



THE PERIAPICAL PERIODONTIUM CAN TOTALLY REGENERATE AFTER LEGA ARTIS ROOT CANAL THERAPY



The regeneration of the marginal periodontium is much more difficult and a complex process

PERIODONTAL WOUND HEALING

Biological Problems

 Open system (continuous bacterial contamination)

 Scarce blood supply to the cementum and dentin **REATTACHMENT** – THE UNIFICATION OF THE SHARPEY FIBERS OVER THE DENUDED ROOT SURFACE AFTER AN ACUTE SHORT TERM SEPARATION

NEW ATTACHMENT - THE TOTAL REFORMATION OF THE SHARPEY FIBERS AND CEMENTUM

COMPLETE NEW ATTACHMENT - THE TOTAL REFORMATION OF CEMENTUM, BONE AND SHARPEY FIBERS

PERIODONTAL WOUND HEALING

Reparation

- LONG JUNCTIONAL EPITHELIUM (LJE)
- CEMENTUM RESORPTION
- ANKYLOSIS
- Regeneration
 - NEW CONNECTIVE TISSUE
 ATTACHMENT(CEMENTUM AND PDL)
 - COMPLETTE PERIODONTAL REGENERATION (BONE, PDL, CEMENTUM)

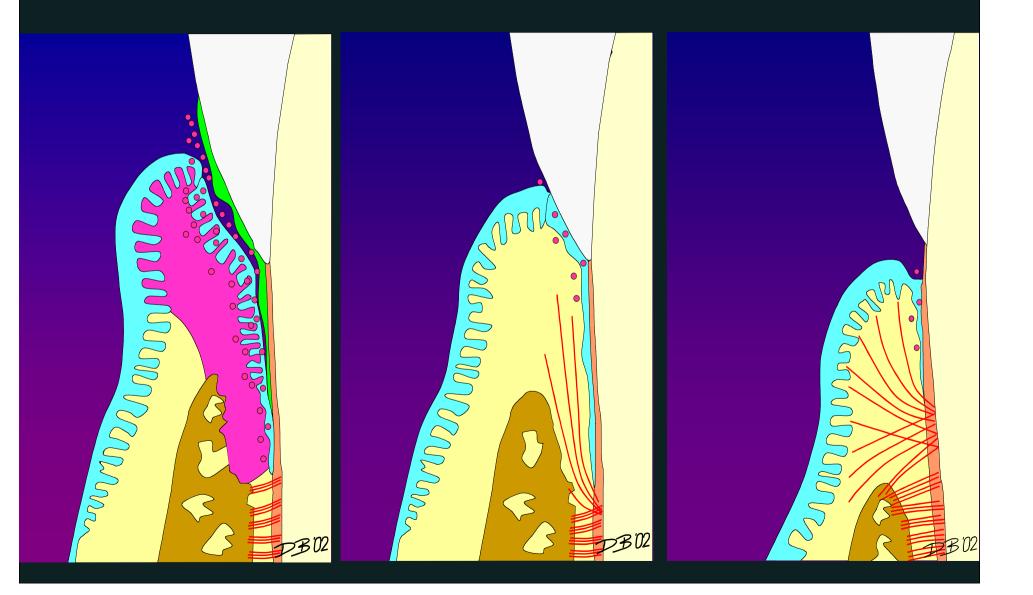
Reparation

- LONG JUNCTIONAL EPITHELIUM (LJE)
- CEMENTUM RESORPTION
- ANKYLOSIS

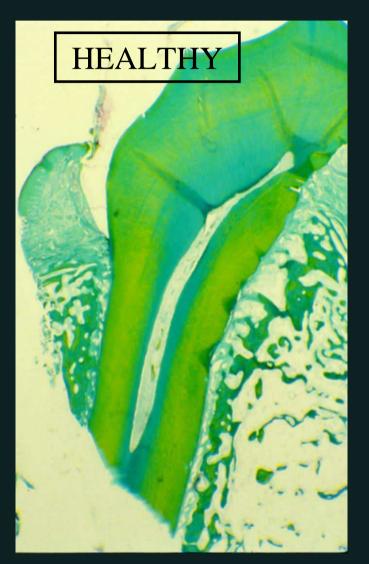
Periodontitis

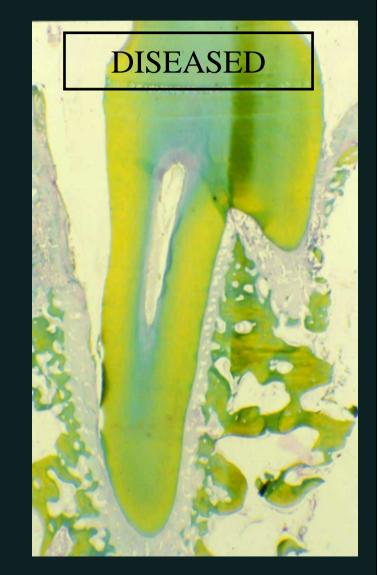
Reparation

Regeneration



PERIODONTIUM





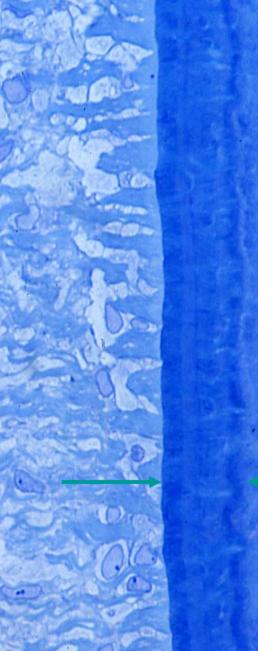
PERIODONTAL REGENERATION INVOLVS THE REFORMATION OF ALL THE THREE ELEMENTS OF THE ATTACHMENT APPARATUS:

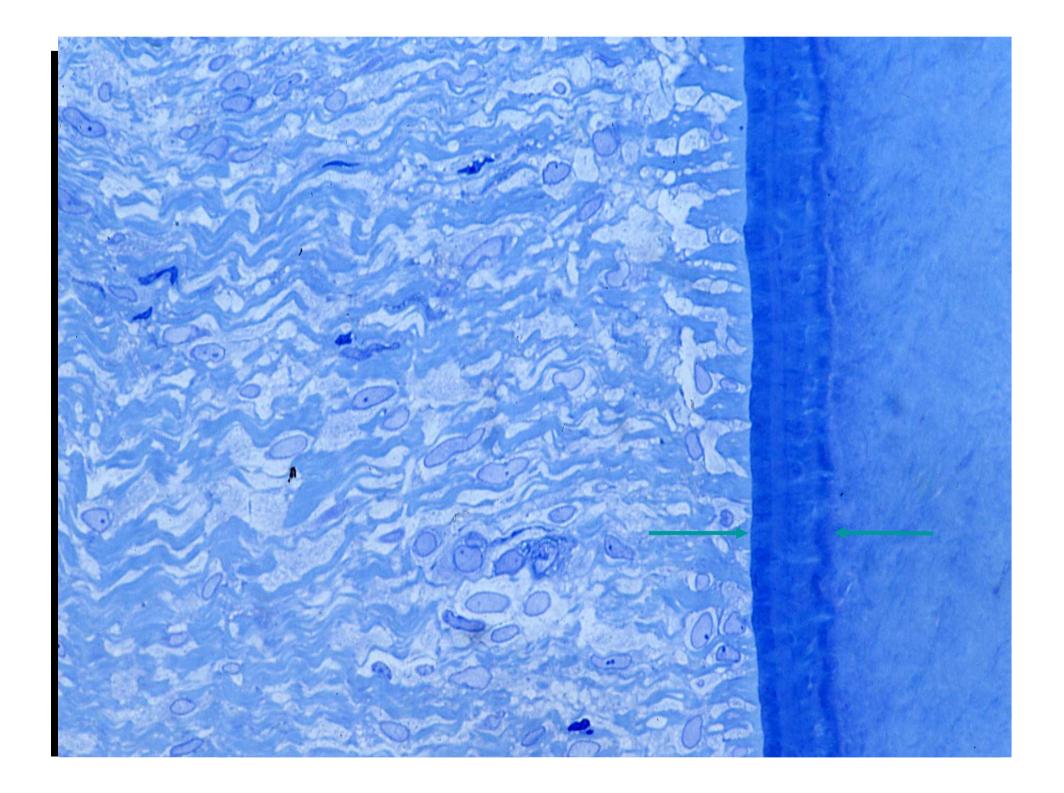
- CEMENTUM
- BOME
- PERIODONTAL
 LIGAMENT

This goal is attained by the presence of specific cell types, that occupy the defect area and form structures typical of the periodontal tissue during wound healing.

At the same time gingival cell types must be prevented from proliferating into the defect region

(Caton, 1976).





REATTACHMENT – THE UNIFICATION OF THE SHARPEY FIBERS OVER THE DENUDED ROOT SURFACE AFTER AN ACUTE SHORT TERM SEPARATION

NEW ATTACHMENT - THE TOTAL REFORMATION OF THE SHARPEY FIBERS AND CEMENTUM

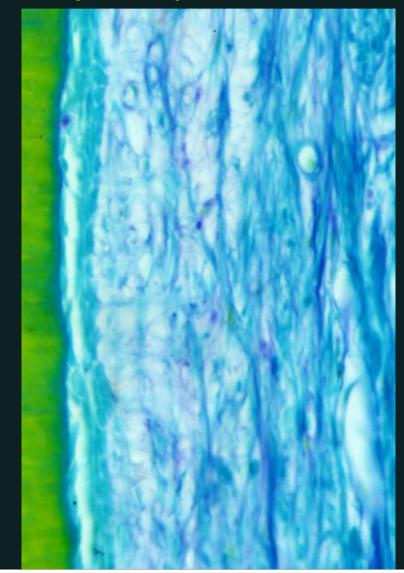
COMPLETE NEW ATTACHMENT - THE TOTAL REFORMATION OF CEMENTUM, BONE AND SHARPEY FIBERS

Reparation

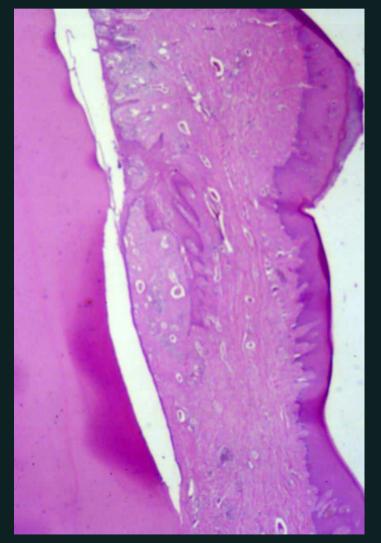
 AFTER CONVENTION PERIODONTAL SURGERY THE PERIODONTAL POCKET HEALS WITH LONG EPITHELIAL ATTACHMENT (LJE)

