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Evaluation on Shear Bond Strength of Different Glass Ionomer and Hydroxy Apatite Cements Used in Ossiculoplasty

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Background: Glass ionomer cements (GIC) have been widely used in dentistry for many years. In recent years, GIC have also been used for ossiculoplasty. The bond strength of GIC used in ossiculoplasty and the way they may change over the years in the cementation area are being questioned. The bonding strength of the substance may be of importance for long-term outcomes.

Aims: The aim of this study was to investigate the bond strength of different GIC on ossicles.

Study Design: *In vitro* study.

Methods: Twenty ossicles were obtained from patients who had undergone ear surgery. All specimens were randomly divided into four subgroups. All specimens were inserted into a specially designed apparatus for shear bond strength (SBS) testing. The tested materials [Aqua Meron (AM), Aqua Cem (AC), Ketac Cem (KC), and Otomimix CPB (OH)] were prepared and applied according to the manufacturer's instructions. The SBS was tested using a universal testing machine at a crosshead speed of 0.5 mm/min.

Results: The mean SBSs were found to be 13.28 MPa, 23.43 MPa, 8.51MPa, and 1.78 MPa for AM, AC, KC, and OH, respectively. AC had the highest SBS, which was statistically significantly different from that of KC and OH ($p<0.05$). Both AM and KC had higher SBS than OH ($p<0.05$).

Conclusion: The results obtained in this study by investigating the bone-bonding strength of cements widely used in ossiculoplasty demonstrate that some of these substances have a greater ability to bond to ossicles compared to others. Further clinical investigations are needed to test different parameters.

Keywords: Bone cement, bond strength, ossicles, ossiculoplasty

The natural ossicular chain plays an important role in transporting sound efficiently from the environment to the oval window (1). Diseases of the middle ear, such as chronic otitis media, cholesteatoma, and trauma, can interrupt this transfer. Incudostapedial joint discontinuity is the most common ossicular defect encountered in tympanoplasty (2, 3). Other problems of the ossicular chain include a defective stapes superstructure with or without a defective incus, and defective malleus (4).

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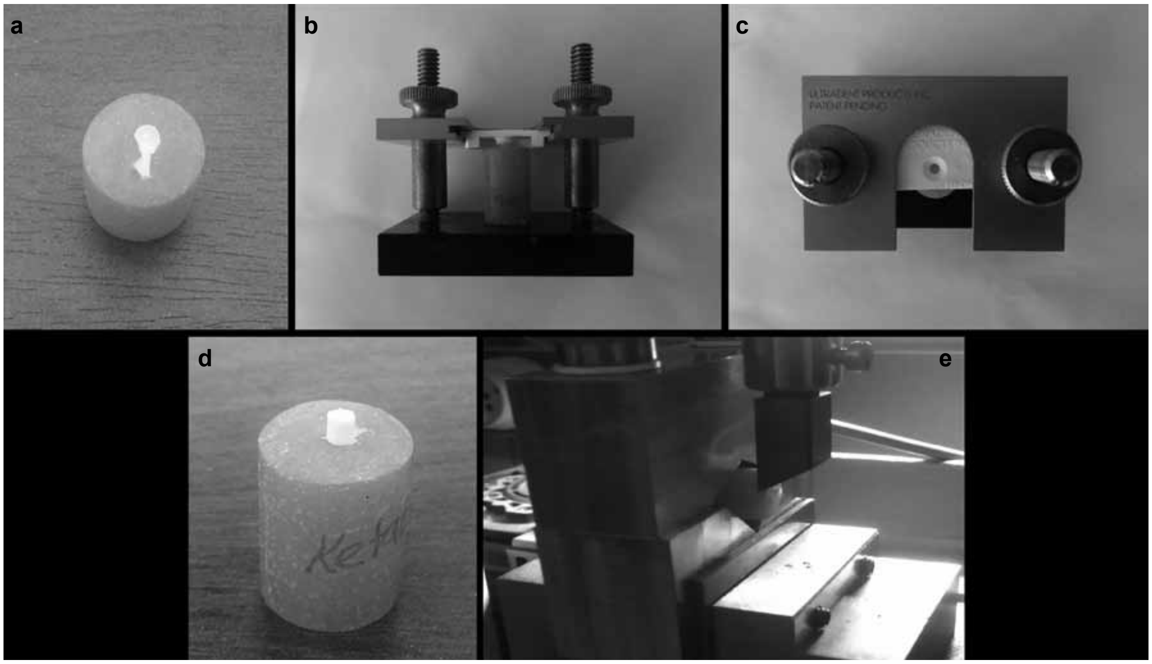

