Histologic study of furcation perforations treated with MTA or Super EBA in dogs' teeth

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Objective. The aim of this study was to investigate the histologic response to MTA or Super EBA when used for the repair of furcation perforations in dogs' teeth.
Study design. Ninety mandibular premolar and molar teeth of 9 mongrel dogs were used in this study. The teeth were divided into 3 groups. Seventy-two teeth were repaired with either MTA or Super EBA (36 each), and 18 teeth were not repaired and used as negative controls. All groups were histologically examined at 1 month, 3 months, and 6 months after treatment. Histologic evaluation was done with regard to inflammation and type of healing.
Results. The Super EBA group showed moderate inflammation in 1 month; the inflammation decreased over time, but most of specimens showed inflammatory reaction from mild to severe at the end of 6 months. The perforation area was filled by connective tissue in specimens in which no inflammation was seen. In the MTA group, mild inflammation was seen in 1 month, it decreased in 3 months, and no inflammation was detected in 6 months. New cementum formation was taken in place in 4 specimens in 1 month, in 8 specimens in 3 months, and in all specimens in 6 months.
Conclusions. MTA showed less inflammation than Super EBA. MTA specimens showed healing with new cementum formation in the perforation area, whereas Super EBA specimens in which no inflammation was seen showed connective tissue healing.
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A root perforation is an artificial communication between the root canal system and the supporting tissues of the tooth. Perforations can result from a resorptive process or can be produced iatrogenically. Iatrogenic perforations in the furcation area may occur throughout the course of endodontic access opening due to an incorrectly directed bur, during post-space preparation, or when trying to locate calcified pulp chambers and canals. A perforation has serious clinical consequences and requires intervention. The trauma of the perforation and subsequent inflammation may rapidly produce a communication with the gingival sulcus and an irreversible periodontal lesion.[1] Although these defects are sealed off immediately, unfavorable tissue reactions may occur.[2,3]

Long-term success of a perforation repair has been shown to depend upon the duration of septic exposure, the size and location of the perforation, and the degree of insolubility of the material and the ability to seal the defect.[4-6] However, the major difficulty with non-surgical repair is extrusion of the filling material into the periodontal space, which interferes with the periodontal reattachment.[7] Although many materials have been used for repair of furcation perforation none of them so far have produced satisfactory results.

Super ethoxybenzoic acid (EBA) is reinforced zinc oxide–eugenol cement, which has high compressive and tensile strengths, neutral pH, high moisture resistance,[8] and low cytotoxic effect, releasing only 2% eugenol while curing.[9] Pitt Ford et al[10] used Super EBA as root end filling material on healing after replantation in dog's teeth and found it superior to amalgam and intermediate restorative materials. Moloney et al[11] and Gencoglu and Mentes[12] both found EBA cement superior to amalgam in lateral root perforations in microleakage studies. Oynick and Oynick[13] advocated the clinical use of Super EBA for furcation perforation and Bogaerts[14] also reported good clinical results from root perforation repairs using Super EBA with a calcium hydroxide barrier.

Mineral trioxide aggregate (MTA) was developed for creating apical plugs to prevent the extrusion of filling materials, as a root end filling material and for repairing furcation perforations.[15,16] At present MTA has been

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Fig 1. Representative radiogram showing repaired furcation perforation.

widely used in endodontic treatment. It has shown good results when used as a repair material.

In perforation treatment, Nakata et al[17] found the sealing ability of MTA to be superior to amalgam, and Lee et al[18] found MTA superior to both amalgam and Super EBA. In addition to these in vitro studies, Torabinejad et al[19] found MTA material superior to amalgam as a root end filling material in monkeys. They also reported new cementum formation over the MTA material. Pitt Ford et al[16] investigated intentionally perforated dog's teeth in the furcation area and found MTA to facilitate superior healing to amalgam, also reporting new cementum formation around the MTA material. Although 1 study[16] regarding MTA material has investigated histologically for repair of furcation perforation, no such study has been performed for Super EBA material.

The purpose of this study was to compare histologic tissue responses to Super EBA and MTA used to repair experimentally induced furcal perforations in dogs' teeth.

