From:
The Dental Institute of Experimental Research and Department of Anatomy,
Dental Faculty,
University of Oslo,
Oslo, Norway

A MICRORADIOGRAPHIC, LIGHT MICROSCOPIC AND ELECTRON MICROSCOPIC STUDY OF THE CEMENTUM FROM DECIDUOUS TEETH OF PIGS

by
RANDI FURSETH

INTRODUCTION

True cementum being part of the attachment apparatus of the teeth, is characteristic for all mammals and certain reptiles (*Kronfeld*, 1938). The structure and development of cementum from different animals have been investigated. *Selvig* (1963, 1964) found that the formation of cementum in mice was initiated by the deposition of crystals around the fibers from the periodontal membrane simultaneously with the appearance of an amorphous ground substance. In bovine teeth as well as in sheep and in rabbit molars a considerable layer of cementum has been found to cover the enamel (*Glimcher*, 1964; *Weinreb* & *Sharav*, 1964; *Mills* & *Irving*, 1967; *Listgarten*, 1968; *Listgarten* & *Kamin*, 1969).

The fine structure of rat cementum in incisors (*Stern*, 1964) as well as in molars (*Lester*, 1969a, b) has been studied, and *Lester* reported the incorporation of epithelial cells in the cementum, particularly at the cemento-dentinal junction. *Boyde* and *Jones* (1968) in a comparative study of cementum observed a densely mineralized zone at the cemento-dentinal junction, and found the walls of the lacunae to be more highly mineralized than the interlacunar cementum.

The structure of human cementum has also been studied by several authors utilizing different techniques (e.g. *Lorber*, 1951; *Haim*, 1961;

Received for publication, May 14, 1970.

Dreyfuss & Frank, 1964; *Selvig*, 1965; *Furseth*, 1967, 1969a; *Furseth & Johansen*, 1968, 1970).

Thus, while human cementum as well as cementum from rats, mice, and several herbivorous animals have been studied, information about pig cementum is scarce. The pig is omnivorous, possesses both deciduous and permanent sets of teeth, and the miniature pig has been found to be well suited as an experimental animal in dental research (*Weaver, Sorenson & Jump*, 1962).

The purpose of the present study was to investigate the structure and mineral distribution of pig cementum to see how it differed from cementum from other species generally and human cementum in particular. Knowledge of the structure and mineral distribution of the normal cementum was also desirable in order to create a basis for evaluation of experimental alterations. Miniature pigs were not available, and pigs of the Norwegian land race were therefore used.

MATERIALS AND METHODS

The material consisted of 80 teeth from 5 pigs of the Norwegian land race ranging in age from 9 to 15 weeks. At this age the functioning dentition consists entirely of deciduous teeth. The formula for the deciduous teeth of the pig is 3 Id, 1 Cd, 3 Md, and is the same for the upper and lower jaw. The third incisor and the canine are cone-shaped. A fourth molar (Md 1) may be missing, or if present, is not erupting. The incisors as well as the molars are teeth of limited growth. It should be kept in mind however, that breeding of pigs has lead to changes in tooth development as well as times of eruption (*Nickel, Schummer & Seiferle*, 1960).

The pigs were killed, the head cut off, and the teeth removed. The teeth which were intended for electron microscopic studies were removed first. Often the teeth were split into two halves, one part being reserved for decalcified and one for undecalcified sections. Moreover, some teeth represented the contralateral side of experimental teeth or areas located apical to the experimental area (to be described subsequently). The teeth were fixed in 2.5 % cacodylate-buffered glutaraldehyde at pH 7.2 or 2 % cacodylate-buffered formalin at pH 7.2 to which had been added 7.5 % sucrose (*Holt & Hicks*, 1961). Phosphate buffered fixatives were not used in order to avoid possible precipitation of atypical crystals (*Furseth*, 1969b).

Light microscopy. Thirty three teeth or parts of teeth, were decalcified for 24—36 hrs in 5.2 % nitric acid, dehydrated in graded alcohols and embedded

in paraffin. The teeth were sectioned on a Leitz sliding microtome and stained with hematoxylin and eosin.

Microradiography and electron microscopy. After fixation, 47 teeth were washed in cacodylate buffer and then placed in 1 % cacodylate-buffered osmium tetroxide at pH 7.2 for two hours. The teeth were dehydrated in graded series of acetone solutions and embedded in Vestopal W (*Ryter & Kellenberger*, 1958).

Ground sections parallel to the long axes of the teeth were cut with a diamond wheel on a Gillings-Hamco Thin Sectioning Machine under constant water spray. The thickness of some of the sections was further reduced by grinding on abrasive papers. The thickness of the sections varied from 50—350 μ with an average of 170 μ.

Contact microradiographs were produced on Kodak Spectroscopic plates 649—0 with a Philips X-ray diffraction unit, type PW 1009, supplied with a fine focus tube. Nickel-filtered copper radiation at 20 kV and 20 mA was employed.

The microradiographs were studied in the light microscope, and parts of 14 ground sections from 11 teeth were re-embedded for electron microscopy. In addition ultrathin sections were also cut from 3 teeth without making microradiographs first.

The ultrathin sections were cut with a diamond knife on an LKB Ultrotome III and floated on 2 % acetone adjusted to pH 7.5 with NaOH. The sections were collected on formvar- and carbon-coated grids and the time during which the sections were in contact with water was reduced as much as possible to avoid dissolution of mineral (*Boothroyd*, 1964).

Some sections from each specimen were floated on a saturated solution of uranyl acetate in 30 % alcohol for 25 min followed by lead citrate (*Reynolds*, 1963) for 5 min, while some sections were examined unstained. Other sections were decalcified on the grid with 1 % phosphotungstic acid (PTA) in 50 % alcohol or 2 % PTA in distilled water. The decalcification time varied from 15 to 30 min. The sections were examined in a Siemens Elmiscope Ia electron microscope operated at 80 kV. Selected-area electron diffraction was carried out using 200 μ condenser and 50 μ diffraction apertures.

FINDINGS

Light microscopy

The cementum was chiefly of the cellular variety, (Figs. 1, 2, 4—6), but areas with acellular cementum were also observed in a few instances in the molars (Figs. 1, 3). The width of the cementum layer varied with the different teeth

PLATE I

Fig. 1. Photomicrograph from incisor showing the cemento-enamel junction. The cementum (C) overlaps the enamel for a short distance, and it is mainly cellular, but areas of acellular cementum can also be noted. Pocket epithelium (PE), dentine (D). Hematoxylin and eosin. × 75.

Fig. 2. Photomicrograph from molar showing wide layer of cellular cementum. Cementocyte nuclei are present in most of the lacunae. A cavern (K) in the cementum containing several cells can be noted. Periodontal membrane (PM), cemento-dentinal junction (CDJ). Hematoxylin and eosin. × 75.

Fig. 3. Photomicrograph from molar, showing hematoxyphilic zone at the cemento-dentinal junction (CDJ). The cementum is mainly acellular. Incremental lines located close together can be noted in the outer part of the cementum. Periodontal membrane (PM). Hematoxylin and eosin. × 790.

Fig. 4. Photomicrograph from molar. A narrow hematoxyphilic zone is seen at the cemento-dentinal junction (CDJ). The outermost layer of the dentine stains lighter than the main part of the dentine. Cementocyte nuclei are present in all the lacunae. Close to the cementum surface several clusters of epithelial cells (EC) can be noted. Hematoxylin and eosin. × 220.

Fig. 5. Photomicrograph from molar showing a wide cementum layer and a few incremental lines. The continuity between the periodontal and Sharpey's fibers can be noted. Cemento-dentinal junction (CDJ). Hematoxylin and eosin. × 220.

Fig. 6. Photomicrograph from incisor showing light staining zone in the dentine abutting the cemento-dentinal junction (CDJ). Cementocytes can be seen in all the cementum lacunae, and there is a wide layer of precementum (PC). Clusters of epithelial cells (EC) partly embedded in the cementum can be noted. Hematoxylin and eosin. × 350.

