

ORIGINAL ARTICLE

A resin-modified glass ionomer cement barrier for treating degree II furcation defects: A pilot study in dogs

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Abstract

Objective. The aim of this study was to evaluate in an animal model the healing of degree II furcation defects treated with: an experimental barrier of resin-modified glass-ionomer cement (GIC), a polylactic acid barrier (GUI), and flap surgery (CTR). **Material and Methods.** In 3 beagles, 18 class II furcation defects were surgically produced in mandibular and maxillary premolars and exposed to plaque accumulation for 21 days. Following a full flap, notches were made at the base to the bone defect. GIC barriers were prepared immediately before use from a commercial product and fit to place with the same product. The GIC barriers were removed after 30 days and the dogs euthanized after 120 days. Histologic sections were analyzed in a computer-assisted microscope. Epithelium, new cementum with inserting fibers, and connective tissue lining the root surface in-between notches were measured and medians of percentage values calculated. **Results.** In the GIC, epithelium constituted 3.5% (median values) of the notch-to-notch root area; new cementum was 83.6% and connective tissue 12.9%. These values were 0%, 73.6%, and 26.4% for the GUI group and 35.6%, 43.2%, and 0% for the CTR group. Bone fill median values were 54.3% for GIC, 20.6% for GUI, and 24.6% for CTR. **Conclusion.** GIC and GUI prevented epithelial migration and promoted the formation of new periodontal tissues in experimentally induced class II furcation defects in dogs.

Key Words: Biocompatibility, class II furcation defect, guided tissue regeneration, periodontitis

Introduction

Furcation involvements are recognized as a challenge to periodontal therapy. Guided tissue regeneration has been proposed as an attempt to solve furcation defects of different magnitudes by the formation of new periodontal tissues. Results from animal studies have been promising. Degree II and III defects, either induced or naturally occurring, have been shown to heal by apposition of bone, cementum, and periodontal ligament [1–4]. Several materials have been used as barriers, including expanded polytetrafluorethylene and absorbable barriers, such as collagen, polylactic and polyglycolic acids among others. More recently, absorbable and non-absorbable barriers have been compared, with inconclusive results [5,6]. Results from clinical trials have been more modest and furcation resolution restricted mostly to degree II defects in lower molars [7–11]. Different

reasons have been proposed to explain the limitations so far observed in clinical studies. Besides excluding cells, barriers should stabilize the wound and protect the clot in order to improve clinical results after regenerative therapies [12]. Additionally, they should create an adequate space where progenitor cells can migrate to and produce new periodontal attachment [13]. Another restricting factor is the occurrence of gingival recessions and consequently barrier exposure to the oral environment [11]. In addition, a negative association between the presence of microorganisms and the clinical attachment levels achieved have been pointed out in some clinical trials [14–16].

Considering the properties of glass ionomer cement (GIC), its use as a barrier might overcome some of these limitations. GIC adheres to the dental surface, thus fixing the barriers, maintaining

a healing space sealed from the outer environment, and stabilizing the clot. GIC is biocompatible and fluoride release has been associated with a potential antibacterial effect [17]. Furcation areas show a high degree of anatomical variation [18]. GIC barriers would be easily adaptable to these variations due to its adhesive capacities and malleability.

The hypothesis is that experimental GIC barriers would prevent epithelium migration and promote formation of periodontal ligament and bone in an experimental model of furcation defects in dogs. The aim of the present pilot study was to histologically analyze the periodontal healing response in degree II furcation defects in dogs after the use of an experimental GIC barrier.

Material and methods

Sample description

Ethical permission was obtained from the Ethics Committee of the Dental School of the Rio Grande do Sul Federal University. Three female, 12-month-old, systemically and periodontally healthy beagle dogs were used. In each animal, the third upper premolars and the third and fourth lower premolars were selected bilaterally. These teeth were randomly distributed in three groups, according to the following treatments: in each dog, four teeth were treated using resin-modified GIC barrier; one by a polylactic acid absorbable barrier (Guidor®; W. L. Gore & Associates, Inc., Flagstaff, Az., USA) (GUI) and one by flap surgery only (CTR).

