# TISSUE CHANGES INCIDENT TO TWENTY-ONE DAYS OF TOOTH MOVEMENT

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# INTRODUCTION

Ever since Sandstedt[1] first reported his histologic findings on a dog in 1901, the complex biologic mechanisms involved in the phenomenon of tooth movement have remained somewhat obscured. In 1911, Oppenheim[2] published results of his tooth movement study in baboons which differed slightly from Sandstedt's findings. It was not until the late 1920's and early 1930's that a number of authors, notably Stallard,[3] Schwarz,[4] and Gottlieb[5] contributed markedly to Oppenheim's work. One of the first histologic studies done on human tooth movement was reported by Herzberg in 1932.[6] Oppenheim[7,8] and Stuteville[9,10] also did some significant work on human studies. Recently (1951 to present) Reitan[11,12,13] has done the majority of histologic studies involving human tooth movement.

The experimental model for the following investigation was similar to that utilized by Church (1971).[15] This model was based on Reitan's[11] work in which a 70 gram continuous tipping force was placed on a human tooth using a fixed appliance, similar to that used by Stuteville.[9] Church reported morphologic evidence of fat-like cells or "signet ring cells" within the periodontal ligament space after 21 days of tooth movement. He described the phenomenon as similar to "fatty degeneration." Due to this unforeseen result, he was not able to confirm the actual presence of lipid within the "signet ring cells," since his specimens were subjected to laboratory procedure which obliterated all existing fat. Therefore it was difficult to use a specific lipid stain, which best utilizes a frozen section technique.

This was not the first time this histologic pattern was observed.

Oppenheim[7] observed an abundance of large vessels and capillaries in the periodontal membrane that suggested an "angioma-like" appearance in some of his specimens of human tooth movement. Stuteville[10] and Reitan[11] also observed this result in some of their specimens but declared them artifacts. Kronfeld[16] reported a similar situation in human necropsy material. However, to our knowledge, this was the first time that the term "fatty degeneration" had been used to describe this histologic pattern in a tooth movement study.

The purpose of this investigation was to repeat the experiment done by Church[14] at the 21 day time interval and to ascertain the presence of fat or fibrin within these vacuoles or spaces, if they existed, by using a more appropriate specific histologic staining technique. The presence of fibrin would suggest the morphologic evidence offered by Church to have been in error and implicated the presence of blood vascular channels. Lastly, the possibility of these spaces to have been unknown artifacts would be investigated.

## REVIEW OF THE LITERATURE

In 1901, Sandstedt published the first histologic investigation dealing with the orthodontic movement of teeth in his book, Nagra bidrag till tandregleringens teori.[1] In his experiments on dogs, he exerted an undetermined lingual tipping force on the upper incisors by means of a labial wire attached to jackscrews on the canine teeth. After a histologic examination of the specimen, he found deposition of bone on the side of pull with both "weak and strong" forces. The newly formed bone spicules followed the direction of the strained periodontal fibers. On the side of a weak force pressure, there was an even alveolar bone resorption. However, when strong forces were placed on the tooth, the PDL was compressed on the side of pressure and there was no alveolar bone resorption. He claimed that the PDL had lost its vitality due to interruption of its normal blood supply. In order to allow tooth movement, this "necrotic" material had to be removed by the active resorption from the underlying marrow spaces which he called "undermining resorption."

Oppenheim published the second major paper on the histology of tooth movement in 1911.[2] Using an expansion arch first designed by Angle, he initiated a tipping movement on the deciduous teeth of baboons. He reported that the architecture of the alveolar bone on the pressure side was "transformed" to spongy bone, when a "weak" force was applied to the tooth. The bony spicules were orientated vertically to the long axis of the tooth and parallel to the fibers of the PDL. When an "intense" force was placed on the tooth, the pressure side of the alveolar bone was

unaltered. He attributed this to the injury of the cells caused by the severe compression of the PDL. On the side of pull, he found this transformation of the bony architecture as well as active bone formation on the surfaces of the bony spicules closest to the tooth. Later, Stallard[3] offered a critique of Oppenheim's work. He refuted Oppenheim's conclusions because of methodology, although he presented no original histologic research to verify his conclusions.

In 1932, the first quantitative study of force compared to the biologic reaction of the PDL was published by Schwarz.[4] He placed continuous tipping forces on the teeth of a dog. Using a variable force spring for a period of five weeks, he found the reaction of the PDL varied as to the amount of force. With forces of 3.5 and 17 grams, there was active resorption on the side of pressure. The PDL remained vital with no necrotic regions. However, when a force of 67 grams was placed on a tooth, he found an area of the PDL which was not nucleated and was changed into a "structureless mass." He noted that the PDL was so compressed that blood could not circulate in the region of pressure and hence the tissue had "suffocated" and had been "destroyed." From this he concluded that the force that was most favorable to move teeth orthodontically was that "not greater than the pressure in the blood capillaries." In man and most mammals, this pressure is 15 to 20 mm. Hg or about 20 to 26 grams for one square cm. of surface area.

In a series of investigations on dogs and humans under traumatic occlusion, Gottlieb[5] corraborated Sandstedt's findings. He found frontal resorption on the pressure side with light forces, but necrosis occurred when heavy forces were used. These areas of compression were removed by the process of undermining or rear resorption. On the tension side he

found bone formation.

Herzberg in 1932,[6] published the histologic results of movement of a premolar in an 18-year-old female. The significance of this report was that it was one of the first on human tooth movement. Initiating a lingual tipping movement with a continuous elastic force, he accomplished a two millimeter movement of the lingual cusp tip over a period of 70 days. Due to technical difficulties, the pressure side was mutilated during surgery and was inadequate for histologic examination. On the side of pull, he found the alveolar bone was arranged in spicules at right angles to the long axis of the tooth similar to the findings of Oppenheim in baboons. He concluded that on the basis of the material obtained, the reaction of human alveolar bone under active orthodontic therapy was similar to that of baboons as described by Oppenheim.[2]

The next major publication of a human tooth movement study was presented by Oppenheim in 1935.[7] He moved the maxillary first bicuspids of patients requiring extractions for orthodontic purposes by a variety of methods. They included an Angle expansion arch, a straight spring soldered to the expansion arch, a spring on a lingual arch, and a closed loop spring. These appliances placed either a continuous or intermittent tipping force on the tooth and were activated at varying time intervals from 30 to 115 days. He, like everyone else, experienced a variety of biologic reactions between all specimens, even those on the same patient using the same appliance. However, there were some similarities in the specimens so that he could form some basic conclusions about the different tissues of the periodontium. The reaction of the alveolar bone differed from his findings in baboons. On the side of pressure, there was frontal resorption by osteoclasts, and on the side of traction there was an even

apposition of "osteoid." He also found in all specimens some degree of cemental resorption. This would be better termed lateral root resorption since the resorption bays extended into dentine. In the PDL, he noted the formation of a "buffer," consisting of increased and enlarged blood-filled vessels and capillaries. He concluded that this "angioma-like" formation intercepted the pressure acting like a sponge or cushion. He found this phenomenon present only when weak intermittent forces were placed on the tooth and never with either continuous or strong intermittent forces. He claimed these latter forces interfered with the vitality of the cells of the PDL preventing resorption to take place. He also concluded that the advantage of having this "angioma-like" formation is the favoring of bone resorption due to the proximity of the blood vessels to bone. However, this could also have a deletrious effect in favoring the resorption of cementum and dentine.

In 1938, Stuteville[9] published his tooth movement studies on humans and dogs. By use of a fixed lingual arch and attached finger springs, he initiated a continuous buccal tipping force on the maxillary first bicuspids of his human patients. He varied the force between 5 and 150 grams and the time interval between 29 and 98 days. In one specimen in which a 100 gram force was exerted over a 29 day interval, the tooth was pressed against alveolar bone causing a "necrosis" of the PDL. Apically and occlusal to this necrotic area, undermining resorption was taking place. He interpreted this reaction as the result of the compression of the PDL, thus obliterating the flow of blood. This resulted in anoxemia of the tissue and necrosis followed if the pressure was maintained.

In a later article, Stuteville[10] commented on the "angioma-like" formation reported by Oppenheim. He interpreted these as "areas of

autolysis of the necrotic connective tissue in the periodontal membrane together with tearing during removal followed by shrinkage during preparation." He based this on his own histologic material in which he found no endothelial lining in these areas, as well as the lack of blood cells within these areas. He also could not trace these into blood vessels by serial section.

In 1942, Oppenheim[8] commented on Stuteville's assertions on his findings. He claimed that these "angioma-like" areas were not artifacts; that they were formed under a light intermittent force and not under either a strong or continuous force. When a strong force was placed on a tooth, he said necrosis occurred. This was removed by undermining resorption thus relieving the pressure. Under these conditions, it was not necessary to form this "protective cushion of vessels." He stated that this phenomenon could be considered a peculiarity of human PDL. He also suggested that this could be some kind of "fibroid degeneration" as described by Hopewell-Smith.[18] Characteristic of this degenerative process was the "areolisation" of the PDL. Also the spaces had no definite walls of other blood vessels and all traces of osteoblast and epithelial rests of Malassez had disappeared.

Probably the most significant and extensive studies of human tooth movement were published by Reitan starting in 1951. His first studies were done on dogs, and he used these findings as a model for his human tooth movement evaluation.

Based on his experiments on dogs, Reitan[14] reported the formation of "necrotic pressure areas" that must be removed by undermining resorption. He stated that a "continuous force may result in stasis of blood capillaries and 'hyalinization' of the collagenous fibers with diminution in the

number of cells."[14] In his later publications, any cell-free area of the PDL was said to be "hyalinized."[12] Actually, this was not a new finding for all he did was to substitute his own term for a histologic pattern originally described by Sandstedt[1] as "necrotic" tissue.