PLATE II

Fig. 7. Microradiograph from molar showing the distribution of cementum (C) in the cervical area. The cementum is cellular, and is fairly thin close to the cemento-enamel junction while it increases in thickness in the apical direction. Cemento-dentinal junction (CDJ), enamel (En). × 280.

Fig. 8. Microradiograph from molar. A fairly X-ray dense zone can be noted at the cemento-dentinal junction (CDJ). Next to the cemento-dentinal junction a layer of acellular cementum with a fairly low mineral content can be noted followed by a layer of cellular cementum with a somewhat higher mineral content. × 60.

Fig. 9. Microradiograph from incisor showing the cemento-enamel junction and enamel pearl. The cementum (C) is entirely cellular and overlaps the enamel (En) for a short distance. Cemento-dentinal junction (CDJ). × 60.

Fig. 10. Microradiograph from incisor showing a wide layer of cellular cementum with numerous lacunae. Giant lacunae or caverns (K) can also be noted. Cemento-dentinal junction (CDJ). × 80.

Fig. 11. Microradiograph from molar showing a wide layer of cellular cementum and several caverns close to the cementum surface. Cemento-dentinal junction (CDJ). × 80.

Fig. 12. Microradiograph from the furcation area of molar. A radiodense zone can be noted at the cemento-dentinal junction (CDJ). The cementum is entirely cellular and several caverns (K) can be noted, particularly close to the surface. × 30.

Fig. 13. Microradiograph from molar showing cellular cementum with numerous lacunae (L). The walls of the lacunae seem to be mineralized to the same degree as the remaining cementum. × 150.

PLATE III

Fig. 14. Electron micrograph from incisor showing electron-dense layer at the cemento-dentinal junction. Cementum (C), dentine (D). No staining. × 6700.

Fig. 15. Electron micrograph from molar showing parts of two cementum lacunae (L) approximately 15 μ from the cementum surface. The walls of the cementum lacunae have the same electron density as the interlacunar cementum. No staining. × 15,600.

