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Article

Characterization of properties for modern dental materials and bordering tissues. Part 1. Mechanical properties

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Abstract. In the present paper, an *ex vivo* nanoindentation study of the mechanical properties of fillings made of composite resin and glass ionomer cement, as well as infiltrated enamel and tissues in their vicinity, was carried out, followed by a comparison of the results with the properties of the sound tissues. For a more in-depth interpretation of the obtained experimental data, optical images of the sample surface structure were obtained. Composite fillings have been shown to be superior to glass ionomer ones due to greater similarity in mechanical properties to the sound enamel and fewer internal structure artifacts. The possibility of the polymerization stress appearing in dentine adjacent to the filling was demonstrated. Pathologically altered demineralized enamel treated with polymer infiltrant, despite a slight decrease in the values of properties compared to sound tissue, turned out to be generally close to it both in terms of the mechanical characteristics, which indicates the high potential for the use of polymer infiltration in dental practice for the treatment of early caries.

Keywords: tooth, enamel, dentine, composite resin, glass ionomer cement, polymer infiltrant, filling, nanoindentation

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Характеризация свойств современных стоматологических материалов и тканей в их окрестности. Часть 1. Механические свойства

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Аннотация. В настоящей работе было проведено *ex vivo* исследование механических свойств пломб из композитного материала и стеклоиономерного цемента, а также инфильтрированной эмали и тканей в их окрестности с использованием наноиндентирования с последующим сравнением результатов со свойствами здоровых тканей. Для более подробной интерпретации полученных экспериментальных данных были получены снимки структуры поверхности образцов с использованием оптического микроскопа. Исследование показало, что пломба из композитного материала имеет ряд преимуществ перед стеклоиономерным цементом ввиду большего сходства её механических свойств со свойствами здоровой эмали и меньшего количества артефактов внутренней структуры. Показана возможность возникновения полимеризационных напряжений в дентине в окрестности пломбы. Патологически изменённая деминерализованная эмаль, обработанная полимерным инфильтратом, несмотря на некоторое снижение значений свойств по сравнению со здоровой тканью, оказалась в целом близка к ней по механическим характеристикам, что свидетельствует о высоком потенциале применения полимерной инфильтрации в стоматологической практике при лечении раннего кариеса.

Ключевые слова: зуб, эмаль, дентин, композитный материал, стеклоиономерный цемент, полимерный инфильтрант, наноиндентирование

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Introduction

The state of the tissues of the human oral cavity is an important component of the health of the body as a system, significantly affecting quality of life in general. At the same time, untreated caries of permanent and primary teeth is one of the most common diseases in the world — according to the global report of the World Health Organization, it affects about 2 billion 510 million people all over the world [1]. Moreover, a number of studies have shown that pathological changes in the tooth hard tissues contribute to the development of respiratory diseases [2], lower respiratory tract infections and influenza [3], as well as a number of other diseases [4].

Caries occurs as a result of the interaction of acidogenic oral bacteria with components of the human diet within the plaque biofilm, resulting in the formation of organic acids, mainly lactic acid. The appearance of acids in the oral cavity can also occur when consuming a number of foods and drinks [5, 6], which contributes to the demineralization process — partial dissolution of the main structural elements of enamel — hydroxyapatite crystallites [7]. Early caries (stage of the white spot lesions, WSLs) is characterized by the process of demineralization of tooth enamel without cavitation [8]. A dental clinician plans a treatment of WSLs depending on a list of factors (the size of the demineralized area, its location, degree of activity, and others). With early manifestations of WSLs, treatment may be limited to the methods of non-invasive dentistry [9]. In more complex cases, it becomes necessary to use methods of preparing the area of demineralization and filling it or minimally invasive treatment (such as polymer infiltration [10]). Glass ionomer cements (GICs) can be used in a wide range of clinical situations due to the ability to control their mechanical properties by changing the powder/liquid ratio or the chemical composition of the material [11]. Treatment of WSLs by the infiltration method consists of acid etching of the damaged enamel surface, drying of the subsurface porous area, and impregnation of it with liquid polymer material, which modifies the microstructure of the enamel prisms [12]. However, the efficacy of this clinical procedure has not been fully investigated: in a number of clinical cases, high efficiency has been reported [13], whereas in other studies, it did not show the desired result for the practicing dental clinician [14].

