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ORIGINAL RESEARCH

The effect of the use of the deproteinization agent hypochlorous acid and two different pit and fissure sealant self-adhesive flowable composites upon its bonding with the enamel

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Abstract

This study evaluates the effect of the deproteinization agents hypochlorous acid and sodium hypochlorite upon the bonding of the two different pit and fissure sealant, self-adhesive flowable composites with the enamel. Thirty-six third molars were randomly divided into six different groups. The groups were formed as follows: Group 1: 37% phosphoric acid + Vertise^{TM} Flow; Group 2: 200 ppm hypochlorous acid + 37% phosphoric acid Vertise^{TM} Flow; Group 3: 5.25% sodium hypochlorite + 37% phosphoric acid + Vertise^{TM} Flow; Group 4: 37% phosphoric acid + Constic; Group 5: 200 ppm hypochlorous acid + 37% phosphoric acid + Constic; Group 6: 5.25% sodium hypochlorite + 37% phosphoric acid + Constic. In each group, samples were obtained that were rectangular prisms in shape (n = 12). Groups to which a deproteinization agent was applied (Groups 2, 3 and 5, 6) showed statistically higher microtensile bonding strength than Group 1, Group 4. There was no statistically significant difference in terms of microtensile bonding strength values between the Groups 3 and the Group 6. The study found that the groups to which deproteinization agents were applied had statistically higher microtensile bonding strength values compared with those groups to which acid and fissure sealants were applied. In this study, it was concluded that the use of fissure-sealing self-adhesive flowable composites after acid application to permanent tooth enamel provides an acceptable bond strength given the limitations of *in vitro* studies. In line with the results obtained, it was observed that in addition to the removal of the inorganic structure with the application of acid, the removal of the organic structure with the use of deproteinization agent increased the bond strength to the enamel.

Keywords

Enamel; Deproteinization; Sodium hypochlorite; Phosphoric acid; Etching; Permanent teeth

1. Introduction

Oral and dental health are an inextricable part of our general health. Tooth decay is a chronic and preventable condition in whose formation many factors play a part and which results in physical and chemical destruction of the hard tissues of the teeth [1]. The sealant acts as a physical barrier in preventing oral bacteria and dietary carbohydrates from creating the acid conditions that result in caries. The resin tag forms a bonding which is mechanically interlinked between the resin material and the tooth surface [2]. Occlusal surfaces of the posterior teeth are the spots more vulnerable to decay formation due to their deep and narrow pits and fissured anatomies, which may harbour plaque bacteria and are difficult to reach during brushing [3]. Pit and fissure sealants form barriers against plaques and acids, serving as hard shields that stop food and bacteria from entering these sensitive parts on the occlusal surfaces of the teeth, thus preventing them from causing decay. Current studies show that fissure sealants prove to be an effective preventive approach when placed upon decay lesions without cavities, preventing decay progression [4].

Fissure sealant is a common method to arrest incipient caries by pretreatment with phosphoric acid and application of a resin-based sealant material [5]. Retention is a significant factor that affects the longevity of sealants. Theoretically, unfilled sealants can penetrate deeper into the fissures due to their low viscosity so that they might exhibit better retention [6]. The preventive role of pits and fissure sealants using resin-based sealants is well described in the literature. There is evidence that pits and fissure sealants reduce the incidence of caries lesions in 76% of healthy occlusal faces versus no preventative treatment over a 3-year follow-up period. Moreover, resin-based sealants procedures are a rapid and minimally invasive treatment for resin-based sealants restorations with marginal defects. This is also a less complex procedure for dentists than a complete replacement of the restoration. More importantly, when resin-based sealants fail, it does not mean immediate development of secondary caries; the treatment can also be repeated [7].

