

Immediate reimplantation of primary teeth: a histological study in dogs

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Abstract – Reimplantation of primary teeth has been the subject of various clinical and histological studies, but very little is known about the effect of this treatment on primary teeth. The aim of the present study was to histologically evaluate the biological response of dog primary teeth after immediate reimplantation. Twelve dogs were divided into two groups according to postoperative time: 1 week and 3 weeks. Twenty-one upper and lower intermediate incisors were extracted and submitted to endodontic treatment, reimplantation and splinting. The 21 homologous teeth not submitted to treatment served as controls. The animals were killed after the respective experimental periods, and the obtained specimens were processed for histological analysis. Most dogs of the 1-week group exhibited the following alterations: gingival epithelium was inserted in the cemento-enamel junction, with a small number of chronic inflammatory cells being observed in the gingival corium; the periodontal ligament was partially reestablished, with a more expressive chronic inflammatory infiltrate being observed in the apical third; small root resorption was observed on the palatine/lingual side in the apical third. In contrast, in 3-week animals, gingival epithelial insertion was predominantly absent on the buccal side, and a moderate chronic inflammatory infiltrate was present; the periodontal ligament generally showed no bone-cementum reinsertion especially on the palatine/lingual side, with an expressive inflammatory infiltrate in the apical region, and large root resorption was mainly observed on the palatine and lingual sides. In conclusion, reimplantation causes histological alterations in the tooth and its supporting periodontal structures that suggest the impossibility of its maintenance.

Traumatic lesions are unexpected and inconvenient at any age or time of life. Children are always eager to discover the world around them, which is inherent to their psychological and motor development, challenging their powers often without noticing the dangers they are exposed to.

In the primary dentition, traumatic injuries accompanied by dental luxations are common (1, 2) due to the greater resilience of the alveolar bone which probably confers a greater elasticity, favoring displacements rather than fractures. Among dental luxations, avulsion or exarticulation leads to the complete displacement of the tooth from its alveolar socket, (3) with the upper central incisors being most affected (4, 5).

Reimplantation has been indicated as treatment for avulsed permanent teeth (2, 6–11) in an attempt to reestablish their integrity and function in the dental arch (7, 8). In this respect, many studies have evaluated an ample number of variables that might interfere with the tissue repair process, which is unquestionably related to the success of treatment, i.e., the maintenance of a functional tooth in the arch (2, 12–14). In contrast, controversies exist regarding the indication or not of reimplantation of primary teeth. These controversies are based on the existence of risk factors that might damage the permanent tooth germ, and therefore reimplantation

has been counterindicated by most authors (2, 4, 10, 15–17). Among the factors that can damage the permanent successor are the processes of repositioning of the tooth in its alveolar socket itself in which the blood clot is pressed against the dental follicle of the involved permanent tooth, and/or possible infection (4, 16, 18). In addition, a possible lack of collaboration on the part of the child during care might interfere with the quality of treatment (19).

Thus, experimental studies on primary tooth reimplantation are important because most investigations in the literature deal with the developmental alterations that occur in permanent teeth as a consequence of trauma to the primary predecessor and not with reimplantation itself. Very little is known about the histological alterations that can occur in the primary tooth itself after reimplantation. Therefore, the aim of the present study was to histologically evaluate the biological response of primary teeth and their periodontal structures in dogs after immediate reimplantation.

Materials and methods

Twelve mongrel dogs with an initial age of 52 ± 3 days were studied. The animals were divided into two groups according to postoperative time: group A (1-week

experimental period) and group B (3-week experimental period).

The four intermediate incisors were used for the experiment, for a total of 42 teeth. The experimental teeth consisted of two intermediate incisors from each animal, the upper and lower one on the same side, which were extracted, treated endodontically, and reimplanted. Their respective intermediate homologs, which were not submitted to treatment, served as controls.

All surgical procedures were performed under general anesthesia consisting of the intravenous administration of 3% sodium pentobarbital at the dose of 1 ml kg⁻¹ body weight. The dogs were maintained on saline solution to facilitate anesthetic complementation when necessary.