In the present work, an *ex vivo* study of the mechanical properties of fillings made of the composite material and the GIC, the enamel surrounding these fillings, dentine in their vicinity (i.e., dentine adjacent to the dentine-enamel junction, DEJ), as well as sound dentine and enamel on the opposite side of the tooth was carried out. According to a similar scheme, a study was made of the properties of enamel modified by the infiltrant and dentine in its vicinity, as well as sound dentine and enamel on the opposite medial side of the tooth (the procedure was repeated on two samples each demonstrating the manifestations of WSLs in order to gather more statistical data).

1. Materials and methods

Four human molars were extracted for orthodontic reasons in the dental department of the Rostov State Medical University clinic, Rostov-on-Don, Russia. The local independent ethical committee of the university approved the research (extract 14/21 dated September 23, 2021), and the patients provided informed consent. The following dental materials were used in the study according to the manufacturers' protocols: Vitremer glass ionomer cement (3M ESPE, St. Paul, USA), Estelite Flow Quick composite material (Tokuyama Dental, Tokyo, Japan), and Icon infiltrant (DMG Chemisch-Pharmazeutische, Berlin, Germany).To form thin sections of the surface of the samples containing the areas under study, they were prepared in longitudinal section using an Isomet 4000 precision saw (Buehler, Lake Bluff, USA), then ground and polished.

The mechanical properties of the areas under study were assessed using a nanoindentation test machine (NanoTest 600 Platform 3, Micro Materials, Wrexham, UK) using a calibrated Berkovich diamond indenter. In each test, the load increased linearly for 20 s, remained constant for 30 s, then decreased linearly for 20 s. The maximum load Pmax was 50 mN. To prevent damage to the tooth tissues caused by uncontrolled dehydration, the specimens were kept moist with drops of distilled water using an infusion pump (Terufusion TE-332, Terumo, Leuven, Belgium). Reduced Young's modulus E_r and indentation hardness H for each of the areas were obtained utilizing the Oliver — Pharr method [15]. In nanoindentation experiments, creep meant a change in the deformation of the material during the experiment when the maximum load was held for 30 s. For each area under study, from 8 to 12 identical indentations were made, and the results were averaged afterwards.

Microscopy of fillings and tissues in the vicinity of indenter prints was carried out using the optical system of a nanoindentation system. Overview images of the prepared cross-sections were made using a Zeiss StereoDiscovery V.20 stereomicroscope (Carl Zeiss Microscopy GmbH, Oberkochen, Germany) according to the Abbe scheme. Zeiss ZEN software (Carl Zeiss Microscopy GmbH, Oberkochen, Germany) was used for image processing.

The Shapiro — Wilk normality test was used to examine whether the nanoindentation datasets were normally distributed. The test statistic (D) provided a measurement of the divergence of the dataset distribution from the normal one. The one-way analysis of variance (ANOVA) was used to detect statistically significant differences between the means of the two or three groups under study. Specifically, the null hypothesis was tested: $H_0 : \mu_1 = \mu_2 = \cdots = \mu_k$, where μ is a group mean and k is a number of groups. The above hypothesis was tested for three sets of indentation data for the composite filling case, three sets for the glass ionomer filling case, and two sets for each polymer infiltration case using the F-ratio at a significance level of $\alpha = 0.05$. The Tukey – Kramer test was then used to identify specific groups that differed from each other.

2. Results and discussion

Figure 1 shows optical images of surface sections of all samples, indicating both the areas of interest and the anatomical features of the crowns. Indenter imprints at a distance of 60 μm from the interface with the filling, in its internal and external parts (the central region of the enamel in Fig. 1, *a*, the approximate boundaries of the filling are outlined by a burgundy dotted line), demineralized dentine, and sound tooth tissues are shown in Fig. 2. The results of the property evaluation are given in Table 1.