Although flowable composites lack mechanical properties due to lower filler content, they are commonly used as a pit and fissure sealant in Class III, Class V and Class I restorations that do not involve areas of occlusal loading, or as a liner in large restorations and so on [8]. Self-adhesive flowable composite resins are indicated as bases under higher-viscosity restorative materials in Class I, Class III or Class V restorations to block out undercuts and as pit and fissure sealants [9].

The adhesion provided is an important factor for a successful restoration. This property is usually measured using bond strength tests [10]. In addition, a study shows that brackets placed on teeth treated with Ozone and Tooth Mousse before bonding agent application produce bond strengths that are clinically adequate and superior to teeth treated with Fluorine. During pre-orthodontic treatment with fluoride lacquer or varnish, it causes lower bond strengths, which is likely to result in a higher frequency of early separation of the bracket [11].

The mechanism of self-adhesion is provided by the demineralization of acidic monomers and their ability to penetrate the tooth substrate. This causes micromechanical retention and chemical interaction between phosphate acidic groups and hydroxyapatite [12]. This interaction with dental hard tissues occurs only through limited. The bonding mechanism of adhesion in such systems is nanointeraction, similar to the mechanism of self-adhesive resin cements [13]. The main advantage of self-adhesive materials is the simple and fast application procedure [12].

Acid etching of the enamel as a pre-treatment is a significant step during the application of pit and fissure sealants. This application helps create micro-porosities on the enamel, and as the enamel has higher wettability, the bonding strength of the fissure sealant increases. The efficiency of a fissure sealant is measured on the basis of the level of adhesion of the material to the teeth and its ability to enable impermeability [14]. The level of adhesion shows variations depending on the appropriate isolation of the work area, the preparation of the enamel surfaces, the viscosity of the fissure sealant and additional processes applied to the surface of the enamel [15].

Before acid etching, it has been shown that the deproteinization agent applied to the enamel increased the bonding strength of the fissure sealant to the enamel. The removal of organic materials from the surface of the enamel before acid etching increases the bonding strength, providing a better acid etch pattern. Hypochlorous acid (HOCI) and sodium hypochlorite (NaOCl) are a perfect protein denaturant. Sodium hypochlorite is a non-specific proteolytic agent that eliminates organic compounds efficiently at room temperature. The application of HOCl and NaOCl, known as deproteinization agents, may be a potential strategy to increase adhesion, eliminating the organic components of both the enamel structure and the pellicle before acid etching of the enamel [16].

Although it is thought that it is difficult to provide saliva isolation during dental treatments in children, recently self-adhesive flowable composites have been introduced as a new generation fissure sealant. It was suggested by the manufacturer that acid should be used before the self-adhesive flowable composite used as a fissure sealant is applied to the enamel. It has been proven that the deproteinization process applied before acid removes the organic structure in addition to the inorganic structure from the enamel surface. As a result of the researches, it was seen that the acid roughened enamel surface, which is exposed after the sealant is poured, is more susceptible to bacterial damage than the normal unroughened enamel surface. As long as the sealants adhere to the enamel, the current protection will continue. As a result, sealant retention and microleakage resistance become the real determinants [17]. In this study, evaluates the effect of the deproteinization agents hypochlorous acid and sodium hypochlorite upon the bonding of the two different pit and fissure sealant, self-adhesive flowable composites with the enamel. The null hypothesis was that the self-adhesive flowable composite used as a fissure sealant would have similar bond strengths only to the tooth surface treated with acid and the enamel after the deproteinization agent applied before acid.

2. Materials and methods

2.1 Specimen preparation

Following the approval of the ethics committee by the Faculty of Dentistry, Gazi University, thirty-six extracted, the caries-free third molars with no defects linked to hypomineralization were used in this study. The composition and production locations of the materials used are as shown in Table 1.

