Pulpal status after autogenous transplantation of fully developed maxillary canines

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By means of histologic and histochemical studies of oxidative enzymes, the pulps of transplanted fully developed maxillary canines have been evaluated after an observation period of 3 to 25 months. No pulp had survived the transplantation trauma, and endodontic treatment of fully developed transplanted teeth was considered to be a prerequisite to an optimal clinical result.

Autogenous tooth transplantation is the transplantation of teeth from one site to another within the mouth of the same individual. The use of this technique is currently increasing. For the most part, third molars with incompletely developed roots are placed into the extraction sites of cariously involved first molars.[7] It is also common to transplant other teeth—for instance, an impacted canine into a surgically preformed alveolus.[14, 16] Histologic studies of the pulpal status of transplanted teeth have mostly been carried out in animals, but there are some reports of such studies in man.[8]

Öhman[10] has reported that, within 35 days after replantation of teeth, no pulp reaction to electric stimuli can be demonstrated. However, when the observation period was extended, an increasing number of teeth were found to give a positive reaction to the electric stimuli. It has been assumed that a new supply of blood is established long before the regeneration of the nerves has reached an extent sufficient to give a response to electric tests, and that such teeth might thus be vital but nonsensitive.[2]

Severe pulpal damage after replantation has been found to occur more frequently in teeth with fully developed roots than in teeth with incomplete root formation.[10] Concerning the capacity of the pulp of fully developed teeth to

Table I. Clinical data on transplanted maxillary canines

Patient	*Sex*	*Age (yr.)*	*Maxillary canine*	*Fixation period (wk.)*	*Interval (mo.) between transplantation and endodontic treatment*	
1	F	48	Right	6	25	
2a	F	42	Left	6	13	
2b	F	42	Right	6	5	
3	F	21	Right	5	16	
4	M	41	Left	6	6	
5a	F	49	Left	4	5	
5b	F	49	Right	4	5	No tissue found in the root canal
6a	M	38	Right	5	3	
6b	M	38	Left	5	3	
7a	F	31	Right	6	3	
7b	F	31	Left	6	3	
8	F	27	Right	6	4	
9	F	32	Left	5	4	

recover from transplantation trauma, different opinions are presented in the literature.[11]

The present investigation was undertaken to study the pulps of fully developed teeth subsequent to autogenous tooth transplantation after a long period of observation and to examine the vitality of the pulp tissue by means of an enzyme histochemical technique.

MATERIAL AND METHODS

The material comprised thirteen autotransplanted teeth from nine adult patients, 21 to 49 years of age. The autografts consisted of eleven impacted and two partly erupted maxillary canines transplanted into correct position in the dental arch (Table I).

During a period from 1 day before to 6 days after the operation, the patients were given 4.8 million I.U. of penicillin V orally each day.

The surgical procedures were performed with the use of local anesthesia. The new alveolus was prepared by means of round burs under constant irrigation with saline solution. After the tooth was extracted, the preparation of the alveolus was completed to fit the shape of the root. In the meantime (a few minutes), the tooth was kept in saline solution.

The transplanted teeth were immobilized by means of an acrylic splint, and the time of fixation was 4 to 6 weeks. During the first 10 days of the fixation period the patients rinsed their mouths with a 0.2 per cent chlorhexidine solution twice a day.

Three to 25 months postoperatively, endodontic treatment of the transplanted teeth was carried out under aseptic conditions. In no instance was there need for local anesthesia. With the use of Hedström files the pulp tissue was removed and then divided into two pieces. The coronal part of the pulp was fixed in formalin and examined by means of routine histologic techniques, with sections from an intact tooth used for comparison.

The apical part of the pulp tissue was embedded in an aqueous solution of

carboxymethylcellulose on a microtome stage and frozen in hexane cooled with solid CO_2 (−75° C.). According to the method devised by Ullberg,[15] frozen sections, 10 to 15 μm thick, were taken and Scotch tape No. 850* was used to stabilize the tissue during sectioning.