FIG. 1. a-e. Preparation of test specimen and SBS testing. Ossicles embedded into the auto-polymerised acrylic resin (a). Specimen placed onto the special apparatus designed for SBS (b), specimen placed onto the special apparatus designed for SBS (c), specimen ready for SBS testing (d), SBS testing with universal testing machine (e)

Ossicular chain reconstructions with total and partial ossicular replacement prostheses and incus repositions are the most frequently used treatment options for these problems (5-7). Most of these treatment options have aimed to transport the sound into the oval window via a prosthesis. Successful ossicular reconstruction requires a stable connection between the vibrating tympanic membrane and the inner ear. Although replacement with a prosthesis is extensively used, this prosthesis can dislocate over time, resulting in decreasing long-term hearing quality. It was reported that, in patients requiring revision of ossiculoplasty, half of the failures result from displacement of the prosthesis (4). While the incus reposition is simple and cost effective, the prosthetic rehabilitations are time consuming and highly costing.

Glass ionomer cements (GIC) have been widely used in dentistry for many years. Among the different treatment options, GICs have also been used for ossiculoplasty for two decades. Geyer and Helms first reported its use in middle ear surgery, in the 1990s (8). It is now used in ear surgery for various indications, such as ossicular reconstruction, bony external ear canal repair, stabilisation of cochlear implants, repair of dural defects, and reconnecting of gaps created by incudostapedial necrosis (9). Several studies have shown that there was a considerable increase in patients' hearing quality when ossicular chain was rebuilt with GIC (4, 10-14). The ideal bone cement used for this purpose should be malleable, easily applied, rapidly setting, non-toxic, fluid resistant, capable of osteointegration, and able to incite minimal inflammation (1).

There are different types of GICs which have different powders used for ossiculoplasty in the literature. The bond strength of GICs used in ossiculoplasty and the way in which they may change over the years in the cementation area are being questioned. The bonding strength of the substance may be of importance for the long-term outcomes. The aim of this *in vitro* study was to evaluate the shear bonding strength of three different GIC used in literature and Hydroxyapatite (HA) for ossicular chain reconstruction materials.

MATERIALS AND METHODS

In this study, we used twenty ossicles removed from the patients, who were operated upon due to advanced ear disease, after local ethic committee approval (2012/61) and after obtaining written informed consent from all subjects.

Specimen preparation

In total, twenty ossicles were prepared and kept in alcohol solutions until the shear bond strength testing (SBS) was performed. Aqua Meron (AM), Aqua Cem (AC), Ketac Cem (KC), and Otomimix CPB (OH), which are frequently used bone cements for ossiculoplasty, were used as testing materials in the current study (Table 1). Bone specimens were embedded into the auto-polymerised acrylic resin block in a cylindrical plastic mould. After the complete setting of resin,

TABLE 1. Used materials and manufacturers and contents information's

Luting Agent	Manufacturer LOT number	Type	Powder	Liquid
Aqua Meron	Voco, Germany 1118221	GIC	Na-Ca fluorosilicate glass, Polyacrylic acid	Distilled water
Aqua Cem	Dentsply, USA 1102002368	GIC	Na-Ca fluorophosphoroaluminiumsilicate, Polyacrylic acid, Tartaric acid, Yellow Ferric	Distilled water
Ketac Cem	3M ESPE, USA 458814	GIC	Glass powder, Polycarboxylic acid, Pigments	Water, tartaric acid, conservation agents
Otomimix CPB	Biomet, USA 307750	Bone filler	Calcium phosphate powder, sodium citrate dihydrate	anhydrous citric acid, distilled water

GIC: Glass ionomer cement

TABLE 2. Manipulative variations of used materials

Material	P/L	Mixing Time	Working Time	Setting Time
Aqua Meron	3.3-3.8/1	30 sec	3 min	6-7 min
Aqua Cem	3.3/1	15 sec	2:30 min	3.5-5 min
Ketac Cem	3.8/ 1	1 min	3:30 min	3:4.5 min
Otomimix CPB	1/1	30-45 sec	3-4 min	4-6 min

