Bulgarian Journal of Veterinary Medicine (2003), 6, No 3, 179–186

DENTAL PULP RESPONSE IN DOGS FOLLOWING INDIRECT CAPPING WITH POLYCARBOXYLATE CEMENT CONTAINING 5% POTASSIUM NITRATE

I. BORISSOV[1], S. TSANOVA[3] & D. SIVREV[2]

[1]Faculty of Veterinary Medicine, [2]Faculty of Medicine, Trakia University, Stara Zagora; [3]Faculty of Dentistry, Medical University, Plovdiv; Bulgaria

Summary

Borissov, I., S. Tsanova and D. Sivrev, 2003. Dental pulp response in dogs following indirect capping with polycarboxylate cement containing 5% potassium nitrate. *Bulg. J. Vet. Med.*, **6**, No 3, 179–186.

Histomorphological studies upon canine dental pulp response following indirect capping with polycarboxylate cement containing 5% potassium nitrate were performed on 42 experimental teeth. The experiment included also another 6 control teeth from dogs. All animals were mixed-breed. Fourteen experimental and 3 control teeth were extracted at post treatment days 7, 14 and 30. The microscopical preparations were done according the routine histological methods. The light microscopic study showed that the reactive inflammation of the dental pulp disappeared gradually and after the 30th day, the normal structure of the odontoblastic layer was restored and areas with reparative dentin were formed. Histomorphometric data evidenced that polycarboxylate cement containing 5% potassium nitrate stimulated both pulpal defense and regeneration which resulted in a better recovery in the experimental groups than in controls.

Key words: indirect capping, polycarboxylate cement, pulpal response

INTRODUCTION

Classical nonvital methods for pulp inflammation treatment are widely used, but are not sufficiently adequate (Borissov and Sivrev, 2002; Ward, 2002). A lot of studies aiming at improving these methods have been performed (Sinai *et al.*, 1967; Lambjerg-Hansen, 1974; Jokinen *et al.*, 1978; Swift and Trope, 1999; de Souza *et al.*, 2001).

Various medications are applied on pulpal tissue either directly (Pavlica *et al.*, 2000) or indirectly (Falster *et al.*, 2002) with a various success, depending on the character of dental pathology. After the middle of the 20th century, the methods of treatment preserving the vitality of the dental pulp became widely recommended as appropriate from the biological point of view (Santini, 1985, 1986; Sazak *et al.*, 1996; Smith, 2002).

Attempts for vital pulp treatment of cariously exposed non-inflammated pulp in permanent teeth were made (Ward *et al.* 2002) and the pulpal response after the application of restoring composites for direct capping of pulp chamber was evaluated (Kitasako *et al.*, 2002). It was concluded that those materials were not approproate because of the toxicity of their monomers.

During the last 50 years the calcium hydroxide preparations were most com-

monly used for treatment of caries (Green, 1977; Staehle, 1990; Schuurs *et al.*, 2000; Faraco and Holland, 2001) not only because of their antibacterial effect, but also because of the stimulation of dentin genesis in the peripheral pulpal layer. At the end of the last century, the deep caries with toxic pulpal response was treated with a remarkable success by calcium phosphate (Maksimovskii and Zemskova, 1994).

At the same time, during the last decade of the 20th century, many authors revised the protective properties of calcium hydroxide (Green, 1977; Schroder, 1985; Staehle, 1990; Murray *et al.*, 2001) that dissolved after an year and created prerequisites for secondary bacterial invasion and consequently, a pulp inflammation.

The aim of the present study was to evaluate the dental pulp response by histological methods after indirect capping with polycarboxylate cement containing 5% potassium nitrate.

MATERIALS AND METHODS

Experimental animals

The studies were performed on 48 teeth in 6 mixed-breed dogs aged between 2 and 3 years and weighing 15−20 kg. The teeth (canines, premolars and molars) were divided as followed: 42 experimental teeth divided in 3 subgroups with 14 teeth in each and 6 control teeth divided in 3 subgroups with 2 teeth in each. Two weeks prior to the experiment and during the study, the animals were fed and housed uniformly.

Anaesthesia

Following premedication with 0.02-0.04 mg/kg atropine sulfate (Sopharma, Sofia, Bulgaria) for reducing the salivation and preventing the respiratory and cardiac failures (Borissov and Sivrev, 2002), an intravenous anaesthesia with xylazine (Rometar, Spofa, Prague, Czech Republic) and ketamine hydrochloride (Imalgène, Rhône Mérieux, France) was performed according to the scheme of Zetner (Borissov and Sivrev, 1995). The anaesthetic effect occurred after 15−20 min and lasted for 30−60 min.

