

Psammoma Bodies Associated with Experimental Thyroid Hyperplasia in the Rat. (30339)

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During the course of studies on experimental goiter produced by feeding iodine-deficient diets or by administration of chemical agents to several animal species (hamsters, rats, mice, and monkeys) (1,2), psammoma bodies have been noted within the lumens of the thyroid follicles of the rat. To our knowledge such bodies, so familiar in certain disease processes in man, have not been heretofore described in experimental animals. The purpose of this communication is to record the circumstances under which these bodies have been observed to occur, to describe their morphological characteristics, and to indicate a possible mode of development.

Materials and methods. The material available for study consists of the thyroid glands of over 900 hamsters, 85 mice, 200 rats, and 30 monkeys. The basic diet has been the Remington type: whole ground maize, 76; wheat gluten, 20; calcium carbonate, 2; and sodium chloride, 2. Supplements have consisted of either 500 mg of a vitamin mixture per kilo of diet or substitution of 3 parts powdered yeast for the same amount of ground maize. A complete salt mixture, except for iodine, has been substituted for the sodium chloride and calcium carbonate in some experiments. In certain animals chemical goitrogens: (thiouracil, sulfadiazine and potassium thiocyanate) have been added to the basic diet at levels of .3, 2.0, and .5%, respectively. Some rats and monkeys have received whole ground maize alone. Animals have been fed these diets for periods up to one year and have been either sacrificed at the end of a given period or returned to a diet containing adequate amounts of iodine.

Upon sacrifice, the thyroid glands were removed, fixed in neutral formalin, sectioned, and stained with the following techniques: hematoxylin and eosin, PAS, von Kossa, Feulgen, and prussian blue. Sections were

microradiographed utilizing Phillips CMR equipment.

Observations. Psammoma bodies were first observed in the hyperplastic thyroid glands of rats which had been maintained for prolonged periods on a diet consisting only of whole ground yellow maize. Such a ration is deficient in certain inorganic elements (calcium, sodium, iodine), contains protein of poor quality, and is lacking in certain vitamins. Further studies revealed that the bodies continued to occur when the cornmeal ration was fortified with wheat gluten, calcium carbonate, sodium chloride, and a vitamin mixture, or when a complete salt mixture, except for its iodine content, replaced the sodium chloride and calcium carbonate of the less complete ration. No psammoma bodies occurred when iodine was added to any of these diets; the thyroid gland remains normal on all. The psammoma bodies could be produced more rapidly if a goitrogen, such as thiouracil, was added to a low iodine ration. When iodine was added to a diet which had already led to production of psammoma bodies, the thyroid follicles, as expected, filled up with colloid in which the bodies were then found. It would appear that the prerequisite for development of the bodies is thyroid hyperplasia induced by excess TSH resulting from iodine deficiency.

As already noted, in 3 other species (mice, hamsters, and monkeys), from which a large number of thyroids have been examined during the course of other studies in which hyperplasia of the gland has been produced by iodine-deficient diets of goitrogens, no psammoma bodies have ever been observed.

The fully formed body is generally round, characteristically, though not always, laminated, and measures approximately 30-40 μ in diameter. In hematoxylin and eosin-stained sections, the central portion is intensely basophilic, with varying degrees of

basic to acidic staining about the periphery. Von Kossa preparations show the most in-

tense silver deposition to be in the central core of the body. In some of the structures,