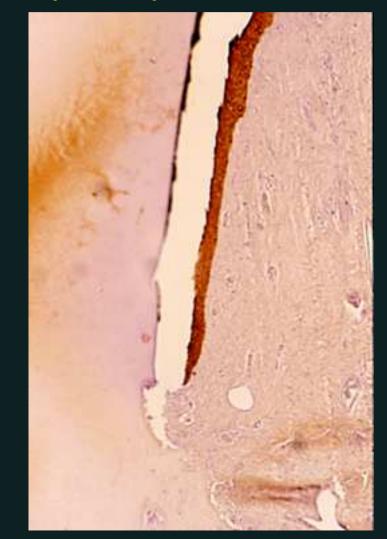
LONG JUNCTIONAL EPITHELIAL ATTACHMENT (LJE)





LONG JUNCTIONAL EPITHELIAL ATTACHMENT (LJE)



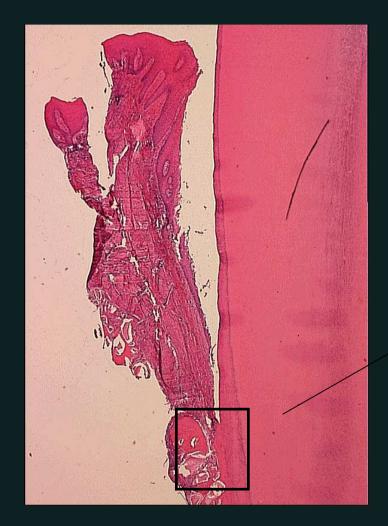


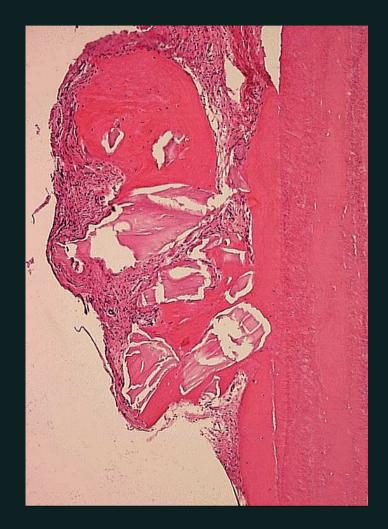
LONG JUNCTIONAL EPITHELIAL ATTACHMENT (LJE)



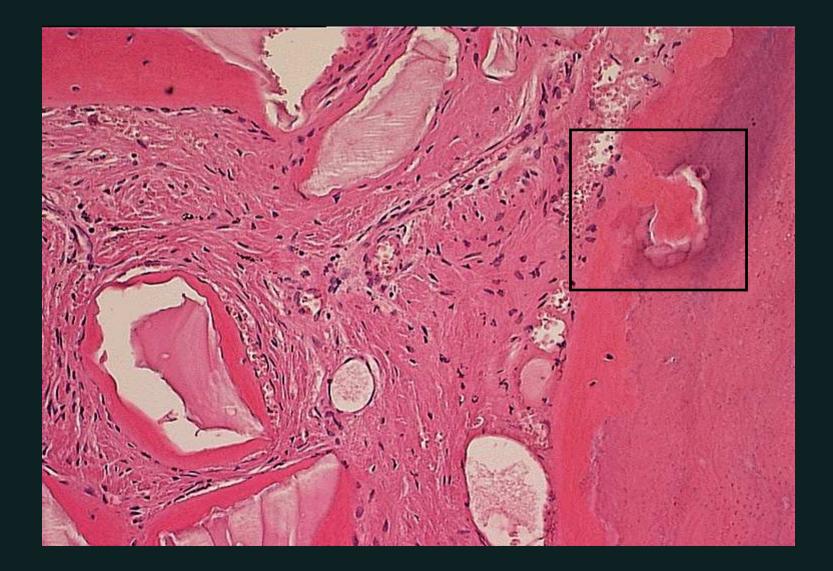
Cytokeratin histochemical detection

ANKYLOSIS

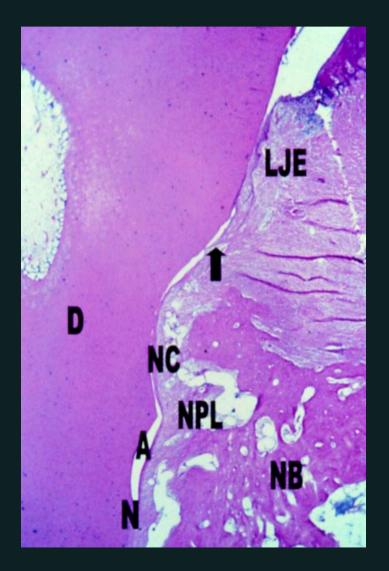


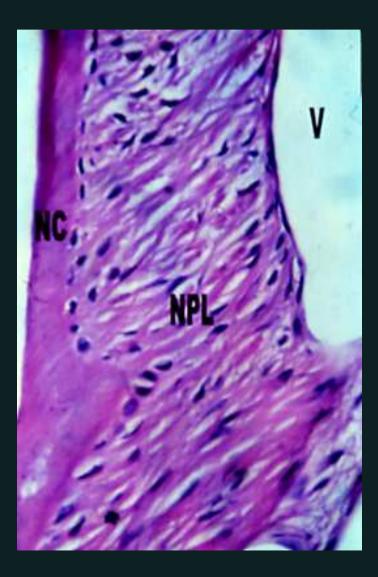


CEMENTUM RESORPTION



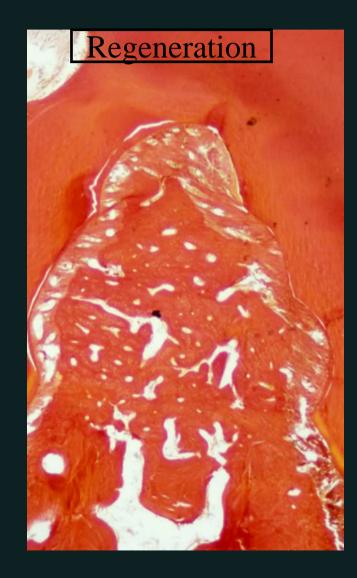
PERIODONTAL REGENERATION



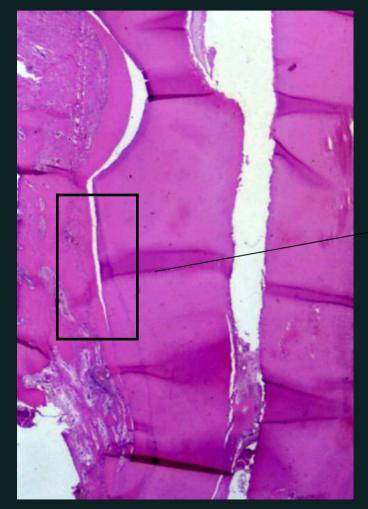


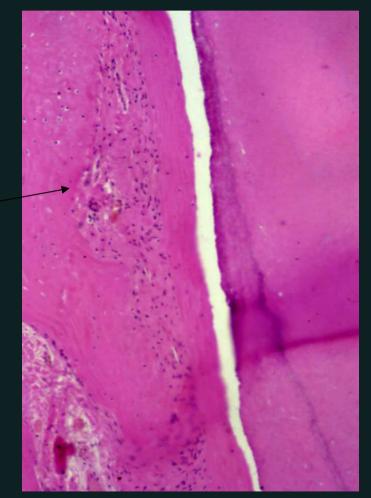
WOUND HEALING





REGENERATING PERIODONTAL ATTACHMENT

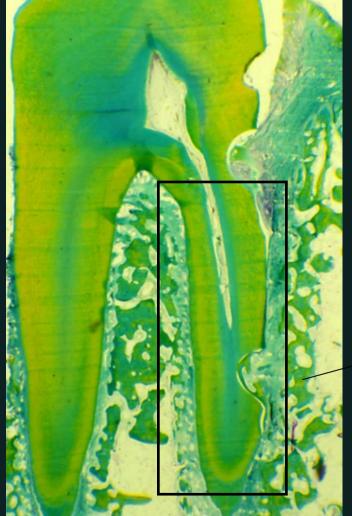


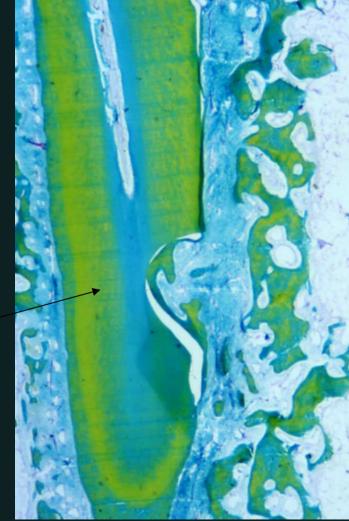


THE CARACTERISTICS OF THE REGENERATING ATTACHMENT APPARATUS

- NEW CEMENTUM FORMATION WITH
 MAINLY CELLULAR CEMENTUM
- NEW SHARPY'S FIBERS FORMATION
- REGENERATING OXYTALAN FIBERS
- NON REGENERATING MALASSEZ
 CELLS

REGENERATING ROOT CEMENTUM

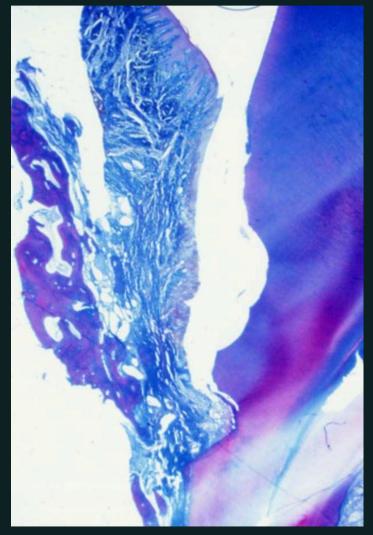




NEW BONE FORMATION WITH LONG EPITHELIAL ATTACHMENT

 The regeneration of the alveolar bone can occur without the reformation of new PDL and root cementum

BONE REGENERATION WITH LJE



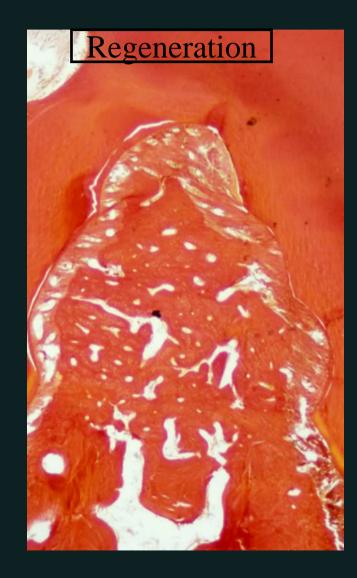


Charakteristics for the regenereted PDL

- Strong Osteopontin (OP) expression
- Strong Bone Sialoprotein (BSP) expression
- Strong Collagen type I and III expression
- No Cytoskeratin expression
- Strong Vimentin expression (i.e. Mesenchymal cell proliferation)