Radiography was performed before surgery, after reimplantation and fixation of the teeth, and at the end of the 1- and 3-week experimental periods. Periapical radiographs of the incisors were taken using a pediatric positioning device for the radiographic films. After syndesmotomy, the incisors were luxated and extracted using a forceps indicated for the extraction of human upper primary incisors. Each extracted tooth was wrapped in a soft tissue and immersed and constantly moistened with physiological saline. Each tooth was carefully handled between the index finger and thumb only at its most coronary portion during endodontic treatment.

The pulp chamber was accessed through the buccal side using No. ¼ and/or No. ½ carbide spherical burs moved at high rotation and cooled with saline and air. After an opening was made in the crown, the root pulp was removed with barbed broaches, and the root canal was instrumented using Hedström files under successive rinsing with physiological saline and subsequent aspiration. The root canal was then dried with absorbent paper cones and filled with Sealapex cement. After rinsing the alveolar sockets with saline and reimplantation of the teeth, the Sealapex (Sybron Kerr, Romulus, MI, USA) was overlaid with a thin layer of Dycal (Dentsply, Rio de Janeiro, Brazil).

The reimplanted teeth were stabilized for 1 week with 0.25-mm brass wire fixed side-by-side to all incisors using compound resin. The animals received a pasty diet during the postoperative period, and cephalixin (30 mg kg⁻¹ body weight) was administered orally at 12-h intervals for 3 days.

After each experimental period, 1 week (group A) and 3 weeks (group B), approximately 6-µm thick specimens were processed for histological analysis and stained with Masson's trichrome and hematoxylin-eosin.

Results

The following parameters were evaluated upon histological analysis: insertion and inflammation of gingival epithelium, insertion and inflammation of periodontal fibers, root and bone resorption, and presence of ankylosis and replacement resorption.

Group A

The gingival epithelium was adhered to the cemento-enamel junction, showing few chronic inflammatory cells

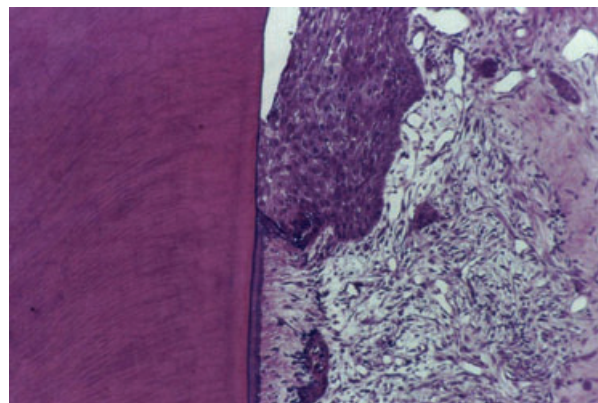


Fig. 1. Group A – reinsertion of gingival epithelium. Hematoxylin-eosin. 250×.

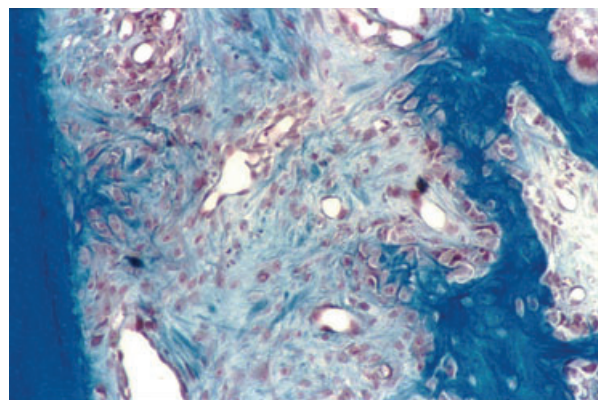


Fig. 2. Group A – disorganized periodontal fibers. Trichrome Masson 250×.

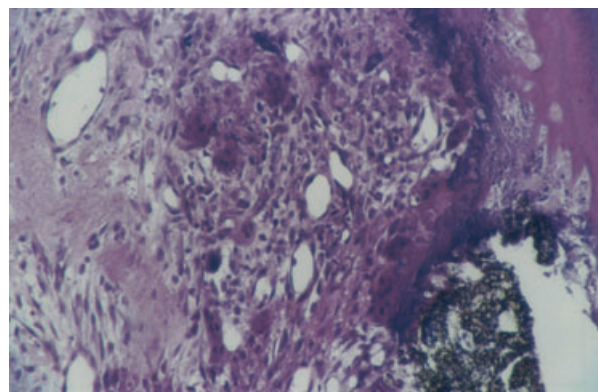


Fig. 3. Chronic inflammatory infiltrate in the apical region. Hematoxylin-eosin 250×.