P/L: powder/liquid; sec: second; min: minute

TABLE 3. Mean SBS values and standard deviations

Groups	N	SBS (Mean±Std. Error)	Bonforonni adjusted with Mann Whitney U*
Aqua Meron	5	13.28±4.13	A, B
Aqua Cem	5	23.43±7.71	A
Ketac Cem	5	8.51±2.31	B
Otomimix CPB	5	1.78±1.21	C

*same letter indicates that there is not significantly difference at a 0.05.

N: number; SBS: shear bond strength

bone surfaces were ground with a dental diamond bur instrument under water (Figure 1a). Then, after all specimens were divided into four groups randomly (n=5) by using the block randomisation method (15) and cemented with three different GICs and one bone void filler. The tested materials and their compositions are summarised in Table 1. Before the cementation, all specimens were washed with water and gently dried with oil free air, but were not dehydrated, following insertion into the specific apparatus designed for SBS testing (Ultradent Production Inc., South Jordan, USA), in a manner that the cementation area (2 mm in height and diameter) was completely inside of bone border (Figure 1b, c). All materials were proportioned and handled according to the manufacturer's instructions (Table 2). After the mixing of powder and liquid, cement was carried into the hole of the apparatus with a hand instrument and moderate pressure was applied until the initial setting of cement was completed (Figure 1d).

Shear bond strength testing

The SBS of the specimens which set completely was measured after 24 hours of storage at 37°C. The specimens were placed in a universal testing machine (Model 5565; Instron Co Ltd, Canton, Mass). Load was applied parallel to the long axis of the specimens and as close as possible to the interface between ossicles and cement, at a constant crosshead speed of 0.5 mm/min until failure (Figure 1e). The SBS was calculated by dividing the failure load (N) by the bonding area (mm^2) as megapascal (MPa).

SEM analysis of bonding interface

To thoroughly assess the bonding interface, a new specimen was prepared for each material. The ossicles were embedded into the auto-polymerised acrylic resin which was 2 mm in thickness. Then, the surface of ossicles was coated with GICs and HA bone cement. After completely setting, specimens were cut along their length using a low speed diamond saw (Isomet 1000, Buehler, Lake Bluff, IL, USA). The specimens were sputter-coated with 15 nm gold-palladium (Bal-Tec, SCD 050, Scotia, NY, USA) for 130 seconds at a current of 10-15 mA and a vacuum of 130 mTorr and then examined by scanning electron microscope (LEO, EVO 40 XVP, Cambridge, UK) with an acceleration voltage of 20 KV and different magnifications.

Failure mode analysis and SEM evaluation

All specimens submitted to the shear bond testing were observed using an optical light microscope (SMZ 800, Nikon, Tokyo, Japan) at 40x magnification. Failure modes were classified based on the following criteria: adhesive failure between the ossicles and the cement, cohesive failure in the resin cement, and mixed failure that included the cohesive failure of the ossicles and the cement.

To analyse the types of failure, demonstrative specimens were selected and SEM images were obtained as mentioned above.

Statistical analyses

Statistical analysis was performed using a computerized statistical software program (SPSS Statistic for Windows, Version 17.0, Chicago: SPSS Inc., USA) A Kruskal-Wallis one way ANOVA analysis was used to determine whether there were significant differences in SBS among the groups, followed by the Mann-Whitney U Test with Benforroni adjustment to allow comparisons between the groups. The statistical significance was set at 5%.

