Remodelling of Collagen Fibers In the Periodontal Ligament and the Supra-Alveolar Region

W. BEERTSEN D.D.S., Ph.D.

It is generally assumed that a stable position of the teeth in the dental arch after orthodontic tooth movement can only be established when the connective tissues of the periodontium have been allowed to adapt to the newly created situation. If adaptation does not occur, the teeth may show a tendency to return toward their original positions after removal of the orthodontic appliance. To decrease relapse, many authors recommend overcorrection of the teeth and the use of retention appliances for a considerable period of time to enable the periodontal tissues to readjust themselves. Since it is thought that, in response to certain types of orthodontic tooth movement, it is especially the supra-alveolar structures that contribute to the relapse, some authors also advocated the removal of the interdental gingiva by means of a simple gingivectomy or cutting the transseptal fiber system which interconnects adjacent teeth in the dental arch.10,21 Thompson38 was able to show that, after orthodontic tooth movement in monkeys, gingivectomy may indeed reduce relapse to a considerable extent. Similar observations have been described by other authors.5,11

Reitan was one of the first who attempted to study the mechanisms underlying rearrangement of the periodontal tissues after orthodontic tooth movement. Examination of the periodontal tissues of the dog immediately after rotation of a tooth revealed a marked displacement and stretching of fibrous structures in the free gingival fiber group, thereby demonstrating resistance against rotational forces.²⁴

After rotation a number of teeth were maintained in position by means of a retentive appliance. According to Reitan no rearrangement of fibrous structures had taken place in the marginal region of the periodontium after retention periods of four weeks. Some of the gingival fiber bundles remained displaced and stretched even after a retention period of 33 weeks. The fiber groups of the periodontal ligament, however, underwent readjustment at a very rapid rate. They were shown to exhibit their normal pattern of organization within a period of four weeks. These observations strongly suggest that where soft connective tissues serve as an attachment, no rapid rearrangement of displaced fibers occurs.

Analyzing various factors that are possibly responsible for differences in adaptability of the collagen component between the gingiva and the periodontal ligament, several questions emerge. Can differences in the rate of collagen turnover explain the remarkable difference in overall behaviour between gingiva and periodontal ligament? Or are there specific sites within the periodontal ligament that have a relatively high remodelling capacity, thereby enabling rapid rearrangement of the collagenous framework? Could it be that rearrangement of periodontal collagen fibers is influenced by remodelling processes of alveolar bone to which the fibers are attached?

It should be stated at the outset that little is known about the factors that control rearrangement of collagen fibers in the periodontal tissues in response to both experimentally induced and physiological tooth movements. However, a number of studies on collagen metabolism have recently been described in the literature which deal mainly with physiological processes in the periodontium and may relate

From the Department of Periodontology, School of Dentistry, University of Amsterdam, The Netherlands.

Presented before the European Orthodontic Society, The Hague, 1978.

to the questions mentioned above. For the sake of brevity and clearness the present paper is mainly limited to a discussion of these studies. It was not intended to provide a comprehensive review of the literature dealing with changes in the periodontium in response to orthodontic tooth movement.

COLLAGEN TURNOVER IN THE PERIODONTIUM

The collagenous framework of the periodontium constitutes a major component of this connective tissue. It occupies almost sixty percent of the volume of the periodontal ligament and the gingiva. Using radioactively labeled amino acids, many authors have attempted to determine the rate at which the collagen fibers in these tissues are synthesized and degraded. The majority of the studies reported on this subject have been based on the use of radioautography. By using this histological method it appears that, shortly after injection of a radioactive protein precursor such as tritiated proline, the label is predominantly located in the connective tissue cells, whereas a few hours later most of it is found in the extracellular space. 35 Thereafter, the labeling intensity decreases gradually as a result of breakdown processes. From various radioautographic studies it appears that turnover of ³Hproline-containing proteins in the periodontal ligament, a substantial part of which is collagen, occurs at a rapid rate. Half-lives from 2 - 12 days have been reported by a number of authors in various animal species.3,7,23,26,27 However, a problem in radioautographic studies of collagen metabolism in periodontal tissues is the lack of sufficient specificity for collagen of the labeled amino acid used.^{3,33} Although it is possible to localize radioactive proteins in radioautographs, it is not possible to determine their chemical nature. Therefore, one should be cautious with the interpretation of data obtained from the radioautographic studies referred to in the foregoing.

