
CG Height	CG Length		0.75	0.16	1.33	0.015	0.751	0.422	2.29	0.034*
	CG Width		1.03	0.33	1.73	0.006				

CI: Confidence Interval. Type 1 was the reference category for CG morphology. \*P < 0.05; \*\*P < 0.01; \*\*\*P < 0.001.

Predictor	Estimate SE		Z OR		%95 CI		2
Fredictor	Estimate	JL	2	UK	Lower	Upper	– p
CG length	-0.57	0.21	-2.66	0.57	0.36	0.84	0.008**
CG width	-0.47	0.38	-1.24	0.62	0.28	1.31	0.214
CG height	0.21	0.20	1.04	1.23	0.86	1.91	0.298
Age	0.07	0.05	1.53	1.07	0.98	1.18	0.126
Gender (Male - Female)	2.81	0.82	2.62	8.54	1.096	51.02	0.009**
CG Morphology (Type 2-1)	-1.76	0.95	-1.84	0.17	0.02	1.07	0.066
CG Morphology (Type 3-1)	-4.46	2.38	-1.87	0.01	0.00	0.69	0.061
Keros Classification (Type 2-1)	-0.13	1.06	-0.12	0.88	0.11	7.11	0.904
Keros Classification (Type 3-1)	-0.16	1.36	-0.12	0.85	0.05	11.91	0.905
Pneumatization (Present-Absent)	1.70	1.63	1.04	5.47	0.23	197.55	0.297

Table 4. Table for ordinal logistic regression analysis between skeletal class and crista galli morphometry and morphology measurements.

**Deviance:** 78.11. AIC: 102.11,  $R^2_{McFadden's}$ : 0.21,  $R^2_{cox&Snell's}$ : 0.14,  $R^2_{Nagelkerke's}$ : 0.27. **Overall Model Test:** X<sup>2</sup>, 20.77, p=0.023. CI: Confidence Interval. CG: Crista Galli. OR: Odds Ratio. Reference category: Skeletal Class I. \*P < 0.05; \*\*P < 0.01; \*\*\*P < 0.001.

#### Discussion

The ossification of the CG, which is seen as a roostercomb-like structure on the midline of the cribriform plate of the ethmoid bone in the second month of fetal development, generally begins in the second month after birth and increases until the 14th month, and then gradually progresses until completion in the 24th month.<sup>1,8,19</sup> This structure, which is closely related to the central structures in the frontal skull base, especially the falx cerebri that separates the right and left cerebral hemispheres, has an extremely important place in endoscopic sinus and skull base surgeries due to its location, shape, and dimensions.<sup>2,20</sup>]. The skull base has an important place in the classification and definition of skeletal malocclusions.<sup>21</sup> Therefore, a relationship may exist between structures involving the skull base and skeletal malocclusions.<sup>21,22</sup> This study thus aimed to examine the relationship between the CG, located in the anterior part of the skull base, and skeletal malocclusions.

The review study of Gong et al. examined cranial base characteristics in various anteroposterior skeletal malocclusions and reported that cranial base length differed morphologically.<sup>21</sup> Cranial base lengths were smaller in skeletal class III malocclusion. However, in that study, an examination was provided only from lateral cephalometric radiographs, which provide a twodimensional view. In our current study, the fact that the anterior-posterior length of the CG of Class I individuals is larger than that of Class III individuals, but has a similar length to Class II individuals, adds another dimension to this information. This adds new information to the literature. Again, in the study of Gong et al.<sup>21</sup>, the cranial base length and angle were smaller in class III individuals than in class I individuals, which somewhat supports our findings.

Amano et al. examined the activity of the neural crest cells that form the anterior craniofacial skeleton and facial structure. They reported that the developmental effect between the nasomaxillary complex and skeletal malocclusions was significant.<sup>23</sup> They revealed that the genes causing hypoplastic nasomaxillary complex are

related to skeletal malocclusion. This may lead us to think that variations in the nasomaxillary complex, which includes the CG and is in proximity, may vary according to skeletal malocclusions. In this context, although a relationship between CG and skeletal malocclusions may be predicted, an exact relationship could not be revealed in our study. Only a difference between skeletal malocclusions in the linear anteroposterior length of the CG was demonstrated. Additionally, the regression analysis determined that the CG length could be seen as a low risk factor in the development of skeletal malocclusion. The main factor affecting the linear measurements of the CG was CG morphology. Among the cephalometric values, only the SN/GoGn angle had a low impact on CG length. Moreover, the presence of a positive correlation between CG width and height in skeletal malocclusion control supports these findings and strengthens the definition of the relationship between them. However, further studies are needed.

A recent study by Chou et al. examined the relationship between cranial base measurements and sagittal skeletal malocclusions.<sup>24</sup> In that study, the CG length was examined, and it was like the findings of our study. Consistent with our work, the width of the cribriform plate of the ethmoid bone did not differ between skeletal malocclusions. In the study of Rai et al., referring to the vertical length of the face, they reported that cranial base lengths were longer in individuals with long faces.<sup>25</sup> This literature information may provide the idea that there may be a relationship between cranial base lengths and facial length, and that the crista galli located in the anterior cranial base may also be indirectly affected by this. In this study, various relationships were found between CG dimensions and vertical measurement parameters of the face. Additionally, although no correlation existed between the CG and cribriform plate measurements in that study, a significant correlation was found between CG height and width in our study. Our study found a positive correlation between CG length and Co-Gn and S-Go cephalometric measurements and between N-ANS and CG height. These findings suggest a possible weak relationship between skeletal facial length and height and CG dimensions. Furhermore, there is insufficient information in the literature about the relationship between crista galli and maxillomandibular dimensions. This study constitutes one of the first examples of attribution.

Many studies have examined the relationships between CG morphology and morphometry and gender and the role of this structure in gender determination.<sup>6-8</sup> Different from those studies, our study found no difference in CG morphology and morphometry between genders. Therefore, comparisons were made between skeletal malocclusion groups. Additionally, some studies examining gender determination have involved CT examinations, with no examination of skeletal malocclusions.<sup>6,7</sup> The reason for the lack of gender differences in our study can be considered the number of samples. This provides a limitation, but the fact that our research question was not about gender and that the study represents a guide for further research supports its contribution to the literature. Our study used CBCT, which causes lower radiation exposure than conventional CT devices.<sup>8,26</sup> The protection of individuals from radiation in line with the ALARA principle was considered <sup>27</sup>, and the study also aimed to raise awareness for clinicians providing CBCT examinations. In our study, unlike the literature, CG pneumatization was examined but not encountered. The reason for this may be the very low frequency of pneumatization, as stated in the literature, and the sample size of our study. Future studies should focus more on this.

#### Limitations

One of the main limitations of our study is its low sample size. As a precaution, a power analysis was performed before the study. Additionally, image acquisition in a wide FOV range to enable the CG to be visible is limited, especially in individuals with skeletal class I malocclusion and who have received CBCT for various diagnostic reasons. Our knowledge of health care services, which primarily aims to protect individuals, foresees minimal radiation exposure in line with the ALARA principle.<sup>27</sup>

#### Conclusions

- The CG length of skeletal class I individuals is greater than that of class III individuals. The CG length of skeletal Class II individuals is like both Class I and Class III individuals.
- The CG width and height do not differ according to skeletal malocclusion classes.
- CG morphology did not differ according to sagittal skeletal malocclusions.
- A positive correlation existed between the effective mandibular length (Co-Gn) and the anteroposterior length of the CG.
- A weak relationship may exist between certain cephalometric parameters and CG dimensions.

#### Acknowledgments

Author Contributions Conception or design of the work: Taner Ozturk; Data collection: Yunus Emre Ucaker, Aykagan Cukurluoglu; Data Analysis: Taner Ozturk, Aykagan Cukurluoglu; Drafting the article: Taner Ozturk, Yunus Emre Ucaker; Critical revision of the article: Taner Ozturk; Final approval of the version to be published: Taner Ozturk, Yunus Emre Ucaker, Aykagan Cukurluoglu.

#### **Ethical Approval**

The ethical approval required for this retrospective study was obtained from the Erciyes University Clinical Research Ethics Committee (Approval no: 2023/338; Approval date: May 10<sup>th</sup>, 2023). The study was carried out in accordance with the Declaration of Helsinki, and informed consent forms were obtained from all individuals at the beginning of the study.

#### **Consent to participate**

Informed consent was obtained from all individual participants included in the study

#### **Conflict of Interest**

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

#### Funding

None.

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## **Cumhuriyet Dental Journal**

| cdj.cumhuriyet.edu.tr |

Founded: 1998

Available online, ISSN: 1302-5805 e-ISSN: 2146-2852

Publisher: Sivas Cumhuriyet Üniversitesi

## Assesment of Youtube<sup>™</sup> Videos About all-on-four Implant Supported Fixed Prostheses Maintenance Using the Global Quality Score, the DISCERN Tool, and Journal of American Medical Association Benchmarks

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Research Article	ABSTRACT
History	Objectives: The aim of the study is to evaluate the information on "all-on-four fixed implant prostheses maintenance" videos shared on YouTube™. Material-Methods: A search was made on YouTube™ at 09:00 on August 14, 2023, with the search term "all-on-
Received: 29/04/2024 Accepted: 25/10/2024	four fixed implant prostheses maintenance". Videos uploaded in the last year are sorted by relevance. The first 60 videos were viewed and analyzed for content by 2 independent prosthodontists. GQS (Global Quality Score), DISCERN, and JAMA (Journal of the American Medical Association) scales were used for the evaluation of the videos. <b>Results:</b> Some of the videos obtained for the study were excluded from evaluation because they did not meet the "all-on-four fixed implant prostheses maintenance" requirements (n=39). A statistically significant relationship was found between GQS scores and DISCERN scores (p<0.05) that as the DISCERN score increases, the GQS score increases. Also, videos with DISCERN scores of 4 and 5 are longer than videos with scores of 3. JAMA scores of videos with a longer duration and more time since they were uploaded were found to be statistically significantly higher (p<0.05). <b>Conclusion:</b> The quality of YouTube™ videos are generally poor. Patients should consider this when they want to obtain information about "all-on-four fixed implant prostheses maintenance" from YouTube™.

Keywords: All-On-Four, Fixed Implant Prostheses, Maintenance, Dental Implant.

## All-On-Four İmplant Destekli Sabit Protezlerin İdamesi Hakkındaki YouTube™ Videolarının Global Kalite Skoru, DISCERN ve Journal of American Medical Association Araçları ile Değerlendirilmesi

Research Article	ÖZ
	Amaç: Çalışmanın amacı YouTube™'da paylaşılan "all-on-four implant destekli sabit protez idamesi" videolarına
Süreç	ilişkin bilgilerin değerlendirilmesidir.
	Gereç-Yöntemler: 14 Ağustos 2023 saat 09:00'da YouTube™'da "all-on-four sabit implant protez idamesi"
Geliş: 29/04/2024	anahtar sözcüğü ile arama yapılmıştır. Son bir yıl içinde yüklenen videolar alaka düzeyine göre sıralanmıştır. İlk
Kabul: 25/10/2024	60 video 2 bağımsız protez uzmanı tarafından değerlendirilmiş ve içerik açısından analiz edilmiştir. Videoların
	değerlendirilmesinde GQS (Global Quality Score), DISCERN ve JAMA (Journal of the American Medical
	Association) ölçekleri kullanılmıştır.
	Bulgular: Çalışma için elde edilen videolardan bazıları "all-on-four sabit implant protez idamesi" gerekliliklerini
	(n=39) karşılamadığı için değerlendirme dışı bırakılmıştır. GQS puanları ile DISCERN puanları arasında istatistiksel
	olarak anlamlı bir ilişki bulunmuş (p<0,05), DISCERN puanı arttıkça GQS puanın da arttığı tespit edilmiştir. Ayrıca
	DISCERN puanı 4 ve 5 olan videolar, puanı 3 olan videolara göre daha uzun olduğu tespit edilmiştir. Süresi daha
License	uzun olan ve yüklendikten sonra daha uzun süre geçen videoların JAMA puanları istatistiksel olarak anlamlı
License	derecede yüksek bulunmuştur (p<0,05).
	<b>Sonuçlar:</b> YouTube™ videolarının kalitesi genellikle düşüktür. Hastalar YouTube™'dan "all-on-four sabit implant
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	Anahtar Kelimeler: All-On-Four, İmplant Destekli Sabit Protezler, İdame, Dental İmplant.
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How to Cite: Akcin ET, Ates G. (202	(4) Assesment of Youtube™ Videos About all-on-four Implant Supported Fixed Prostheses Maintenance Using the Global

#### Introduction

Dental implants are a frequently preferred treatment option in the rehabilitation of edentulous jaws.<sup>1</sup> However, when the maxillary sinuses in the upper jaw the mandibular canal, mental foramen in the lower jaw approach the residual crest due to crestal bone loss, implant surgical applications bring difficulties.<sup>2</sup> The all-on-four technique has been used safely for years to overcome these difficulties and ensure dental implant placement in atrophic edentulous jaws without damaging the anatomical structures.<sup>3</sup>

In the all-on-four technique, four implants are placed between the mental foramens in the mandible. For the maxillary arch, four implants are placed in the area anterior to the maxillary sinuses. The necks of the distal implants are placed at an angle towards the distal, the apexes towards the mesial so that the emergence profile of the abutments is brought to the level of the first molar teeth.<sup>4</sup> After osseointegration, implant-supported fixed prostheses that mimic both soft tissue and hard tissue are delivered to the patient.

It is extremely important to fulfill oral hygiene requirements after prosthesis delivery. Tooth brushing, the use of dental floss and interdental brushes, oral irrigators, and mouthwashes play an active role in all-on-four implantsupported fixed prosthesis care. If daily oral hygiene requirements are not applied and proper care is not taken, inflammation may occur in the soft tissues surrounding all-onfour fixed implant prostheses.<sup>5</sup> As inflammation progresses to the alveolar bone, bone loss occurs around the implant, and implant loss may occur.<sup>6</sup> If the all-on-four implant-supported fixed prostheses, which are applied with a small number of implants to patients with already limited bone tissue, are not properly maintained by the patient and regular dentist checkups are not performed, irreversible problems may arise. Due to bone loss, the patient may lose the chance to have new dental implants.

YouTube™ (https://www.youtube.com) is a video-sharing with a large user network that allows users to create and watch videos containing health information.<sup>7</sup> Every day a large amount of videos are uploaded and viewed by millions of people. There are a lot of videos about all-on-four fixed implant prosthesis maintenance on YouTube<sup>™</sup> and they have great effects on patients who wear all-on-four fixed implantsupported fixed prostheses. Patients view these videos to learn how to clean their prostheses to use their prostheses for a long time. So, correct and appropriate information should be given on videos for a long life of all-on-four fixed implant prostheses maintenance. Videos are made available without the information which they contain being evaluated by an independent and blind referee system. They may be problems such as incorrect diagnosis and/or treatment.<sup>8</sup> Previous studies reported that YouTube<sup>™</sup> videos about various health tissues often contain misleading and inadequate information.9-13

This study aimed to evaluate the information on "all-onfour fixed implant prostheses maintenance" videos shared on YouTube<sup>™</sup>. The hypothesis of the present study was that YouTube<sup>™</sup> videos on all-on-four fixed implant prostheses maintenance contain misleading or incomplete information.

