

Enzymatic Pretreatment Of Enamel And Dentin As A Stage In Optimizing The Adhesive Protocol For Esthetic Restoration

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Abstract: Modern esthetic dentistry is based on the principles of minimally invasive preparation, preservation of hard dental tissues, and the use of adhesive technologies that provide a strong bond between restorative material and enamel and dentin. Despite substantial advances in adhesive systems, the problem of adhesive interface durability remains relevant, especially in the restoration of anterior teeth, where mechanical stability, marginal adaptation, color integration, and prevention of postoperative sensitivity are simultaneously important. One of the factors limiting the effectiveness of the adhesive protocol is the presence of the smear layer, organic residues, collagen matrix, and surface contaminants that can impair the penetration of adhesive monomers and the formation of a complete hybrid layer.

In recent years, enzymatic pretreatment of enamel and dentin using proteolytic enzymes, particularly bromelain and papain, has attracted increasing interest. Data from international studies show that bromelain, at certain concentrations, can increase the strength of the adhesive bond and improve the marginal adaptation of restorative material, whereas the results of papain application remain more variable and depend on concentration, type of adhesive system, dentin depth, and the sequence of clinical steps.

Keywords: Enzymatic treatment, enamel, dentin, adhesive protocol, esthetic restoration, bromelain, papain, smear layer, hybrid layer, marginal adaptation.

Introduction: Esthetic restoration of anterior teeth is one of the leading areas of modern operative dentistry, since restoration of anatomical shape, color, translucency, and optical properties of the tooth is directly related not only to function but also to the patient's quality of life. In clinical practice, direct composite restorations are widely used for enamel and dentin defects, discoloration, traumatic injuries, diastemas, fractures, non-carious lesions, and the need to correct the shape of anterior teeth. However, the long-term success of such restorations is determined not only by the quality of the composite material but also by the stability of the adhesive bond at the "dental tissue - adhesive - restorative material" interface [4].

Over recent decades, adhesive dentistry has undergone considerable development: from multistep etch-and-rinse systems to self-etch and universal adhesives. Modern universal adhesive systems allow the clinician to select the etching mode depending on the clinical situation: total-etch, self-etch, or selective enamel etching. Nevertheless, even with modern materials, there is no absolutely universal protocol that would provide equally high bond strength to enamel and dentin under all clinical conditions [6,8].

Enamel and dentin differ fundamentally in their structure and biological properties. Enamel is a highly mineralized tissue with a low content of organic components and water; therefore, after acid etching, it

creates favorable conditions for micromechanical retention. Dentin, by contrast, is a hydrated biological tissue containing a mineral phase, collagen matrix, dentinal tubules, and dentinal fluid. For this reason, adhesion to dentin is considered more complex and less predictable than adhesion to enamel [12].

One of the key factors affecting the quality of adhesion is the smear layer - a surface layer formed on enamel and dentin after mechanical instrumentation. It consists of hydroxyapatite particles, denatured collagen, organic detritus, microorganisms, salivary residues, and products of preparation. When etch-and-rinse systems are used, the smear layer is largely removed by acid etching, whereas in self-etch and some universal systems it is partially dissolved and incorporated into the hybrid layer. If the smear layer is excessively thick, heterogeneous, or highly enriched with organic components, it may hinder complete adhesive infiltration and reduce bond strength [2,12].

In this regard, methods for preliminary modification of the dentin and enamel surface have been actively investigated in recent years. These include mechanical treatment, chemical deproteinization, the use of sodium hypochlorite, hypochlorous acid, laser treatment, plasma technologies, matrix metalloproteinase inhibitors, collagen cross-linkers, and enzymatic agents. Among these methods, enzymatic treatment attracts attention because of its potential biocompatibility, selective action on the organic component, and ability to provide mild surface modification without excessive aggressiveness [1,7].

Bromelain and papain - plant-derived proteolytic enzymes - are of particular interest. Bromelain is obtained from pineapple, whereas papain is obtained from papaya. Both enzymes are able to degrade protein components, which theoretically allows them to be used for removing organic residues from the surface of hard dental tissues. In dentistry, papain has previously been used as part of atraumatic systems for chemo-mechanical removal of caries-affected dentin, while bromelain has been considered a possible agent for dentin deproteinization before adhesive restoration [10,15].