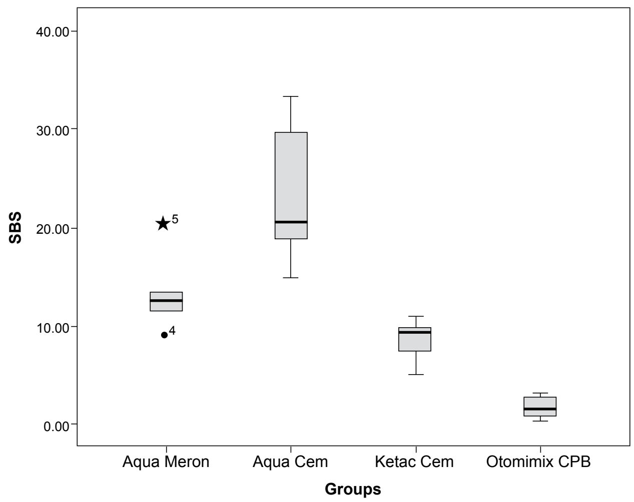

FIG. 2. SBS values of tested cements

RESULTS

The mean SBS for the cements are represented in Table 3 and Figure 2. Kruskal-Wallis analysis showed that the cements used affected the shear bond strength on the ossicles. The bone cement, OH (1.78MPa), had significantly lower SBS than all other GIC groups. AC had the highest SBS (23.43MPa) among all of the groups and was significantly different from the KC (8.51 MPa) and OH (1.78 MPa), but not AM (13.28 MPa) ($p<0.05$). AM showed higher SBS than KC, but this was not statistically significant ($p<0.05$). Both AC and AM had a higher SBS than OH ($p<0.05$) (Table 3).

The ossicles' surfaces were covered continuously by all of the tested cements. Cements penetrated into the ossicles in the form of tags. All of the ossicle surfaces showed a smooth fibrous appearance (Figure 3a-c).

Surface characteristics of AC were similar and most like the ossicles (Figure 3b) and there were little micro voids between the cements and ossicles. On the other hand, a well-developed bonding surface with cement tags penetrating into the ossicles was also observed along the interface walls.

KC and AM provided a sharper and more distinct picture of the situation and enabled a morphological condition in greater

