



Revista Portuguesa de Estomatologia, Medicina Dentária e Cirurgia Maxilofacial

www.elsevier.pt/spemd



Investigation

Safe use of ozone gas in enamel adhesion

Patrícia T. Pires^{a,*}, João C. Ferreira^a, Sofia A. Oliveira^b, Mário J. Silva^a, Paulo R. Melo^a

^a Faculty of Dental Medicine, University of Oporto, Oporto, Portugal

^b Faculty of Dental Medicine, University of Lisbon, Lisbon, Portugal

ARTICLE INFO

Article history:

Received 13 February 2012

Accepted 7 May 2012

Available online 12 July 2012

Keywords:

Shear strength

Dental enamel

Dentin-bonding agents

Ozone

ABSTRACT

Purpose: Evaluate the shear bond strength of a self-etching system to enamel pretreated with ozone and its type of fracture.

Material and methods: Thirty sound bovine incisors were bisected and polished just before the application of the adhesive system. The adhesion area was limited to a 3-mm diameter. The specimens were randomly assigned to the experimental groups ($n=15$) and composite resin cylinders were added to the tested surfaces, after the application of the adhesive according to the manufacturer's instructions. Group G1 (AdheSE[®] with ozone) was previously used with ozone gas from the HealOzone unit (Kavo[®]) for 20 s, groups G2 (AdheSE[®]) was used as control. The specimens were stored in distilled water for 24 h at 37 °C with 100% humidity, before being thermocycled. The type of fracture was analyzed under scanning electronic microscope and the data were submitted to Shapiro-Wilk, Student's t-test and Chi-squared statistical analyses.

Results: The mean bond strengths were G1: 15.0 MPa (77.8% of adhesive fractures between enamel and adhesive) and G2: 13.1 MPa (36.4% of adhesive fractures between enamel and adhesive).

Conclusion: The shear bond strength of a self-etching system was not influenced by the previous application of ozone gas.

© 2012 Sociedade Portuguesa de Estomatologia e Medicina Dentária. Published by Elsevier España, S.L. All rights reserved.

Segurança da aplicação de ozono na adesão ao esmalte

RESUMO

Objectivo: Avaliar as forças de resistência adesiva de um sistema adesivo auto-condicionador no esmalte pré-tratado com ozono e o tipo de fractura.

Material e métodos: Trinta incisivos hígidos de origem bovina foram seccionados de forma a separar a coroa da raiz e polidos antes da colocação do sistema adesivo. A área de adesão foi limitada a 3 mm de diâmetro. Os espécimes foram aleatoriamente divididos ($n=15$) e cilindros de resina composta foram adicionados às superfícies de teste e após cada sistema adesivo ter sido aplicado de acordo com as instruções do fabricante. O grupo G1 (AdheSE[®] com ozono) foi condicionado com gás de ozono gerado pelo aparelho HealOzone (Kavo[®]),

Palavras-chave:

Força de resistência adesiva

Esmalte dentário

Adesivos dentários

Ozono

* Corresponding author.

E-mail address: patriciapires75@gmail.com (P.T. Pires).

durante 20 segundos, G2 (AdheSE®) funcionou como controlo. Os espécimes foram mantidos em água destilada durante 24 horas numa estufa a 37 °C com 100% de humidade, antes da termociclagem. O tipo de fractura analisado ao MEV e os dados submetidos à análise estatística Shapiro–Wilk, Student's t-test e Chi-squared.

Resultados: As médias de resistência adesiva foram: G1:15,0MPa (77,8% de fracturas adesivas entre o esmalte e o adesivo) e G4: 13,1MPa (36,4% de fracturas adesivas entre o esmalte e o adesivo).

Conclusão: Os valores de resistência adesiva do sistema adesivo auto-condicionador não foram influenciados pela aplicação prévia de gás de ozono.

© 2012 Sociedade Portuguesa de Estomatologia e Medicina Dentária. Publicado por Elsevier España, S.L. Todos os direitos reservados.

Introduction

Currently, it is not possible to assure that a tooth cavity is bacteriologically aseptic, thus an antibacterial treatment of the dental surface previous to restoration has been advised.¹ Indeed, some authors have started to apply Ozone as a disinfecting agent.² Ozone, with its antibacterial action due to its strong oxidizing activity, is an important disinfecting agent.³⁻⁵

Recent research reveals the bactericidal action of ozone against *Streptococcus mutans* and other bacteria commonly found in cervical caries.^{5,6} However, there are very few data concerning its effect on dental adhesion.⁷ Previous studies demonstrated that oxygen and other oxidant agents (such as whitening agents) have a negative influence on bond strength values of dental-enamel adhesives.