PLATE I

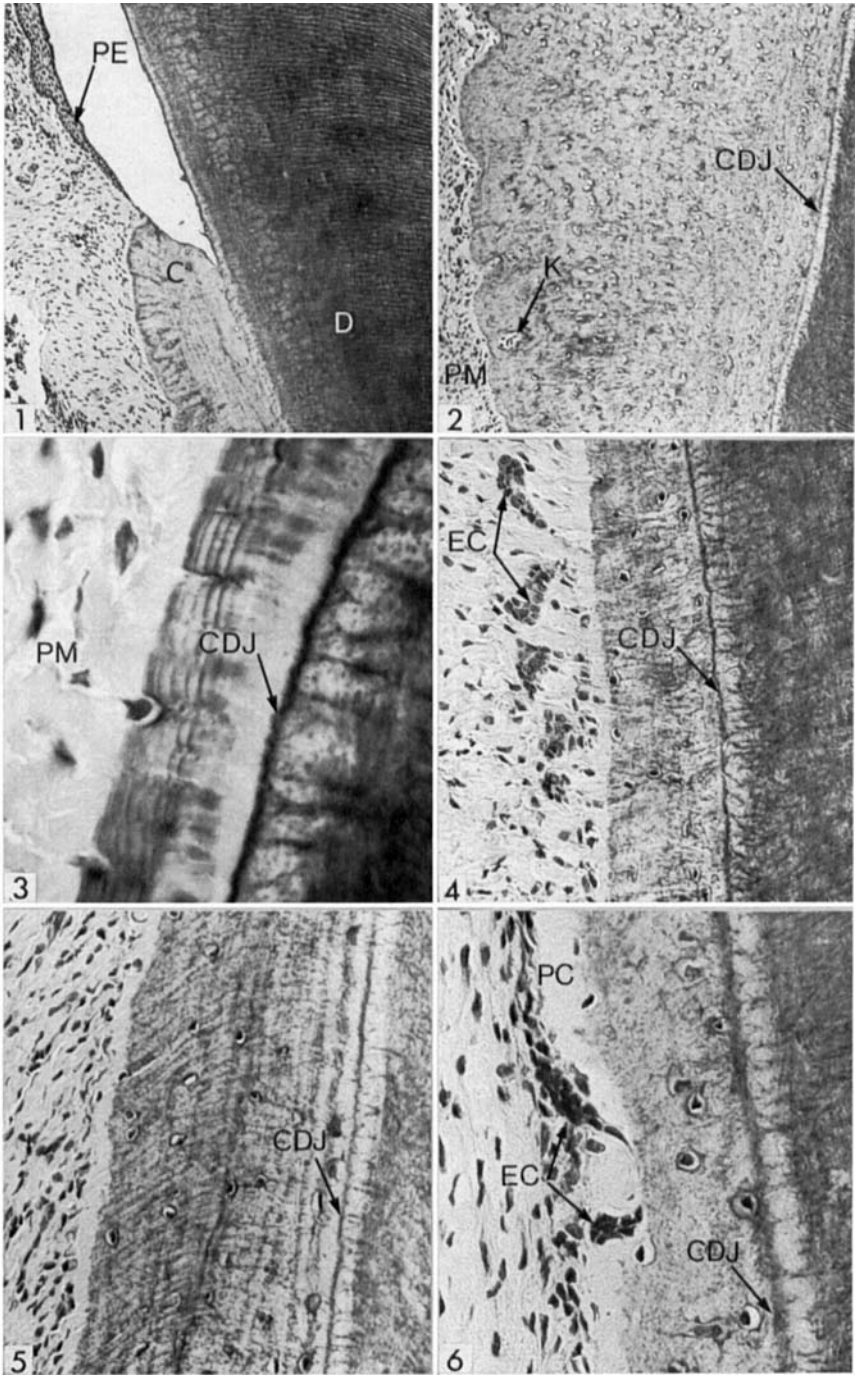


PLATE II

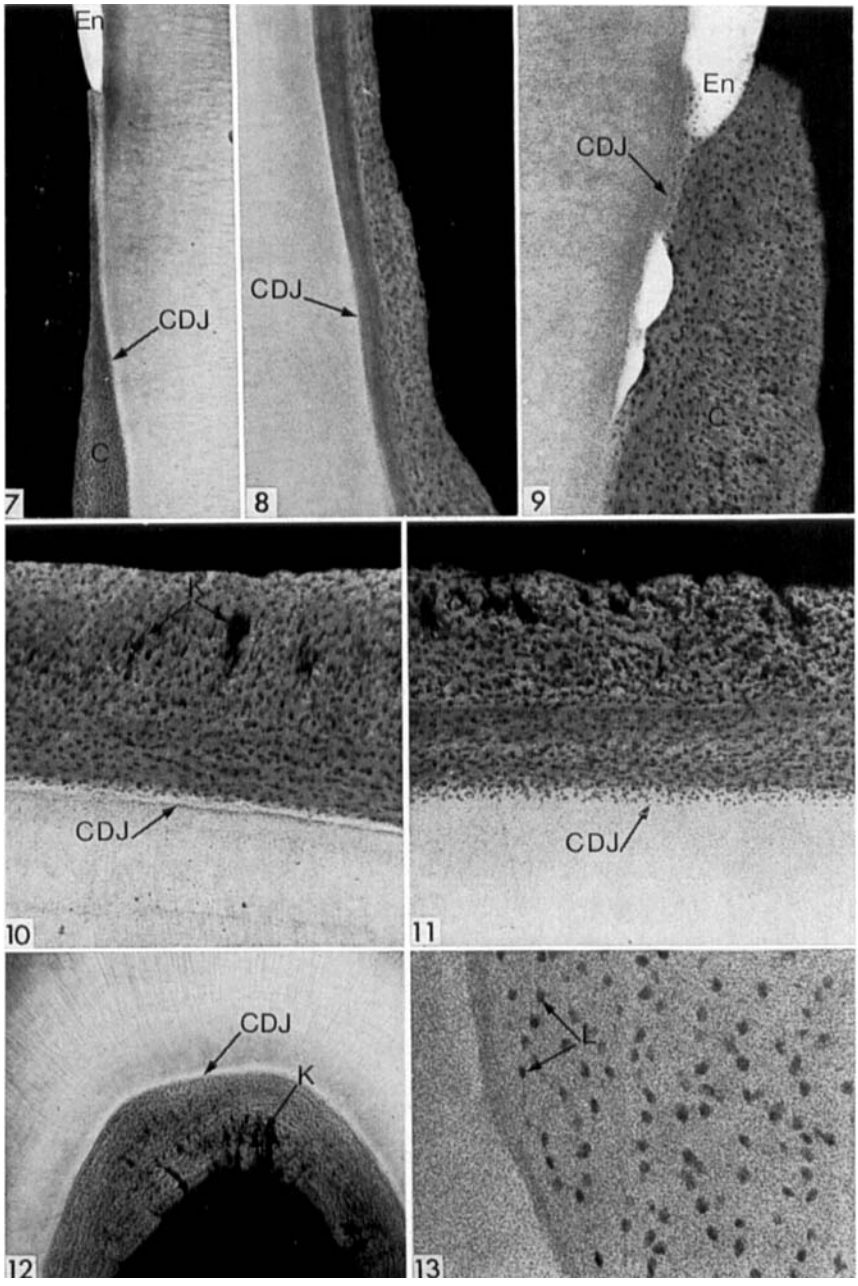
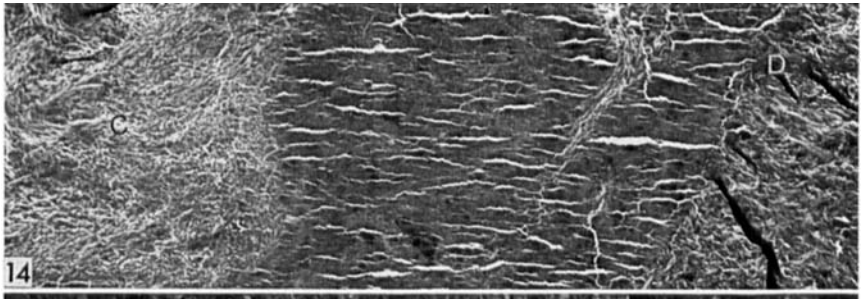