All the plaques and soft tissue debris on the teeth were brushed away with the help of running water. After being stored in distilled water containing 0.2% thymol crystals at room temperature for purposes of disinfectation until the time of the experiment, the teeth were used within 3 months of extraction. All the extracted teeth were cleaned and dried for 15 seconds before the experiments. The roots of the teeth before abrasion, at least six teeth in each group, were cut 2 mm below the enamel-cement junction before being buried in cold caryl with their buccal surfaces left open. The buccal enamel surfaces of the teeth underwent abrasion for 30 seconds with the 600–1200 grit silicon carbide paper (Gripo 2V Grinder-Polisher, Metkon Instruments Ltd, Bursa, Turkey) under running water to create a standardized smear layer. Samples from the experiment were randomly divided into six groups. A rectangular mould 6 mm in width and 5 mm in height was used so that fissure sealants could be applied in a standard fashion to the enamel surfaces. By doing so, the enamel-bonding region was narrowed in which the fissure sealant was applied. In Group 1, 37% phosphoric acid and Vertise^{TM} Flow; in Group 2, 200 ppm hypochlorous acid plus 37% phosphoric acid and Vertise^{TM} Flow; in Group 3, 5.25% sodium hypochlorite plus 37% phosphoric acid and Vertise^{TM} Flow; in Group 4, 37% phosphoric acid and Constic; in Group 5, 200 ppm hypochlorous acid plus 37% phosphoric acid and Constic; in Group 6, 5.25% sodium hypochlorite plus 37%

TABLE 1. The materials used in our study and the firms that produce them.

Material	Composition	Manufacturer
Vertise^{TM} Flow (Flowable composite, Self-etch, Self-adhesive)	GPDM, HEMA, prepolymerized filler, nano-sized ytterbium fluoride, 1-μm barium glass filler, nano-sized colloidal silica	Kerr Dental, Italy
Constic (Flowable composite Self-etch, Self-adhesive)	Matriks; Bis-GMA, EBADMA, UDMA, HEMA, TEGDMA, MDP Filling; Ba-glass 0.02–2.3 μm	DMG, Germany
HOCl (Deproteinization agent)	6.2 pH and 200 ppm active chlorine	SuperOx, Anolit Kimya, Ankara, Turkey
NaOCl (Deproteinization agent)	5.25% sodium hypochlorite	Promida, Turkey
Acid Etching	37% phosphoric acid	NovaDFL, Rio De Janerio, Brazil

Bis-GMA: bisphenol A glycidyl methacrylate; GPDM: glycerol phosphate dimethacrylate; UDMA: urethane dimethacrylate; HEMA: hydroxyethyl methacrylate; TEGDMA: triethylene glycol dimethacrylate; EBADMA: ethoxylated bisphenol A dimethylacrylate; MDP: methacryloyloxydecyl dihydrogen phosphate; HOCl: hypoclorous acid; NaOCl: sodium hypochlorite.

phosphoric acid and Constic were applied. Hypochlorous acid and sodium hypochlorite solutions were applied, using a disposable microbrush before being rinsed off with water for 20 seconds. After, the teeth in each group were applied a 1 mm fissure sealant, they were built up to a composite of 2 mm in 2 layers. Each 2 mm layer of the applied composite resins was polymerized through light-curing for 20 seconds.

2.2 Microtensile bond strength test

After the application of the fissure sealant in each group, the composite resin was applied and polymerized in pieces of 2 mm up to the highest section of the rectangular mould and samples were prepared for the microtensile bond strength test. The samples were kept in an étuve at 37 °C for a period of 24 h in distilled water until the process of cross-section. Following this, all the samples were turned into sticks in rectangular prisms with a surface area of 1 mm^2, after being placed into a cross sectioning tool (Micracut Precision Cutter, Metkon Instruments Ltd, Bursa, Turkey). The edges of the samples were measured with a digital caliper (Shinwa Co, Osaka, Japan) with the bonding surfaces of the samples being calculated in mm^2. The ends of the obtained samples were fixed to a microtensile testing device (Micro Tensile Tester, Bisco Inc., Schaumburg, IL 60193, USA) with cyanoachrylate adhesive. After the samples were fixed to a metal block in the device, 1 mm tensile strength per minute was applied until the fissure sealant was broken away from the surface of the enamel. All the values during this separation were recorded in Newton on the digital screen of the device. The microtensile bond strength values recorded in Newton were later converted into "Megapascal" using the following formula: Newton/Surface Area = MPa.