WOUND HEALING





REATTACHMENT – THE UNIFICATION OF THE SHARPEY FIBERS OVER THE DENUDED ROOT SURFACE AFTER AN ACUTE SHORT TERM SEPARATION

NEW ATTACHMENT - THE TOTAL REFORMATION OF THE SHARPEY FIBERS AND CEMENTUM

COMPLETE NEW ATTACHMENT - THE TOTAL REFORMATION OF CEMENTUM, BONE AND SHARPEY FIBERS

HISTORY

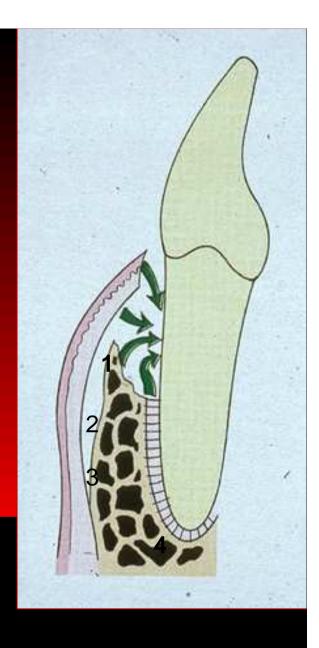
Regenerative potential of the periodontal structures

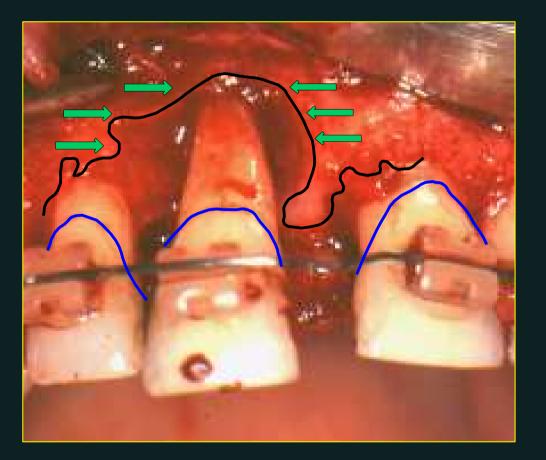
In 1976, Melcher postulated that, the cells which repopulate the root surface after surgery determine the nature of the attachment that will form.

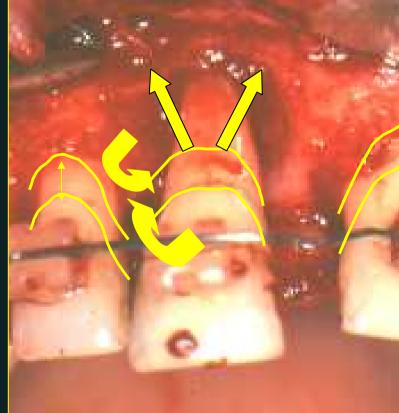
Root surface may be repopulated by 4 different types of cells:

- 1. Epithelial cells.
- 2. Cells originating from the gingival connective tissue.
- 3. Cells originating from the bone tissue.
- 4. Cells originating from the periodontal ligament.

Melcher, AH. On the repair potential of periodontal tissues. J Periodontol 1976;47:256-260.







PERIODONTAL WOUND HEALING

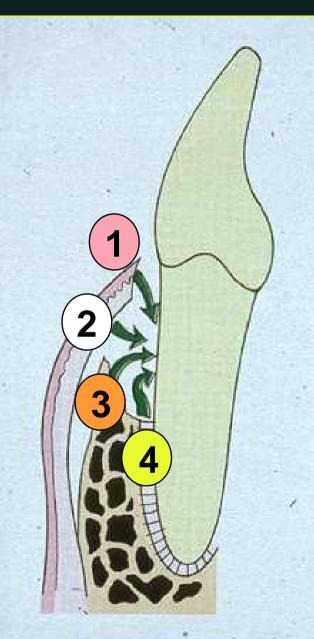
AFTER FLAP OPERATION THE WOUND CAN BE REPOPULATED BY FOUR GROUP OF CELLS:

Epitheleial cells

Gingival connective tisseue

Alveolar bone cells

Periodontal ligament cells



• If the cells from the epithelium proliferate apically to its presurgical level, an epithelial attachment (long junctional epithelium) will result.

The contact between the root surface and this junctional epithelium is maintained by cuticular structures and hemidesmosomes.



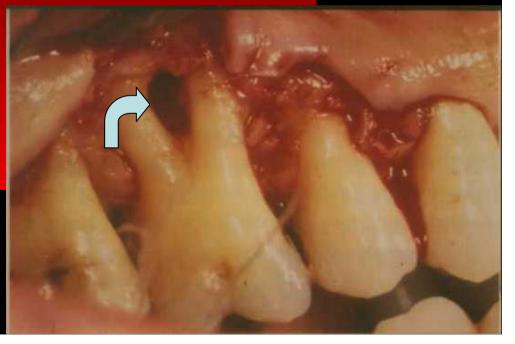
• If cells originating from the gingival connective tissue migrate into contact with the root surface, it has been assumed that **some kind of connective tissue adhesion or** attachment between the soft and hard tissues may be established.



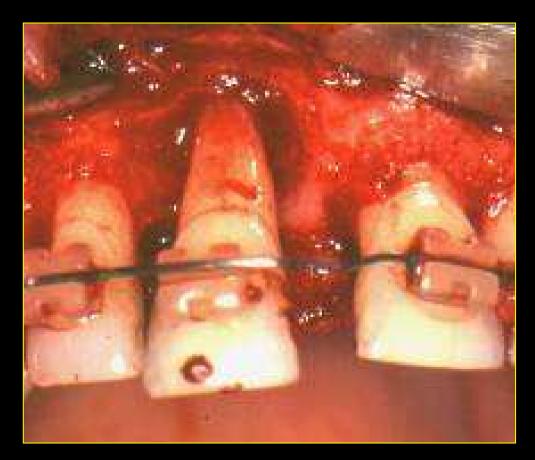
• If cells originating from bone tissue are migrating into contact with the curetted root surface, root resorption and ankylosis can occur.

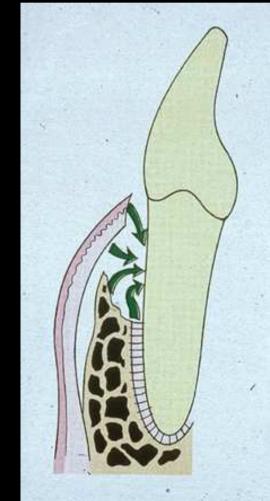


 Ideal conditions for healing may develop in situations where cells originating from the periodontal ligament proliferate to cover the previously diseased root surface. Such cells have, according to Melcher
 (1976), the capacity to form cementum and periodontal ligament fibers.



EXPERIMENTAL EVIDENCES





GINGIVAL EPITHELIUM

Björn 1965–

MWF SURGERY

POSTOPERATIVE REGULAR GINGIVECTOMY TO REMOVE GINGIVAL EPITHELIUM TO ANTICIPATE THE APICAL MIGRATION OF THE SULCUS EPITHELIUM

SIGNIFICANTLY LARGER POCKET REDUCTION THAN IN CONTROLS.

CONCLUSION

HEW CONNECTIVE TISSE ATTACHMENT CAN ONLY OCCURE IF THE POCKET EPITHELIA DOES NOT INTERFERE WITH THE CONNECTIVE TISSUE HEALING.

Björn H, Hollender L, Lindhe J. Tissue regeneration in patients with periodontal disease. Odontologisk Revy 1965;16:317-326.

Caton, J. & Zander, H. A.: Osseous repair of an intrabony pocket without new attachment of connective tissue. 1976. J. Clin. Periodontol. 3:54-58.

Epithelium

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to study the healing of the periodontal structures after different modalities of periodontal therapy. monkey model,

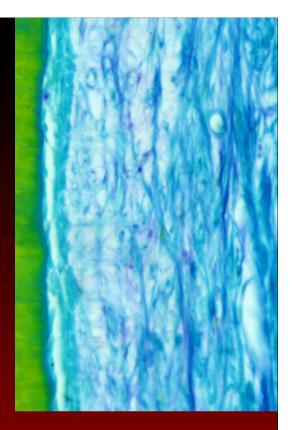
1. Periodic closed curettage.

- 2. Modified Widman flap operation without osseous surgery.
- 3. Modified Widman flap, + autogenous cancellous bone.
- 4. Modified Widman flap + bone substitute, (beta tricalcium phosphate).

Histometric analysis:

all 4 treatment procedures resulted long junctional epithelium

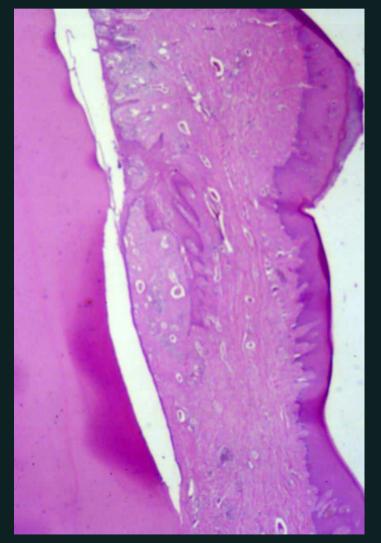
The results of the studies: new attachment formation is inhibited by the apically migrating dentogingival epithelium.

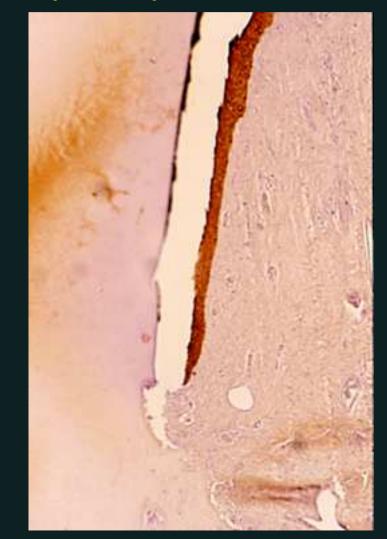


The rapid proliferation of the epithelium prevent the gingival connective tissue from establishing a close contact with the root surface.

An early contact between the gingival connective tissue and the root has been regarded as the prime prerequest for the formation of a new connective tissue attachment

LONG JUNCTIONAL EPITHELIAL ATTACHMENT (LJE)

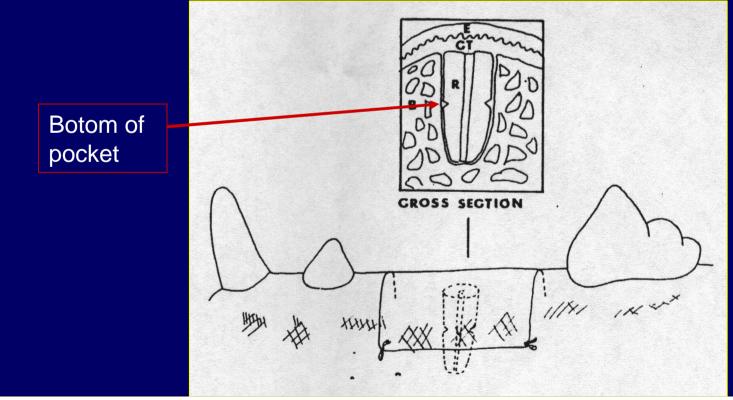




Karring T, Nyman S, Lindhe J. Healing following implantation of periodontitis affected roots into bone tissue. J Clin Periodontol 1980;7:96-105.