#### **Materials and Methods**

Ethical approval was not required because of the use of the publicly available data. A search was made on YouTube<sup>TM</sup> at 09:00 on August 14, 2023, with the search term "all-on-four fixed implant prostheses maintenance". Videos uploaded in the last year are sorted by relevance. It was reported in previous studies that when searching on YouTube<sup>TM</sup>, users could look at a list between 60-200<sup>14</sup>, but the majority would choose to watch only the first 30 videos listed so that the first 60 videos were evaluated.<sup>15,16</sup>

All videos were viewed and analyzed for content by 2 independent prosthodontists (E.T.A. and G.A.). Videos' information (duration, date of upload, number of likes&dislikes... etc) were recorded. The viewing rates (Number of views/Number of days since upload ×100%) and the interaction index [(Number of likes-Number of dislikes) / Total number of views×100%)] are also calculated according to the formula used in a previous study.<sup>17</sup>

Videos in languages other than English, implant training videos for dental professionals, implant-supported prostheses clinical try-in videos, and irrelevant videos were excluded. After that, all remaining videos were analyzed and GQS (Global Quality Score), DISCERN, and JAMA (Journal of the American Medical Association) scales were used for the evaluation of the videos.

GQS<sup>18,19</sup> was used to evaluate the general quality of the videos. The score description is listed in Table 1. DISCERN tool<sup>20</sup> consists of 16 questions (Table 2) and each questions are scored 1 to 5. Using DISCERN, videos were divided into 5 groups according to total score: very poor(16 to 20), poor (27 to 38), fair (39-50), good (51 to 62), and excellent (above 63).<sup>21</sup> JAMA (Journal of the American Medical Association) evaluation tool<sup>22</sup> is used for reliability and usefulness of health-related information. JAMA benchmark criteria have 4 sections and each one is scored 0 to 4.<sup>21</sup> The criteria and descriptions are listed in Table 3.

Data were analyzed with IBM SPSS 25. The Shapiro-Wilk Test was used to assess conformity to the normal distributon. Homogenity of variance was assessed with The Levene Test. Independent Sample-T test was used for comparing two independent groups with normal distribution. In cases where the assumption was not met, Mann Whitney U test was used. ANOVA test was used to compare the means of three or more groups with normal distribution, and the Kruskal Wallis test was used when the assumption was not met. The Post Hoc Tamhane T2 test was applied to reveal the group or groups that made the difference. Kendall's Tau correlation was used to examine the relationship between ordered categorical variables and continuous variables. To examine the relationship between categorical variables, Fisher's Exact test was used when the sample size assumption was not met. The significance level was taken as *p*<0.05.

#### Table 1. Global Quality Score (GQS) Five-Point Scale.

Score D	Description
1	Poor quality, poor flow of video, most information missing, not at all useful for patients
2	Generally near quality and near flow, some information listed but many important tonics but of limited use to pat

Generally poor quality and poor flow, some information listed but many important topics but of limited use to patients
 Moderate quality, suboptimal flow, some information is adequately discussed but others poorly discussed, somewhat

- 3 useful for patients
- 4 Good quality generally good flow, most relevant information is covered, useful for patients

5 Excellent quality and flow, very useful for patients

#### Table 2. DISCERN Questions.

Section 1: Is the publication reliable?

1. Are the aims clear?

2. Does it achieve its aims?

3. Is it relevant?

4. Is it clear what sources of information were used to compile the publication (other than the author or producer)?

5. Is it clear when the information used or reported in the publication was produced?

6. Is it balanced and unbiased?

7. Does it provide details of additional sources of support and information?

8. Does it refer to areas of uncertainty?

Section 2: How good is the quality of information on treatment choices?

9. Does it describe how each treatment works?

10. Does it describe the benefits of each treatment?

11. Does it describe the risks of each treatment?

12. Does it describe what would happen if no treatment is used?

13. Does it describe how the treatment choices affect the overall quality of life?

14. Is it clear that there may be more than one possible treatment choice?

15. Does it provide support for shared decision-making?

16. Based on the answers to all of the above questions, rate the overall quality of the publication as a source of information about treatment choices

#### Table 3. JAMA benchmark criteria.

Criteria	Description
Authorship	Authors and contributors, their affiliations, and relevant credentials should be provided
Attribution	References and sources for all content should be listed clearly, and all relevant copyright information noted
Disclosure	Web site "ownership" should be prominently and fully disclosed, as should any sponsorship, advertising, underwriting, commercial funding
Currency	Dates that content was posted and updated should be indicated

#### Results

Some of the videos obtained for the study were excluded from evaluation because they did not meet the "all-onfour fixed implant prostheses maintenance" requirements (Table 4). Characteristic features of the included videos (duration in minutes, days since upload, number of likes, etc.) are given in Table 5. 38.1% (n=8) of the evaluated videos were uploaded by dentists and 61.9% (n=13) were uploaded by dental clinics. Global Quality score distributions of the included videos are shown in Table 6.

Kendall's Tau correlations were applied to examine the relationships between the characteristics of the videos and the scores. As a result of the analysis, no statistically significant relations were obtained between the characteristics of the videos and GQI, JAMA, and DISCERN scores (p>0.05) (Table 7).

Fisher's Exact tests were performed to investigate the relationships between the characteristics of the videos and GQS scores. As a result, a statistically significant relationship was found between GQS scores and DISCERN

scores (p<0.05). It has been determined that as the DISCERN score increases, the GQS score increases.

Anova and Kruskal Wallis tests were applied to compare the characteristics of the videos according to DISCERN scores. A statistically significant difference was found between the lengths of the videos according to the DISCERN groups (p < 0.05). According to Tamhane tests, a statistically significant difference was detected between scores 3 and scores 4 and 5 (p=0.002). Videos with DISCERN scores of 4 and 5 are longer than videos with scores of 3.

Independent Sample T Test and Mann Whitney U Test were performed to investigate the differences between the characteristics of the videos and JAMA scores. Statistically significant differences were found between the JAMA scores, the duration of the videos and the average time elapsed since the date they were uploaded (p<0.05). The duration of videos with JAMA scores of 3 and 4 and the average of the time since the date they were uploaded are higher than the average of the duration and the time since the date of upload of the videos with JAMA scores of 1 and 2.

#### Table 4. Reasons for exclusion.

	n	%			
Excluded videos (not related to subject)	39	65.0			
Included videos	21	35.0			
Total	60	100.0			

#### Table 5. Distribution of YouTube videos according to their characteristic features.

	Minimum	Maximum	Average	Standard Deviation	Median
Duration in minutes	0.32	17.30	6.43	5.23	4.20
Days since upload	36.00	300.00	196.10	73.15	191.00
Number of likes	1.00	562.00	100.38	139.39	46.00
Number of comments	0.00	62.00	16.14	20.22	6.00
Number of subscriptions	105.00	941.00	360.17	301.74	286.50
Number of views	22.00	35872.00	7293.38	10331.77	1991.00
Viewing Rate	16.06	17329.47	3757.25	5089.08	1217.84
Interaction Index	0.23	4.55	2.21	1.14	1.94

#### Table 6. Distribution of Global Quality Score (GQS) Five-Point Scale Scores.

	n	%
Poor quality, poor flow of video, most information missing, not at all useful for patients	0	0
Generally poor quality and poor flow, some information listed but many important topics but of limited use t patients	<sup>0</sup> 4	19.0
Moderate quality, suboptimal flow, some important is adequately discussed but others poorly discussed, somewhat useful for patients	at 3	14.3
Good quality generally good flow, most relevant information is covered, useful for patients	3	14.3
Excellent quality and flow, very useful for patients	11	52.4

#### Table 7. Relationships between the characteristic features of the videos and the scores.

		GQS	JAMA	DISCERN
Duration in minutes	r	-0.221	-0.150	0.111
Duration in minutes	р	0.209	0.396	0.540
Days since upload	r	-0.229	-0.073	0.185
Days since upload	р	0.249	0.714	0.367
Number of likes	r	0.260	0.194	0.034
Number of likes	р	0.137	0.267	0.848
Number of comments	r	0.179	0.103	-0.034
Number of comments	р	0.306	0.557	0.848
Number of subscriptions	r	-0.306	-0.148	0.034
Number of subscriptions	р	0.080	0.396	0.848
Number of views	r	-0.221	-0.150	0.111
Number of views	р	0.209	0.396	0.540
Viewing Rate	r	-0.229	-0.073	0.185
	р	0.249	0.714	0.367
Interaction Index	r	0.260	0.194	0.034
	р	0.137	0.267	0.848

#### Discussion

Daily home care is very important for the safe use of all-on-four implant-supported fixed prostheses by the patient for many years. Setti *et al.*<sup>6</sup> investigated the use of angled toothbrushes in implant-supported full-arch dentures and reported that the symptom of bleeding on probing in the experimental group decreased statistically significantly. The study emphasized that there is a serious deficiency in the literature regarding the cleaning and maintenance of implant-supported fixed prostheses and long-term validated home-care oral hygiene procedures should be established.

Post-operative information about the care of the prostheses and patient education provided by dental professionals. In addition to verbal explanations, videos,

and visual representations make it easier for patients to apply the oral hygiene requirements. In this context, YouTube<sup>™</sup> videos serve patients as an important data source.<sup>12</sup> According to some researchers, YouTube<sup>™</sup> videos contain personal opinions and they are not based on scientific findings.<sup>23–25</sup> For this reason, the content and the quality of information in the videos should be evaluated.

Yağcı *et al.* evaluated 200 YouTube<sup>™</sup> videos providing information about cleaning dentures and compared their GQS scores. As a result of the study, it was emphasized that videos were insufficient and should not be considered the only source for denture cleaning.<sup>19</sup> Menziletoğlu *et al.*<sup>26</sup> examined whether dental implant videos on YouTube<sup>™</sup> were useful for patient education and compared the GQS scores of the videos. They reported that the duration of excellent videos was longer than others. Consistent with our study, GQS and DISCERN scores increased as video duration increased. According to the results of the study, it was reported that important parameters related to implant treatment, such as implant maintenance, oral hygiene requirements, periimplantitis, and implant loss were not included in the videos.<sup>26</sup> This situation also makes the job of professionals providing dental implant services difficult. 38% of doctors think that the information obtained by patients negatively affects doctor-patient appointments, and the possible reason for this situation may be videos with poor content.<sup>27</sup>

In a previous study, the viewing rates, likes, GQS, DISCERN, usefulness scores of videos uploaded by healthcare professionals were found to be statistically significantly higher than those of videos uploaded by individual users.<sup>28</sup> All of the videos evaluated in our study were uploaded by dental professionals, and consistent with this study, interaction index and GQS scores increased as the duration of the videos increased.<sup>28</sup> Kurian et al.,<sup>29</sup> searched with the keyword "Complete arch fixed treatment using dental implants" and reached a total of 508 videos and evaluated 89 videos that met the inclusion criteria. They reported that dental implant contraindications, survival rates, prognoses, and possible complications were not emphasized in the videos. Only 1% of the videos received an excellent score according to GQS. Most of the videos did not mention oral hygiene, implant maintenance, and complications of prosthesis. 78% of the videos were described as poor. Contrary to existing studies, 52.4% (n=11) of the videos in our study received an excellent GQS score. A possible explanation for this may be that all the videos evaluated had good content because they were uploaded by dentists or dental clinics.

In a study examining YouTube<sup>™</sup> videos about dental implants, 117 videos were evaluated according to the "Information for patient" available at the American Academy of Implant Dentistry, the European Association of Osseointegration, and the British Society of Restorative Dentistry.<sup>17</sup> According to the results of the study, it was reported that 35 of the videos contained incorrect information about dental implant prognosis and maintenance. It was emphasized that the videos were low quality and unreliable. In our study, GQS, JAMA, and DISCERN tools, which are frequently used to evaluate YouTube<sup>™</sup> videos, were used and 14.3% (n=3) of the videos received moderate, 14.3% (n=3) good and 52.4% (n=11) excellent scores.

In a previous study, 20 videos were evaluated and their GQS scores were compared. After 1 month the videos were re-evaluated and consistency and reliability were analyzed.<sup>19</sup> In another study, a pilot study was conducted for analysis and calibration by observers then 20 videos were re-watched at different times. Both intraobserver and interobserver reliability were examined.<sup>28</sup> In another study, before evaluating the videos, evaluators were trained to perform the study, and evaluation calibration was performed. Kohen's kappa coefficient was used to

evaluate this calibration.<sup>17</sup> In our study, the evaluators were not subjected to prior training and calibration, and no pilot study was conducted. All videos were evaluated once by both researchers. Unlike studies comparing interrater reliability<sup>23,29</sup>, this comparison was not made in our study.

Limitations of this study included that, because YouTube<sup>TM</sup> is a dynamic platform, these results only reflected information available at the time of the search. Also, these results are specific to keywords. If the keyword changes or extra keywords are added, results change.<sup>17,19,23,26</sup> Although searches with a single keyword yield better<sup>9,30</sup> the scanning area should be improved by adding extra keywords.<sup>28</sup>

#### Conclusions

The quality of YouTube<sup>™</sup> videos is generally poor. Patients should take this into consideration when they want to obtain information about "all-on-four fixed implant prostheses maintenance" from YouTube<sup>™</sup>. If dental professionals use scales such as GQS, JAMA, and DISCERN when uploading videos to YouTube<sup>™™</sup>, better quality content will be enhanced. So, patients can access better-quality information.

#### Acknowledgments

The authors declare that they have o conflict of interest.

No source of funding were used to assist in the preparation of this paper.

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## The Relationship Between Stress and Depression Levels in Patients with Temporomandibular Disorder

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Research Article	ABSTRACT
History	<b>Objectives:</b> The aim of this study was to examine the relationship between anxiety and depression levels in patients with temporomandibular disorder (TMD). <b>Material-Methods:</b> A total of 100 patients who applied to the Oral and Maxillofacial Surgery Clinic between
Received: 26/07/2024	March and June 2024 were included in the study. Fonseca anamnestic index (FAI) was used to evaluate signs
Accepted: 09/12/2024	and symptoms related to TMD and to determine symptom severity. Beck anxiety inventory (BAI) was used to evaluate anxiety. Beck depression inventory (BDI) was used to evaluate depression. Mean scores were evaluated between patients with and without TMD. Statistically significant differences were evaluated as p<0.05. <b>Results:</b> A total of 100 patients, 52 female and 48 male, aged between 20 and 65 (mean 29.12±9.19) were included in the study. The average FAI values of all patients were found to be 30.85±22.82, and TMD was found in 56% of the patients according to the FAI values. The mean BDI and BAI values of patients with TMD were found to be 12.84±6.11 and 13.61±10.29, respectively. The mean BDI and BAI values of patients without TMD were found to be 9.25±6.88 and 7.16±6.34, respectively. The mean BDI and BAI values in TMD patients were found to be significantly higher (p<0.05) than in patients without TMD. <b>Conclusion:</b> There is a strong connection with TMD, depression and anxiety. High anxiety and depression have been observed in individuals with TMD.

Keywords: Anxiety, Depression, Temporomandibular Disorder.