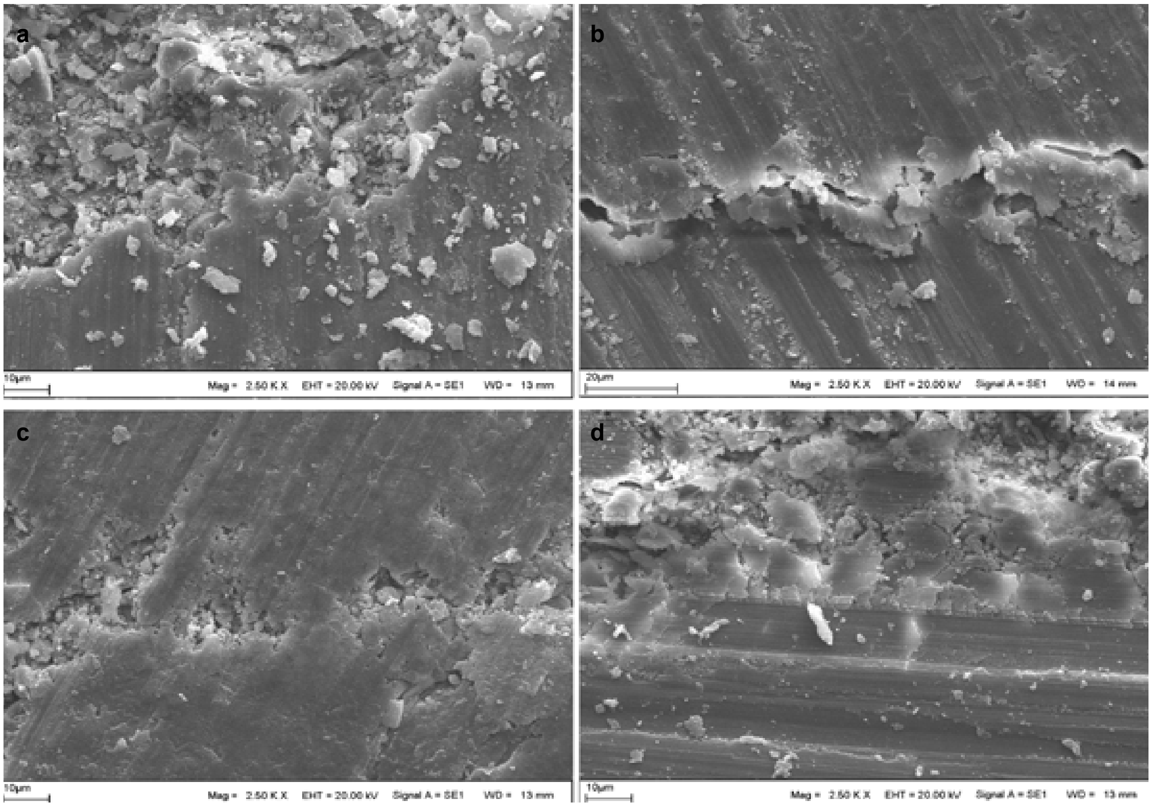

FIG. 3. a-d. SEM evaluation (x2500) of ossicle and cement interface for Aqua Meron (a), SEM evaluation (x2500) of ossicle and cement interface for Aqua Cem (b), SEM evaluation (x2500) of ossicle and cement interface for Ketac Cem (c), SEM evaluation (x2500) of ossicle and cement interface for Otomimix CPB (d)

details. The differences between the materials used, quality of set material, and character of filler particles were better demonstrated by the application technique (Figure 3a, c). Co-polymerisation of OH was found to be good and without voids. However, there are significant differences of internal structures between the ossicles and OH. In addition, OH presented porous and weak textures (Figure 3d).

The failure mode distribution for all specimens is presented in Figure 4. For AM, failure mode distribution was adhesive (40%), cohesive (40%), and mixed (20%). For the AC and KC groups, the failure modes were mainly adhesive (60%) and mixed (40%). For the OH group, failure modes were completely adhesive (100%). SEM images of the different failure types are presented in Figure 5a, b, c.

DISCUSSION

The natural ossicular chain conducts sound efficiently from the environment to the inner ear (1). The prosthesis aims to transmit sound to the oval window through the natural ossicular route. Although an incudostapedial clips prosthesis ensures the functional success for rehabilitation of the incus long arm defect, it increases the cost of operation (16).

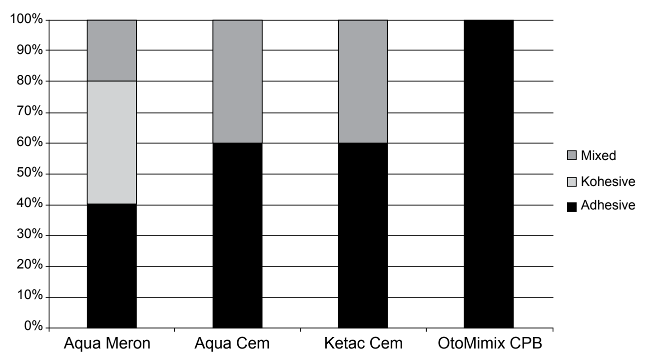

FIG. 4. Failure type analysis of tested materials

Glass ionomer cements have been widely used in dentistry from the 1960s as a luting and filling restorative material due to their adhesive and other advantageous properties. In general, GICs contain organic acids, a glass component and additives. The setting reaction of GIC involves the acid-base reaction of a polyacrylic acid and glass particles and ions (Al^{3+}, Ca^{2+}) located in the network of the glass (17). The adhesion of GICs to the dental hard tissues appears to be via mechanical interlocking and the development of an 'ion-exchange layer' adjacent to the dentine (18). GICs have been used in dentistry for four decades; and took place in otology practice, especially in ossiculoplasty operations since the 1990s. The GICs are cost effective and alternative materials for the treatment of different types of defects. Reconstruction of incus long arm defects with GICs especially contribute to the transfer of sound by the natural ossicular chain.

Hydroxyapatite bone cements have also been chosen as prosthetic materials for incudostapedial rebridging ossiculoplasty. The use of HA is increasing, and very satisfying results have been reported by many papers. OH is a calcium phosphate powder that forms HA bone cement when mixed with a setting solution (1). It is used for several conditions, as a reconstructive material for incus erosion, total or partial ossicular replacement material, in revision surgery with incus erosion and, in primary stapedotomy to fix the crimped prosthesis to an intact incus (1).

Mechanical testing allows evaluation of the bonding strength of the biomaterials before clinical practice and to compare the materials against each other. The SBS test is one of the most commonly used bond strength tests. *In vitro* testing conditions differ within the same study as well as between different clinical *in vitro* studies. For this reason, well standardised specimens were prepared using a specific apparatus for the SBS test, and stored in the same conditions before and after the preparation of testing with 0.5 mm/min crosshead speed.