Experimental periodontitis in dogs

- Periodontitis- affected teeth after rootplaning and total decoronation were reimplanted in artificial sockets prepared in the edentulous ridge
- The gingival flap was tightly sutured to prevent the apical migration of the epithelia (totally submerged roots).



Karring et al. (1980)

New connective tissue attachment could develop around decoronated roots if the gingival flap was tightly closed during the whole period of postoperative healing

If the suture was not tight enough and the gingival flap opened up the gingival epithelium could migrate apically,

new connective tissue attachment never occurred.

conclusion:

The epithelial migration is the biggest obstacle of the connective tissue regeneration

Karring T, Nyman S, Lindhe J. Healing following implantation of periodontitis affected roots into bone tissue. J Clin Periodontol 1980;7:96-105.

8

The role of gingival connective tissue and alveolar bone

Periodontal tissue breakdown was produced around certain teeth, (experimental teeth) in monkeys and dogs

according to Caton and Zander (1975).

Caton J ,Zander HA. Primate model for testing periodontal treatment procedures: I. Histologic investigation of localized periodontal pockets produced by orthodontic elastics. J Periodontol 1975; 2:71-77.

EXPERIMENTAL PERIODONTITIS IN DOGS AND MONKEYS

50% ATTACHMENT LOSS

recipient sites in the edentulous mandible for subsequent implantation of the diseased roots

crowns of the experimental teeth were resected.

Nyman S, Karring T, Lindhe J, Planten S. Healing following implantation of periodontitis-affected roots into gingival connective tissue. J Clin Periodontol 1980;7:394-401.

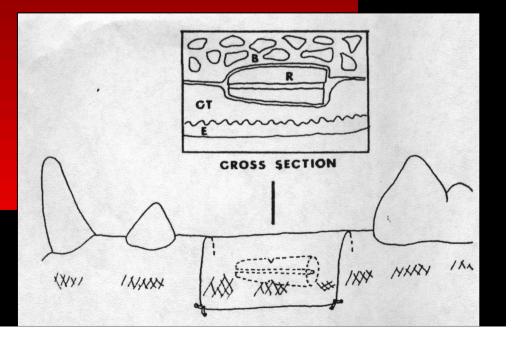
9/a

Full-thickness MW flaps

the diseased parts of the root scaled and rootplaned.

Notches at the level of the marginal bone crest.

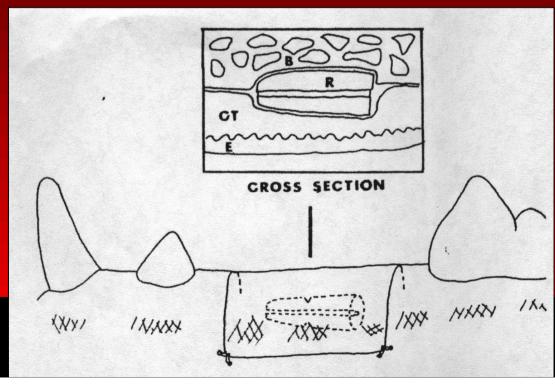
Recipient site : full thickness flap horizontal grooves for roots to be implanted

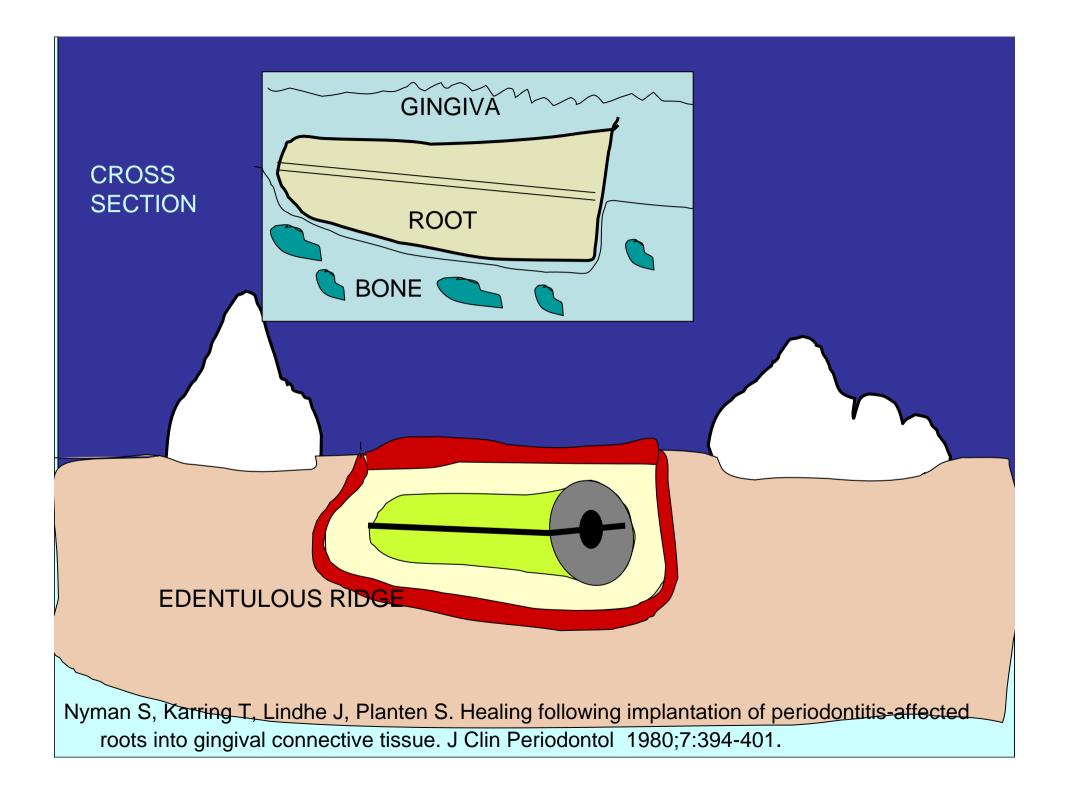


7. The roots of the experimental teeth were extracted and placed in an arteficial socket in the alveolar bone.

8. The flaps were repositioned and sutured to completly cover the implanted root and the surrounding bone.

With this experimental design, the epithelium was prevented from migrating into the wound.



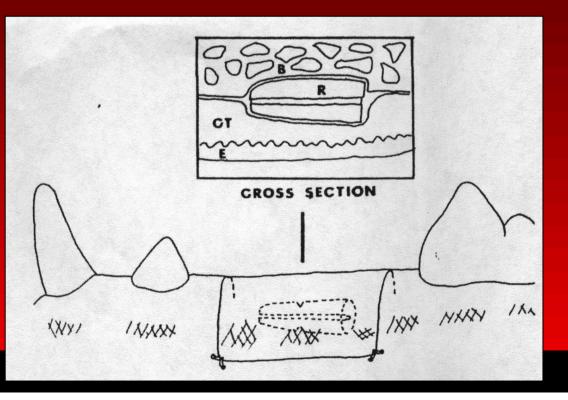


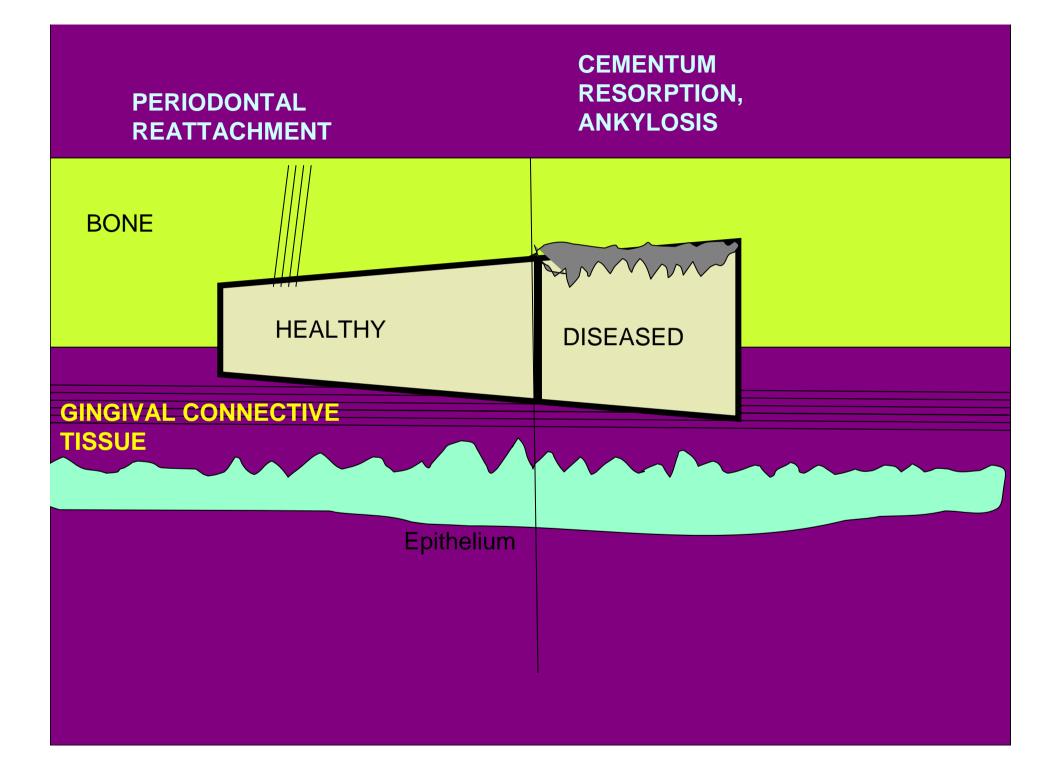
10/a

Each implanted root was embedded to half its circumference into bone, and the remaining part was covered by the gingival connective tissue of the flap.

After 3 months of healing, -

histological examination.





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The diseased root faced the gingival connective tissue

Collagen fibers parallel to the root surface. No de novo formation of root cementum Cementum resorption with multinucleated

The diseased root facing the bone tissue

Healing with extensive root resorption and ankylosis.

11/a

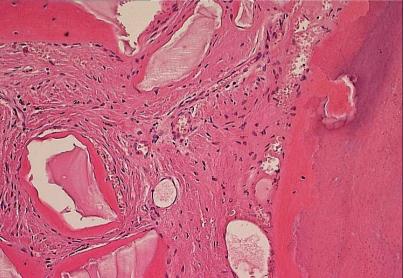
Granulation tissue derived from gingival connective tissue produced root resorption

Granulation tissue derived from bone produced resorption and ankylosis.