Teeth preparation

Under both water and air cooling for prevention of thermal irritation of the pulp, 3 ×5 mm cavities of class V were prepared in buccal surfaces of experimental teeth near to the gingival edge. The cavity floor was thinned to a pulpal dentin thickness of 0.5 mm using a round sterile borer. After washing of cavity with sterile saline and drying with sterile cotton buds, the floor of the cavity was capped with polycarboxylate cement containg 5% potassium nitrate, shaped as a protective obturation base. The same methods of preparation were used in control groups, but with a calcium hydroxude paste (Calxyd, Spofa Dental, Prague, Czech Republic) as a protective base. All cavities were hermetically closed with a temporary seal, containing zinc oxide 30.8 g, calcium sulfate 30.8 g, zinc sulfate 15.4 g, vaseline 22.6 g and menthol oil 0.4 g (Cavit, Spofa Dental, Prague, Czech Republic).

Teeth specimens

At post treament days 7, 14 and 30, fourteen experimental and 2 control teeth were extracted using the same schedule of premedciation and anaesthesia. The specimens were processed immediately after their obtaining.

Histological studies

With a diamond separator and water cooling, a significant amount of dental roots was removed. The rest of the tooth was cut into 3 mm slices and fixed in 10% neutral buffered formalin for a month. The fixed slices were decalcinated with bichromium trioxide and were further processed by the routine histological procedure.

Decalcinated dental slices were embedded in paraffine blocks in a thermostat at 56 ^{o}C with a subsequent cooling and used for preparation of 5-6 μm histological sections via a microtome. After deparaffination, hydratation and clearing, the sections were stained using three classical histological staining techniques: with haematoxillin/eosin (for histomorphological examination), with acid picrofuchsin according to Van Gieson (for observation of collagen fibres) and according to Malori (for observation of reticular fibres). After fixation and covering on standard observation glasses, the stained experimental and control histological sections were observed on an Amplival light microscope at magnifications of 6.3 ×10; 10 ×10 and 40 ×10.

Assessment of findings

The changes in dental pulp and the degree of inflammation were evaluated using the Negm scale as followed: weak changes (hyperaemia, stasis), medium changes (exudation, cellular migration) and considerable changes (cellular infiltration, tissue acidosis).

The reduction in odontoclast counts was calculated according to Qvist indexes: 0 − no reduction; 1 − insignificant reduction and 2 − medium to significant reduction.

The quantity of reparative dentin was determined by the method of Stanley on a 3−degree scale: 0 − no reparative dentin; ½ − initial formation of reparative dentin and 1− presence of a thick layer of reparative dentin.

RESULTS

The results obtained from the evaluation of teeth with experimental traumatic pulpitis and indirect capping with polycarboxylate cement with 5% potassium nitrate (experimental teeth) and Calxyd (control teeth) were as followed:

By the 7th day, the experimental teeth were characterized by a weak degree of

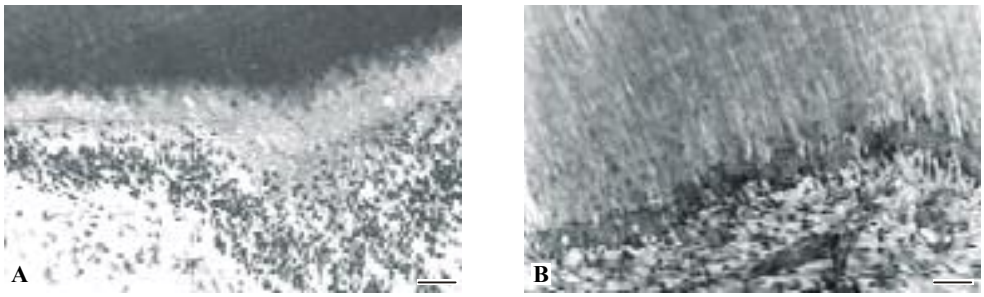

Fig. 1. Morphological changes in the dental pulp of a dog 7 days after indirect capping with polycarboxylate cement containing 5% potassium nitrate. A − experimental tooth; B − control tooth. Bar = 5 μm.