This *in vitro* study evaluated the SBS of three different GICs and one HA to the ossicles. The most important result of this

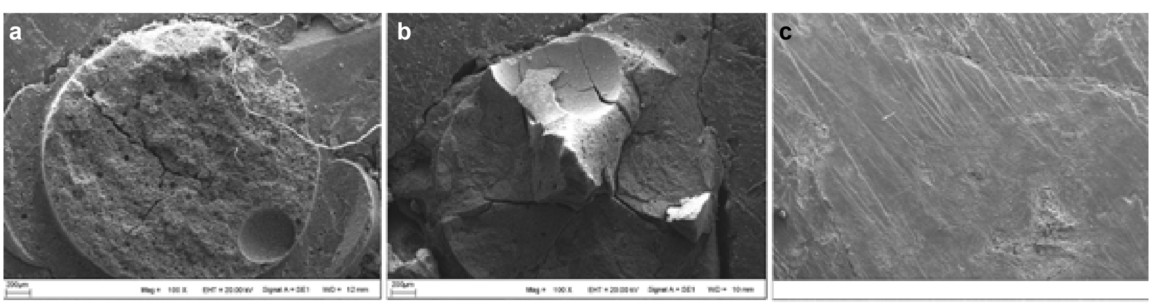

FIG. 5. a-c. Representative SEM images of different failure modes after SBS testing. SEM images (x100) of cohesive failure into the Aqua Meron GIC (a). Representative SEM images of different failure modes after SBS testing; A mixed failure of Ketac Cem GIC (x100 magnification). A part of cement crack is seen at half part of the ossicle surface (b). Representative SEM images of different failure modes after SBS testing; Adhesive failure of OtoMimix CPB specimen (x250 magnification) (c)

study is that all GICs had higher SBS values than the HA. GICs used in this study have significantly superior SBS values (AM; 13.28MPa, AC; 23.43MPa, KC; 8.51MPa) to the ossicular bones than the bone filler (HA, OH; 1.78 MPa). A variety of GICs with different contents have been used in ossiculoplasty (4, 10, 11). It is considered that the differences among the SBS values of different GICs are due to small variations in their contents. GIC is bonded chemically to the dental hard tissue during the setting reaction. It is thought that the bonding mechanism involves the chelation of carboxyl groups of the polyacids with calcium and phosphate ions from the enamel and dentin. When the GIC is used for ossiculoplasty, an ionic interaction between the GIC and ossicles probably results in strong chemical bonds. Since GICs bond firmly to stapes, it may be difficult to remove it during a revision surgery. Thus, an inappropriate manipulation may cause stapes to fracture (19).

The use of GICs for different ossicular reconstructions such as incus stapes re-bridging, incus augmentation, and the fixation of stapes implants have been shown by several studies as the reliable method (4, 12, 19, 20). Baglam et al. reported a significant hearing improvement after incudostapedial re-bridging ossiculoplasty with KC (20). Bayazit et al. presented a significant improvement in the hearing results of different aural pathologies and surgeries after two years of follow-up with bone cement ossiculoplasty (19). Righini-Grunder et al. reported their long-term experiences by using Sereno Cem and KC GICs for different kinds of ear surgeries (9). They noted some revision surgeries due to cement breakage. This study shows that the cement connection could break or debond; thus, a revision surgery may be needed.

Hydroxyapatite has been recognised as a well-tolerated material in cranioplasty and in the production of ossicular prostheses (1). In this study, OH showed less bonding strength to ossicles. While the GICs are mainly used as a luting material, HA is a bone filler material and could not form tight and rigid structures from the initial setting time until testing. Due to its fragile structure, there was a problem during the SBS testing for HA specimens. HA creates about 70% of bone and HA cements harden as a microporous HA. It has been shown in animal studies that HA is an osteoconductive material. Many retrospective studies also confirmed the long-term clinical success of HA. It is likely that connection at the interface between the HA and ossicles gets stronger over time due to the osteoconductive activity of HA. Goebel et al. used the Mimix, another form of OH, to repair a variety of ossicular problems, and also reported significant improvements in functional outcomes after an 11 month follow-up (1).