In 1951, he reported his findings on the maxillary premolars of 23 young patients aged 11 to 13 which had been subjected to a variety of forces for varying amounts of time. In one specimen in which a fixed appliance exerted a 70 gram continuous tipping force for two days, he noted the appearance of hyalinized PDL on the side of pressure. He also noted that the capillaries were partly compressed, and there was a presence of "pyknotic" nuclei. When heavy forces were placed on a tooth by an activator containing a mid-palatal jackscrew, hyalinized areas on the side of pressure were also found. He concluded that this hyalinized connective tissue was more resistant to resorption and the avoidance of a force which could cause this hyalinization would result in the faster movement of teeth. He also commented that he found several specimens with an "angioma-like" appearance (Oppenheim) but called them obvious artifacts, since they appeared in only two of his histologic specimens.

In later publications, Reitan[12,13] formulated some clinical applications based on his histologic findings. He believed that during tipping movements the PDL was characterized by the formation of cell-free areas and that these areas prevented the resorption of bone. However, in bodily movements there was less of a tendency to form compressed areas, for the force was distributed over the whole alveolar wall. He stated that light forces between 50 and 75 grams could prevent the formation of additional cell-free areas after the removal of the initial hyalinized areas by undermining resorption. He explained tooth movement on a

biological basis. The initial movement was the compression of the PDL which took four to seven days. This was followed by a lag phase of about two to three weeks in which there was no movement due to the formation of hyalinized areas. After this time period, which was about 30 days, the hyalinized areas were removed by undermining resorption, and frontal resorption and active tooth movement were initiated.

In 1971, Church[15] conducted a study on human tooth movement. He placed a 70 gram continuous buccal tipping force on the upper premolars of selected orthodontic patients. The appliances used were fixed lingual arches with attached finger springs. He then obtained his pressure side specimens of alveolar bone and tooth, at fixed time intervals of seven, 14, 21, and 28 days.

His findings were as follows: At the end of seven days, he noted the presence of cell-free areas along with undermining resorption from marrow spaces in areas of compression. At 14 days, most specimens showed limited cell-free areas and the presence of immature fibrous bone. His most significant findings occurred at the 21 day interval. Most of his specimens showed a widened PDL space with stasis of blood vascular channels. There was little or no osteoclastic activity, although there was some osteoblastic activity along with the presence of fibrous immature bone. Reorganization of the PDL was evident by the capillary budding in areas of previous thrombosis and the return of the new PDL cells. Also, the fiber bundles were widely separated and the presence of occasional myelinated nerve bundles was reported. In three of the specimens, he reported the morphologic evidence of "fatty degeneration." He based this assumption on the appearance of "vacuoles in the PDL created by the characteristic 'signet ring cells' in which the nucleus has been forced to the periphery

of the cells, leaving a large round vacuole for fat storage." He attributed the formation of these areas of "fatty degeneration" to local ischemia, brought about by a "change in the resident cells due to anaerobic glycolysis from reduced blood flow rather than an infiltration of cells from other areas." Also associated with this fatty degeneration was a large amount of lateral root resorption. At the end of 28 days the primary finding was of PDL reorganization. He also noted the presence of macrophages in locations which appeared to be former areas of fat deposit.

The biologic phenomenon in which there is an increase of histological demonstrable fat in the parenchymal cells of an organ or tissue has been described by a number of similar terms. They include fatty metamorphosis, fatty infiltration, lipomatosis, fat phanerosis and fatty degeneration. The interchange of these terms by various authors has led to some confusion as to their exact definition.

Smith and Gault[19] divided fatty metamorphosis into two categories, fatty infiltration and fatty degeneration. They claimed that fatty infiltration or lipomatosis was caused by the incomplete combustion of excess ingested fat, either through insufficient exercise or some change in physiology. This resulted in the excessive deposition of fat in various tissues of the body. It could be thought of as a normal physiological process and it only became pathological in extreme cases. They stated that fatty degeneration was an intracellular pathological process which was due to cell injury. This injury was most likely due to tissue anoxia caused by either the direct mechanical interference of blood flow, severe anemia, or circulating toxins which disrupted the normal metabolism of the cell, thus interfering with normal oxygen supply. They could not explain the actual mechanisms involved in fatty degeneration

but considered the source of the fat as the unmasking of previously present lipids, a process known as fat phanerosis. They described the typical cell as one in which the cytoplasm was finely granular or vacuolated. They reported that Virchow had this similar finding giving the cell a foamy appearance in the usual H and E stained preparation.

Tallis[20] also called this increase in fat, fatty metamorphosis. He described the involved cells as having "small or large clear cytoplasmic vacuoles" in routine H and E sections. The cells often showed other degenerative changes, expecially cloudy swelling and hydropic degeneration. It was usually found in the liver, heart, or kidney, but was occasionally seen in other tissues. In the kidney and heart, the chief cause was cellular injury due to toxic agents or anoxia, and he termed this fatty degeneration. In the liver, fatty metamorphosis could be caused by two different methods. When there was an "excessive mobilization of depot fat," he called it fatty infiltration. When the cause was the "impairment of hepatic utilization and secretion of lipids" due to injury of the liver cells either by carbon tetrachloride poisoning, the toxemia of infection, or anoxia, he reported it as fatty degeneration.

Anderson,[21] Florey[22] and Cameron[23] all stated that when the fatty change was due to cell injury, the appropriate term to use was fatty degeneration. One of the main causes for this cell injury was severe anoxia in which the cells were denied an adequate supply of oxygen to maintain their metabolic functions.

In a study of 270 patients proven to have fatty metamorphosis of the liver on biopsy, Leevy[24] examined the possible causes for this fatty change. He found it difficult to determine the specific mechanisms involved, although he stated that the fatty change was the result of an

overall disparity between the rate of deposition and removal of neutral fats. Among the many possible causes he listed for this change, alcohol, heart disease and diabetes mellitus were the major findings.

Another reason for the presence of fat within various tissues could have been the regeneration of fat. Maximow and Bloom[25] believed that any fibroblast had the potential to be turned into a fat cell, although this had not been proven conclusively. Another possible source could have been from undifferentiated mesenchymal cells.

Clark and Clark[26] using clear "round table chambers" in the ears of rabbits noted the appearance of intracellular fat in half of their samples when examining regenerating tissues in this cell-free area. These fat cells were in close proximity to regenerated capillaries in areas of slow or sluggish circulation. Since serial studies were possible, they found that these fat cells formed from fibroblast-like cells. At first the fat appeared as small droplets which continued to increase in size. Finally, it displaced practically all visible cytoplasm, forcing the nucleus and cytoplasm to the periphery to the cell, forming the "signet-ring" form of adipose tissue. They also noted the reverse of this process. However, the cells never reverted to their original spindle-like appearance due to stretching of the cell wall. The fat from these cells was derived from either a soluble form or from intracellular activity but never from phagocytosis of extracellular fat droplets.

The only other published reports on human material involving either fat or "angioma-like" formation within the PDL was by Kronfeld and Boyle on necropsy material. Kronfeld[16] examined the jaws of a ten-year-old Negro child who had died of sickle cell anemia. On examination of the distal side of the upper right cuspid, he found a great number of

enlarged blood vessels within the compressed PDL. He deduced that this was an area similar to the "angioma-like" formation described by Oppenheim. He claimed that this phenomenon was produced by the intermittent forces of eruption and lip pressure on the tooth. Boyle[17] stated that embedded teeth or teeth without an antagonist for a long time would transform their fibrous PDL into fat tissue as a result of inactivity. He showed an upper second molar which had lost its antagonist for several decades. Part of the PDL had been completely replaced by fat tissue, similar to that found in the marrow spaces. The former alveolar bone seemed to run through the fat tissue like a trabecule of cancellous bone.

In summary, the earliest histologic study of tooth movement material was initiated by Sandstedt (1901). Other studies on animals were conducted by Oppenheim (1911), Schwarz (1932), and Gottlieb (1930's). After the first human orthodontic tooth movement was published by Herzberg (1932), a number of other authors conducted studies in this field including Oppenheim (1935), Stuteville (1938), Reitan (1951 to present) and Church (1971). Also various mechanisms concerning the possible presence of fat within the PDL were examined.

From these studies it can be concluded that following the application of force on a tooth, there is a resulting compression of the PDL on the side of pressure resulting in the formation of a cell-free area. This area is removed by a process of undermining resorption from the adjacent marrow spaces with the resultant re-establishment of the normal PDL space. However, the associated changes seen within this re-established PDL space remains in question. Whether or not these observed vacuoles found in PDL of some specimens represent fat, capillary spaces, or artifacts remains unanswered.

## MATERIALS AND METHODS

Four orthodontic dental patients who required the extraction of maxillary bicuspids were utilized in this study. They included two males and two females aged 12 to 14. A lingual appliance (fig. 2) which created buccal tipping of the maxillary first bicuspids was activated with a force of $70 \pm 5$ grams and stabilized by cemented molar bands. The finger springs were calibrated by a dead weight loading device and fabricated from .016 inch standard stainless steel orthodontic wire (Unitek Corp.). These springs were designed with a double helix and a five millimeter lever arm to yield the required load at a deflection of 4-5 mm. The springs were checked in the mouth at the time of insertion with a calibrated elastic tension gauge (fig. 3).

On the tip of the lingual cusp of the maxillary first bicuspids, small indentations were placed with a dental handpiece using a No. $\frac{1}{4}$ round bur. The distances between the indentation on one side and an occlusal pit on the opposite side maxillary bicuspid were taken using a divider and transferred to an index card. The occlusal pits used were marked on the study cast for later identification. Also, the distances between opposite side indentations were recorded. The patients were seen on the seventh, 14th, and 21st days after placement of the appliance to record the distances between the previously mentioned reference points. At the time of surgical removal, the finger springs were checked with the elastic tension gauge to determine the amount of force exerted by the finger springs.