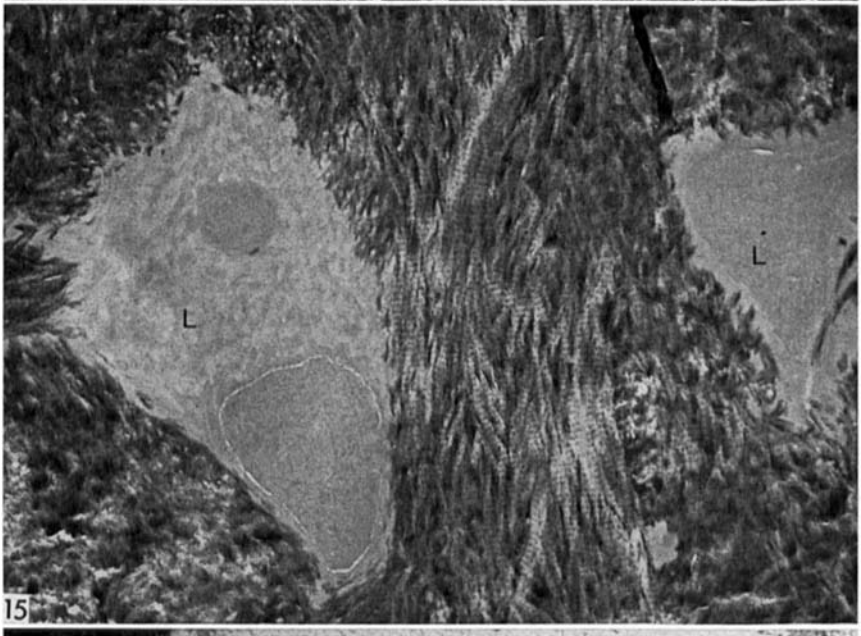


PLATE III





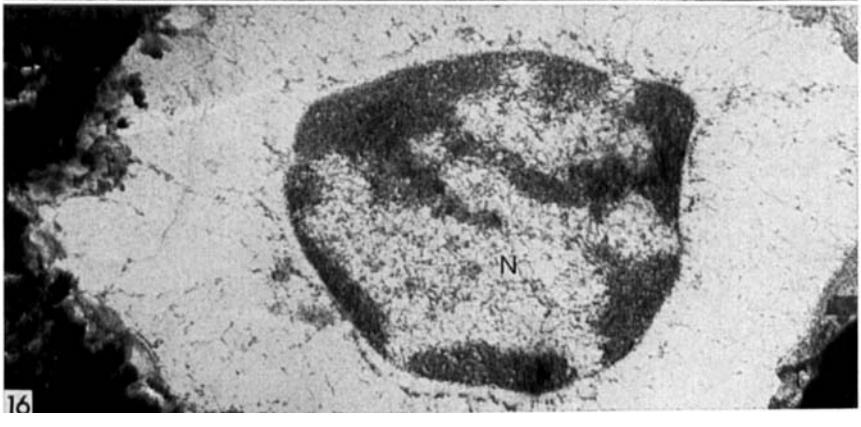
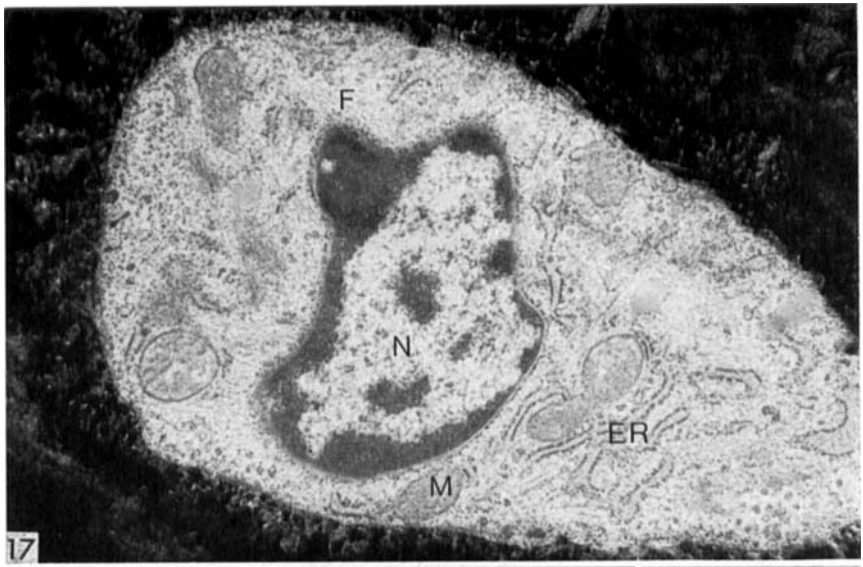
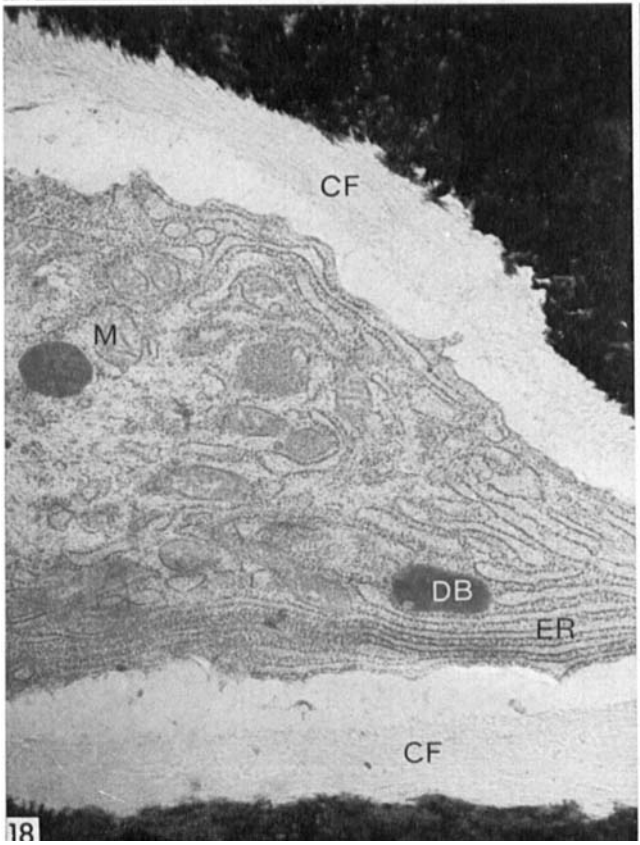