Manipulative variations such as mixing time, setting time, consistency and easy application are the preferred factors for shortening the operating time for ossicular surgery. There are slight manipulative variations (Table 3) among the tested GICs and HA. Short working and setting times of GICs and HA can shorten the operation time. On the other hand, the GICs develop a putty-like consistency that makes it easy to handle and they can be applied as accurately as HA. However, ossiculoplasty with GIC is cost effective, which is an advantage (19).

Mechanical and bonding properties of GICs could be altered by the powder/liquid (P/L) ratio. In general, increasing the P/L ratio results in higher viscosity decreasing the wetting ability of GIC on the ossicles. In contrast, the cement's properties would weaken and fluidity would increase when the ratio is lowered. Consequently, changing the P/L ratio from the manufacturer's recommendation makes it complicated to connect the two ends of ossicles and has a negative effect on the retention and survival of the re-built part.

The hearing quality after ossiculoplasty is affected by the prostheses quality and environmental factors such as the presence or absence of malleus, dry or wet ear, or primary or revision surgery. In contrast, the GICs are moisture-sensitive materials, especially in the early setting (10 min.) phase. This means that a gain or loss of water from the surface in the early setting phase in clinical conditions can severely affect the final mechanical properties of the GIC. For this reason, the application area has to be dried during the operation for reliable and strong bonding and long-term clinical success. Therefore, the operator should pay a great deal of attention and effort.

The high success rate of ossiculoplasty with GICs and HA were reported by many authors. In patients requiring revision of surgery, Geyer and Helms reported that half of the failures resulted from the displacement of prosthesis (8). However, there is no information about the character of bonding failures of bone cement ossiculoplasty. If the missing part of incus is very long, the re-built part could fail adhesively or cohesively over time. To overcome this problem, establishing more reliable and stronger bonding between the remainder of the long process of the incus and the head of the stapes is preferable, instead of using a total or partial ossicular chain reconstruction prosthesis. In this study, all GICs showed firmer and more compact structures than the HA in the short-term. While OH showed complete adhesive failure between the ossicles, HA, AC and KC were broken off mainly adhesive (60%) and mixed (40%). Only AM showed cohesive failure of the cement. That means that AM could prevent the ossicles from re-breaking during functioning.

According to the author's knowledge, this is the first study evaluating the bonding strength of GICs and HA to the ossicles. Nonetheless, there is still not enough knowledge in the literature about the optimum bonding criteria such as minimum bonding strength for short- and long-term success and vibration character of the prosthetic materials used during the sound transmission. Erosion of the incudostapedial joint is the most common ossicular defect encountered in chronic middle ear disease (20). However, behaviour of the re-built part under function has not been clarified. Several studies have reported the results of clinical performance of different GICs and bone cement by means of pre- and post-operative hearing quality for patients with various middle ear defects. However, there is no *in vitro* or *in vivo* comparative study about the bonding strength of different GICs or bone cement to the ossicles. Furthermore, there are also no stated numeric values about the bonding strength of prosthetic materials to the ossicles. Despite deep discussion on the choice of materials, Demir et al. suggested that the success rate does not depend on the type of replacement material but is extremely related to the pathophysiological status of the middle ear (3). Therefore, new comparative prospective clinical studies should be planned to verify whether the *in vitro* test results are compatible with the clinical conditions or not.

Ethics Committee Approval: Ethics committee approval was received for this study from the ethics committee of İnönü University.

Informed Consent: Written informed consent was obtained from patients who participated in this study.

Peer-review: Externally peer-reviewed.

Author contributions: Concept - M.T.K., İ.H.U., M.Y.; Design - M.T.K, İ.H.U.; Supervision - M.T.K.; Resource - M.T.K., İ.H.U., M.Y., M.A.M., A.T.O., F.M.H.; Materials - M.T.K.; Data Collection&/or Processing - M.T.K., İ.H.U., A.T.O.; Analysis&/or Interpretation - İ.H.U., M.Y., M.A.M.; Literature Search - M.T.K., İ.H.U., M.Y., M.A.M., F.M.H., A.T.O.; Writing - İ.H.U., M.A.M., F.M.H.; Critical Reviews - M.T.K., İ.H.U., M.Y., M.A.M., F.M.H., A.T.O.

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