(Fig. 1). The periodontal ligament was partially recovered, presenting poorly dense fibers without a defined spatial orientation which were rich in fibroblasts (Fig. 2). Periodontal inflammation was a frequent finding only in the apical region of most specimens and was characterized by a more intense chronic inflammatory infiltrate, with a predominance of lymphocytes, macrophages containing intracytoplasmic Ca(OH)₂ particles, and multinucleated cells (Fig. 3).

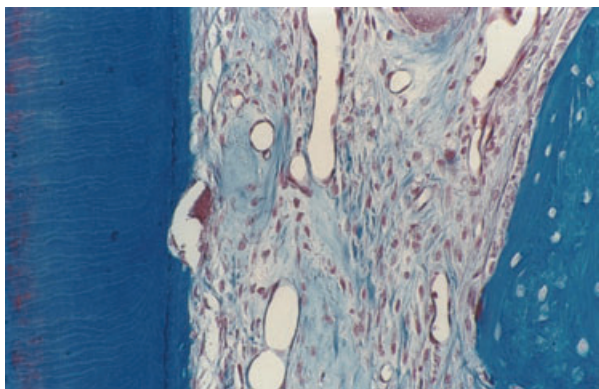


Fig. 4. Group A – small area showing active primary root resorption. Trichrome Masson 125 \times .

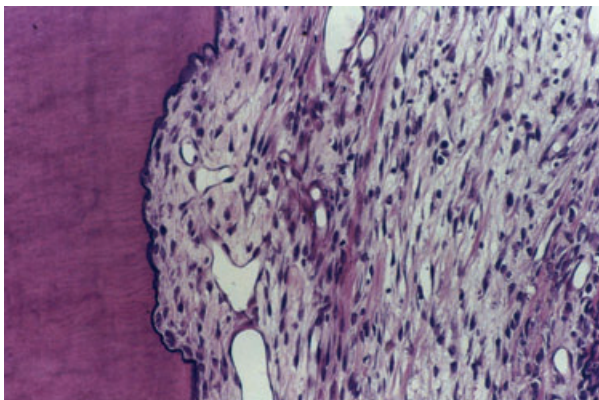


Fig. 5. Group A – area of root resorption showing a thin layer of cementum and the absence of multinucleated cells. Hematoxylin-eosin 250 \times .

Areas of root resorption, when present, were small and characterized by multinucleated cells (Fig. 4). Areas of active resorption predominantly located on the buccal side of the middle and apical thirds were observed in one of the specimens. However, in most specimens, areas of active resorption predominated in the apical third and were accompanied by the presence of a chronic inflammatory infiltrate mainly consisting of lymphocytes and macrophages (Fig. 5). Signs of healing in the areas of resorption included the deposition of a thin layer of basophilic cementum and the absence of multinucleated cells. Bone resorption was observed in most specimens, but was more evident on the palatine and lingual surfaces.

Group B

In most specimens, no reinsertion of the gingival epithelium was observed, especially in the buccal region, and a moderate to severe chronic inflammatory infiltrate was noted in the gingival corium (Fig. 6). In general, no bone-cementum reinsertion of the periodontal ligament was observed in the palatine or lingual regions. In addition, an intense chronic inflammatory infiltrate was present in the apical region.

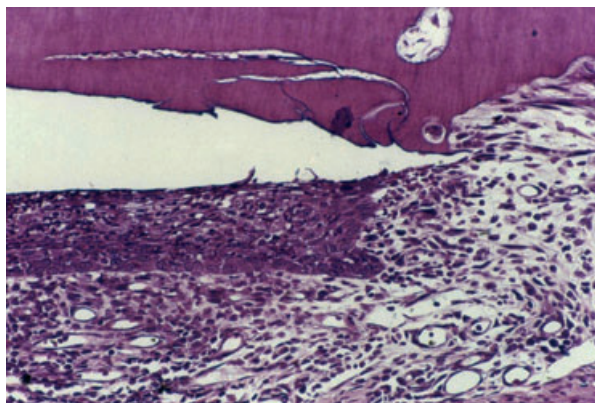


Fig. 6. Group B – note the absence of gingival epithelial reinsertion and the presence of a severe chronic inflammatory infiltrate. Hematoxylin-eosin 250 \times .