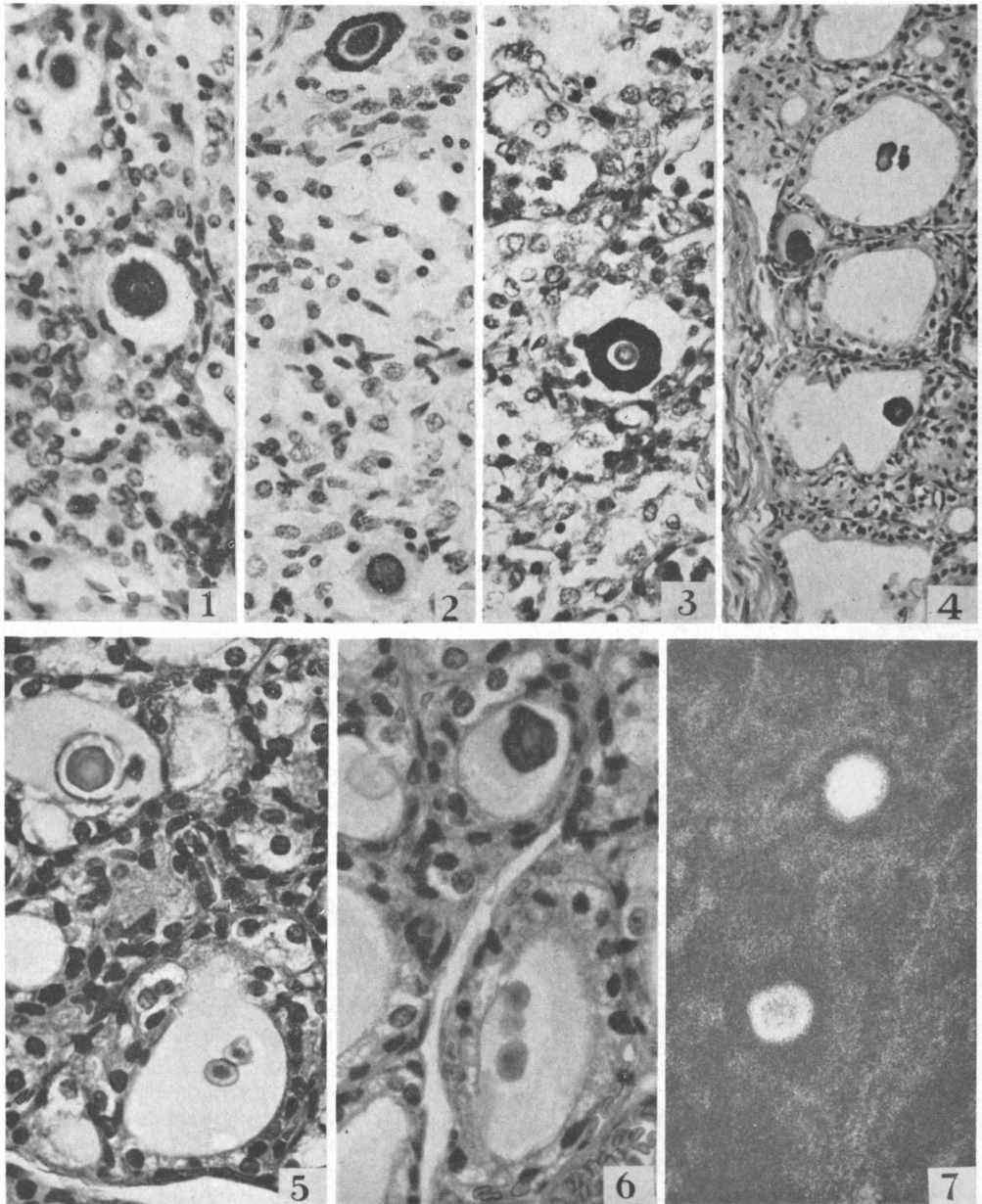


FIG. 1. Typical psammoma body in hyperplastic thyroid gland of rat. H. and E. $\times 400$.

FIG. 2. Two bodies stained by Von Kossa technique. $\times 400$.

FIG. 3. Body in hyperplastic thyroid stained by PAS technique. $\times 400$.

FIG. 4. Bodies within colloid-filled follicles of previously hyperplastic gland which has now involuted following iodine administration. $\times 300$.

FIG. 5. One large and several smaller bodies, the latter having the approximate dimensions of hypertrophied epithelial cells. H. and E. $\times 400$.

FIG. 6. Large and smaller bodies which are basophilic with a more deeply staining center. $\times 400$.

FIG. 7. Microradiograph of 2 bodies to show radioopacity, *i.e.*, presence of inorganic material. $\times 400$.

however, the central portion is paler and one or two concentric rings of argyrophilic material are found about this paler center. The presence of inorganic material is brought out in microradiographs. In PAS preparations the entire body is stained intensely pink to purple. The central portions are basophilic with the hematoxylin counterstain, depending apparently on how much inorganic material has been removed by the periodic acid treatment. In general, the bodies do not give a positive prussian blue reaction. However, occasionally a bluish tinge is found in the calcified areas, indicating the presence of some iron. The bodies are isotropic in polarized light. They are always found within the lumen of the follicle, and have not been observed in the interstitial area between follicles (Fig. 1-7).

In those thyroids in which fully-formed psammoma bodies are present, extraneous materials are found in the lumens of other follicles. These consist of small amorphous basophilic bodies of varying sizes ranging from approximately $10\ \mu$ up to the size of a mature psammoma body. These are not calcified; they stain positively with the PAS reaction. When the Feulgen technique is applied, there is no evidence of a positive DNA reaction.

An attempt has been made to follow the development of the fully formed bodies. As just noted, small, basophilic, usually rounded and amorphous, bodies are prominent in the lumens of the follicles in thyroid glands which contain psammoma bodies. Some of these appear to be formed by an aggregation of several smaller ones which have the approximate diameter of a lining epithelial cell. Very occasionally, a central, more basophilic core is present. Sometimes more dense basophilic particles are present, scattered through the bluish staining matrix. One gains the impression that the initial change may be a desquamation of epithelial cells into the lumens of the follicles. These take on a homogenous basophilic appearance and are generally PAS positive. Some would then appear to aggregate, with this mass eventually rounding up. In time, inorganic materials appear to deposit within a certain number,

sometimes in a cyclic manner, as evidenced by the concentric rings.