Figure 2, *b* shows large pores found on the examined surface of the composite filling. Diagrams of the dependence of the indentation depth on the applied load for the case of this filling are shown in Fig. 3. For the GIC filling (Fig. 1, *b*) from the cervical area to the pulp horn, an assessment was made of its mechanical properties, enamel at the border with the filling (about 400 μm from the interface due to the presence of a crack in the immediate vicinity), bordering dentine, as well as the sound enamel and dentine on the opposite medial side of the tooth. The measurement results are shown in Table 2.

The results of optical microscopy of the interface of a filling made of GIC and dentine are shown in Fig. 2, *c*, *d*. Diagrams of the dependence of the indentation depth on the applied force for the case of a GIC filling application are shown in Fig. 4.

Considering that in the current research, polymer infiltration was carried out on the two samples, further, the first and second cases of using this material will be mentioned.

For the first case of the WSL infiltration (Fig. 1, *c*), the mechanical properties of the treated enamel, dentine in its vicinity, as well as sound enamel and dentine on the opposite medial side of the tooth were assessed.

A visual examination of the infiltrated enamel showed the presence of a WSL focus, extending from the enamel surface to the DEJ. In this regard, the mechanical properties of the infiltrated enamel were measured close to the DEJ. On the opposite side of the junction, presumably pathologically altered dentine was examined. The test results are given in Table 3. Diagrams of the dependence of the indentation depth on the applied force for the first case of the WSL infiltration are shown in Fig. 5. For the second case of polymer infiltration of the molar in the upper third of the enamel in height (Fig. 1, *d*), the mechanical properties were assessed similarly to the first case (see Table 4).