2.3 Failure mode analysis

After the microtensile bond strength test, the failure area of the specimens was observed using a light microscope (Leica DM 4000B, Germany) at 40× magnification for failure mode determination. Failure modes were classified as cohesive failure inside the resin material, adhesive failure between the enamel and the resin material and mixed failure when it occurred both within the material and between the enamel surface and the resin material.

3. Results

Mean values of the micro tensile bond strength (MPa) for the two self adhesive flowable composite according to the pretreatment protocol are summarized in Table 2.

Application of deproteinization agents NaOCl and HOCl increased the microtensile bond strength of self-adhesive flowable composite Vertise^{TM} Flow compared to Group 1. For each specimen in which Vertise^{TM} Flow was applied as a fissure sealant, a statistically significant difference ($p < 0.001$) in microtensile bond strengths was observed as shown in Table 2, Fig. 1 and Group 2 (200-ppm HOCl + acid) and Group 3 (5.25% NaOCl + acid) was found to have a higher microtensile bond strength value than Group 1 ($p < 0.001$ and $p = 0.023$) as shown in Fig. 1. There was no statistically significant difference in terms of microtensile bond strength values between Group 2 (200-ppm HOCl + acid) and Group 3 (5.25% NaOCl + acid) as shown in Table 2, Fig. 1 ($p = 0.498$).

While the deproteinization agent NaOCl increases the bond strength of the self-adhesive flowable composite Constic according to Group 4 (the acid-only Constic Group); there was no statistically significant difference in microtension bond strength of Constic compared to Group 4 (the acid-only Constic Group) of HOCl application. For the samples in which Constic was applied as a fissure sealant, a statistically significant difference was observed as shown in Table 2. Fig. 1 ($p = 0.008$) and Group 6 (5.25% NaOCl + acid) was found to have a higher microtensile bond strength value than Group 4 in which acid was applied as shown in Table 2, Fig. 1 ($p = 0.006$).

There were no statistically significant differences in microtensile bond strength values both between the acid group (Group 4) and 200 ppm HOCI + acid group (Group 5) and between 200 ppm HOCl + acid group (Group 5) and 5.25% NaOCl + acid group (Group 6) as shown in Table 2, Fig. 1 ($p = 0.447$ and $p = 0.299$).

When the bond strength of the fissure sealant self-adhesive

TABLE 2. Mean values of the micro tensile bond strength (MPa) for the two self adhesive flowable composite according to the pretreatment protocol.

Pretreatment	Vertise Flow	Constic	*p*-value[†]
Acid	13.70 (12.97–15.77)[a,b]	16.60 (14.25–18.42)[b]	0.014
Acid + 200 ppm HOCl	19.10 (16.82–26.30)[a]	18.65 (15.20–22.02)	0.242
Acid + %5.25'lik NaOCl	17.05 (15.32–22.10)[b]	21.30 (18.15–24.72)[b]	0.045
p-value[‡]	<0.001	0.008	

Abbreviations used in this table: HOCl: hypoclorous acid; NaOCl, sodium hypochlorit.

The same superscripts in the rows and columns indicate the statistically significant differences. It was expressed as the median (25%–75%).

