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## STUDIES ON MINERALIZED DENTAL TISSUES<sup>1</sup>

### V. Microradiographic investigation of experimentally produced caries and attrition in white rats

by

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In carious lesions in man, there occurs a demineralization of both enamel and dentine. This demineralization can be demonstrated by means of x-ray absorption measurement, i.e. the lesions absorb less radiation than the surrounding normal tissue. In certain parts of the lesions, however, an increased content of mineral salts may be observed (cp. *Bergman & Engfeldt*, 1954). It has been shown by *Van Huysen et al.* (1937), that an increase in x-ray absorption may also result from attrition.

A suitable experimental diet will produce caries in rats. The diet most often used is the one composed by *Hoppert, Webber & Canniff* (1931), which *inter alia* contains coarsely ground maize (HWC diet). Upon this diet ensues, together with caries, attrition of the cusps.

The aim of the present investigation is to study, by means of a microradiographic technique, the changes occurring in enamel and dentine as a result of experimental caries in white rats. In connexion with this investigation the dentinal changes associated with attrition have also been studied.

#### MATERIALS AND METHODS

White rats were used as experimental animals. They were of the same breed as in earlier experiments by *Bergman* (1953). At

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Fig. 1. Microradiogram showing early stage of fissure caries in the enamel with low x-ray absorption near the bottom of the fissure. No visible change in the dentine.  $\times 200$ .

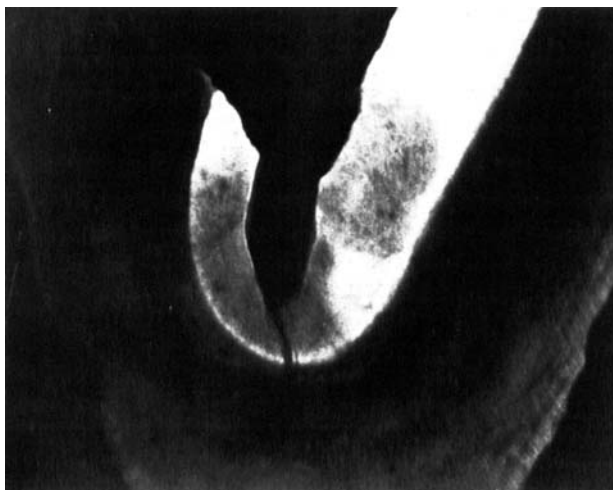


Fig. 2. Microradiogram showing more advanced fissure caries. Lamella or crack in the enamel with surrounding zone with low x-ray absorption.  $\times 100$ .

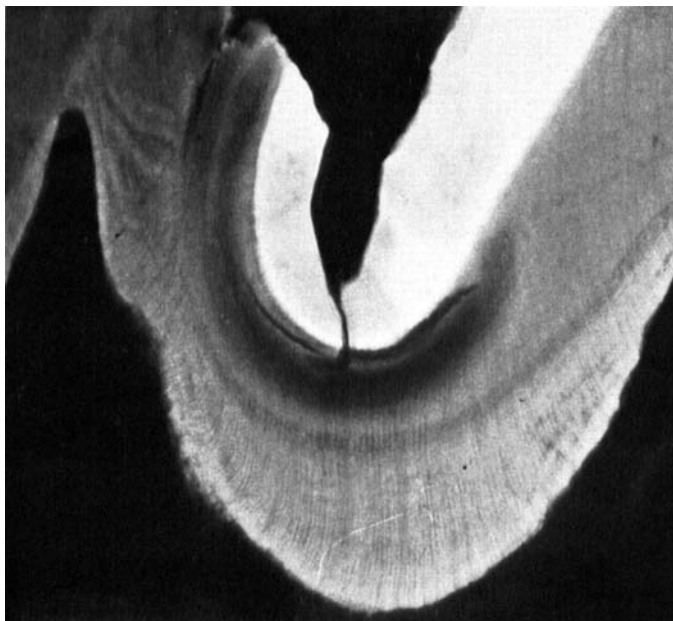


Fig. 3. Microradiogram. Fissure caries in the dentine with low x-ray absorption in a crescent-shaped zone beneath the enamel.  $\times 100$ .

the age of 30 days the rats were put on a HWC diet, and they were killed after 100 days on this diet. Ground sections about  $100 \mu$  in thickness were prepared from the molars in such a way that all three molars in one half of the jaw were cut together (longitudinal sections).

The content and distribution of mineral salts was determined by microradiography. The microradiograms were obtained by placing the ground sections in close contact with a fine-grained photographic emulsion and exposing them to soft x-rays. A Machlett OEG 50 X-ray tube with a 0.2 mm Be window served as radiation source. The tube was operated with 24 kV fully rectified d.c. The microradiograms were taken on Eastman Kodak Spectroscopic Plate No. 649, which has a resolving power of more than  $1 \mu$ . They were at unit magnification and were enlarged by photomicrography on Kodak 0 250 plates. For a general survey and discussion of the technique, see *Bergman & Engfeldt (1954)*.



Fig. 4. Ground section in transmitted light. Note opaque and transparent zones in the abraded cusps.  $\times 40$ .

#### RESULTS

The microradiograms of the molars disclosed areas of fissure caries in our animals (Figs. 1--3). The enamel at the bottom of the fissures showed a lower x-ray absorption than the surrounding normal enamel which indicates a dissolution of mineral salts. This is in agreement with results obtained by *Applebaum & Adams* (1938). In the early phase of fissure caries shown in Fig. 1, the x-ray absorption of the dentine was not appreciably changed, while in more advanced alterations (Fig. 3) a definitely lowered absorption of the dentine beneath the fissure was observed. The carious lesion in this latter instance had spread down to the dentine, the mineral salts of which are being dissolved. In both the above described types of fissure caries no areas with increased x-ray absorption were found in the dentine.