## Temporomandibular Bozukluğu Olan Hastalarda Stres ve Depresyon Düzeylerinin İlişkisi

Research Article	ÖZ
	Amaç: Bu çalışmanın amacı temporomandibular bozukluğu (TMB) olan hastalarda anksiyete ve depresyon
Süreç	seviyelerinin ilişkisini incelemektir.
	Gereç-Yöntemler: 2024 yılının mart ve haziran ayları arasında Ağız, Diş ve Çene Cerrahisi Kliniğine başvuran 100
Geliş: 26/07/2024	hasta çalışmaya dahil edildi. Fonseca anamnestik indeksi (FAI), TMB'ye bağlı belirti ve semptomları
Kabul: 09/12/2024	değerlendirmek ve semptom şiddetini belirlemek için kullanıldı. Beck anksiyete ölçeği (BAÖ), anksiyeteyi
	değerlendirmek için kullnıldı. Beck depresyon ölçeği (BDÖ), depresyonu değerlendirmek için kullanıldı. Ortalama
	puanlar TMB olan ve TMB olmayan hastalar arasında değerlendirildi. İstatistiksel olarak anlamlı farklılık için p<0.05 olacak şekilde değerlendirildi.
	<b>Bulgular:</b> Çalışmaya yaşları 20 ile 65 arasında değişen (ortalama 29,12±9,19), 52 kadın ve 48 erkek toplam 100
	hasta dahil edildi. Tüm hastaların FAI değerleri ortalaması 30,85 $\pm$ 22,82 olarak bulundu ve FAI skoruna göre
	hastaların %56'sında TMB bulundu. TMB olan hastaların ortalama BDÖ ve BAÖ değerleri sırasıyla 12,84 $\pm$ 6,11
License	ve 13,61 $\pm$ 10,29 olarak bulundu. TMB olmayan hastaların ortalama BDÖ ve BAÖ değerleri sırasıyla 9,25 $\pm$ 6,88
	ve 7,16 $\pm$ 6,34 olarak bulundu. TMB hastalarındaki ortalama BDÖ ve BAÖ değerleri TMB olmayan hastalara oranla
	anlamlı derecede (p<0,05) daha fazla bulundu.
This work is licensed under	Sonuçlar: TMB, depresyon ve anksiyete ile kuvvetli bir bağlantı bulunmaktadır. TMB olan bireylerde yüksek
Creative Commons Attribution 4.0	anksiyete ve depresyon gözlenmiştir.
International License	
	Anahtar Kelimeler: Anksiyete, Depresyon, Temporomandibular Bozukluk.
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	024) The Relationship Between Stress and Depression Levels in Patients with Temporomandibular Disorder, Cumhuriyet
Dental Journal, 27(4):2	(30-239.

#### Introduction

Temporomandibular disorders (TMD) are multifactorial conditions that involve a variety of clinical problems, including pain and functional limitations, affecting the joint, the masticatory muscles in the joint area, and the bones and nerves in the area.<sup>1</sup> Although the etiology of TMD is not known with certainty, it is assumed to be a multifactorial condition involving the interconnections of psychological and physiological factors, factors such as occlusion and trauma, parafunctional habits such as teeth grinding, and genetic factors. The combination of these factors can disrupt the static balance of the masticatory system and cause the signs and symptoms of TMD.<sup>2</sup> The most common signs and symptoms of TMD are chronic pain, pain in the jaw muscles, restriction of movements, and sounds coming from the jaw temporomandibular joint.<sup>3</sup> TBM affects 15% to 20% of adult patients, with the highest incidence occurring between the ages of 20 and 40. It tends to be more common in female.<sup>4</sup>

Psychological/psychosocial and stress factors also attract attention in temporomandibular joint disorders, as it is a multifactorial structure that is open to more than one etiology. Therefore, the role of stress and personality factors in the etiology of TMD is subject to a comprehensive examination.<sup>5</sup> The onset of TMD symptoms has been associated with high levels of anxiety, stress, and.<sup>6</sup> Psychological studies have shown that patients with TMD have similar psychological definitions to patients with chronic pain disorders such as tension-type headaches and back pain that affect the musculoskeletal system.<sup>7</sup> Although the DC/TMD classification is a standardized and widely used test for the differential diagnosis of TMDs, the complexity and difficulty of its use have led to the frequent use of less difficult diagnostic tests such as the Fonseca Anamnestic Index (FAI).<sup>8</sup> FAI identifies the presence of TMD. It is a simple and low-cost patient-reported method used to differentiate between and absent.<sup>9</sup>

The aim of this study is to compare the depression and anxiety levels between individuals with TMD and healthy individuals. While TMD diagnosis was evaluated with FAI, anxiety level was evaluated with Beck anxiety inventory (BAI), and depression level was evaluated with Beck depression inventory (BDI).

#### **Materials and Methods**

#### Ethical Aspects and Study Design

Participants were fully informed about the study and took part voluntarily, adhering to the principles outlined in the Declaration of Helsinki. Approval for the study was obtained from the Eskişehir Osmangazi Non-Interventional Clinical Research Ethics Committee (Decision no: 2024/10). The study included 100 patients who visited the Eskişehir Osmangazi University Faculty of Dentistry, Department of Oral and Maxillofacial Surgery clinic, between March and June 2024. Patients who were younger than 18 years of age, had systemic health problems, were missing survey data, and did not fully fill out the evaluation scales were excluded from the study. An experienced oral and maxillofacial surgeon who was trained on this subject examined all patients participating in the study and administered the surveys regarding the study.

#### **Diagnosis of TMD**

The diagnosis of TMDs was determined using the Turkish version of the FAI. The psychometric properties and diagnostic accuracy of this index have been validated by comparison with the Diagnostic Criteria for TMD (DC/TMD).<sup>9,10</sup> FAI is an index used to evaluate TMD-related pain frequency, psychological stress, jaw functional limitations, and parafunctional behaviors. It is a survey consisting of questions. "No", "sometimes" and "yes" answers correspond to 0, 5 and 10 points, respectively. FAI shows the severity of TMD between 0 and 100.<sup>11</sup>

#### **Evaluation of Anxiety**

Anxiety scores were evaluated using BAI.<sup>12</sup> This questionnaire contains a total of 21 items that reflect anxiety symptoms somatically, emotionally and cognitively. Validation and reliability of the scale in Turkish was carried out by Ulusoy et al. <sup>13</sup> BAI is a survey consisting of 21 multiple-choice questions regarding the participant's anxiety levels regarding how they have been feeling lately. The answers to the questions are scored between 0 and 3. High scores from the answers given indicate high levels of anxiety symptoms.<sup>14</sup>

#### **Evaluation of Depression**

Depression was assessed using the BDI questionnaire. The validity and reliability of this tool and its Turkish version were confirmed by Hisli et al.<sup>15</sup> to predict the level of depression. This scale evaluates the emotional state of the patient through 21 questions related to his/her mental state in the last 24 hours. The total score is calculated by summing the responses.<sup>16</sup>

#### **Statistical Analysis**

SPSS 22.0 Windows (IBM Corporation, Armonk, NY, USA) program was used in the analysis of the data obtained in the study. Mean, standard deviation and minimum-maximum values were taken into account in descriptive statistics. The suitability of variables for normal distribution was assessed using the Shapiro-Wilk test. In comparisons between two groups, independent sample t-test was applied for data showing normal distribution and Mann-Whitney U test was applied for data not showing normal distribution.

#### Results

100 patients aged between 20 and 65 (mean: 29.12±9.19) were included in the study. According to the FAI score, TMD was observed in 56 patients, while TMD was not observed in 44 patients. 52% of the participants were female and 48% were male patients. Minimum maximum mean and SD FAI, BDI and BAI values of all patients are shown in Table 1.

Shows the mean/SD BDI and BAI scores for the TMD and WTMD groups for all participants. The TMB group showed significantly higher BDI and BAI scores than the WTMD group (p<0.05). (Table 2)

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	Variables	Minimum	Maximum	Total (n=100)
	Variables	Minimum	Waximum	Mean±SD
	BDI	0	32	11.26±6.67
	BAI	0	55	10.77±9.31
	FAI	0	90	30.85±22.82
	Age	20	65	29.12±9.19

Table 1. Minimum maximum mean and SD values of FAI, BDI and BAI scores.

Table 2. Relationship between BAI and BDI scores of individuals with TMD (FAI).

Variables	Total (n=100) Mean±SD	TMD (n=56)	WTMD (n=44)	p-value
BDI	11.26±6.67	12.84±6.11	9.25±6.88	<0.005*
BAI	10.77±9.31	13.61±10.29	7.16±6.34	<0.005*
TMD tomporomondibular	licardary W/TMD without tomas	romondihular dicardaru *stati	stigal significance, DDL Deck de	arassian inventory DAL Book

TMD, temporomandibular disorder; WTMD, without temporomandibular disorder; \*statistical significance; BDI, Beck depression inventory; BAI, Beck anxiety inventory

#### Discussion

Although many factors are involved in the etiology of TMD, psychosocial factors play a role in its onset or progression.<sup>17</sup> Literature reviews have found a correlation between TMD and psychological factors.<sup>18</sup> A recent study also reported a relationship between TMD and depression and anxiety.<sup>19</sup> Another study indicated that the prevalence of TMD among university students was 46.8%, and emotional stress was 30.5%.<sup>20</sup> In a study conducted on adolescent individuals in Asia, it was found that those with TMD were more stressed than individuals without TMD. In a study including 578 Chinese individuals, they reported that 61.4% showed more than one symptom, anxiety rates were 74.4%, and depression rates were 38.3%.<sup>21</sup> Another study conducted on 400 young individuals in Asia reported that individuals with TMD had high levels of stress, a high correlation with anxiety and depression.<sup>22</sup> In Ekici's study<sup>5</sup>, it was observed that 27.3% of participants experienced mild anxiety, 16.9% had moderate anxiety, and 19.3% suffered from severe anxiety. Regarding depression levels, mild depression was found in 26.4% of participants, moderate depression in 22.6%, and severe depression in 4.7%. In general, abnormal levels of anxiety (63.5%) and depression (53.6%) were observed in more than fifty percent of patients with chronic temporomandibular joint disorders. This study also thoroughly examined the relationship between depression and anxiety in TMD patients.

FAI is an index used to classify TMD.<sup>23</sup> It consists of a total of 10 items. hese items evaluate the limitations in mouth opening during jaw movements, painful mouth opening situations, crepitation and sound in the joint area, presence of teeth grinding accompanying bruxism, occlusion that may cause the disorder, and emotional changes.<sup>11</sup> First implemented in Portugal and later translated into English, FAI has been applied in Brazil, China, and Spain due to its high reliability and validity.<sup>8,10,24</sup>Arıkan et al.<sup>25</sup> translated the FAI, which has high validity and reliability, into Turkish and evaluated it. In their study, they reported that it is a consistent, reliable, and valid method for diagnosing TMD diseases. They verified the validity of this index by comparing it with other indexes.The Turkish translated version of FAI is a

simple, easy-to-understand, easy-to-use and applicable scale. It is recommended to use it together with all its substances to evaluate and understand the individual's experiences. Based on these results, it has been reported that the Turkish version of FAI can be used in clinical practice and research.<sup>25</sup> In our study, we used the Turkish version of the FAI due to its high reliability in diagnosing TMD.

The Beck Depression Inventory (BDI) and Beck Anxiety Inventory (BAI), developed by Beck et al., are reliable scales used to assess depression and anxiety levels in TMD patients.<sup>16</sup> These scales are used to measure the level of depression and anxiety and to determine the frequency and level of symptoms.<sup>12</sup> The validity of both methods has been confirmed by various studies in the literature<sup>26,27</sup>. A study by Maślak-Bereś et al.27 evaluated patients diagnosed with TMD according to TMD/RDC criteria with patients without TMD using the Beck Depression Inventory (BDI) and Stress Scale and found that the values were higher in individuals with TMD than in individuals without TMD. Çebi et al.28 conducted a study on 125 patients using the BDI and BAI in TMD patients and found higher scores on both scales in those with TMD. In our study, we utilized BDI and BAI due to their validity and reliability. The average BDI and BAI scores were found to be higher in individuals with TMD compared to those without.

The results discussed in the literature underscore the importance of assessing anxiety and depression in TMD patients. The diagnosis and treatment of TMD require a multidisciplinary approach to address all aspects of the problem. Therefore, using questionnaires that measure anxiety, depression, and other psychological factors in the initial evaluation of patients can assist dentists in identifying the need for treatments by other specialists. In this regard, further studies incorporating various tools are crucial for comparing results and determining the most suitable tool for clinical applications.<sup>29</sup>

One of the important aspects of this study is that it includes a large number of patients with and without TMJ disease. This study also increases the accuracy of the research. However, it is not possible to definitively determine whether anxiety and depression contribute to the onset of TMD or if TMDs lead to these conditions. Longitudinal cohort studies with specific research designs are needed to assess causality.

#### Conclusions

According to the results of our study, it can be said that there is a connection between anxiety and depression and TMD. It shows that patients with high levels of anxiety and depression are more likely to have TMD. This highlights the need to focus on the various factors that can cause TMD.

#### Acknowledgments

None

#### **Conflicts of Interest Statement**

The authors declare no conflicts of interest

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## Dental Implant Procedures Performed in Gazi University Faculty of Dentistry, Department of Oral and Maxillofacial Surgery for the last six years: Prevalence and Demographic Distribution

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Research Article	ABSTRACT
History	<b>Objectives:</b> The primary goal of dental implant treatment is to restore functional integrity, which is vital for oral health in patients with tooth loss, as well as to address aesthetic and psychological concerns. Demographic data from retrospective studies conducted in clinical settings with a high patient volume can be used as additional
Received: 02/08/2024	parameters when evaluating the success of dental implants.
Accepted: 05/12/2024	Material-Methods: In this study, the distribution of data by treatment years, patient gender and age, tooth regions and number of teeth (FDI system) were reviewed retrospectively for a total of 1892 patients among 7345 patients who underwent dental implant procedures at Gazi University Faculty of Dentistry, Department of Oral and Maxillofacial Surgery between January 2017 and June 2022. <b>Results:</b> It was found that female patients (51.7%) showed a higher prevalence than male patients for dental implant treatments, the mandible and maxilla posterior regions were the most common sites used for dental implants, and the age range of 50-59 years (5 <sup>th</sup> decade) had the highest number of dental implants (31.1%). Moreover monthly figures showed that the majority of the dental implant procedures were performed in 2019, prior to the COVID-19 pandemic.
	<b>Conclusion:</b> Based on these results, it was concluded that patient demographics are an important factor in planning dental implant treatment.
	Keywords: Dental Implant, Demographic Distribution.