[a] *: The difference between the Acid group and the Acid + 200 ppm HOCl group was statistically significant ($p < 0.001$); [b]: The difference between the Acid group and the Acid + 5.25% NaOCl group statistically significant ($p < 0.025$).*

[†] *Comparisons between fissure sealants in each solution, Mann Whitney U test, results for $p < 0.0167$ according to Bonferroni correction were considered statistically significant.*

[‡] *Comparisons between solutions in each fissure sealant, Kruskal Wallis test, Bonferroni correction. The results were considered statistically significant for $p < 0.025$.*

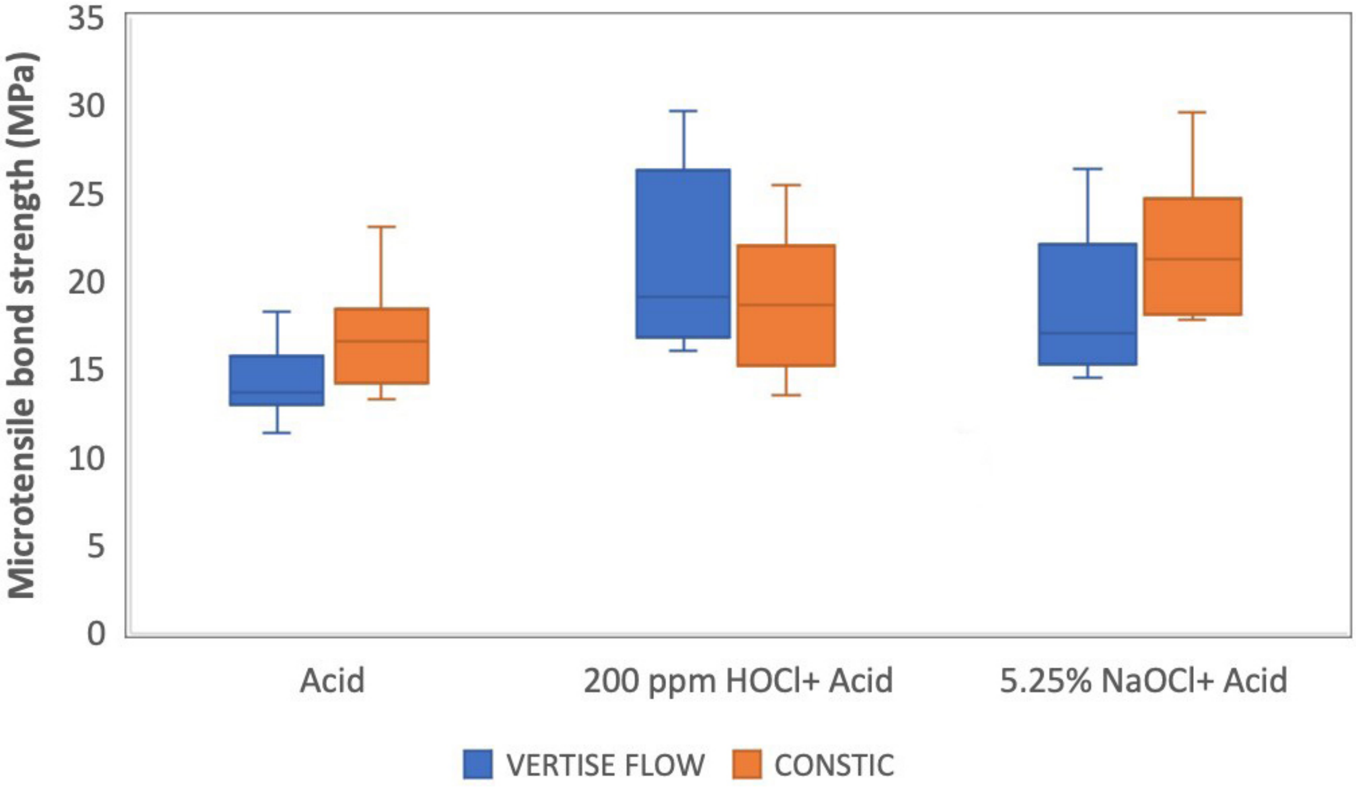

FIGURE 1. Microtensile bond strengths value of each group. Apart from the application of acid and two different self-adhesive flowable composites to the permanent tooth enamel, the effectiveness of the use of HOCl and NaOCl on the bond formed between the permanent tooth and the self-adhesive flowable composite before the acid application is seen. HOCl: hypoclorous acid; NaOCl, sodium hypochlorite.

flowable composite Vertise^{TM} Flow and Constic was evaluated only after acid application, no statistically significant difference was found. The acid-only Constic Group (Group 4) was found to have a statistically higher microtensile bond strength value than the acid-only Vertise^{TM} Flow (Group 1) as shown in Table 2, Fig. 1 (p = 0.014).