MATERIALS AND METHODS

A total of 90 teeth of 9 adult mongrel dogs in the mandibular premolars and molars were used. The experimental protocol was approved by the Marmara University animal ethics committee. These teeth were divided into 3 groups: Super EBA group (n = 36), MTA group (n = 36), and control group (n = 18). The control group received no treatment, to evaluate the effects of external variables, such as periodontal disease, that could have developed during the experiment.[20] Access into the pulp chamber was obtained through the occlusal surface, using a #4 fissure bur under general anaesthesia using 20 mg/kg ketamine hydrochloride (Ketalar, Eczacibasi, Turkey). The root canals of mandibular molars and premolars were cleaned, shaped and obturated with laterally condensed gutta-percha and

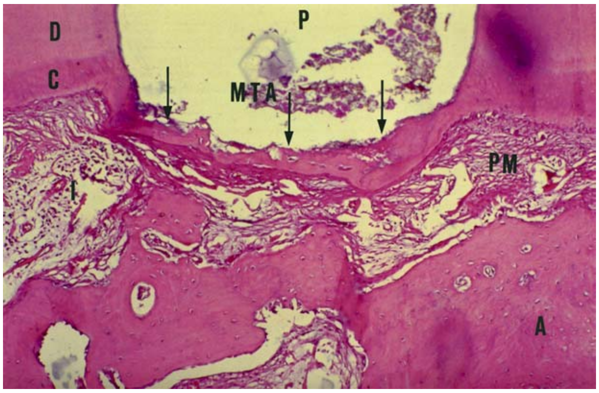

Fig 2. Perforation repair with MTA. Observation period is 1 month. The specimen shows mild inflammatory reaction (*I*) in the periodontal tissue. New cementum covers the perforation site (*arrows*). *D* = dentin; *P* = perforation area; *C* = cementum; *PM* = periodontal membrane; *A* = alveolar bone. HE, 100×.

sealed with Grossman's sealer. One week later, a perforation was made through the floor of the pulp chamber into the furcation area using a round bur (ISO size 014) at low speed until bleeding was observed. The bleeding was controlled by paper point.

In each dog 1 side of the mandible was treated with MTA and the other with Super EBA according to the manufacturer's recommendation. Radiographs were taken of each tooth (Fig 1) and the access cavities were filled with amalgam. The animals were killed by use of an overdose of sodium pentobarbital at 1, 3, and 6 months. The jaws were perfused with 10% buffered formalin and after the removal of the mandibles they were postfixed for 48 hours. After fixation the specimens were demineralize for 10 weeks in formic acid with sodium citrate. Each tooth with surrounding bone was separated. The specimens were dehydrated through graded alcohols and embedded in paraffin. Serial sections were cut through the perforation sites with the microtome set at 5 μm. Slides were stained with hematoxylin-eosin (HE) and Masson's trichrome.

The sections were examined by 2 examiners. Inflammation and type of healing were scored as follows[21]:

Inflammation:

0 = Absent—no infiltration of inflammatory cells

1 = Mild—inflammation including a few inflammatory cells, neutrophilic leukocytes, lymphocytes, and macrophages

2 = Moderate—accumulation of macrophages, lymphocytes, and plasma cells

3 = Severe—massive infiltration of macrophages, lymphocytes, and plasma cells, sometimes with abscess formation throughout the perforation

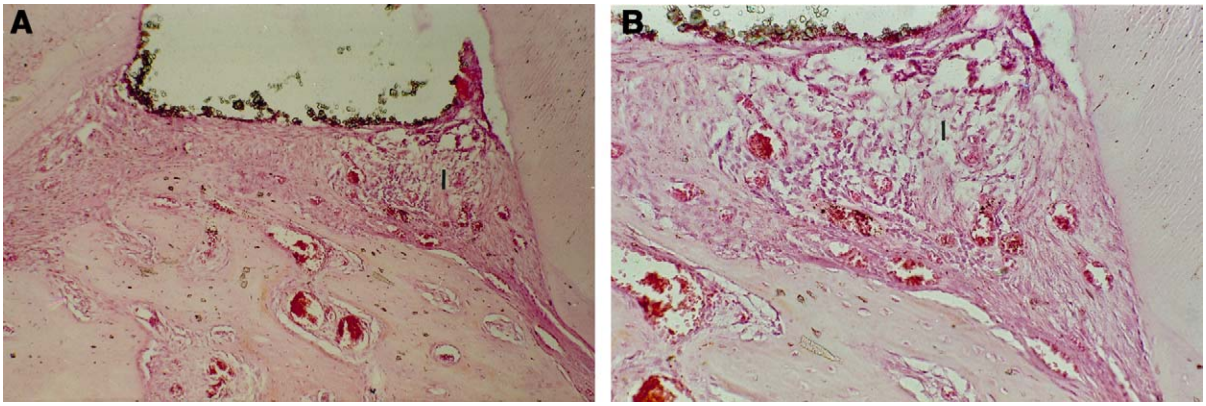

Fig 3. **A,** Perforation repair with Super EBA. Observation period is 6 months. Moderate inflammatory reaction (*I*) and healing with connective tissue. HE, 100×. **B,** Inflamed area in *A* at higher magnification. HE, 200×.