## Gazi Üniversitesi Diş Hekimliği Fakültesi Ağız Diş ve Çene Cerrahisi Anabilim Dalı'nda Son Altı Yılda Gerçekleştirilen Dental İmplant Operasyonları; Sıklık ve Demografik Dağılım

**Research Article** ÖZ Amaç: Dental implant tedavilerinin temel amacı, diş kaybı yaşamış hastaların ağız sağlığında önemli olan Süreç fonksiyonel bütünlüğün sağlanmasının yanı sıra estetik ve psikolojik sorunların da ortadan kaldırılmasına yöneliktir. Hasta yoğunluğunun fazla olduğu kliniklerde yapılan restrospektif çalışmalarla birlikte sağlanacak Gelis: 02/08/2024 demografik bilgiler dental implantların başarı kriterlerinin değerlendirilmesinde kullanılacak diğer parametreler Kabul: 05/12/2024 için bir temel oluşturmaktadır. Gereç-Yöntemler: Bu çalışmada Ocak 2017-Haziran 2022 yılları arasında Gazi Üniversitesi Diş Hekimliği Fakültesi Ağız Diş ve Çene Cerrahisi Anabilim Dalı'nda implant tedavisi gören 7345 vaka arasından 1892 vakanın yıllara, cinsiyete, yaşa, diş bölgeleri ve numaralarına göre dağılımı retrospektif olarak incelenmiştir. Bulgular ve Sonuçlar: İmplant uygulaması yapılan 1892 hastadan kadın hastaların %51,7 ile erkek hastalardan daha fazla implant tedavisi gördükleri saptanmıştır. Bununla beraber, alt çene ve üst çene posterior bölgelerin en fazla implant uygulanan bölgeler olduğu saptanmıştır. En çok implant yapılan yaş aralığının ise %31.1 ile 50 ila 59 yaş aralığında (5.dekatta) olduğu gözlemlenmiştir. Aylık ortalamaya bakıldığında ise en fazla implantın 2019 License yılında pandemi öncesinde yapılmış olduğu da bulgular arasındadır. Bu sonuçlara dayanarak, hasta demografisinin dental implant tedavisinin planlanmasında önemli bir faktör olduğu sonucuna varılmıştır. <u>c</u> 0 S This work is licensed under Anahtar Kelimeler: Anksiyete, Depresyon, Temporomandibular Bozukluk. Creative Commons Attribution 4.0 International License \* shurmuzlu9090@gmail.com bttps://orcid.org/0000-0002-8750-1697 • One of the book of the boo https://orcid.org/0000-0003-2686-2955 How to Cite: Hurmuzlu M, Mollaoglu N. (2024) Dental Implant Procedures Performed in Gazi University Faculty of Dentistry, Department of Oral and

Maxillofacial Surgery for the last six years: Prevalence and Demographic Distribution , 27(4):240-244.

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#### Introduction

Tooth loss is an irreversible condition that is associated with adverse outcomes. Complete tooth loss or edentulism is defined as "the ultimate indicator of the disease burden for oral health".<sup>1</sup> Although the prevalence of tooth loss has decreased over the last decade, it is still a major concern worldwide, particularly among adults and the elderly population.<sup>2</sup>

Tooth loss, either partial or complete, has numerous effects on oral functions and, consequently, on oral health. From a physiological perspective, bone loss is an ongoing process that continues following tooth loss. Bone loss resulting from teeth loss is four times greater in the mandible than in the maxilla. Additionally, edentulism has been found significantly influence the alveolar bone resorption, which can lead to a reduction in alveolar crest height. This reduction affects facial height following tooth loss, thereby altering the facial appearance. Alveolar resorption can also cause significant changes in the soft tissue profile of both the lower lip and mandible.<sup>3-5</sup>

The number of teeth is regarded as the key determinant of oral functions and overall oral health status. A systematic review examining the relationship between oral function and the number of teeth has shown that having fewer than 20 teeth is associated with impaired chewing ability and performance.<sup>6</sup> Furthermore, it is also known that the thickness of the masseter muscle is reduced in edentulous patients, which in turn decreases bite force and directly affects chewing efficiency.<sup>7</sup>

In some cases, edentulism may be associated with functional and sensory deficiencies of the oral mucosa and musculature, and salivary glands. Edentulous population also shows reduced tissue regeneration, which can impair the function of the oral mucosa. A positive correlation between edentulism and oral mucosal disorders; aging; denture use; denture stomatitis; oral candidiasis and traumatic ulcers, has been previously reported.<sup>8,9</sup> Although most oral mucosal disorders affecting adults and older individuals are benign, some may become malignant.<sup>10</sup> Edentulism can also induce oral dyskinesia, which is characterized by abnormal, involuntary, stereotypic orofacial movements.<sup>11</sup>

The concept of osseontegration was first introduced by Brånemark nearly 50 years ago. Since then, dental implantology has evolved from experimental efforts to a successful and predictable treatment modality for replacing missing teeth.<sup>1</sup> Dental implants are of significant importance as they can restore natural function, protect the alveolar bone, provide additional support, preserve surrounding tissues, and enhance aesthetics. Consequently, dental implant treatment has become a routine clinical practice in modern dentistry. Several studies have reported that dental implants improve chewing function and enhance the quality of life in patients with complete or partial edentulism.<sup>2-5</sup> As a result, dental implants have become an excellent treatment option for the restoration of missing teeth. Despite their long-term survival, the rate of failure in dental implants increases over time. Patient-related factors such as overall health, age, gender, socioeconomic status, smoking habit, bone quality, oral hygiene, and untreated infections, along with implant-related factors including implant size, surface characteristics, location, and loading protocol, as well as clinician experience, have been reported to influence the success or failure of implants.<sup>5,6</sup>

While the implant design, surface characteristics, framework materials, and surgical protocols have been welldocumented in the literature, there is still a need for detailed demographic data and further information on the sites of implant placement. Therefore, the aim of this study was to retrospectively analyze patient demographic data and dental implant localizations to provide information on the location and number of dental implants by age and gender. For this purpose, demographic and implant-specific data of patients who underwent dental implant treatment at Gazi University Faculty of Dentistry, Department of Oral and Maxillofacial Surgery between January 2017 and June 2022 were analyzed.

#### **Materials and Methods**

Approval for the study was obtained from the Ethics Committee for Non-Interventional Clinical Trials of Gazi University Faculty of Dentistry (Date: 06/10/2022, No. E-21071282-050.99-519934). In this study, archived data of a total of 7345 patients who referred to Department of Oral and Maxillofacial Surgery at Gazi University Faculty of Dentistry between January 2017 and June 2022 were reviewed retrospectively, and 1892 patients were included in this study with total of 6486 dental implants placed over a 6-year period. Only patients age of 18 years or older having dental implant treatment were included to study. Demographic data (age and gender) and location of implants were reviewed.

All the patients included in this study were classified according to age and gender and statistical analysis was performed according to the position of the dental implants placed. To determine the statistical significance of the study, a 95% confidence interval was established using the single proportion confidence interval approach. Demographic parameters were statistically analyzed by comparing the mean values.

Analysis by age was done both by decades and by specific age ranges (Table 1):

- 1. 18-29 age group
- 2. 30-39 age group
- 3. 40-49 age group
- 4. 50-59 age group
- 5. 60-69 age group
- 6. 70-79 age group
- 7. 80-89 age group

#### *Table 1. Distribution of dental implants by age group.*

Variable (n=6486)	n	%
Age group		
18-29	310	4.8
30-39	594	9.2
40-49	1358	20.9
50-59	2019	31.1
60-69	1719	26.5
≥70	486	7.5

3.

4.

Implant location were divided into four regions as follows (Table 2):

- 1. Anterior maxillary region, including the teeth number 13 to 23.
- Anterior mandibular region, including the teeth 2. number 33 to 43.

Table 2. Distribution of	f all dental im	plants by l	location.
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Table 2. Distribution of all dental implants by location.		
Variable (n=6486)	n	%
Implant Location		
Anterior maxilla	942	14.5
Posterior maxilla	2388	36.9
Anterior mandible	774	11.9

Initially, an analysis was performed based on the total number of dental implant procedures carried out over the 6year period, including patient age, gender, tooth number, and dental region. Data analyses were then carried out for the following years:

1. Year 2017

**Posterior mandible** 

- 2. Year 2018
- Year 2019 3.
- Year 2020 4.
- 5. Year 2021
- 6. Year 2022 (first 6 months).

#### **Statistical Analysis**

Considering the error rate and standard deviation, the mean value of each parameter was evaluated quantitatively. Statistical comparative analyses for each parameter were conducted using the R software (Version 4.0.4 (2021-02-15) --"Lost Library Book" Copyright (C) 2021-The R Foundation for Statistical Computing). Data visualization was performed using MS Excel and R software packages. The normality of the data distribution was checked using the Shapiro-Wilk test. For variables that followed a normal distribution, a paired samples t-test was used to compare two groups. Two-way ANOVA was used to compare the means of two independent variables or factors from two or more populations. Unless stated otherwise, p<0.05 was considered statistically significant.

#### **Results**

This study included 1892 patients with mean age of 53. 21 ± 12.51 years (18 to 87 years). Total of 51.7% of the

2382 36.7 patients (n= 978) were female, and 48.3% (n=914) were

Posterior maxillary region, including the teeth

Posterior mandibular region, including the teeth

number 14 to 17 and 24 to 27.

number 34 to 37 and 44 to 47.

male. A total of 310 implants (4.8%) were placed in patients in the 18-29 age group, 594 implants (9.2%) in the 30-39 age group, 1358 implants (20.9%) in the 40-49 age group, 2019 implants (31.1%) in the 50-59 age group, 1719 implants (26.5%) in the 60-69 age group, and 486 implants (7.5%) in the  $\geq$ 70 age group (Table 1). Among a total of 6486 implants, 3282 implants (50.6%) were placed in female patients and 3204 implants (49.4%) in male patients.

It was found that 942 implants (14.5%) were placed in the anterior maxillary region, 2388 implants (36.9%) in the posterior maxillary region, 774 implants (11.9%) in the anterior mandibular region, and 2382 implants (36.7%) in the posterior mandibular region (Table 2).

When the distribution of implants according to tooth numbers was analyzed, it was found that the highest number of implants were placed in teeth numbered 26,16,24,14 in the maxilla, and in the mandible, in teeth numbered 36,46,33,43, respectively (Table 3).

There was no statistically significant relationship between gender and dental implant placement (p>0.05). In both genders, dental implants were most frequently placed in the region of teeth numbered 36, 46, 16 and 26, respectively. However, there was a significant association between gender and the year of implant placement ( $\chi^2$ =107,166; p=0.000) (Table 4). Moreover, statistically significant relationship was also observed between gender and age group ( $\chi^2$ =25,785; p=0.000) (Table 5).

Table 3. Distribution of dental implants according to FDI tooth numbering
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	n	%		n	%
Tooth Number (#)			Tooth Number		
11	126	1.9	31	39	0.6
12	157	2.4	32	99	1.5
13	186	2.9	33	256	3.9
14	333	5.1	34	220	3.4
15	265	4.2	35	234	3.6
16	398	6.1	36	469	7.2
17	175	2.7	37	270	4.2
21	131	2.0	41	37	0.6
22	153	2.4	42	94	1.4
23	198	3.1	43	256	3.9
24	345	5.3	44	231	3.6
25	252	3.9	45	219	3.4
26	414	6.4	46	469	7.2
27	202	3.1	47	258	4.0

#### Table 4. Distribution of patients by gender between 2017 and 2022.

		5			
Sex (Variable)	Female	Female (n=3282)		n= <b>3204)</b>	Statistical analysis*
	n	%	n	%	Probability
Year					
2017	609	18.6	652	20.3	χ²=107,166 <b>p=0.000</b>
2018	931	28.4	720	22.5	
2019	993	30.3	829	25.9	
2020	215	6.6	307	9.6	
2021	157	4.8	307	9.6	
2022	377	11.5	389	12.1	

\* Pearson's Chi-Square cross-tabulations were used to examine the relationships between two categorical variables.

*Table 5. Distribution of gender by age group.* 

Sex (Variable)	Female (n=3282)		Male (n=3204)		Statistical analysis*
	n	%	n	%	Probability
Age groups (years)					
18-29	160	4.9	150	4.7	
30-39	301	9.1	293	9.1	χ <sup>2</sup> =25,785 <b>p=0.000</b>
40-49	767	23.4	591	18.4	
50-59	973	29.6	1046	32.6	
60-69	843	25.7	876	27.3	
≥70	236	7.3	248	7.7	

\*Pearson's Chi-Square cross-tabulations were used to examine the relationships between two categorical variables.

#### Discussion

Osseointegrated dental implants represent a highly effective and predictable treatment method for tooth loss. Following the introduction of the "osseointegration" concept in the 1950s, which revolutionized the field of dentistry, dental implants have become a routine procedure. Since then, numerous scientific studies have been conducted on the use and effectiveness of dental implants.<sup>12-16</sup> Dental implants are a successful, highly effective, and predictable treatment modality used to address problems caused by tooth loss and to restore the function of missing teeth.<sup>17</sup> Despite the increase in dental implant procedures in Turkiye over the past decade, studies failed to provide quantitative data.<sup>18</sup> The challenges in collecting quantitative data may be attributed to the newly developing automation and hospital systems, as well as difficulties in tracking surgical procedures within the existing system in Turkiye.

The need for implant treatment has been correlated with tooth loss and advancing age.<sup>19</sup> Our study included 1892

patients with a mean age of 53.21 years (range, 18 to 87 years). More than half of the patients (51.7%) were female. The mean age of the male patients was significantly greater than that of female patients (Table 1). Similarly, in a retrospective study by Noack et al., which examined approximately 2000 dental implants placed between 1981 and 1997, the 50-59 age group received the greatest number of implants, in both males and females.<sup>20</sup> According to the results of this study, implant procedures were most frequently performed in the 5<sup>th</sup> decade (31%), followed by the 6<sup>th</sup> decade (26%) in both sexes (Table 3). Also, a significant association was observed between sex and age group (Table 5) (p<0,05). It was observed that 23.4% of the female patients were in the 40-49 age group, while 32.6% of the male patients were in the 50-59 age group. Furthermore, the patients in the 18-29, 30-39, and 40-49 age groups were predominantly female, whereas those in the 50-59, 60-69, and ≥70 age groups were predominantly male. Previous studies from Turkiye also reported quite variable mean ages.<sup>21</sup> In our study, the age range of patients who received implants was 18-87, with the most common age for receiving implant treatment being 57 years for women and 60 years for men. The slight age variations observed might explained by differences in sample size, methods used for age categorization, or socioeconomic status of the populations examined. A study at Yonsei University involving 1814 implants and 640 patients found that 49% of the patients who received implants were in their 40s and 50s<sup>22</sup> much younger than our study population. In agreement with our findings, published studies also show that dental implant treatment is more common among female patients compared to male patients (Table 4).<sup>23-25</sup> It was found that 993 (30.3%) females received dental implants in 2019, while 652 (20.3%) males received implants in 2017. Males mostly received dental implants in the years 2017, 2020, 2021, and 2022, while females mostly underwent implant treatment in the years 2018 and 2019 (Table 5) (p<0,05). The distribution of implants by year shows that the most implant procedures were performed in 2019, just before the COVID-19 pandemic started in Turkiye.

The distribution of implants by dental region was also evaluated in detail. In our patients, dental implants were most commonly placed in the posterior maxillary region, followed by the posterior mandibular region (Table 5). The lowest number of implants were placed in the anterior mandibular region. When examining the posterior maxillary region by individual dental regions, the tooth number 26 was the most frequent implant location (Table 3). There was no significant association between sex and implant regions (p>0,05). The distribution of implant regions was homogeneous irrespective of gender. Likewise, in a retrospective study, It was reported that the majority of dental implants were placed in the maxillar molar region and that the mandibular molar region was the second most common implant site (Table 2).<sup>26-29</sup> In contrast, Drago et al. reported in a retrospective study that dental implants were predominantly placed in the mandible (87.3%) and that a significant portion of implants (75.4%) were placed in the anterior region.<sup>28</sup>

A significant relationship was found between sex and the year of implant placement, with female patients receiving a higher number of dental implants than male patients (p<0.05). Our study revealed that 71 (31.6%) female patients underwent dental implant procedures in 2019, while in 2017, 2020, and 2021, the majority of patients receiving implants were male, and in 2018, 2019, and 2022, the majority were female (Table 4).