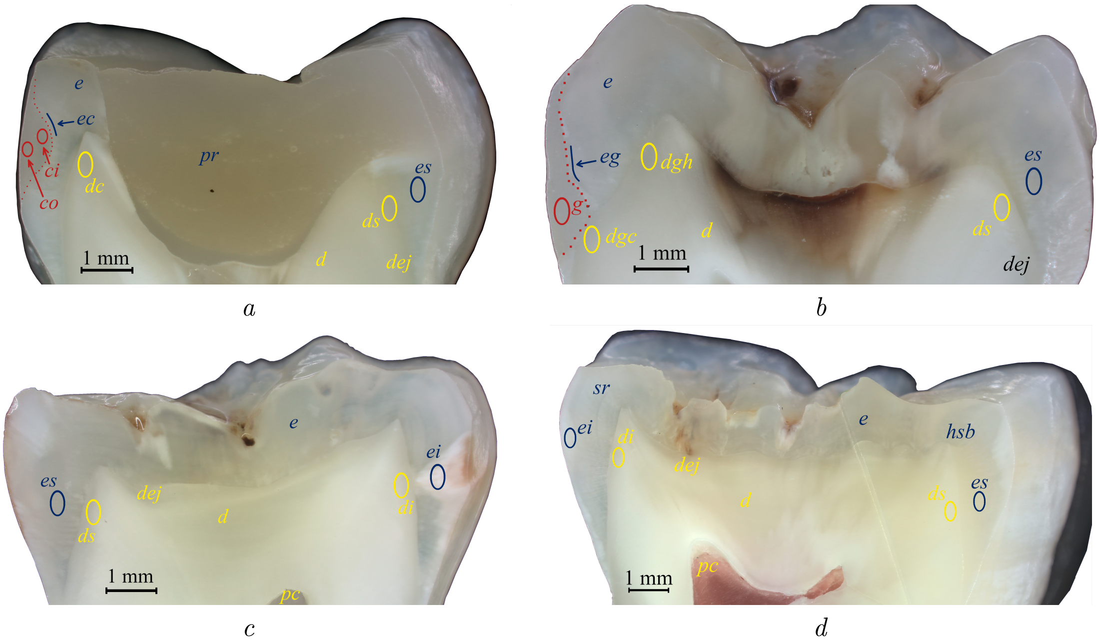


Fig. 1. Optical microscope images of samples after *ex vivo* dental treatment: *a* shows composite filling; *b* shows GIC filling; *c* shows enamel infiltration, first case; *d* shows enamel infiltration, second case; *e* stands enamel, *d* stands for dentine, *dej* stands for DEJ, *es* stands for sound enamel, *ds* stands for sound dentine, *ci* stands for internal part of the composite filling, *co* stands for external part of the composite filling, *ec* stands for enamel at the interface with the composite filling material, *dc* stands for dentine in the vicinity of a filling made of composite material, *pr* stands for previous crown restoration, *g* stands for GIC filling, *dgc* stands for dentine in the vicinity of a filling of GIC in the cervical region, *eg* stands for enamel on the interface of a filling of GIC, *dgh* stands for dentine in the vicinity of the GIC filling in the area of the pulp horn (crown part), *sr* stands for stria of Retzius, *hsb* stands for Hunter-Schreger bands, *ei* stands for infiltrated enamel, *di* stands for dentine in the vicinity of infiltrated enamel, *pc* stands for pulp chamber (color online)

Table 1

Mechanical properties of the sample treated with the composite material

Area	Red. Young's modulus E_r, GPa	Hardness I, GPa	Creep, nm
Filling outer part	11.47 ± 1.05	1.11 ± 0.17	236.3 ± 56.1
Filling inner part	11.51 ± 0.32	0.93 ± 0.05	293.5 ± 30.0
Enamel bordering filling	40.55 ± 3.85	2.48 ± 0.65	229.7 ± 65.7
Sound enamel	97.77 ± 5.50	5.98 ± 0.37	36.5 ± 11.1
Dentine bordering filling	23.50 ± 2.82	1.16 ± 0.13	162.3 ± 33.7
Sound dentine	26.48 ± 3.34	1.14 ± 0.15	101.5 ± 15.4

To assess statistically significant differences in the indentation results, three sets of Young's modulus values were selected for the composite material and GIC (the inner part of the composite filling, the enamel in the vicinity of the filling, and sound enamel with sample sizes of 11, 9, and 9 tests, respectively; the GIC filling, the enamel in the vicinity of the GIC, and sound enamel with sample sizes of 8, 6, and 9 tests, respectively), and two sets of values for the samples treated with the infiltrant (treated and sound enamel with sample sizes of 7 and 11 tests for the first sample, 12 and 12 tests for the second sample, respectively). In all cases, from the sample information, we obtained that the test statistic F was not in the 95% acceptance region, based on which a conclusion was made to reject the null hypothesis (at $p < 0.05$), thus, at least two group mean values for each material were statistically significantly different from each other. Additionally, Tukey – Kramer test subsequently revealed a significant difference between all three pairs for filling materials.

