Effect of a Papain–based Gel for Chemomechanical Caries Removal on Dentin Shear Bond Strength

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ABSTRACT

Purpose: The aim of this study was to assess the shear bond strength of an adhesive restorative system on sound and demineralized dentin after the use of a papain-based agent.
Methods: Forty human dentin slabs were randomly distributed into 4 groups: (1) sound dentin slabs that received an application of papain-based gel (N=10); (2) sound dentin slabs that did not receive a papain-based gel application (N=10); (3) demineralized slabs that received an application of the agent (N=10); and (4) demineralized slabs that did not receive an application of the agent (N=10).
After manual excavation and use of the chemomechanical agent, the slabs were restored with a total etch adhesive system and microhybrid resin composite. The test specimens were individually stored in a damp environment for 7 days, and the shear bond strength test was performed using a universal test machine at a speed of 0.5 mm/min. The fragments were observed under a stereoscopic microscope to assess the fracture mode.
Results: Fracture mode assessment showed adhesive and cohesive type fractures in resin for all the groups. The analysis of variance and the Tukey test showed that there were no differences in the shear bond strength means among the groups (P>.05).
Conclusions: The use of a papain-based gel to remove dental caries did not interfere in the bond strength of restorative materials to dentin. (J Dent Child 2007;74:93-7)

Keywords: Shear bond strength, chemomechanical caries removal, total etch adhesive system

One of the challenges in dentistry is to reduce caries lesions.[1] This disease still presents a high prevalence in some groups, however, characterizing a group polarization.[1-3]

Several chemomechanical methods for dental caries removal have been developed, beginning with the use of 5% sodium hypochlorite (NaOCl), which was shown to be an easily way to remove carious lesions.[4,5] The solution was called GK-101 or n-monochloroglycine composed of: sodium hydroxide (NaOH), sodium chloride (NaCl), glycine, and 0.05% of NaOCl was efficient at removing carious lesions.4,6 and later modified by the addition of an ethyl group renamed GK-101E or N-monochloro-D,L-2-aminobutyrate, which was shown to be more efficient.

It was given the brand name Caridex and applied using the same technique used for GK-101.[5,7] The Caridex system was proposed to:

1. minimize the need for anesthesia;
2. reduce the use of rotary instruments; and
3. preserve the remaining dental structure by removing only the carious lesion.[8,9]

In the 1980s, because of the chemomechanical agent's advantages, Swedish researchers tried to eliminate the disadvantages of Caridex.[10] They patented the product, which

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was initially called "Demex" and, later, "Carisolv."[9] This product is composed of amino acids and NaOCl, developed in the form of gel and used in small quantities (0.5-1 ml) per carious lesion.[9,11] It is presented in the form of 2 gels:

1. one gel is transparent and contains 5% NaOCl; and
2. the other gel is red and composed of 3 amino acids (glutamic acid, leucine, and lysine), NaCl, and water.

Considering the advantages of Carisolv due to its longer action time,[12] a gel was developed in Brazil in 2003. It was based on papain, chloramines, and toluidine blue and called "Papacárie" (a word that means "eating caries"). This development allowed a reduction in costs, due to the use of low-cost Brazilian raw materials.[13] The gel is applied to the contaminated dentin and its proteolytic, chlorinating, and oxidating properties act on the affected collagen,[14] without acting on the sound dentin.

There are few studies, however, that relate its clinical effectiveness in removing caries from dentin.[15,16] It is known when Carisolv is used, that it will lead to the removal of organic material as well as the smear layer. Consequently, the dentin becomes more permeable, which facilitates adhesive system penetration and may increase resin composite bonding to the tooth.[17] The effectiveness of adhesive material bonding after the use of papain-based gel, however, is unknown.

This present study's aim was to assess the shear bond strength of an adhesive restorative system on sound and demineralized dentin after the use of a papain-based gel.

METHODOLOGY

EXPERIMENTAL DESIGN

The factors studied in this experiment were:

1. the dentin caries removal method:
   a. chemomechanical removal—use of a papain-based gel called Papacárie for chemomechanical removal of dentin caries, which is composed of papain, chloramine, toluidine blue, salts, conservative, and thickener associated with removal using manual excavators;
   b. mechanical removal—removal with manual excavators only; and
2. degree of dentin mineralization:
   a. healthy dentin slabs; and
   b. demineralized dentin slabs.

