Histometric evaluation of periodontal surgery
I. The modified Widman flap procedure

JACK CATON AND STURE NYMAN

Department of Periodontology, Eastman Dental Center, Rochester, NY, USA
Department of Periodontology, Faculty of Odontology, University of Göteborg,
Göteborg, Sweden

Abstract. The present investigation was performed in the Rhesus monkey to determine the effect of the modified Widman flap procedure on the level of the connective tissue attachment and supporting alveolar bone. Two adult male Rhesus monkeys were used. Eighteen contralateral pairs of periodontal pockets were produced in a standardized manner. Surgical treatment of the pockets was performed around experimental teeth and the contralateral teeth were used as the unoperated controls. Twelve months following treatment the animals were sacrificed and histological sections obtained. Using the cemento-enamel junction (CEJ) as a fixed reference point, linear measurements along the root surface were made to the most apical cells of the junctional epithelium (JE), to the crest of the interproximal alveolar bone (CR), and to the apical extent of angular bony defects (AAD). These measurements from operated and unoperated pockets were then compared.

The data revealed that treatment of periodontal pockets using the modified Widman flap procedure produced no gain in connective tissue attachment and no increase in crestal bone height. In angular bony defects a certain degree "bone fill" was noted. This bone repair was never accompanied by new connective tissue attachment.

Regeneration of the periodontal tissues has long been an endeavor in the treatment of periodontal diseases. A variety of therapeutic techniques, such as root planing and curettage, gingivectomy-curettage, flap curettage, and flap procedures including transplantation of various materials into periodontal defects, have been used to obtain regeneration of the periodontium. (For an extensive review of the literature in this field, see Adell 1974.) Most studies on regeneration procedures describe a possibility of achieving de novo formation of the supporting tissues, particularly within infra-bony pockets, and present varying degrees of success in the treatment.

Methods utilized for assessing regeneration of the periodontal tissues have been discussed in recent years. Thus, criticism has been directed toward results presented from trials in humans (Friedman 1958, Nambers et al. 1967, Caton & Zander 1975, Armitage et al. 1977, van der Velden & de Vries 1978, Spray et al. 1978), because clinical assessments (pocket depth and attachment level measurements, radiographic analyses) do not provide proof of new attachment (aposition of root cementum, re-
THE MODIFIED WIDMAN FLAP PROCEDURE

dure
generation of periodontal ligament and alveolar bone). Clinical assessments, even in conjunction with reentry operations, can provide evidence for repair of alveolar bone but not for the new formation of cementum and a functioning periodontal ligament.

In order to overcome the limitations of clinical registrations as evidence of new attachment, various animal models have been developed. By the use of such models, new attachment can be evaluated histologically, which for ethical reasons is difficult in man. Certain prerequisites should, however, be fulfilled for an animal model to be optimal for testing the effect of new attachment procedures (Ramfjord 1971, Caton & Zander 1975): (1) the anatomy of the teeth as well as the morphological relationship of the tooth to the jaw should be similar to man, (2) it should be possible to experimen-tally produce periodontal pockets, including angular bony defects, with the characteristics of the human periodontal pocket, i.e. loss of connective tissue attachment, apical proliferation of the junctional epithelium and marginal alveolar bone resorption, (3) the pockets produced should not heal spontaneously, and (4) the animal model should permit the production of nearly identical pockets on contralateral teeth in order to facilitate intrasubject comparisons between treated and untreated areas. A model utilizing the Rhesus monkey as the experimental animal, meeting the above-mentioned demands, has been developed and described by Caton & Zander (1975) and Caton & Kowalski (1976).

The modified Widman flap procedure is frequently used in periodontal therapy (for review see Ramfjord 1977) Its use alone, or
combined with the insertion of various grafting materials into the pockets, has been advocated for the regeneration of the supporting tissues destroyed by periodontitis (Ellegaard 1976). The periodontal literature, however, appears to lack precise information regarding the effect of the modified Widman flap procedure on the connective tissue attachment level and the alveolar bone.

The purpose of the present investigation was to determine, in the Rhesus monkey, the effect of the modified Widman flap procedure as described by Ramfjord & Nørgaard (1974) on the connective tissue attachment level and supporting alveolar bone.