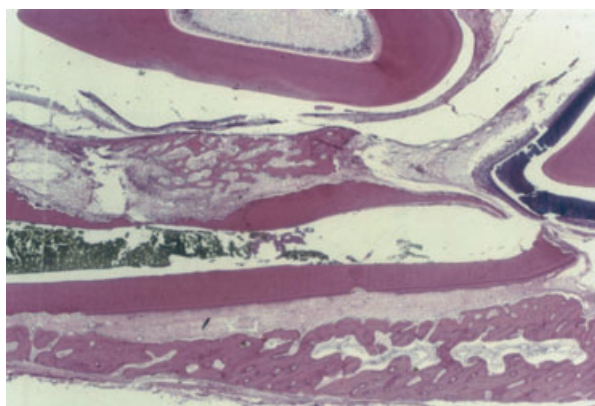


Fig. 7. Areas of severe root resorption in the middle and apical thirds of the palatine and lingual regions. Hematoxylin-eosin 16 \times .

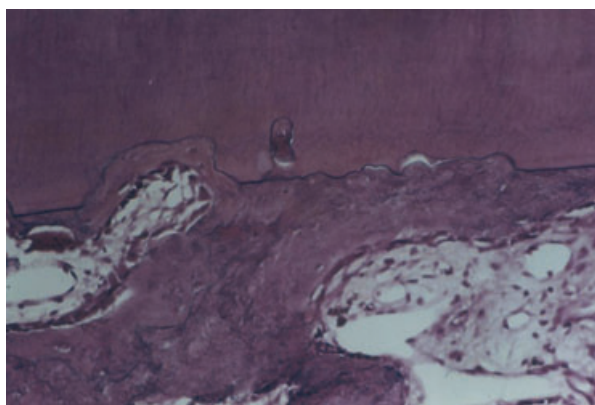


Fig. 8. Areas showing replacement resorption and ankylosis. Hematoxylin-eosin 125 \times .

Large areas of root resorption mainly affecting the palatine and lingual sides were observed in all specimens analyzed (Fig. 7). Two dogs presented areas of replacement resorption and ankylosis (Fig. 8).

Discussion

In the literature, most authors advise against the reimplantation of primary teeth (2, 4, 15, 17, 20, 21) because of the difficulty in handling very young children and the risk of damaging the germ of the permanent successor. This risk is related to the possibility of local infection and the process of reimplantation itself in which the blood clot that forms in the alveolar socket might be pressed against the dental follicle and the permanent tooth germ. These events can lead to damage ranging from stains and structural defects to altered eruption and even loss of the permanent successor (2, 4, 16–18).

In the present experiment, the risk of damage to the permanent tooth due to the pressure exerted by the blood clot, as reported by Andreasen & Andreasen (4), was avoided by rinsing the alveolar socket with physiological saline to remove the clot formed before reimplantation of the extracted primary tooth. We observed a new bleeding soon after this procedure and, at that time, the tooth was gently repositioned in its original socket, similar to the experiment of Sheppard & Burich (22). The position and height of the reimplanted teeth were clinically verified by comparison with the neighboring teeth and confirmed by radiography after fixation.

Infection has also been associated with structural defects in the enamel of the permanent successor due to the fact that any infection generates an acid pH which may interfere – depending on the developmental stage of the permanent tooth germ – with both deposition of organic matrix and calcification of crown enamel (18). In the present study, to prevent infection all animals received cephalixin antibiotic therapy as indicated by the veterinarian. We were unable to detect any bacteria because no specific staining method was used, for example the Brown and Brenn stain, which would have been of great help in the interpretation of the results. However, it should be emphasized that none of the dogs showed signs of local infection upon clinical examination.