In this study, implant treatments were more commonly applied to the molar teeth compared to other teeth. Regarding the tooth numbers of the teeth undergoing implant treatment, the maxillary and mandibular first molars (teeth #16, 26, 36 and 46) were the most commonly treated teeth. The least frequently treated teeth were the teeth # 31, 32, 41, and 42 (Table 3). Similar findings were reported in a 2018 study from Turkiye involving 1000 patients.<sup>30</sup> Another retrospective study by Bornstein et al. reported that implants were most frequently placed in the posterior mandible region, with

the most common implant site being the tooth #36, and the least common implant site being the anterior mandibular region.<sup>31</sup> The distribution of dental implants by implant location observed in our study is consistent with Bornstein et al.'s findings.<sup>31</sup> In addition, this study emphasizes that the prevalence of dental implant surgery is higher in the 50-59 age group in both genders.

#### Conclusions

This study revealed that the demand for dental implants increased significantly in certain age groups for both genders, as well as the number of dental implant placements being higher in the posterior mandibular region. Based on these results, it was concluded that patient demographics are an important factor in planning dental implant treatment. However, further research is needed in different geographic and demographic settings to confirm these results.

#### **Conflicts of Interest Statement**

In this study, there is no conflicts of interest.

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### Investigation of Antifungal Effects of Different Remineralization Agents on Salivary Candida amount in children with Early Childhood Caries

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\*Corresponding author **Research Article** ABSTRACT Objectives: In recent years, Candida has been linked to dental caries in children, with Candida albicans being particularly associated with the development of early childhood caries (ECC). In children with ECC, Candida History species, along with mutans streptococci and lactobacilli, are commonly present in the oral cavity. This study Received: 25/11/2024 aimed to evaluate the effects of different remineralization agents on Candida counts in children diagnosed with Severe Early Childhood Caries (S-ECC). Accepted: 13/12/2024 Material and Methods: Fifty-four healthy children, aged 3 to 5 years, diagnosed with S-ECC, were assessed, and 21 Candida-positive children were selected for inclusion in the study. The children were randomly divided into three groups: a control group using 500 ppm NaF toothpaste, a 10% Casein Phosphopeptide-Amorphous Calcium Phosphate (CPP-ACP) group, and a Calsium Glycerophosphate (CaGP) + 12% Xylitol group. Oral hygiene instructions were provided, and participants were advised to apply the remineralization agents three times daily for 3-5 minutes over a 2-week period. Unstimulated saliva samples were collected and cultured to quantify Candida counts. The number of Candida colonies was measured at baseline, the second week, and at the first and fourth months. Data analysis was performed using SPSS Statistics software (version 23), with statistical significance set at a p-value of less than 0.05. Results: A significant difference was observed in Candida counts between baseline and the first and fourth months in the CaGP + 12% Xylitol group (p < 0.05). The CaGP group exhibited a notable decrease in salivary Candida levels (P = 0.028) after 4 months of treatment. However, while reductions in Candida counts were seen in the NaF toothpaste and CPP-ACP groups, these changes did not reach statistical significance (p>0.05). Conclusion: These findings indicate that remineralization agents may reduce Candida counts, suggesting their potential effectiveness in caries management. Keywords: Early Childhood Caries, CPP-ACP, CaGP, Xylitol, Candida Albicans. Erken Çocukluk Çağı Çürüğü Olan Çocuklarda Farklı Remineralizasyon Ajanlarının Tükürükteki Candida Miktarı Üzerine Antifungal Etkilerinin İncelenmesi

Research Article	02				
	Amaç: Son yıllarda Candida türlerinin çocuklarda diş çürüğü ile ilişkili olduğu bildirilmiştir. <i>Candida albicans'</i> ın				
Süreç	Erken Çocukluk Çağı Çürüğü (EÇÇ) başlangıcındaki rolü iyi bilinmektedir. Mutans streptokokları ve				
	laktobasillerin yanı sıra, Candida türleri de EÇÇ' li çocukların ağız boşluğunda sıklıkla bulunmaktadır. Bu				
Geliş: 09/04/2024	çalışmanın amacı, Şiddetli Erken Çocukluk Çağı Çürüğü (Ş-EÇÇ) olan çocuklarda farklı remineralizasyon				
Kabul: 16/10/2024	ajanlarının tükürükteki Candida miktarına etkisini karşılaştırmaktır.				
	Gereç ve Yöntemler: Bu araştırmada, yaşları 3-5 arasında değişen, Ş-EÇÇ tanısı konmuş 54 sağlıklı çocuk				
	değerlendirildi ve Candida-pozitif olan 21 çocuk çalışmaya dahil edildi. Çocuklar rastgele 3 gruba ayrıldı: 500				
	ppm sodyum fluorid (NaF) diş macunu grubu (kontrol), %10 kazein fosfopeptid–amorf kalsiyum fosfat (CPP-				
	ACP) grubu ve kalsiyum gliserofosfat (CaGP)+ %12 ksilitol grubu. Tüm çocuklara ağız hijyeni eğitimi verildi ve				
	remineralizasyon ajanlarını iki hafta boyunca günde üç kez, 3-5 dakika süreyle kullanmaları istendi. Candida				
	kolonilerinin kültürü için uyarılmamış tükürük örnekleri toplandı. Tükürükteki Kandida değerlendirilmesi,				
	başlangıç, 2. hafta, 1. ay ve 4. ayda toplanan tükürüklerde gerçekleştirildi. Tüm analizler SPSS Statistics Version				
	23 ile yapıldı ve anlamlılık düzeyi $\rho$ < 0,05 olarak kabul edildi.				
License	Bulgular: CaGP + %12 Ksilitol grubunda, başlangıç ile 1. ay ve 4. ay arasındaki Candida miktarı açısından				
	istatistiksel olarak anlamlı bir azalma bulundu ( $p < 0,05$ ). Ayrıca, CaGP grubu, 4 aylık uygulama sonrasında				
	tükürükteki Candida miktarında anlamlı bir azalma gösterdi ( $p = 0,028$ ). Ancak, NaF diş macunu ve CPP-ACP				
	gruplarındaki Candida miktarındaki azalmalar, istatistiksel olarak anlamlı düzeyde değildi ( $\rho > 0.05$ ).				
This work is licensed under	Sonuc: Sonuclar, remineralizasyon ajanlarının Candida miktarını azaltabileceğini ve bu nedenle diş çürüğü				
Creative Commons Attribution 4.0	yönetiminde etkili olabileceğini göstermektedir.				
International License	Anahtar Kelimeler: Erken Çocukluk Çağı Çürüğü, CPP-ACP, CaGP, Ksilitol, Candida Albicans.				
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How to Cite: Eren F, Guven Ozsahin C, Topcuoglu N, Kargul B. (2024) Investigation of Antifungal Effects of Different Remineralization Agents on Salivary Candida amount in children with Early Childhood Caries, Cumhuriyet Dental Journal, 27(4):245-251.
#### Introduction

Early childhood caries (ECC) is defined by the occurrence of at least one decayed lesion (either cavitated or non-cavitated), a tooth lost due to caries, or a restored tooth surface in any primary tooth of a child aged 71 months or younger. For children under the age of 3, the detection of caries on smooth surfaces signifies severe early childhood caries (S-ECC).<sup>1</sup> ECC remains the most prevalent chronic disease in children, with nearly 1.8 billion new cases reported globally each year.<sup>2</sup> S-ECC develops at an earlier age than ECC, often leading to more severe health complications and typically necessitating extensive dental treatment under general anesthesia, which may include several tooth extractions, restorations, and crowns.<sup>3</sup> Although not life-threatening, disease significantly affects individuals and the communities by diminishing quality of life due to pain and functional impairment, and it negatively impacts a child's growth, body weight, and overall development.<sup>4</sup> Therefore, S-ECC represents a significant challenge to public health, necessitating a deeper understanding of its etiopathogenesis and the development of more effective treatment strategies to reduce recurrence and associated costs.<sup>5</sup>

In germ-free animal studies, it was observed that caries did not occur despite a high carbonhydrate diet, and the primary effect of oral microorganism in caries etiology has been proven.<sup>6</sup> The microbial origin of S-ECC is often linked to a polybacterial infection of the teeth. However, microbiological studies also indicate that fungal organisms play a role in this pediatric oral condition. Candida species were found in significantly higher levels in the oral cavities of children with S-ECC compared to caries-free children, with their presence correlating positively with both the severity of caries and the colonization of Streptococcus mutans. In the studies, researchers stated that C. albicans is highly acidogenic and has the ability to dissolve hydroxyapatite crystals of enamel more than S.mutans.<sup>7,8</sup> These factors show that C.albicans contributes to the severity and aggresive nature of the S-ECC. Research on chemotherapeutic approaches to reduce or prevent Candida levels in ECC is limited, emphasizing the need to assess the effectiveness of various agents in decreasing caries incidence in children.9

A common approach to minimizing the risk of ECC involves brushing teeth twice daily using fluoride toothpaste. In addition to fluoride, chlorhexidine, silver, povidone iodine, xylitol, probiotics and caseine containing preparations (Casein Phosphopeptide-Amorphous Calcium Phosphate (CPP/ACP) and Casein Phosphopeptide-Amorphous Calcium Fluoride Phosphate (CPP/ACFP)) are also used. In a meta-analysis study, it has been reported that, these antibacterial and remineralization agents effect is low in the prevention of ECC, and there is not enough evidence about the effectiveness of the agents.<sup>10</sup>

There is limited research evaluating the antimicrobial effects of CPP-ACP on primary teeth.<sup>11-14</sup> However, no research has compared the antifungal effect of CPP- ACP,

xylitol containing CaGP paste and low-fluoride containing tooth paste in children with ECC. Therefore the aim of this study was to compare three approaches: (1) brushing twice daily with low-fluoride toothpaste; (2) brushing twice daily combined with the application of 10% CPP-ACP paste three times daily; and (3) brushing twice daily combined with the application of a paste containing CaGP, MgCl2, and 12% xylitol three times daily, to evaluate their effects on *Candida* counts in children with S-ECC.

#### **Materials and Methods**

#### **Ethical considerations**

The study protocol received approval from two authorities: (1) the Clinical Research Ethics Committee of Marmara University, School of Dentistry (approval number 2019-283) and (2) the Turkish Medicines and Medical Devices Agency, Republic of Turkey Ministry of Health (approval number 20-AKD-167). The research adhered to the principles outlined in the Declaration of Helsinki (1964). Written informed consent was obtained from the parents or guardians of all participants.

#### Sample size calculation

The sample size was determined using G\*Power 3.1 software, with the significance level set at 0.05 and a statistical power of 0.8. The calculation indicated a total of 18 participants (at least 6 per group) was required. To account for an anticipated dropout rate of 15%, the recruitment target was set at a minimum of 21 participants, with 7 individuals in each group.

#### Study design

This randomized clinical trial adhered to the Consolidated Standards of Reporting Trials (CONSORT) guidelines.<sup>15</sup> The study was carried out with systemically healthy, cooperative children aged 3-5 years diagnosed with S-ECC, who were patients at the Department of Pediatric Dentistry, Marmara University, Istanbul, Turkey. All participants met the American Academy of Pediatric Dentistry's criteria for the definition of S-ECC. Exclusion criteria included the presence of a systemic disease, recent antibiotic or anti-inflammatory use within the previous month, or receiving specialized dental care such as orthodontic treatment, the use of space maintainers, or other dental appliances. After being informed about the study, the parents of 54 children provided consent, and these children were included in the trial. The CONSORT flow diagram outlining the study process is presented in Figure 1.

Sociodemographic and oral hygiene behavior characteristics were recorded. All children were examined by a single examiner (CG). Before the study commenced, the examiner underwent training and calibration, and the initial patients enrolled were assessed through a peer-reviewed consensus process. Caries experience in children was evaluated using visible light, a standard dental mirror, and a probe designed for the Community Periodontal Index.



#### Randomization and blinding

Randomization and blinding procedures were conducted using sealed, opaque envelopes, each individually numbered and limited to single selections. These envelopes contained details about group allocation and identification numbers. Participants were asked to draw a sealed envelope at random from the collection of concealed envelopes. The maintenance of allocation concealment was independently overseen by FE. The trial intervention remained blinded to patients, the microbiologist, and the data analyst. The microbiologist was not involved in the investigative team conducting the trial intervention, did not participate in salivary sample collection, and had no access to randomization sheets. Salivary samples were uniformly collected by a single investigator (CG), who was unaware of the patients' group allocations.

#### Saliva collection

Saliva samples were collected from 54 children at 9:00 to 11:00 am to reduce potential circadian variations. Prior to collection, participants were asked to swallow to eliminate any saliva that had accumulated in the mouth. Unstimulated saliva samples were collected at baseline from 54 children using a dental saliva ejector. The collected samples were forwarded to the laboratory at the Department of Basic Medical Sciences, Faculty of Dentistry, Istanbul University, for *Candida* culture testing. The study population was constituted by children with a positive Candida value ( $\geq$ 100 cfu/ml) as determined by the results of the Candida culture.

#### Intervention

Baseline saliva samples were obtained from all participants, and the 21 children were randomly divided

into three groups, each comprising 7 participants. Participants received (Group 1) tooth paste including 500 ppm NaF, (Colgate Oral Pharmaceuticals, New York, NY) (Control Group); (Group 2) 500 ppm NaF + 10% CPP-ACP containing paste (GC Tooth Mouse, Europe, Tokyo, Japan) and (Group 3) 500 ppm NaF + CaGP and 12% Xylitol containing paste (R.O.C.S Medical Minerals gel, DRC Group, Moscow, Russia). Application procedure for remineralization agents were 2 weeks, 3 times per day; 1) after brushing the teeth in the morning after breakfast, 2) at any time of the day and 3) after brushing the teeth before going to bed at night as chickpea size and advised to applied to all tooth surfaces under parent's supervision. Parents were advised to wait 60 minutes between tooth brushing and gel application and to avoid eating or drinking any solid or liquid items for 40-50 minutes after applying the gel. All participants received the same toothpaste as the control group and hygiene education. The participants were oral continuously monitored to ensure adherence to the prescribed regimen. Throughout the intervention phase (2 weeks) and follow-up periods, the subjects were advised to continue their regular oral hygiene routine during each check-up session. Saliva samples were obtained at baseline, followed by collections after 2 weeks, 1 month, and 4 months, and the salivary Candida counts were measured. Each sample tube was marked with a code that included the participant's identification number and the corresponding control session of the study.

#### Salivary Candida counts

Two milliliters of unstimulated saliva were utilized for microbiological analysis. Sabouraud Dextrose agar (Merck KGaA, Darmstadt, Germany) medium was utilized for *Candida* isolation in the study. A volume of 0.01 ml from collected saliva samples was inoculated by spreading onto the agar surface. Subsequently, incubation was carried out in an aerobically controlled environment at 37°C for 48. At the end of the appropriate time, the typical colonies seen in the medium were counted and the colony count per milliliter (cfu/ml) was calculated. For *candida*, counts less than <10<sup>3</sup> cfu/ml were considered as low level, between >10<sup>3</sup> - <10<sup>4</sup> cfu/ml as moderate level, and >10<sup>4</sup> cfu/ml as

high level. The count below the detection limit of 10 cfu/ml with no detectable growth was accepted as '0'.  $^{\rm 16}$ 

#### **Statistical Analysis**

Data analysis was conducted using SPSS software, version 23 (SPSS Inc., Chicago, IL, USA). The results were presented as frequencies (%), as well as mean  $\pm$  standard deviation (SD) with descriptive statistics presented as mean  $\pm$  SD. The normality of the data distribution was assessed using the Kolmogorov-Smirnov test. Values that did not have normal distribution and were followed at 4 different times were analyzed with the Friedman's two-way ANOVA test. To identify which specific pairs of groups differed significantly, All Pairwise tests were used. The level of statistical significance (*p*-value) was set at <0.05.