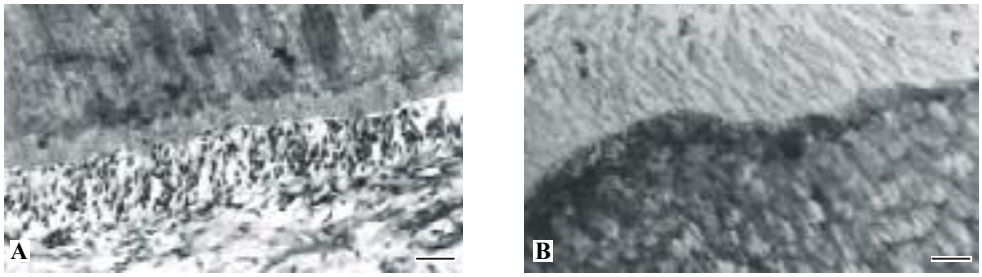

Fig. 2. Morphological changes in the dental pulp of a dog 14 days after indirect capping with polycarboxylate cement containing 5% potassium nitrate. A – experimental tooth; B – control tooth. Bar = 8 μm.

inflammation with hyperaemia, insignificant cellular infiltrate and a slight pulpal oedema (according to Negm) (Fig. 1A). The Qvist index was 1 with desorganization and insignificant reduction in some areas of odontoblasts. No cell penetration, related to odontoblast layer inflammation was detected. The Stanley degree was 0 − a reparative dentin was lacking, but the predentin band was thinned and the structure of dentin tubules at the cavity floor were changed.

The histological picture in the control teeth (Fig. 1B) was not significantly different from that in the experimental one.

By day 14, the experimental teeth (Fig. 2A) manifested a weak degree of inflammation (according to Negm) with a slight oedema, primarily in vicinity to the cavity, under the odontoblasts. In the remaining part of the dental pulp, a hyperaemia was observed, related to vasodilatation of vessels, tightly packed with erythrocytes. The Qvist index was 0, with zones of desorganization and increase in odontblast rows, arranged regularly by 10−12 in an close vicinity to the cavity. The Stanley degree was zero again − the thickness of the prodentin band was regular and well defined and in some areas was widened with infiltration of odontoblastic cells.

The Van Gieson staining revealed bundles or single collagen fibers that were in a lesser amount immediately under the cavity floor, probably because of the preexisting oedema in this part of the dental pulp. Increased fibroblast counts, due to enhanced fibroplastic function of the pulp were noticed.

In control teeth (Fig. 2B), the inflammation was weak with a persistent hyperaemia and a minor inflammation infiltrate including mainly fibroblasts and round cells. The Qvist index was 0 (no reduction in odontoblast counts). The Stanley degree was also zero (no formation of reparative dentin).

By day 30 (Fig. 3A), there was no inflammation by Negm (no hyperaemia, no oedema, vasodilatation and inflammation infiltrate) in experimental teeth. The Qvist index was 0. The odontoblasts were arranged evenly in 3−4 rows in a close proximity to the cavity. No reduction in odontoblast counts was present. The Stanley degree of reparative dentin amount was ½ to 1. The predentin band was wide with droplet-like shaped calcium deposits in some areas. Signs of dentinopoesis were visible as large odontoblasts with deeply coloured nuclei as well as newly formed reparative dentin. It was

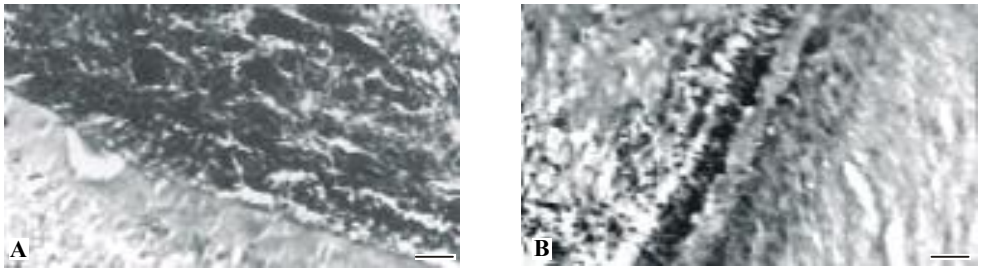

Fig. 3. Morphological changes in the dental pulp of a dog 30 days after indirect capping with polycarboxylate cement containing 5% potassium nitrate. A – experimental tooth; B – control tooth. Bar = 12.5 μm.

with bigger channeles compared to the normal one. The channels were round, oblique or S-shaped depending on the direction of the section.

