





Analysis of enamel and material wear by digital microscope: an in-vitro study

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Abstract: The objective of the study was to analyze the surface area (SA) of the wear caused by simulated chewing on human enamel and opposing restorative material, namely: composite resin (CR), porcelain fused to metal (PFM), lithium disilicate (LD), or monolithic zirconia (MZr). Forty-eight premolars were selected as enamel specimens and divided randomly into 4 groups ($n = 48$; $n = 12$) used as antagonists in chewing simulation (250,000 loading cycles) against one of the four selected test materials. Enamel and material specimens were scanned and evaluated under digital microscope, and wear SA (mm^2) were recorded. Descriptive statistics, paired t-test, one-way ANOVA, and post-hoc Tukey-HSD tests were used for statistics ($p < 0.05$). The smallest and largest SA were exhibited by enamel against LD (0.80 mm^2) and PFM (1.74 mm^2), respectively. PFM (3.48 mm^2) showed the largest SA and CR (2.28 mm^2) showed the smallest SA. Paired t-test for SA values showed significant difference ($p < 0.05$) in all wear comparisons between materials and enamel antagonists. The wear of materials were greater than that of their respective enamel antagonists ($p < 0.05$). One-way ANOVA of the logarithmic means of wear SA revealed significant differences ($P < 0.05$). Post-hoc Tukey test revealed significance for PFM ($p < 0.05$) with other materials. Wear of all test materials was greater compared to the wear of enamel antagonists. PFM and LD caused the largest and the smallest enamel wear, respectively. CR, LD, and MZr are more resistant than PFM to wear after simulated chewing against enamel.

Keywords: Tooth Wear; Dental Restoration Wear; Tooth Attrition; Ceramics.

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Introduction

Tooth wear is an irreversible process of tooth surface loss having multifactorial etiologies. Many researchers have studied its associations with age, gender, bite force, bruxism, craniofacial morphology, mouth breathing, and malocclusion. It is a common multifactorial phenomenon resulting from direct contact between teeth, restorations, or prostheses during mastication or para-functional habits, effects of abrasive substances, or effects of acids from various sources. The condition progresses rapidly if the etiological factors are not identified and addressed promptly.¹ Tooth wear is becoming an ever-increasing problem as more natural teeth are retained in old age and is likely to continue increasing as patients' demands and expectations rise. The condition causes a myriad of clinical problems including enamel and dentin loss, hypersensitivity, loss of

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vertical dimension, temporo-mandibular disorders, impaired mastication, and compromised esthetics. The treatment for tooth wear is extensive, expensive, and complex. Early diagnosis and timely intervention can result in prevention of this otherwise progressive and destructive condition of the human teeth.²

The diagnosis and management of tooth wear presents a number of problems for dentists. It is complicated to identify and modify the etiological factors contributing to various forms of tooth wear and it is difficult to select appropriate restorative strategies.³ Demands for esthetic alternatives have led to an increased development of new restorative materials. The wear of human enamel and of the restorative material are often a functional and esthetic concern when selecting a material for clinical restorative treatment. Ceramic and composite restorations are known to cause various levels of wear of opposing enamel.^{3,4,5} Surface loss of natural dentition may get aggravated due to increased hardness and wear properties of opposing restorations as the rates of enamel wear of antagonist teeth may differ.⁴ Seghi et al. recommend the selected material to have a hardness degree similar to that of the enamel.⁵

Over the years, many in vivo and in vitro methods were developed to describe and quantify the extent of tooth and restorative material wear. Advancing technology and reproducibility within and between observers have been among the factors that contributed to the development of new methods. Each method has both advantages and disadvantages. The selection of the appropriate measurement method requires due consideration to the type of experiment, accuracy, time, and cost. Although numerous in vitro wear studies have been conducted on restorative materials, few studies have provided detailed information to characterize and predict the wear behavior of a range of materials.^{6,7}