#### Results

This study included 54 children diagnosed with S-ECC who consented to participate in the research. In forming the research group, initial Candida counts were considered, and to assess the significance of changes in Candida counts, 21 children with counts greater than 100 cfu/ml were included. The mean age of these 21 children was 4.08  $\pm$  0.68 years, with 14 boys (66.7%) and 7 girls (33%).

Table 1 shows the the changes in low, moderate and high level of *Candida* at baseline, after 2 weeks, 1 month and 4 month of intervention. When comparing baseline and 4th-month results, a reduction in the moderate level of *Candida* count was observed across all groups.

The changes in the median *Candida* levels of the children in the study group according to the follow-up sessions were evaluated for each group. No statistically significant difference was observed between baseline and follow-up values in the 500 ppm NaF and 10% CPP-ACP groups (p > 0.05). However, a statistically significant difference was found between baseline and follow-up values in the CaGP + 12% Xylitol group (p = 0.012) (Table 2).

The difference between the baseline mean values and the first-month mean values in the CaGP + 12% Xylitol group was statistically significant (p = 0.018), as was the difference between the baseline mean values and the fourth-month mean values (p = 0.028) (Table 3).

*Table 1.* The evaluation of changes in Candida count between the groups.

Candida	500ppm NaF (Control)			10% CPP-ACP n=7		CaGP + 12% Xylitol n=7			
Baseline	Low <10 <sup>3</sup> cfu/ml n(%)	n=7 Moderate >10 <sup>3</sup> - <10 <sup>4</sup> cfu/ml n(%) 3(42.9%)	High >10 <sup>4</sup> cfu/ml n(%)	Low <10 <sup>3</sup> cfu/ml n(%)	Moderate >10 <sup>3</sup> - <10 <sup>4</sup> cfu/ml n(%)	High >10 <sup>4</sup> cfu/ml n(%)	Low <10 <sup>3</sup> cfu/ml n(%)	Moderate >10 <sup>3</sup> - <10 <sup>4</sup> cfu/ml n(%)	High >10 <sup>4</sup> cfu/ml n(%)
2nd-week	3(42.9%) 4(57.1%)	3(42.9%)	1(14.3%) 0(0%)	2(28.6%) 4(57.1%)	5(71.4%) 3(42.9%)	0(0) 0(%0)	4(57.1%) 6(85.7%)	2(28.6%) 1(14.3%)	1(14.3%) 0(0%)
1st-month	5(71.4%)	1(14.3%)	1(14.3%)	4(57.1%)	3(42.9%)	0(0%)	5(71.4%)	2(28.6%)	0(0%)
4th-month	4(66.7%)	0(0%)	2(33.3%)	5(83.3%)	1(16.7%)	0(%0)	5(83.3%)	1(16.7%)	0(0%)

Candida counts (lo	ogCFU/ml)	Median [%25-%75]	Р
	Baseline	1800 [700 -9200]	
500ppm NaF	2nd-week	100 [0 -3800]	0.169
(Control)	1st-month	200 [100 -2000]	0.168
	4th-month	800 [0 -19000]	
	Baseline	3300 [300 -4800]	
	2nd-week	700 [200 -1200]	0.000
10% CPP-ACP	1st-month	100 [0 -1200]	0.093
	4th-month	600 [100 -1100]	
	Baseline	800 [500 -9000]	
	2nd-week	200 [0 -800]	0.042*
CaGP+12% Xylitol	1st-month	200 [0 -1900]	0.012*
	4th-month	400 [100 -700]	

*Table 2.* Distribution of changes in Candida counts (median [%25-%75]) at the baseline, 2nd week, 1st and 4th month according to the groups.

Friedman's two-way ANOVA test, \*p<0,05 statistical significance.

Table 3. Statistical chang	e in Candida count in the CaGP + 1	12% Xylitol aroup	according to control sessions.

Candida	CaGP+12% Xylitol	
(CFU/	ml)	p
	2nd-week	0.128
Baseline	1st-month	0.018*
	4th-month	0.028*
and weak	1st-month	0.833
2nd-week	4th-month	0.528
1st-month	4th-month	0.686

All pairwise test, \*p<0.05 statistical significance.

#### Discussion

S-ECC is a widespread public health issue that necessitates intricate treatment procedures and results in significant costs. It has been suggested that the prevention of S-ECC is positively correlated with the understanding of etiopathogenesis involving cariogenic bacteria.<sup>7</sup> However, it has long been recognized that the microbiota of caries-associated biofilms consists of a diverse range of microorganisms, including Fusobacterium, Bifidobacterium, Actinomyces, Prevotella, Scardovia, Veillonella, Atopobium, and Candida species.<sup>17</sup>

Raja et al., in their study of healthy children in the mixed dentition period, showed that dental caries was associated with Candida carriage.<sup>18</sup> Given their ability to colonize tooth surfaces, infiltrate dentinal tubules, contribute to microbial biofilm formation, and generate substantial amounts of acid that lead to enamel demineralization and hydroxyapatite crystal dissolution, Candida species are hypothesized to play a significant role in the advancement of caries lesions.  $^{19,20}\ \mbox{In their}$ study, de Carvalho et al. demonstrated a significant association between Candida albicans and the occurrence of ECC.<sup>21</sup> Akdeniz et al., investigated Candida carriage in children with and without caries in their study and stated that environmental factors such as oral hygiene status, high concentration of dietary sugar and the presence of widespread caries may be an important factor for the high prevalence of Candida.<sup>22</sup> On the other hand, certain clinical studies have found no notable differences in oral Candida prevalence between cariesfree and caries-active populations, nor a clear positive correlation between the presence of Candida albicans and caries risk in children.<sup>7</sup> Due to the controversial results of studies on *Candida* and ECC, this study evaluated the antifungal effect of two remineralization agents and low-fluoride toothpaste on *Candida* known to be causative in S-ECC.

Two systematic reviews have demonstrated that numerous studies have investigated either a single or combination of various agents (e.g., chlorhexidine, iodine, fluoride, silver compounds, xylitol, CPP-ACP, probiotics and triclosan) combined with different application methods (e.g., m outh rinses, gels, varnishes, cleaning wipes, restorative materials) for the prevention of ECC.<sup>23,24</sup> In our study, remineralizing gels containing CaGP+12% Xylitol or 10% CPP-ACP, and a toothpaste containing 500 ppm NaF were used. A literature review revealed studies investigating the remineralization effects of the agent containing 10% CPP-ACP<sup>25</sup> but it was found that research on its antimicrobial effects was limited.<sup>14</sup> However, our study is the first to investigate the antifungal effect of the 10% CPP-ACP and 12% Xylitol + CaGP-containing remineralizing agents on ECC.

The effectiveness of fluoride-containing toothpaste in preventing dental caries among children and adolescents has been consistently confirmed by numerous systematic reviews.<sup>26,27</sup> The anticariogenic effects of low fluoride levels in toothpaste are unclear; however, most studies have shown that toothpastes containing less than 500 ppm fluoride are less effective compared to those containing 1000 ppm fluoride.<sup>13</sup> Zaze *et al.* investigated the effects of varying concentrations of calcium glycerophosphate (CaGP) in low-fluoride toothpastes on enamel demineralization. Their findings indicated that a

low-fluoride toothpaste (500  $\mu$ g F/g) supplemented with 0.25% CaGP could achieve a level of efficacy comparable to that of a higher-fluoride toothpaste (1,100  $\mu$ g F/g). This suggests that adding CaGP to low-fluoride toothpastes may effectively enhance their ability to prevent enamel demineralization, potentially offering a valuable alternative in oral care formulations.<sup>28</sup> Cavazana et al. examined the effect of CaGP, with or without fluoride, on the pH of dual-species biofilms consisting of Streptococcus mutans and Candida albicans. They concluded that CaGP had an impact on the dual-species biofilm formed by S. mutans and C. albicans.<sup>29</sup> In this study, the changes in Candida count during follow-up sessions after regular use of toothpaste containing 500 ppm fluoride were also evaluated. A decrease in Candida count was observed between sessions, but this reduction was not statistically significant. In both developed and developing countries, regular use of fluoride toothpaste has been identified as a key factor in reducing dental caries.<sup>30</sup> However, discussions on fluoride use have been increasing in recent years, and the emerging need has led to an increase in studies focused on developing alternative agents that are as effective as fluoride.<sup>31</sup>

Research suggests that the anticariogenic properties of CPP-ACP result from its combined effects on remineralization and bacterial displacement.<sup>32</sup> The literature is conflicting on CPP-ACP's antibacterial properties. In their in vitro study, Erdem et al. showed that CPP-ACP reduced S. mutans biofilm formation, while Rahiotis et al. found that the presence of CPP-ACP delayed bacterial biofilm formation.<sup>11,12</sup> In an in vivo study, Schüpbach et al. demonstrated that CPP reduced the ability of mutans streptococci (MS) to adhere to treated enamel surfaces.<sup>33</sup> In a study by Plonka et al., it was shown that the daily application of a CPP-ACPcontaining remineralizing agent from the eruption of the first tooth until the child reached approximately 24 months was more effective than chlorhexidine in reducing the presence of MS.<sup>13</sup> In contrast, Grychtol et al. reported that a CPP-ACP-containing preparation had no significant effect on the initial bacterial colonization of enamel and dentin, and thus could not be recommended for biofilm prevention and management.<sup>34</sup> While the antibiofilm potential of S. mutans is established, the antibacterial properties of casein and calcium phosphate have not yet been confirmed.

Al-Batayneh et al. evaluated the combined effects of fluoride, CPP-ACP, and both agents on *S. mutans* in children at high risk of caries at the 3rd and 6th months. They found a significant reduction in the number of *S. mutans* positive children from baseline to the 3rd month in all groups except the CPP-ACP group, but no significant difference between the groups throughout the study period. However, at the 6-month follow-up, the CPP-ACP group exhibited the most significant reduction in *S. mutans* levels. These findings were associated with the better performance of CPP-ACP over time, with a significant difference reported in the fluoride group at the 3-month mark and a similar trend observed in the combination group. It was reported that the similarity in the combination group was due to fluoride being applied before CPP-ACP.<sup>14</sup> In this study, the effect of a remineralising agent containing 10% CPP-ACP on *Candida* was examined after being used three times a day for two weeks. A decrease in *Candida* count between sessions was observed, with a slight increase at the 4-month follow-up session, which did not reach baseline levels; however, these changes between sessions were not statistically significant. A literature review revealed no studies evaluating the antimicrobial efficacy of CPP-ACP against *Candida*, making it impossible to compare the results. Further studies are required to assess the antibacterial effects of CPP-ACP.

Antibacterial activity in materials used for the remineralization of dental hard tissues is a valuable property for preventing and reversing dental caries. To achieve this, various antibacterial agents can be incorporated into remineralization agents.<sup>35</sup> In our study, two different remineralization agents containing CaGP+12% xylitol and 10% CPP-ACP were used for this purpose.

Since the 1960s, xylitol has been approved by the US. Food and Drug Administration for use in foods, and research has demonstrated its effectiveness as an anticaries agent.<sup>36</sup> Studies on xylitol have included various applications such as tablets, gum, syrup, and cleaning wipes in children ranging from 6 months to 5 years old.<sup>37-39</sup> Li and Tanner, in their meta-analysis, demonstrated that xylitol-containing agents generally led to a significant reduction in mutans streptococci (MS) colonization in young children.<sup>23</sup> Milgrom et al found that applying xylitol syrup (8g/day) twice a day from the appearance of the first primary tooth could prevent up to 70% of dental caries.<sup>37</sup> In our study, the effects of a remineralization agent containing 12% xylitol were evaluated after its use three times a day for two weeks. A statistically significant change in Candida count was observed between sessions. Regular use of the 12% xylitol-containing agent led to a significant reduction in Candida count compared to baseline. The changes between baseline and the 1-month follow-up session, as well as between baseline and the 4-month follow-up session, were statistically significant. In light of these findings, it was found that the effect of the mentioned remineralization agent on Candida continued for up to 4 months following two weeks of use. The use of this remineralization agent every 4 months for two weeks in preschool children at high caries risk appears to be a reasonable approach.

To summarize, the results of this study, which investigated the antifungal effects of different remineralization agents on children with S-ECC, indicate that the effects of CaGP + 12% Xylitol containing gel on *Candida* are significantly acceptable throughout all sessions. This antimicrobial efficacy is thought to be due to the xylitol content in the used agent. Additionally, no studies have been found in the literature investigating the effect of this remineralization agent on *Candida* in vivo. Therefore, our study is significant as it is the first to address this issue. We believe that further research involving larger sample sizes, different application and examination methods, and longer follow-up periods is needed to obtain more definitive data on the effects of the agent containing CaGP + 12% Xylitol on other bacterias. Considering regular oral hygiene, the use of CaGP + 12% Xylitol, which is easy to use, effectively acceptable, and economically advantageous compared to other remineralization agents, appears appropriate to be included in preventive application protocols for preschool children at high risk of caries.

#### Conclusions

These results suggest that remineralization agents can reduce the role of Candida in the pathogenesis of caries by reducing the count of Candida, thus it can be effective in caries management.

#### Acknowledgments

This study received financial support from the Marmara University Scientific Research Projects (BAP) Unit under project number SAG-C-DUP-080519-0177. The part of this manuscript was presented as oral poster presentation at the 70<sup>th</sup> ORCA and European Federation for Conservative Dentistry Congress held in July 2023 at Egmond aan Zee, Netherlands.

#### **Conflict of Interest**

The authors have no conflicts of interest to declare.

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# Treatment of Gingival Recession in the Mandibular Anterior Region with an Amnion Chorion Membrane Using the Papilla Access Tunnel Technique – 2 Clinical Case Reports

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Case Report	ABSTRACT
History	The tunnel technique for root coverage and soft tissue augmentation in the mandibular anterior area can pose several challenges, including shallow recession, thin soft tissue, pronounced roots, and alveolar undercuts. These
Received: 20/02/2024 Accepted: 06/12/2024	factors can complicate the surgical procedure and lead to poor outcomes. In this context, a papilla access technique has been developed to facilitate the preparation of the tunnel site and the placement of grafts under challenging anatomical conditions. This surgical access method is particularly useful in patients with minimal recession of the mandibular central incisors, where extending the tunnel to include the lateral incisors and canines is necessary for augmentation purposes. This approach offers improved surgical maneuverability for both site preparation and graft placement while minimizing the risk of perforation or trauma to delicate tissues. Overall, the papilla access technique represents a useful addition to the armamentarium of periodontal surgeons for root coverage in the mandibular
License	anterior region and soft tissue augmentation procedures.
Creative Commons Attribution 4.0 International License	Keywords: Papilla access tunnel technique, Amnion membrane, Chorion membrane, Root coverage, Gingival recession.
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How to Cite: Roshni KB, Shetty NJ. (2024) Treatment of Gingival Recession in the Mandibular Anterior Region with an Amnion Chorion Membrane Using the Papilla Access Tunnel Technique – 2 Clinical Case Reports, Cumhuriyet Dental Journal, 27(4): 252-259.