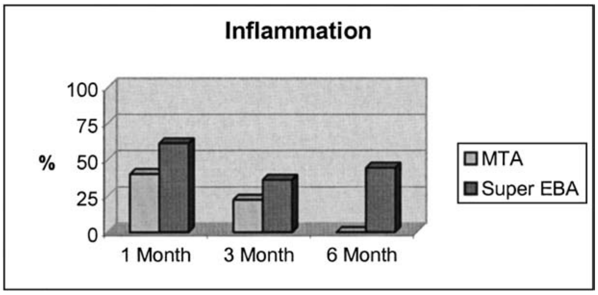

Fig 4. Samples (in %) with inflammatory response after perforation repair with MTA or Super EBA.

Type of healing:
Soft tissue healing
Hard tissue healing

RESULTS

Three specimens in the MTA group were lost during preparation process (2 specimens in the 1 month and 1 specimen in the 6 month group).

The MTA group showed mild inflammatory reaction in 1 month (Fig 2) which decreased at 3 months. No inflammatory reaction was seen in any of the specimens at 6 months.

The Super EBA group showed moderate inflammatory reaction in 1 month. It decreased between 1 and 3 months but remained unchanged between 3 and 6 months. At the and of 6 months only 3 cases showed no inflammation and the others showed inflammatory reaction from mild to severe (Fig 3).

The inflammatory reactions of both materials in furcation area at 1, 3, and 6 months are summarized in Table I and Fig 4.

In the MTA groups, the perforation area was filled by new cementum tissue in 3 specimens at 1 month, 7 specimens at 3 months, and all specimens at 6 months (Fig 2). In overfilling cases, new cementum tissue formation was detected over the material in 5 of 6 specimens. Also, in 1 specimen, although deep perforation was performed, again hard tissue healing had taken in place (Fig 5).

In Super EBA specimens where inflammatory reaction was seen, the perforation area usually was filled with irregular connective tissue (Fig 3). However, specimens with no inflammation showed dense connective tissue formation (Fig 6).

The control group showed normal periodontal tissue with no inflammation.

DISCUSSION

The use of 2 materials for the immediate treatment of mechanical perforations in the furcation area was evaluated in this study. MTA and Super EBA have been recommended for use as repair materials in furcation perforations,[13-16] but no comparative or controlled studies have been reported in the literature. For this reason, we intended to determine whether the use of those materials was suitable for the repair of perforated defects and whether their use led to increased rate of success over existing materials.

Dogs were used for this study because their teeth have well developed roots and the root furcations provide good accessibility and visibility. Their teeth are large enough to facilitate the study of tissue reaction and to allow ample room for perforation.[22] However, the morphologic character of the dog's tooth is different from that of the human teeth. The dog is a demanding experimental model, having 2 rooted lower premolars where furcation is often as close as 1 to 2 mm from the cementoenamel junction. The furcation lies more deeply within the alveolus in humans.[21] Thus, any technique shown to produce favorable results in dogs may have a more favorable response in humans.[21]

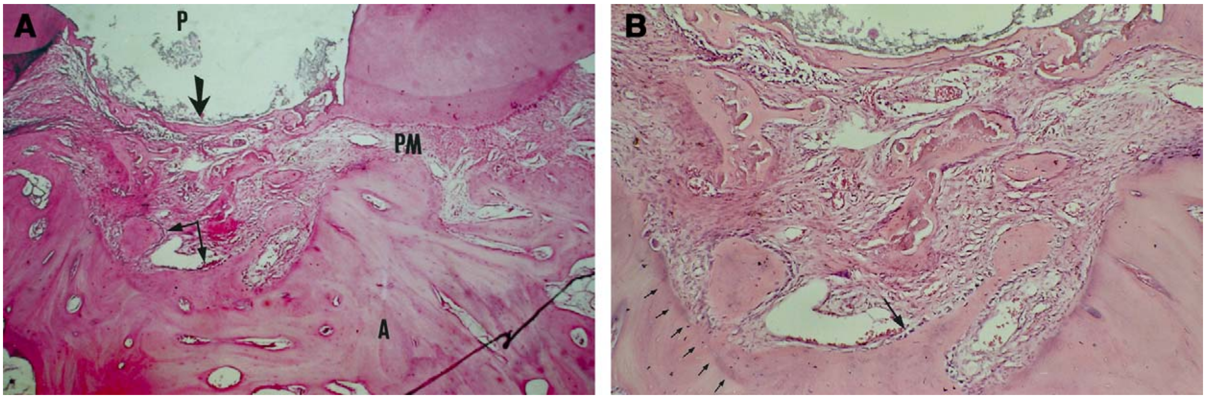

Fig 5. **A,** Perforation repair with MTA. Observation period is 1 month. The perforation was extended into furcation bone. New cementum covers the MTA in the perforation area (*big arrow*). The bone cavity is filled with newly formed bony tissue. *P* = perforation area; *A* = alveolar bone; *PM* = periodontal membrane. HE, 40×. **B,** Higher magnification of bone cavity in *A*. Intense new bone growth can be seen (*big arrow*). The border of the prepared bone lesion can still be (*small arrows*) HE, 100×.