Ceramics and composite resin (CR) are the most commonly used tooth-colored restorative materials; however, the abrasive effect of these materials against enamel is still a clinical concern. Several studies have demonstrated that in general, ceramic material causes greater enamel wear compared with any other restorative materials or enamel.^{8,9,10,11} Enamel wear caused by ceramics or CR is also

a multifactorial condition because the wear of a material is influenced by numerous factors, including contact geometry, surface roughness, microstructural features, grain size, fracture toughness, velocity, load, temperature, duration, environment, and lubrication.^{12,13} Lambrechts et al.,¹³ using clinical measurements, reported that enamel vertical wear was between 20-40 microns per year when opposing enamel in premolar and molar regions.¹³ The wear behavior of enamel and ceramics is different from that of metal and CR; enamel and ceramics wear through micro fracture mechanism and metal and composites wear by adhesion.¹⁴ The complexity of the wear mechanism and its measurement in the oral environment makes it difficult to conduct in vivo tooth wear studies. In order to overcome the difficulties of in vivo studies, in vitro methods such as wear simulators have been developed to study the wear behavior of dental materials. Advances in current technology have enabled simulation of the human chewing cycle in a laboratory using specific loads and frictional forces exerted by a chewing simulator and determination of the surface profile of worn materials by using advanced digital scanning methods.^{15,16,17,18}

The aim of this in vitro research study was to investigate and compare the surface area (SA) of wear facets caused by simulated chewing on human enamel and on opposing restorative materials, namely: composite resin (CR), porcelain fused to metal (PFM), lithium disilicate (LD), and monolithic zirconia (MZr). The null hypothesis tested was that enamel and its opposing restorative materials would show similar simulated chewing wear behavior.

Methodology

This in vitro research study was reviewed and approved by the Ethical Committee of the College of Dentistry Research Center, King Saud University, Riyadh.

Preparation of enamel specimens

A total of 48 maxillary first premolar teeth from patients aged between 18–25 years that were recently extracted for orthodontic purpose were used as enamel specimens. Only teeth with similar sizes and height by visual inspection and with sound and

non-abraded enamel over the cusps were selected, whereas teeth with worn-out cusps, fractured, or carious teeth were excluded. Teeth were collected immediately after extractions, cleaned, disinfected, and stored in water containing 0.05% thymol to simulate the intra-oral condition, and used as samples within 30 days. To overcome anatomical variations in the sizes and shapes of the teeth and to standardize the mounting in the wear simulation machine holder, each tooth was embedded in an acrylic resin mold (Ortho-Resin, DeguDent GmbH, Hanau, Germany) up to the level of cemento-enamel junction so that the buccal cusps remained prominent (Figure 1A). All 48 samples were distributed equally among the four groups by random draw method. The sample size per group ($n = 12$) was selected based on previous studies.^{18,19,20}

Fabrication of material specimens

Four commonly used restorative materials were selected as four test materials: CR (Nanohybrid Filtek Z250XT, 3M ESPE), PFM (Ivoclar Vivadent AG, Schaan), LD (IPS E-max, Ivoclar vivadent AG, Schaan), and MZr (Zolid FX preshade, Aman GIRRbach, Austria), with 12 samples per group. Twelve discs of each material measuring 10 mm diameter and 3 mm thickness were fabricated and glazed and/or polished according to the manufacturer's instructions (Table 1). For the PFM specimens, the thickness of the metal and veneering ceramic was kept at 1 and 2 mm respectively. Discs were cleaned in an ultrasonic cleanser for 10 min and embedded in an acrylic resin (Ortho-Resin, DeguDent GmbH, Germany) mold to facilitate locking the samples in the chewing simulation machine (Figure 1B).

Table 1. Tested materials with their polishing / glazing protocols.

Composite resin	Brand	Manufacturer	Polishing / Glazing system	Polishing / Glazing instructions
Composite resin	Nano hybrid Filtek® z250	3M ESPE Dubai United Arab Emirates	PoGo (Dentsply Caulk, Milford, USA)	Apply light intermittent pressure at moderate speed for 30 seconds
Porcelain fused to metal	Porcelain fused to metal	Ivoclar Vivadent AG Schaan (Principality of Liechtenstein)	IPS Classic Glazing material	Application of material with brush and firing at 900°C for 1 minute.
Lithium disilicate	IPS E-max®	Ivoclar Vivadent AG Schaan (Principality of Liechtenstein)	IPS E.max Ceram Glaze	Application of glazing paste and firing at 800°C for 1-2 minutes.
Monolithic zirconia	Zolid fx Preshade®	Amann GIRRbach (Koblach, Austria)	Zolid allbright diamond paste	Gentle polishing with Sinter State polishing kit for finishing zirconium restorations.