The experimental units consisted of 40 human dentin slabs randomly distributed (N=10) into 4 groups:

1. sound slabs that received an application of a papain-based gel (N=10);
2. sound slabs that did not receive an application of a papain-based gel (N=10);
3. demineralized slabs that received an application of the agent (N=10); or
4. demineralized slabs that did not received an application of the agent (N=10).

Shear bond strength was quantitatively assessed and expressed in MPa. The 3 experimental design principles, according to Montgomery (2003)[18]–repetition, randomization, and blockage–were complied with.

PREPARATION OF SLABS

After approval by the Research Ethics Committee of Hermínio Ometto University Center (Uniararas), (protocol no. 05/232), human third molars—extracted for reasons not related to those of the research and stored in thymol (0.1%; pH=7.0) after extraction—were used in this experiment. The teeth were cleaned with scalpel blades and periodontal curettes. A cross section was made, separating the root and coronal portions. The apical third of the root was discarded, and only the cervical region was used. Longitudinal cuts allowed slabs measuring 4 mm x 4 mm to be obtained. Those presenting cracks or stains were excluded, resulting in 40 dentin slabs being obtained.

The slabs were embedded in polystyrene resin with PVC molds, leaving the external dentin uncovered by the resin. After 24 hours, the test specimens were removed from the molds for flattening to obtain smooth surfaces needed for making test specimens for the shear bond strength tests. The slabs were flattened with decreasing granulations (400, 600, and 1200) of water abrasive paper under constant water cooling and then randomly distributed (N=10) into 4 groups.

OBTAINING THE DEMINERALIZED SLABS

To obtain artificial caries lesions (demineralized slabs), a dynamic model similar to that used by Hara et al (2004),[19] was used. Each slab was individually immersed in demineralizing solution for 1 hour. Next, each slab it was washed in distilled and deionized water and placed in a remineralizing solution to complete a 23-hour cycle. After this time, the slabs were washed again, dried and immersed in the demineralizing solution to start a new cycle. Three cycles were performed, maintaining a temperature of 37°C during the procedures.

CHEMOMECHANICAL AGENT APPLICATION AND DENTIN CARIES REMOVAL

For Groups 1 and 3, the dentin slabs received an application of the chemical agent. The manufacturer's recommendations were followed, and the product was placed on the dentin surface and allowed to act for 30 seconds (acute lesions). The dentin was scraped with the noncutting edge of the curette, and 5 to-and-fro movements were made. The gel was applied again, and manual excavation was repeated with another 5 to-and-fro movements. Residual gel was removed with absorbent paper.

For Groups 2 and 4, the dentin slabs did not receive any application of the chemical agent. The demineralized dentin was submitted to manual excavation only with the noncutting edge of the curette, and 5 to-and-fro movements were made. Manual excavation was repeated with another 5 to-and-fro movements. The cleaned dentin was then washed with water.

After cleaning, the fragments were kept in their individual receptacles containing distilled water at relative humidity for 1 day.

SHEAR BOND STRENGTH TESTS

The location of the shear bond strength tests was demarcated with adhesive paper, leaving an exposed area 3.0 mm in diameter.

The test specimens were made in a random sequence—to which the investigators were blind—with a total etch adhesive system (Single Bond, 3M ESPE, Saint Paul, Minn) and microhybrid resin composite (Filtek Z 100, 3M ESPE), in accordance with the manufacturers' recommendations.

A Teflon mold 5.0 mm high and 3.0 mm in diameter was placed over the fragment after the adhesive system had been applied to make a resin composite cylinder in 2 layers. The first layer was placed into the mold with a thickness of approximately 2.0 mm and photocured for 20 seconds with a light curing unit (Ultralux EL, Dabi Atlante, Ribeirão Preto, São Paulo, Brazil). The second layer was placed and was also photocured for 20 seconds. The mold was removed and the resin cylinder was cured for 20 seconds on both sides. After every 5 test specimens made, the light curing unit was assessed with a radiometer (Newdent, São Carlos, São Paulo, Brazil).

The test specimens were stored in individual vials in a damp environment for a period of 7 days.

Shear bond strength tests were also performed blindly in a universal test machine (Emic DL 200, São José dos Pinhais, Paraná, Brazil) at a speed of 0.5 mm/minute. The active tip of the machine was placed close to the test specimen so that the shear strength force was directed toward the bond area. The shear strength values attained were expressed in Mpa.