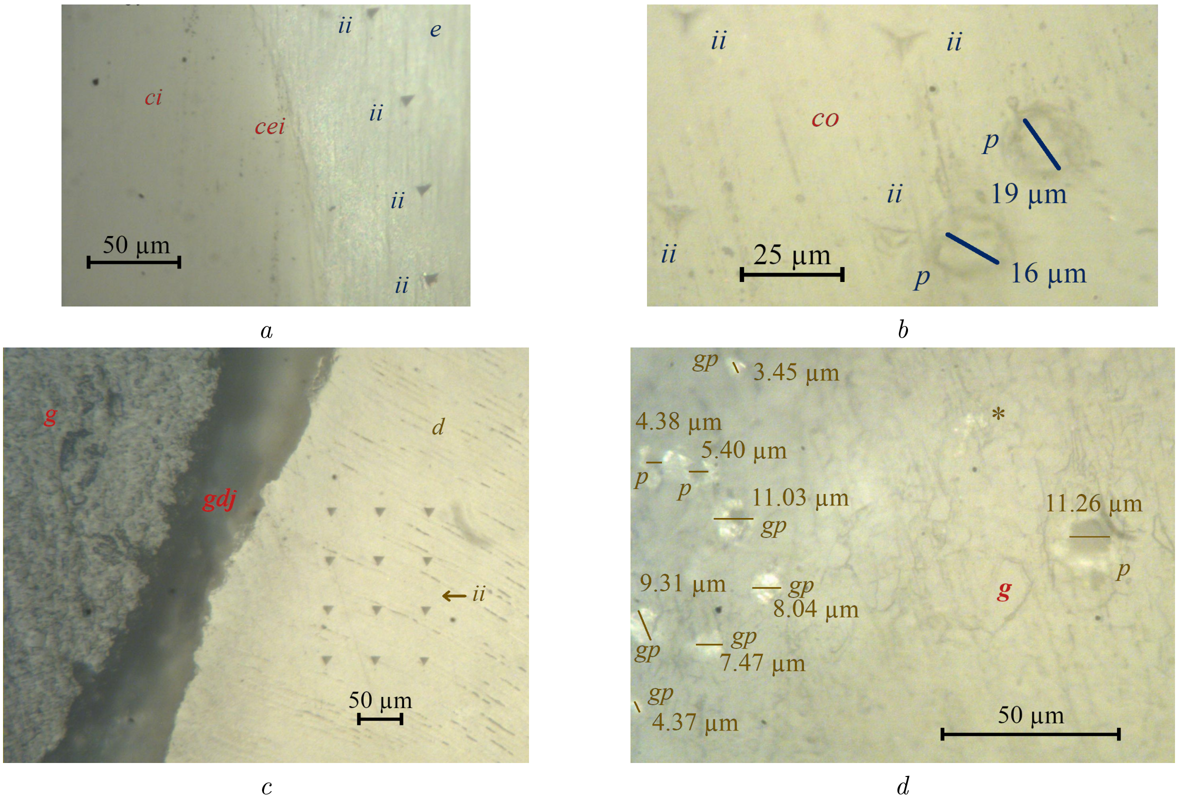


Fig. 2. Optical microscopy of fillings and surrounding tissues: *a* shows indenter marks on the surface of a composite filling; *b* shows indenter marks and pores in the outer layer of the filling; *c* shows indenter marks on dentine in the immediate vicinity of the filling; *d* shows surface of the GIC filling; *g* stands for GIC filling surface, *e* stands for enamel, *ci* stands for inner layer of the filling, *co* stands for outer layer of the filling, *ii* stands for indenter imprint, *cei* stands for interface between the filling and enamel, *p* stands for pore, *d* stands for dentine, *g* stands for GIC, *gdj* stands for interface between filling and dentine, *gp* stands for glass particle; symbol $*$ marks the agglomerate of glass microparticles (color online)

Table 2

Mechanical properties of the sample treated with the GIC

Area	Red. Young's modulus E_r, GPa	Hardness I, GPa	Creep, nm
Filling	1.57 ± 0.34	0.14 ± 0.05	927.0 ± 159.5
Enamel bordering filling	16.27 ± 7.51	1.25 ± 0.93	502.5 ± 285.5
Sound enamel	91.18 ± 4.89	5.61 ± 0.99	49.5 ± 27.7
Dentine (pulp horn)	39.37 ± 0.96	2.03 ± 0.07	116.3 ± 9.0
Dentine (cervical area)	25.36 ± 1.04	1.48 ± 0.10	182.9 ± 25.3
Sound dentine	23.33 ± 4.15	1.14 ± 0.01	124.6 ± 45.9

The analysis of experimental data of nanoindentation shows that the outer and inner layers of the composite filling are characterized by practically identical values of the Young's modulus and a decrease in the value of indentation hardness of the inner layer by 16.2% compared to the outer one, as well as an increased creep value by 24.2%. This observation partially agrees with the studies of the mineral density of composite fillings [16]. At the same time, the values of the reduced Young's modulus and the indentation hardness of the inner part of the filling are significantly lower than both the surrounding enamel, noted in Fig. 2, *a* (by 3.5 and 2.7 times, respectively), and sound enamel (by 8.5 and 6.4 times, respectively). The observed features are important from a practical point of view for creating a strong filling and high adhesion to natural enamel. For the enamel in the filling area, both the reduced Young's modulus and the indentation hardness were equal to 41.5% of the similar characteristics for sound enamel, and creep was 6.3 times higher, practically not inferior in creep value to the outer part of the composite filling (Fig. 2, *b*).