#### Introduction

Gingival recession, also known as soft tissue recession, is defined as the displacement of the gingival margin apical to the cemento-enamel junction (CEJ) of a tooth or the platform of a dental implant. This condition has been associated with compromised aesthetics, noncarious cervical lesions (NCCLs) and dentinal hypersensitivity.<sup>1,2</sup> Clinical challenges arise, particularly in cases of recession defects located in the mandibular area, due to anatomical considerations such as vestibular depth, tooth position, muscle pull, frenulum, and the thickness of hard and soft tissues.

In order to address these challenges and enhance postsurgical outcomes for such defects, more complex treatments, such as soft tissue grafting, are considered the "gold standard".<sup>3</sup> A newer surgical technique, the papilla access tunnel technique, has emerged as a promising option for treating mandibular recessions.<sup>4</sup> Since its introduction by Allen in 1994, the tunnel technique has undergone several modifications <sup>5</sup> and has demonstrated more favorable results than alternate methods in terms of aesthetic outcomes and patient experience.<sup>6,7</sup> The advantages of this tunneling approach include an incision-free design and preservation of papilla integrity. Papillary access minimizes the risk of injury or perforation of thin tissue while improving surgical access for site preparation and graft insertion. However, in areas where

recession is shallow, the intrasulcular approach might provide challenges, especially in the mandibular anterior region where roots are narrow and sulcus dimensions are small, rendering intrasulcular access insufficient.<sup>8</sup>

Gingival tissue shares similarities with fetal tissues in terms of structure, fibroblast phenotype, and scarless healing. Recently, placenta-derived tissues, such as the amnion and chorion, have gained popularity as guided tissue regeneration membranes in dentistry. These tissues contain stem cell reserves; various growth factors; and proteins, such as fibronectin, laminin, proteoglycans, and glycosaminoglycans, that accelerate healing and regeneration. The chorion is particularly rich in collagen proteins.<sup>9</sup> Placental allografts offer advantages due to their immune-privileged composition, antibacterial and antimicrobial properties, and provision of a protein-enriched matrix facilitating cell migration<sup>10</sup>. Both amnion and chorion membranes are now utilized in root coverage procedures. In particular, the amniotic membrane serves as a cost-effective, allogenic substitute for subepithelial connective tissue grafts for achieving root coverage. Based on the biological properties of placental membranes and their potential, it is theorized that they could be effective at managing gingival recession.<sup>10</sup>

#### **Case Report**

In the first case, a male patient aged 30 years reported sensitivity in the lower mandibular anterior teeth. Clinical examination revealed Miller's class II gingival recession affecting teeth #31 and #41. The recession depth was measured at 3 mm for tooth #31 and 2 mm for tooth #41, with

a recession width of 2 mm. The patient exhibited a thin biotype (Figure 1). In the second case, a female patient aged 42 years presented with sensitivity in teeth #31 and #41. Clinical examination indicated Miller's class I gingival recession involving teeth #31 and #41, with a recession depth and width of 2 mm. Similar to the first patient, the patient also displayed a thin biotype (Figure 2).



Figure 1: Baseline - Miller's class II gingival recession involving 31 and 41 with recession depth 3mm irt 31 and 2mm irt 41 and a recession width of 2mm.



Figure 2: Baseline - Miller's class I gingival recession involving 31 and 41 with a recession depth and width of 2 mm.

With the patient's signed informed consent, the treatment plan was discussed. Due to the presence of a thin biotype of the gingiva in both patients, the Papilla access tunnel technique with the amnion and chorion membrane was planned for both patients. Following local anesthesia administration (2% lidocaine with 1:80000 epinephrine), the papilla access tunnel was incised between the lateral incisors and canines on both the right and left sides. This incision separates the facial layer of the papilla on either side of the midline, effectively detaching the papilla from the tooth surface (Figure 3).



Figure 3: Papilla was incised between the lateral incisors and canines on both the right and left sides. The facial layer of the papilla was seperated on either side of the midline, effectively detaching the papilla from the tooth surface.

Using a #15 BP blade, an incision was initiated, commencing at the lateral incisor and progressing toward the canine to separate the facial layer of the papilla from its interdental papilla. Subsequently, an intrasulcular incision was extended in both the mesial and distal directions from the incised papilla, both on the mesial line angle of the lateral

incisor and the distal line angle of the canine, traversing facially to the central incisors. The incision spanned from the base of the sulcus to the crest of the bone. Furthermore, it was extended proximally to ensure detachment of the papillae from the proximal surfaces of the adjacent teeth (Figure 4).



Figure 4: An intrasulcular incision was extended in both the mesial and distal directions from the incised papilla, both on the mesial line angle of the lateral incisor and the distal line angle of the canine, traversing facially to the central incisors. The incision spanned from the base of the sulcus to the crest of the bone. Furthermore, it was extended proximally to ensure detachment of the papillae from the proximal surfaces of the adjacent teeth.

A full-thickness mucoperiosteal dissection was extended apically by approximately 4 mm from the tissue margin. This dissection also proceeded laterally beneath the facial aspect to create a tunnel for the placement of the membrane (Figure 5). To address any potential impediments to the unobstructed passage of the graft through the tunnel, a modified Orbans knife was used. This approach was utilized to traverse the prepared tunnel and incise any residual fibrous attachments that could hinder easy advancement of the graft.



Figure 5: A full-thickness mucoperiosteal dissection was extended apically by approximately 4 mm from the tissue margin. This dissection also proceeded laterally beneath the facial aspect to create a tunnel for the placement of the membrane.

A placental-derived, freeze-dried, irradiated chorion and amnion membrane (Tata Memorial Hospital Tissue Bank) (Figure 6) was inserted into the prepared tunnel after tunnel preparation, coronally advanced with the tunnel to cover the recession site (Figure 7), and sutured using interrupted sling sutures (Figure 8).



Figure 6: A placental-derived, freeze-dried, irradiated chorion and amnion membrane (Tata Memorial Hospital Tissue Bank)



Figure 7: A placental-derived, freeze-dried, irradiated chorion and amnion membrane (Tata Memorial Hospital Tissue Bank) was inserted into the prepared tunnel.



*Figure 8: Coronally advancement done with the tunnel to cover the recession site and sutured using interrupted sling sutures* 

A periodontal pack was placed on both the labial and lingual aspects of the treated site (Figure 9). Postoperative instructions and care were given to the patient. Amoxicillin 500 mg TID was administered for five days, analgesics were prescribed, and the patients were advised to use a mouth rinse containing 0.2% chlorhexidine gluconate for up to 14 days for plaque control. Patients were advised to maintain oral hygiene and follow a soft diet. Tooth brushing was discontinued near the surgical site for 14 days following the surgical procedure.



*Figure 9: A periodontal pack was placed on both the labial and lingual aspects of the treated site .* 

Patients were recalled after 2 weeks for suture removal, and follow-up was performed 3 months after root coverage. The clinical outcomes of the two patients who underwent surgery performed using this technique showed 100% gain in root coverage (Table 1) after baseline and after 3 months with an increase in biotype. (Figure 10), (Figure 11)

RECESSION DEPTH	,	AT BASELINE	AT 3 MONTHS
CASE I		3 mm	0 (complete root coverage)
CASE II		2 mm	0 (complete root coverage)
Percentage of root coverage	=	Recession depth at baseline	Recession depth at 3 months
gain	=	Recessio	n depth at baseline

Table 1: Root coverage achieved at the end of 3 months follow-up



Figure 10: Follow up of Case 1 at 3 months post root coverage.



Figure 11: Follow up of Case 2 at 3 months post root coverage.

#### Discussion

Enhancing both periodontal health and appearance is the goal of ideal periodontal care. As esthetics is one of the main complaints in patients with gingival recession, a pivotal strategy to improve esthetic results involves addressing gingival recession through therapeutic measures. The initial step of therapy for gingival recession is contingent upon the severity of the gingival defect and the etiological cause.<sup>11,12</sup>

The PATT is a relatively new tunneling technique introduced by Allen.<sup>9</sup> The technique of papilla access presents an alternative method for preparing tunnel sites, specifically in the mandibular anterior region. As per earlier research, the gingival sulcus is used to facilitate the tunnel access technique.<sup>13,14</sup> The papilla access technique improves the efficiency of tunnel site preparation and reduces the risk of surgical trauma or perforation that can occur with intrasulcular dissection without additional access. Additionally, this approach allows for the inclusion of posterior teeth with lateral extension of the tunnel as needed and access for easier, less traumatic graft insertion.<sup>15</sup>

The papilla access technique has several drawbacks, but its main downside is that suturing and positioning grafts on incised papillae require special care. To avoid crossing the papillary bed, it is crucial to place the graft at the level of the CEJs, which is crucial when allografts or xenografts are used. Additional limitations include the risk of tearing the delicate papillary tissue during manipulation, difficulty in achieving proper adaptation of the graft material in patients with thin biotypes, and challenges in ensuring adequate blood supply to the graft in cases of extensive recession or reduced interproximal tissue. Furthermore, the technique is technique-sensitive, requiring advanced surgical skills and experience, which may limit its applicability in routine clinical settings. Postoperative discomfort and prolonged healing times can also occur due to the manipulation of multiple papillae.<sup>16</sup>

In this case report, 100% root coverage was observed at every site treated with the pouch and tunnel technique. The initial recession height varied between 3 and 4 mm, with a width ranging from approximately 2 to 4 mm. After a one-year follow-up, all the sites displayed comprehensive root coverage. The tunnel technique, devised as a modification of the envelope technique, yielded positive outcomes in terms of root coverage. The application of the tunnel technique ensured superior graft adaptation to the recipient site, resulting in aesthetically pleasing outcomes and an increase in the thickness of the keratinized gingiva.<sup>17</sup>

The advantages of the tunneling technique over conventional flap techniques include the fact that the former is a miniamally invasive procedure that maintains blood supply to the gingiva, which results in a greater success rate in terms of esthetic outcomes and graft adaptability at the donor site.18 The papilla access technique is a more efficient way of preparing the tunnel site, as it reduces the risk of surgical trauma or perforation to the tissue that may occur with intrasulcular dissection. Additionally, this approach allows less traumatic and easier graft insertion and enables the inclusion of additional posterior teeth by lateral extension of the tunnel when necessary.<sup>9</sup> Its disadvantages include that it is technique sensitive. The primary drawback is the need for special attention while placing the graft and suturing the incised papillae, as it is crucial to align the graft level

with the CEJs to prevent it from extending over the papillary bed, notably when using allografts.<sup>19</sup>

The envelope flap has been shown in several trials to offer more stability and better results when treating multiple teeth than a flap with vertical releasing incisions.<sup>20</sup> The papilla access approach facilitates more effective preparation of the tunnel site and reduces the risk of surgical trauma or perforation to the tissue that might arise from intrasulcular dissection without extra access. It also allows access for lateral tunnel expansion to accommodate more posterior teeth as necessary, making the insertion of grafts simpler and less painful. The graft material used in these two cases was the amnion chorion membrane, whereas the acellular dermal matrix was used in the original study <sup>9</sup> because it was unavailable in India and because a 100% improvement was achieved in terms of complete root coverage at recession site defects in both cases.

Gingival tissue has a similar structure to fetal tissues. In dentistry, placenta-derived tissues are becoming increasingly popular as guided tissue regeneration membranes. Stem cell reserves and various growth factors and proteins, such as fibronectin, laminin, proteoglycans, and glycosaminoglycans, present in the amnion accelerate healing and regeneration, whereas the chorion is rich in collagen proteins.<sup>21</sup> The amniotic epithelium, basement membrane, compact layer, fibroblast layer, and intermediate/spongy layer make up the amnion membrane. Chorion membrane consists of the reticular layer, basement membrane, and trophoblasts.<sup>22</sup>

Placental allografts have an advantage because they are composed of immune privileged tissue, possess antibacterial and antimicrobial properties, and provide a protein-enriched matrix to facilitate cell migration. Amnion and Chorion membranes are now being used in root coverage procedures and are utilized as costeffective allogenic substitutes for subepithelial connective tissue grafts to achieve root coverage, thereby eliminating a second surgical site.<sup>23</sup>

#### Conclusions

This report outlines a surgical access approach that enables the use of a tunnelling technique in the mandibular anterior region, particularly in the presence of minimal to no recession or thin tissue that may pose a challenge in carrying out routine intrasulcular tunnel site preparation followed by graft placement without the risk of injury or perforation.

#### **Informed Consent**

Informed consent was obtained from the patient prior to the start of the treatment and for publication of this case report.

#### **Ethical Statement**

Approval was obtained from the institutional ethics committee (Manipal College of Dental Sciences, Mangalore, Manipal Academy of Higher Education) on the 10th of March, 2023 with protocol reference number: 23001.

#### **Conflict of Interest**

The authors declare that they have no conflict of interest.

#### Acknowledgement

None

#### Funds

The authors declare that no funds, grants, or other support were received during the preparation of this manuscript.

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### **Cumhuriyet Dental Journal**

Available online, ISSN: 1302-5805 e-ISSN: 2146-2852

Publisher: Sivas Cumhuriyet Üniversitesi

## Non-Syndromic Bilateral Dentigerous Cyst in 9-Year-Old Boy: A Case Report and Literature Review

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Case Report	ABSTRACT
History Received: 27/02/2024 Accepted: 15/12/2024	Dentigerous cyst is a developmental odontogenic cyst that often develops in the second and third decades of life. This is one of the most common odontogenic cyst types that involve developing, or impacted teeth, partially erupted, accounting 20% of all jaw cysts. Dentigerous cysts are usually unilateral, bilateral presentations have been associated with syndromes such as Cleidocranial Dysplasia and Maroteaux–Lamy syndrome. However, they can occur without any syndrome association, but are extremely rare. This article reports the case of bilateral dentigerous cysts that is non-syndromic in a 9-year-old boy.
License Discontinuity This work is licensed under Creative Commons Attribution 4.0 International License	Keywords: Bilateral Dentigerous cyst, Developmental cyst, Decompression, non-syndromic, Unerupted teeth.
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How to Cite: Vanguru R, Pasupuleti S, Manyam R, Tatapudi R, Pampana S. (2024) Non-Syndromic Bilateral Dentigerous Cyst in 9-Year-Old Boy: A Case Report and Literature Review, Cumhuriyet Dental Journal, 27(4): 260-268.

#### Introduction

Dentigerous cyst (DC), which Paget first identified in 1863, are slow-growing odontogenic tumors of developmental origin that are most frequently found on the crown of an impacted or unerupted third molar.1 They are the most common type of jaw developmental cyst, accounting for approximately 20% of all jaw cysts.<sup>2</sup> It is derived from the reduced enamel epithelium and has been described as an epithelium lined cavity. It is usually asymptomatic although it can increase in volume causing pain and compression of nerve structures.<sup>3</sup> These cysts are usually unilateral, developing around the crowns of impacted mandibular third molars.<sup>2</sup> Radiographically, a unilocular radiolucency is associated with the retained or embedded tooth. Syndromes like Cleidocranial Dysplasia and Maroteaux-Lamy have been linked to bilateral dentigerous cysts. It is unusual to find bilateral dentigerous cysts that are not syndromic, especially in the mixed dentition.<sup>2</sup> So far, less than twenty such cases have been reported.<sup>1</sup> According to the study conducted by Nuraini H, DC occurred more commonly in permanent teeth than primary. He also stated that among the primary dentition, DC is more often due to the impaction of mandibular molar followed by maxillary canines, maxillary second molar, and mandibular canines.<sup>4</sup> We report a rare case of bilateral non-syndromic dentigerous cysts in a 9-year-old boy and Literature review to emphasize the importance of early diagnosis and management of this condition to prevent potential complications.