This experiment revealed that granulation tissue originating from bone or gingival connective tissue lacks the ability to establish a new connective tissue attachment.

• Root resorption is not a common complication following traditional flap surgery,

• Ankylosis is a rare complication following treatment of infrabony pockets.

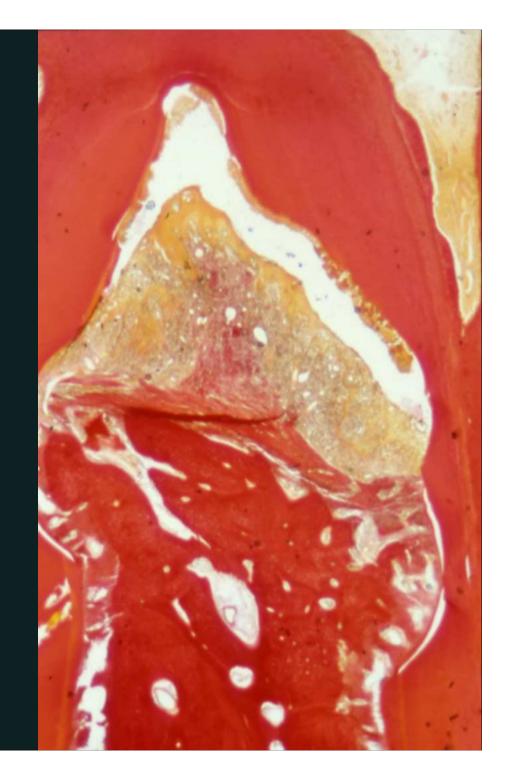


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In practice the newly formed epithelial lining acts as a barrier protects the root from granulation tissue from the alveolar bone and the gingiva.

The migrating epithelium functions as a protective barrier towards root resorption and ankylosis during healing following conventional periodontal therapy.

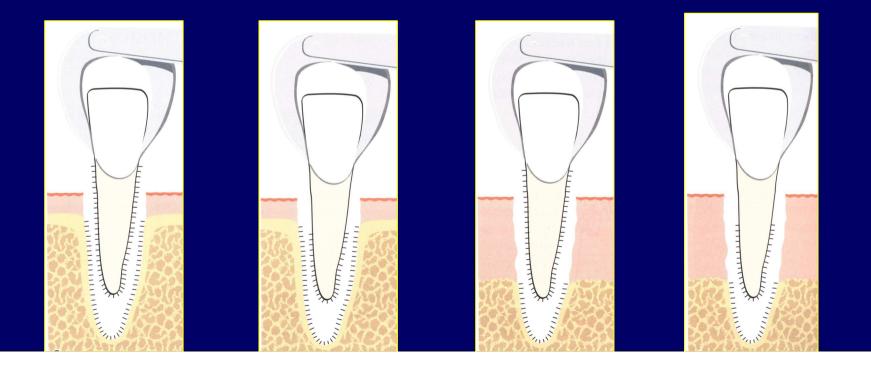




Lindhe J, Nyman, S, Karring, T. Connective tissue attachment as related to presence or absence of alveolar bone. J Clin Periodontol 1984;11:33-40.

INCISORS WERE EXTRACTED AND LATER REIMPLATEN IN THEIR OWN SOCKET :

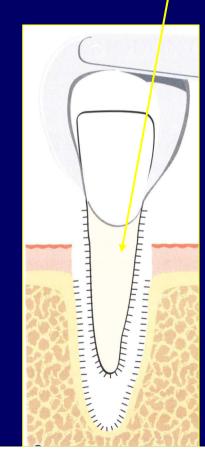
- Non- root planed teeth in sockets with nomral bone height
- Root planed in the coronal third and reimplanted in sockets with normal bone height
- Non- root planed teeth in sockets with reduced bone heigth
- Root plaed teeth reimplanted in sockets with reduced height

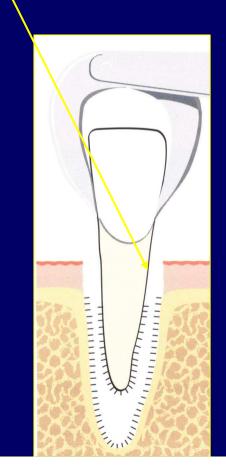


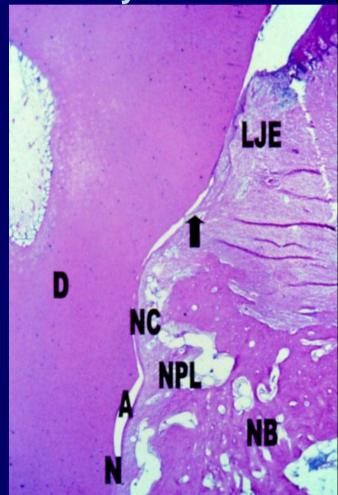
Histology after 6 months

fibrous reunion was established in areas where the original Sharpey fibers were preserved

In areas where the Sharpey fibers were destroyed long epithelial attachment occurred







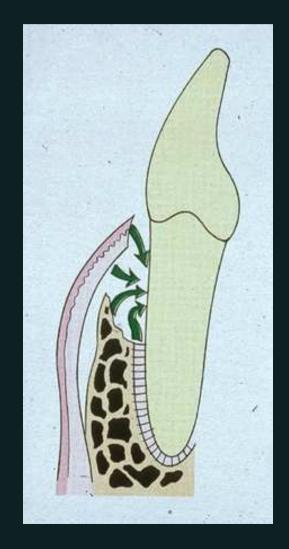


Result from animal experiments by (Melcher 1970, Lindhe 1974) have provided support that cells derived from the periodontal ligament posses an ability to produce new cementum.

> research model was developed (Nyman 1982) dentogingival epithelium granulation tissue, derived from the gingiva,

was inhibited from reaching contact with the root surface during healing.

PERIODONTAL LIGAMENT



15 PERIODONTAL LIGAMENT CELLS

Monkey model 1. U shaped incision was made through the oral mucosa.

2. A mucoperiosteal flap was raised on the coronal side of the incision to expose the buccal alveolar bone.

3. Within the area 3 to 5mm buccal and approximal alveolar bone was resected and the root surface thoroughly planed to remove all cementum.

Gottlow, J., Nyman, S., Karring, T., Lindhe, J. New attachment formation as the result of controlled tissue regeneration. J Clin Periodontol 1984; 11:494-503.



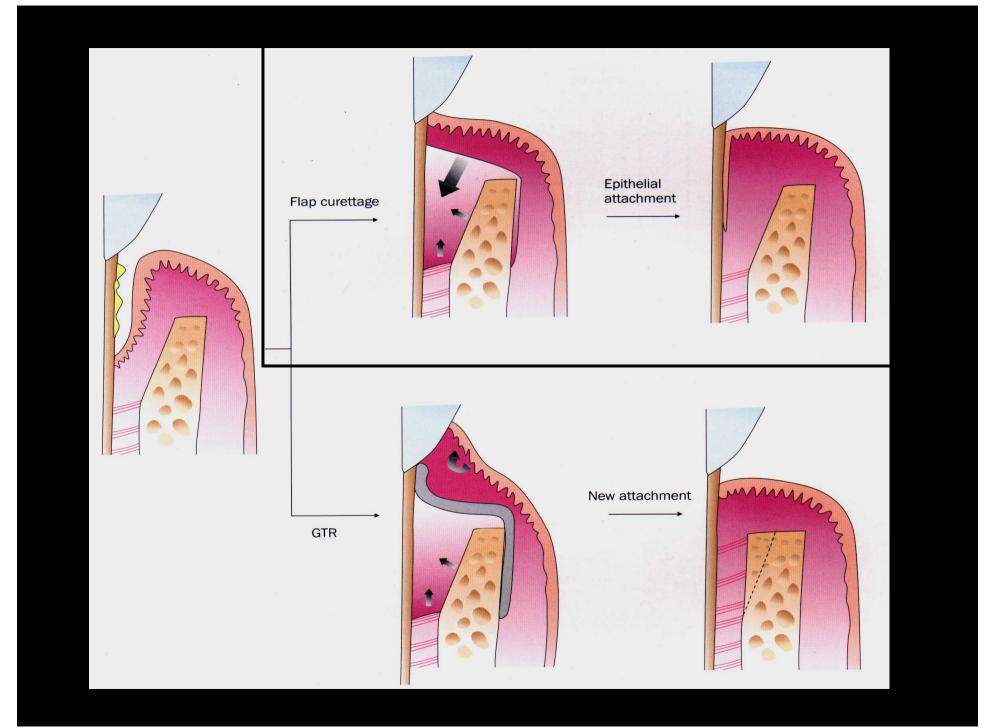
16

4. By preserving the most marginal portion of the periodontium, wound healing was allowed to occur without interference from an apically migrating dentogingival epithelium.

5. In order to prevent the gingival connective tissue from reaching contact with the root surface, a membrane was placed to cover the fenestration in the alveolar bone.

6. The flap was repositioned to the facial surface of the membrane and sutured





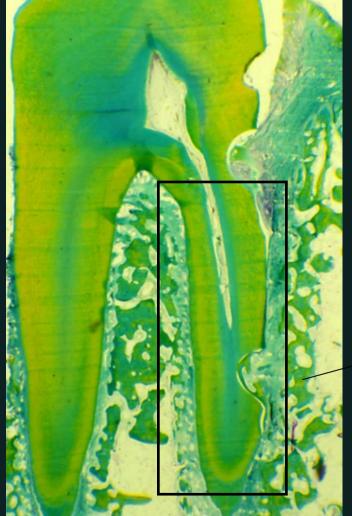
17 The histological analysis of block sections representing 3 mounths of healing disclosed that :

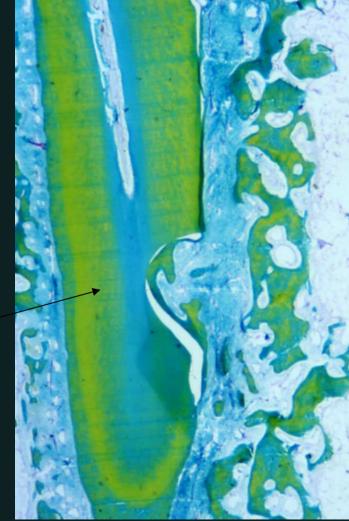
New formation of attachment had occurred, including newly formed cementum with inserting collagen fibers and also supporting alveolar bone.

This finding demonstrates that periodontal ligament cells have a considerable potential for regeneration but only if the epithelium and the gingival connective tissue are prevented from occupying the wound area adjacent to the root.

In the study described, new attachment was obtained on the root surfaces, which were indeed previously deprived of their periodontal ligament and cementum but had not been exposed to plaque or to a periodontal pocket.

