Human pulp response after direct pulp capping with an adhesive system

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ABSTRACT

Background: The adhesive systems are experimentally used in direct pulp capping procedure. The aim of this study was to evaluate the histological responses of pulp tissues after direct pulp capping with an adhesive agent.

Materials and Methods: The pulps of twenty human teeth scheduled for extraction were mechanically exposed and capped with an adhesive system and calcium hydroxide cement for 60 days. The teeth were extracted and prepared for examination under light microscope for inflammatory responses and dentin bridge formation.

Results: The slides showed that there were variable responses which ranged from moderate to severe and progressive extension of tissue necrosis with time for teeth capped with the adhesive system. After the application of calcium hydroxide paste, only mild tissue response was elected with complete hard tissue bridging.

Conclusion: Based on the experimental conditions, the application of the adhesive system in direct contact with mechanically exposed pulp cannot lead to acceptable repair of dentin-pulp complex.

Keywords: Adhesive, pulp capping, calcium hydroxide, pulp response. **(J Bagh Coll Dentistry 2006; 18(1) 25-29)**

INTRODUCTION

Direct pulp capping is a therapeutic method aimed at treating reversible pulpal injury whenever dentine and pulp is affected by caries, restorative procedures or trauma. It is generally accepted that wound healing after pulp capping treatment is not unique to a given treatment modality [(1)].

An ideal direct pulp capping material would maintain the vitality and function of the dental pulp, form a dentine bridge, have appropriate mechanical properties and adhesion to dentine (to prevent microleakage), and be simple to handle clinically. No direct pulp capping material has completely satisfied these requirements [(2)].

Traditionally, various formulations of $Ca(OH)_2$ -containing materials represent the more usual treatment modalities in capping treatment, due to their favorable effects (experimentally and clinically) for more than 4 decades and ability to stimulate reparative dentin formation [(3)].

However, the physical limitations of $Ca(OH)_2$ 4 (non-adherence to dentine, dissolution in tissue fluids, degradation) have led scientists to seek new approaches.

A tight seal between the exposed pulp and the capping material is essential to pulpal healing.

During the last decade, dentine adhesives have been proposed as an alternative to $Ca(OH)_2$ based materials in direct pulp capping treatment. It has been claimed that a properly hybridized dentine-adhesive interface can seal both dentine and pulp effectively, thus protecting the wound area from additional injury and post-operative infection and permitting complete tissue healing through the inherent capacity of the dentine-pulp complex 5-7.

However, acid etching is a short procedure that is completely rinsed off by water and buffered by surrounding dentin and/or dentinal fluids. Therefore, the rationale behind the use of dentin bonding materials instead of the commonly used calcium hydroxide compounds has been that prevention of leakage by micromechanical interlocking of adhesive systems to cavity walls is more critical for the healing processes than the material used for capping [(8, 9)].

The aim of this study was to address and compare the effect of the adhesive on the pulp response of human teeth.

MATERIALS AND METHODS

Twenty human premolars scheduled to be extracted for orthodontic reasons were selected from patients ranging from 14 to 18 years old. All teeth were clinically and radiographically examined to assure absence of proximal caries and periapical lesions. Thermal testing was applied for 5 seconds on the buccal surface of the teeth scheduled for the pulp therapy, and adjacent teeth. After local anesthesia, rubber dam isolation was installed. Mesio-occlusal cavities, with 0.5 mm beyond the cementum–enamel junction, were prepared by means of sterile diamond burs at high speed under water/spray coolant, the cavity dimensions

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were: occlusal depth, 2 mm; axial depth, 4 mm; box width, 3.5 mm 6.

Pulp exposure was performed in the center of the pulpal floor by means of a round diamond bur, with a tip diameter 0.3 mm, under water cooling. One bur was used for each cavity. The teeth were then divided into two experimental groups (n=10) .

The pulp hemorrhage was controlled by abundant irrigation with saline solution followed by the application of a damp cotton pellet embedded in saline solution and held in place for one minute. Paracetamol 500 mg was prescribed as an analgesic for pain relieving that may be happened post-operatively.

In group 1, a metal matrix band was installed and enamel, dentin and pulp were conditioned with phosphoric acid for 20 s. The acidic agent was rinsed off, slightly dried in such way that the dentin stayed visibly moist with a shiny surface. Two coats of the adhesive agent (Quadrant Uni One Bond; Cavex, Holland, Batch # 10060) were subsequently applied and light-cured for 20 s. Increments of composite (Quadrant Universal LC. Cavex, Holland, Batch # 44098) were used to restore the cavities. Each increment was light-cured (Coltolux 50 Colten –France) for 40 s.