There was no statistically significant difference between Group 2 (200-ppm HOCI + acid + Vertise^{TM} Flow) and Group 5 (200-ppm HOCl + acid + Constic) in terms of microtensile bond strength values as shown in Table 2, Fig. 1 (p = 0.242).

No statistically significant difference was found between the microtensile bond strength values of Group 3 (5.25% NaOCl + acid + Vertise^{TM} Flow) and Group 6 (5.25% NaOCl + acid + Constic) as shown in Table 2, Fig. 1 (p = 0.045).

4. Discussion

While the deproteinization agent NaOCl increases the microtensile bond strength of both self-adhesive flowable composites (Vertise^{TM} Flow and Constic); HOCl increased the microtensile bond strength of the self-adhesive flowable composite Vertise^{TM} Flow. Therefore, the null hypothesis was rejected.

In the light of this information, it is suggested that the use of self-adhesive flowable composites together with the deproteinization agents NaOCl or HOCl before applying them as fissure sealant may increase the bonding to the enamel surface.

Self-adhesive flowable composites have recently been introduced as materials aimed at ushering in innovations in restorative procedures, bringing together the features of not only adhesive but also restorative material technologies. Self-adhesive flowable composites allow for fewer application steps even for patients for whom much restoration is fulfilled in the same visit while creating less room for failure. Additionally, they reduce the amount of visit time, thereby proving to be extremely useful when dealing with non-cooperative patients [18].

Vertise™ Flow is a restorative material that can bond with the hard dental tissue without using an adhesive system. The bonding mechanism of Vertise^{TM} Flow relies on the functional monomer glycerol phosphate dimethacrylate (GPDM) whose acidic phosphate groups bond with calcium ions and whose methacrylate functional groups copolymerize with other methacrylate monomers and thus increase the cross bonding strength and mechanical endurance [19].

Vertise^{TM} Flow contains both GPDM, which has the capacity to etch the enamel and the dentin and hydroxyethyl methacrylate, a hydrophilic monomer. There is a chemical and micromechanical bonding between functional monomers and hydroxyapatite and polymerized resin and collagen fibers, respectively [20]. Glycerol phosphate dimethacrylate has a stronger etching effect compared to other self-adhesive monomers, a very short intermediate chain that brings about more satisfactory wettability, as well as a higher hydrophilic potential [21].

Vertise^{TM} Flow is used as a repair material in Class I and Class V cavities, in the restoration of caries-free cervical lesions and in Class I and Class II restorations [19]. This material is also clinically used as a pit and fissure sealant. Because caries-free pits and fissures have a prismatic enamel layer, the producer recommends that Vertise^{TM} Flow be placed on the enamel layer after the application of phosphoric acid for appropriate adhesion of Vertise^{TM} Flow.

Another self-adhesive flowable composite introduced recently, Constic, is a resin material with a bonding system based on an adhesive monomers GPDM. Additionally, similar to the content of Vertise^{TM} Flow, Constic contains a 10-Methacryloyloxydecyl dihydrogenphosphate (MDP) monomer with a longer and more hydrophobic intermediate chain. This monomer also forms stable 10-MDP-Ca salts, which provide a strong chemical bonding with hydroxyapatite [21]. Thanks to the phosphate group of GPDM, acid application to the enamel is feasible. Recommended for etching and rinsing systems, the abrasion of the enamel is aimed at increasing the surface energy of the enamel, removing the smear layer. This step is not necessary when the self-adhesive flowable composite is applied to the surface of the tooth and has no effect on marginal adaptation at all [22].