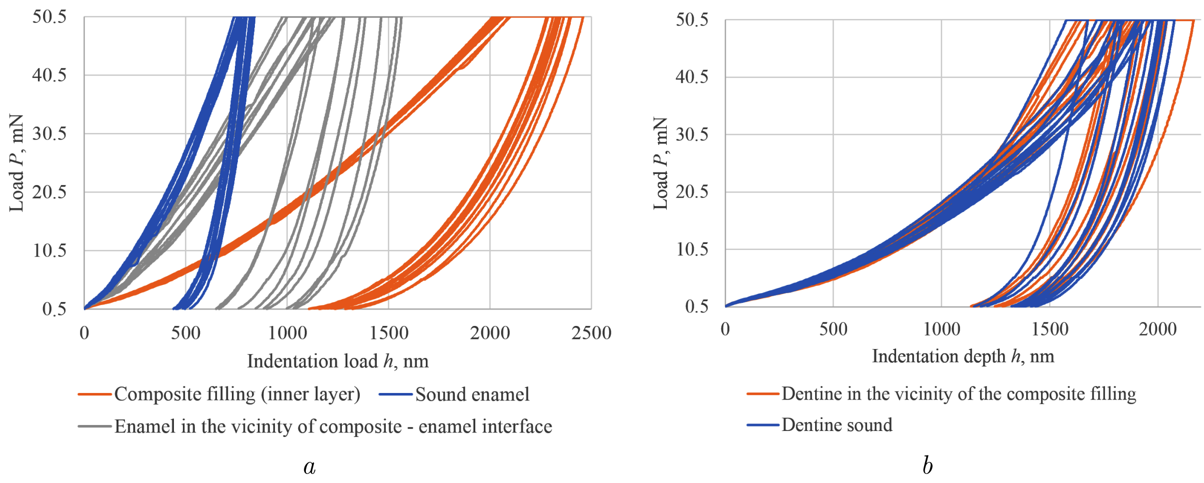


Fig. 3. Diagrams of the dependence of the indentation depth on the applied force for the case of a composite filling: *a* shows composite filling, enamel in its vicinity, and sound enamel; *b* shows dentine in the vicinity of the composite filling and sound dentine (color online)

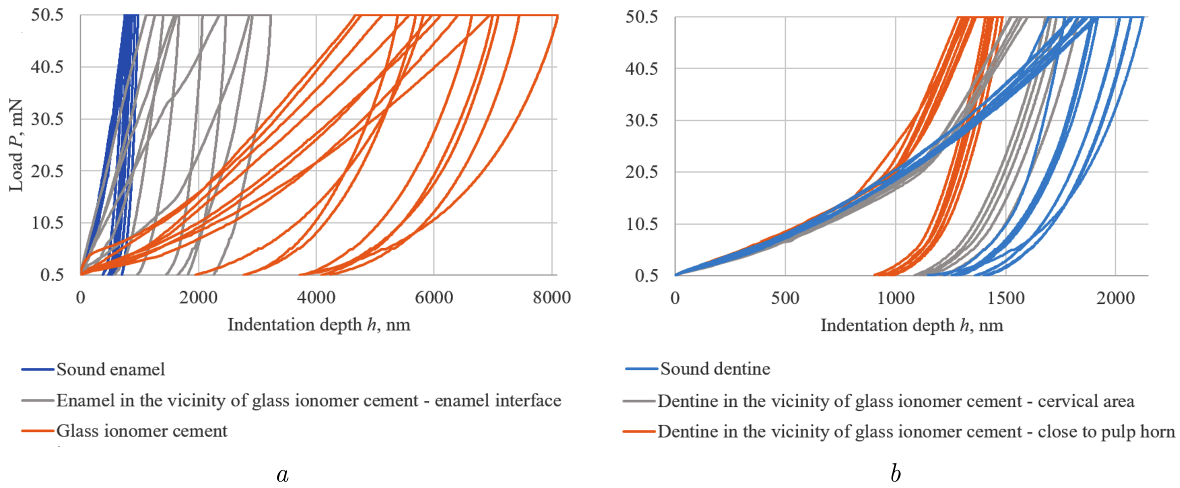


Fig. 4. Diagrams of the dependence of the indentation depth on the applied force for the case of a GIC filling: *a* shows GIC, enamel in its vicinity, and sound enamel; *b* shows dentine in the vicinity of the GIC filling close to the cervical area and to the pulp horn, sound dentine (color online)