Discussion. To my knowledge, this is the first time that psammoma bodies have been described in hyperplastic thyroid glands which have been produced by iodine-deficient diets in the rat. Species differences would appear to be important in that the bodies have not been observed in thyroid glands from iodine-deficient mice, hamsters, or monkeys in this laboratory.

It is my experience and that of others that psammoma bodies are not encountered in association with the hyperplastic thyroid glands of Graves' disease. So, too, in material from cases of non-toxic diffuse or nodular, sporadic and endemic goiters, these bodies are uncommon. This is borne out by studies of Klinck and Winship(3) wherein only one example of tissue containing psammoma bodies was found in 2153 cases of non-toxic nodular goiters. In my own experience, psammoma bodies have been seen in only one case of endemic goiter obtained through the courtesy of Dr. P. Corera, Cali, Colombia. A recent report by Sauter(4) from an endemic goiter area in Switzerland substantiates this situation. On the other hand, in a large series of 473 thyroid cancers reported by Klinck and Winship(3), psammoma bodies were found in 48%. Sauter(4) reports an incidence of 18%.

Although a number of observations on thyroid tumors occurring in rats and hamsters are on record, psammoma bodies do not appear to have been noted in this material. In a few small nodules which have been observed to occur in the thyroids of animals studied in this laboratory, no psammoma bodies were present.

Klinck and Winship(3) have reviewed the various mechanisms which have been advanced to explain the formation of psammoma bodies in thyroid tissue. Most observers, including Klinck and Winship, have suggested that they arose from necrotic epithelial cells. The observations reported herein would be in keeping with this hypothesis which cannot be proved, however, at present.

Summary. Psammoma bodies have been noted in the hyperplastic thyroid glands of

rats which were fed iodine-deficient diets. Such bodies have not been observed in hamsters, mice, and monkeys maintained on similar diets.

Med., 1959, v100, 203.

2. ———, unpublished observations.

3. Klinck, G. H., Winship, T., *Cancer*, 1959, v12, 656.

4. Sauter, C., *Virch. Arch.*, 1964, v337, 328.

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Morphological Changes in Rats Fed Navy Beans.* (30340)

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It has been reported that raw navy beans are toxic to rats when fed at a 10% protein level and that autoclaving the beans destroyed the toxic effect(1). The gross changes observed in rats fed raw navy beans were (a) growth inhibition, (b) reduced food consumption, (c) excessive flow of urine, (d) diarrhea, and ultimately (e) death. Booth *et al* (2) studied the morphological changes of heart, lungs, thyroid, testes, spleen, liver, pancreas, kidney, adrenals and intestine and found that all organs of the animals fed raw soybean meal were normal except the pancreas, which was hypertrophic. These authors suggested that the growth inhibition of rats fed raw soybean meal may be due to direct stimulation of the pancreas resulting in excessive flow of critical amino acids contained in the pancreatic enzymes, which are then excreted in the feces. However, studies made by Saxena *et al*(3) indicate that the growth depression in chicks fed raw soybean meal may be due to inability of the hypertrophic pancreas to supply the necessary digestive enzymes. This investigation was undertaken to determine the morphological changes associated with growth inhibition in rats fed raw navy beans.

Experimental. Composition of the basal diet and the details of experimentation have been described(4). Rats were divided into 5 groups of 6 each and the protein component of the diet fed each group was: (a) raw navy

beans, (b) autoclaved navy beans, (c) raw navy beans plus 0.6% DL methionine, (d) autoclaved navy beans plus 0.6% DL methionine and (e) casein. The remainder of the diet was the same for all groups. The navy beans and casein were included in the diet to provide a level of 10% protein ($N \times 6.25$). At the end of the experimental period of 20 days the rats were subjected to euthanasia and the selected organs were excised, blotted and weighed. Sections of tissues were fixed in 10% neutral formalin. Representative samples of each tissue were embedded in paraffin, sectioned at 6 μ and stained with hematoxylin and eosin. Special stains were used where needed.

Results and discussion. The results of the growth rates of the rats fed the diets were similar to those reported previously(4). For comparative purposes, Table I gives weight changes and food consumption of the rats fed raw navy beans, autoclaved navy beans and casein. In this experiment the rats fed the raw navy beans lost weight and consumed considerably less food compared to those fed the autoclaved bean diet. Although protein efficiency ratio value for autoclaved bean diet is greatly increased over that of raw bean diet, it is significantly less than that of the animals fed the casein diet.

Rats fed the casein or the autoclaved navy bean diets appeared to be normal. However, those fed the raw navy bean diet were emaciated, debilitated, had rough hair coats and were wet around the genitalia. There was paleness of the mucous membranes and the livers were yellowish in color. The abdominal

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