Resin-enamel adhesion is one of the most significant advances in the history of Dentistry⁸ and it is in our days as a simple effective procedure, when using a total-etch technique.⁹ Nevertheless, the enamel etching concept has been improved through the years and new adhesive systems have been developed and released.^{10,11}

Self-etching systems were developed to simplify and eliminate some of the clinical steps associated to total-etch.¹¹ Self-etching adhesives are based on acidic monomers that simultaneously condition and prime enamel.¹² The primer is applied on the enamel and resin tags are formed. Smear layer is dissolved and incorporated into the bonding process, therefore the tooth no longer requires rinsing, as it does with etch-and-rinse.^{13,14}

One of the questions that arise is whether the acidic monomer used in self-etch adhesive systems is capable of promoting enamel demineralization, making it a reliable and durable adhesion.^{15,16} Shear bond strength tests aim to establish a numeric value in order to determine how strong that bond is.¹⁷ In addition, since no rinsing occurs after the application of the self-etching, we may speculate that self-etch systems are more susceptible to Ozone residual oxygen.

This study aimed to determine whether ozone gas is safe to use in bovine enamel regarding its effect on Shear Bond Strength (SBS) when using a self-etching adhesive (AdheSE®, Ivoclar vivadent AG, Liechtenstein).

Materials and methods

Thirty sound bovine incisors were extracted for no longer than a month and kept in distilled water at 4 °C. After this period

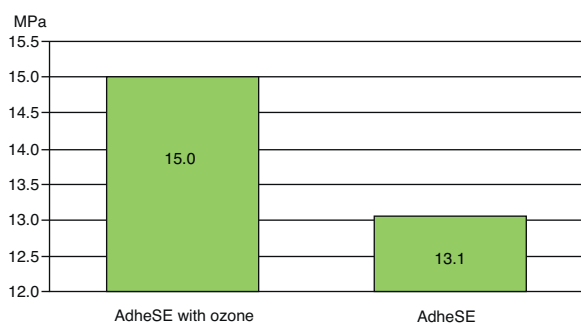
of time, the teeth were kept in a 0.5% chloramine solution for a week and bisected with a microtome (Accuton-Struers, Copenhagen, Denmark) to separate the crown from the root. They were then polished with a 240-grit sandpaper to create a flat surface and polished, again, with a 320-grit sandpaper (Carbimet Buehler-met, Buehler, Lake Bluff, IL) to simulate the smear layer just before the application of the adhesive system. Polyester film (Mylar, Dupont Corp., DE, USA), with a 3-mm diameter hole was used to restrict the adhesion area. Specimens were randomly assigned to one of two experimental groups (n=15) and composite resin cylinders were bonded to the tested surfaces, after the application of the adhesive according to the manufacturer's instructions: AdheSE Primer was applied with a brush. Once the surface was completely coated, the primer was brushed into the entire surface for another 15 s. The total reaction time was not shorter than 30 s. The primer was dispersed with a strong stream of air until the mobile liquid was no longer visible. Then, AdheSE Bond was applied and dispersed with a weak stream of air and polymerized for 10 s. Group G1: AdheSE® with ozone (Ivoclar vivadent AG, Liechtenstein) was conditioned for 20 seconds with ozone gas from the HealOzone unit (Kavo®, Germany) using a 5-mm delivery cup (green). Groups G2 (AdheSE®) was used as control, not receiving ozone before the application of the adhesive system. The adhesive materials used in this study are listed in Table 1 along with the manufacturers' compositions, batch numbers and codes. After this application, specimens were kept in distilled water for 24 h at 37 °C with 100% humidity (Hemmet, Schwabach, Germany) in order to obtain the maximum resin polymerization, before being thermocycled (Aralab, mod 200E, Cascais, Portugal) for 500 cycles at 5 °C and 55 °C for 20 s¹⁸ in each bath and submitted to shear testing at a crosshead speed of 0.5 mm/min (Instron, model 4502, series H3307, Instron Ltd, Bucks, England). The type of fracture was analyzed under SEM (JEOL JSM 6301F, Tokyo, Japan). Fractures were classified by a single experienced investigator, as either adhesive, cohesive (resin or enamel) or mixed¹⁹ and the data were submitted to Shapiro–Wilk, to evaluate the normality and Student's t-test to compare both groups. Chi-squared statistical analyses were used to compare the type of fractures.