The fractured test specimen surfaces were examined visually under a stereoscopic magnifying glass at X30 magnification to classify the fracture. The fractures were classified as: (1) adhesive (lack of adhesion); (2) cohesive in dentin (failure of the tooth substrate); (3) cohesive in resin (failure of the resin composite); and (4) mixed (adhesive and cohesive failure).

The results obtained in MPa were statistically analyzed by analysis of variance (ANOVA) followed by the Tukey test (P<.05).

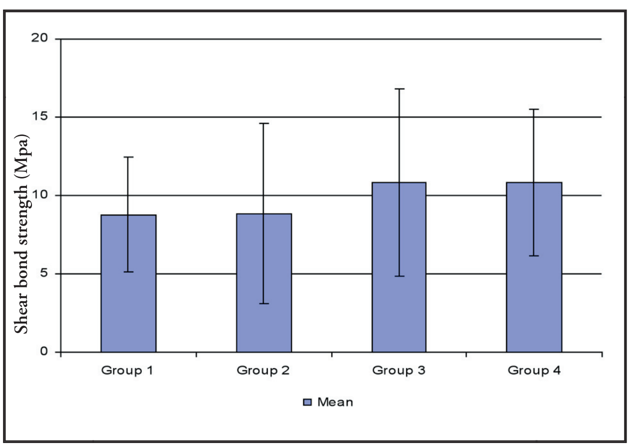

Figure 1. Means and standard deviations of the study groups.

RESULTS

The ANOVA and the Tukey test showed that there were no differences in the mean shear bond strength values among groups, showing that this product did not affect the bonding of different adhesive systems to dentin (Figure 1 and Table 1). Fracture mode assessment showed fractures of the adhesive and cohesive type in resin for all the groups (Table 2), showing a preservation of dentin substrate followed by resistance fracture.

Table 1. Means and Standard Deviations of the Study Groups

	Group 1 (n=9)	Group 2 (n=10)	Group 3 (n=6)	Group 4 (n=9)
Mean±(SD)	8.78±3.67[a]	8.86±5.77[a]	10.87±5.97[a]	10.83±4.69[a]

** Values followed by the same letters do not differ by ANOVA (P=.7128).*

Table 2. Fracture Modes of Test Specimens Submitted to Shear Bond Strength Test

Fracture mode	GROUPS			
	Sound slabs		Demineralized slabs	
	Chemomechanical removal	Mechanical removal	Chemomechanical removal	Mechanical removal
Adhesive	60%	67%	44%	33%
Cohesive in dentin	0%	0%	0%	0%
Cohesive in resin	40%	33%	56%	67%
Mixed	0%	0%	0%	0%

DISCUSSION

In view of the need for adopting conservative treatments to preserve a larger amount of sound structure in teeth affected by caries lesions, it is noted that conventional dentin caries removal with rotating instruments may cause unnecessary wear, leaving the tooth remainder fragile.[20-22] Therefore, products were developed for chemical carious lesion removal, which would remove affected dentin and preserve demineralized dentin.[23]

These systems do not eliminate the use of manual instruments, however, helped to reduce the patient's stress regarding noise, as they almost completely eliminated the use of rotary instruments.[24,25]

Carisolv, a product that contains NaOCl, ruptures the cross links between the dentinal collagen fibrils, denaturing them and dissolving the necrotic tissue. The bond between NaOCl and the amino acid reduces the effect of whole collagen denaturing and rupturing only the link between the affected collagen fibrils, without any molecular alterations occurring.[11] According to Banerjee, et al (1999),[26] the results with the use of Carisolv showed to be equivalent to those of the rotary system after 1 year of clinical follow-up. This product allows only the infected and necrotic dentin, no longer capable of being remineralized, to be removed, and allowed the bottom uninfected layer to be preserved. Thus, the technique is able to remove the smear layer, which facilitates penetration of substances such as adhesives,[27] enhancing the adhesional properties of restorative materials.[17]

Papacárie, a gel based on papain, chloramines, and toluidine blue, however, is inexpensive compared with Carisolv and has a similar use, indication and chemomechanical caries removal efficiency.[13] Papain makes it easy to clean necrotic tissues and secretions, and reduces tissue repair time, in addition to not affecting sound tissues close to the lesion. This substance lacks a plasmatic antiprotease—antitrypsin—which prevents its proteolytic action on tissues that are considered normal. It is also considered a chemical debridement agent, which helps in the healing process and acts as an anti-inflammatory agent.[13,16] Chloramine helps to soften the carious dentin, thus facilitating its removal since the degraded portion of the carious dentin collagen is colored by the solution used in chemical and mechanical caries removal.[12] Results have been favorable after 1-year clinical follow-up of 60 teeth in children from 5 to 9 years old and of 30 molars in adolescents and adults up to 23 years of age.[16]

