Regeneration of gingival microvascular architecture on the interface of endosseous titanium implants

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Introduction

It is well known that a local anti-inflammatory mechanism or a defensive immune response exists in peri-implant oral mucosa (gingiva) that is directly in contact with substantia adamantina. This response is due either to infiltration of the exudate or to the migration of polymorphonuclear leukocytes to the gingival sulcus, both of which involve a characteristic capillary network which is distributed beneath the inner marginal epithelium. An implantation method is now commonly used to recover occlusal function by placing an artificial root into the maxilla or mandible, and then covering it with filler. Using this technique, it is extremely important to prevent local infection at the interface between the implant and the gingiva after the implant has been placed.

In this study, we sought to determine whether a capillary network would form beneath the newly formed gingiva surrounding the implant in a canine mandible as well as it would form in natural teeth. After placing an experimental titanium endosseous implant into a canine mandible, the three-dimensional architecture of the peri-implant gingiva was studied using microvascular corrosion casts.

Materials and methods

The right 3rd and 4th mandibular premolars were extracted in a group of adult mongrel dogs under pentobarbital sodium (i.v., 25 mg/kg) anesthesia. A total of eight dogs that had healed well 3 months after the extraction were used for the experiment. An ITI benefit implant was used (3.5 mm in diameter, 11 mm long, 8 mm implantation depth: Strauman Co.)

Implantation Method

Following an incision in a mesio-distal direction at the mucosa of the alveolar crest which was restored after the tooth extraction, the alveolar bone was exposed by exfoliating the mucosa and the periosteal flap. An implant cavity was prepared using a special drill with chilled sterile saline as a coolant. The ITI implant was then placed into position. Finally, the mucosa and the flap were replaced and sutured, and the top margin of the implant was covered with a healing cap.

Microvascular corrosion casts were prepared for the gingival specimens obtained from three dogs each at 2 and 4 weeks post-implantation, using the method described previously by Kishi et al., and were examined by three-dimensional scanning electron
microscopy. In addition, histologic sections of the gingiva with the implant were also prepared, and observed under a light microscope after staining with Hematoxylin-Eosin.

**Results**

Figure 1 shows the histologic structures of the peri-implant gingiva obtained 4 weeks after implantation. The surgical wound margin of the oral mucosa was lined with elongated junctional epithelium at a height of one-half to two-thirds of the gingiva as a result of the downgrowth of mucosal epithelial cells. From the gingival margin to one-half of the upper margin of the inner marginal epithelium, rete pegs were formed by the sidegrowth of epithelial cells after the downgrowth. A layer of connective tissue extended from the lower margin of the epithelium to the apex radit. No evidence of bone absorption was found in the alveolar crest.

Figure 2 shows a microvascular corrosion cast representing inside of the cavity after removing the implant.

Vascularization of the capillary network in the peri-implant soft tissue is seen 2 weeks after implantation. The upper half of the plate reveals a newly-formed capillary network in the peri-implant gingiva and the formation of rete arteriosum and rete venousum, in the deeper layer. Generally, the capillary network seemed to be sparsely distributed. Two types of capillary networks could be identified in the gingiva. One type beneath the inner marginal epithelium was seen in the upper two-thirds of the gingiva, and the other in the layer of connective tissue was seen in the lower one-third.
In the layer of connective tissue, the vessels ran transversely to surround the implant, thus producing a ring-shaped formation, while the vessels ran irregularly beneath the inner marginal epithelium.

The capillary network in the lower margin of the connective tissue layer often anastomosed with vessels which had been newly formed in the granulation tissue between the implant and the alveolar socket wall. Some of these vessels had already entered the newly formed bone (arrow in Fig. 2).