Material and Methods

Two adult male Rhesus monkeys (Macaca mulatta) with a full compliment of teeth and in good physical condition were used for the study. Blood, stool and T.B. cultures were performed at 3-month intervals; the animals were under the supervision of veterinary primatologists. The animals were maintained on a diet of Monkey Chow (Ralston Purina Co., St. Louis, Mo.) augmented with fresh fruit and water ad libitum.

Periodontal pockets were produced in animals by the method described by Caton & Zander (1975) and Caton & Kowalski (1976). Orthodontic elastics were placed bilaterally around the maxillary and mandibular central incisors and first molars, maxillary first premolars and the mandibular second premolars. In order to sufficiently deepen the periodontal pockets, posterior teeth, additional elastics were placed around the roots as bone loss progressed. When the probing depth was 8 mm (2–6 months after placement), the elastics were removed.

Three months after removal of the orthodontic elastics, all teeth were thoroughly

Fig. 3. The distances which were measured in the histologic sections:
1. Cemento-enamel junction (CEJ) to the apical cells of the junctional epithelium (IE).
2. CEJ to the crest of the alveolar bone (CR).
3. CEJ to the base of the bony defect (AAD).
(Dental-diastema section; orig. magn. X 8).

Die Abstände, die an den histologischen Sektionen gemessen wurden.
1. Schmelz-zementenerschritt (CEJ) bis zu den apikalen Zellen des Saumepithels (IE).
2. CEJ bis zur Lamina dura des Alveolarknochens (CR).
3. CEJ bis zum Grund des Knochendefekts (AAD). Mesio-distale Sektion; Orig. X 8 vergr.

Les distances mesurées sur les coupes histologiques:
1. De la jonction émail-cément (CEJ) aux cellules apicales de l’épithélium de jonction (IE).
2. De CEJ à la crête de l’os alvéolaire (CR).
3. De CEJ à la base de la lésion osseuse (AAD). (Coupe mesio-distale; grossissement orig. X 8).
of various pockets, has erosion of the bone and exposed root surfaces. The modified Widman flap procedure was described in detail by Widman, Pjetursson, and colleagues (1990). The procedure involved the removal of the gingival margin and the subsequent closure of the wound using interrupted sutures. The results of this procedure showed a significant reduction in pocket depth and an increase in the percentage of healed sites.

Fig. 4. Clinical appearance of an experimental site prior to surgery. Klinisches Bild einer Versuchsseite vor der chirurgischen Behandlung. Aspect clinique d'un site expérimental avant l'intervention chirurgicale.

scales utilizing curettes, which had been reduced in size. Plaque control was instituted at this time and included toothbrushing, interdental flossing and a 3-min topical application of 2% chlorhexidine gluconate solution three times a week (Monday, Wednesday, and Friday). This plaque control regimen results in the establishment and maintenance of clinically healthy gingiva in this species (Caton 1979). The mean GI score was found to be 0.22 (Loe & Silness 1963). These plaque control measures were continued on both sides of the jaws until the animals were killed.

Three weeks after the institution of plaque control, the surgical procedures were performed around the experimental teeth on one side of the jaw. The contralateral teeth remained as unoperated controls. Sulcular incisions were made to the alveolar crest on the facial and lingual aspects of the teeth. The incisions extended one tooth mesial and distal to the experimental teeth. A mucoperiosteal flap was elevated and all granulation tissue was removed (Fig. 1). The inner surface of the flap was thoroughly curetted and trimmed with tissue scissors in order to remove the epithelial lining of the pocket. Following planning of the exposed root surfaces, the surgical site was irrigated with sterile saline, and the flap was replaced to its preoperative position and secured with 4-0 silk interrupted, interdental mattress sutures to accomplish complete coverage of the alveolar bone (Fig. 2). The animals were placed in a restraining chair for 2 weeks to prevent unwanted interference with the surgical sites and 600,000 units of penicillin G was administered intramuscularly once a day for 14 days. The ani-

Fig. 5. Clinical appearance of an experimental site after 1 month of healing. Klinisches Bild einer Versuchsseite nach 1-monatlicher Heilung. Aspect clinique d'un site expérimental après un mois de cicatrisation.
nals were fed a modified diet postoperatively. For the first 2 weeks they were maintained on soft bananas, a liquid high protein supplement and water ad libitum.

For the following 3 weeks, the protein supplement was replaced with fine ground Monkey Chow mixed with water. From the sixth week after surgery the animals were fed their normal diet. The regular plaque control regimen was altered for the initial 2 weeks after operation. During this period only chlorhexidine solution was applied 3 times a week. Starting 2 weeks after surgery, the teeth on the operated and operated sides were scaled and polished every 60 days until the animals were killed, which was 1 year after surgery.