Papacárie has been tested at various concentrations. It has not been shown to be cytotoxic and is biocompatible with oral tissues.[28] This product has antimicrobial effectiveness, mainly regarding *Streptococcus mutans* and *Lactobacillus*.[29]

In the present study, Papacárie did not interfere in resin composite bonding, as there were no significant differences between the tested groups or regarding fracture mode distribution. These data corroborate the studies conducted by Frankenberg et al[30] and Haak et al (1999)[31] with Carisolv, showing that this product did not affect the bonding of different adhesive systems to dentin. Hosoya et al,[32] however, found that the application of Carisolv before acid etching might alter the bond strength of primary and permanent dentin, depending on the adhesive system.

It is suggested that changes in bond strength values after the use of chemical agents to remove carious lesion may occur due to micromorphologic alterations. Rodrigues et al[33] assessed primary tooth dentin submitted to carious lesion removal with burs and with a chemomechanical agent. The dentin submitted to rotary instrument treatment presented a smooth, uniform appearance with the presence of a smear layer. The use of chemomechanical agents led to irregular surfaces, however, with the predominance of an amorphous layer in flakes covering the dentinal tubules. In some areas, locations similar to the presence of a smear layer were observed, but with microfractures. Nevertheless, the present study used an adhesive system that involves the application of phosphoric acid on all of the dental substrate.[34] This may remove the entire smear layer, giving the dental substrate an irregular appearance after the use of the chemomechanical agent. Self-etching adhesive systems requiring the use of a weak acid, however, do not completely remove the smear layer to form the hybrid layer. When using these systems, different results can be expected than those found in this research.[35,36]

CONCLUSION

A papain-based chemomechanical agent does not interfere in the shear bond strength of restorative materials when a total etch adhesive system is required. Thus, this agent can safely be used as a method for caries removal when employing conventional adhesive systems.

REFERENCES

1. Marthaler TM. Changes in dental caries 1953-2003. Caries Res 2004;38:173-81.
2. Mattos-Graner RO, Rontani RMP, Gavião MBD, Bocatto, HARC. Caries prevalence in 6- to 36-month-old Brazilian children. Community Dent Health 1996;13:96-8.
3. Antunes JL, Jahn GM, de Camargo MA. Increasing inequalities in the distribution of dental caries in the Brazilian context in Finland. Community Dent Health 2005;22:94-100.
4. Goldman M, Kronman JH. A preliminary report on a chemomechanical means of removing caries. J Am Dent Assoc 1976;93:1149-53.
5. Schutzbank SG, Galaini J, Kronman JH, Goldman M, Clark RE. A comparative in vitro study of GK-101 and GK-101E in caries removal. J Dent Res 1978;57:861-4.
6. Habib CM, Kronman J, Goldman M. A chemical evaluation of collagen and hidroxyproline after treatment with GK-101 (N-chloroglycine). Pharrmacol Ther Dent 1975;2:209-15.
7. Kronman JH, Goldman M, Habib CM, Mengel L. Electron microscopic study of altered collagen structure after treatment with N-monochloro-DL-2-aminobutyrate (GK-101E). J Dent Res 1979;58:1914.
8. McCune RJ. Report on symposium on chemomechanical caries removal: A multicenter study. Compend Contin Educ Dent 1986;7:151-4.
9. Medi Team Annual Report. Stockholm, Sweden: Medi Team Dentalutveckling I Göteborg; 1998.
10. Meinbach E. A chemomechanical caries removal system: Painless dentistry versus procedural inconveniences. Dentistry 1990;10:10-2.
11. Ericson D. In vitro efficacy of a new gel for chemomechanical caries removal. J Dent Res 1998;77:1252.
12. Maragakis GM, Hahn P, Hellwig E. Clinical evaluation of chemomechanical caries removal in primary molars and its acceptance by patients. Caries Res 2001;35:205-10.
13. Bussadori SK, Castro LC, Galvao AC. Papain gel: A new chemo-mechanical caries removal agent. J Clin Pediatr Dent 2005;30:115-9.

