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The Prevalence Morphology and Distribution of Epithelial Rests in the Periodontal Ligament of Humans

Charles McCormick Reeve

Loyola University Chicago

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THE PREVALENCE MORPHOLOGY AND DISTRIBUTION OF
EPITHELIAL RESTS IN THE PERIODONTAL
LIGAMENT OF HUMANS

by

Charles McCormick Reeve

A Thesis Submitted to the Faculty of the Graduate School
of Loyola University in Partial Fulfillment of
the Requirements for the Degree of
Master of Science

June
1960
LIFE

Charles M. Reeve was born in Elbow Lake, Minnesota, September 6, 1933.

He was graduated from Elbow Lake High School, Elbow Lake, Minnesota, June, 1951. From September, 1951, to June, 1954, he attended the University of Minnesota.

In September, 1954, he began his studies at Loyola University School of Dentistry and received the degree of Doctor of Dental Surgery in June, 1958. His graduate studies began in the Department of Oral Anatomy of Loyola University in June, 1958. He was awarded a Research Fellowship in the Department of Periodontics in the School of Dentistry in December, 1958. In May, 1959, he was appointed a Research Fellow by the National Institute of Dental Research, United States Public Health Service.
ACKNOWLEDGEMENTS

To Doctor Frank M. Wentz, whose ideals will always be inspiring, I wish to acknowledge his untiring efforts and guidance in this investigation.

I sincerely thank Doctor Harry Sicher for his many suggestions and constructive criticisms.

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To the Foundation for Dental Research of Loyola University I am indebted for the use of the human autopsy specimens which are part of the collection of the late Rudolph Kronfeld.

To my wife, Louise, I am grateful for her many sacrifices which made this work possible.
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CHAPTER I

INTRODUCTION

This study was an attempt to provide evidence on the fate of the epithelial rests in the periodontal ligament of humans between the ages of one and seventy-seven years. Specific objectives included classification of the different types of epithelial rests, and investigation of the prevalence and distribution of these structures in the periodontal ligament.

Wentz, Weinmann, and Schour conducted a study on the prevalence, distribution, and morphologic changes in the epithelial rests in the molar region of the rat. By applying their established method to the study of epithelial rests in human teeth, the biology, and age changes of this persistent embryonal debris may be revealed. Some lead may be afforded in this study to better understand a probable role of the epithelial rests in periodontal disease.
CHAPTER II

REVIEW OF THE LITERATURE

A. Morphology of the Epithelial Rests

Robin and Magitot (1860) seem to have been the first to describe "epithelial debris" around a developing tooth.

Malassez (1885) was the first to describe epithelial rests in the periodontal tissues.

Black (1886) described a very peculiar grouping of cells in the periodontal ligament which he believed were part of a lymphatic system. He failed to identify these structures as epithelial rests. His study of the morphology, location, and incidence of these elements however, was the most exacting at the time. Black later (1899) identified the lymphatic like structures as glands and attempted to trace their ducts. However, he failed in this. The lymphatics of Black, were without doubt, the epithelial rests, Malassez had described a year before.

It was the work of v. Brunn (1887) that finally clarified derivation of the epithelial rests. v. Brunn stressed the importance of the epithelial sheath in the development of the tooth as the determining factor in the shaping of the root. He
described the epithelial sheath as being perforated by connective tissue of the primitive periodontal ligament giving rise to the epithelial rests.

Talbot (1898) described the epithelial cells in the periodontal ligament as being arranged in single rows, loops, double rows, and sometimes in round or oblong groups without characteristic shape or form.

B. Location of the Epithelial Rests

Black (1886) found epithelial rests in young subjects in great profusion between connective tissue fibers of the periodontal ligament. The structures according to Black, were always located close to, but not in contact with the cementum. He described a strand formation which anastomosed with other strands to form a network. This network completely surrounded the surface of the tooth root.

Talbot (1899) described the epithelial rests as arranged along the surface of the cementum, and that they were more numerous near the gingival border than in the apical area. In 1921 Talbot seems to have abandoned any ideas as to any function of the epithelial rests and closed his work with the statement "the epithelial rests are a useless scaffolding remaining in the body".