#### **Case Report**

A 9-year-old male patient presented to the institution complaining of swelling in the right and left lower back tooth region for one month. He was apparently well two months back, later a swelling developed that gradually attained the present size. He also complained of occasional pus discharge, associated with pain which was intermittent, dull, non-radiating, aggravating on eating hard foods relieved on taking medication. There was no history of paraesthesia, fever and salty discharge. Patient visited private dental clinic 10 days ago for the same complaint where he was given medication and referred to our hospital. On extra oral examination there was no obvious swelling bilaterally but intra orally there are bilateral ovoid swellings in the vestibular region. On palpation, a swelling of 2x3 cm is noted extending 1cm away from symphysis region, 4cm away from ear lobule lying in 74 region mesial to 36 on the left side and a 2x2 cm swelling extending from 84 tooth region to mesial aspect of 46 on right side. (Figure1) Based on these features he was provisionally diagnosed as Radicular cyst in relation to 74,75,85 and Chronic apical periodontitis irt 84.

For radiographic evaluation, a CBCT was advised. Axial section of mandibular arch showed multiple well defined hypodense areas surrounded by hyperdense tooth like structures irt 85, 46, & 74,75 regions respectively with thin expanded corticated borders. (Figure 2) Panoramic view showed multiple well defined hypo dense areas were seen and displacement of tooth were also noticed.(Figure 3) Based on these details, it was given as dentigerous cyst. Marsupialization of tissue was done irt to 75, 84,85 along with extraction of 75 and submitted for histopathological examination.

On microscopic examination, tissue bits from the right side showed, focal areas of odontogenic cell clusters resembling cell rests with inflammatory areas whereas those from left side showed odontogenic islands with acanthomatous change and odontogenic cell clusters resembling cell rests with inflammatory areas. (Figure 4) Correlating clinically and radiographically, it was diagnosed as bilateral inflamed dentigerous cyst. Five days after marsupialization the patient was given lingual arch space maintainer.

In the follow-up visit of one week, Clinical examination showed tissue overgrowth in 84 region that was surgically removed as it interrupts the eruption. Histopathological examination revealed granulation tissue with odontogenic epithelial lining.

In further follow up visit of one week, OPG showed a bilateral well-defined radiolucency involving the premolar regions of 34, 35, 45 with intact inferior border of mandible. (Figure 5) The tissue bits obtained from right and left premolar region. A decompression tubes were placed on either side attached to a denture. The histopathological examination of specimen showed features consistent with that of first biopsy and was diagnosed as bilateral dentigerous cyst. The patient is under the follow-up.



Figure 1: Clinical image



Figure 2: Axial section of CBCT showing multiple well defined hypodense areas.



Figure 3: Panoramic image showing multiple well defined hypo dense areas



Figure 4: Histopathology showing odontogenic epithelial islands and cell clusters.



Figure 5: OPG showing bilateral well-defined radiolucencies in premolar areas

#### Review of cases reported in literature: 5-68

Table 1: Review of non- syndromic bilateral DC cases reported so far	
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S No	)	Authors	Age	Sex	Location	Treatment
			14	F	15,18,28,37,38,48	
	1.	lvy et al (1939)	18	F	48, 47, 35,38	Enucleation
	1.	ivy et al (1959)	12	F	43 48, 32, 33, 37	Endcleation
			11	Μ	13,23,34	
	2.	Myers (1943)	19	F	38,48	Enucleation
	3.	Catania et al.(1952)	19	F	18,28,32,33,38,42,43,48	Enucleation
	4.	Henefer (1964)	52	F	18,28	Enucleation
	5.	Stanback (1970)	9	М	36,46	Enucleation
	6.	Callaghan (1973)	38	М	38,48	Enucleation
	7.	Burton (1980)	57	F	38,48	Enucleation
	8.	Swerdloff (1980)	7	F	38,48	Enucleation
	9.	Crinzi RA (1982)	15	F	38,48	Enucleation
	10.	Norris et al (1987)	14	F	13,14,15, 17,23,24,25,27	Marsupialisation
		McDonnell DG (1988)	15	М	35,37,45,47	Enucleation
		Eidinger GB (1989)	15	М	36,46	Enucleation
		O Neil DW et al (1989)	5	М	36,46	Enucleation
		Banderas JA et al (1996)	38	М	38,48	Enucleation
		Sands and Tocchio (1998)	3	F	31,36,41,46	Enucleation
		Ko kS et al (1999)	42	M	38,48	Enucleation
		De Biase et al & others (2001)	8	M	36,46	Enucleation
		Shah N et al (2002)	39	M	38,48	No treatment
		Ustuner et al (2003)	6	M	13,23	Enucleation
		Batra et al (2004)	15	F	35,38,45,48	Enucleation
		Freitas et al (2006)	14	M	18,28, 35,45	Enucleation
		Lung and Ganatra (2006)	77	M	38,48	Enucleation
		Sumita M et al (2006)	13	M	35,45	Enucleation
		Mahajan et al (2006)	13	M	35, 44	Enucleation
		Yamalik et al (2007)	51	M	NA	Enucleation
		Farahani &Lotfalian (2007)	37	M	13,23,33,43,32	Enucleation
		Fregnani et al (2008)	5	M	36,46	Enucleation
		Carter et al (2008)	81	M	38,48	Enucleation
		Chew YS et al (2008)	30	F	38,48	No treatment
		Cury et al (2009)	5	M	38,48	Enucleation
		Grewal HK et al (2010)	11	M	13,43	Enucleation
	51.		11	141	14,15,24,25,34,35,44,45	Endeleation
	32.	Saluja et al (2010)	22	Μ	14,15,24,25,54,55,44,45	Enucleation
	22	Tikekar S et al (2010)	11	м	34,35,44,45	Marsupialization
		Gaur G et al (2010)	7	F		Enucleation
			7 32	M	16,26 13,23	Enucleation
		Kannan et al (2010) Prasad LK (2010)	52 12	F		Enucleation
		Shiva S et al (2011)	12		13,23 35,45	
				M	35,45 34,35,44,45	Marsupialization Enucleation
		Reddy et al (2011) Brabbakar y et al (2011)	11	F		Enucleation
		Prabhakar v et al (2011)	10	F	13,23, 11,21	
		Tamgadge et al (2011)	10	M	14,15,24,25,13,23	Enucleation
		Kanth et al (2011)	9	F	34,35,44,45	Enucleation
		Ozkan et al (2011)	NA 10	M	38,48	Enucleation
		Gonzalez SM (2011)	10	F	33,43	NA
	44.	Shirazian and Agha-Hosseini (2011)	10	M	34,35,44,45	Marsupialization
			7	F	35,45.	
			8	F	35,45	
	45	$A_{\rm kov}$ at al. (2011)	7	M	35,45	
	45.	Akay et al (2011)	8	М	15,25,35,45.	Marsupialization
			9	F	13,23	
			8	F	13,23, 14,15,24,25	
			9	Μ	35,45	- · · · ·
		Ishihara et al (2012)	NA	NA	14,15,24,25,34,35,44,45	Enucleation and
	46.				, , , -, - , - , -, -	auto-transplantation
						- I
	47.	Aher V et al. (2013)	24	М	18,28,38,48	Enucleation
	47. 48.	Aher V et al. (2013) Jia et al (2013)	61	F	38,48	Enucleation
	47. 48. 49.	Aher V et al. (2013)				

54		NA	NA	38,48	
51.	Lin et al (2013)	NA NA	NA NA	13,23 15,23	Enucleation
		INA		15,25	Marsupialization &
52.	Imada et al (2014)	42	F	NA	Enucleation
53.	Cura et al (2014)	45	М	44, 43,33 14,15,24,25	Enucleation
		34	М	33,43	Enucleation
54.	Kaushik A, et al. (2014)	17	F	13,23	Marsupialisation
55.	Vassiliou et al (2014)	38	F	36,37,46,47	Enucleation
					Marsupialisation (rt)
56.	Morais et al (2014)	15	Μ	37,47	and enucleation of
					(It) cyst
	Naik et al (2014)	25	Μ	38,48	Enucleation
58.	Rocha et al (2014)	43	F	13,23	Enucleation
	Shetty et al (2014)	12	Μ	33,43	Enucleation
	Swain et al (2014)	9	F	13,23	Enucleation
	Devi P et al (2015)	17	Μ	33,43,34,35,44,45	Enucleation
	Hansford JT (2015)	6	F	NA	NA
63.	Sanjay CJ (2015)	24	F	33,43	NA
64.	Singh et al (2015)	8	М	33,43	Enucleation
65.	Bansal et al (2015)	34	Μ	33,43	Enucleation
66.	Jae-Yun jeon (2016)	15	Μ	18,28,38,48	Enucleation
67.	Kashyap et al (2016)	23	Μ	Mx right and left supernumeraries and 23	Enucleation
68.	Gnanaselvi et al (2016)	14	Μ	13,14,23,24,33,34,43,44	Enucleation
69.	Dhupar A (2017)	8	Μ	13,14,15,23,24,25,34,35,44,45	Enucleation
70.	Khandeparker RV et al (2018)	10	Μ	15, 23	Enucleation
71.	Moturi K et al (2018)	19	F	Multiple teeth in Mx and Md arches	Enucleation
72.	Vasiapphan H (2018)	27	М	38,48	Enucleation
73.	Pant B et al (2019)	10	М	13,23	Enucleation
74.	AlKhudair B (2019)	19	М	Mx sinus	Enucleation
75.	Sindi AM (2019)	44	М	37,47	Enucleation
76.	Sharma S, (2019)	27	F	18,28	Enucleation
77.	Sethi A (2020)	12	М	35,45	NA
78.	Bang KO et al (2021)	55	F	38,48	NA
79.	Arici M (2022)	32	F	18,28	Enucleation

\*NA- not available, F- Female, M- Male, Md- mandibular, Mx- maxillary, Rt-right, It-left, Location - Tooth Numbering system according FDI (Federation Dentaire Internationale) System.

#### Discussion

A dentigerous cyst can be defined as "a cyst that encloses the crown of an unerupted tooth, expands the follicle and is attached to the cemento-enamel junction of the unerupted tooth".<sup>68</sup> They tend to occur across a wide age range, peaking between the second and fourth decade. Dentigerous cysts' precise histogenesis is unknown; however the majority of publications favour a developmental origin from the tooth follicle.<sup>69</sup> Two different forms of dentigerous cysts were put forward by Al Talabani and Smith. One results from early stellate reticulum degeneration, which is linked to hypoplastic teeth, and the other develops after the crown completion through fluid buildup between the layers of decreased enamel epithelium.<sup>70</sup>

Racial, ethnic, sexual, and individual variables can affect how normally teeth erupt into the oral cavity, and these are typically taken into account when establishing standards for proper eruption.<sup>71</sup> There are numerous

eruption regulating molecules like EGF, EGF-R, CSF-1R, CSF-1, IL-1R, IL-1, c- NFB, Fos, TGF-b1, MCP1, PTHrP, OPG, Cbfa-1, and RANK/RANK L that reside in dental follicle with a few in Stellate reticulum.<sup>72</sup> The anomalies in the eruption, however, might be caused by a gene abnormality.<sup>73</sup>

Syndromes such as Cleidocranial Dysplasia, Maroteaux-Lamy Syndrome, and Mucopolysaccharidosis, are frequently observed in conjunction with the bilateral and multiple cysts. Without these characteristics, bilateral dentigerous cysts are uncommon.<sup>74</sup> All of these circumstances disrupt tooth development or eruption in some way, which accounts for the presence of many cysts. The age and sex predilections for non-syndromic cases according to literature are given in the Graph (1). In terms of etiology, Batra P reported Chromosomal polymorphism in bilateral dentigerous cyst whose karyotyping revealed a large secondary constriction in chromosome 1qh+ 2.<sup>75</sup> The combined effect of cyclosporin and a calcium blocker was highlighted by Yamalik K et al.<sup>76</sup> in the development of bilateral dentigerous cysts. But there is no proof linking them to the bilateral prevalence of dentigerous cysts, and they could simply be coincidental findings. In 2001, De Biase A et al. observed that long-term, concomitant usage of cyclosporine A and calcium channel blockers can result in bilateral mandibular dentigerous cysts.<sup>77</sup>

The most common site of occurrence is Mandibular, Maxillary third molars and maxillary cuspids but the literature search showed other combinations as well.<sup>78</sup> Aher V et al and jae Yun jeon reported occurrence of bilateral DC in both maxillary and mandibular third molars.<sup>79,80</sup> According to the cases reported thus far, Mandibular third molars (20.65%) are the most frequently affected site for Dentigerous Cysts, followed by mandibular premolars (11.95%) and maxillary canines (10.86%). Sizable fractions (44.56%) of cysts develop in other combinations at different jaw regions.<sup>5-68</sup>

Only 91 cases of multiple dentigerous cysts that are not syndromic have been documented thus far. (Table 1) In non-syndromic patients, multiple dentigerous cysts are regarded to be incredibly unusual. In a study of 2944 cases of odontogenic cysts, Ochsenius et al. found 546 patients with dentigerous cysts, 61 (11%) of whom had synchronous dentigerous cysts. None of these patients had any metabolic or syndromic conditions, but it is unclear how many of these cysts were bilateral or led to root resorption.  $^{\ensuremath{81}}$ 

A complicated interdisciplinary strategy is required for the management of many impacted teeth linked to dentigerous cysts. When planning the course of treatment, the patient's age, the location of their teeth, the number of impacted teeth associated with dentigerous cysts, and any accompanying metabolic, genetic, or syndromic disorders must all be taken into account. Patients with many impactions require guided eruption of numerous teeth with the use of coordinated multidisciplinary care.82 Enucleation of minor lesions can prevent harm to the implicated permanent teeth. It is possible to decompress and marsupialize larger lesions to release the pressure inside the cysts, followed by enucleation-also known as Waldron's procedure-when the size has shrunk.83,84 The management procedures followed so for the cases reported in literature was given in the Table 2.

Dentigerous cysts does not pose any challenges after complete removal as they do not reappear.<sup>85</sup> This is due to the diminished enamel epithelium's fatigued state after differentiating and forming tooth crown enamel before developing into a cyst.<sup>86</sup> Shah et al suspected spontaneous regression of these cysts after 3 years, without any intervention.<sup>87</sup>





Table 2: Treatment modalities followed for bilateral DC according literature

S no.	Treatment	Percentage (%)
1.	Enucleation	75
2.	No treatment	2.17
3.	Marsupialization	15.21
4.	No data available	6.52
5.	Marsupialization + Enucleation	1.08

#### Conclusions

The occurrences of non syndromic dentigerous cysts are very rare and are challenging to draw conclusions on their etiopathogenesis due to their rarity and the dearth of evidence available in the literature.

#### **Declaration by Patient**

The authors attest to obtaining all necessary patient consent papers. The patient has agreed on the form to the publication of his photos and other clinical data in the journal. The patient is aware that while every attempt will be made to keep their identity a secret and that their name and initials won't be published, anonymity cannot be guaranteed.

#### **Authorship Contribution**

RTV, SP conceived and designed the study, RTV acquired the data; and SP, RM conducted data analysis and interpreted the results. RTV, SP wrote the initial and final drafts of the manuscript, and RM, RT, SRP, provided logistic support. All the authors drafted the manuscript and designed the tables with critical revisions. All authors discussed the results and commented on the manuscript. All authors have critically reviewed and approved the final draft and are responsible for the content and similarity index of the manuscript.

#### **Conflict of Interest**

None

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