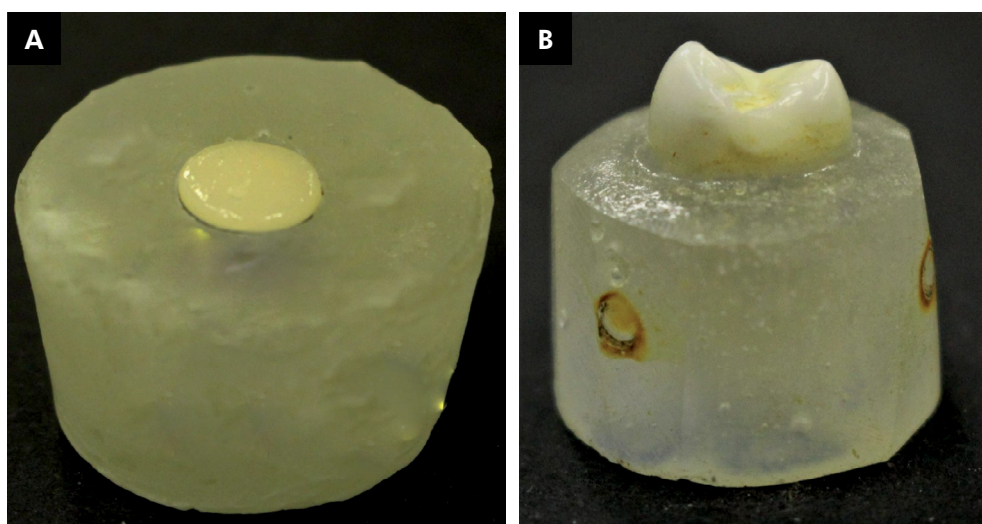


Figure 1. Material specimen embedded in a resin block (A); Tooth specimen embedded in a resin block (B).

Chewing simulation

The chewing simulation machine (Chewing Simulator, CS-4.8 professional line, SD Mechatronik GMBH, Westerham, Germany) was used to perform wear tests of enamel against material specimens. The machine has eight chambers that simulate the vertical and horizontal chewing movements simultaneously in a thermodynamic condition. Basically, the machine operates by exerting a vertical load from the enamel antagonist onto the material specimen, sliding horizontally and then repeating the cycle (Figure 2A). The enamel specimens embedded in resin were fastened with a screw in the upper sample holder and the material specimens embedded in resin were fixed in the lower sample holder (Figure 2B). According to previous studies, a weight of 5 kg is comparable to 49 N of chewing force and 250,000 loading cycles in a chewing simulator are comparable to approximately one year of chewing from a clinical perspective.^{18,19} Therefore, those parameters were used, accompanied by thermo-cycling with distilled water, which were unchanged. The specific parameters for this wear test such as cold bath temperature of 5°C, hot bath temperature of 55°C, dwell time of 60 s, vertical movement of 6 mm, horizontal movement of 0.3 mm, rising speed of 55 mm/s, forward speed of 30 mm/s, descending speed of 30 mm/s, backward speed of 55 mm/s, vertical load per sample of 5 kg, cycle frequency of 0.8 Hz, kinetic energy of $2,250 \times 10^{-6}$ J, and 250,000 loading cycles were adjusted to simulate the natural chewing function.^{18,19,20,21,22,23,24,25}

Wear scar SA evaluation

After the wear cycles, specimens were cleansed in ultrasonic bath to eliminate the debris or wear particles and dried. Then, each specimen was evaluated under digital microscope (HIROX, KH-7700, Digital microscope system, Tokyo, Japan) and post-wear digital photographs of the specimen's surfaces were taken at $\times 50$ magnification (Figure 3). While recording the digital photographs, every attempt was made to keep the wear surface parallel to the base of the microscope. For each enamel and material specimen, the SA was outlined and the SA in square micrometers (μm^2) of each wear surface was calculated digitally with the 3D Viewer Software (HIROX-USA, 100 Commerce Way, Hackensack, NJ, USA) programmed in the digital microscope (Figure 4). The readings of SA in μm^2 were converted to mm^2 for each specimen. To eliminate the inter-examiner variability, all the scans were recorded and wear facet areas traced twice by a trained technician.²⁵

Statistical analysis

The obtained data were tested statistically using SPSS software (Ver. 21.0, SPSS, Chicago, USA) to calculate means and standard deviations. Data were evaluated with Shapiro-Wilk test for normality and found to be normally distributed among all groups. The statistical analysis included descriptive statistics, comparisons of the mean difference post-wear cycle with paired t-test, comparisons of group means with one-way ANOVA, and multiple comparisons among the groups with Tukey-HSD post hoc test at a significance level of $p < 0.05$.