Orban (1932) found epithelial rests within the bony canals containing the dental arteries, veins, and nerves; in the pulp;
and in the interdental alveolar septum.

Meyer (1932) found that epithelial rests show in younger individuals a net like arrangement. Meyer also observed that the epithelial rests are located within the bundles of the principle fibers and not in the spaces between the bundles.

Fischer (1932) observed a net like arrangement rarely in human teeth, but regularly in the dentition of herbivores. He stressed the frequency of epithelial rests at the apex, the bifurcation, and the cervical region of the tooth.

Aisenberg (1943) found epithelium in the dental pulp, which entered through the apical foramen from activated epithelial rests.

C. Prevalence of the Epithelial Rests

Black (1886) counted one hundred to two hundred cell groups in a cross section of an incisor. Apparently this tooth was procured from a young individual because in his study of a man forty years of age, the number diminished. When he examined the periodontal ligament of a man seventy, only a few groups of the cells were found. From these data Black believed it probable that the peculiar cell groups he described diminish with advancing age.

Kronfeld (1937) in his description of the epithelial rests in the periodontal ligament states that the rests are particularly abundant in young human teeth, and that they may form a
network surrounding the entire root. In the periodontal liga-
ment of older teeth they seem to be scarce and appear as indi-
vidual rounded groups of epithelial cells. The number of the
rests is subject to individual variation, but no specimen of a
human tooth has been found in which epithelial rests were entire-
ly absent.

D. Epithelial Rests in Dental Pathology

McConnell (1928) was of the opinion that the epithelial
cells present in granulomas are derived from the embryonal rests
of the outer layer of the enamel organ, which are so frequently
found in the periodontal ligament.

Lartschneider (1929) claimed that the epithelial rests
originating from the dental lamina give rise to multiloculated
cysts and enamel-forming tumors, whereas the epithelial rests
originating from the epithelial root sheath might develop into
simple cysts.

Hill (1930) in a study of granulomas found epithelial tis-
sue in every case of the forty-two granulomas he studied. He
demonstrated the morphologic similarities between the prolifera-
ted epithelium of dental granulomas and the epithelium from
the embryonal enamel organ, and that actual transition from
enamel-organ rests to proliferated epithelium in dental granu-
Iomas occurred.

Hill later (1949) discusses the importance of the prolifer-
ation of epithelium in the development of granulomas. The early proliferation of connective tissue is accompanied by an increase in the number of cells of the epithelial rests of Malassez. As the granuloma develops in size these epithelial rests develop into large islands.

Orban and Weinmann (1942) found enlarged epithelial rests in a jaw with periodontosis.

Stafne (1947) in his description of myxomatous tumors associated with an unerupted tooth stated that within the connective tissue around an unerupted tooth there were scattered narrow cords of epithelial cells which were sometimes branched. This epithelium resembled the epithelium of the dental lamina or the epithelial debris of Malassez.

Aisenberg (1952) is of the opinion that after trauma and infection the epithelial rests can proliferate and change from simple epithelium into cysts and tumors.

Boyle (1955) described the transition from epithelial rests to proliferated epithelium in granulomas and cysts.

E. Possible Function of the Epithelial Rests

In 1922 Gottlieb stated that excementosis and cementicles of the periodontal membrane are probably the result of calcification of disintegrated epithelial rests in the periodontal ligament.

Robinson (1925) suggested that the epithelial rests have
the function of a ductless or endocrine gland. He was not able to prove this hypothesis.

Orban (1928) indicated epithelial rests may have some influence on the deposition of cementum.