In group 2, calcium hydroxide cement (Urbical; Promedica, Germany, Lot # 480865) was applied on pulp exposure by means of a Dycal applicator after homestasis in the occlusal cavity floor. Then the restorative procedure was conducted as described in group 1.

During the observation period (60 days), the patients were frequently asked about the presence of sensitivity. After this period the extraction of each tooth was performed under local anesthesia 10.

The roots were sectioned in order to facilitate fixation in 10 %buffered formalin solution for 72 h. The samples were prepared according to normal histological techniques and embedded in paraffin. Six-micron thick sections were cut with a Microtome (Reichert Jung 2030, Japan) parallel to the main vertical axis of the tooth. The sections, mounted on glass slides were stained with hematoxylin and eosin (H/E). The sections were blindly evaluated by an experienced pathologist according to the criteria described in Table.1, using conventional light microscope (Olympus; Japan). The scores attributed to each group were subjected to non-parametric Kruskal–Wallis test which is used for independent samples ($P<0.05$).

Table 1: Scores criteria used for the examination.

Scores	Inflammatory cell response
1	Few scattered inflammatory cells beneath the exposure site
2	Acute & chronic inflammatory cells aggregation
3	Severe inflammatory reaction (abscess or dense infiltrate)
4	Completely necrotic pulp tissue
Scores	**Dentin bridge formation**
1	Formation of barrier tissue directly adjacent to the material
2	Formation of incomplete dentin bridge distance from the material
3	Absence of any dentin tissue formation

RESULTS

The two groups studied demonstrated different pulp tissue reactions to the materials, which are capped with. Pulp tissue was mainly infiltrated by polymorphnuclear cells, macrophages lymphocytes and plasma cells. Giant cells were rarely found only in a few specimens.

The percentage of scores for each group is shown in Table 2. The histological feature from group 1 was quite inferior to group 2.

Inflammatory infiltrate: An intense and chronic inflammatory response was observed in group 1 (60%). In 10%of the cases from group 1, none, or a few scattered inflammatory cells (Polymorphonuclear leukocytes (acute) or mononuclear lymphocytes (chronic)) were presented in the pulp beneath the exposure site. Completely necrotic pulp was observed in 20% of the cases in group 1 (Figure 2).

Dentin bridge formation: There was no evidence of any dentin tissue formation in specimens of groups 1. In group 2, an amorphous complete dentin bridge formation with irregular contour and varied mineralization degree was observed adjacent to the material in (90%) of specimens and in one case (10%) the dentin bridge was incomplete and slightly far

away from the material. Along with the dentin bridge, a similar odontoblastic layer was also observed Figure 1.

Pain: 30 % of the cases in group I were suffered from mild to moderate pain, only one case (10 %) in group II, and the pain was relieved by the analgesic. Two cases in group I were excluded because of severe pain post-operatively during the first week, the teeth had to be extracted, and did not enter the statistics.

Table 2: Percentage of scores for each group.

Groups	Inflammatory response					Bridge formation				Pain
Scores	1	2	3	4	(*)	1	2	3	(*)	
Adhesive System	10	60	10	20	S	0	0	100	NS	30%
Calcium hydroxide	80	20	0	0	NS	90	10	0	NS	10%

(*) Statistical significance (Kruskal–Wallis); S= Significant difference; NS= No Significant difference.

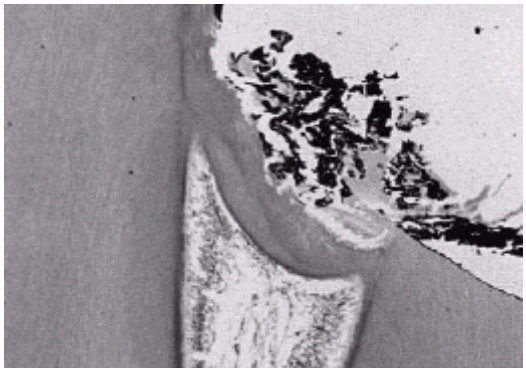

Figure 1: Dentin bridge formation for the $Ca(OH)_2$ group after 60 days (x100).