The use of self-adhesive flowable composites as fissure sealants is based on adhesion-forming capacities without the need to prepare any cavity or any adhesive system in hard dental tissues; however, bonding to the enamel is extremely difficult for these materials even on buccal surfaces. As such, when self-adhesive flowable composites are used as fissure sealants, acid application is recommended [23]. In our study, 37% phosphoric acid was applied for a period of 30 seconds before the application of 2 different self-adhesive flowable composites to the enamel surface.

Studies have shown that enamel etching patterns depend on the type and concentration of the substance used, as well as the etching time and the composition and morphology of the enamel surface to be roughened [24]. Successful adhesion of composite materials requires sufficient etching to provide a good etching pattern as well as bonding capacity. Phosphoric acid has been used for this purpose for a long time. However, phosphoric acid alone does not form a total adhesion surface [24].

Espinosa *et al.* [25] found that phosphoric acid etched less than 50% of the enamel area, while Hobson *et al.* [26] found up to 69% of the surface intact, with only 2% of the surface ideally etched. In our study, it was observed that the deproteinization agents (NaOCl, HOCl) used before the fissure sealant application had higher enamel binding values than the fissure sealant applied only with the acidification process.

This weak bond strength is usually a result of phosphoric acid acting only on the mineralized surface (inorganic material) and not on the organic material covering the enamel. Therefore, it is necessary to remove organic material from the enamel surface before starting the etching protocol. Etching is often hindered by the presence of proteins found between the enamel crystals. Thus, to achieve a more permanent etching, Espinosa *et al.* [27] developed a pretreatment based on enamel deproteinization for one minute with 5.25% NaOCl for acidification of the entire enamel. This process step is successful in increasing fissure sealant retention.

According to our study, both the self-adhesive flowable composites applied to the enamel surface with 5.25% NaOCl for 60 seconds before acid etching were found to have a statistically more significant microtensile bonding strength than the control group.

In another study that explored the effect of the application of the deproteinization agent with 5.25% NaOCl before and after acid etching upon the bonding of the adhesive resin to the enamels of the primary, immature and mature permanent teeth, researchers found that deproteinization significantly increased the bonding effect on the primary and immature permanent teeth [28].

In contrast to sodium hypochlorite, HOCI solutions can be rinsed easily, which explains why the dentin surfaces pre-treated with HOCI have relatively higher μGBD values compared to dentine pre-treated with NaOCl [29]. A mildly acidic HOCI solution, in comparison with an NaOCl solution, displays more effective antibacterial and oxidizing behaviors even at low chlorine concentrations [30].

The incomplete polymerization of the resin monomers is induced because of the competition between the vinyl free radicals generated during photo-irradiation and the reactive free radicals that are formed following the oxidizing reactions of deproteinization agents such as sodium hypochlorite and hypochlorous acid [31]. For this reason, 37% phosporic acid was applied to the enamel surface following the application of deproteinization agents sodium hypochlorite and hypochlorous acid.

A study by Valencia *et al.* [32] has revealed that occlusal enamel deproteinization performed with 5.25% NaOCl for 60 seconds before 37% phosphoric acid application to the permanent teeth enamel helped remove the organic material in pits and fissures, which significantly increased the bonding strengths of adhesive restorations. Considering this research, our study found that 5.25% NaOCl deproteinization that we achieved on the enamel before the 37% phosphoric acid application (Group 3 and Group 5) had statistically more significant microtensile bonding strength compared with the control group (Group 1 and Group 4).