Table 3

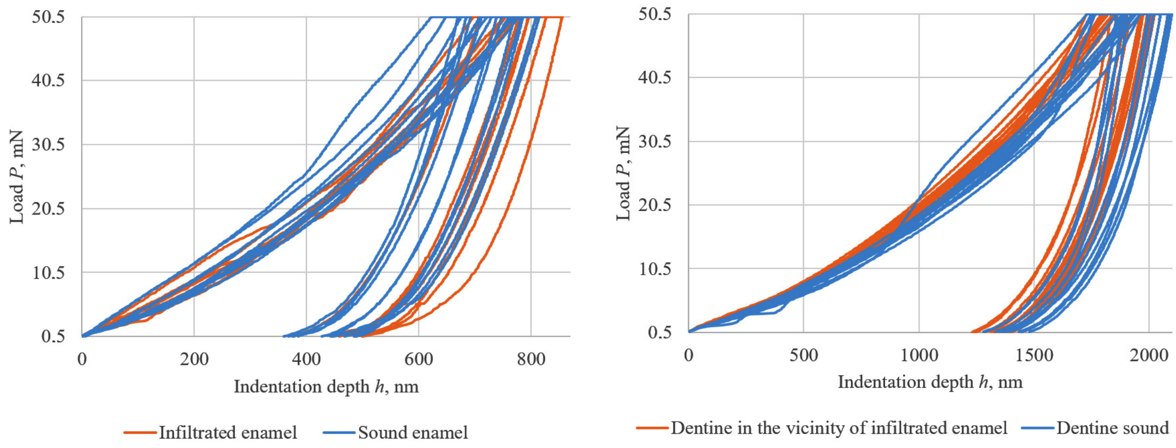
Properties of the sample treated with the infiltrant — first case

Area	Red. Young's modulus E_r, GPa	Hardness Í, GPa	Creep, nm
Infiltrated enamel	94.54 ± 6.53	6.32 ± 0.95	42.7 ± 27.8
Sound enamel	107.67 ± 11.89	7.02 ± 0.90	29.0 ± 11.9
Dentine bordering infiltrated enamel	23.38 ± 1.76	1.16 ± 0.07	119.4 ± 241.0
Sound dentine	26.38 ± 5.60	1.07 ± 0.07	132.1 ± 26.0

Table 4

Properties of the sample treated with the infiltrant — second case

Area	Red. Young's modulus E_r, GPa	Hardness Í, GPa	Creep, nm
Infiltrated enamel	77.9 ± 3.99	4.90 ± 0.55	58.5 ± 10.9
Sound enamel	88.84 ± 4.88	6.62 ± 1.32	42.0 ± 21.2
Dentine bordering infiltrated enamel	19.37 ± 2.89	1.09 ± 0.17	204.0 ± 38.6
Sound dentine	21.22 ± 1.62	1.25 ± 0.17	152.2 ± 21.9



a *b*

Fig. 5. Diagrams of the dependence of the indentation depth on the applied force for the first case of WLS infiltration: *a* shows infiltrated and sound enamel; *b* shows dentine in the vicinity of the infiltrated enamel and sound dentine (color online)

Observations of the dependence of the indentation depth on the applied force ($P-h$ diagrams) show that the nature of both the loading and unloading branches of the curves for enamel at the border with the filling is generally similar to that for the sound enamel, but an increase in the indentation depth is observed. These facts indicate a change in the mechanism of resistance to loads of such enamel, despite the fact that during optical microscopy, no visual signs of pathological changes were noted (homogeneous tissue, without color changes and distinguishable cavities [17], Fig. 1, *a*). At the same time, a small number of large defects in the form of pores with a diameter of up to 19 μm are observed on the surface (this pore is shown in Fig. 2, *b*). Similar effects are observed for dentine: its Young's modulus in the filling area decreases by 11.3%, and creep increases by 59.9% in relation to the sound tissue, while the hardness values are comparable.