14. Flindt M. Allergy to a-amylase and papain. Lancet 1979;1:430-2.
15. Silva LR, Motta LJ, Reda SH, Façanha RAA, Bussadori SK. Papacárie–um novo sistema para a remoção química e mecânica do tecido cariado–relato de caso clínico. Rev Paul Odontol 2004;XXVI:4-8.
16. Pereira AS, Silva LR, Motta LJ, Bussadori SK. Remoção Química de Cárie Por Meio do Gel Papacárie. RGO 2004;52:385-8.
17. Emanuel R, Broome MJC. Surface energy of chemomechanically prepared dentin. Quintessence Int 1988;19:369-72.
18. Montgomery DC. Design and Analysis of Experiments. 4th ed. New York: Jonh Wiley and Sons; 1996:704p.
19. Hara AT, Magalhães CS, Rodrigues Jr AL, Serra MC. Efeito cariostático de restaurações adesivas em superfícies radiculares: Estudo in vitro. Pesqui Odont Bras 2004;14:113-8.
20. McNierny HD, Petruzillo MA. A gentle approach to operative dentistry: The Caridex caries removal system. Gen Dent 1986;34:282-4.
21. Cavel WT, Kelsey WP 3rd, Barkmeier WW, Blankenau RJ. Clinical evaluation of chemomechanical removal of cervical caries. Gen Dent 1988;36:405-8.
22. Gwinnett JA, Barkmeier WW. Morphology of dentin surfaces in chemomechanically prepared cavities. Am J Dent 1988;1:101-4.
23. Fusayama T. Two layers of carious dentin: Diagnosis and treatment. Oper Dent 1979;4:63-70.
24. Akyuz S, Pince S, Hekim N. Children's stress during a restorative dental treatment: Assessment using salivary cortisol measurements. J Clin Pediatr Dent 1996;20:219-23.
25. Aartman IH, Dejongh A, Van Der Meulen MJ. Psychological characteristics of patients applying for treatment in a dental fear clinic. Eur J Oral Sci 1997;105:384-8.
26. Banerjee A, Watson TF, Kidd EAM. Carious dentine excavation using carisolv gel: A quantitative, autofluorescence assessment using scanning microscopy. Caries Res 1999;33:313.
27. Naressi SCM, Araújo MAM. Comparação da infiltração marginal de restaurações adesivas empregando instrumento rotatório ou sistema químico-mecânico na remoção do tecido cariado. RPG Rev Pós Grad 2001;8:313-21.
28. Silva LR, Tonolli G, Santos EM, Bussarori SK. Avaliação da biocompatibilidade in vitro de um novo biomaterial para a remoção química e mecânica da cárie. Pesq Odontol Bras 2003;17(suppl):93.
29. Pereira SA, Silva LR, Piccinini DPF, Santos EM, Bussadori SK. Comparação in vitro do potencial antimicrobiano de dois materiais para remoção químico-mecânica da cárie. Pesqui Odontol Bras 2004;18(suppl):78.
30. Frankenberg R, Oberschachtsiek H, Kramer N. Bond Strength to dentin after mechanical and chemical removal of simulated caries. J Dent Res 1999;78:232.
31. Haak R, Fritz UB, Faber FJ, Noack MJ. Influence of chemomechanical caries removal on dentin bonding. J Dent Res 1999;78:369.
32. Hosoya Y, Kawashita Y, Marshall Jr. GW, Goto G. Influence of Carisolv for resin adhesion to sound human primary dentin and young permanent dentin. J Dent 2001;29:163-71.
33. Rodrigues CRMD, Arana-Chaves VE, Oliveira MDM, Mathias RS. Estudo em MEV da dentina de dentes decíduos após tratamento com carisolv. Pesqui Odontol Bras 2000;14(suppl):126.
34. Pashley DH. The effects of acid etching on the pulpodentin complex. Oper Dent 1992;17:229-42.
35. Chain MC, Chiarelli M, Dunker C. Análise do mecanismo de ação de sistemas adesivos autocondicionantes através de microscopia eletrônica de varredura. Anais do Encontro Grupo Brasileiro de Professores de Dentística 2001;14:76.
36. Sinhoretti MAC, Góes MF, Consani S, Correr Sobrinho L. Resistência ao cisalhamento da união de sistemas adesivos sobre dentina. Rev Assoc Paul Cir Dent 1997;51:251-5.