When removed from their cages, the
and a typically
chirurgisch be
nach apikal der
sion. Org. 10 X
jonction (JE) o
de flèche) et à
for the initial 2
ning this period
weeks after sur
graphic and un
polished and ani

cages, the ani
mals were sedated with phenacyclidine HCl.
2 mg/kg bodyweight. For the surgical proce-
dures, pentobarbital sodium was adminis-
trated intravenously and titrated to obtain
maximum sedation while retaining the eye
blink reflex.

The animals were killed with an intra-
vavenous overdose of pentobarbital sodium
and the jaws dissected free and fixed in
Lavdovsky's solution. After 72 h of fixa-
tion, the jaws were washed for 24 h in fil-
tered running tap water and placed in 4 %
nitric acid for decalcification. After 7 days,
the jaws were sectioned, each section
(block) containing the teeth under investi-
gation and those adjacent to them. When
decalci cation was complete, the blocks
were again washed for 24 h in filtered run-
ning tap water, dehydrated in increasing con-
centrations of ethyl alcohol and embedded
in celloidin. The blocks were mounted for
sectioning in a mesio-distal plane starting
on the buccal surface. The sections were
cut at 12 μm thickness and step serial sec-
tions at 96 μm intervals were mounted and
stained with hematoxylin and eosin.

Histological analysis was performed on
step serial sections at 190 μm intervals with
a calibrated grid mounted in the eyepiece of
a light microscope at 35 X. Using the ce-
mento-enamel junction (CEJ) as a fixed re-
ference point, linear measurements along the
root surface were made to the most apical
cells of the junctional epithelium (JE), to
the crest of the interproximal alveolar bone
(CR), and to the apical extent of angular

Fig. 7. In this specimen, 0.25 mm of new
cementum (C) has formed coronal to the
curette mark (arrow); JE = apical end of
junctional epithelium. (Mesial-distal sec-
tion; orig. magn. × 10).

Dieses Präparat hat neuen Zement (C) in
einer Breite von 0.25 mm coronal der Kü-
rettenmarke (Pfeil) gebildet; JE = apikale
Grenze des Saumepithels. (Mesio-distale
Sektion; Orig. 10 X × verg.).

Dans ce spécimen, il s'est formé 0.25 mm
de nouveau cément (C) à un niveau coro-
nal par rapport à la marque Iaste par
la curet (pointe de flêche); JE = extré-
mité apicale de l'épithélium de jonction.
(Coupe métio-distale; grossissement orig.
× 10).
Table 1. Statistical analysis of the measured distances CEJ-JE, CEJ-CR, and CEJ-AAD in the surgically treated and corresponding control sites.

<table>
<thead>
<tr>
<th>Distance measured</th>
<th>X Operated sites in mm (+)</th>
<th>X Control sites in mm (-)</th>
<th>d</th>
<th>SE</th>
<th>t</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>CEJ-JE</td>
<td>3.38</td>
<td>3.51</td>
<td>-0.13</td>
<td>±0.12</td>
<td>-1.08</td>
<td>0.3-0.2</td>
</tr>
<tr>
<td>CEJ-CR</td>
<td>3.09</td>
<td>3.20</td>
<td>-0.11</td>
<td>±0.09</td>
<td>-1.13</td>
<td>0.3-0.2</td>
</tr>
<tr>
<td>CEJ-AAD</td>
<td>3.60</td>
<td>3.82</td>
<td>-0.22</td>
<td>±0.11</td>
<td>-2.00</td>
<td>0.1-0.05</td>
</tr>
</tbody>
</table>

CEJ = cemento-enamel junction (Schmelz-zementgrenze, Jonction cément-émail), JE = apical extent of the junctional epithelium (apikale Zellen des Saumepithels, Niveau apical de l'épithélium de jonction), CR = crest of the interproximal alveolar bone (Lamina dura des Alveolknochen, Crête de l'os alvéolaire interproximal), AAD = apical extent of angular bone defect (Defekt des Knochendefektes, Niveau apical de la lésion ostéaire angulaire), d = mean of the differences (Mittelwert der Unterschiede, Moyenne des différences), Distance measured (gemessene Abstände, distance mesurée), operated sites in mm (operierte Seiten in mm, sites opérés en mm), control sites in mm (Kontrollseiten in mm, sites témoins en mm).

bony defects (AAD) (Fig. 3). The data derived from these measurements of operated and unoperated sites were subjected to statistical analysis as described previously (Caton & Kowalski 1976). Eighteen sites from the operated side of the animals were compared with corresponding unoperated sites by the paired sample t-test (Chilton 1967).