After freeze-drying, the sections were brought to room temperature in an air-tight box, and incubated for the histochemical demonstration of the following enzymes:

1. Succinate dehydrogenase (E.C. 1.3.99.1) as a marker of the aerobic glycolysis or the citric acid (Krebs) cycle.
2. Lactate dehydrogenase (E.C. 1.1.1.27) as a marker of the anaerobic glycolysis.
3. Glucose-6-phosphate dehydrogenase (E.C. 1.1.1.49) as a marker of the pentose phosphate pathway.
4. Cytochrome oxidase (E.C. 1.9.3.2) as a marker of electron transport in the cytochrome chain.

The incubations for the three dehydrogenases were performed according to the methods described by Barka and Anderson,[1] and the incubations for cytochrome oxidase were carried out according to the method of Seligman and associates.[14] Sections from rat and monkey teeth were incubated as positive controls, together with the sections from the human pulps. After being washed in distilled water, the sections were mounted in glycerin jelly.

Clinical data on the transplanted teeth are summarized in Table I.

RESULTS

Pulp tissue was recovered from twelve of the thirteen transplanted maxillary canines by means of endodontic instruments, whereas one pulp chamber was found to be empty.

Histochemical observations

No enzyme activity could be demonstrated in the apical part of the extirpated pulps. In sections incubated for the four oxidative enzymes, staining for enzyme activity was found in all control sections.

Histologic observations

The predominant histologic finding in the coronal part of the pulp of the transplanted teeth was the lack of normal connective tissue architecture (Figs. 1 to 3). Odontoblasts could not be identified at the surface of the pulpal tissue, where, however, small flakes of dentin were a frequent finding (Fig. 1). Empty or partly filled vascular spaces consistently lacked an identifiable endothelial lining (Figs. 1 and 2). Cellular outlines were lost also in the extravascular tissue. Pyknotic nuclei were seen in a structureless or slightly fibrillar, weakly stained stroma (Figs. 1 and 2). Only a few inflammatory cells could occasionally be identified in this stroma. Diffuse calcifications were observed in some pulps, and they were readily distinguished from previously described dentin flakes by being structure-

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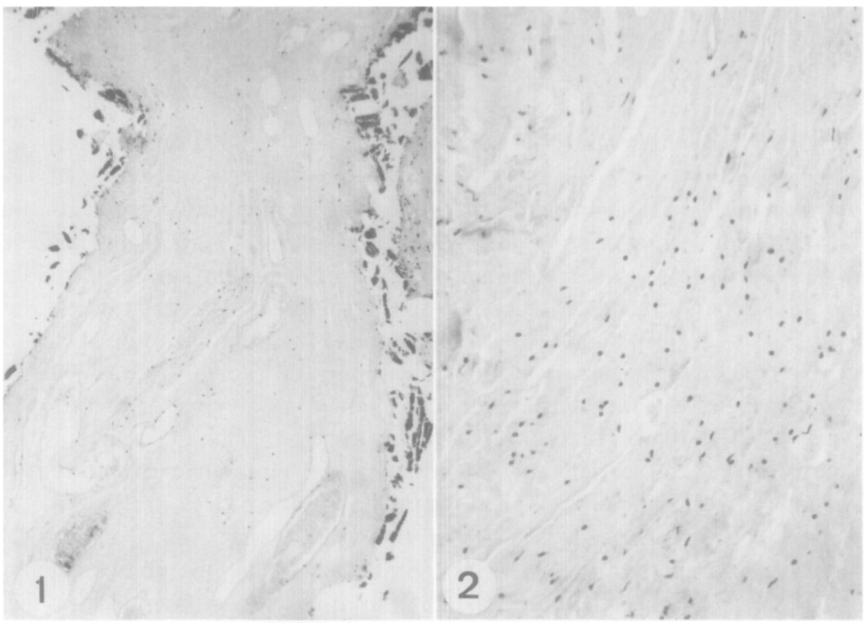

Fig. 1. Section of coronal pulp tissue extirpated 4 months after transplantation. Pyknotic nuclei are present in a structureless, weakly stained stroma. Empty or partly filled vessels lack an identifiable epithelial lining. Flakes of dentin are present at the surface of the tissue. (Hematoxylin and eosin stain. Magnification, ×110.)

Fig. 2. Section of coronal pulp tissue extirpated 3 months after transplantation. No cellular outlines can be seen in the structureless stroma. Pyknotic nuclei are evident. (Hematoxylin and eosin stain. Magnification, ×200.)