Few authors have studied turnover of collagen in the periodontal ligament by using biochemical methods. Guis¹⁵ and Slootweg,³² in studies of the guinea pig incisor, found that the half-life of collagen in the ligament is about 12 days which is considerably less than the half-life of collagen in the skin of the same animal (43 days). That collagen in the periodontal ligament may have a very high turnover rate has also been demonstrated by Sodek³³ in a biochemical study of rat molars.

Several authors have attempted to compare the rate of collagen turnover in the connective tissue of the gingiva with that in the periodontal ligament. Skougaard and co-workers,31 in a radioautographic investigation of marmosets, found that incorporation of ³H-proline in gingival connective tissue was only half of that in the periodontal ligament. A similar observation has been reported by Kameyama¹⁶ for the rat molar. This could, at first glance, suggest a lower metabolic activity in the gingiva than in the periodontal ligament. However, Skougaard and colleagues noted that the rate at which the injected amino acid disappeared from the tissues was about the same in gingiva and periodontal ligament. Thus, it is conceivable that the higher incorporation of ³H-proline in the ligament was related to a higher concentration of proline-containing proteins in this tissue. Also Engstrom and Minkoff¹² concluded from a radioautographic study of mouse molar periodontium that a rapid turnover of proline-containing proteins occurs in both the periodontal ligament and the supracrestal region. Sodek,33 in a biochemical study of rat molars, found that the rate of collagen turnover in attached gingiva, though much higher than in skin corium, was substantially lower than in periodontal ligament. However, since in the latter investigation no distinction was made between the attached gingiva and the gingiva in the marginal region, it is still not clearly understood how turnover of collagen in the supracrestal region relates to that in the periodontal ligament.

When all these studies are considered together, it appears that the turnover rate of collagen in both periodontal ligament and gingiva is relatively high as compared with other connective tissues, such as skin corium. In the periodontal ligament the rate of collagen turnover is possibly somewhat higher than in the supra-alveolar region. It is questionable, however, whether this difference is the only factor that may explain why the periodontal ligament seems much more capable of remodelling than the gingiva in the supra-alveolar region.

Several authors have attempted to study, radioautographically, metabolic activity in the periodontal ligament in response to experimental tooth movement. Crumley8 has observed that incorporation of ³H-proline on the tension side of the ligament of orthodontically moved rat molars was only slightly increased as compared with the control teeth. Baumrind and Buck2 noticed an initial decline in the uptake of radioactively labeled amino acid both on the tension and pressure sides of the ligament in the rat molar. Such an initial decline was not observed by Diaz,9 also in a study of rat molars. Since no biochemical studies of collagen metabolism have been described in the literature. we may conclude that it is still not entirely clear whether and to what extent orthodontic tooth movement influences collagen metabolism in the periodontal tissues.

SITE OF COLLAGEN TURNOVER

Although collagen turnover in periodontal soft connective tissues apparently occurs at a rapid rate, the question remains whether it occurs throughout the entire soft connective tissue compartment or in specific sites with a relatively fast turnover enabling certain areas of the periodontium to undergo rapid rearrangement. It has been suggested that, in the periodontal ligament, the site of collagen rearrangement is located in a so-called intermediate

plexus, 29,30 Within this plexus, collagen fiber bundles anchored in the cementum and collagen bundles anchored in the alveolar bone are thought to split, together forming a meshwork of delicate fibers which continuously become rearranged. thereby allowing the tooth to change its position. Although there is some evidence in support of the view that an intermediate plexus exists in the periodontal ligament of continuously erupting teeth.3 it is unlikely that a similar structure is present in the ligament of teeth of limited eruption. Several authors have argued that in teeth of limited eruption the periodontal ligament exhibits two compartments, one related to the tooth and the other to the alveolar bone. In the bone half of the ligament a higher uptake of ³H-proline has been observed than in the tooth half. 1,8,9,35 However, it is not clear from these studies whether this incorporation was related to protein synthesis in the periodontal ligament or to synthesis of bone matrix. From a quantitative radioautographic study of the mouse molar³⁴ it appeared that, under normal functional demands, the rates of protein synthesis as revealed by incorporation of ³H-proline, were similar among the bone and tooth halves of the periodontal ligament. In the latter study, care was taken by the authors to determine only the fibroblastic contribution to matrix formation in the periodontal ligament.