Noyes, Schour, and Noyes (1938) in their textbook state that the epithelial rests may degenerate, become calcified and give rise to cementicles, or, in inflammatory processes may lead to root cyst formation. They state epithelial rests are more numerous in young people, but persist throughout life. In one instance they demonstrated epithelial rests in the periodontal ligament of a man seventy years old. They described the epithelial rests as composed of cords or rows of epithelial cells surrounded by an extremely thin delicate basement membrane. In some cases there was a slight indication of a capsule arrangement of connective tissue around them. The authors do not ascribe any particular function to the epithelial rests, but regard it as unlikely that embryonal debris would persist throughout life if it served no function.
CHAPTER III

MATERIAL AND METHODS

The material for this study consisted of thirty-one human jaws obtained at autopsy from individuals ranging in age from one to seventy-seven years, who had died of various diseases: scarlet fever, endocarditis, enteritis, pneumonia, tuberculosis, cancer and coronary thrombosis.

One thousand forty sections, of two hundred eighty permanent teeth cut in mesio-distal, labio-lingual, and transverse directions, and stained with hematoxylin and eosin were subjected to examination. All the sections exhibited an infiltration of inflammatory cells in the gingiva. Signs of resorption of the alveolar crest were evident in many of the specimens after the second decade. However, teeth that presented evidence of gross periodontal disease were not included in the study. The degree of inflammation, the age changes (loss of bony support, gingival recession, apposition of cementum) were noted.

The periodontal ligament was divided into three zones for study (Plate I). Zone I included the area of the dento-gingival junction. Zone II included the middle portion of the periodontal ligament. Zone III included the apical portion of the peri-
dental ligament. The adjacent bone marrow spaces and dental pulp were also examined.

A methodical scanning search for the epithelial rests in each zone was accomplished. Utilizing the serial sections a three dimensional morphologic sketch was made when possible. The location of the rests and a rest count of each zone was recorded for comparative analysis with other specimens.
CHAPTER IV

FINDINGS

A. Morphology of the Epithelial Rests

Three different morphologic types of epithelial rests could be distinguished: (1) resting type (2) degenerated type (3) proliferated type.

Resting type. The majority of this type of epithelial rest was observed close to the cementum. (Plate II) Occasionally the epithelial structures were located in the bone marrow spaces of the alveolar septum and beyond the apex of the teeth. (Plate III) A small and large variation of this type was found.

Tangential sections of the teeth and adjacent tissues revealed the small resting type of epithelial rests to be in a strand arrangement. The strand arrangement was observed frequently in individuals in the first and second decades. In several instances the strands communicated with one another forming a net like arrangement. (Plate IV)

In cross section the cells of the strands appeared in oval groups which consisted of an average of 10 polyhedral cells. (Plate V) The cells were uniform in both size and staining. The connective tissue adjacent to this type of epithelial rest
did not form a capsule.

The large resting type was found in ovoid or spherical groups composed of an average of 28 cells. These groups are isolated and do not form strands. (Plate VI) The nuclei of the cells in the large resting type were slightly larger than those of the small type, and the chromatin material of the nuclei was not as deeply stained. The cells were not as closely arranged as those of the small resting type. In contrast to the small resting type the connective tissue fibers and cells surrounding this type appeared to have a loose concentric arrangement.

An occasional variation of the large resting type was observed in which the cells were arranged in a duct like manner. The center of the epithelial rest contained an amorphous mass of eosinophilic staining material, with the periphery of the rest surrounded by a thin, delicate "basement" membrane. (Plate VII)

Degenerated type. This type of epithelial rest was observed most frequently in teeth of young individuals. The majority of the degenerated forms were located in the apical and middle zones of the periodontal ligament, and in close proximity to the cementum. (Plate VIII) In cross section these rests appeared oval in shape, with an average of 10 cells in each rest. The individual cells exhibited changes characterized by an ap-
parent hydropic degeneration. The nuclei appeared dark and pyknotic. The cell groups had distinct boundaries and a definite shape.

In several specimens obtained from individuals in the later decades the beginning of calcification of these islands could be observed. Epithelial cells undergoing calcification were observed, with an eventual calcification of the entire epithelial rest. In other instances fragments of cells were visible within the calcified epithelial rests. The epithelial rests now appeared to be similar to that of cementicles. (Plate IX)

The connective tissue adjacent to either form of degenerating epithelial rest showed no visible signs of change.

**Proliferated type.** Rests of this type were observed in the later decades, located close to the cementum. This type was the largest and averaged eight to ten times the size of the small resting type.