Fig. 3. Section of coronal pulp tissue extirpated 13 months after transplantation. Deeply stained diffuse calcifications are distributed throughout the tissue. (Hematoxylin and eosin stain. Magnification, ×110.)

less and diffusely distributed within the pulpal tissue proper rather than at the surface (Fig. 3).

DISCUSSION

The freeze-sectioning and freeze-drying method of Ullberg[15] has previously been used in enzyme histochemistry.[4] It makes possible the sectioning of tissues without previous fixation, which is advantageous in the study, for example, of oxidative enzymes, since these are sensitive to fixatives.[3] Mejàre and associates[7] recently showed that this sectioning method in combination with oxidative-enzyme histochemistry makes it possible to distinguish between vital and devitalized pulp tissue in pulpotomized teeth. Therefore, the same techniques were used in the present investigation, and the apical part of the pulps was chosen for enzyme histochemical examination since the chance for survival of the pulp tissue of a transplanted tooth was expected to be greatest near the apical foramen. The incubations of the sections for the oxidative enzymes demonstrated that there was no metabolic activity in the apical part of the pulps.

The histologic features registered in the pulps extirpated from the transplanted teeth are not compatible with a vital tissue. The lack of identifiable endothelial linings and cellular outlines and the appearace of pyknotic nuclei are all indicative of pulpal necrosis.

Various patterns of necrosis exist, the differences being due to different causes.[12] Loss of blood supply usually induces coagulation necrosis. Lack of oxygen would be expected to seriously affect the mitochondrial function, with subsequent loss of enzyme (including succinate dehydrogenase) activities. Even though anaerobic glycolysis characterized by lactate dehydrogenase activity may provide some energy, this energy cannot meet the total requirements of the cell. The result will be a denaturation and coagulation of the cell proteins.[12] No activity of the two enzymes mentioned above could be registered in any of the pulps. Furthermore, there was the total lack of cellular outlines, with pyknotic nuclei in a structureless, coagulated tissue. The picture thus suggests a coagulation necrosis, presumably due to a lack of blood supply caused by the rupturing of the vessels when the teeth were removed from their original alveoli.

However, the histologic appearance of the necrotic tissue of the extirpated pulps is not completely compatible with a coagulation necrosis, as defined by Robbins.[12] Normally, in this type of necrosis, the cellular outlines can be identified at a stage at which cytoplasmic and nuclear details are obscured. This appearance of the tissue may persist for days or weeks, before further changes lead to removal of the dead cells. The fact that pyknotic nuclei were seen in the pulpal tissue several months after transplantation, whereas a loss of cellular outlines was otherwise evident, would indicate that ischemic infarction of the pulp may be modified by factors that so far are unknown. It is obvious that, being completely deprived of a supply of blood, the pulp can no longer be invaded by inflammatory cells, and will not be subjected to the action of released lysosomal enzymes from such cells. However, a release of lysosomal enzymes from the dying pulpal cells would be expected to accelerate the breakdown of the nucleoproteins as well.

On the basis of the present histologic and enzyme histochemical observations, we may thus conclude that the pulpal tissue will readily respond to an ischemia with a necrosis, which, however, does not follow the normal pattern of a coagulation necrosis. There is a rather low acid hydrolase activity in the pulpal tissue,[1-6] and the persistence of pyknotic nuclei over long periods of time may thus be related to the relative absence of lysosomal enzymes in the dying pulpal cells.

The lack of pyknotic nuclei, associated with the odontoblasts, could then be associated with the high acid hydrolase activity recorded in these cells.[4-7, 17, 18]

The calcific deposits observed in five of the extirpated pulps of the present study show similarities to the diffuse type of pulpal calcifications described by Sayegh and Reed,[13] who suggested that the deposition of these calcified structures was associated with degenerative changes of the tissue. The present findings are in accordance with the concept that pathologic calcification is more apt to take place in degenerate or dying tissues.

In conclusion, the findings of the present study indicate that the pulp of transplanted fully developed teeth would not be expected to recover from or survive the transplantation trauma. Thus, endodontic treatment of such teeth would be considered to be a prerequisite to the achievement of an optimal clinical result, a concept already advanced by Widman[16] in 1915.

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