Thus, studying enzymatic pretreatment of enamel and dentin is a relevant direction because it may complement existing adhesive protocols and improve the predictability of esthetic restoration. It is especially important that this problem lies at the intersection of clinical dentistry, materials science, biochemistry, and morphology of hard dental tissues, which makes it promising for dissertation research.

The aim of the present review is to analyze current international scientific data on the influence of

enzymatic pretreatment of enamel and dentin surfaces on the effectiveness of the adhesive protocol in esthetic restoration, with an assessment of its possible effects on the smear layer, hybrid layer formation, adhesive bond strength, microleakage, and marginal adaptation of restorative materials.

Main body / discussion

Adhesion of restorative material to hard dental tissues is based on a combination of micromechanical retention, chemical interaction of functional monomers, and formation of the hybrid layer. During phosphoric acid etching of enamel, partial demineralization of the surface layer occurs and microporosities are formed, into which adhesive resin penetrates. After polymerization, a strong micromechanical bond is created, ensuring stable retention of the restoration.

Dentin adhesion is more complex. After acid etching, the smear layer is removed, dentinal tubules are opened, and the superficial dentin is demineralized with exposure of collagen fibrils. Subsequently, the primer and adhesive must penetrate this zone and, after polymerization, form the hybrid layer. If monomer infiltration is incomplete, areas of exposed collagen and water remain in the deeper part of the hybrid layer. These areas become vulnerable to hydrolysis, nanoleakage, activation of endogenous proteases, and gradual degradation of the adhesive interface [2].

Modern universal adhesives contain functional monomers, in particular 10-MDP, which are capable of chemically interacting with calcium in hydroxyapatite. This increases bond stability, especially when mild self-etch systems are used. However, even universal adhesives remain sensitive to clinical conditions: dentin moisture, smear layer thickness, degree of etching, application time, active rubbing of the adhesive, solvent evaporation, and quality of polymerization [4,6].

For esthetic restoration of anterior teeth, the quality of adhesion is of particular importance. Even minimal deficiencies in marginal fit may lead to marginal staining, secondary caries, alteration of the optical transition between dental tissues and composite, increased sensitivity, and reduced esthetic outcome. Therefore, the search for additional methods of enamel and dentin preparation is a justified direction for optimizing the clinical protocol.

The smear layer is formed during preparation of dental tissues with diamond and carbide burs, abrasive discs, finishing instruments, and other tools. Its thickness, density, and composition depend on the type of instrument, rotational speed, cooling, applied

pressure, condition of the dental tissues, and depth of dentin. In enamel, the smear layer is usually more mineralized, whereas on dentin it contains more organic components and collagen fragments.

On the one hand, the smear layer may temporarily reduce dentin permeability and decrease dentinal fluid movement. On the other hand, it can prevent full contact between the adhesive and the intact dentin surface. This problem is especially significant when self-etch systems are used, since they do not completely remove the smear layer but rather modify and incorporate it into the hybrid complex. If the smear layer is dense, uneven, or saturated with organic detritus, it may reduce monomer penetration and impair the quality of hybridization [12].

This is why different approaches to smear layer modification are discussed in the literature. Chemical deproteinization with sodium hypochlorite makes it possible to remove the organic component, but it can be aggressive, induce changes in the collagen matrix, and require strict time control. Hypochlorous acid is considered a milder alternative, although its clinical use also requires standardization. Enzymatic agents such as bromelain and papain can potentially provide a more selective effect on protein components of the smear layer while preserving the mineral basis of the surface [1,7].

Enzymatic pretreatment is based on the ability of proteolytic enzymes to break down protein structures. In relation to hard dental tissues, this may have several effects. First, enzymes can partially remove organic residues present on the surface after preparation. Second, they may reduce the amount of denatured collagen that mechanically blocks adhesive penetration. Third, enzymatic treatment may improve surface wettability and promote more uniform distribution of the adhesive system.