The nuclei of the cells of the proliferated rests were pale, larger, and more vesicular than those of the resting type. Chromatin granules could be distinguished. The cells adjacent to the connective tissue resembled the cells of a basal epithelial cell layer in both arrangement and structure. Some of the more centrally located cells were not similar. Some were loosely arranged and others were more densely packed. The depth of staining varied. The nuclei were either deeply stained with
hematoxylin, or were more pale. The cytoplasm varied in quantity, staining reaction, and shape. In some areas the cytoplasm of the cells appeared to possess granules which stained more deeply. In other areas, the cells were arranged in a form resembling the stellate reticulum of the enamel organ.

The reaction of the connective tissue surrounding the proliferated epithelial rests varied. The rests were often found surrounded by a well differentiated fibrous capsule, with a concentric arrangement of cells and fibers. (Plate X) In other instances the capsule changed into a hyaline mass in which fibers and cells could not be distinguished. (Plate XI) Occasionally defects were observed in the cementum containing these islands of proliferated epithelium. (Plate XII)

B. Distribution of the Different Types of Epithelial Rests

Epithelial rests were found around all examined teeth. The distribution of the different types depended partly upon the age of the individuals. The resting type of epithelial rest was observed most frequently, and was present in all specimens. The degenerated type was observed primarily in individuals in the first and second decades. The proliferated type was seen in the later decades -- fifty years and older.

The incidence of the different types of epithelial rests varied with the age of the individuals. The small resting type
and degenerated type decreased with age, while the large resting type and proliferated type increased with age.

When comparing the total epithelial rest count of the different age groups it was found that the number of epithelial rests decreases with an increase in age.

C. Distribution of Epithelial Rests in the Different Zones

The distribution of epithelial rests varied with the age of the individuals. To determine this distribution the specimens were divided into eight age groups with each group representing a decade. The number of rests in each age group was expressed as a percentage of the total rest count. (Table I)

The rests were most prevalent in Zone III, the apical zone, during the first and second decades. When the third to seventh decades were considered, Zone I composing the cervical one-third of the teeth, contained 47% of all rests. Zone II composing the middle one-third of the teeth, contained 31% of all rests. Zone III composing the apical one-third of the teeth, contained 22% of all rests.

From the foregoing data the age distribution variation of the epithelial rests in relation to the teeth is expressed as follows: In the young individuals there is a greater apical distribution, while in the twenty to seventy-seven group there is a shift in the distribution -- now the greatest number of
epithelial rests are located near the cervical area of the teeth.

D. Topography of the Epithelial Rests

The epithelial rests were located close to the cementum within the principle fibers of the periodontal ligament. The rests were observed occasionally in the fibrous marrow spaces distally to the teeth, and beyond the apex.
<table>
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<th>Zone II</th>
<th>Zone III</th>
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<tr>
<td>1-10</td>
<td>17%</td>
<td>36%</td>
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<td>11-20</td>
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<td>22%</td>
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<td>26%</td>
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<tr>
<td>71-80</td>
<td>51%</td>
<td>35%</td>
<td>14%</td>
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CHAPTER V

DISCUSSION

A. Incidence and Prevalence of the Morphologic Types

The findings of this study were consistent with those of Black (1886) and Kronfeld (1937) who found epithelial rests to be more abundant in young human teeth, than old. The greatest frequency of occurrence was in the first and second decades. After this the total number of epithelial rests observed diminished until in the seventh decade the incidence was approximately only one-fourth the number that had been observed in the first twenty years of life.

The descriptions of the epithelial rests by the early investigators were fairly exact. They described epithelial net formations in loops and rows, located in the connective tissue bundles near the cementum. The question of the function of the epithelial and why they persist was unanswered. Though persistence of the rests over seven decades was observed, an accurate description of morphologic age changes was lacking.

The findings of this study indicate that the morphologic structure of these epithelial structures varies with the age of the individual. The small resting type is found in all age
groups. The large resting type is found to a greater extent in the fourth and later decades. The transformation of some of the small resting types into the larger form by proliferation, may account for their occurrence in the later decades.