Table I. The inflammatory scores of perforation repairs with MTA or Super EBA

	1 Month				*3 Months*				*6 Months*			
	None	*Mild*	*Moderate*	*Severe*	*None*	*Mild*	*Moderate*	*Severe*	*None*	*Mild*	*Moderate*	*Severe*
MTA	1	7	1	1	8	2	0	2	11	0	0	0
S-EBA	0	4	6	2	5	3	2	2	3	4	3	2

In this study all perforations were the same diameter (ISO 014 bur), although the perforation-to−tooth width ratios were higher for premolars than molars. Alhadainy et al[23] and Himel et al[20] reported a higher success rate in molars than premolars. However, Salman et al[21] found no difference in success rate between molars and premolars. Our study supports the findings that there is no particular relationship between tooth width and success rate.

Furthermore, it is reported that large perforations make it difficult to completely seal the defect with the material, thus allowing irritant to continuously penetrate into the furcation area.[24] When perforations have been allowed to remain open to oral environment, the prognosis is much poorer.[25,26] Perforations close to the gingival sulcus produce persistent inflammation and/or a downgrowth of sulcular epithelium into the defect.[1,4,6] In the present study, no epithelial migration was detected in any specimen. The probable explanation for the high rate of repair in the present study is that both MTA and Super EBA materials were biocompatible.

Because most investigators recommend immediate sealing of endodontic perforations, all perforations were repaired at the same time they were created. Pitt Ford et al[16] repaired furcation perforations immediately with either amalgam or MTA or exposed to oral contamination for 6 weeks before repairing them. They found that 5 out of 6 teeth immediately treated with MTA showed no inflammation whereas 4 out of 7 teeth repaired after 6 weeks of exposure were inflamed. They also reported that teeth repaired with amalgam were always associated with inflammation and abscess formation and also epithelial downgrowth.

The sealing ability of the repair material and its extrusion into furcation areas are considered major problems when repairing furcation perforation repairs,[27] and several in vitro studies investigated the sealing ability of different materials. Most of the materials used for furcation perforation repair exhibit slight to severe leakage. However, investigators have reported less leakage with MTA than with amalgam[17,18,28] and Super EBA.[18,28,29]

Although several microleakage studies on MTA and Super EBA material have been done, few histologic studies are published.[1,30]

Traditional zinc-oxide−eugenol cement has been used in furcation perforation repair in dog's teeth.[1,30] The results reported include severe inflammatory reaction with abscess formation in furcation area. In the present study, however, the Super EBA material showed moderate inflammation at 1 month, decreased between 1 and 3 months, but remained unchanged between 3 and 6 months. In Super EBA specimens where no inflammatory reaction was seen, the perforation area healed with connective tissue. Resorption activities were replaced with apposition. In contrast to

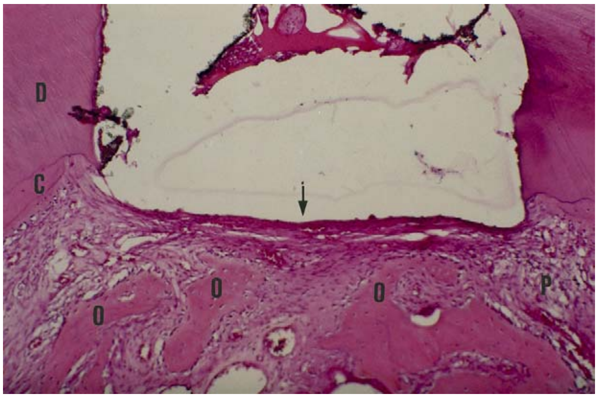

Fig 6. Perforation repair with Super EBA. Observation period is 6 months. Super EBA is demarcated with fibrous soft connective tissue (*arrow*). New bone formation in perforation area (*O*). *D* = dentin; *C* = cement; *P* = periodontal membrane. HE, 100×.

the findings with zinc oxide–eugenol cements, Super EBA showed good histologic results. These different findings may be due to the fact that traditional zinc oxide–eugenol cements release 50% eugenol while curing whereas Super EBA releases only 2%.[9] Thus, material toxicity may be an important factor for tissue healing.

In conclusion, the findings support the idea that MTA is superior to Super EBA for the repair of furcation perforations.

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