REGENERATING ROOT CEMENTUM





In order to test the hypothesis that new cementum with inserting principal fibers also could be formed on a previously diseased root surface, the following experiment was performed (Nyman 1982).

18

A male patient (47 years of age) suffered from advanced periodontal disease of a generalized character.

1. A mandibular incisor with deep pockets of long standing and scheduled for extraction was treated using a flap technique.

2. The attachment level at the buccal surface of this tooth was found to be located 11mm apical to the cemento-enamel junction (CEJ).

3. Following elevation of soft tissue flaps, a notch was prepared in the buccal root surface at the level of the alveolar bone crest the distance between (CEJ) and this notch was 9mm.



Nyman S, Lindhe J, Karring T, Rylander H. New attachment following surgical treatment of human periodontal disease. J Clin Periodontol 1982; 9:290-296.

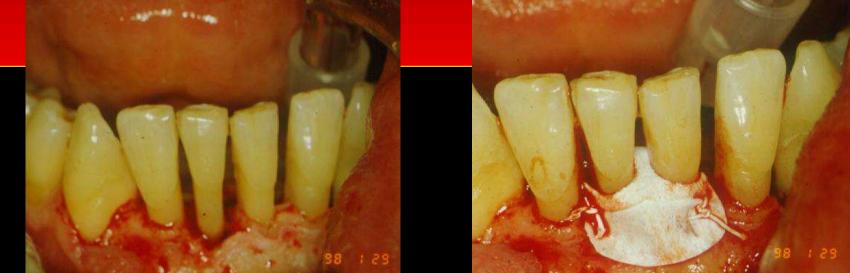
4. An angular bony defect, 2 mm deep was found to be present apical to the notch.

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5. Following removal of all granulation tissue and careful scaling and root planing, a membrane was placed in position

6. The buccal flap was then repositioned to the facial surface of the membrane and secured to the lingual flap by interdental sutures.

The membrane prevented the gingival connecting tissue from contacting the facial and proximal parts of the root. The proliferating oral epithelium was thus during healing guided along the outer surface of the membrane rather than along the root surface.



19/a

6. The buccal flap was then repositioned to the facial surface of the membrane and secured to the lingual flap by interdental sutures.

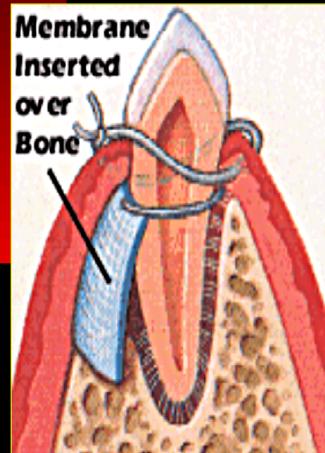
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19/b

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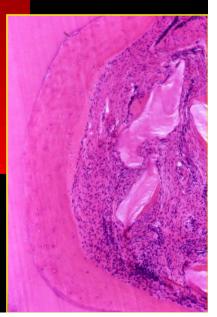


After 3 months:

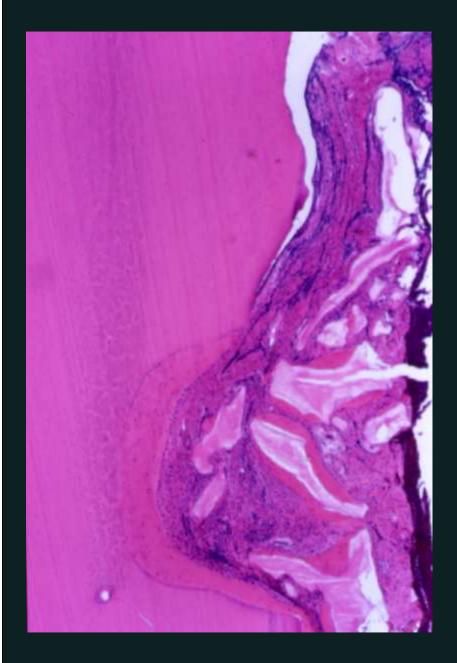
The histological section illustrates that the epithelium had proliferated along the facial surface of the membrane. New cementum with inserting collagen fibers was observed on the facial root surface extending in coronal direction from the apical level of the previous angular bony defect via the notch area to a level 5mm coronal to the apical border of the notch.

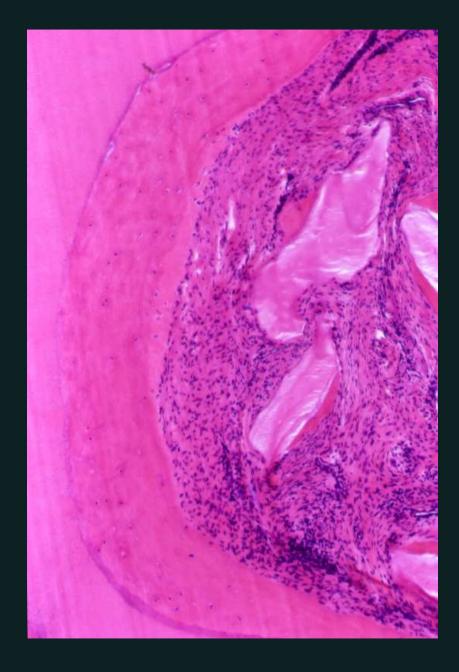
From the finding of this experiment it was concluded that: New attachment can in fact be established on a previously diseased root surface.

Hence, the concept that the periodontitis affected root surface is a major preventive factor for new attachment seems to be invalid.



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A subsequent study (Gottlow 1984) was designed in order to evaluate the predictability of new attachment formation on previously plaque exposed root surfaces using the method of guiding the periodontal ligament cell (PDL) into the wound (Guided Tissue Regeneration).

Gottlow J, Nyman S, Karring T, Lindhe J. New attachment formation as the result of controlled tissue regeneration. J Clin Periodontol 1984; 11:494-503.

Gottlow J, Nyman S, Lindhe J, Karring T, Wennström J. New attachment formation in the human periodontium by guided tissue regeneration. J Clin Periodontol 1986; 13:604-616.

21/a

This study was carried out in the monkey.

1. A soft tissue flap was raised at the buccal aspect of the experimental teeth.

2. 50-70% of the buccal alveolar bone, between the mesio-buccal and disto-buccal line angels of the root, was removed.

3. Before flap repositioning the coronal portion of the flap was excised so that its cut margin coincided with the reduced bone crest. Hereby the coronal portion of the roots was left open to plaque accumulation



4. After 6 months of experimentation, a notch was prepared in the buccal root surface at the level of the gingival margin.

5. A soft tissue flap was again raised at the buccal aspect of the experimental teeth.

6. A root surface coronal to the notch was thoroughly scaled.

22/a

7. The crowns of the teeth were then resected and the flaps sutured to the lingual wound edge to accomplish complete coverage of the root.

8. Before suturing, a teflon membrane was placed over the denuded root surfaces on one side of the jaw (test roots) in order to prevent granulation tissue from the soft tissue flap from reaching contact with the roots.
No membrane was placed over the contralateral roots (control roots).



New cementum was observed on the curetted surface of all test roots and on 5 of 6 control roots.

The newly formed cementum, was always in continuity with the cementum of the apical part of the roots, the newly formed cementum was thicker in the apical than in the coronal portions.

In the control roots the length of the newly formed fibrous attachment varied between 0 and 2.5mm, while in the test roots the range was 0.9 to 6.8mm.



• In 4 of the 9 test roots, new cementum covered the entire length of the root portion coronal to the notch.

• Only 3 of the 9 test roots exhibited a new fibrous attachment, which covered less than 60% of the length of the previously exposed root surface.

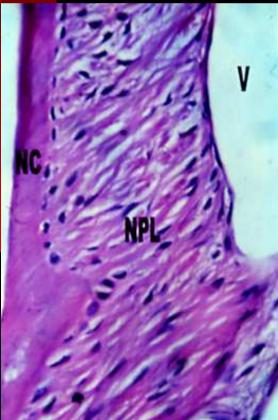
• Only 1 control root exhibited a new attachment which exceeded 60% of corresponding root portion.

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• In all control specimens, the surface coronal to the newly formed cementum was characterized by the presence of multinucleated cells and resorption cavities. However, areas without sign of root resorption were also observed. The root surface in such areas was covered by a non-attached connective tissue with collagen fibers running parallel to the surface.

• Coronal regrowth of alveolar bone had occurred to a varying extent adjacent to7 of the 9 test roots and 2 of the 6 control roots.

No relationship was found between the amount of new cementum formation and the degree of bone regrowth.





The most important information derived from this study:

• Gingival connective tissue lacks the ability to form a new attachment.

• Prevention of the gingival tissues (both the connective tissue and the epithelium) from interfering with healing facilitates new attachment formation by giving preference to the periodontal ligament cells to repopulate the wound area adjacent to the root.

HUMAN CLINICAL STUDIES

Gottlow J, Nyman S, Lindhe J, Karring T, Wennström J. New attachment formation in the human periodontium by guided tissue regeneration. J Clin Periodontol 1986; 13:604-616.

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Clinical studies with non bio-absorbable barrier membranes:

The classic animal studies discussed above prompted the initiation of many clinical trials in the mid 80s and led to the widespread clinical use of different non bioabsorbable barrier membranes.

The objective of the first clinical studies was to assess if new attachment formation would predictably form also in human teeth when treatment is based on the principle of GTR.

GTR model of human teeth (Gottlow 1986):

In this study 12 periodontally involved teeth in 10 patients were used.

1. The comprehensive treatment plan for the patients called for extraction of 5 of the 12 teeth.

• 4 of these teeth were treated with (GTR) technique.
• 1 of the 5 teeth was surgically treated without placement of membrane.

Gottlow J, Nyman S, Lindhe J, Karring T, Wennström J. New attachment formation in the human periodontium by guided tissue regeneration. J Clin Periodontol 1986; 13:604-616.





