# THE EFFECT OF STORAGE SOLUTIONS ON ENAMEL OF BOVINE TEETH

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## ABSTRACT

This study evaluated the influence of storage solution on bovine enamel. Forty freshly extracted bovine incisor teeth were used. The teeth were divided into four groups (n=10) following conditions, for 90 days: G1 = distilled water (control group); G2 = artificial saliva; G3 = 0,1% thymol; G4 = saline solution. After storage, the specimens were observed under a scanning electron microscope. The comparative analysis of the specimens revealed a great variation in the structure pattern of the normal enamel. The immersion in artificial saliva caused a larger precipitation of salts in the enamel surface. Increase of superficial porosity, characterized by larger amount of depressions was observed in the group of the saline solution. The specimens submerged in 0,1% thymol, revealed a flat, polished superficial aspect. The authors conclude that the storage solutions altered the dental enamel substrate.

Keywords: Storage solution. Thymol. Teeth.

#### **RESUMO**

O objetivo deste estudo foi analisar in vitro as possíveis alterações estruturais do esmalte bovino, após o armazenamento em diferentes soluções. Foram utilizados 40 espécimes bovinos que foram divididos em quatro grupos (n=10) e armazenados nas seguintes condições, por 90 dias: G1= água destilada (controle); G2= saliva artificial; G3= timol a 0,1%; G4= soro fisiológico. Decorrido o prazo experimental os corpos-de-prova foram analisados por meio da microscopia eletrônica de varredura. A análise comparativa dos corpos-de-prova revelou uma grande variação no padrão morfológico da superfície do esmalte normal. A imersão em saliva artificial causou uma maior precipitação de sais na superfície do esmalte. Aumento de porosidade superficial, caracterizado por maior quantidade de depressões foram observados no grupo do soro fisiológico. Os corpos-de-prova que ficaram imersos em timol a 0,1% revelaram um aspecto superficial liso, polido. Os autores concluem que as soluções de armazenamento alteraram o aspecto morfológico da superfície do esmalte dentário.

Palavras-chave: Armazenamento de materiais. Timol. Dente.

#### **INTRODUCTION**

Various researches were developed using extracted human or bovine teeth to test dental materials. After extraction it is important to prevent dehydration of teeth in storage solutions. However, the storage may affect the results of experimental studies in vitro interfering in the dental surface of the extracted teeth, modifying its physical and optical properties, besides alteration in the permeability and the dentin bond strength (RETIEF et al., 1989; GOODIS et al., 1991; GHERSEL et al. 2001; PIMENTEL et al., 2002). These solutions include water in various forms (distilled, deionized, tap, with or without antibacterial agents), formalin, alcohol, thymol, sodium hypochlorite, glutaraldehyde and saline solutions (GOODIS et al., 1991; BRANNSTROM et al., 1992; NAKABAYASHI et al., 1992; STRAWN et al., 1996; CAMPS et al., 1996).

In general terms storage solutions are not standardized and their effects on structure are not clear. Therefore, this study aims to evaluate structural alterations of the bovine enamel in scanning electron microscopy after storage in commonly used solutions on the period of 90 days. MARANHÃO, Kalena de Melo, et al. The effect of solutions on enamel of bovine teeth. *Salusvita*, Bauru, v. 28, n. 2, p. 129-134, 2009. MARANHÃO, Kalena de Melo, et al. The effect of solutions on enamel of bovine teeth. *Salusvita*, Bauru, v. 28, n. 2, p. 129-134, 2009.

### MATERIALS AND METHODS

Twenty freshly extracted bovine incisor teeth obtained from the abattoir were used. Immediately after extraction, the teeth were autoclaved and divided randomly into four groups (n=5) and stored in the following conditions, for 90 days: G1 = distilled water (control group); G2 = artificial saliva; G3 = 0,1% thymol; G4 = saline solution. After storage, the teeth were decoronated, the coronal pulps removed with a dental probe, and the crows thoroughly washed free of blood and organic debris in running tap water. Subsequently the specimens were stored in an incubator, maintained at 37°C for 24 hours. Next, the specimens were placed into a vacuum and the surface was gold sputter-coated and observed under a scanning electron microscope at different magnifications.

#### **RESULTS AND DISCUSSION**

Bovine teeth were chosen as the substrate because of the convenient size of the teeth and they may be, to some extent, a substitute for human teeth (NAKAMICHI et al., 1983; SCHILKE et al., 2000).

Studies on the effects that storage media or methods have on dentine have been focused on dentine permeability, chemical composition and structure. The present study evaluated the influence of four of the most commonly used storage solutions on enamel substrate over a longer storage period. The comparative analysis of the specimens revealed a great variation in the morphologic surface pattern of the normal enamel. The immersion in artificial saliva caused a larger precipitation of salts in the enamel surface (Fig. 1B). Salt precipitates may occur in dentine stored in phosphate-buffered saline, which could explain decreases in permeability over time. Decrease could also be due to dissolution and redeposition of either the mineral or organic components of the dentine (GOODIS, 1991).