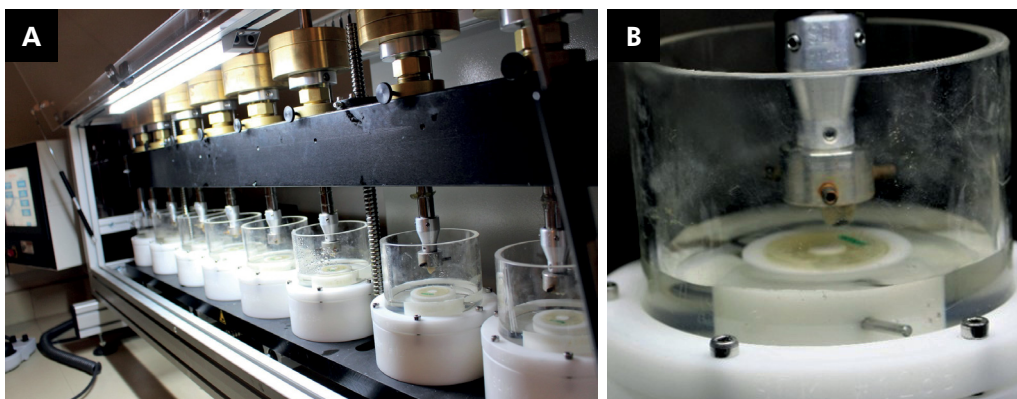


Figure 2. Wear simulator machine with specimens mounted for wear test (A); Close up view of specimens mounted in upper and lower holders (B).

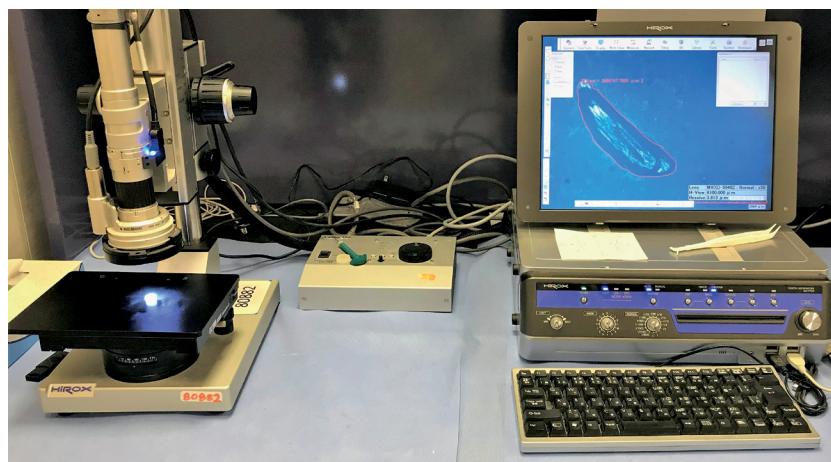


Figure 3. Digital microscope (HIROX, KH-7700) with mounted specimen for scanning.