Surgical production of the defects (Day 0)

A buccal mucoperiosteal flap was raised and degree II furcation defects were created through osteotomy with the dimensions of $2 \times 2 \times 2$ mm [10]. Bone removal was restricted to the furcation area, leaving the interproximal areas intact. The exposed root surfaces were scaled. Impressions of the furcation areas were obtained with a silicon material in order to obtain acrylic resin models used for prefabrication of the GIC barriers. The furcations were then filled with the same impression material, to prevent spontaneous regeneration of the defects, and the flaps were sutured. Plaque was allowed to accumulate over the course of 21 days. After this period, the impression material was removed with the aid of curettes. Supragingival scaling was performed. At this moment, a chemical plaque control regimen, consisting of a daily application of 2% chlorhexidine gel applied with a soft toothbrush, was started and lasted until the experimental surgical procedure (Day 42).

Barrier production

Resin-modified GIC (Vitremer; 3M Dental Products, St. Paul, Minn., USA) was prepared following the manufacturer's instructions. The material was pressed between two glass plates under a 500 g weight for 30 s and light-cured for 30 s. A 0.2-mm layer of GIC was obtained and adjusted to cover the furcation area and the adjacent bone onto the acrylic resin models. These procedures were performed 24 h before the experimental surgery. Barriers were kept in a sterile dry environment until use.

Surgical experimental procedure (Day 42)

After sedation and anesthesia, mucoperiosteal flaps were raised and the exposed root surfaces were scaled and planed. Reference notches were made at bone level with a $33\frac{1}{2}$ burr as a guideline to histometric analysis. Barriers were placed as follows: (1) GIC group: following drying of the surfaces with cotton pellets, the primer was applied at the root surface. Barriers were fixed to the root surface with the aid of a portion of freshly prepared GIC applied over the borders of the barrier in contact with the root surface followed by light curing for 30 s; (2) GUI group: polylactic barriers were adjusted and sutured covering the furcation defect and part of the bone. In both groups, flaps were coronally positioned to provide full coverage of the barriers and subsequently sutured. In the CTR group, the same surgical procedures were performed, but with no barrier placement.

Post-surgical phase (Day 42 to Day 162)

The animals received amoxicillin 500 mg, once a day, for 7 days, and chemical plaque control continued as described above. They were clinically examined once a week. GIC barriers were removed after 30 days.

Euthanasia (Day 162) and histological procedures

The animals were euthanized with an overdose of anesthetic and immediately perfused with 10% formalin. Block sections were decalcified in Morse solution [19] and routinely processed for paraffin embedding. Six-mm-thick step serial sections were obtained in the mesio-distal direction and HE stained. Six to nine sections per tooth, corresponding to the first and last sections that contained notches in both roots, and four to seven intermediate sections selected between them, were recovered for histological measurements [2].

Analysis of the results

Histological sections were analyzed in a computer-assisted microscope and specific software (Standard

20; Carl Zeiss Inc., USA; ImageLab, Brazil) by a blinded and calibrated examiner. Epithelium, new cementum with inserting fibers, and connective tissue lining the root surface between the notches were measured. Means and standard deviations were calculated for each treatment and the dog was considered the unit of analysis. Median percentages of the coverage by epithelium, new cementum with inserting fibers, and connective tissue in relation to the total linear notch-notch extension were calculated for each treatment. The area of bone fill expressed as a percentage of the total area of the furcation defect was measured and medians/percentiles were calculated.

Results

All the furcation defects, independently of the treatment group, showed similar behavior during the healing phase (Days 42 to 162). The animals were free of gingivitis and marked gingival recessions were not observed. One out of 12 GIC barriers was lost one week after its placement.

Descriptive histology

In all furcations of the CTR group (Figure 1A), epithelium was observed lining almost the entire length of the root surface between notches. Newly formed cementum with inserting periodontal fibers was noted only at the level of the notches. The defects were almost completely filled with connective tissue and varying amounts of bone.