At the end of 21 days, the maxillary first bicuspids with

approximately 10 mm. of buccal alveolar bone (fig. 4) were surgically removed. Healing following the extraction was uneventful.

After a running water rinse, the specimen of tooth and bone was fixed in a ten percent neutral buffered formalin solution. After 30 minutes in the solution, the crown of the tooth was sectioned from the root and bone by a high-speed dental handpiece. The specimen was then returned to the fixative for 24 hours. Decalcification was accomplished in ten days by citric acid-sodium citrate solution. Twelve micron frozen sections were cut on a clinical microtome employing $CO_2$ as the freezing agent. These sections were floated on water prior to staining. Representative sections of each specimen at the cervical area, 0.5 mm., 1.0 mm., 1.5 mm. and 2.0 mm. apical to the cervical area were obtained. The sections were routinely stained by Hemotoxylin and Eosin, Sudan Black B lipid method, and an Acid Picro-Mallory method for fibrin.

All slides were coded and read by two independent observers using a Zeiss GL 4 brightfield microscope and photomicrographs were made on a Zeiss photoscope. The data was compiled in Table 1 according to subjective evaluation of the slides by the observers.

## HISTOLOGIC OBSERVATIONS

The sample for this study consisted of seven teeth with attached buccal alveolar bone obtained from four orthodontic patients who required the extraction of maxillary first premolars. One specimen was damaged during histologic sectioning and was unsatisfactory for study. All specimens were of tooth and pressure side alveolar bone which had been subjected to a continuous tipping force for 21 days.

There was much individual variation in the reaction of the tissues of the periodontium to the forces placed upon them. This made a single explanation of what actually happened during tooth movement difficult. However, there were similarities in the reactions so that some basic conclusions could be made.

Six of the seven specimens showed areas of compression of the PDL. Associated with this compression was the appearance of cell-free areas characterized by the condensation of the collagen fibers, the absence of all PDL cells, and the obliteration of all blood vascular channels and nerve bundles (figs. 5,6). This created the characteristic cell-free zones described by Reitan[11,12,13] as "hyalinized" PDL tissue. These zones were limited to the alveolar crest region of the PDL. In some specimens, the compression was so severe that the alveolar bone almost contacted the tooth. On sectioning, these areas experienced some tearing or separation artifacts (fig. 6). This gave the impression that the tissue was necrotic and hence tore easily in the frozen section technique.

All specimens showed areas of active frontal bone resorption (fig. 7). This was evidenced by the presence of numerous osteoclasts lying within

Howship's lacunae. Adjacent to the cell-free areas, the resorption of the bone from the PDL was in an undermining fashion (fig. 5). In some specimens, spicules of resorbing bone seemed suspended in the middle of the PDL, due to the joining of these resorption pockets from both sides of the cell-free areas. Most specimens showed osteoclastic activity within the marrow spaces resembling the "undermining resorption" first described by Sandstedt[1] (fig. 8). In some areas these pockets of undermining resorption had broken through the alveolar bone into the PDL space.

Lateral root resorption was present in all specimens which exhibited cell-free areas. The resorption was in an active state at the time of extraction due to the presence of dentinoclasts in their resorption bays (figs. 7,9). The root resorption was seen in areas of the tooth adjacent to cell-free zones. No root resorption was seen in the one specimen that did not exhibit a cell-free area.

Adjacent to these cell-free areas, the PDL space was widened. The major finding in these areas was the vigorous reorganization of the PDL (figs. 7,9). This was characterized by the presence of numerous blood vascular channels, which were identified by the presence of red blood cells within their lumen. In some cases these areas of capillary budding resembled the "angioma-like spaces" described by Oppenheim[7] (fig. 8). The collagen fiber bundles were disorganized and widely separated. This made the identification of glomera of the PDL, as described by Provenza,[27] very easy (fig. 10). In normal PDL, these glomera, or groups of arteriovenous shunts surrounded by a fibrous capsule, appeared to be masked by the fibers. Very few areas of osteoblastic activity were present. Newly formed fibrous immature bone was present in these areas.

On examination of the Sudan B stain for lipid, only one specimen

showed evidence of fat within the PDL space (fig. 11). It consisted of Sudan B lipid positive cells located in an area where a marrow space communicated with the PDL space. Within this marrow space, positive evidence of adipose tissue was present. The myelin sheaths surrounding nerve fibers were seen within the reorganizing PDL with this lipid stain (fig. 12).

The Acid-Picro Mallory stain was a selective stain for fibrin. In none of the sections was there any evidence of fibronoid, a derivative of fibrin. Areas of hemorrhage were found adjacent to cell-free zones. The disorganization of the fibers and the presence of glomera of the PDL were well demonstrated by this staining method (fig. 10).

## DISCUSSION

The appliance consisting of a fixed lingual arch with attached finger springs proved generally satisfactory for this study. However, some disadvantages became apparent. The finger springs often slipped under the lingual cervical gingiva and had to be re-bent to minimize this displacement. No appliances were broken or lost and the patients reported no apparent displacement of the finger springs between appointments. Some springs lost force during the 21-day interval. When the springs were checked in the mouth with the elastic tension gauge, approximately half of the force had disappated in some, while the force remained unchanged in others. The possible cause for the loss of force could have been the fatigue of the wire due to stresses placed on the springs while in the patient's mouth. This variety of forces could have been a possible cause for the variety of tissue reactions exhibited by the specimens. A method of placing a constant continuous force on a tooth could eliminate this source of error. Oppenheim[7] listed bent or broken springs and the slipping of the springs under the gums as disadvantages of the lingual arch appliance during his investigations. This was also reported by Church.[15]

The method used to record the amount of tooth movement was adequate. The estimated range of total error was $\pm$0.2 mm. This was the approximate error obtained on repeated measurements on a single specimen. Compared to the actual movement recorded, this was relatively a large amount. The sources of error were many. The main reason was the difficulty in establishing the passive position of the extremely mobile teeth. Other reasons were the difficulty in locating the exact reference pit on the

apposing second premolar, and the error in the transference and measurement of the distances on index cards. These recorded movements were of the cusp tips and just reflected the actual movement at the cervical area of the tooth.

The movements of each specimen at the end of the seventh, 14th, and 21st day were recorded in Table 1. They were also interpreted graphically (fig. 1). These measurements were used to show the relative amount of tooth movement, and to help explain the possible biologic reasons for this movement. This was in no way an empirical study of tooth movement, and statistical analysis of the data was not undertaken due to inadequate sample size. However, the total range of possible error was reported (fig. 1).

The individual variation of tooth movement was probably the most significant finding. Even on different teeth of the same individual, the movement varied. However, six of the eight specimens reacted in a similar manner. Based on these reactions, some conclusion could be made. The amount of movement was greatest during the first seven days. Most of this movement could be attributed to compression of the PDL. There was also the possibility of bone bending. During the next seven days, there was relatively little or no movement recorded. This was probably due to the presence of cell-free areas that had not yet been removed by undermining resorption. These findings were in agreement with those of Reitan.[12,13] During the last seven days, there was a slight amount of movement. This could have been due to the elimination of the original cell-free areas and the formation of new cell-free areas apical to the original areas. Another possible reason could have been the frontal resorption of the more cervical portions of the cell-free zones, leaving only the more apical portions. These conclusions were based on the

presence of cell-free zones in five of the six specimens and the presence of PDL tissue cervical to these cell-free zones with no attached buccal alveolar bone. Since one specimen was unsuitable for histologic examination, definite evidence of this reaction was not possible. This differed from Reitan's conclusions. He stated that once a cell-free "hyalinized" area was removed by undermining resorption, the formation of additional "hyalinized" areas was unlikely if the forces used were not greater than 75 grams.[13] He also stated that active movement by frontal resorption of alveolar bone began after about 30 days.[12,13]

The other two specimens showed very little movement during the 21 days. However, the reactions of their PDL's were quite different. In one specimen there were no cell-free areas and active frontal resorption was taking place. The other showed a large cell-free area along with an extensive amount of undermining and root resorption. The possible reasons for the first reaction could have been the loss of activation of the finger spring, the blocking out of the tooth preventing movement, or just the normal reaction for that particular PDL. The reason for the second reaction was probably caused by the formation of a large cell-free area. Due to its extensive manner, this area had not yet been removed by undermining resorption resulting in little or no tooth movement.

There were no complications associated with the surgical removal of the specimens. The attached buccal bone was not separated from any of the specimens as evidenced by the lack of separation artifacts on histologic examination. At the time of the surgical procedures, it was the subjective evaluation of the oral surgeon that there was an increase in vascularity of the bone specimen. He also commented that the tissue at the cervical portion of the specimen resembled granulation tissue. Close

examination of the specimens showed the cervical portions of alveolar bone to have an eroded and frayed appearance. Also the height of the bony attachments seemed more apical than normal. All this gave the impression that the most cervical parts of the alveolar bone had been resorbed. However, the lack of controls made this evaluation strictly subjective. The need for controls for this and other reasons, should be considered for future studies.

There were many problems associated with the histologic techniques used. During the frozen section technique, one specimen was damaged. The buccal alveolar bone was separated from the tooth and the section was unsatisfactory for study. There was also difficulty in obtaining sections at exactly the areas indicated for study. Increased experience in the use of this technique may eliminate these errors in the future. On routine H and E staining of the frozen sections, the specimens showed shrinking or tearing artifacts (fig. 13). The stains were repeated with a modified technique and these artifacts were avoided. In the sections stained with Sudan Black B, not all fat cells within the marrow spaces were positively stained (fig. 14). This was probably due to the obliteration of the lipid contents of the cells on sectioning. This made the possibility of some fat cells within the PDL space not being positively identified. The increase in thickness of the sections to about 20 to 25 micra could eliminate this artifact. There were also some false positives when the stain was trapped within blood vascular channels. Evidence of a positive reaction of the stain was the staining of some fat cells within marrow spaces and the staining of the myelin sheaths surrounding nerve fibers (fig. 12). Other possible sources of error in this staining technique were pointed out by Louchard.[28] He showed that the increase in pH

greatly enhanced the positive reaction of the stain. He also explained the instability of commercial preparations of Sudan Black B. The use of another lipid stain in addition to Sudan Black B should be evaluated for future studies in which the identification of fat is indicated.