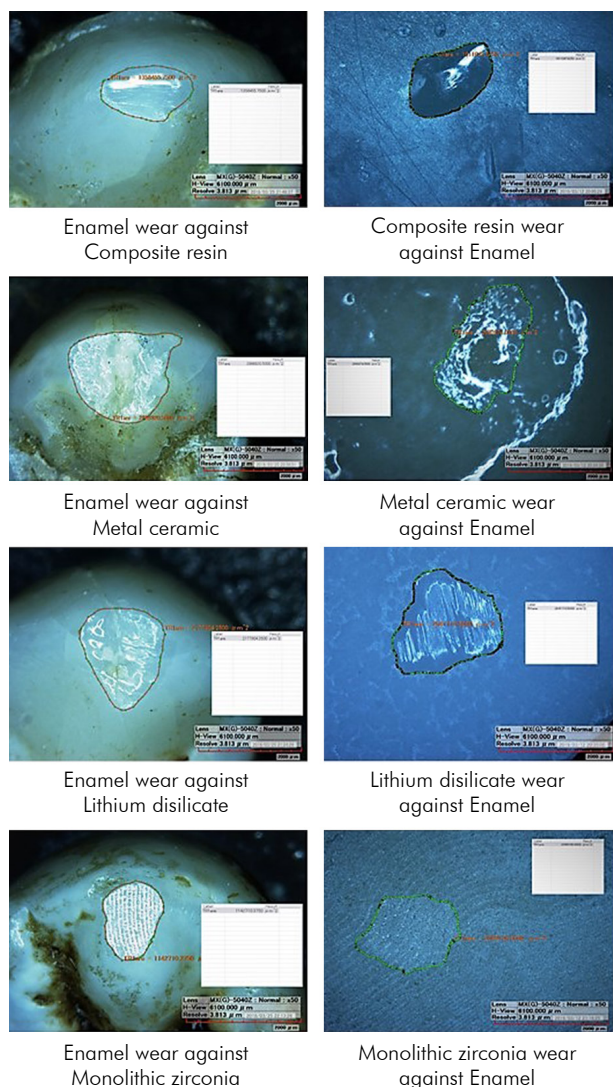


Figure 4. Post wear scans of enamel and material specimens and calculated wear facet surface areas (μm^2) using digital microscope.

Results

In this study, the SA of wear facets of four restorative materials and their antagonist enamel were measured using a digital microscope and compared.

The descriptive statistics and ANOVA results are presented in Table 2. Among the overall mean SA values of the tested enamel, the least wear was found for enamel specimens tested against LD ($0.80 \pm 0.54\text{mm}^2$) and highest wear was found for enamel against PFM ($1.74 \pm 0.66\text{mm}^2$). Among materials, PFM ($3.48 \pm 0.58\text{mm}^2$) showed the largest SA and CR ($2.28 \pm 0.54\text{mm}^2$) showed smallest SA. The least and highest standard deviations were observed for enamel against LD (0.54mm^2) and enamel against MZr (0.87mm^2), respectively. One-way ANOVA of the enamel SA means ($p = 0.001$) and materials ($p = 0.000$) revealed statistically significant differences. Comparison between the materials SA minus enamel SA revealed more wear for the material specimens than that for antagonist enamel specimens ($p = 0.000$).

Multiple comparisons among the material and enamel groups revealed statistically significant differences for PFM ($p < 0.05$) compared with the other materials, and LD showed statistically significant difference ($p < 0.05$) compared to enamel and the other materials, as presented in Table 3.

Table 4 describes the paired samples analysis, which revealed significant differences for the SA of each test material and their enamel specimens ($p \leq 0.05$).

Table 2. Descriptive statistics and ANOVA results of the wear surface areas of tested materials and antagonist enamel specimens.

Surface area	Mean	Standard deviation	Lower bound (95%CI)	Upper bound (95%CI)	Minimum	Maximum	*p- value
Enamel (n = 48)							
CR	1.67	0.07	1.63	1.72	1.60	1.80	
PFM	1.74	0.66	1.31	2.16	0.86	2.69	
LD	.80	0.54	0.46	1.15	0.18	1.95	0.001
MZr	1.63	0.87	1.07	2.19	0.14	3.11	
Total	1.46	0.70	1.26	1.67	0.14	3.11	
Materials (n = 48)							
CR	2.28	0.54	1.93	2.63	1.74	3.73	
PFM	3.48	0.58	3.10	3.85	2.89	4.41	
LD	2.66	0.16	2.55	2.76	2.48	2.92	0.000
MZr	2.66	0.66	2.24	3.08	1.96	4.32	
Total	2.77	0.67	2.57	2.96	1.74	4.41	
Material minus Enamel							
CR	0.60	0.58	0.23	0.97	0.06	2.13	
PFM	1.73	0.79	1.23	2.24	0.20	3.01	
LD	1.85	0.59	1.47	2.22	0.53	2.68	0.000
MZr	1.03	0.94	0.42	1.63	0.03	2.96	
Total	1.30	0.88	1.04	1.56	0.03	3.01	

*p-value is significant at $p < 0.05$.

Figure 5 shows the comparison of mean SA of enamel and materials tested. Enamel against PFM showed the highest (1.74 mm²) and enamel against LD (0.80 mm²) showed the smallest surface wear area. Amongst the materials, PFM had the highest (3.48 mm²) wear, and LD and MZr (2.66 mm²) had the least similar surface wear areas.

Table 3. Multiple comparisons among the tested materials by post hoc Tukey HSD test for wear differences.

Surface area	CR	PFM	LD	MZr
Materials				
CR	-	0.000	0.038	0.301
PFM	0.000	-	0.002	0.003
LD	0.308	0.002	-	1.000
MZr	0.301	0.003	1.000	-
Enamel				
CR	-	0.994	0.006	0.998
PFM	0.994	-	0.003	0.973
LD	0.006	0.003	-	0.011
MZr	0.998	0.973	0.011	-

*p-value is significant at $p < 0.05$.

