Mineralization of the Growing Tooth as Shown by Radio-Phosphorus Autographs.* (17692)

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The deposition of radio-phosphorus in growing molar teeth was examined in young hamsters with the help of the autographic technic. An injection of carrier-free radio-phosphorus (P^{32}) was given at the age of 4 days, that is, soon after the appearance of dentin and enamel in the germs of the first and second molars. The animals were sacrificed 1 hour, 6 hours, 1 day, 2 days or 4 days later. The jaws were fixed in a neutral formalin-alcohol mixture, embedded in celloidin without decalcification, sectioned as usual, autographed by coating the sections with photographic emulsion according to the "inverted method"(1), and stained with basic fuchsin. Dentin and enamel, but not soft tissues, showed autographic reactions. These were attributed to the presence of radio-phosphorus in a form sufficiently insoluble to be retained in the sections during the histological procedure. Presumably, the radio-phosphorus had become part of the complex phosphate salt making up the crystalline structures of the tooth (apatite?).

At 1 and 6 hours after injection, the autographic reactions were of 2 kinds: *diffuse* reactions extending throughout dentin and enamel, and *localized* reactions restricted to definite parts of these structures. The presence of diffuse reactions was in itself proof that the circulating phosphate must have been able to enter dentin and enamel. Immature enamel is known to be permeable to dyes and other solutions(2), and dentin to radio-sodium(3) and radio-iodine(4). These findings indicate that the "extra-cellular" spaces extend throughout these structures and provide the pathway for the diffusion of the circulating phosphate. Presumably, the radio-phosphorus responsible for the diffuse reactions was deposited by a process of "exchange" between the labeled phosphate circulating in the extracellular spaces and the surface phosphate groups of the crystals making up dentin and enamel(5). The gradual decrease and disappearance of the diffuse reactions, which was observed at 1, 2 and 4 days after injection, would then be due to an exchange in the reverse direction, as the labeled phosphate of the crystal surfaces was replaced by the circulating phosphate which, by that time, no longer contained significant amounts of radio-phosphorus.

At one day after injection, the decrease in the diffuse reactions caused the localized reactions to appear more prominent. In the cross-sections of dentin, the reactions were in the form of a dark line, limited to the inner dentinal region, close to the odontoblasts. At 2 days, the reaction line was in the substance of the dentin at some distance from the odontoblasts; at 4 days, this distance was approximately doubled (Fig. 1). Obviously, the reaction line in Fig. 1 was due to the radio-phosphorus incorporated in the dentin formed during the first 24 hours after injection, while the light area separating it from the odontoblasts consisted of P^{32}-free dentin formed between the first and fourth day. There was little decrease in the intensity of the reaction line with time, presumably because the crystals of dentin formed soon after injection, were labeled throughout, while only the phosphate groups of their surface were

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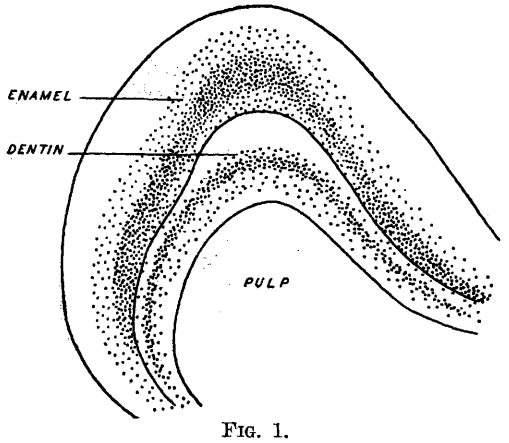

FIG. 1.
Inverted autograph of a section of a molar cusp of an 8-day-old hamster injected with P^{32} 4 days previously. The dentin shows an autographic reaction as a rather narrow dark line of granules in its mid-portion. This line moved from the inner edge of the dentin (1 day after injection) to its present position. The layer of enamel shows a reaction as a broad, dark line of granules at a distance from the ameloblasts and the dentino-enamel junction.

liable to lose their label by exchange. It was concluded that dentin grows by continuous apposition of layers of insoluble phosphate in the area close to the odontoblasts,—a process similar to the formation of periosteal and endosteal bone(6).

The deposition of phosphate in young enamel occurred at a rather considerable distance from the ameloblasts, the reaction being spread over a much broader line than that of the dentin (Fig. 1). The distance between the reaction line and the ameloblasts appeared to increase with time. The reaction did not significantly decrease in intensity with time and, therefore, was attributed to newly formed enamel crystals. Parallel studies with the von Kossa stain for calcium phosphate revealed a progressive increase in calcification from the outer (ameloblastic) limit to the inner (dentinal) limit of the enamel; there was an area of maximum calcification near the apex of the dentino-enamel junction. The radio-phosphate was deposited with a similar gradient, which, however, did not extend as far inwards as the area of maximum calcification. These findings were in agreement with the concept that the formation of enamel is due to the elaboration by the ameloblasts of an organic matrix, which is mineralized much later(7).

The results gave no information as to why the crystals of dentin and enamel form in the indicated areas.

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Synthesis of Sulfur Amino Acids from Inorganic Sulfate by Ruminants.* (17693)

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The conversion of nitrogen of simple compounds in the diet to body protein of the ruminant, probably through the mediation of the micro-organisms of the paunch, is now generally accepted(1,2). However, there have

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