The epithelial rests undergoing hydropic degeneration were found primarily in teeth just completing root formation. The transformation of epithelial rests into cementicles occurs in the later years, with the epithelial rests acting as a nidus for beginning calcification.

The proliferated rests were in close contact with the cementum, and in some instances were contained within depressions of the cementum. From this one could assume that these proliferated rests were responsible for some disturbance in cementoblastic activity. If proliferated epithelial rests are located close to the epithelial attachment, there is the possibility that they may fuse with the epithelial attachment as it shifts apically, and contribute to its width. In degenerative periodontal disease the fusion of epithelial rests with the down-growing of the epithelial attachment has been thought to be responsible for the sudden appearance of deep periodontal pockets (Orban 1942).

The reduction of the epithelial rests in number with age occurs by degeneration with consequent disappearance, or by degeneration followed by calcification and the formation of cemen-
articles. Some of the epithelial rests persist through seven decades of life. During this period they may even proliferate.

B. Location of the Epithelial Rests

Early descriptions of the location of the epithelial rests show them to be located close to the cementum within the bundles of the principal fibers of the periodontal ligament. (Black 1886, Meyer 1932). The findings of this study are in agreement with these earlier investigators. The fact that the epithelial rests are located within the cemental group of periodontal fibers, and not within the alveolar group, or in the mid-portion of the periodontal ligament may lead one to assume that the presence of an intermediate plexus may only allow some of the epithelial rests to escape to other locations, i.e., bone marrow distally to the tooth, or the pulp chamber.

The distribution of the epithelial rests in relation to the tooth varies with the age of the individual. In young individuals the epithelial rests are more numerous in the apical one-third of the tooth, but in the later decades this distribution is reversed, with the epithelial rests occurring in greater numbers in the cervical one-third of the tooth. A possible explanation for this may be that a significant fact was overlooked by the earlier investigators. This fact was the presence of a persistent inflammatory reaction in the gingiva. The influence of this inflammatory reaction upon the epithelial rests was not
considered by these earlier investigators. The proliferative nature of the epithelial lining of the gingival sulcus in the presence of inflammation is well known. The constant chronic inflammation around the gingival sulcus may also cause proliferation or at least promote a persistence of the epithelial rests in this area of the tooth.

C. Epithelial Rests in Dental Pathology

Proof of the effects of inflammation upon epithelial rests located in the periodontal ligament is the proliferation of the epithelium in periapical granulomas. Several investigators (Kronfeld 1928, McConnell 1928, Hill 1930) have shown epithelium to be present in periapical granulomas that is derived from epithelial rests found within the periodontal ligament. If this epithelium proliferates in the presence of periapical inflammation, it is logical to assume that the same phenomenon may occur elsewhere within the supporting tissues of the teeth. The proliferative nature of epithelium in the presence of inflammation and the predominance of these epithelial rests in the cervical area of the tooth may be significant in certain phases of periodontal disease.
CHAPTER VI

SUMMARY AND CONCLUSIONS

The purpose of this study was to examine the epithelial rests between the ages of one and seventy-seven years; to classify the different types of epithelial rests; to investigate the prevalence and distribution of these structures in the periodontal ligament of humans, and to consider their possible role in dental pathology.

Conclusions:

1. Epithelial rests were found in all specimens regardless of age.

2. The incidence of the epithelial rests decreased with increasing age.

3. Three types of epithelial rests were observed:
   a) A resting type, of varying size. The small resting type occurred in all age groups. The large resting type appeared in the fourth and later decades.
   b) A degenerated type was found in all age groups, but was most frequent in the first and second decades.
   c) A proliferated type which appeared in the fifth and later decades.
4. Some forms of the epithelial rests persisted throughout life; some proliferated; others degenerated and disappeared; still others calcified and persisted as cementicles.

5. The majority of the epithelial rests were located in the cervical area of the teeth in all ages except during the first and second decades. In the younger individuals the greatest number was found in the apical area.

6. The greater persistence of the epithelial rests in the cervical area may be correlated to and influenced by the constant inflammatory reaction present in the area of the gingival sulcus.