The major finding in the histologic examination was the presence of cell-free areas within the PDL. In some areas of severe compression, the tissue seemed to be necrotic. This observation was based on the fact that there were tearing or separation artifacts in these areas. This gave the impression that the tissue was necrotic and hence tore very easily with the frozen section technique. This probably was caused by the prolonged obliteration of blood supply by the compression of blood vascular channels in this area. This resulted in local ischemia and the resultant cell death and necrosis. There was no evidence of an infiltration of inflammatory cells and hence the necrosis was aseptic in nature. This result was similar to that reported by Sandstedt[1] in dogs. Oppenheim[7] and Stuteville[9] also reported this finding in humans using the same experimental model at similar time intervals. Reitan[10,11,12,13] found this similar phenomenon in his studies, but disregarded the older terms of "necrosis" and "degeneration" and called it "hyalinization." This term means the formation of a substance which on gross or microscopic examination appears glassy, homogenous and translucent.[30] Although this was a more convenient term, it did not accurately describe the histologic pattern observed. He later stated that these "hyalinized" areas remained for a period of about 30 days when a continuous force was placed on a tooth. After the removal of these cell-free zones by undermining resorption, additional "hyalinized" areas would not be formed with forces between 50 to 75 grams. Church[15] reported that only one of

his specimens exhibited a cell-free area after 21 days. The rest showed a widened PDL space along with reorganization of the PDL. The results of the present study closely resembled Church's findings at the 14-day interval. This could have been attributed to the wide variation of reaction of the PDL during tooth movement. The presence of cell-free areas at the end of 21 days could also have been due to another reason. These areas could have been formed subsequent to the removal of the original cell-free areas. Initially, the PDL lacked the resistance to absorb the 70-gram force. Therefore, a continuous constant force would likely form other cell-free areas once the initial pressure zones were relieved. Since in some cases the movement recorded was much more than could be achieved by compression of the PDL space and bone bending, this explanation seemed plausible. This interpretation would make this type of tooth movement an uneven, erratic process and not an even and continuous one.

Frontal bone resorption in non-cell-free areas and undermining resorption from marrow spaces were in harmony with the findings of previous authors.[1,4,5,9,12,13,15]

The finding of lateral root resorption in all specimens with cell-free areas and not in the one specimen without a cell-free area implied a cause-effect relationship. These areas of root resorption were located adjacent to the cell-free areas. This gave the impression that the clastic multinucleated giant cells could not distinguish between tooth or bone tissue in their attempt to relieve the pressure in these areas.

In areas of the PDL which were widened, the major finding was the vigorous reorganization of the tissue of the periodontium. Capillary budding was evidenced by the presence of numerous blood vascular channels.

Some of these areas resembled the "angioma-like" appearance described by Oppenheim.[7] He stated that this formation was a product of weak intermittent forces and never of either a strong or continuous forces. He claimed these latter forces caused necrosis of the PDL. In this study both strong and continuous forces were used and the formation of these "angioma-like" spaces resulted. The increase in vascularity appeared to be in direct correlation to the increase in activity within the PDL. This change was probably brought about by the increased demands of the reorganizing PDL following the removal of pressure cell-free areas. This was demonstrated by the absence of increased vascularity in the one specimen which exhibited no cell-free areas and the increased vascularity in the other specimens. However, this increase was the subjective evaluation of the investigator. In order to substantiate the relative increase in vascularity, controls from the same patients were needed.

The finding of glomera of the PDL in all specimen was a surprising finding. Provenza[27] described these as arteriovenous shunts surrounded by a fibrous coat. These were located throughout the PDL but were more numerous apically. He stated that the presence of these glomera might have some pathologic implications. Since the blood was shunted to the efferent vessels by-passing the microcirculation, prolonged shunting could prevent the "metabolic interchange occurring in the capillary bed." In this study, these glomera were seen in areas of vigorous reorganization of the PDL. The need for an adequate blood supply in these areas made Provenza's explanation for their presence somewhat doubtful. The glomera were easier to identify than in normal PDL tissue. This was probably due to the disorganization of the fiber bundles, which probably mask the glomera in normal PDL. The need for controls would help in the

explanation of this phenomenon.

Very few isolated areas of osteoblastic activity were noted. The presence of fibrous immature bone was the subjective evaluation of the investigator. Frost[29] stated that this bone or "osteoid seam" could best be seen in fresh undecalcified sections. He also stated that this type of bone was not seen in decalcified sections and the pink borders seen in the normal H and E stained sections usually represented another phenomenon. These isolated areas of bone formation seemed to indicate an attempt by the alveolar bone to remodel or repair itself due to changes of pressure within the PDL. However, the major bone remodelling process exhibited was active frontal resorption.

Only one specimen showed any evidence of Sudan B lipid positive cells within the PDL. These cells were located in one isolated area which communicated with a marrow space containing fat cells. This finding indicated that fat cells could have appeared within the PDL space as a result of movement of a tooth into fat-filled marrow spaces. However, since only a few specimens exhibited fat cells within their marrow spaces of buccal alveolar bone, and lipid-positive cells within the PDL were demonstrated in only one specimen, a valid conclusion could not be made. In order to confirm this plausible explanation, additional studies at the same or longer time intervals are needed.

Church described the presence of "signet-ring" cells within the PDL of his 21-day specimens. He concluded that these specimens exhibited morphologic evidence of fatty degeneration. These cells were found apically to former cell-free areas where there had been "prolonged ischemic without complete necrosis." This ischemia was brought about by the compression and "occlusion of a majority of vessels."[15] Compression

of the PDL was seen in the present study but this fatty change was not seen. This made Church's conclusions highly unlikely, although it cannot be totally discounted. His explanation as to the cause of this change was in agreement with the cause of fatty degeneration in other tissues of the body as described by other authors.[19,20,21,22,23] Finally, the possibility of Church's histologic finding to be artifacts cannot be discounted by the results of this study.

## SUMMARY AND CONCLUSIONS

The purpose of this study was to verify the histologic findings of previous authors dealing with human tooth movement. Of utmost concern was the finding of vacuoles within the PDL following 21 days of tooth movement. Also, possible explanations for the presence of these vacuoles, if they exist, were explored.

The sample for this study consisted of four orthodontic patients who required the extraction of maxillary first bicuspids for treatment. Continuous buccal tipping forces of 70 grams were placed on these teeth utilizing finger springs attached to fixed lingual arches. Following 21 days of tooth movement, these teeth along with attached buccal alveolar bone were surgically removed and prepared for histologic examination. Since one specimen was damaged during histologic sectioning, the sample yielded seven specimens suitable for histologic study.

The histologic findings were not unlike those reported by previous authors. Following the application of forces that compressed the PDL, the blood vascular channels became occluded. Continued application of these forces resulted in "necrosis" or "hyalinization" of the PDL, as evidenced by the formation of cell-free areas. These areas were removed by frontal resorption from the adjacent PDL and undermining resorption from marrow spaces.

Based on the results of this study there were three possible explanations for the presence of vacuoles within the PDL. The first was the presence of a large number of capillaries within the reorganizing PDL giving the "angioma-like appearance" as described by Oppenheim.[7]

The second explanation was concerned with the positive identification of fat cells within the PDL, using a specific fat-staining histologic technique. Church's[15] explanation of fatty degeneration as the possible cause seemed to be in error. Similar conditions that he stated were necessary for this process to take place were observed without the appearance of this fatty change. The more likely explanation was the movement of the tooth into fat-filled marrow spaces, resulting in fat cells being incorporated into the newly established PDL space. The last possible explanation was that these vacuoles were artifacts of the histologic staining techniques.

Other conclusions based on the present study include:

1. Additional cell-free areas appeared to be formed following the removal of the original cell-free areas. This was based on the relatively large amounts of tooth movement in some specimens that was more than could be attributed to compression of the PDL space and bone bending and the presence of cell-free areas on histologic examination of these specimens.

2. Lateral root resorption seemed to be a side-effect of the formation of cell-free areas. The clastic multinucleated giant cells could not distinguish between tooth or bone in their attempt to relieve the pressure in this area. Hence, areas of lateral root resorption were found adjacent to the cell-free zones.

3. There was a great increase in vascularity of the reorganizing PDL. Besides the large number of capillaries being present, the finding of many glomera of the PDL was surprising. Their identification was made easier primarily due to the disorganization of the fiber bundles of the PDL which unmasked the previously obscure glomera.

# BIBLIOGRAPHY


1. Sandstedt, C.: Nagra Bidrag Till Tandregleringens Teori. Stockholm, 1901 (as reported by Reitan[11]).

2. Oppenheim, A.: Tissue Changes Particularly of the Bone Incident to Tooth Movement. Amer. Orthodont. 1911 and 1912.

3. Stallard, H.: Criticism of Oppenheim's 1911 Report. Int. J. Ortho. 14:872, 1928.

4. Schwarz, A.: Tissue Changes Incidental to Orthodontic Tooth Movement. Int. J. Ortho. 18:331, 1932.

5. Gottlieb, B.: Some Histological Facts Useful in Orthodontic Practice. A.J.O. & O.S. 28:167, 1942.

6. Herzberg, B.L.: Bone Changes Incident to Orthodontic Tooth Movement in Man. J.A.D.A. 19:1777, 1932.

7. Oppenheim, A.: Biologic Orthodontic Therapy and Reality. Angle O. 1935 and 1936.

8. _____: Human Tissue Response to Orthodontic Intervention of Short and Long Duration. Am. J. Ortho. 28:263, 1942.