Some authors have attempted to determine the site of collagen turnover in a somewhat different way, namely, by obtaining information on the breakdown of collagen rather than on its synthesis. Morphologic and histochemical studies have indicated that one pathway of collagen resorption may be via phagocytosis of collagen fibrils by connective tissue cells.20,22,36,39 Beertsen and co-workers4 found that in the rodent molar fibroblasts with intracellular collagen fibrils occur more or less uniformly across the width of the periodontal ligament. No major differences were observed among the cemental, middle and alveolar zones. This

would seem to indicate that resorption of collagen in the ligament is evenly distributed across the width of this tissue which is consistent with the observation of Stahl and Tonna³⁴ that, in the mouse molar, incorporation of radioactively labeled proline is similar between the bone and tooth halves of the ligament.

On the basis of these findings it may be concluded that, under physiological conditions, resorption and probably also synthesis of collagen occur evenly throughout the periodontal ligament of teeth of limited eruption.

With regard to the supra-alveolar structures, it may be of interest to mention that Ten Cate et al.³⁷ noted the presence of a significant number of fibroblasts containing phagocytosed collagen fibrils in the lower part of the transseptal fiber system of mouse molars. However, as far as I am aware, no studies have been described in the literature providing quantitative data with respect to collagen metabolism at various sites in the supra-alveolar region.

REMODELLING OF SOFT CONNECTIVE TISSUE FIBERS AND ALVEOLAR BONE

As mentioned before, Reitan²⁴ noted that the collagen fibers in the periodontal ligament, but not in the gingiva, undergo rapid reorientation after orthodontic rotation. Tentatively, it may be suggested that the observed difference in behaviour is related to the circumstance that in the gingiva the majority of the collagen fibers are probably not attached to bone whereas in the periodontal ligament they insert in the bone of the tooth socket.²⁴

Many authors have described changes in the supporting periodontal tissues in response to orthodontic tooth movement. It has been reported that in areas of compression a process of hyalinization occurs in the periodontal ligament.^{6,17,18,25,28} The hyalinized tissue is resorbed by activity of the cells and subsequently replaced by granulation tissue. It is thought that the tissue will then be restructured according

to the functional demands made upon the teeth in their newly occupied positions.

On the basis of data reported in the literature it seems that mechanisms of readjustment of periodontal ligament may exist which are based on a combined action of fibroblasts in the ligament and bone cells along the wall of the alveolar bone. Several authors have described morphologic changes in the periodontal ligament of animals held under experimental conditions, changes that could possibly be related to altered metabolic processes along the alveolar bone. Melcher and Correia19 have shown that, during reactivated eruption of rat molars, remodelling in the ligament may particularly involve the bone half of the tissue. To check for eruption, these authors made a wound penetrating the alveolar process and the periodontal ligament. It is conceivable that both wounding and eruption could have influenced metabolic activity in the periodontal tissues, particularly along the surface of the alveolar crypt. The observation of Melcher and Correia, in a way, agrees with observations reported earlier by Goldman14 and by Waerhaug.40 The latter author observed that in monkeys maintained on a vitamin C-deficient diet, destruction of periodontal ligament fibers occurred. This destruction was particularly evident in the bone half of the ligament. Similar changes have been described by Goldman in the periodontal ligament of rat molars and in the ligament of teeth of monkeys maintained on a protein-deficient diet. Both Goldman and Waerhaug also observed signs of an increased resorption of bone along the wall of the tooth socket. That resorption of collagen fibers in the periodontal ligament may be related to breakdown processes along the alveolar bone is further suggested by recent electron microscopic studies. Garant, 13 in a study of collagen phagocytosis by fibroblasts in the periodontal ligament of the rat molar, noted that fibroblasts in the vicinity of osteoclasts contain increased numbers of intracellular collagen fibrils. A

similar observation has been reported by Beertsen and co-workers⁴ who quantitatively studied the distribution pattern of collagen phagocytosis by fibroblasts, also in the ligament of the rat molar. This finding may suggest that a relationship exists between the resorptive activity of fibroblasts in the periodontal ligament and that of osteoclasts along the alveolar bone.