Discussion

In this in vitro study, wear SA of enamel and four commonly used restorative materials was tested in a simulated oral environment using a chewing simulator machine. Test specimens of identical shape and size were used under the standardized testing conditions. The two-body wear of the chewing simulator provided a combined action of impact load and sliding that matched the natural mastication of one year of chewing function under normal chewing force.^{18,19}

The results of this study indicate that there is significant difference in the wear SA of the antagonist enamel as well as the materials tested. Thus, based on the results, the null hypothesis of similar wear among the materials and antagonist enamel after the wear simulation test was rejected. Previous studies in the literature have shown the difference in wear behavior of various restorative materials against natural enamel,^{16,17} but most of the wear tests provide only limited correlation with clinical data,^{19,20} even though they allow a comparative evaluation of different materials under standardized testing

Table 4. Results of paired samples statistics for comparison of material and enamel surface area for each test materials.

Test materials	Surface area (n = 12)	*Mean ± SD	Paired statistics		
			Correlations	Differences	**t-test
				*Mean ± SD	p-value
CR	Enamel	1.67±.07	-0.423	-0.60 ±.58	0.004
	Material	2.28±.54			
PFM	Enamel	1.74±.66	0.205	-1.73 ±.79	0.000
	Material	3.48±.58			
LD	Enamel	.80±.54	-0.138	-1.85 ±.59	0.000
	Material	2.66±.16			
MZr	Enamel	1.63±.87	0.270	-1.03 ±.94	0.003
	Material	2.66±.66			

*Mean surface area was calculated in mm²; **p-value significant at p ≤ 0.05.

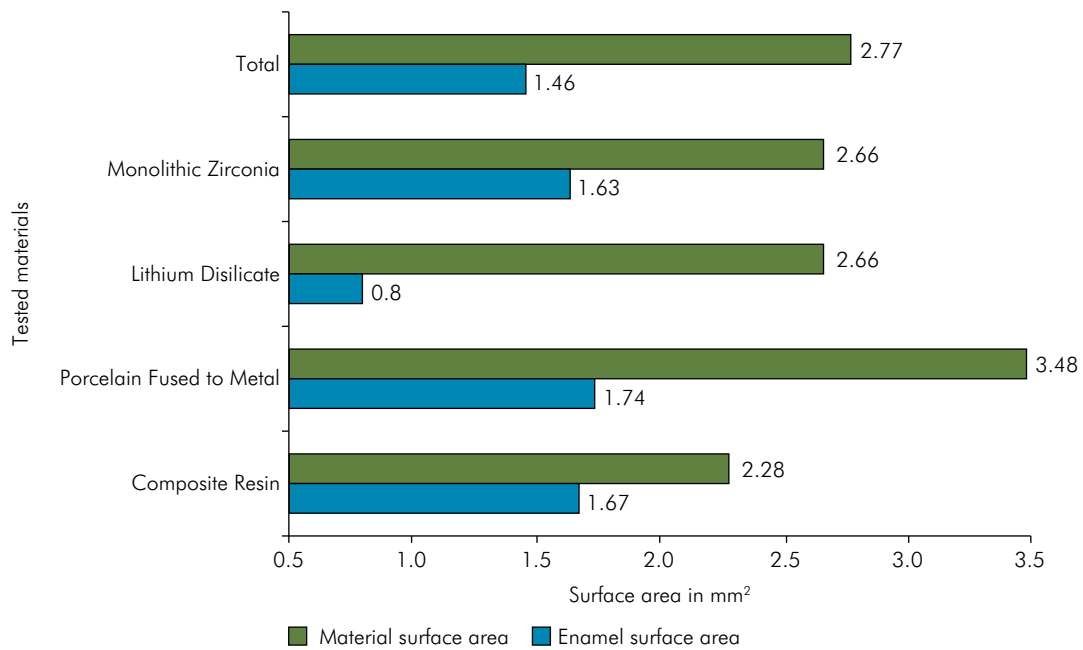


Figure 5. Graphical comparison of mean surface area of enamel and materials.

conditions.^{21,22} Therefore, testing conditions closely simulating clinical situations are preferred.^{23,24,25,26}

In this study, it was expected that continuous thermo-cycling with water would remove wear debris from the specimen’s surface; specimens were kept wet during the whole course of the test, which caused additional aging of the specimens. Enamel properties vary depending on the location in the tooth and histological structure.²⁷ Enamel on the cusps is stronger and withstands high occlusal

forces parallel to the direction of enamel rods.²⁷ In this study, only the cusp tip of tooth specimens were held in contact with material specimens during the wear test. Therefore, the interpretation of the results of this study should consider this factor.