The Van Gieson staining showed red coloured, uniformly arranged bundles of collagen fibers, many of them immature with fibrocytes within and a well pronounced fibroblast proliferation. The observation of Malori-stained preparations showed that the pulpal reticular network was preserved and the reticular fibres − grouped around the blood vessels.

In controls (Fig. 3B), the predentin band was well visible, the odontoblasts − uniformly arranged in 5-6 rows, with droplet-like calcium deposits (Qvist index 0). Sometimes, sideways from the cavity, a persistent hyperaemia was present. The Stanley degree of reparative dentin was ½ and its channels were wider than normal and evenly arranged.

DISCUSSION

Our histomorphometrical results showed clearly that following the indirect capping with polycarboxylate cement containing 5% potassium nitrate, the dental pulp reacted by a local exudative inflammation manifested by hyperaemia, insignificant cellular infiltrate and a slight pulpal oedema. The changes in normal pulpal structure were observed immediately under the dentin wall in cavity floor and were most probably due to traumatic and in a lesser extent − to thermal challenge during the cavity preparation, but not to the effect of polycarboxylate cement upon the pulp. Similar changes following mechanical manipulation of hard dental tissues were reported by Ivanova *et al.* (1990), Smith (2002) and Murray *et al.* (2002). Within the period between the treatment and the 7th post treatment day, there were no differences between experimental and control teeth.

At the 14th day, the odontoblastic layer began to recover with an even arrangement of odontoblasts. The predentin band in expeirmental teeth was wider and clearly outlined compared to control specimens, there were also areas with increased odontoblastic rows. Signs of reversibility of dental pulp inflammation (increased fibroblast counts) were detected.

By day 30th, the inflammation was disappeared in both groups of teeth − there were no signs of hyperaemia, vasodilatation, oedema and inflammation infiltrate in dental pulp. The normal structure of

odontoblastic layer was restored in both groups, but reparative dentin was better manifested in experimental teeth. Its dentin channels were wider and the boundary with the dentin was irregular compared to control preparations, where reparative dentin was well outlined by predentin and dentin.

Our histomorphometrical data support and add to the observations of Hodosh *et al.* (1983a) indicating a strong antiacidotic and anti-inflammatory effect of alkaline medium created by potassium nitrate after a biological treatment of pulp inflammation with potassium nitrate and polycarboxylate cement. The same authors (Hodosh *et al.*, 1983b; Hodosh *et al.*, 1991) followed out only the clinical effects of the treatment without objective data about the response of dental pulp or periapical tissue. In our experimental studies, the advantages of polycarboxylate cement containing 5% potassium nitrate as a preferred alternative to calcium hydroxide preparations for dental pulp capping were objectively evidenced by histomorphometry.

REFERENCES

Borissov, I. and D. Sivrev, 1995. Veterinary Dentistry. NIS, Stara Zagora, pp. 32–33.

Borissov, I. and D. Sivrev, 2002. Veterinary Dentistry and Oral Surgery. Universe, Stara Zagora, pp. 51–53; 101–106.

de Souza Costa, Q. A., A. B. Lopes do Nascimento, H. M. Teixeira and U. F. Fontana, 2001. Response of human pulps capped with a self-etching adhesive system. *Dental Materials,* **17**, No 3, 230–240.

Falster, C. A., F. B. Araujo, L. H. Straffon and J. E. Nor, 2002. Indirect pulp treatment: *in vivo* outcomes of an adhesive resin system vs calcium hydroxide for protection of the dentin-pulp complex. *Pediatric Dentology*, **24**, No 3, 241–248.

Faraco, I. M. Jr and R. Holland, 2001. Response of the pulp of dogs to capping with mineral trioxide aggregate or a calcium hydroxide cement. *Dental Traumatology,* **17**, No 4, 163–166.

Green, B., 1977. Calcium hydroxide and potassium nitrate as desenzitizing agents for hypersensitive root surfaces. *Journal of Periodontology,* **48**, No 10, 667–672.

Hodosh, A. J., S. H. Hodosh and M. Hodosh, 1983a. Potassium nitrate-zinc oxide eugenol temporary cement for provisional crowns to diminish post-preparation tooth pain. *Journal of Prosthetic Dentistry,* **70**, No 6, 493–495.

Hodosh, M, S. H. Hodosh, G. Shklar and A. J. Hodosh, 1983b. Potassium nitrate: an effective treatment for pulpitis. *Oral Surgery, Oral Medicine, Oral Pathology*, **55**, No 4, 419–420.