In another study conducted by Paing *et al.* [31], it was found that the deproteinization agent HOCI applied to the dentin had higher microshear bonding strength than the no pre-treatment group. While there are a limited number of studies on the application of the deproteinization agent hypochlorous acid, according to our study, the groups to which we applied the deproteinization agent HOCI to the enamel for 30 seconds (Group 2 and Group 5) were found to have a statistically more significant microtensile bonding strength than the control groups (Group 1 and Group 4).

Another study by Peterson *et al.* [33] demonstrated that in Vertise^{TM} Flow and Constic self-adhesive flowable composites were applied to the enamel, following failures occurred in decreasing order of frequency: adhesive type failures, mixed type failures and cohesive type failures.

Our test results, consistent with those of other studies, showed that the failures that occurred with Vertise^{TM} Flow were characterized both by adhesive and by mixed types [34, 35].

Despite the success achieved in the retention of sealants in deep pits and fissures vulnerable to decay, other factors that can interfere with the decay etiology were kept out of the scope of this study in this study. For this reason, more comprehensive studies must explore the other factors that play a role in the retention of pit and fissure sealants.

In 1955, Dr. Buonocore developed the procedure acid application to improve the adhesion of restorative materials to the enamel [36]. Bonding to enamel is referred to as a mechanism by which resin tags are formed and micro-mechanic bonding is achieved thanks to micropores that are obtained through etching [37].

At the same time, it was shown in a study that adhesion values were clinically acceptable when demineralized enamel was remineralized using biomimetic nano-hydroxyapatite. This study is a guide for treatments that can be applied on demineralized enamel in the future [38].

In *in vivo* and *in vitro* studies, the contamination of the surface enamel with saliva was shown to be the most significant factor that affected both the retention and the clinical success of the resin-based fissure sealants [39].

According to the results of a meta-analysis by Lo *et al.* [40], before the fissure sealant application, acid etching must be applied for at least 30 seconds on the permanent molars to achieve sufficient fissure sealant retention. Considering this meta-analysis, before the application of the fissure sealants, we performed etching on the enamel surfaces with 37% phosphoric acid in gel form for a period of 30 seconds.

The results observed for self-adhesive flowable composite resins should be interpreted with caution because the analysis showed heterogeneity and there is insufficient evidence to support a clinical decision. Self-adhesive dental materials are the promising future of dental adhesives, it is important to examine available materials and identify problems that may help improve the bonding mechanism and efficacy to dental tissues. Randomized clinical trials with long-term follow-up are recommended to obtain strong clinical evidence that can assist clinicians in their decision-making on dental practice.

5. Conclusions

In this study, the effects of hypochlorous acid and sodium hypochlorite, which are deproteinization agents, on the bonding of two different pit and fissure sealant, self-adhesive flowable composites with enamel were evaluated. Given the limitations of *in vitro* studies, the results of this study demonstrated that the use of NaOCl and HOCl prior to acid application to permanent tooth enamel provides an acceptable increase in enamel bond strength of Vertise^{TM} Flow and Constic self-adhesive flowable composites. Deproteinization with 5.25% NaOCl and 200 ppm HOCl before etching offers a non-invasive, efficient and economical method to improve adhesion of materials to permanent tooth structure. Thus, according to the results obtained, it was observed that in addition to the removal of the inorganic structure with acid application, the removal of the organic structure with the use of deproteinization agent increased the bond strength to the enamel. Further laboratory and clinical studies are necessary to investigate the retention of fissure sealants on enamel following enamel deproteinization. *In-vivo* studies with a large sample size and long-term follow-up are needed.

AVAILABILITY OF DATA AND MATERIALS

The data presented in this study are available on reasonable request from the corresponding author.

AUTHOR CONTRIBUTIONS

SP and ÇÇ—designed the research study. SP—performed the research. SP and ÇÇ—analyzed the data. All authors contributed to editorial changes in the manuscript. All authors read and approved the final manuscript.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

The study protocol was approved by the Gazi University Faculty of Dentistry Ethics Committee (E-21071282-050.99-186369, 07.10.2021).

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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