In the GUI group (Figure 1B), besides the formation of new cementum, a small amount of newly formed bone was observed in the area adjacent to the furcation fornix. Granulomas located in the apical area of the defects were observed in two out of three specimens that received the absorbable barrier. In one of the defects of this group, epithelium proliferation was present without evidence of cementum formation.

In the furcation defects treated with GIC barriers (Figure 1A,B), newly formed cementum with a cellular pattern could be observed. Periodontal fibers were present inserting perpendicularly and obliquely in the cementum and in the bone. Bone formation was variable. In some specimens, although new cementum was observed, there was little or no bone formation. Some areas of the defects presented connective tissue with collagen fibers running parallel to the root surface without evidence of cementum. Resorption areas were observed in all three groups and in most cases the lacunae were filled with cementum. Areas showing ankylosis were not identified in any of the groups.

Histometric assessments

Of the 18 teeth initially treated, 2 from the GIC group were excluded from the histometric evaluation, 1 because the barrier was lost and the other because the notches could not be identified.

Means and standard deviations of the histometric measurements for each treatment are presented in Table I. The median of the percentage values of newly formed cementum in relation to the total

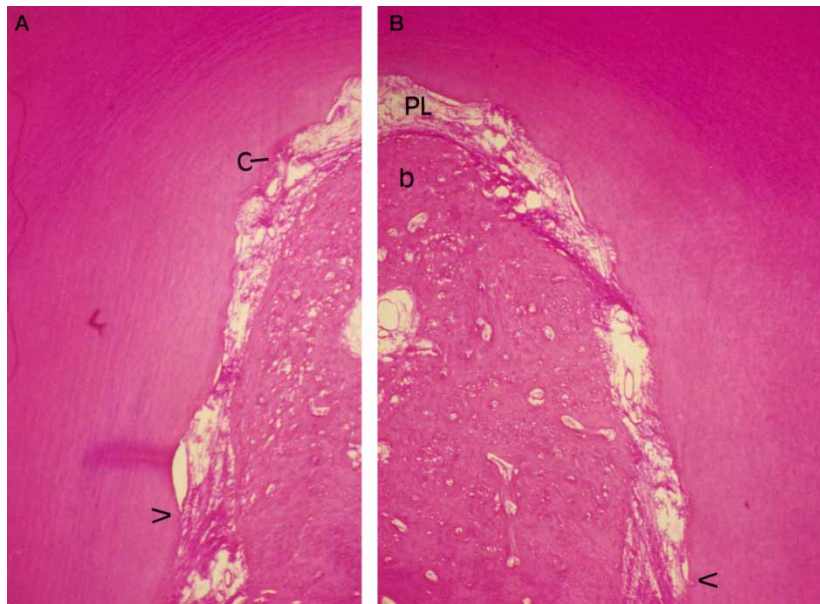


Figure 1. A. Mesio-distal aspect of the furcation defect in the control group (E: epithelium, CT: connective tissue). Arrows indicate apical border of notches. H/E staining, original magnification 25 ×. B. Mesio-distal aspect of the furcation defect in the GUI group (C: cementum, PL: periodontal ligament, B: bone tissue, G: granuloma). Arrows indicate apical border of notches. H/E staining, original magnification 25 ×. A,B. Mesio-distal aspect of the furcation defect in the GIC group (C: cementum, PL: periodontal ligament, B: bone tissue). Arrows indicate apical border of notches. H/E staining, original magnification 25 ×.

Table I. Means (\pm standard deviations, in mm) of the linear extension of cementum, connective tissue, and epithelium present in the total linear extension of the furcation defect, according to groups

Group	n	Total linear extension	Cementum	Connective tissue	Epithelium
CTR	3	5.08 \pm 0.20	2.35 \pm 2.12	0.32 \pm 0.56	2.39 \pm 1.24
GUI	3	4.16 \pm 0.67	2.36 \pm 2.04	0.95 \pm 0.84	0.85 \pm 1.47
GIC	3	4.51 \pm 0.74	3.61 \pm 1.00	0.79 \pm 0.54	0.12 \pm 0.10

linear extension between the notches was 43.2% for the CTR group, 73.6% for the GUI group, and 83.6% for the GIC group. In the control group, the median of connective tissue present was 0% of the linear extension between notches, 26.4% in the GUI group and 12.9% in GIC group. Epithelium lined 35.6% of the linear extension in the CTR group, 0% in the GUI group, and 3.4% in the GIC. The median percentages of newly formed bone tissue in relation to the total area of the defects were 24.6% in the CTR group and 20.6% and 54.3% in GUI and GIC groups, respectively.