In the present study an increase of superficial porosity, characterized by larger amount of depressions, was observed in the group of the saline solution (Fig. 1D). It has been shown that the hardness of enamel decreased when teeth were stored in physiological saline, presumably due to the loss of surface calcium (MUHLEMAN, 1964). Changing the storage solution might also induce the loss of calcium from the dentin, resulting in further exposure of the dentin collagen. Conversely, equilibrium of calcium ion transfer between the dentin and unchanged storage solutions would be established in the solution (KITASAKO, 2000). The specimens preserved in 0.1% thymol revealed a flat, polished superficial aspect (Fig. 1C). It is known that the greatest demineralization occurs in enamel stored in thymol solution (MOURA et al., 2004). In addition, Fujisawa and Kadoma (1992) demonstrate that the thymol act as an oxidizing agent and react with free radicals causing polymerization inhibition of the methacrylate monomers. However, no significant difference in bond strength to dentin in relation to storage in thymol and distilled water was reported by Aquilino et al. (1987), Titley et al. (1998) and Ziskind et al. (2003).

Water is the most popular storage solution, which caused the smallest changes in enamel substrate over time (Fig. 1A). This data may present an appropriate storage solution for adhesion and permeability experiments.





## CONCLUSION

According to the methodology used, it was possible conclude that the storage solutions may alter the dental enamel substrate. Further studies with longer observation periods are required in orMARANHÃO, Kalena de Melo, et al. The effect of solutions on enamel of bovine teeth. *Salusvita*, Bauru, v. 28, n. 2, p. 129-134, 2009. MARANHÃO, Kalena de Melo, et al. The effect of solutions on enamel of bovine teeth. *Salusvita*, Bauru, v. 28, n. 2, p. 129-134, 2009. der to investigate the permeability and the dentin bond strength in dental enamel.

#### REFERENCES

AQUILINO, S. et al. The effect of storage solutions and mounting media on the bond strengths of a dentinal adhesive to dentin. *Dent Mat*, v. 3, p. 131, 1987.

BRANNSTROM, M. et al. Effect of tooth storage and cavity cleansing on cervical gap formation in Class 11 glass ionomer/composite restorations. *Dent Mater*, v. 8, p. 327-31, 1992.

CAMPS, J. et al. Influence of cryopreservation and storage time on microleakage. *Dent Mater*, v. 12, p. 121-26, 1996.

FUJISAWA, S.; KADOMA, Y. Effect of phenolic compounds on the polymerization of methyl methacrylate. *Dent Mat,* v. 8, p. 324, 1992.

GHERSEL, E. L. A. et al. Influência do modo de armazenamento na microinfiltração de dentes decíduos restaurados com diferentes sistemas adesivos: estudo in vitro. *Pesqui Odontol Brás*, v. 15, p. 29-34, 2001.

GOODIS, H. E. et al. The effects of storage after extraction of the teeth on human dentine permeability in vitro. *Arch Oral Biol*, v. 36, p. 561-66, 1991.

GOODIS, H. E. et al. Storage e€ects on dentin permeability and shear bond strengths. *Dent Mater*, v. 9, p. 79-84, 1993.

KITASAKO, Y. et al. The influence of storage solution on dentin bond durability of resin cement. *Dent Mat*, v. 16, p. 1–6, 2000.

MOURA, J. S. et al. Influence of storage solution on enamel demineralization submitted to ph cycling. *J Appl Oral Sci*, v. 12, p. 205-8, 2004.

MUHLEMAN, H. R. Storage medium and enamel hardness. *Helv Acta Odont*, v. 8, p. 112–17, 1964.

NAKABAYASHI, N. et al. Dentin adhesion of "modified" 4-META/ MMA-TBS resin: function of HEMA. *Dent Mater*, v. 8, p. 259-64, 1992.

NAKAMICHI, I. et al. Bovine teeth as a possible substitute in the adhesion test. *J Dent Res*, v. 62, p. 1076-81, 1983.

PIMENTEL, E. et al. Armazenamento de dentes extraídos para estudos in vitro: revisão da literatura. *RBO*, v. 59, p. 224-26, 2002. RETIEF, D. H. et al. The effect of storage media and duration storage of extracted teeth on the shear bond strength of Scotchbond 2/Silux to dentin. *Amer J Dent*, v. 2, p. 269-73, 1989.

SCHILKE, R. et al. Comparison of the number and diameter of dentinal tubules in human and bovine by scanning electron microscopic investigation. *Arch Oral Biolog*, v. 45, p. 355-61, 2000.

STRAWN, S. E. et al. Spectroscopic changes in human dentine exposed to various storage solutions-short term. *J Dent*, v. 24, p. 417-23, 1996.

TITLEY, K. C. The effect of various storage methods and media on shear-bond strengths of dental composite resin to bovine dentine. *Arch Oral Biolog*, v. 43, p. 305-11, 1998.

ZISKIND, D. et al. Evaluation of cetylpyridinium chloride for infection control in storage solution. *J Oral Rehab*, v. 30, p. 477–81, 2003. MARANHÃO, Kalena de Melo, et al. The effect of solutions on enamel of bovine teeth. *Salusvita*, Bauru, v. 28, n. 2, p. 129-134, 2009.