A digital microscope was used to evaluate and measure the SA of wear facets as it has been recommended by a previous study.¹⁸ The microscope offered a good resolution and accurate measurements of the traced wear facets on enamel and material

specimens. A key advantage of digital microscopy is its ability to easily and quickly autofocus an image. The intuitive software of the microscope allows the user to automatically detect the optimal focus point.

According to the results of this study, PFM created the highest enamel wear, followed by CR and MZr, and LD showed the least wear of opposing enamel. The high wear of antagonist enamel by PFM has been also reported by Etman et al.²⁵ The wear mechanism of PFM is by fracture and creating sharp asperities on its surface that potentiate enamel wear by acting as a three-body wear.^{22,23} MZr was introduced to overcome the complications of chipping of veneer ceramics and to reduce wear of the opposing enamel,^{21,22,26} although the results of this study revealed high enamel wear caused by the MZr compared to the other tested materials. The possible reasons for high enamel wear caused by PFM and MZr could be related to the high surface hardness values of veneer ceramics (420 HV) and zirconia (1,250 HV), respectively. In addition, the removal of the glaze layer and exposure of the underlying rough surface during wear cycles can potentiate the wear mechanism and cause more abrasion of the opposing enamel.^{28,29}

One of the findings of this study is that CR caused enamel wear comparable to PFM and MZr. The possible reasons for nanohybrid CR causing high wear of the opposing enamel could be related to its high surface roughness and microhardness levels attributed to filler content.³⁰ The degree of wear is affected by surface microstructure, material roughness, and environmental factors.³¹ In contrast to other ceramics, LD exhibits a unique microstructure, composed of 70% of small interlocking, randomly oriented LD glass crystals. These glass crystals cause micro cracks to deflect, branch, or blunt reducing their propagation and resulting in less abrasiveness. This property of LD may be attributed to least wear SA of antagonist enamel.³²

When comparing the overall SA mean of materials and antagonist enamel it was evident that PFM, LD, and MZr wear were highest and CR was lowest compared to their antagonist enamel. These differences could be related to the different composition and microstructure of restorative materials and enamel.²⁷

The wear of composites depends mainly on the content and the size of filler particles in the matrix.³⁰ The CR tested in this study (Filtek Z250XT) contains nanohybrid filler particles that improve wear resistance because of less inter-particle spaces and a more protected resin matrix. This resulted in decreased wear, most probably due to better load transfer between matrix and filler.²⁹

Another possible explanation for the differences in the wear behavior of the various restorative materials tested could be the different effects of hydrothermal aging within the chewing simulation machine. This could affect occlusal wear, as LD (glass-ceramics) and PFM (dental porcelains) are susceptible to slow crack growth³³, MZr (zirconia) to low temperature degradation³⁴, and CR (dental composites) to hydrolytic degradation³⁵.

In this study, the variation in the results could be attributed to the inhomogeneity in enamel antagonists. Human tooth tissues show variations in physical properties, histological structure, and thickness of enamel layers at different locations within the same tooth as well among the different tooth types.²⁶ In vitro wear studies have some inherent shortcomings; they evaluate only one or two wear mechanisms under limited chewing simulation conditions. Ideally, simulation tests should present clinical conditions of enamel antagonists. However, morphological and structural differences of enamel complicate standardizing wear testing leading to high variations in the obtained data. Despite being simple, the digital photographs taken for the measurement of SA recorded the flat area of the wear scar. Due to these limitations, the results of the current study should be interpreted with caution.

Conclusions

Within the limitations of this study it can be concluded that:

- a. The wear area of all the tested materials was higher compared to their respective antagonist enamel specimens.
- b. Among the tested materials, PFM and LD caused the highest and least enamel wear, respectively.

- c. CR, LD, and MZr were more resistant to wear against enamel, whereas PFM had higher wear against enamel.
- d. The clinical implication of the study is that wear properties of restorative materials should be considered while selecting the appropriate material in clinical situations.

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