In addition to this possible relationship. radioautographic studies would seem to suggest that a relationship may also exist between bone matrix formation along the alveolar wall and synthetic activity of fibroblasts in the periodontal ligament. Crumley8 in an experimental investigation of tooth movement in the rat observed, on the tension side of maxillary molars, a relatively high uptake of ³H-proline in the periodontal ligament adjacent to the bone. Three days after administration of the label he noted this area still contained the greatest amount of radioactivity while, at the same time, intense labeling was found over the adjoining bone matrix. Similar observations have been reported by Diaz.9

Coordination between the remodelling of alveolar bone and the collagen fibers attached to it could be of considerable significance for the periodontal ligament to readjust itself at a rapid rate without the need for a complete reorganization of collagen fibers throughout the entire tissue. Collagen fibrils, detached from the bone in the course of resorptive activity of both osteoclasts and fibroblasts, may subsequently be reattached to bone and pre-existing periodontal collagen fibers by local synthetic activity of fibroblasts and osteoblasts. It is conceivable that such a local mechanism might contribute to a rapid adaptation of the periodontal ligament fibers to changes in the position of the tooth during physiologic and possibly also during certain types of orthodontic tooth movement. Since in the supra-alveolar region most of the soft connective tissue fibers are not attached to bone, rapid readjustment of collagen fibers in this region may not be accomplished via the mechanism outlined above for the periodontal ligament.

CONCLUSIONS

Although the rate of turnover of collagen in the periodontal ligament may be somewhat higher than in the gingiva, it is doubtful whether this is the only factor that may explain why rearrangement of collagen fibers in the ligament occurs much more rapidly than in the supraalveolar region under certain experimental conditions. It may well be that there are other factors that determine the ability of the periodontal tissues to rearrange themselves. On the basis of the distribution pattern of collagen phagocytosis by fibroblasts and the pattern of incorporation of ³H-proline it seems that turnover of collagen, at least in the periodontal ligament, is evenly distributed across the width of the tissue. However, local variations in the presence of cells containing intracellular collagen fibrils may occur, as indicated by the observation that a relatively high concentration of ingested collagen fibrils is seen in fibroblasts in the direct vicinity of osteoclasts. It is suggested that coordinated action of fibroblasts and osteoclasts near the alveolar bone surface may represent part of a local mechanism through which rapid remodelling of certain areas in the periodontal ligament may be accomplished.

School of Dentistry
University of Amsterdam
Louwesweg 1
Amsterdam, The Netherlands

REFERENCES

- Anderson, A. A.: The protein matrixes of the teeth and periodontium in hamsters: a tritiated proline study. J. Dent. Res. 46:67-78, 1967.
- Baumrind, S., and Buck, D. L.: Rate changes in cell replication and protein synthesis in the periodontal ligament incident to tooth movement. Am. J. Orthod. 57:109-131, 1970.

- Beertsen, W., and Everts, V.: The site of remodelling of collagen in the periodontal ligament of the mouse incisor. *Anat. Rec.* 189:479-498, 1977.
- Beertsen, W., Brekelmans, M., and Everts, V.: The site of collagen resorption in the periodontal ligament of the rodent molar. *Anat. Rec.*, 192:305-318, 1978.
- Brain, W. E.: The effect of surgical transsection of free gingival fibers on the regression of orthodontically rotated teeth in the dog. Am. J. Orthod. 55:50-70, 1969.
- Buck, D. L., and Church, D. H.: A histologic study of human tooth movement. Am. J. Orthod. 62:507-516, 1972.
- Carneiro, J., and Fava de Moraes, F.: Radioautographic visualization of collagen metabolism in the periodontal tissues of the mouse. *Arch. Oral Biol.* 10:833-848, 1965.
- Crumley, P. J.: Collagen formation in the normal and stressed periodontium. *Perio*dontics 2:53-61, 1964.
- Diaz, E: Periodontal ligament collagen response to tooth movement: Histochemical and autoradiographic reactions. Am. J. Orthod. 73:443-458, 1978.
- Edwards, J. G.: A study of the periodontium during orthodontic rotation of teeth. Am. J. Orthod. 54:441-461, 1968.
- A surgical procedure to eliminate rotational relapse. Am. J. Orthod. 57:35-46, 1970.
- Engstrom, T., and Minkoff, R.: Autoradiographic study of protein turnover in the mouse periodontiuim. *J. Dent. Res.* 57 (A):223, 1978 (Abstract).
- Garant, P. R.: Collagen resorption by fibroblasts. A theory of fibroclastic maintenance of the periodontal ligament. *J. Perio*dontol. 47:380-390, 1976.
- Goldman, H. M.: The effects of dietary deprivation and of age on periodontal tissues of the rat and spider monkey. J. Periodontol. 25:87-96, 1954.
- Guis, M. B.: Collagen in the periodontal ligament of the continuously erupting guinea pig incisors. A biochemical study on its presence, properties and metabolism. Thesis, *University of Utrecht*, 1976.
- Kameyama, Y. Autoradiographic study of ³H-proline incorporation by rat periodontal ligament, gingival connective tissue and dental pulp. J. Period. Res. 10:98-102, 1975.
- 17. Kvam, E.: A study of the cell-free zone