The presence of epithelium at the buccal, middle, and lingual thirds of the furcation defects was analyzed and results are given in Table II.

Discussion

The present study reports preliminary observations of the healing response of degree II furcation defects following the use of experimental GIC barriers in an animal model. The presence of new bone and cementum with inserting periodontal fibers was observed both when GIC and polylactic acid barriers were used and, to a limited extent, also in the control sites.

The number of animals used is admittedly small, but judged adequate both for ethical and experimental purposes of a pilot study. One of the limitations of the dog model is the spontaneous regeneration reported in different studies [1,2]. In the present study, 43.2% of the linear extension between notches was covered with new cementum with inserting fibers in the control group. This is in accordance with other results using a similar model [2]. Spontaneous healing is reported to occur also following treatment of naturally diseased sites [1]. The amount of newly formed cementum was enhanced when both GUI and GIC barriers were used, resulting in 73.5% and 83.6% of the linear notch-to-notch extension, respectively. These values are in

accordance with the figures reported by different authors employing both resorbable and non-resorbable barriers [1,5,20].

The median values of bone formation were 24.6% in the CTR group and 20.6% and 54.3% in the GUI and GIC groups, respectively. There is a difference in the apposition timing of new bone and new cementum. Possibly, a longer period of observation could elucidate this. The restricted result of the GUI group may be related to the granuloma-like tissue observed. Gottlow et al. [21] observed that macrophages and multinucleated cells were present 3 to 6 months after GUI barrier placement. However, in the 24-month analysis, a complete periodontal regeneration was observed. Araujo et al. [4] observed that the foreign-body reaction elicited by this barrier may have hindered bone formation without significantly reducing the formation of new cementum. In the present study, it was observed that areas far from the granuloma showed new bone formation.

Epithelium could not be identified at the furcation sites treated with GUI barriers, whereas at 4 out of 10 sites where GIC was used epithelium was present in the outer third (buccal). Only one of these sites showed epithelium in areas deeper than that. Restricted epithelial proliferation has been reported in other studies using non-resorbable barriers [5,6]. The time period that the membrane is left in place may be of importance, even though epithelium was observed when barriers were left on site for 30 days or 3 months [5]. Tissue accommodation following removal of the membrane may also be associated with the presence of epithelium in these areas.

No adverse reactions, such as abscesses or edema, were observed in the sites treated with the experimental barrier. It is possible that the antibiotics used post-operatively and the continuous chemical plaque control regimen might have been associated with this observation. The reported antibacterial properties of the GICs may also be of importance [22].

Regenerative attempts started with the use of millipore filters [23]. Other alternative materials tested have included a rubber dam [6]. A case reported by Abitbol et al. [22] has shown the clinical potentialities of GIC membranes in the treatment of furcation defects. The good clinical response observed by those authors was confirmed in this study and may be associated with the histological events described here. Several of the limitations

Table II. Presence of epithelium in the buccal, middle, and lingual thirds of degree II furcation defects, according to groups

Groups	Buccal	Middle	Lingual
CTR	3 (3)*	3 (3)	3 (3)
GUI	1 (3)	1 (3)	1 (3)
GIC	4 (10)	1 (10)	0 (10)

*Teeth with presence of epithelium (teeth treated).

associated with soft barriers could be overcome with the use of a GIC membrane. It can easily adapt to the furcation area even when high trunks are present, as sutures are not needed to secure the barriers. Healing space and cloth protection could be provided by the stiff nature of the material. In the present report, the principle of tissue regeneration in the furcation area was shown to occur with the use of GIC barriers in a limited experimental protocol. Further studies are needed, however, before a clinical recommendation can be made.

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