GTR model of human teeth (Gottlow 1986):

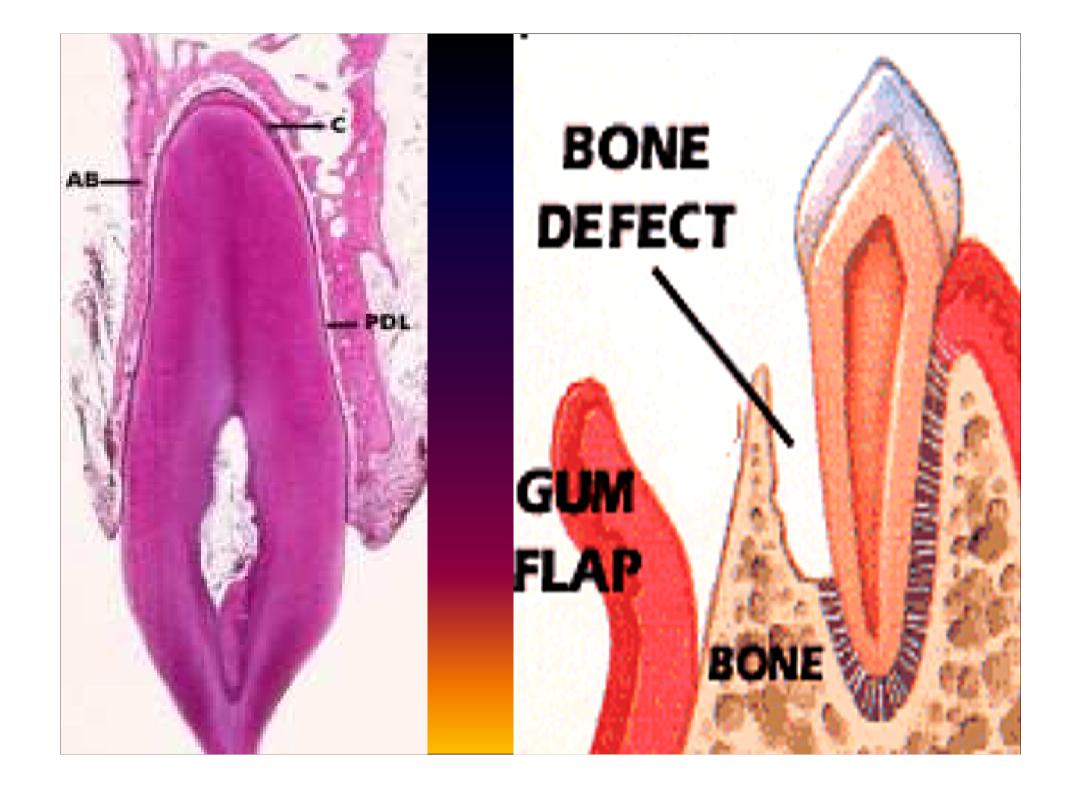
2. The remaining of 7 teeth were treated with the (GTR) method and retained. The result of healing in these teeth was evaluated using clinical parameters such as:

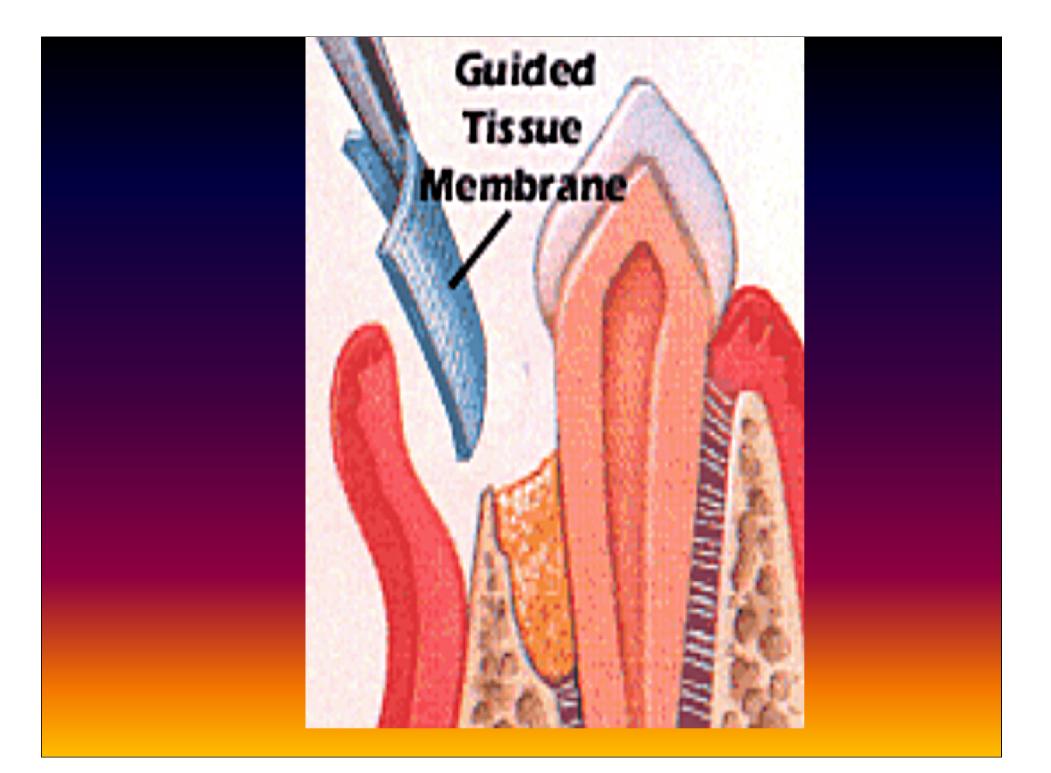
• Probing attachment level (PAL).

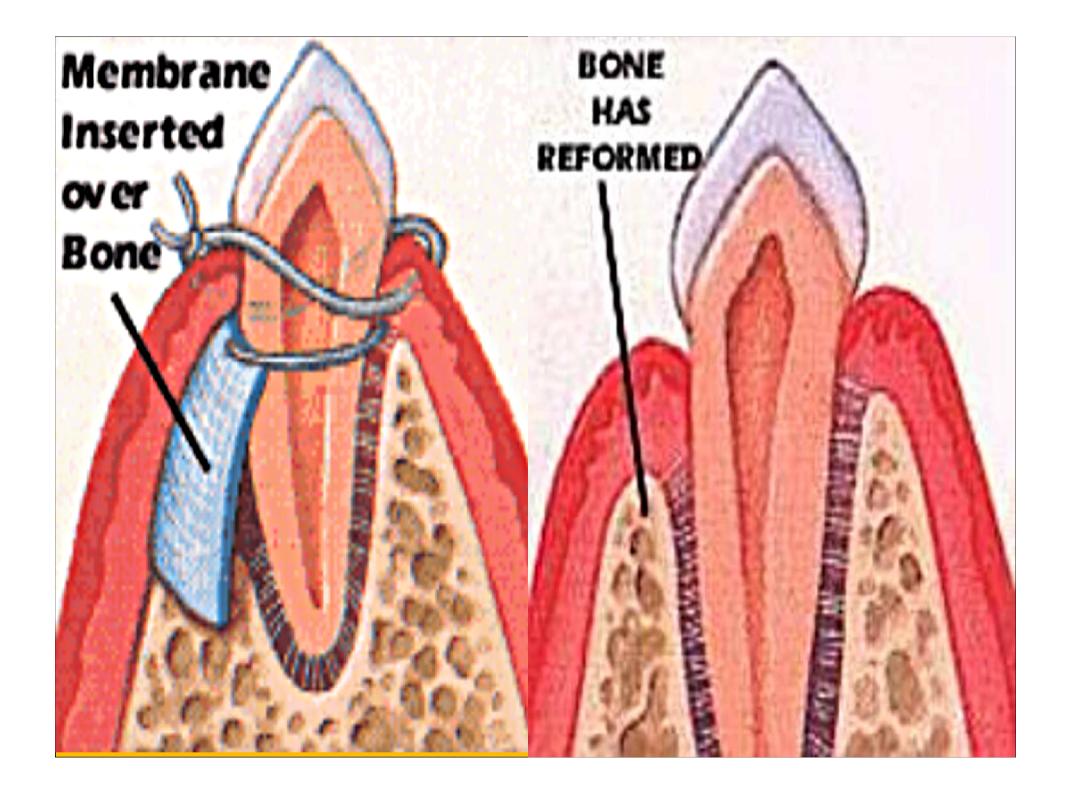
• Probing bone level (PB).

Radiographic analysis.

Clinical inspection during reentry operation.









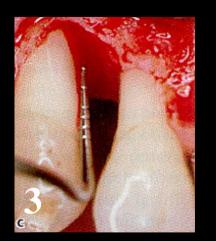
3. Full thickness flaps were raised Epithelium and granulation tissue were removed. The root surfaces were scaled and planed.

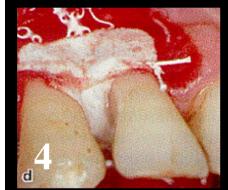
4. In 11 teeth a teflon membrane (Gortex) was adjusted to cover the root surfaces The membrane was retained in this position and the mucoperiosteal flaps were placed on the outer surface of the membrane and secured with interdental sutures

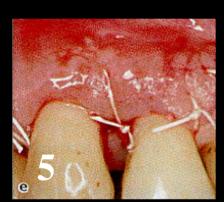
5. After 3 month of healing, the 5 teeth, which had been scheduled for extraction were removed together with surrounding periodontal tissues.



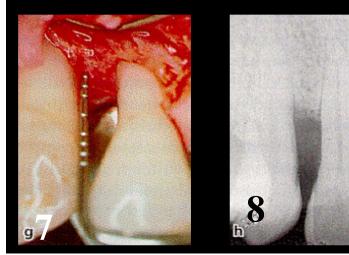






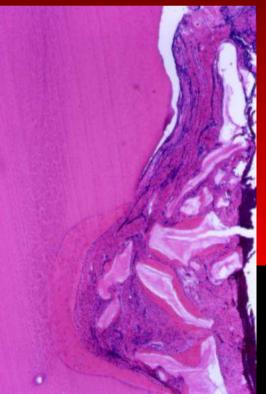




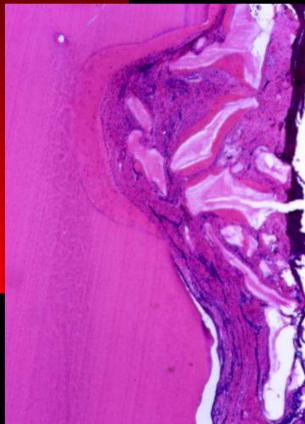


The biopsies were subjected to routine histological preparation:

• The amount of new attachment (the linear distance from the apical extension of the root planed surface to the coronal level of newly formed cementum with inserting collagen fibers) was assessed in 5 sections obtained from the central portion of each experimental root.



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- In the group of 7 teeth which were retained a second surgical procedure was carried out after 3 months of healing for the removal of the membranes.
- The level of the alveolar bone crest and the apical extension of angular defects were determined by probing

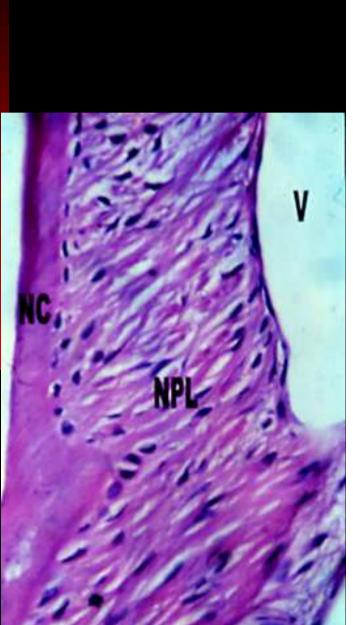
• After another 3 months of healing, a final examination was made including measurements of the probing attachment levels (PAL).