In general, inflammation, particularly in the middle third of the roots, was not a noteworthy finding in the present study. However, inflammation was observed in some situations, for example, in the apical region of cases presenting leakage of the Sealapex cement from the root canal. In this region, inflammation was clearly visible and was characterized by the expressive presence of macrophages containing material that resembled the cement particles and by a predominance of lymphocytes. Andreasen & Kristerson (23) also observed active resorption of calcium hydroxide by multinucleated cells when calcium hydroxide paste leaked into the periodontal region.

The histological observation of this expressive chronic inflammation leads us to reflect on the definition of a successful outcome of primary tooth reimplantation, as clinical examination of the dogs throughout the experimental period did not identify any sign of inflammation that would have suggested this histological finding. In view of this consideration, one may question the cases of successful outcomes reported in the literature (24–27).

Clinical success – i.e., to maintain the tooth in the mouth for a longer period of time without symptoms (4, 12) – may not indicate the absence of a local chronic inflammatory reaction. The latter can be considered a risk factor for damage to the germ of the permanent successor due to the close anatomical relationship between these two teeth as demonstrated in humans (10).

One aspect that should also be discussed is why we chose endodontic treatment for the immediate reimplantation of young teeth, because Andreasen et al. (28) showed that pulp revascularization can be achieved more easily. These authors observed pulp revascularization in 34% of young permanent teeth with open apices which had been reimplanted immediately. This percentage seems to show an uncertain prognosis to risk the most expected consequence – necrosis – and thus the occurrence of inflammatory resorption (29–33) which would markedly complicate the whole experiment as this event shows a rapid progression and the tooth of interest may therefore be lost. On the basis of the studies (28, 31) which related pulp necrosis to a higher percentage of root resorption in reimplanted permanent teeth, we decided to treat the canals immediately after extraction (before reimplantation), as proposed by Heithersay (34) and Einsenberg (1995) and not before extraction because the structure of the small tooth would become even more fragile and more prone to fracture during extraction. Root canal treatment was also not performed after reimplantation because the splint would limit the access to the canal and thus increase the surgical difficulties. Andreasen & Andreasen (4) recommended endodontic treatment with calcium hydroxide within an interval of 1 week after immediate reimplantation of permanent teeth. However, variations in this time interval have been reported in the literature, ranging from immediately after reimplantation (35) to a period of more than 20 days (36). In the present experiment, endodontic treatment was performed in a single session to prevent the animal from being exposed to more than one anesthesia. Another reason was that the time interval between immediately after extraction and reimplantation ranged from 10 to 12 min, thus not exceeding the 15 min recommended by Donaldson & Kinirons (37) or the 30 min as proposed by Kawashima & Pineda (26) and Filippi et al., (27) characterizing the present reimplant as immediate. In clinical practice, attending a child of young age in a single session would also be a more practical and less stressful option for all persons involved, the professional, parents and mainly the child.

Analysis of the control specimens from groups A and B (1 and 3 weeks) revealed a normal pattern along the periodontium in all of them. In addition, the permanent tooth germ was always surrounded by tissue with a 'lace-like' aspect, with basophilic cells being found throughout its extension from the crown to the gingival corium. This tissue, called gubernaculum dentis, is a loose tissue located in the gubernacular canal, the latter being related to the predetermined eruption path of the permanent tooth, as reported by Cahill et al. (38). According to these authors, the gubernacular tissue corresponds to the dental follicle.

No physiological root resorption was observed in 1- and 3-week control specimens, a finding somehow indicating that this is a period of resorption-free tooth development in the dog. It was clear that the age of the animals used here, which was established based on the studies of Arnall (39) and Shabestari et al. (40) on the development and chronology of tooth eruption in dogs, was opportune for the present investigation. Probably, at a more advanced age when resorption is already present, it would be more complicated to establish a difference between physiological resorption and resorption caused by the experiment due to the lack of a reference. The fact that very young animals were used permitted the demonstration that the root resorption observed was indeed triggered by the treatment as no resorption was observed in control teeth.

Histological analysis of the experimental teeth of group A (1 week) demonstrated probable reorganization due to the presence of numerous fibroblasts throughout the periodontium, as well as osteoblasts and osteoid tissue indicating bone neoformation. Similar findings have been observed by other investigators, such as Andreasen & Andreasen (41), within a period of 3 to 4 days after reimplantation of permanent teeth. This similarity might indicate that during this period of up to 1 week, the biological responses are the same for primary and permanent teeth.