PLATE IV





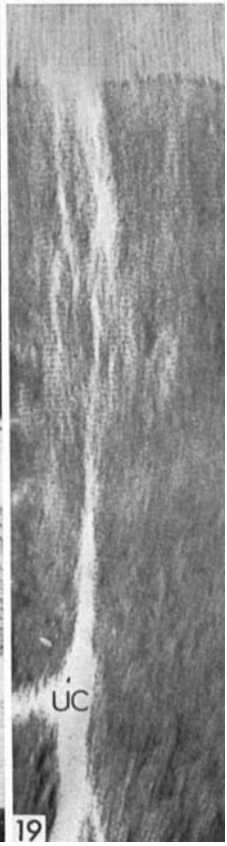


PLATE V

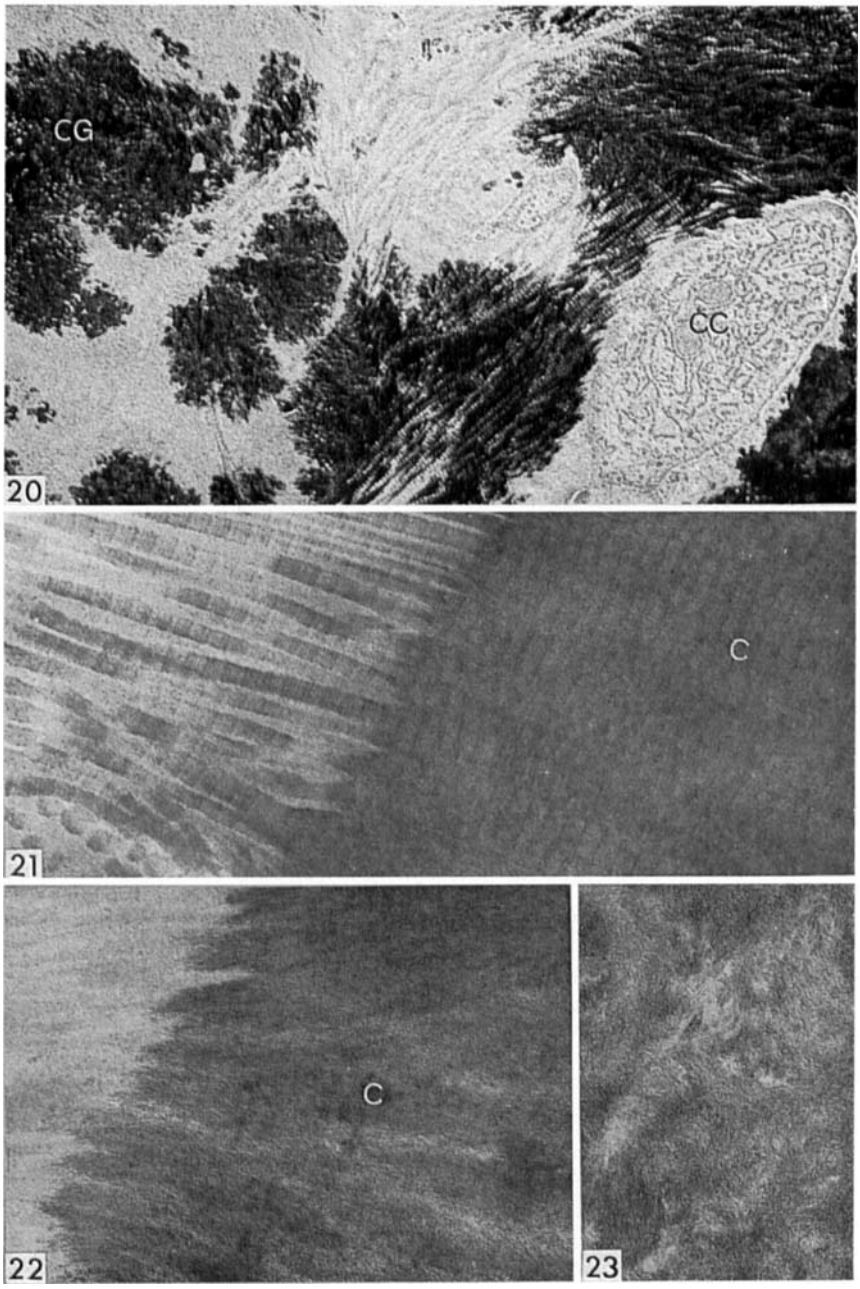


PLATE VI

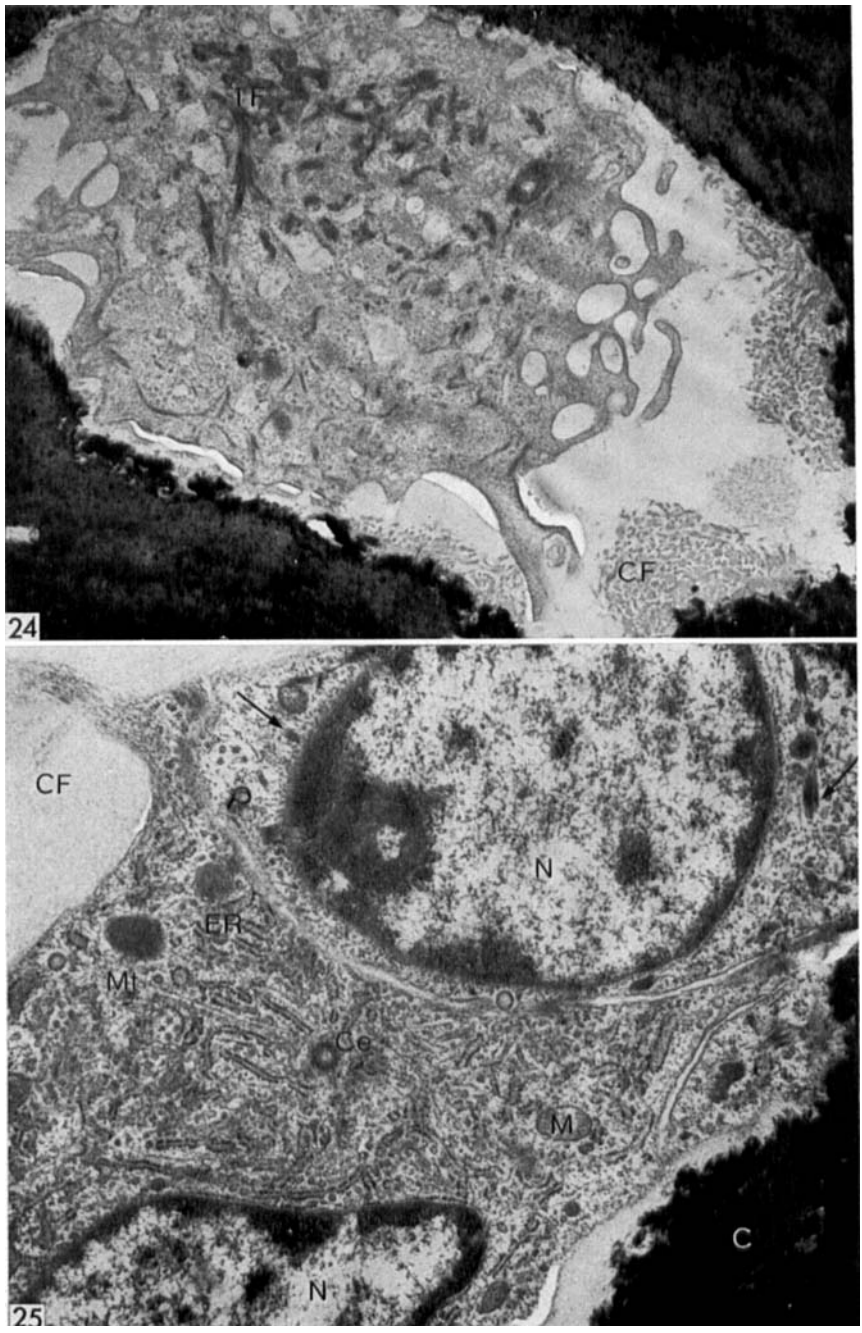


PLATE VII

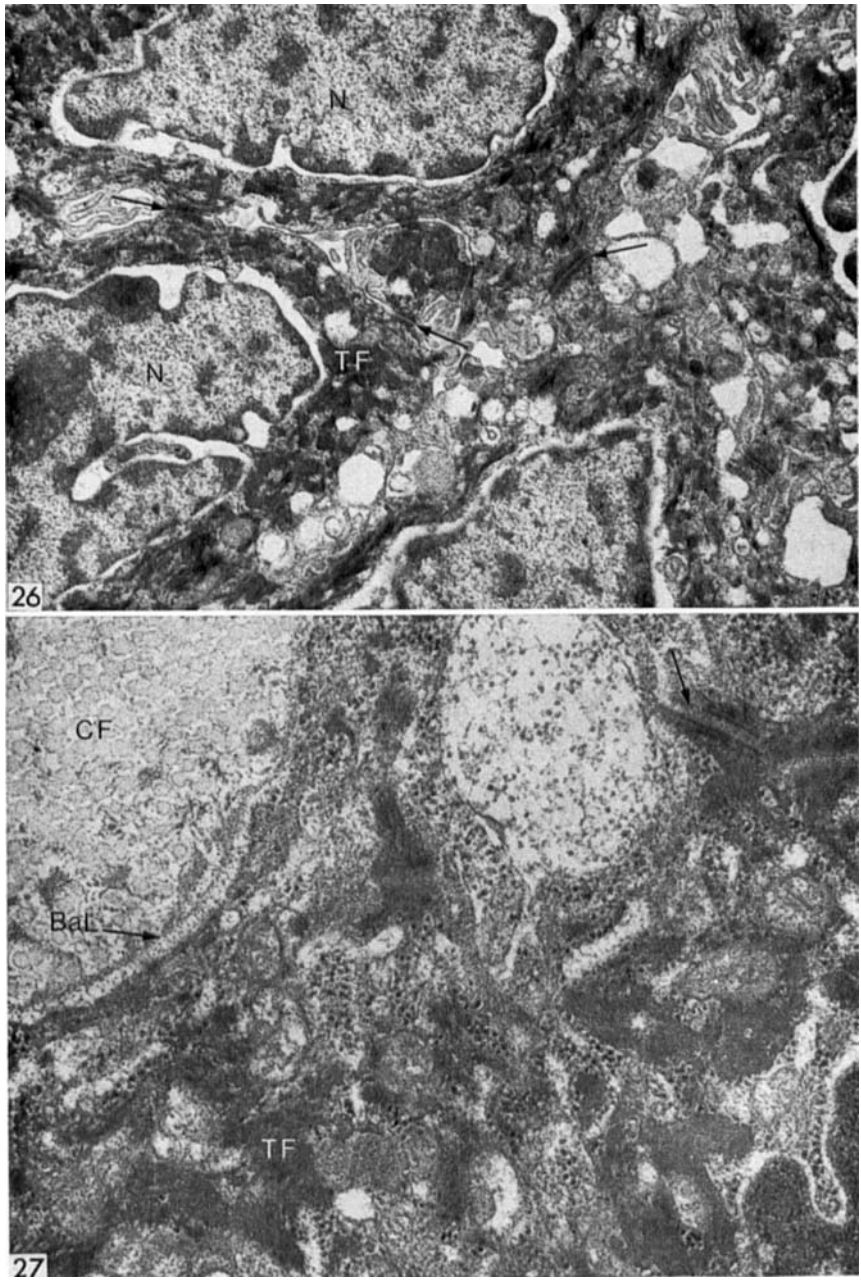


Fig. 16. Electron micrograph from molar showing cementocyte approximately 75 μ from the cementum surface. The nucleus (N) is fairly well preserved while only scattered filaments are seen in the cytoplasm. Lead citrate, uranyl acetate. × 26,700.

PLATE IV

Fig. 17. Electron micrograph from molar showing cementocyte from an area close to the cementum surface which occupies the entire lacuna in the plane of section. Mitochondria (M), endoplasmic reticulum (ER), fine fibrillar material (F), nucleus (N). Lead citrate, uranyl acetate. × 16,300.

Fig. 18. Electron micrograph from molar showing cementocyte located approximately 20 μ from the cementum surface. An abundant endoplasmic reticulum (ER), several mitochondria (M), and dense bodies (DB) are seen in the cytoplasm. The lacunar wall is lined by a layer of unmineralized collagen fibers (CF). Lead citrate, uranyl acetate. × 17,000.

Fig. 19. Electron micrograph from incisor showing unmineralized core (UC) in Sharpey's fiber in the cementum. No staining. × 13,600.

PLATE V

Fig. 20. Electron micrograph from the apical portion of molar showing several calcific globules (CC) in the precementum and part of cementocyte (CC). Lead citrate, uranyl acetate. × 11,200.

Fig. 21. Electron micrograph from incisor showing periodontal fibers entering the cementum (C). The cementum surface has a serrated appearance. The crossbanding of the collagen fibers is clearly visible. The section has been treated with PTA. × 47,300.

Fig. 22. Electron micrograph from molar showing the outer layer of the cementum (C). The cementum surface has a serrated appearance and the crystals in the surface layer seem to be oriented with the long axes parallel to the long axes of the collagen fibers. No staining. × 42,600.

Fig. 23. Electron micrograph from molar showing small, irregularly oriented crystals from an area in the surface layer. No staining. × 47,300.

PLATE VI

Fig. 24. Electron micrograph from incisor showing epithelial cell embedded in the cementum. Bundles of tonofilaments (TF), unmineralized collagen fibers (CF). Lead citrate, uranyl-acetate. × 13,300.

Fig. 25. Electron micrograph from molar showing parts of two cells embedded in the cementum close to the surface. Small bundles of tonofilaments (arrows) are seen in the cytoplasm of one of the cells, indicating that it is of epithelial origin. The other cell contains an abundant endoplasmic reticulum (ER), some mitochondria (M), but no tonofibrils. Nucleus (N), centriole (Ce), microtubuli (Mt), unmineralized collagen fibers (CF), cementum (C). Lead citrate, uranyl acetate. × 17,800.

PLATE VII

Fig. 26. Electron micrograph from incisor showing part of cluster of epithelial cells located approximately 25 μ from the cementum surface. The cytoplasm contains bundles of tonofilaments (TF) and several desmosomes (arrows) are present between the cells. Nucleus (N). Lead citrate, uranyl acetate. × 17,500.