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The result of the present study indicate that:

- Regenerative surgical therapy, based on the principle of GTR. predictably resulted in a new connective tissue attachment.
- The amount of newly formed fibrous attachment was found to vary considerably from one tooth to the other.
- In some teeth, treatment resulted in a complete new attachment, while in other sites only a few mm of new cementum formed.





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The membrane allowed both periodontal ligament tissue and bone tissue to invade the wound area adjacent to the root.

However, no signs or symptoms of ankylosis were found in any of the treated teeth as evaluated histologically in the extracted teeth and by percussion and mobility tests in the retained teeth.

This means that the periodontal ligament tissue had established contact with the root surface before granulation tissue originating from the bone reached the tooth.

This observation indicates that the migration rate of periodontal ligament cell is at least as high as that of bone cells.

Several factors may influence this variability

• The degree of gingival recession that occurs during healing. *The more the gingiva recedes, the shorter the root surface portion that is available for PDL-cells repopulation.*

Several factors may influence this variability

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• The morphology of the periodontal defect may influence the degree of new attachment formation:

In the situation of a "horizontal" type defect circumscribing the entire root, new attachment is exclusively dependent on a coronal upgrowth of PDL-tissue from remaining periodontium.

while in angular defects and furcation involvement of partial character, PDL-cells may migrate into the wound area not only from the apical part of the defect but also from its lateral borders.

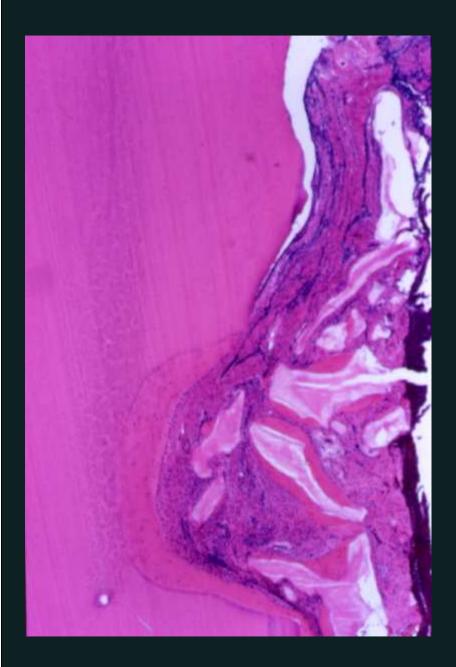
This may enhance the potential of new attachment formation.

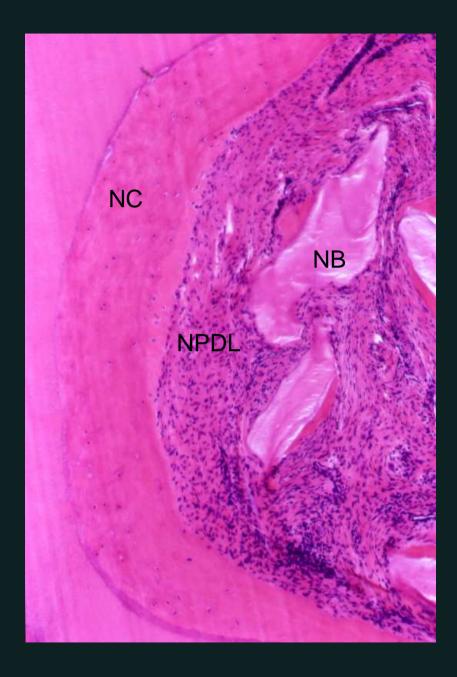
33 GUIDED TISSUE REGENERATION

GTR was developed based on a series of studies on periodontal wound healing performed in Scandinavia during the early 1980s by Nyman and coworkers.

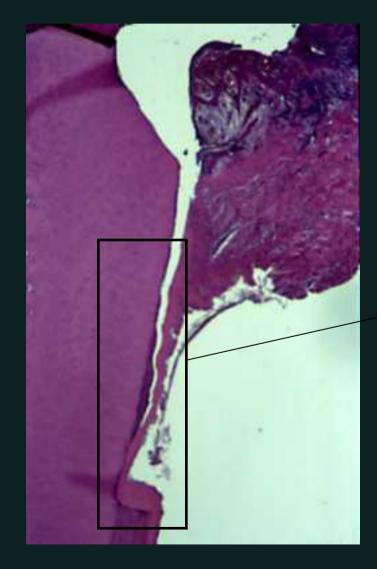
The results of these studies, strongly suggested that the exclusion of epithelial and gingival connective tissue cells from the healing area by placing a physical barrier, between the periodontal defect and the gingival flap before suturing, may allow (guide) PDL cells to repopulate the detached root surface.

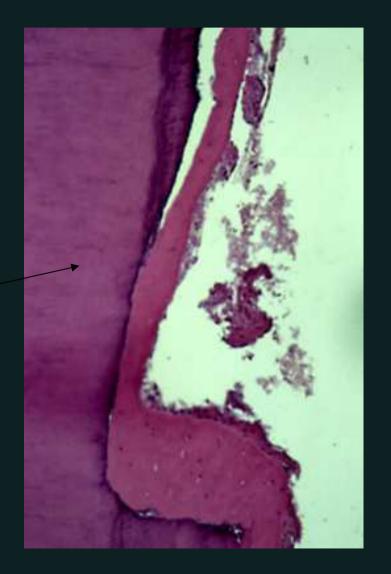
Their finding in animals were also histologically confirmed in humans.





NEW ATTACHMENT

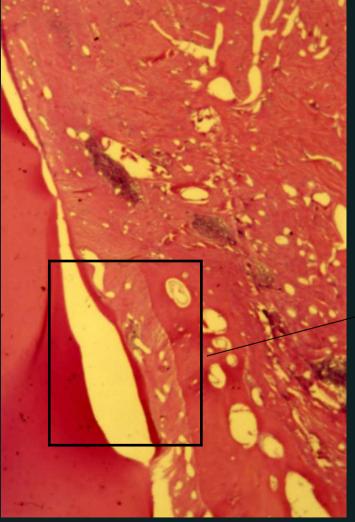


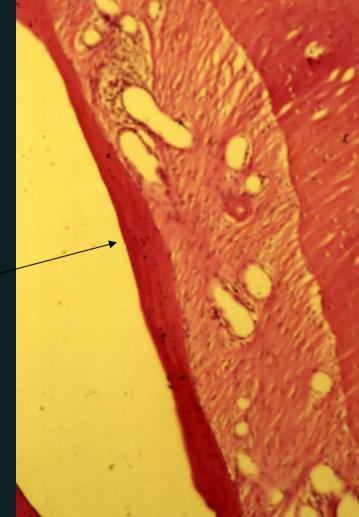


Summary

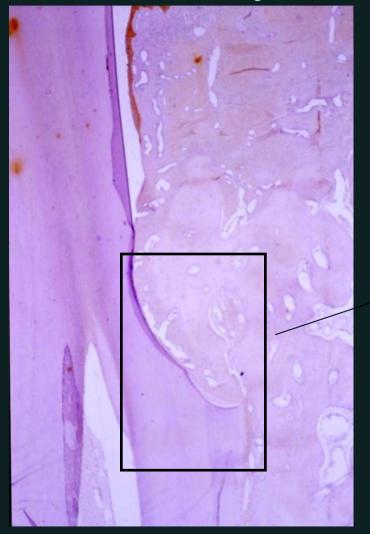
- The conventional non-surgical and surgical periodontal therapy result no real periodontal regeneration
- The periodontal regeneration is a complex phenomena followed by the reformation of new cementum, PDL and alveolar bone
- The condition of the remaining PDL and root cementum play critical role in the periodontal regenerative process

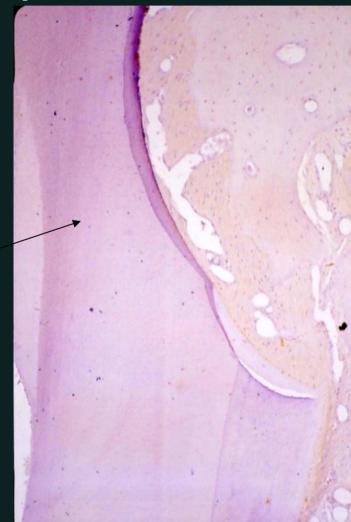
Regenereting Periodontal Ligements



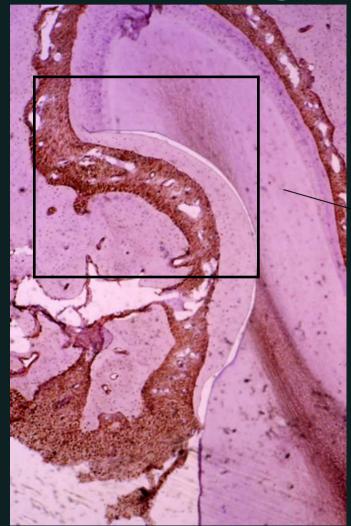


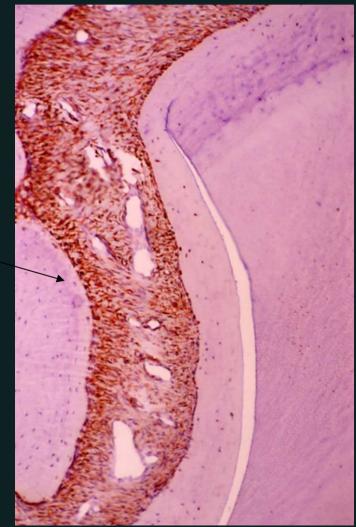
No cytokeratin expression



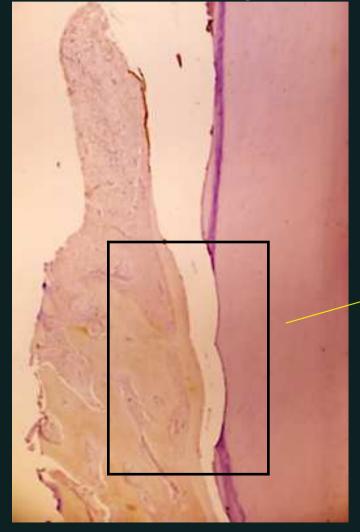


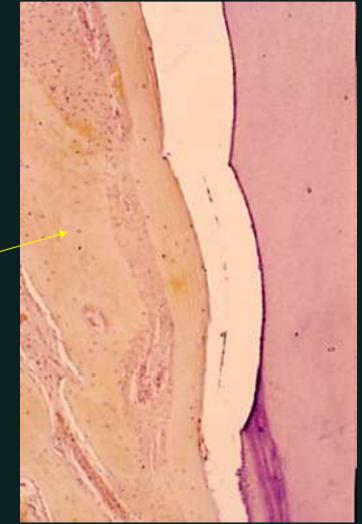
Strong Vimentin expression





No Cytokeratin expression





Strong Vimentin expression