9. Stuteville, O.H.: Injuries Caused by Orthodontic Forces and the Ultimate Results of These Injuries. A.J.O. & O.S. 24:103, 1938.

10. _____: A Summary Review of Tissue Changes Incident to Tooth Movement. Angle O. 8:1, 1938.

11. Reitan, K.: The Initial Tissue Reaction Incident to Orthodontic Tooth Movement. Acta Odont. Scand. Suppl. 6:1-240, 1951.

12. _____: Tissue Behavior During Orthodontic Tooth Movement. A.J.O. 46:881, 1960.

13. _____: Clinical and Histologic Observations on Tooth Movement During and After Orthodontic Treatment. A.J.O. 53:721, 1967.

14. _____: Continuous Bodily Movement and Its Histologic Significance. Acta Odont. Scand. 6:115, 1957.

15. Church, D.: Histologic Changes Occuring During Human Tooth Movement. Certificate Paper, University of Oregon Dental School, 1971.

16. Kronfeld, R.: Histologic Analysis of a Child With Malocclusion. Angle O. 8:21, 1938.

17. Boyle, P.E.: Kronfeld's Histopathology of the Teeth and Their Surrounding Structures. Lea & Febiger, 1955.

18. Hopewell-Smith, A.: The Normal and Pathological Histology of the Mouth. The Blakiston Co., 1918 (as reported by Oppenheim[5]).

19. Smith, L.W. and Gault, E.S.: Essentials of Pathology. The Blakiston Co., 1948.

20. Wallis, B.D.: Textbook of Pathology. McGraw Hill Book Co., New York, 1964.

21. Anderson, W.A.D.: Pathology. C.V. Mosby Co., 1966.

22. Florey, H.: General Pathology. W.B. Saunders Co., 1962.

23. Cameron, G.R.: Pathology of the Cell. Charles L. Thomas Co., 1951.

24. Leevy, C.M.: Fatty Liver: A Study of 270 Patients With Biopsy Proven Fatty Liver and a Review of the Literature. Medicine. 41:249, 1962.

25. Maximow, A.A. and Bloom, W.: A Textbook of Histology. W.B. Saunders Co., 1942.

26. Clark, E.R. and Clark, E.L.: Microscopic Studies of the New Formation of Fat in Living Adult Rabbits. Amer. J. of Anat. 67:255, 1940.

27. Provenza, D.V.: Oral Histology, Inheritance and Development. J.B. Lippincott Co., 1964.

28. Louchard, W.E.: Improving the Spècificity of Sudan Black B Staining. Dental Student. Vol. 50, No. 7:43, 1972.

29. Frost, H.M.: Bone Remodelling Dynamics. Charles C. Thomas, 1963.

30. Asimov, I., et al.: Stedman's Medical Dictionary. Williams & Wilkens Co., 1966.

| | Specimen Number | Patientage yrs. & mos. | Extent of Cell-free Zone | Extent of Undermining Resorption | Extent of Root Resorption | Presence of Blood Vascular Channels in PDL | Presence of Fatty Tissue in PDL | Presence of Fibrinoid in PDL | Amount of Movement in mm. | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | | | 7th | 14th | 21st |
| 1 | 17-R | 12-6 | 0 | 0 | 0 | + | 0 | 0 | 0.3 | 0.0 | 0.0 |
| 2 | 17-L | 12-6 | Unsuitable for study due to damage during histologic sectioning | | | | | | 1.0 | 0.2 | 0.4 |
| 3 | 18-R | 13-8 | + | +++ | + | + | 0 | 0 | 0.6 | 0.2 | 0.2 |
| 4 | 18-L | 13-8 | +++ | +++ | +++ | +++ | 0 | 0 | 0.1 | 0.2 | 0.0 |
| 5 | 19-R | 12-4 | + | + | + | + | 0 | 0 | 0.9 | 0.1 | 0.8 |
| 6 | 19-L | 12-4 | ++ | + | ++ | ++ | 0 | 0 | 1.3 | 0.0 | 0.4 |
| 7 | 20-R | 12-0 | +++ | + | + | +++ | + | 0 | 1.0 | 0.0 | 0.4 |
| 8 | 20-L | 12-0 | + | + | + | +++ | 0 | 0 | 1.0 | 0.1 | 0.1 |

Legend:
0 - Not present or not applicable
\+ - Mild
++ - Moderate
+++ - Extensive

Table 1. Summary of Histologic Observations

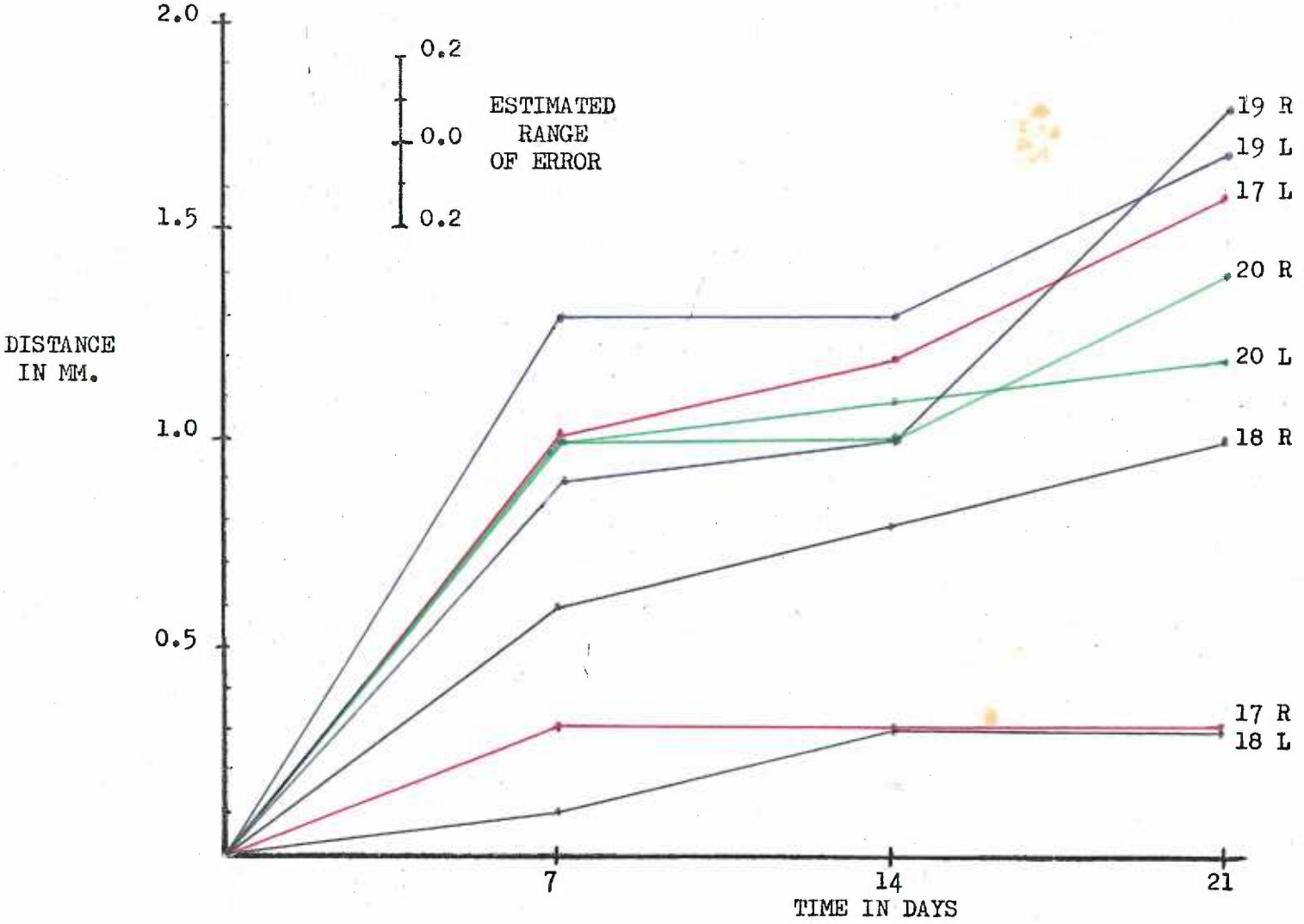


FIG. 1. MOVEMENT OF MAXILLARY FIRST BICUSPIDS

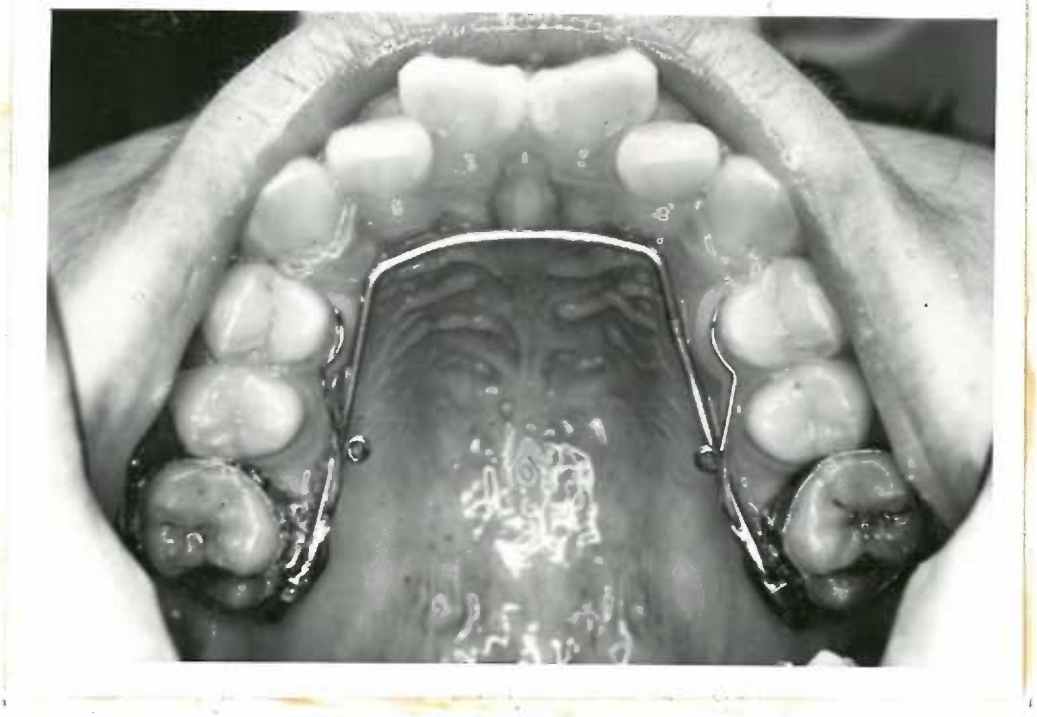

Fig. 2 An Occlusal View of the Appliance Used in This Study.