Fig. 27. Electron micrograph from molar showing parts of epithelial cells located in the periodontal membrane. Characteristic dense bundles of tonofilaments (TF) giving the cytoplasm a dark texture are seen. The cells are surrounded by a basement lamina (BaL). Desmosome (arrow), collagen fibers (CF). Lead citrate, uranyl acetate. × 43,700.

and the stage of development. In most of the teeth studied the root development had not been completed. Wide cementum layers were generally seen, but in the cervical regions of the molars thin cementum layers were also noted. The cementum stained lighter than the dentine with the exception of a narrow zone of dentine bordering the cemento-dentinal junction (Figs. 1—6). A hematoxyphilic line was seen at the cemento-dentinal junction (Figs. 3—6). Generally, incremental lines were absent, but they were observed in a few areas in some specimens (Figs. 3, 5).

The cementum often overlapped the enamel for a short distance, particularly in the incisors, and the pocket epithelium could be seen to cover the enamel for some distance (Fig. 1). In some specimens the continuity between periodontal and Sharpey's fibers could be observed (Figs. 3—5). The appearance of the cementocytes varied. In some instances cementocyte nuclei seemed to be well preserved (Figs. 4, 6), while in other specimens the nuclei seemed pycnotic, and empty lacunae were sometimes observed (Fig. 2). However, there was a tendency for the cementocytes in the surface to be better preserved than those in the deeper layers. Caverns or giant lacunae in the cementum containing more than one cell were also observed (Fig. 2).

Quite often epithelial rests were seen close to the cementum surface. These were characterized by the fact that the cells were located close together and had a cytoplasm which stained darker than the surrounding cells (Fig. 4). In a few instances epithelial rests partly embedded in the cementum were seen (Fig. 6).

Microradiography

Also with this method it was evident that the cementum was almost exclusively of the cellular variety. In a few instances however, acellular cementum was observed, particularly in the coronal root portion of the molars (Fig. 8). The acellular cementum seemed to have the same or a slightly lower degree of mineralization than the interlacunar matrix of the cellular cementum.

Figure 7 demonstrates the distribution of cementum in the cervical area of a lower third molar. Close to the cemento-enamel junction the cementum layer was fairly thin, while the cementum thickness increased in the apical direction. Enamel pearls were also observed in some specimens (Fig. 9).

In some areas an X-ray dense zone was observed at the cemento-dentinal junction (Figs. 10, 12) while in other areas this zone was lacking (Figs. 11, 13). Highly mineralized incremental lines could only be discerned in a few instances.

Cementum lacunae were quite numerous (Figs. 9—13), and in some

specimens caverns or giant lacunae were also observed (Figs. 10—12). The walls of the cementum lacunae seemed to be mineralized to the same degree as the remaining bulk of the cementum (Fig. 13).

Electron microscopy

At the cemento-dentinal junction an electron-dense zone was often observed while the adjacent cementum and dentine had a lower electron density (Fig. 14). The walls of the lacunae showed the same degree of electron density as the surrounding cementum (Fig. 15).

The appearance of the cementocytes varied, but generally the impression was gained that the cementocytes in the surface layer had the smallest nuclear/cytoplasmic ratio. In the deeper cementum layers alterations in the cells could sometimes be noted. While the nuclei were fairly well preserved, only scattered filaments were seen in the cytoplasm (Fig. 16). The endoplasmic reticulum appeared dilated in some cementocytes (Figs. 17, 20), while in others no marked dilations were observed (Figs. 18, 25). A layer of unmineralized collagen fibers lining the walls of the lacunae was often observed (Fig. 18), but lacunae where the cementocytes seemed to fill the entire unmineralized space in the plane of section were also seen (Fig. 17). The cementocytes, particularly those close to the surface, as well as the cementoblasts, contained an abundant rough-surfaced endoplasmic reticulum, several mitochondria, a well developed Golgi apparatus, and a few dense bodies (Figs. 18, 25). A centriole and microtubuli were also observed (Fig. 25).

In the cervical area the cementum surface often had a serrated appearance with an even mineralization front (Figs. 21, 22). Small, plate-like crystals were present in the surface layer (Figs. 22, 23). Selected-area electron diffraction showed that they were hydroxyapatite. The crossbanding of the collagen fibers was clearly visible in PTA-decalcified sections. After the periodontal fibers had entered the cementum, the crossbanding was more diffuse (Fig. 21). Unmineralized areas in the cementum which might represent unmineralized cores of Sharpey's fibers extending for some distance into the tissue were occasionally seen (Fig. 19). In the apical area the cementum surface had a more irregular appearance with calcific globules in the precementum (Fig. 20).

Epithelial rests were seen close to the cementum surface. The epithelial cells were located close together, and the clusters of epithelial cells were surrounded by a basement lamina. The cells were characterized by dense aggregates of tonofilaments in the cytoplasm and several desmosomes were seen between the individual cells (Figs. 26, 27). Epithelial cells, as indicated

by their bundles of tonofilaments in the cytoplasm, were also found to be embedded in the cementum (Figs. 24, 25). Some of the cells demonstrated only a few bundles of tonofilaments (Fig. 25), while in others the tonofibrils were more abundant (Fig. 24). Unmineralized collagen fibers were also observed in the lacunae containing epithelial cells (Fig. 24).

DISCUSSION

In the teeth of mammals the distribution of cementum shows great variation. The extremes are represented by the herbivorous animals where the entire tooth is covered with a fairly thick layer of cementum, and the carnivorous animals where the relatively thin layer of cementum is confined to the roots (*Kronfeld*, 1938). The pig is omnivorous, and the cementum of the pig seems to represent somewhat of a transition between the two other types, i.e. consisting of a fairly thick layer which is largely limited to the root.

At the cemento-dentinal junction a hematoxyphilic zone was generally observed which made it easy to distinguish between cementum and dentine. This was not the case in human deciduous teeth, where the location of the cemento-dentinal junction could be difficult to determine in areas with cellular cementum (*Furseth*, 1967). The inconsistent presence of an X-ray dense zone at the cemento-dentinal junction agrees with findings in cellular cementum in human teeth (*Dreyfuss & Frank*, 1964; *Furseth*, 1967; *Furseth & Johansen*, 1968). Incremental lines with a high mineral content are a characteristic feature of cementum from human permanent teeth (*Soni, Van Huysen & Swenson*, 1962; *Yamamoto et al.*, 1962; *Selvig, 1965*; *Furseth & Johansen*, 1968). In this respect the pig cementum appeared more related to cellular cementum of human deciduous teeth (*Furseth*, 1967) and repair tissue in deciduous teeth (*Furseth*, 1968) which have a more uniform mineral distribution.

The walls of the cementum lacunae were mineralized to the same degree as the surrounding tissue both as judged microradiographically and electron microscopically. This confirms previous findings in human material, (*Furseth*, 1967; *Furseth*, 1969a; *Furseth & Johansen*, 1968), but disagrees with findings by *Awazawa* (1963) and *Boyde* and *Jones* (1968). This apparent disagreement may however be due to a difference in age of the teeth examined. These factors have been discussed elsewhere (*Furseth*, 1969a).

Radiolucent caverns or giant lacunae were observed in the cementum of the pig, and similar observations have been made in human material (*Dreyfuss & Frank*, 1964; *Furseth*, 1967; *Furseth & Johansen*, 1968). In

a few instances acellular cementum was observed, and this was sometimes less mineralized than the cellular cementum, a fact which is not in accordance with observations made in human material (*Soni et al.*, 1962; *Selvig*, 1965; *Furseth*, 1967). The occasional finding of unmineralized areas believed to represent unmineralized cores of Sharpey's fibers, is in agreement with observations made by *Selvig* (1965) and *Boyde* and *Jones* (1968).