Bromelain is a complex of proteolytic enzymes capable of degrading protein substrates. In dental studies, it has been investigated as an agent for deproteinization of dentin and enamel. According to Alshahli et al. (2020), bromelain pretreatment promoted partial removal of the organic phase of the smear layer, increased microshear bond strength to enamel and dentin, and improved restorative adaptation in class V cavities. These data are particularly important because they link enzymatic treatment not only with laboratory bond strength but also with marginal adaptation, which has direct clinical relevance.

Papain is a plant-derived cysteine protease. Its dental use has previously been associated mainly with chemo-mechanical removal of caries-affected tissues. As a deproteinizing agent, papain may also affect organic

components of the surface; however, findings regarding its influence on adhesion are inconsistent. Some studies have shown increased bond strength after papain treatment, whereas others have reported no significant effect or a dependence of the outcome on concentration and type of adhesive system [14,15].

Differences between bromelain and papain may be explained by their biochemical activity, concentration, exposure time, preparation form, solution pH, dentin depth, and the subsequent adhesive protocol. Therefore, enzymatic treatment cannot be regarded as a universal additional stage without experimental justification. On the contrary, it is necessary to determine the optimal enzyme, concentration, application time, and compatibility with a specific adhesive system.

Bond strength is one of the main laboratory indicators of adhesive protocol effectiveness. It can be evaluated using microtensile bond strength, microshear bond strength, or shear bond strength tests. Although these methods do not fully reproduce clinical conditions, they make it possible to compare different surface preparation methods and identify potentially effective protocols.

In the study by Alshahli et al. (2020), bromelain pretreatment was shown to increase microshear bond strength to enamel and dentin when certain universal adhesive systems were used. At the same time, papain did not demonstrate the same stable positive result. The authors associated the effect of bromelain with partial dissolution of the organic phase of the smear layer, which facilitates penetration of adhesive monomers and improves contact between the adhesive and dental tissue.

Sharafeddin et al. (2024) investigated the influence of different concentrations of bromelain and papain on the bond strength of composite resin to deep dentin using an etch-and-rinse adhesive system. The authors noted that enzymatic deproteinization in general did not produce a universal increase in bond strength; however, differences between concentrations were statistically significant. For example, 15% papain showed higher values compared with 10% papain, while 6% bromelain was more effective than 10% bromelain. This indicates that increasing the enzyme concentration does not always lead to improved adhesion.

Khatib et al. (2020) evaluated the microtensile bond strength of composite resin to etched dentin after the use of sodium hypochlorite, bromelain, and papain. In this study, natural enzymatic agents showed promise as alternatives to traditional chemical deproteinization. At the same time, bromelain demonstrated higher

values compared with other treatment options.

Sharafeddin and Safari (2019) also examined the effect of papain and bromelain on the bond strength of composite to superficial dentin with different adhesive systems. It was shown that the effect of enzymatic treatment depends on the selected adhesive strategy. This is especially important for clinical practice because the same enzyme may produce different results when used with etch-and-rinse, self-etch, or universal adhesives.

Thus, analysis of the available data shows that enzymatic treatment may increase adhesive bond strength, but its effectiveness is not absolute. At present, bromelain appears to be the most promising agent; however, its clinical use requires standardization of concentration, exposure time, and compatibility with a specific adhesive system.

For esthetic restoration, not only bond strength but also the quality of marginal fit is important. Microleakage leads to the penetration of fluids, pigments, bacteria, and their metabolic products between the restoration and dental tissues. Clinically, this manifests as marginal staining, secondary caries, postoperative sensitivity, and reduced restoration longevity.

Studies show that smear layer modification can influence marginal adaptation. Alshahli et al. (2020) demonstrated that bromelain and hypochlorous acid not only increased microshear bond strength but also improved restoration adaptation in experimental cavities. However, this effect depended on the type of adhesive. This means that enzymatic treatment should not be considered in isolation but rather as part of a comprehensive adhesive protocol.

Bedir and Telatar (2023), comparing different deproteinizing agents, showed that their influence on bond strength and microleakage may differ depending on the application mode of a universal adhesive. In particular, some agents improved bond strength in the etch-and-rinse mode but could fail to produce the expected effect or even worsen microleakage parameters in the self-etch mode. This emphasizes the need for a cautious approach to including an enzymatic stage in the clinical protocol.