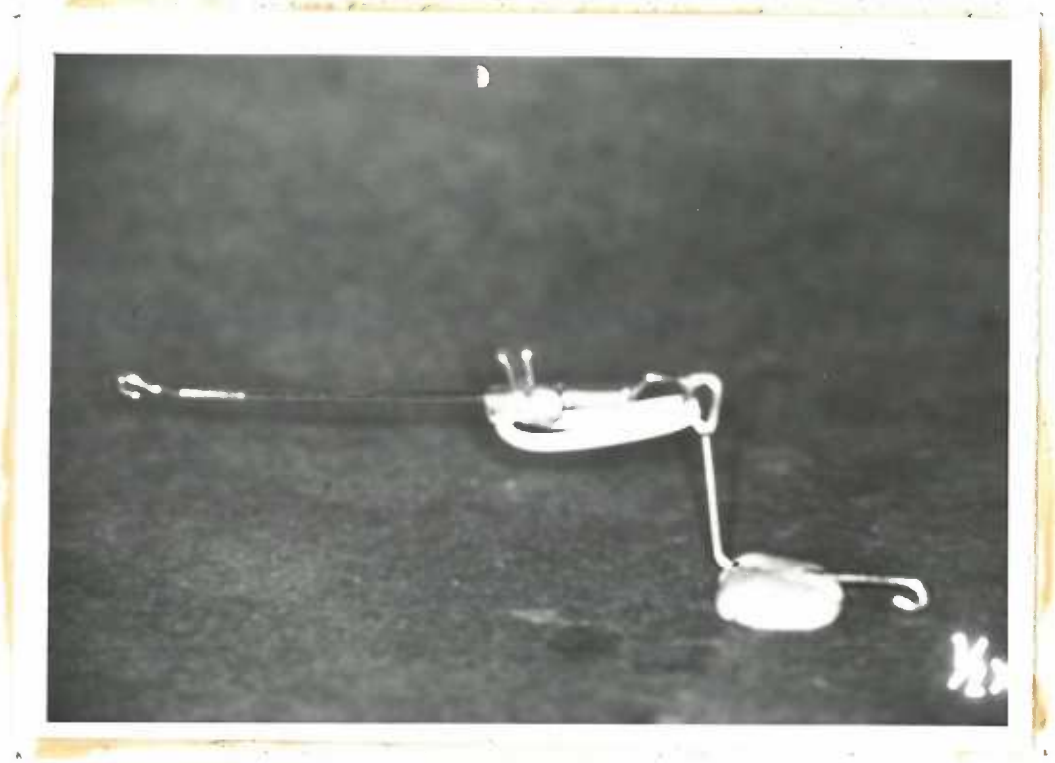

Fig. 3 The Elastic Tension Gauge Used to Check the Force Exerted by the Finger Springs Within the Mouth.

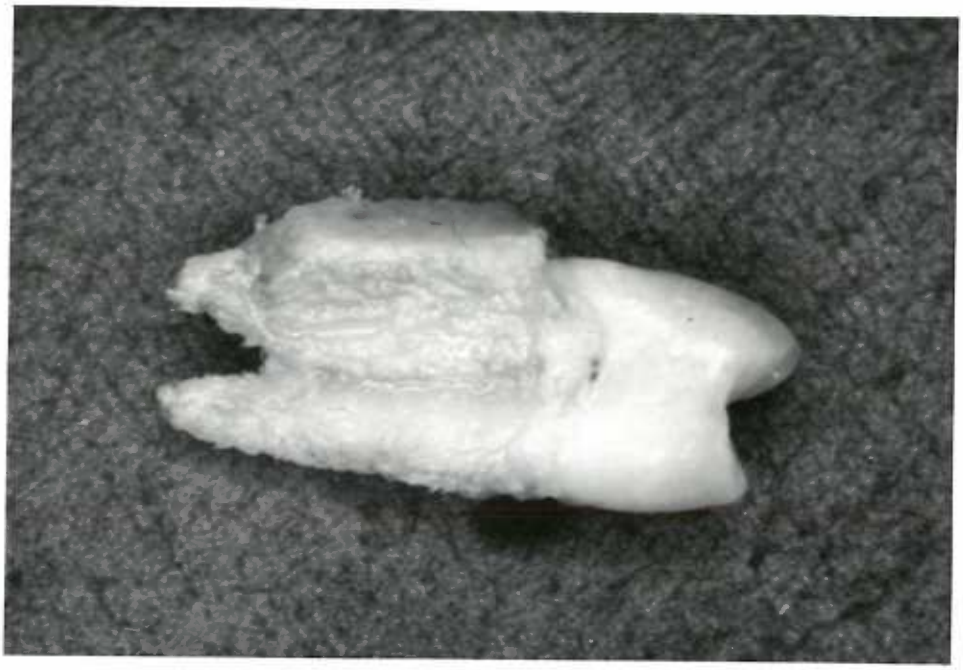

Fig. 4 Proximal View of Extracted Maxillary First Bicuspid and Attached Buccal Alveolar Bohe.

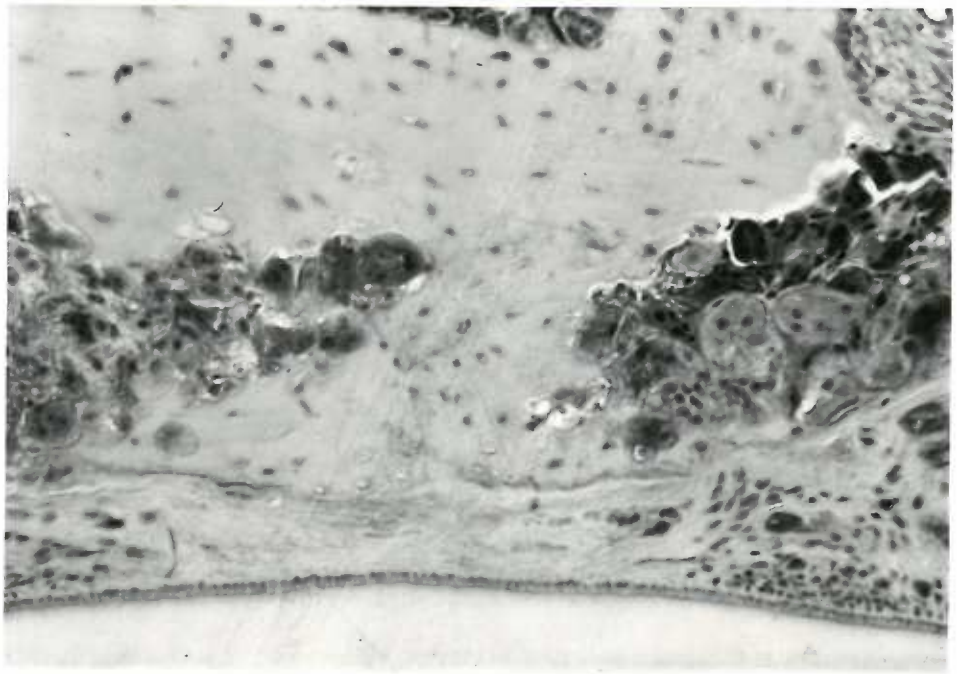

Fig. 5 An Area of Compression of the PDL Showing the Characteristic Cell-free Zone. Also Seen are Numerous Osteoclasts Involved in Widening the PDL Space.