As judged in the light microscope, the appearance of the cementocytes in the different layers of the cementum varied. Generally the cementocytes were better preserved here than those observed in a previous study of human material (*Furseth*, 1967). It should be kept in mind however, that cementum formation in the pig occurs quite rapidly, and therefore the cells would not have been embedded for a long period of time. Also, cementum lacunae are much more numerous in the pig, a fact which might make the passage of nutrients easier. The vitality of the cementocytes is a controversial question. The literature on this subject and the influence of factors like ageing of the cementocytes, poor nutritional conditions for the cells in the deeper layers, and possible fixation artifacts have been discussed in detail previously (*Furseth*, 1967). *Erausquin* and *Muruzábal* (1967) found that the vitality of normal rat molar cementum decreased with age, and senile necrosis in the zone adjacent to the dentine was an almost constant finding after the seventh month. *Listgarten* and *Kamin* (1969) observed the development of lysosome-like bodies and autophagic vacuoles in the cementocytes when the cementum became displaced occlusally by continuous eruption. These authors pointed out that the association of lysosomes and particularly autophagic vacuoles with degenerating cells has been described previously by *Ashford* and *Porter* (1962) and *Novikoff* and *Essner* (1962). In the present study dense bodies were observed in the cementocytes, but they were not particularly numerous.

The cementocytes generally had a fairly small amount of cytoplasm, particularly those which were somewhat removed from the surface, and this agrees with previous findings in human material (*Furseth*, 1967, 1969a) as well as findings in the cellular cementum of rabbit molars (*Listgarten & Kamin*, 1969). This indicates that the cementocytes, once embedded in the cementum have a low activity. However, cementocytes with a fairly abundant endoplasmic reticulum were also seen occasionally, particularly close to the surface. As seen in deciduous teeth of humans (*Furseth*, 1967) cementocytes with altered cytoplasm were also seen in the pig.

Epithelial rests within the periodontal membrane have been described in several species. *Valderhaug* and *Nylen* (1966) described epithelial rests in human material and concluded that on the basis of the ultrastructural

morphology and histochemical evidence the term, resting epithelial cells was an appropriate description for these cells. *Grant* and *Bernick* (1969) found a great prevalence of epithelial rests in the periodontium of miniature swine and this agrees with the present findings.

The epithelial rests were surrounded by a basement lamina and were characterized by numerous desmosomes and bundles of tonofilaments in the cytoplasm and this is in accordance with observations made by other investigators (*Valderhaug & Nylen*, 1966; *Lester*, 1969a,b; *Listgarten & Kamin*, 1969). The bundles of tonofilaments were useful in order to distinguish between epithelial cells and cementocytes when individual cells were observed in lacunae. In the epithelial rests observed in the periodontal membrane spaces were noted between the cytoplasm and the nucleus. This may be a preparation artifact, but it may also indicate degenerative changes, since a dilation of the nuclear sac, together with thicker and more prominent bundles of tonofilaments were some of the degenerative changes noted in incorporated epithelial cells by *Lester* (1969a).

Incorporation of epithelial cells in the cementum were observed, and this is in accordance with findings in other species (*Paynter & Pudy*, 1958; *Diab & Stallard*, 1965; *Grant & Bernick*, 1969; *Lester*, 1969a,b; *Listgarten & Kamin*, 1969).

In rat molars epithelial cells have particularly been observed in the first formed cementum (*Diab & Stallard*, 1965; *Lester*, 1969a,b) and *Lester* reported incorporation of epithelial cells en masse between cementum and dentine at the advancing root edge. This developmental event was coincident with an alteration in the rate of cementum and dentine formation, cellular cementum coming both to precede and to be a greater bulk than dentine at the advancing root edge. This type of root formation was not seen in the pig, and incorporation of single epithelial cells or groups of cells were seen in different layers of the cementum.

Unmineralized collagen fibers were observed in several of the lacunae, and this confirms previous observations in human material (*Furseth*, 1967; *Furseth*, 1969a). Autoradiographic studies by *Frank* and *Frank* (1969) have shown labelling of unmineralized collagen fibers in osteocyte lacunae, showing that collagen is produced after the incorporation of the cells in the calcified tissues. Unmineralized collagen fibers were also seen in lacunae which contained epithelial cells. Since epithelial cells presumably do not produce collagen, this indicates that the unmineralized collagen at least in these lacunae was formed prior to the mineralization of cementum, even if the possibility that epithelial sheath cells may produce collagen cannot be entirely excluded (*Lester*, 1969b).

As mentioned previously pig cementum differed from human cementum in some respects, particularly by being mainly cellular also in the cervical area. The embryological development of the pig is very comparable to that found in man, the pig is omnivorous, and the oral glands are similar to what is observed in man (*Weaver et al.* 1962). The pig is therefore considered to be a suitable experimental animal.

Acknowledgement. The author is greatly indebted to professor, dr. philos. Odd Skjerven and his staff, Department of Obstetrics, The Veterinary College of Norway, Oslo, Norway, for providing housing for the animals and for their help with anesthesizing the animals.

SUMMARY

The structure and mineral distribution of the cementum from deciduous teeth of pigs have been studied. The cementum was chiefly of the cellular variety, and the cementum layer was quite wide. Incremental lines were generally absent, but in some areas an X-ray dense zone was observed at the cemento-dentinal junction. The cementum often overlapped the enamel for a short distance, particularly in the incisors. Cementum lacunae were quite numerous and the walls of the lacunae were mineralized to the same degree as the interlacunar areas. The appearance of the cementocytes varied, but generally the cells close to the surface were better preserved than the cells in the deeper layers. Caverns or giant lacunae in the cementum containing more than one cell were also observed.

Quite often epithelial rests were seen close to the cementum surface. These were surrounded by a basement lamina and the cells were characterized by dense bundles of tonofilaments in the cytoplasm and numerous desmosomes. Epithelial rests partly embedded in the cementum as well as single epithelial cells in lacunae were also seen.

A layer of unmineralized collagen fibers lining the walls of the lacunae was often observed. The cementum contained small, plate-like crystals. Selected-area electron diffraction showed that they were hydroxyapatite.

RÉSUMÉ

ÉTUDE PAR MICRORADIOGRAPHIE, AU MICROSCOPE OPTIQUE ET AU MICROSCOPE ÉLECTRONIQUE DU CÉMENT PROVENANT DE DENTS TEMPORAIRES DE PORCS.

Une étude a été entreprise sur la structure et la répartition des substances minérales dans le cément de dents temporaires de porcs. Le cément était principalement un cément cellulaire, et la couche cémentaire était assez large.

Les lignes incrémentales manquaient en général, mais en certaines parties, une zone radio-opaque était observée à la jonction cémento-dentinaire. Le cément recouvrait souvent légèrement l'émail, particulièrement dans les incisives. Les lacunes du cément étaient assez nombreuses; le degré de minéralisation de leur parois était le même que celui des zones interlacunaires. L'apparence des ostéocytes variait, mais les cellules proches de la surface étaient en général mieux conservées que celles des couches profondes. On observait aussi dans le cément des cavités ou des lacunes géantes contenant plus d'une cellule.

Assez souvent on voyait des débris épithéliaux auprès de la surface du cément. Ces débris étaient entourés d'une lame basale; les cellules étaient caractérisées par des faisceaux denses de tono-fibrilles dans le cytoplasme et par de nombreux desmosomes. On observait aussi des débris épithéliaux inclus en partie dans le cément, ainsi que des cellules épithéliales isolées dans des lacunes.

On observait souvent une couche de fibres collagènes non minéralisées tapissant les parois des lacunes. Le cément contenait de petits cristaux en forme de plaques. La diffraction electronique au niveau de zones déterminées a montré qu'il s'agissait d'hydroxyapatite.