All histologic measurements were made by the same person. Intraexaminer error for histologic measurements was determined before and after the analysis by performing three sets of measurements, 72 h apart, on 10 randomly chosen sections and then calculating the mean for each parameter. The percentage difference from this mean of each set was determined and the intraexaminer error expressed as the average percentage difference.

In addition to the histologic measurements, a histological evaluation of healing was performed. Particular attention was paid to signs of new cementum, extension of the junctional epithelium, and osseous repair in angular bony defects.

Results

Clinical observations

The animals remained healthy for the entire course of the experiment and demonstrated a progressive gain in weight. Blood, stool and T.B. tests remained within normal limits.

Scaling and subsequent plaque control markedly reduced the gingival inflammation prior to the surgical procedure (Fig. 4). A decrease of redness, swelling and bleeding was noted clinically, but the preoperative probing depth remained essentially the same, namely between 6 and 8 mm.

Postoperatively, healing was rapid, so that within 2 weeks the wounds were closed and mechanical means of plaque control could be reinstated. At 1 month, healing was advanced and the gingiva exhibited minimal signs of inflammation (Fig. 5).

Histologic and histometric analysis

Connective tissue attachment level (CEJ-JE). Analysis of the histological specimens
from the surgically treated teeth demonstrated that the most apical cells of the junctional epithelium were consistently located at or near the most apical level to which the roots were planed (Fig. 6A, B). In five operated sites, 0.10–0.25 mm of new cementum had formed coronal to the curette mark (Fig. 7). In these areas a functional periodontal ligament had reorganized. The results of the paired difference t-test (Table 1), which compared the CEJ-CR measurements for the operated and unoperated sites, showed that there was no significant difference between the two.

Crestal bone height (CEJ-CR). The measurements of the distance CEJ-CR disclosed that no increase in crestal bone height occurred during the healing period following surgery. The result of the paired difference t-test comparing the distance CEJ-CR in the operated and unoperated areas (Table 1) showed that there was no significant difference between the two.

Bone repair in angular bony defects (CEJ-AAD). Observation of the histological sections revealed "bone fill" in angular bony defects on the operated side, although extensive fill (Fig. 8) occurred in only two sites. "Bone fill" was, however, in no instance accompanied by new connective tissue attachment. A long junctional epithelium was always found to be interposed between the new bone and the root surface (Fig. 8), and the collagen fibers of the "pe-
periodontal ligament" were arranged parallel to the root surface. When "bone fill" was observed, few inflammatory cells or bone fill was present in the connective tissue adjacent to the bone. The t-test, which compared CEJ-AAD measurements for the operated and unoperated sites, demonstrated no significant difference (Table 1). There was, however, a tendency for this distance to be smaller on the operated side (P = 0.1-0.05), which coincided with the histological observation of extensive "bone fill" in two of the experimental sites.

Intrasaxinex error for the histologic measurements CEJ-IE, CEJ-CR and CEJ-AAD were 3.1, 3.4 and 4.1 %, respectively.

Discussion

New attachment implies the apposition of new cementum and the regeneration of a functionally oriented periodontal ligament and supporting alveolar bone coronal to the level of these tissues before treatment. Therefore, in order to evaluate whether new attachment has occurred following therapy, the coronal level of these tissues components of the periodontium before and after the operation must be determined. This demand was met in the present study by using an animal model which permits the production of nearly identical periodontal pockets on contralateral teeth. In histological sections, the position of the involved tissue components can then be properly assessed on operated and unoperated contralateral periodontal units. This model was described in detail in previous reports (Catton & Zander 1975, Catton & Kowalski 1976).