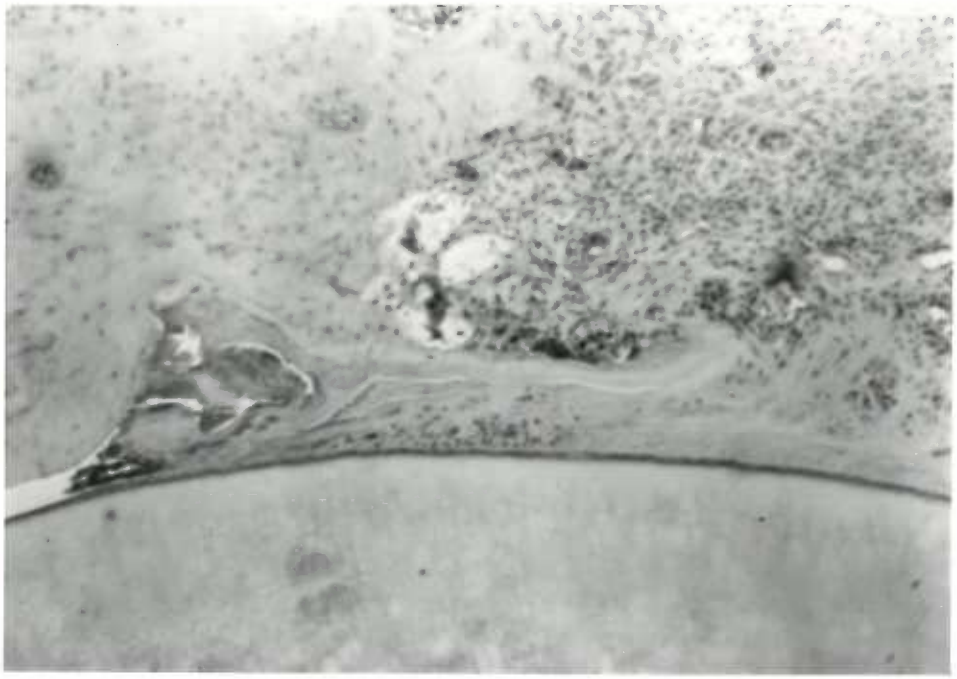

Fig. 6 Area of the PDL Showing Severe Compression -- at Far Left Side is the Separation Artifact Interpreted as Area of Necrosis.

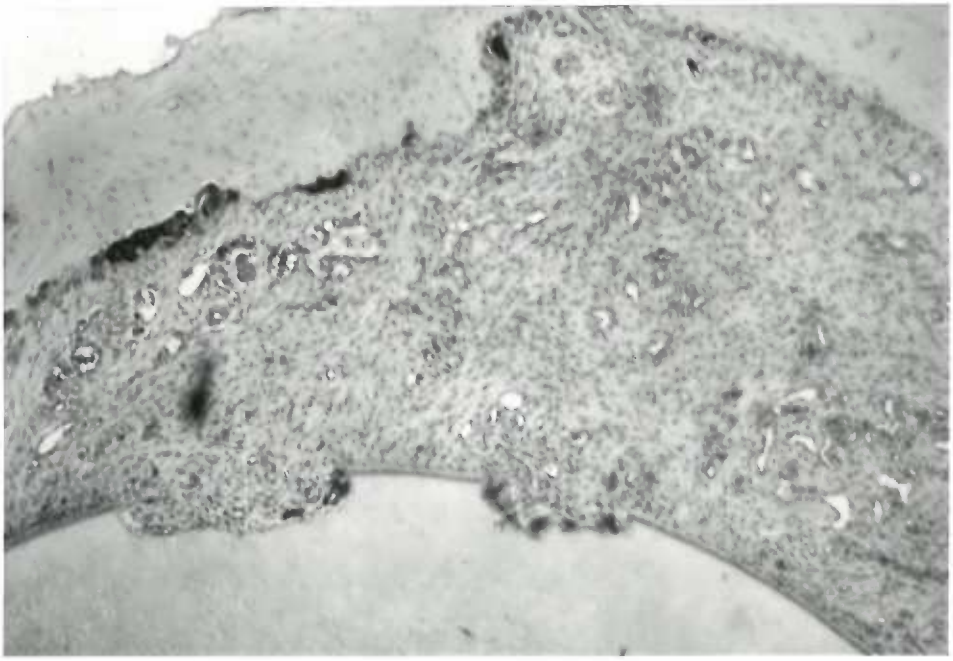

Fig. 7 Lateral Root Resorption and Frontal Resorption of Alveolar Bone as Seen in a Section Apical to a Cell-free Area of the PDL.

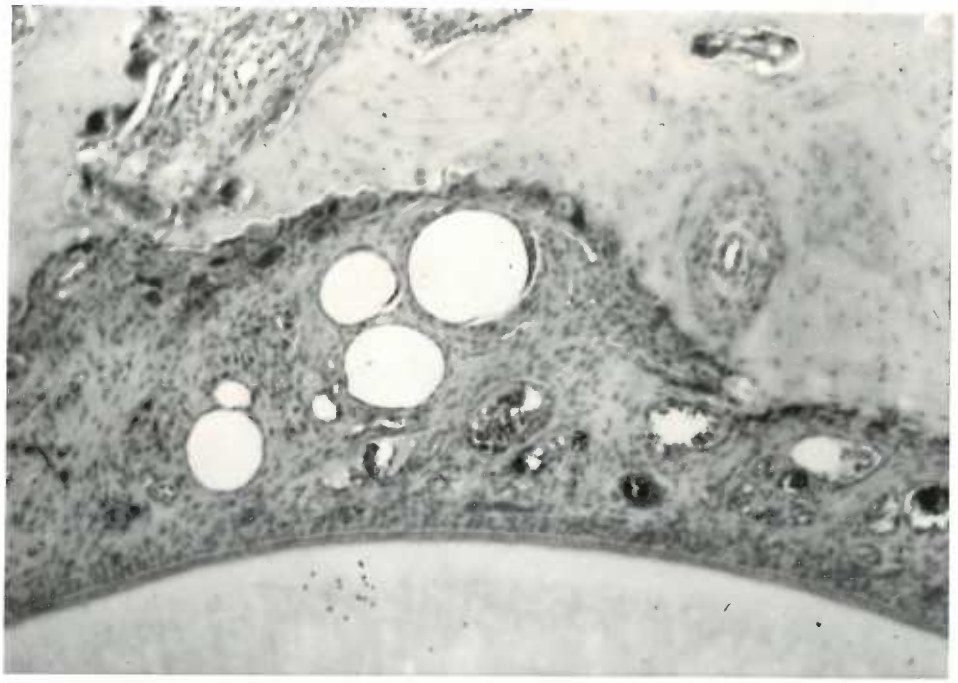

Fig. 8 Area of Pressure Showing "Angioma-like Spaces". Note also Finger of Undermining Resorption from Adjacent Marrow Space.

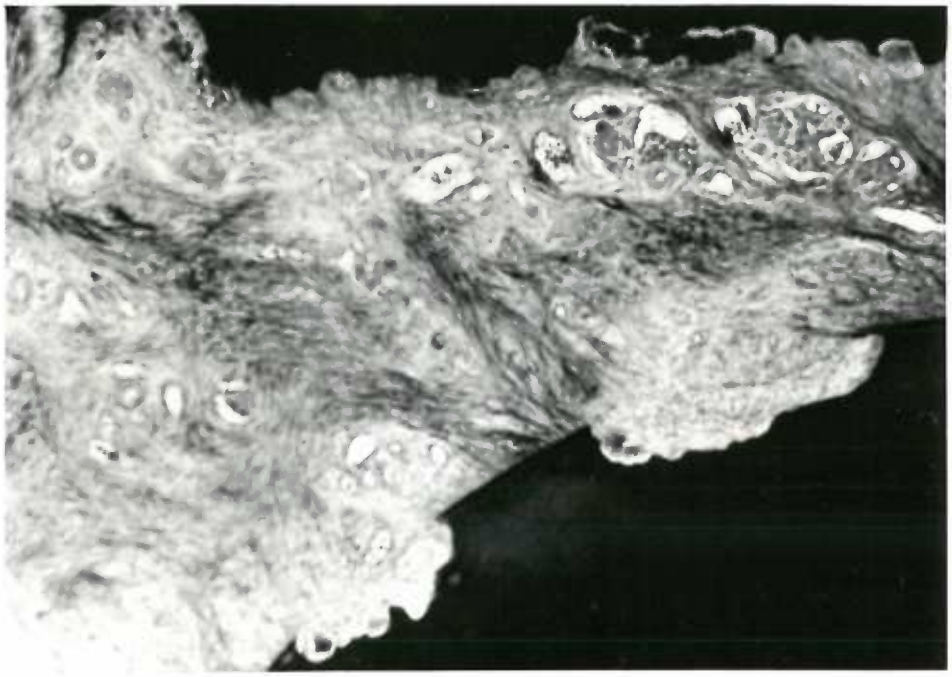

Fig. 9 Acid-Picro Mallory Stain Showing Reorganization of the PDL. Note the Disorganized Fiber Bundles, Capillary Budding, Frontal and Lateral Resorption of Alveolar Bone and Root Resorption.

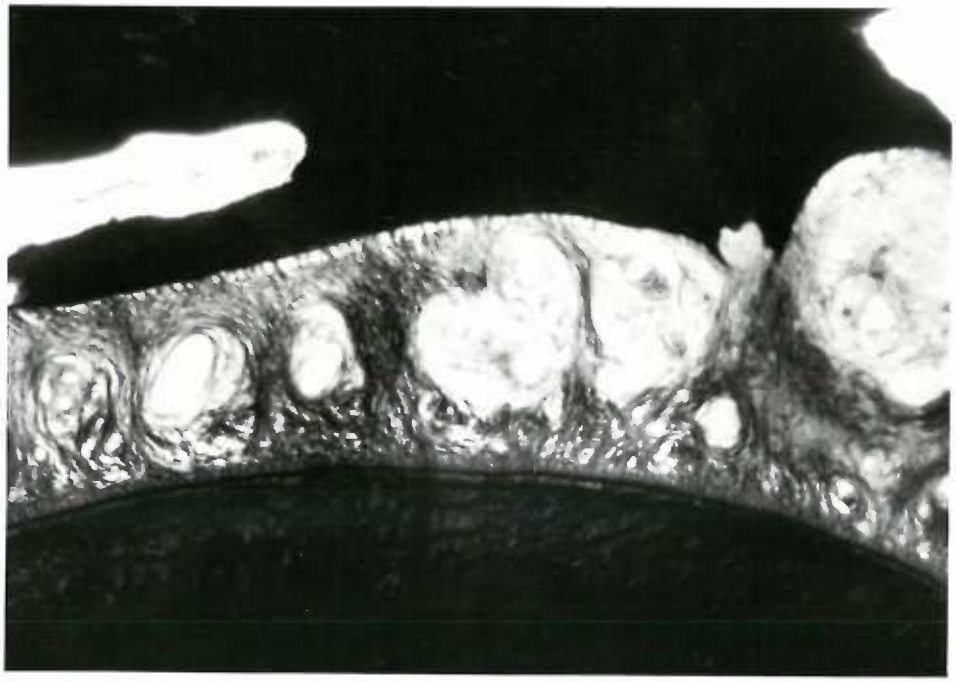

Fig. 10 Acid-Picro Mallory Stain Demonstrating Glomera of the PDL.