The microradiogram shown in Fig. 6 demonstrates a carious lesion rarely occurring in the white rat, viz. proximal caries in two adjacent teeth. In the left tooth large areas of the enamel

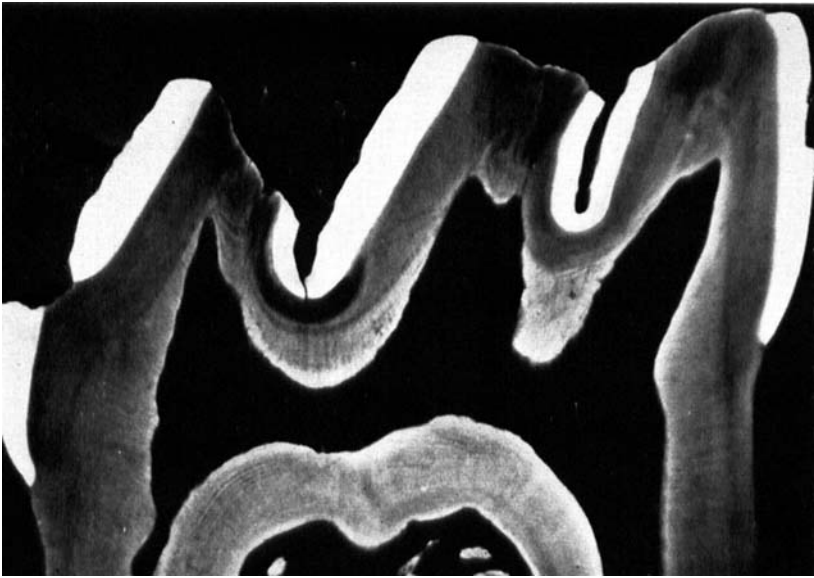


Fig. 5. Microradiogram of the same tooth as in Fig. 4. The cups tips show increased x-ray absorption. No complete correlation between the transparent zones in Fig. 4 and the zones with high absorption in the microradiogram.  $\times 40$ .

and dentine were destroyed. In the right tooth the enamel was apparently normal, while the dentine showed a lowered x-ray absorption in a narrow zone next to the enamel. It seems probable that the section had been cut in such a way that the penetration of the carious lesion through the enamel had taken place in the enamel lying at the side of the section. No areas of increased x-ray absorption of the dentine were found.

The microradiograms of the abraded cusps, however, showed increased x-ray absorption in a narrow zone at the outer surface of the dentine as well as in the deeper parts of the cusps (Fig. 5).

Photomicrographs of the ground sections taken in transmitted light showed both transparent and opaque areas in the cusps. Comparison of the photomicrograph of the ground section (Fig. 4) with the corresponding microradiogram (Fig. 5) shows that there is no clear correlation between opaque and transparent areas, on the one hand, and the distribution of mineral salts, on the other.



Fig. 6. Microradiogram, Proximal caries in two adjacent teeth.  $\times 100$ .

#### DISCUSSION

The microradiographic investigation of the molars in rats with experimental caries disclosed findings in close agreement with the ones in caries in man (cp. *Bergman & Engfeldt, 1954*). Thus, an area with low x-ray absorption of the enamel was observed in the fissure caries lesions, and also the dentine situated beneath carious enamel showed a decreased x-ray absorption. In more advanced fissure caries the area of the enamel with low x-ray absorption was increased. These findings demonstrate that there is a dissolution of calcium salts from the involved enamel and

dentine. These details are similar to what is observed in caries in man and might indicate similarities in the development of fissure caries in rat and man.

As regards the reaction of the dentine to the carious lesions there was a discrepancy, however. The areas with increased x-ray absorption of the dentine observed in human caries have not been seen in the molars of the rats. This negative result can, however, not directly be taken as evidence of such a reaction of the dentine being absent in the rat. On the basis of our material it cannot be decided whether the discrepancy in this respect between human caries and caries in rats is depending upon different modes of reaction, or whether a time factor may be involved. It is not possible to rule out the possibility of longer experimental periods than we have used being required for the occurrence of areas with increased x-ray absorption of the dentine situated beneath the carious lesions. To elucidate this question further studies on these lines will have to be carried out.

In the abraded cusps areas with increased x-ray absorption of the dentine were observed. These areas were situated at the outer surface of the dentine as well as in its deeper parts. It is known that artefacts may occur at the borderline between highly absorbing and non-absorbing areas. However, areas with similar differences in absorption as the cusp tips did not show this effect, and accordingly this observation must be taken to indicate a really increased x-ray absorption in a narrow zone at the surface of the cusp tips. This zone probably depended on an increased mineral content but might also depend on an incorporation of foreign material in the surface of the dentine. The areas with somewhat increased x-ray absorption in the deeper parts of the cusps, however, most probably depended on an increase of the mineral salts of the dentine. These latter changes were similar to what is observed in attrition in man.

From our study it is apparent that there is no clear correlation between the transparency of the dentine and the x-ray absorption. With the technique used in the present study, the x-ray absorption provides evidence as to the content and distribution of mineral salts in mineralized tissues and thus it seems clear that the transparency of the dentine in white rats can not be taken as an indication of its degree of mineralization.

## SUMMARY

In a microradiographic investigation of carious molars from white rats it could be demonstrated that the lesions showed similarities with corresponding lesions in man. However, in the rats no areas with increased mineralization were observed in the dentine.

In attrition, however, areas with increased mineralization were observed in the cuspal dentine.

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