The most conspicuous observation made in the present study was the absence of new attachment 12 months following the treatment of periodontal pockets using the modified Widman flap procedure. This observation appears to be at variance with previous human studies in which the results were evaluated by clinical means (Ellegaard & Løe 1971, Ramfjord et al. 1975, Yukna et al. 1976, Rosling et al. 1976). In these studies, a decreased probing depth following healing was reported. It has recently been shown in a number of experiments, however, that periodontal healing by no means a reliable method for the accurate estimation of connective tissue attachment level (see Linstad 1980). For example, termination of the coronal intact connective tissue attachment by gingival inflammation has resulted in healing and when a long junctional epithelium has been formed. Furthermore, this condition will not identify the apical extent of epithelial attachment when the root surface area of a normal angular bony defect. In the present study, the healing result was assessed by histologic rather than clinical means, and the level of the apical cells of the junctional epithelium was located at or close to the surgical position in all treated teeth. The observation was unrelated to the morphology of the periodontal defects (i.e., alveolar or infrabony pockets). Thus, differences in methodology of assessing the coronal level of the intact connective tissue attachment in the studies by Ellegaard & Løe (1971), Ramfjord et al. (1975), Yukna et al. (1976) and Rosling et al. (1976) and in the present study may explain the different results. This explanation is supported by Yukna's findings in monkeys.

In five operated sites, a small amount (0.10-0.25 mm) of new cementum was found coronal to the apical level of radiographic instrumentation. This finding is consistent with observations by Frank et al. (1972) and Linstad & Karr (1979). They reported deposition of new cementum in the most apical areas of healing periodontal defects in dogs, rats, and rabbits. In these studies, the portion of the root surface exposed to the oral environment was a factor and that the apical level of the healing defect was not an accurate indicator of the extent of new attachment.

This investigation was supported by the National Institute of Dental Research.
periodontal wounds in humans, monkeys and dogs, respectively. It is possible that the formation of new cementum in the apical portion of some pockets occurred on root surfaces exposed as the result of a root planing which extended beyond the presurgical level of the junctional epithelium.

Healing following the modified Widman flap procedure did not produce increased crestal alveolar bone height but bone repair occurred to a varying extent within infrabony pockets. The most pronounced bone regrowth was found where there were no obvious signs of inflammation in the connective tissue adjacent to the bone walls. This finding supports results presented by Rosling et al. (1976) and Polson & Heijl (1978). These investigators demonstrated significant bone repair in infrabony pockets following the modified Widman flap procedure in humans in whom meticulous plaque control was maintained. It should be stressed, however, that in the present material osseous regrowth in infrabony pockets was not accompanied by new connective tissue attachment. In fact, the histologic sections revealed that a junctional epithelium was always interposed between the new bone and the root surface and extended to a level near the apical extension of root planing. This finding confirms previous observations by Caton & Zander (1976).

Acknowledgement
This investigation was supported by USPHS Research Grant D-1648 from the National Institute of Dental Research, National Institutes of Health, Bethesda, Md.

Zusammenfassung
Histometrische Auswertung parodontaler Chirurgie. I

Résumé
Evaluation histométrique des interventions de chirurgie parodontale. I. L'opération à lambeau selon la technique de Widman modifiée
La présente étude a été effectuée sur le singe Rhésus, pour déterminer l'effet de l'opération à lambeau selon la technique de Widman modifiée sur le niveau de l'attachement de tissu conjonctif et de l'os alvéolaire de soutien. Pour cette étude, deux singes Rhésus mâles adultes ont été utilisés. La formation de dix-huit paires contralatérales de culs-de-sac a été provoquée de façon standardisée. Un traitement chirurgical des culs-de-sac a été effectué autour des dents expérimentales, et les dents contralatérales ont servi de témoins sans traitement chirurgical. Douze mois après l'intervention, les animaux ont été sacrifiés et des coupes histologiques ont été préparées. A partir de la jonction cémento-émail (CEJ), utilisée comme point de référence, les mesurations linéaires ont été faites le long des surfaces radiculaires jusqu'aux cellules les plus apicales de l'épitéh-
lum de jonction (JE), jusqu'à la crête de l'os alvéolaire interproximal (CR), et jusqu'au niveau apical atteint par les lésions osseuses angulaires (AOA). Les mesures obtenues au niveau des cuills-de-sac opérés et des cuills-de-sac non opérés ont ainsi été comparées. Les résultats ont mis en évidence que le traitement des cuills-de-sac par l'opérant à lameau selon la technique de Widman modifiée ne produisait pas d'accumulation de l'attachement de tissu conjonctif, ni d'augmentation de la hauteur de la crête osseuse. Dans les lésions osseuses angulaires, on notait un certain degré de remplacement osseux. Cette réparation osseuse n'était jamais accompagnée d'un nouvel attachement de tissu conjonctif.

References