Root resorption has been reported to be the most common and discouraging complication of permanent tooth reimplantation (6, 7, 30–33). The same was observed in the present study; however, a finding that called attention was the fact that the resorptions were frequently located in the palatine and lingual regions of the primary roots.

The severity of root resorption was clearly demonstrated for the 3-week experimental group when compared to the 1-week group, and when comparing both groups to their respective controls. Resorption was always much more pronounced on the palatine and lingual sides and was closely accompanied throughout its extension by the permanent tooth germ. This observation suggests that reimplantation may have somehow accelerated both primary root resorption and eruption of the permanent successor. The permanent successors were located closer to the gingival corium and there seemed to be a direct relationship between the proportion of resorbed root and the position of the permanent tooth germ.

In the present experiment, two specimens of the 3-week experimental group presented replacement resorption and areas of ankylosis located in the apical third. In fact, a higher frequency of this finding would be expected as primary tooth ankylosis is one of the counterindications of reimplantation, and because of the occurrence of leakage of the Sealapex cement into the periodontal region. Analysis of the clinical history of these animals revealed the occurrence of secondary traumas to the reimplanted teeth. In one case, intrusion due to total fixation loss had occurred in the first postoperative week and no procedure was performed. Re-eruption of the intruded tooth was observed during the third week control exam. In the second specimen,

secondary trauma was probably related to the difficulty in performing syndesmotomy of the highly resistant periodontal fibers, which might have caused further damage to both the ligaments and root surface involved. The case of intrusion demonstrates the importance of fixation after reimplantation, as reported by Weisman, among others, thus preventing the risk of displacement, aspiration, (42, 43) or deglutition of the tooth. According to Andreasen & Andreasen (41), healing after large periodontal damage may occur by cellular competition, resulting in ankylosis (3, 6, 7). Thus, we believe that the finding of ankylosis and replacement resorption in these specimens was not only the result of the proposed treatment but also of the sum of injuries to the periodontal region. Although we know that the use of a splint in the control teeth was not ideal, this procedure was necessary to obtain a better force distribution and to increase the resistance of the fixation. The dog primary teeth are very small and do not offer sufficient surface for good adhesion. The splinting time recommended by Andreasen & Andreasen (4) for immediate reimplantation of permanent teeth is 1 week. The same time interval was suggested by Nasjleti et al. (44) who reported that longer periods (30 days) seem to increase the frequency of both ankylosis and root resorption. However, no consensus exists regarding the splinting time for primary teeth, with fixation periods of 7, (45) 10, (18) 15 (25) and 17 days (46) having been reported.

Although all possible care was provided to the animals, the clinical conditions of the fixations had changed after 7 days when compared to those immediately after surgery. Thus, an interval of 1 week was indeed the maximum period that could be chosen for fixation in this experiment. These findings were probably related to the fact that the animals were housed two to a cage, playing and biting each other, and to the small dental surface available which is insufficient for good adhesion of the splint. In addition, the brass wire may not have been the best choice because it is very flexible and easily distorted. In a future study, it would be interesting to have one animal per cage, to fabricate additional mechanical retention in the supporting teeth, and to use another type of wire, for example, an orthodontic wire.

In dental trauma research, avulsion is an event that is difficult to reproduce experimentally. The intentional extraction of animal teeth in laboratory experiments with a forceps does not truly reproduce the direction or intensity of accidental trauma to the tooth and its supporting structures. In addition, extraction is not able to reproduce possible movements that a tooth might undergo during its accidental avulsion. These details may result in differences between experimental tissue repair and the *in vivo* recovery of an accidentally avulsed tooth.

In view of the fact that some variables that we could not control for might have interfered with the present results, further studies are necessary to develop a more appropriate method for the reimplantation of primary teeth and to analyze the pathologies involved. Therefore, the present results do not permit us to indicate primary tooth reimplantation as a routine treatment, although no

clinical aggravating complications were observed in the arch throughout the study period.

Based on the analysis of the results obtained with the method employed, we conclude that reimplantation causes histological alterations both in the tooth and in its periodontal structures that suggest the impossibility of its maintenance.

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