From a practical point of view, it is especially important to evaluate enzymatic treatment at the enamel-dentin interface, since esthetic restorations of anterior teeth often have a complex margin configuration: part of the restoration is bonded to enamel, part to dentin, and part may be located in the cemento-enamel junction area. Consequently, an optimized protocol should provide reliable adhesion to different substrates simultaneously. In esthetic restoration of anterior

teeth, the adhesive protocol must ensure not only strength but also color stability, absence of marginal discoloration, minimal sensitivity, high polishability, and long-term restoration integration. Enzymatic pretreatment may be especially useful when the dentin surface contains a pronounced organic component, when minimally invasive preparation is performed, and when teeth with non-carious defects, erosions, abrasion, or areas of sclerotic dentin are restored [3,5,11].

However, at present enzymatic treatment cannot be recommended as a standard mandatory stage for all clinical cases. The reason is that most studies are *in vitro*, differ in methodology, use different enzyme concentrations, different adhesive systems, and different outcome assessment methods. In addition, there is insufficient evidence on long-term clinical effectiveness, influence on postoperative sensitivity, color stability, and restoration survival.

For enzymatic treatment to be included in an optimized adhesive protocol, it is necessary to determine which enzyme is most effective; which concentration is safe and effective; how long the exposure should last; whether the enzyme should be applied before or after acid etching; how rinsing and drying should be performed; whether the enzyme is compatible with a specific adhesive system; and whether it influences the long-term stability of the hybrid layer.

From the perspective of dissertation research, the most justified approach is to study enzymatic treatment as one of the elements of an optimized protocol. In the laboratory part, microshear or microtensile bond strength, microleakage, marginal adaptation, and surface morphology after treatment can be evaluated. In the clinical part, it is possible to study restoration retention, marginal staining, sensitivity, esthetic integration, and patient satisfaction. Such an approach will make it possible not only to describe the mechanism of enzyme action but also to substantiate its practical significance.

Conclusion

Enzymatic pretreatment of enamel and dentin represents a promising direction for optimizing the adhesive protocol in esthetic restoration. The theoretical rationale for this approach is related to the ability of proteolytic enzymes to act on organic components of the smear layer, improve surface preparation, enhance the penetration of adhesive monomers, and contribute to the formation of a more stable adhesive interface.

Analysis of current international studies shows that bromelain is of greatest interest, as several studies have demonstrated its ability to increase microshear

bond strength to enamel and dentin, as well as improve the marginal adaptation of restorative material. Papain may also be considered a potential deproteinizing agent; however, evidence of its effectiveness is less consistent and depends on concentration, type of adhesive system, and features of the clinical protocol.

At the same time, enzymatic treatment should not be regarded as a universal and automatically effective stage. Its application requires rigorous experimental and clinical substantiation. It is necessary to consider enzyme concentration, exposure time, sequence of steps, condition of enamel and dentin, type of adhesive system, and etching mode. Of particular importance is the study of the influence of enzymatic treatment on restoration longevity, microleakage, marginal adaptation, and postoperative sensitivity.

Thus, enzymatic pretreatment of enamel and dentin can be included in the scientific substantiation of an optimized adhesive protocol for esthetic restoration. Further studies should focus on standardizing the enzymatic stage, comparatively evaluating bromelain and papain, and confirming laboratory findings under clinical conditions.