Figures 3 and 4 show the microvascular corrosion cast obtained 4 weeks after implantation as observed from the same angle as in Figure 2. Judging from the distribution patterns of the capillary networks, the ratio between the epithelium and the connective tissue which occupied the gingival region was nearly identical to that observed after 2 weeks of healing (Fig. 2). The upper two-thirds shows a network similar to that beneath the inner marginal epithelium, while the lower one-third shows a network similar to that in the connective tissue. At this stage of healing, a high density of the newly-formed capillary network was seen beneath the epithelium with a vascular architecture dominated by a highly glomerulus-like network, consisting mainly of dilated capillaries or venules. These ran longitudinally in the upper 1/2~2/3 of the gingiva, while a flat fishnet-like capillary network lined the remaining lower 1/2~1/3 (Fig. 4).

The capillary network in the connective tissue layer was similar to that observed after 2 weeks of healing with regard to both distribution density and the direction in which the vessels ran, which resulted in a ring-shaped formation around the implant. Most of the networks in the alveolar bone wall, as shown in Figure 2, had penetrated and entered the newly-formed bone. Accordingly, like the vascular distribution pattern...
that developed in natural teeth, the network in the lower margin of the connective tissue communicated with that in the alveolar bone only by a few ramus communicans.

Discussion

It is generally accepted that the healing process in peri-implant soft tissue is essentially the same after surgery as it is in a general wound with regard to the regeneration of connective tissue and vascularization.

To elucidate the events that occur during the healing process, we believe it is extremely important to clarify the vascular architecture which forms after implantation.

Although it has been accepted that peri-implant gingiva has a histologic structure similar to that of natural teeth, which consists of non-keratinized sulcular and junctional epithelium, we were unable to confirm this point here. In the present study, no significant difference was found in the height of the epithelium between healing for 2 and 4 weeks. The presence of a large number of inflammatory cells and phagocytes in the epithelium and the connective tissue may indicate that the peri-implant gingiva had not yet begun the healing process at 2 weeks after the implantation. This inflammation persisted during the regeneration of the gingiva at 4 weeks.

Using microvascular corrosion casts, we observed that a dense vascular network, consisting mainly of capillaries, was distributed beneath the inner marginal epithelium, and a capillary network, which had a distribution pattern similar to that in natural teeth, was formed beneath the epithelial layer around the implant. Although it is commonly accepted that these networks show various patterns of vascular distribution, we suspect that those beneath the inner marginal epithelium do not change, except in response to some disturbance, such as inflammation, of the tissue. This speculation was supported by our previous studies in which we observed the distribution patterns which appeared throughout permanent tooth germination, and the patterns which formed in the gingiva after treatment with a plaque control. In these studies, we found that a hexagonal network always appeared following germination, and a very similar vascular architecture was also seen in plaque-controlled gingiva.

It has also been suggested that there is a correlation between the severity of inflammation and the structure of the capillary network. Our previous studies on vascular architecture revealed that the structure of the network beneath the inner marginal epithelium can vary with the severity of the inflammation, as follows: 1. hexagonal shape, 2. hair-pin shape, 3. palisade-like shape, 4. spherical globular shape, and 5. spherical globular shape with a long foot. Based on these categories, the vascular architecture shown in Figure 4 can be classified type 5, since the upper one-half of the area beneath the epithelium was occupied by a network with a spherical globular shape and a long foot. This observation may also indicate that the most severe inflammation occurred in this region. In contrast, in the lower one-half the hexagonal network similar to type 1, together with signs of vasodilatation and the presence of a denser vascular distribution, may indicate the existence of minor inflammation.

Although the area between the lower margin of the junctional epithelium and the alveolar crest has been described as "the connectium tissue layer", we refer to it as "the cervical-periodontium", since it is equivalent to the area which contacts the rentis in natural teeth. The capillary network in the cervical-periodontium has been shown to differ from that beneath the inner marginal epithelium. The former is distributed in a rough capillary network on the extreme surface and by a vascular bundle which consists of capillaries, arteriola and venula, running transversely to surround the radix dentis in the shape of a ring. On the other hand, in the peri-implant soft tissue show in Figure 4, only the transverse-running ring network was formed, and no capillary network developed on the surface as in the cervical-periodontium. The direction in which blood vessels run is closely related to that of connective tissue fibers. Thus, the present results may suggest that peri-implant soft tissue has a different structure than that of connective tissue in natural teeth.
References