Hodosh, M., S. H. Hodosh and A. J. Hodosh, 1991. Maintenance of pulpal vitality using potassium nitrate-polycarboxylate cement cavity liner. *Quintessence International*, **22**, No 6, 485–502.

Ivanova, Z., S. Tsanova and P. Botushanov, 1990. Clinical investigations on the effect of biological treatment of reversible pulpitis with polycarboxylate cement containing 5% potassium nitrate. In: Proceedings of the XV National Dentistry Congress, pp. 10–14.

Jokinen, M. A., R. Kotilainen, P. Poikkeus, R. Poikkeus and L. Sarkki, 1978. Clinical and radiographic study of pulpectomy and root canal therapy. *Scandinavian Journal of Dental Research*, **86**, No 5, 366–373.

Kitasako, Y., P. E. Murray, J. Tagami and A. J. Smith, 2002. Histomorphometric analysis of dentinal bridge formation and pulpal inflammation. *Quintessence International,* **33**, No 8, 600–608.

Lambjerg-Hansen, H., 1974. Vital and mortal pulpectomy on permanent human teeth. An experimental comparative histologic investigation. *Scandinavian Journal of Dental Research*, **82**, No 4, 243–332.

Maksimovskii, M. and M. Zemskova, 1994. The use of a calcium phosphate ceramic in treating deep caries. *Stomatologiya* (*Moskow*), **73**, No 2, 14–17 (RU).

Murray, P. E., I. About, J. C. Franquin, M. Remusat and A. J. Smith, 2001. Restorative pulpal and repair responses. *The Journal of the American Dental Association*, **132**, No 4(10), 482–491.

Murray, P. E., P. J. Lumley and A. J. Smith, 2002. Preserving the vital pulp in operative dentistry: 3. Thickness of remaining cavity dentine as a key mediator of pulpal injury and repair responses. *Dental Update*, **29**, No 4, 172–178 .

Pavlica, Z., P. Juntes and M. Pogacnik, 2000. Defence reaction in dental pulp after pulp capping and partial pulpectomy in dogs. *Acta Veterinaria Hungarica,* **48**, No 1, 23–34.

Santini, A., 1986. Long-term clinical assessment of pulpotomies with calcium hydroxide containing Ledermix in human permanent premolars and molars. *Acta Odontology Pediatrics*, **7**, 2, 45–50.

Santini, A. H., 1985. Intraoral comparison of calcium hydroxide (Calnex) alone and in combination with Ledermix in first permanent mandibular molars using two direct inspection criteria. *Journal of Dentistry*, **13**, No 1, 52–59.

Sazak, H., M. Gunday and C. Alatli, 1996. Effect of calcium hydroxide and combinations of Ledermix and calcium hydroxide on inflamed pulp in dog teeth. *Journal of Endodontics*, **22**, No 9, 447–449.

Schroder, U., 1985. Effects of calcium hydroxidecontaining pulp-capping agents on pulp cell migration, proliferation and differentiation. *Journal of Dental Research,* **65**, No 4, 541–548.

Schuurs, A. H., R. J. Gruythuysen and P. R. Wesselink, 2000. Pulp capping witrj adhesive resin-based composite vs. calcium hydroxide: a review. Endodontic *Dental Traumatology*, **16**, No 6, 240–250.

Sinai, I., S. Seltzer, W. Soltanoff, A. Goldenberg and I. B. Bender, 1967. Biologic aspects of endodontics. II. Periapical tissue reactions to pulp extirpation. *Oral Surgery, Oral Medicine, Oral Pathology*, **23**, 5, 664–679.

Smith, A. J., 2002. Pulpal responses to caries and dental repair. *Caries Research,* **36**, No 4, 223–232.

Staehle, H. J., 1990. Calcium hydroxide in der Zahnheikunde. *Quintessence of Dentistry*, **94**, No 4, 13–16.

Swift, E. J. and M. Trope, 1999. Treatment options for the exposed vital pulp. *Practical Periodontics Aesthetic Dentistry*, **11**, No 6, 735–739.

Ward, J., 2002. Vital pulp therapy in cariously exposed permanent teeth and its limitations. *Australian Endodontic Journal*, **28**, No 1, 29–37.

Paper received 11.04.2003; accepted for publication 09.07.2003

Correspondence:
Assoc. Prof. I. Borissov
Department of Veterinary Surgery,
Faculty of Veterinary Medicine,
Trakia University,
6000 Stara Zagora, Bulgaria