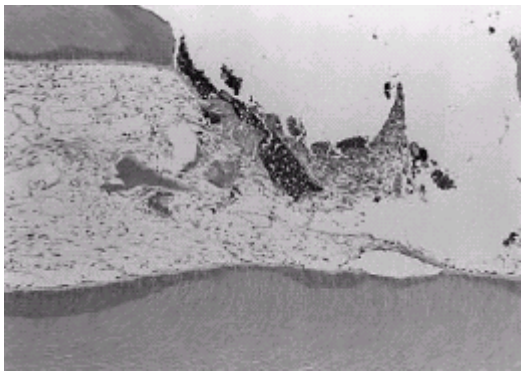

Figure 2: Slight inflammatory response without dentin bridge formation for adhesive capped group after 60 days (x100).

DISCUSSION

Different materials are used for management of direct pulp exposures that may be faced during the different operative procedures. Calcium hydroxide and its compounds are the materials of choice, and the materials against which new candidates for capping are tested because of a long record of success from clinical and histological studies. The ability of calcium hydroxide to stimulate reparative dentin formation, shown in the present investigation, has already been observed in many other clinical studies. In addition its antibacterial effect may be attributed to effects on induction and up-regulation of odontoblast-like cell differentiation for new matrix deposition and especially its effect on growth factors from the dentin matrix [11,12].

In this study, the application of a $Ca(OH)_2$ cement directly on the pulp exposure was characterized by very slight inflammatory cells infiltration, no tissue necrosis but partial to complete hard tissue bridging at the 60 days observation period. From a comparison of the results between the group of teeth treated with dentine adhesive system and this treated with $Ca(OH)_2$ it may be concluded that a low success rate (wound healing with tertiary dentine bridge formation) might be expected from pulp capping with the dentine adhesive system used, and these results are in complete agreement with Chen et al 13& Cox et al [14].

On the other hand, the results of the present investigation confirm that the adhesive solution may result in degenerative pulp alterations when placed directly over pulp exposure sites.

In this study, the application of the adhesive system directly on pulp exposure produced variable responses, characterized by moderate to severe inflammatory reaction in most teeth, progressive extension of tissue necrosis with time and a total absence of continuous hard tissue bridge formation. This is in agreement with several studies where pulp capping was performed with adhesive systems over human pulps [6,7,14].

The main constituents of adhesive solution are Hydroxyethyl Methacrylate (HEMA) and

Bisphenol A glycidyl Methacrylate (Bis-GMA), respectively. The high percentage of HEMA in the adhesive, its low molecular weight and hydrophilic features, facilitate its diffusion through pulp tissue and consequently avoid pulp healing [15].

Although these components were light-cured prior to restoration of the cavity, it is known that the conversion of monomers to polymers is never complete. All the organic components of the adhesive systems, photo-initiators and constituents generated during the setting process, are released, in particular shortly after setting [16,17].

The resin components (mainly HEMA) cause inhibitory effects on DNA synthesis, total protein content and protein synthesis. They may destroy the lipid membrane of pulpal cells if their local concentrations are high18. When resins are placed directly on exposed pulps, the high lipid solubility of the resins in the lipid-phase of biologic membranes may permit the resin monomers to reach cytotoxic concentrations within cell membranes. Furthermore, the resin monomers may affect the immune system adversely and induce immuno-suppression response [19,20].

It seems that the presence of resin particulates can trigger a foreign body response characterized by the presence of mononuclear inflammatory infiltrate as well as multinuclear giant cells. It is reported that as low as 3.6 mmol/L of HEMA can reduce cell metabolism to 50%after 24 h exposure [21,22].

This is likely to be responsible for chronic inflammatory response of pulps and also to necrosis that occurs after longer periods of evaluation. The adhesive resin might promote more intense pulp damage over time 23. Therefore, when the bonding was applied over pulp exposures (groups 1), no dentin bridging was found in this group. These result disagreed with those of Kitasako et al 5& Cox et al [24], who used an adhesive agent for capping of non-human pulp exposures.

Regarding pain experience through the experiment, many causes of this pain may be encountered, like the irritative effects of the materials that the pulp being capped with, the microleakage which may occur from the gingival margin of the box, and the subsequent infection that this microleakage cause 6.

Based on the results of the present experiment, it was concluded that the application of dentine adhesive systems, which are in direct contact with the mechanically-exposed pulp, cannot lead to acceptable repair of the dentine-pulp complex such as wound healing with tertiary dentine bridge formation, and the exposed pulp should be capped with calcium hydroxide liner to ensure successful pulp tissue healing.

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