7. One can assume that the epithelial rests are vestigial structures persisting within the periodontal ligament, with a potential role in dental and periodontal pathology.
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PLATE I

Diagrammatic illustration of the tooth and supporting tissues divided into three zones.
APPENDIX

PLATE I

ZONE 1
ZONE 2
ZONE 3

DENTOGINGIVAL JUNCTION
Figure 1. Photomicrograph X100 of small resting type of epithelial rests in strand formation (A). Note the close proximity of the epithelial rests to the cementum.

Figure 2. Photomicrograph X400 of small resting type of epithelial rest. In this high power of Figure 1 the broken strand arrangement of the epithelial rests (A) is separated from the cementum by the cementoblastic layer and connective tissue cells (B).
PLATE III

Photomicrograph X400 of epithelial rests in alveolar bone marrow. The epithelial rests are located beneath the apex of the tooth.
PLATE IV

Figures 1 and 2. Photomicrograph X100 of strand arrangement of epithelial rests.
Figure 1. Photomicrograph X100 showing spherical groups of epithelial rests (A).

Figure 2. Photomicrograph X400 of Figure 1. Note the proximity of the epithelial rests (A) to the cementoblastic layer and cementum (B).
PLATE VI

Figure 1. Photomicrograph X100 of large resting type of epithelial rest.

Figure 2. Photomicrograph X400 (high power Figure 1) of large resting type of epithelial rest. Note the arrangement of the fibroblasts adjacent to the rest.
PLATE VII

Photomicrograph X600 of an epithelial rest appearing as a duct like structure. Note the peripheral arrangement of the cells (A). The presence of plasma cells and lymphocytes is indicative of chronic inflammation (B).
PLATE VIII

Figure 1. Photomicrograph X400 of degenerating epithelial rests (A). Note pyknotic nuclei and apparent hydropic degeneration.

Figure 2. Photomicrograph X400 of epithelial strand showing beginning degeneration.
PLATE IX

Photomicrograph X100 of cementicle (A) and strand formation of epithelium (B).
Figure 1. Photomicrograph X100 of proliferated epithelial rest (A). Note the inflammatory cells (B) present in the periodontal ligament.

Figure 2. Photomicrograph X400 of proliferated epithelial rest (A in Figure 1). Note the concentric arrangement of cells and fibers surrounding the proliferated mass of epithelium.
PLATE XI

Figure 1. Photomicrograph X100 of proliferated epithelial rest surrounded by a hyaline like capsule.

Figure 2. High power photomicrograph of Figure 1. X400 Note the clear hyaline capsule (A) in which fibers and cells are absent.
PLATE XII

Figure 1. Photomicrograph X100 of proliferated epithelial rest in cemental defect.

Figure 2. High power photomicrograph of Figure 1. X400
Plate XII

Age in Years

1-10
11-20
21-30
31-40
41-50
51-60
61-70
71-80

Totals

Figure 1

Figure 2
### TABLE A

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| 61-70 Years     |        |        |        |       |
| Jaw # 27        | 246    | 148    | 42     | 436   |
| Jaw # 28        | 107    | 58     | 104    | 269   |
| Jaw # 29        | 134    | 61     | 33     | 228   |
| Total           | 487    | 267    | 179    | 933   |

| 71-80 Years     |        |        |        |       |
| Jaw # 30        | 176    | 122    | 45     | 343   |
| Jaw # 31        | 65     | 46     | 19     | 130   |
| Total           | 241    | 168    | 64     | 473   |
APPROVAL SHEET

The thesis submitted by Charles M. Reeve has been read and approved by three members of the faculty of the Graduate School.

The final copies have been examined by the director of the thesis and the signature which appears below verifies the fact that any necessary changes have been incorporated, and that the thesis is now given final approval with reference to content, form and mechanical accuracy.

The thesis is therefore accepted in partial fulfillment of the requirements for the Degree of Master of Science.

Date May 24, 1960

Signature of Adviser

Frank M. Wentz, D.D.S., M.S., PH.D.