Results

The mean Shear Bond Strengths (SBS) shown in Fig. 1 were G1: 15.0 MPa (77.8% of adhesive fractures between enamel and adhesive) and G2: 13.1 MPa (36.4% of adhesive fractures

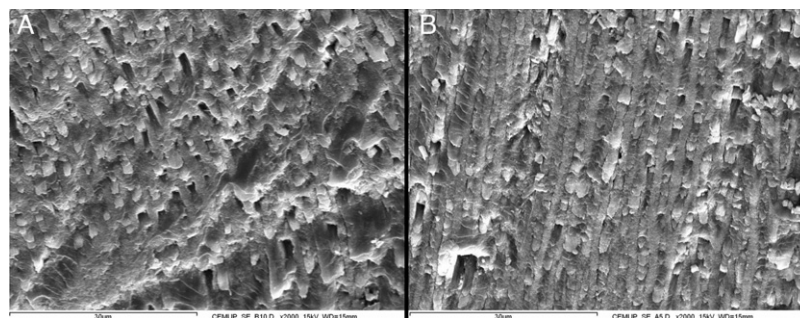
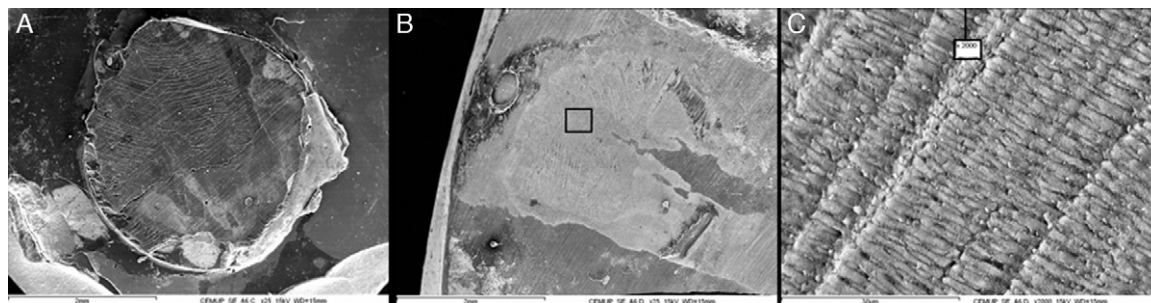
Table 1 – Restorative and adhesive materials.

Materials	Function	Composition	Batch#
AdheSE® (Ivoclar vivadent A G, Liechtenstein)	Adhesive system	Primer: Mixture of dimethacrylate, phosphonic acid acrylate, water, initiators and stabilizers; Bond: Bis-GMA, HEMA, GMDA,	J03385
Synergy D6 (Coltene whaledent GmbH + Co. KG Germany)	Restorative material	Methacrylates, barium glass silanized, amorphous silica	0145721

**Fig. 1 – Mean shear bond strength (MPa).**

between enamel and adhesive). A typical fracture between enamel and adhesive can be observed in Fig. 3.

No statistic differences were found between G1 and G2 ($p > 0.05$). The enamel surface obtained with and without ozone can be seen in Fig. 2.

**Fig. 2 – Enamel surface after being treated with the self-etching AdheSE® with (A) and without (B) Ozone. FESEM, 2000x, Gold. Enamel prisms can be identifiable in all the observed specimens.****Fig. 3 – Adhesive fracture between enamel and adhesive (Caption: A – Composite surface mostly covered by the adhesive; B – Enamel surface not covered by the adhesive; C – Enamel surface with enamel rods).**

Discussion

In the present study, AdheSE® mean SBS values did not differ significantly between Ozone pretreated group and the control group (15.0 MPa and 13.1 MPa with Ozone pretreatment and control, respectively).

The result of this work seems to show no harm influence of ozone in terms of Shear Bond Strength.