- following experimental tooth movement in the rat. *Europ. Orthod. Soc. Trans.* 45:419-434, 1969.
- Cellular dynamics on the pressure side of the rat periodontium following experimental tooth movement. Scand. J. dent. Res. 80:369-383, 1972.
- Melcher, A. H., and Correia, M. A.: Remodelling of periodontal ligament in erupting molars of mature rats. *J. Period. Res.* 6:118-125, 1971.
- Parakkal, P. F.: Involvement of macrophages in collagen resorption. *J. Cell Biol.* 41:345-354, 1969.
- Parker, G. R.: Transseptal fibers and relapse following bodily retraction of teeth: A histologic study. Am. J. Orthod. 61:331-344, 1972.
- Pérez-Tamayo, R.: Collagen resorption in carrageenin granulomas. II. Ultrastructure of collagen resorption. Lab. Invest. 22:142-159, 1970.
- Ramos, A. B., and Hunt, A. M.: Remodelling of the periodontal ligament of the guinea pig molars. In: *The Mechanisms of Tooth Support*. A symposium, 1965. Wright, Bristol, pp. 107-112, 1967.
- Reitan, K.: Tissue rearrangement during retention of orthodontically rotated teeth. *Angle Orthod.* 29:105-113, 1959.
- Reitan, K., and Kvam, E.: Comparative behavior of human and animal tissue during experimental tooth movement. *Angle Orthod.* 41:1-14, 1971.
- Rippin, J. W.: Collagen turnover in the periodontal ligament under normal and altered functional forces. I. Young rat molars. J. Period. Res. 11:101-107, 1976.
- Collagen turnover in the periodontal ligament under normal and altered functional forces. II. Adult rat molars. J. Period. Res. 13:149-154, 1978.
- Rygh, P.: Elimination of hyalinized periodontal tissues associated with orthodontic tooth movement. *Scand. J. Dent. Res.* 82:57-73, 1974.
- Sicher, H.: Bau und Funktion des Fixationsapparates des Meerschweinchenmolaren. Z. Stomatol. 23:580-594, 1923.
- Tooth eruption: the axial movement of continuously growing teeth.
 J. Dent. Res. 21:201-210, 1942.
- 31. Skougaard, M. R., Levy, B. M., and Simpson, J.: Collagen metabolism in skin and periodontal membrane of the mar-

- moset. Scand. J. Dent. Res. 78:256-262, 1970.
- 32. Slootweg, R. N.: Changes of collagen and non-collagenous proteins in the periodontal ligament during acceleration of the eruption. A biochemical and histological investigation of different sectors of the periodontal ligament in the guinea pig incisor. Thesis, University of Utrecht, 1976.
- Sodek, J.: A comparison of the rates of synthesis and turnover of collagen and noncollagen proteins in adult rat periodontal tissues and skin using a micro-assay. Arch. Oral Biol. 22:655-666, 1977.
- 34. Stahl, S. S., and Tonna, E. A.: ³H-proline study of aging periodontal ligament matrix formation: Comparison between matrices adjacent to either cemental or bone surfaces. *J. Periodont. Res.* 12:318-322, 1977.
- 35. Stallard, R. E.: The utilization of ³H-proline by the connective tissue elements of the

- periodontium. Periodontics 1:185-188, 1963.
- Ten Cate, A. R.: Morphological studies of fibrocytes in connective tissue undergoing rapid remodelling. J. Anal. 112:401-414, 1972.
- Ten Cate, A. R., Deporter, D. A., and Freeman, E.: The role of fibroblasts in the remodelling of periodontal ligament during physiologic tooth movement. *Am. J. Orthod.* 69:155-168, 1976.
- Thompson, H. E.: Orthodontic relapses analyzed in a study of connective tissue fibers. Am. J. Orthod. 45:93-109, 1959.
- Usuku, G., and Gross, J.: Morphologic studies of connective tissue resorption in the tail fin of metamorphosing bullfrog tadpole. *Dev. Biol.* 11:352-370, 1965.
- Waerhaug, J: Effect of C-avitaminosis on the supporting structures of the teeth. J. Periodontol. 29:87-97, 1958.