As for filling materials made of GIC, extremely low values of the reduced Young's modulus and indentation hardness are obvious: they are significantly lower than those of the surrounding (10.4 and 8.9 times, respectively) and sound enamel (58.1 and 40.1 times, respectively). At the same time, the creep of GIC was 1.84 times higher than that of the surrounding enamel and 18.7 times higher than that of sound enamel. Such a low value of mechanical characteristics is consistent with the recent results obtained by Petrović et al. [18] during nanoindentation of a number of commercial GICs. It should be noted that at the boundary between the filling and enamel, as well as between the filling and dentine, there was a delamination of the interface (more than 50 μm wide, Fig. 2, *c*), presumably caused by an increased stress concentration due to an excessive difference in the mechanical characteristics of the filling and the surrounding tissue (which can be further studied using the approaches of [19]). Analysis of optical images showed a high content of glass particles on the cement surface with a diameter of 6.68 ± 2.70 μm, as well as pores reaching 15.12 μm, and agglomerates of glass microparticles (Fig. 2, *d*).

For the GIC filling, an even smaller angle of inclination of the unloading branches of the $P-h$ diagrams (Fig. 4) relative to the abscissa axis and a greater spread in depth (due to the heterogeneity of the surface and high porosity) is characteristic compared to the case of the composite filling. The study of dentine in the vicinity of the GIC filling (Fig. 4, *b*) was carried out in two areas: near the pulp horn and in the cervical area. It is noteworthy that, unlike demineralizede dentine or dentine around carious enamel, the values of the reduced Young's modulus and indentation hardness in both areas of dentine in the vicinity of the GIC were higher than the values of the sound tissue: in the cervical area by 8.7% and 29.8%, and near the pulp horn by 68.8% and 78.1%, respectively. Presumably, this phenomenon is associated with tissue deformation caused by polymerization stresses inside the GIC [20, 21]. This observation is consistent with the results of Dias et al. [22], in whose work increased hardness was observed in all areas of direct contact with the GIC in sound and demineralized dentine.

The nanoindentation data of the first case of using a polymer infiltrant show that, despite the lower reduced values of Young's modulus and indentation hardness of the infiltrated enamel compared to sound tissue (by 12.2% and 10.0%, respectively), as well as an increased creep value (by 47.2%), both branches of the $P-h$ diagrams for the pairs "infiltrated enamel — sound enamel" have a similar character, including the angle of inclination (Fig. 5, *a*). As for dentine, we note lower values of the reduced Young's modulus and creep of this tissue in the vicinity of the infiltrated enamel (by 11.4% and 9.6%, respectively), as well as a higher hardness value (by 8.4%) compared to sound dentine (Fig. 5, *b*).

For the second case of using a polymer infiltrant, lower values of the reduced Young's modulus and indentation hardness of the infiltrated enamel relative to the sound tissue were recorded (by 12.3% and 26.0%, respectively), as well as a higher creep value (by 39.3%). The unloading branches of the $P-h$ diagrams for the pairs "infiltrated enamel — sound enamel" demonstrate a similar character, including the angle of inclination, while the loading branches of the infiltrated enamel have a smaller angle of inclination and a noticeable increase in the depth of indentation. In this sample, the behavior of dentine largely repeats the features of the enamel.

Conclusion

In this work, an *ex vivo* study of the mechanical properties of composite and GIC fillings, as well as infiltrated enamel and tissues in their vicinity, was conducted, followed by a comparison of the results with the corresponding properties of the sound tissues. The results showed that a composite filling appears more preferable for use in dentistry than a GIC filling due to greater similarity of mechanical properties, a lower probability of adhesion loss at the enamel border, and a smaller content of internal structure artifacts.

The information collected for both samples treated with the infiltrant allows us to draw several conclusions:

– the mechanical properties of enamel after infiltration are closer to the properties of the sound enamel, in contrast to both types of filling materials;

– given the small difference in the mechanical properties of infiltrated enamel and the sound tissue surrounding it, the probability of undesirable stress growth in the infiltrated DEJ is lower than in the case of fillings.

However, further research is needed to better understand the ability of the infiltrant to penetrate tissues in different areas of the tooth and at different stages of caries progression.

The continuation of the article will be published in the next issue.

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