In this study, ozone was applied on enamel for 20 s. Ozone eliminates 99.9% of bacteria present after this period.^{2,3,20,21} This very short ozone gas application time when compared with other potential oxidants, like bleaching agents, may explain why ozone gas did not decreased SBS values in the present study, as it was expected.^{22,23}

Consistent with our results, Schmidlin et al.⁷ found no statistical differences on SBS values when a total-etch system was used. In a study conducted in February 2008,²⁴ SBS values of orthodontic brackets to enamel had no differences in groups treated with or without ozone. The same results were obtained in a contemporaneous study.²²

Ozone is a promising alternative in modern Dentistry. The application of ozone to enamel may be important to disinfect the surface before applying sealants or orthodontic brackets. Recent studies shown its ability to provide an antibacterial treatment against *S. mutans* even after 8 weeks.²⁵ The current work complements a growing body of work demonstrating the safe usage of ozone in dental adhesion.^{7,22,24}

Investigation focusing on the long-term effects is recommended for further research.

AdheSE[®] is a two-step self-etching adhesive developed with the expectation of decreasing the time and technical sensitivity of the adhesion process. It is intended that they will substitute the total-etching adhesives, as manufacturers claim that these products have a similar behavior and are easier to work with.²⁶ The main concern about self-etch materials is the fact that they may not be able to etch efficiently tooth enamel.¹⁶ The results of published studies on the efficacy of bonding to enamel are not pacific,²⁷ on the contrary to etch and rinse; this was the reason for using a self-etching system in our study.

The results of AdheSE[®] SBS values in enamel were low, however, our findings are consistent with other several studies.^{11,26,28,29} Some authors state that bond strength of self-etch adhesives is not as good as adhesive systems that use total etching with phosphoric acid.³⁰

The main reason might be the total-etch acidic capacity of demineralizing enamel better and deeper than self-etch adhesives.

However, Hannig et al.³¹ using self-etching adhesives, found similar SBS with or without pre-etching enamel showing that a better and deeper demineralization might not mean higher SBS values. Other researchers have shown that although demineralization patterns are not as accentuated as self-etch adhesives, they reach highly satisfactory levels of bond strength.^{11,32,33}

Self-etching adhesives are less susceptible to the operative procedures and might show similar results *in vivo* and *in vitro*, which may not be true for total-etching adhesives that are more susceptible to the technique.

Concerning the type of fracture, it was not possible to withdraw any conclusions in our study, since the differences were not statistically significant.

Conclusions

Gaseous ozone caused no reduction in Shear Bond Strength values in enamel when self-etch systems were used. Self-etching systems were not influenced by the previous application of ozone gas in shear bond strength.

Ethical disclosures

Protection of human and animal subjects. The authors declare that the procedures followed were in accordance with the regulations of the responsible Clinical Research Ethics Committee and in accordance with those of the World Medical Association and the Helsinki Declaration.

Confidentiality of data. The authors declare that no patient data appears in this article.

Right to privacy and informed consent. The authors declare that no patient data appears in this article.

Conflicts of interest

The authors have no conflicts of interest to declare.