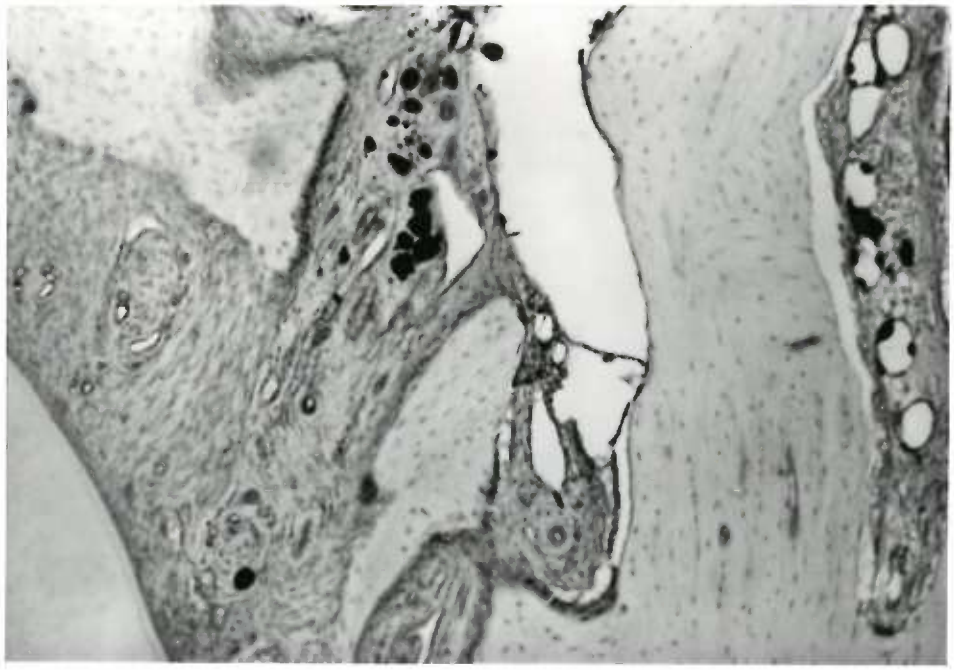

Fig. 11 Sudan Black B Stain Showing the Incorporation of Lipid Material Within the PDL Space Noted by the Presence of Black Areas.

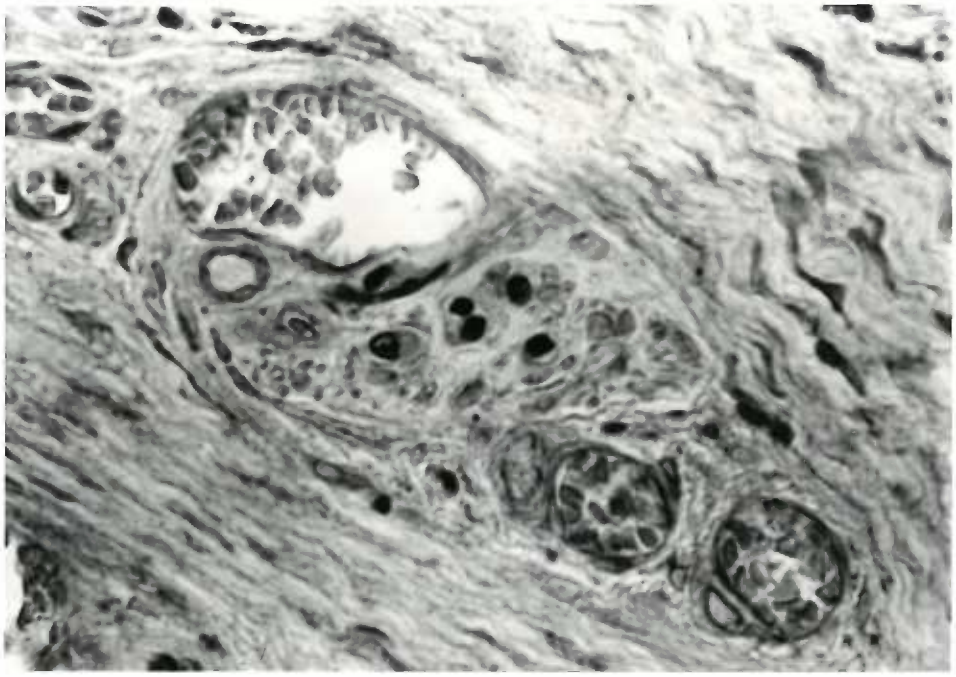

Fig. 12 Sudan Black B Stain of a Neurovascular Bundle of the PDL. Note the Black Areas Indicating the Staining of the Lipid Material Within the Myelinated Nerve Sheath.

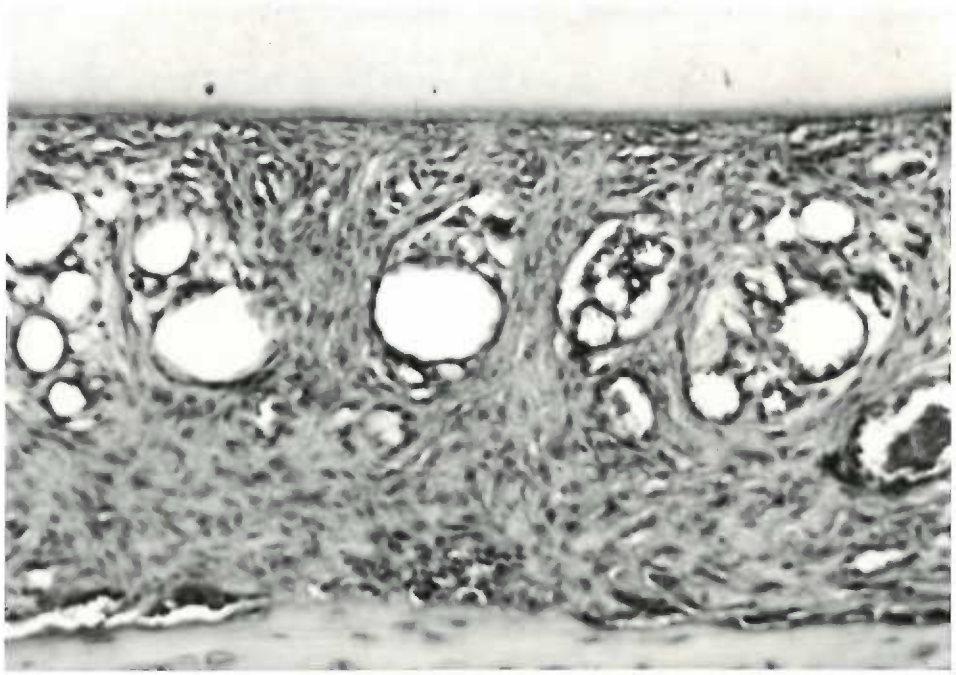

Fig. 13 Shrinking and Tearing Artifacts as Noted with the Original H & E Staining Method.

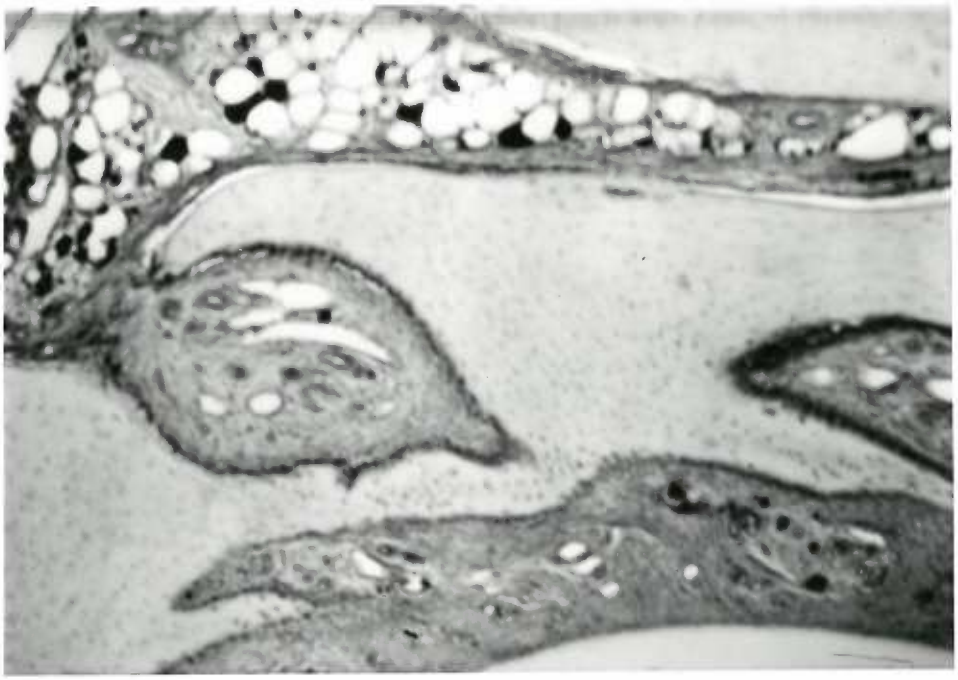

Fig. 14 Sudan Black B Stain Showing a Fat-filled Marrow Space. Note that not All Fat Cells are Positively Stained for Lipid Material.