References

1. Alshahli R. Z., Sato K., Hosaka K., Hatayama T., Chiba A., Foxton R. M., Tagami J., Sumi Y., Shimada Y., Nakajima M. Effect of smear layer deproteinization with enzyme solutions on bonding efficacy of one-step self-etch adhesives // International Journal of Adhesion and Adhesives. — 2020. — Vol. 102. — Article 102672. — DOI: 10.1016/j.ijadhadh.2020.102672.
2. Amin F., Fareed M. A., Zafar M. S., Khurshid Z., Palma P. J., Kumar N. Degradation and stabilization of resin-dentine interfaces in polymeric dental adhesives: an updated review // Coatings. — 2022. — Vol. 12, № 8. — Article 1094. — DOI: 10.3390/coatings12081094.
3. Bedir F., Telatar G. Comparison of different dentin deproteinizing agents on bond strength and microleakage of universal adhesive to dentin // Journal of Advanced Oral Research. — 2023. — Vol. 14, № 1. — P. 44–53. — DOI: 10.1177/23202068231157998.
4. Bourgi R., Hardan L., Cuevas-Suárez C. E., Lukomska-Szymanska M., Cornejo-Ríos E., Tosco V., Monterubbianesi R., Mancino D., Haikel Y. A literature review of adhesive systems in dentistry: key components and their clinical applications // Applied Sciences. — 2024. — Vol. 14, № 18. — Article 8111. — DOI: 10.3390/app14188111.
5. Breschi L., Maravic T., Comba A., Cunha S. R., Loguercio A. D., Reis A., Van Meerbeek B. The evolution of adhesive dentistry: from etch-and-rinse to universal bonding systems // Dental Materials. — 2025. — Vol. 41. — P. 1–16.
6. Cuevas-Suárez C. E., de Oliveira da Rosa W. L., Lund R. G., da Silva A. F., Piva E. Bonding performance of universal adhesives: an updated systematic review and meta-analysis // The Journal of Adhesive Dentistry. — 2019. — Vol. 21, № 1. — P. 7–26.
7. Delgado A. H. S., Belmar da Costa M., Polido M. C., Mano Azul A., Sauro S. Collagen-depletion strategies in dentin as alternatives to the hybrid layer concept and their effect on bond strength: a systematic review // Scientific Reports. — 2022. — Vol. 12. — Article 13028. — DOI: 10.1038/s41598-022-17371-0.
8. Doshi K., Nivedhitha M. S., Solete P., Antony D. P. S., Balasubramaniam A., Jacob B., Siddique R. Effect of adhesive strategy of universal adhesives in non-carious cervical lesions: an updated systematic review and meta-analysis // BDJ Open. — 2023. — Vol. 9. — Article 6. — DOI: 10.1038/s41405-022-00124-6.
9. El-Laithy M. A., Niazy M. A., Abdelhakim S. H. Effect of bromelain enzyme versus sodium hypochlorite on shear bond strength of resin composite to dentin using two adhesive systems // Egyptian Dental Journal. — 2024. — Vol. 70, № 1. — P. 761–767. — DOI: 10.21608/edj.2023.241784.2745.
10. Khatib M. S., Devarasanahalli S. V., Aswathanarayana R. M., Venkateswara A. H., Nadig R. R. Microtensile bond strength of composite resin following the use of bromelain and papain as deproteinizing agents on etched dentin: an in vitro study // International Journal of Clinical Pediatric Dentistry. — 2020. — Vol. 13, № 1. — P. 43–47.
11. Nozari A., Al-Zain A. O., Almutairi A., Alshehri A., Alshahrani A. Influence of different application modes of a universal adhesive system on microshear bond strength of restorative materials // Clinical and Experimental Dental Research. — 2024. — Vol. 10. — Article e947. — DOI: 10.1002/cre2.947.
12. Saikaew P., Sattabanasuk V., Harnirattisai C., Chowdhury A. F. M. A., Carvalho R., Sano H. Role of the smear layer in adhesive dentistry and the clinical applications to improve bonding performance // Japanese Dental Science Review. — 2022. — Vol. 58. — P. 59–66.
13. Sharafeddin F., Haghbin N., Farhadpour H., Vohra F. Comparison of bromelain enzyme, sodium hypochlorite and titanium tetrafluoride on shear

bond strength of restorative composite to dentin: an in vitro study // Journal of International Oral Health. — 2019. — Vol. 11. — P. 281–286.

- 14.** Sharafeddin F., Jowkar Z., Safari M. Effects of different concentrations of bromelain and papain enzymes on shear bond strength of composite resin to deep dentin using an etch-and-rinse adhesive system // Dental and Medical Problems. — 2024. — Vol. 61, № 1. — P. 85–91. — DOI: 10.17219/dmp/133404.
- 15.** Sharafeddin F., Safari M. Effect of papain and bromelain enzymes on shear bond strength of composite to superficial dentin in different adhesive systems // The Journal of Contemporary Dental Practice. — 2019. — Vol. 20, № 9. — P. 1077–1081.