REFERENCES

- Pereira-Cenci T, Cenci MS, Fedorowicz Z, Marchesan MA. Antibacterial agents in composite restorations for the prevention of dental caries. *Cochrane Database Syst Rev*. 2009;CD007819.
- Baysan A, Lynch E. The use of ozone in dentistry and medicine. *Prim Dent Care*. 2005;12:47-52.
- Bocci VA. Scientific and medical aspects of ozone therapy: State of the art. *Arch Med Res*. 2006;37:425-35.
- Stubinger S, Fader R, Filippi A. The use of ozone in dentistry and maxillofacial surgery: a review. *Quintessence Int*. 2006;37:353-9.
- Baysan A, Whiley RA, Lynch E. Antimicrobial effect of a novel ozone-generating device on micro-organisms associated with primary root carious lesions *in vitro*. *Caries Res*. 2000;34:498-501.
- Holmes J. Clinical reversal of root caries using ozone, double-blind, randomised, controlled 18-month trial. *Gerodontology*. 2003;20:106-14.
- Schmidlin PR, Zimmermann J, Bindl A. Effect of ozone on enamel and dentin bond strength. *J Adhes Dent*. 2005;7:29-32.
- Christensen GJ. Bonding to enamel: when does it stand in 2005? *J Am Dent Assoc*. 2005;136:1299-302.
- Pilecki P, Stone DG, Sherriff M, Watson TF. Microtensile bond strengths to enamel of self-etching and one bottle adhesive systems. *J Oral Rehabil*. 2005;32:531-40.
- Miyazaki M, Sato M, Onose H. Durability of enamel bond strength of simplified bonding systems. *Oper Dent*. 2000;25:75-80.
- Paradella TC, Fava M. Bond strength of adhesive systems to human tooth enamel. *Braz Oral Res*. 2007;21:4-9.
- Moszner N, Salz U, Zimmermann J. Chemical aspects of self-etching enamel-bonding adhesives: a systematic review. *Dent Mater*. 2005;21:895-910.
- Kiremitci A, Yalcin F, Gokalp S. Bonding to enamel and dentin using self-etching adhesive systems. *Quintessence Int*. 2004;35:367-70.
- Peumans M, Kanumilli P, De Munck J, Van Landuyt K, Lambrechts P, Van Meerbeek B. Clinical effectiveness of contemporary adhesives: a systematic review of current clinical trials. *Dent Mater*. 2005;21:864-81.
- Moura SK, Pelizzaro A, Dal Bianco K. Does the acidity of self-etching primers affect bond strength and surface morphology of enamel? *J Adhes Dent*. 2006;8:75-83.
- Swift Jr EJ. Dentin/enamel adhesives: review of the literature. *Pediatr Dent*. 2002;24:456-61.
- Yu L, Liu JM, Wang XY, Gao XJ. Evaluation of shear bond strengths of self-etching and total-etching dental adhesives to enamel and dentin. *Zhonghua Kou Qiang Yi Xue Za Zhi*. 2009;44:165-7.
- ISO. Dental materials – testing of adhesion to tooth surface. ISO/TS 2003;11405:2003(E).
- Oliveira SA. Mecanismos de adesão: análise de self-etching materials. Porto: Universidade do Porto; 2004.
- Chahverdiani B, Thadj-Bakhche A. Ozone treatment in root canal therapy. Introduction and general discussion. *Acta Med Iran*. 1976;19:192-200.

21. Fernandes FP. Influência do tratamento de superfícies cavitárias com ozono na infiltração marginal de restaurações. Porto. 2006.
22. Cadenaro M, Delise C, Antoniollo F, Navarra OC, Di Lenarda R, Breschi L. Enamel and dentin bond strength following gaseous ozone application. *J Adhes Dent.* 2009;11:287-92.
23. Barkhordar RA, Kempler D, Plesh O. Effect of nonvital tooth bleaching on microleakage of resin composite restorations. *Quintessence Int.* 1997;28:341-4.
24. Al Shamsi AH, Cunningham JL, Lamey PJ, Lynch E. The effects of ozone gas application on shear bond strength of orthodontic brackets to enamel. *Am J Dent.* 2008;21:35-8.
25. Polydorou O, Halili A, Wittmer A, Pelz K, Hahn P. The antibacterial effect of gas ozone after 2 months of in vitro evaluation. *Clin Oral Investig.* 2012;16:545-50.
26. Ernest CP, Holzmeier M, Willershausen B. In vitro shear bond strength of self-etching adhesives in comparison to 4th and 5th generation adhesives. *J Adhes Dent.* 2004;6:293-9.
27. Goracci C, Bertelli E, Ferrari M. Bonding to worn or fractured incisal edges: shear bond strength of new adhesive systems. *Quintessence Int.* 2004;35:21-7.
28. Atash R, Van den Abbeele A. Bond strengths of eight contemporary adhesives to enamel and to dentine: an in vitro study on bovine primary teeth. *Int J Paediatr Dent.* 2005;15:264-73.
29. Lopes GC, Marson FC, Vieira LC, de Caldeira AM, Baratieri LN. Composite bond strength to enamel with self-etching primers. *Oper Dent.* 2004;29:424-9.
30. Hara AT, Amaral CM, Pimenta LA, Sinhoreti MA. Shear bond strength of hydrophilic adhesive systems to enamel. *Am J Dent.* 1999;12:181-4.
31. Hannig M, Reinhardt KJ, Bott B. Self-etching primer vs phosphoric acid: an alternative concept for composite-to-enamel bonding. *Oper Dent.* 1999;24:172-80.
32. Tay FR, Pashley DH, King NM. Aggressiveness of self-etch adhesives on unground enamel. *Oper Dent.* 2004;29:309-16.
33. Perdigao J, Lopes L, Lambrechts P, Leitao J, Van Meerbeek B, Vanherle G. Effects of a self-etching primer on enamel shear bond strengths and SEM morphology. *Am J Dent.* 1997;10:141-6.