ZUSAMMENFASSUNG

EINE MIKRORADIOGRAPHISCHE, LICHTMIKROSKOPISCHE UND ELEKTRONENMIKROSKOPISCHE UNTERSUCHUNG DES ZEMENTES DER SCHWEINEMILCHZÄHNE

Die Struktur und Mineralverteilung des Zementes von Schweinemilchzähnen wurden untersucht. Das Zement war hauptsächlich vom zellhaltigen Typ, und die Zementschicht ziemlich breit. Wachstumslinien gab es gewöhnlich keine. Eine röntgendichte Zone wurde jedoch in einigen Gebieten nahe der Zement-Dentingrenze beobachtet. Das Zement überlagerte oft den Schmelz ein Stückchen, besonders an den Schneidezähnen. Zementlakunen gibt es zahlreich, ihre Wände waren ebenso mineralisiert wie die interlakunären Gebiete. Die Zementozyten zeigten verschiedenes Aussehen und die Zellen nahe der Oberfläche waren allgemein besser erhalten als die Zellen tiefer im Zement. Es wurden auch Kavernen oder Riesenlakunen die mehr als eine Zelle enthielten im Zement beobachtet.

In der Nähe der Zementoberfläche konnten Epitelreste öfters gesehen werden. Die Epithelreste waren von einer Basalmembran umgeben, und die Zellen wurden durch dichte Tonofilamentbündel im Zytoplasma und zahlreiche Desmosome gekennzeichnet. Epithelreste, die teilweise im Zement

eingelagert waren, und einzelne Epithelzellen in Zementlakunen wurden auch beobachtet.

An den Wänden der Zementlakunen gab es oft eine Schickt von nicht-mineralisierten Kollagenfasern. Das Zement enthielt kleine, plattenähnliche Kristalle. Es wurde mittels Elektronendiffraktion in ausgewählten Gebiete gezeigt, dass es sich um Hydroxylapatitkristalle handelte.

REFERENCES

Ashford, T. P. & K. R. Porter., 1962. Cytoplasmic components in hepatic cell lysosomes. J. Cell Biol. *12:* 198—202.

Awazawa, Y., 1963. Electron microscopy of human cementum. J. Nihon Univ. Sch. Dent. *5:* 127—137.

Boothroyd, B., 1964. The problem of demineralisation in thin sections of fully calcified bone. J. Cell Biol. *20:* 165—173.

Boyde, A. & S. J. Jones, 1968. Scanning electron microscopy of cementum and Sharpey fibre bone. Z. Zellforsch. mikrosk. Anat. *92:* 536—548.

Diab, M. A. & R. E. Stallard, 1965. A study of the relationship between epithelial root sheath and root development. Periodontics *3:* 10—14.

Dreyfuss, F. & R. Frank, 1964. Microradiographie et microscopie électronique du cément humain. Bull. Grpmt int. Rech. scient. Stomat. *7:* 167—181.

Erausquin, J. & M. Muruzábal, 1967. Necrosis of cementum induced by root canal treatments in the molar teeth of rats. Archs oral Biol. *12:* 1123—1132.

Frank, R. M. & P. Frank, 1969. Autoradiographie quantitative de l'ostéogenèse en microscopie électronique à l'aide de la proline tritiée. Z. Zellforsch. mikrosk. Anat. *99:* 121—133.

Furseth, R., 1967. A microradiographic and electron microscopic study of the cementum of human deciduous teeth. Acta odont. scand. *25:* 613—645.

—»— 1968. The resorption processes of human deciduous teeth studied by light microscopy, microradiography and electron microscopy. Archs oral Biol. *13:* 417—431.

—»— 1969a: The fine structure of the cellular cementum of young human teeth. Archs oral Biol. *14:* 1147—1158.

—»— 1969b: The occurrence of atypical crystals in human cellular cementum fixed in phosphate-buffered fixatives. Archs oral Biol. *14:* 1419—1427.

Furseth, R. & E. Johansen, 1968: A microradiographic comparison of sound and carious human dental cementum. Archs oral Biol. *13:* 1197—1206.

—»— 1970: The mineral phase of sound and carious human dental cementum studied by electron microscopy. Acta odont. scand. *28:* 305—322.

Glimcher, M. J., U. A. Friberg, & P. T. Levine, 1964: The identification and characterization of a calcified layer of coronal cementum in erupted bovine teeth. J. Ultrastruct. Res. *10:* 76—88.

Grant, D. A. & S. Bernick, 1969: A possible continuity between epithelial rests and epithelial attachment in miniature swine. J. Periodont. *40:* 87—95.

Haim, G., 1961: Histochemische Untersuchungen des Zementgewebes. Dt. zahnärztl. Z. *16:* 71—81.

Holt, S. J. & R. M. Hicks, 1961: Studies on formalin fixation for electron microscopy and cytochemical staining purposes. J. biophys. biochem. Cytol. *11:* 31—45.

Kronfeld, R., 1938: The biology of cementum. J. Am. dent. Ass. *25:* 1451—1461.

Lester, K. S., 1969a: The incorporation of epithelial cells by cementum. J. Ultrastruct. Res. *27:* 63—87.

—»— 1969b: The unusual nature of root formation in molar teeth of the laboratory rat. J. Ultrastruct. Res. *28:* 481—506.

Listgarten, M. A., 1968: A light and electron microscopic study of coronal cementogenesis. Archs oral Biol. *13:* 93—114.

Listgarten, M. A. & A. Kamin, 1969: The development of a cementum layer over the enamel surface of rabbit molars — a light and electron microscopic study. Archs oral Biol. *14:* 961—985.

Lorber, M., 1951: A study of the histochemical reactions of the dental cementum and alveolar bone. Anat. Rec. *111:* 129—144.

Mills, P. B. & J. T. Irving, 1967: Coronal cementogenesis in cattle. Archs oral Biol. *12:* 929—932.

Nickel, R., A. Schummer, & E. Seiferle, 1960: Lehrbuch der Anatomie der Haustiere. Vol. II. pp. 85—88. Paul Parey, Berlin.

Novikoff, A. B. & E. Essner, 1962: Cytolysomes and mitochondrial degeneration. J. Cell Biol. *15:* 140—146.

Paynter, K. J. & C. Pudy, 1958: A study of the structure, chemical nature, and development of cementum in the rat. Anat. Rec. *131:* 233—251.

Reynolds, E. S., 1963: The use of lead citrate at high pH as an electron-opaque stain in electron microscopy. J. Cell Biol. *17:* 208—212.

Ryter, A. & E. Kellenberger, 1958: L'inclusion au polyester pour l'ultramicrotomie. J. Ultrastruct. Res. *2:* 200—214.

Selvig, K. A., 1963: Electron microscopy of Hertwig's epithelial sheath and of early dentin and cementum formation in the mouse incisor. Acta odont. scand. *21:* 175—186.

—»— 1964: An ultrastructural study of cementum formation. Acta odont. scand. *22:* 105—120.

—»— 1965: The fine structure of human cementum. Acta odont. scand. *23:* 423—441.

Soni, N. N., G. Van Huysen, & H. M. Swenson, 1962: A microradiographic and X-ray densitometric study of cementum. J. Periodont. *33:* 372—378.

Stern, I. B., 1964: An electron microscopic study of the cementum, Sharpey's fibers and periodontal ligament in the rat incisor. Am. J. Anat. *115:* 377—409.

Valderhaug, J. P. & M. U. Nylen, 1966: Function of epithelial rests as suggested by their ultrastructure. J. periodont. Res. *1:* 69—78.

Weaver, M. E., F. M. Sorenson, & E. B. Jump, 1962: The miniature pig as an experimental animal in dental research. Archs oral Biol. *7:* 17—24.

Weinreb, M. M. & Y. Sharav, 1964: Tooth development in sheep. Am. J. vet. Res. *25:* 891—908.

Yamamoto, H., M. Masuda, K. Toeda, & K. Suzuki, 1962: Microradiographic and histopathological study of the cementum. Bull. Tokyo med. dent. Univ. *9:* 139—150.

Address:
Dental Institute of Experimental Research,
University of Oslo,
Blindern, Oslo 3, Norway