Table II it appears evident that adenosine gave no growth response when added to the soy protein type synthetic ration used in this study, or when added to this diet on top of fish meal or fish meal ash supplements. The picture with regard to ash is not quite so clear but certainly, considering all experiments, little response was obtained compared to the response obtained by the addition of 10% of this particular sample of fish meal. Other experiments not reported here have given the same order of response with respect to the intact fish meal.

Summary. It was found that adenosine at 150 mg kg of ration failed to stimulate chick growth when supplementing a 30% protein, soy protein type purified diet. Supplementation of such rations with 2% fish meal ash gave only small and variable responses, while supplementation with 10% fish meal uniformly gave marked growth response.

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Received August 27, 1956. P.S.E.B.M., 1956, v93.

Effect of Replacement Therapy on Dental Caries Experience of Radiothyroidectomized Rats.* (22746)

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Previous work from this laboratory(1-3) has indicated that the thyroid gland in rats is associated with the dental caries experience. By feeding desiccated thyroid it has been possible to repeatedly demonstrate a significant reduction in dental caries while the administration of I^{131} or thiouracil results in a significant increase in dental caries. From these data it appears quite convincing that the thyroid gland is intimately associated with the dental caries experience in rats. It has become a difficult problem to determine accurately the mechanism by which these effects take place. Various facts are known, however, which suggest that the salivary glands are important intermediates in this mechanism. For example, when salivariadenectomized rats are used, there is no appreciable anticariogenic effect observed by feeding desiccated thyroid. Also, it has been possible to demonstrate a direct relationship between thyroid function, the histological structure of the submaxillary gland and the incidence of dental caries. A further correlation has been shown not only between the microscopic structure of the submaxillary glands, their proteolytic enzyme content and dental caries incidence in hypophysectomized rats but also of proteolytic activity and arginase activity in the submaxillary glands with thyroid function(4-6).

To extend these studies on the relationship of the rat thyroid gland and dental caries, it was considered important to determine if it were possible to return the increased incidence of dental caries occurring in radiothyroid-

* This work was supported in part by a grant from the Office of the Surgeon General, Department of the Army, under Contract No. DA-49-007-MD-332.

TABLE I. Comparison of Incidence of Dental Caries in Radiothyroidectomized Rats Receiving Thyroxine and Testosterone.

Group	Sex	No. of animals	Final wt (g)	No. of lesions		Probability*	
Control	♂	21	370	8.9	> 8.8	—	.001
	♀	18	225	8.7			
I^{131}	♂	18	185	12.7	>13.5	.001	—
	♀	19	131	14.3			
Control + thyroxine	♂	16	326	6.8	> 6.1	.05	.001
	♀	17	242	5.4			
Control + thyroxine + testosterone	♂	14	332	5.4	> 5.9	.05	.002
	♀	15	261	6.4			
I^{131} + thyroxine	♂	18	340	7.4	> 7.5	.11	.01
	♀	19	229	7.6			
I^{131} + testosterone	♂	15	201	11.5	>11.6	.01	.05
	♀	17	178	11.7			
I^{131} + thyroxine + testosterone	♂	15	331	8.6	> 8.3	<.5	.02
	♀	16	252	8.0			

* Probability was calculated from No. of lesions with sexes combined, and the probability values should be compared only within each column.

ectomized rats to that of the control value by replacing the thyroid hormone. Thyroxine administered subcutaneously was used for this purpose instead of desiccated thyroid in the diet as used in previous studies since by use of injection technics it is possible to accurately regulate the dosage. Inasmuch as testosterone has been shown to be necessary for the histological maintenance of the submaxillary gland(6), it was also included in these studies.

Materials and methods. A total of 280 weanling Sprague-Dawley strain rats were divided into 7 experimental groups according to sex and initial body weight. Four groups received 500 μc of carrier-free I^{131} by subcutaneous injection; the first of these groups received no hormone supplement while the second, third and fourth groups received respectively 50 μg of thyroxine daily by subcutaneous injection, 2.5 mg/week/animal of testosterone propionate in croton oil injected intra-muscularly and a combination of thyroxine and testosterone at the same levels as used above. Two non-thyroidectomized groups received hormone injections, one receiving thyroxine and the other testosterone at the same levels as used in the radiothyroidectomized rats, while a final group received no radioiodine or hormone supplements and served as a control group. All of the animals were housed in raised screen cages and received the same cariogenic stock corn diet† (Fluorine = 0.5 μg/g) and fluorine-low (F = 0.5 μg/ml) drinking water. The duration of the experiment was 140 days after which time the animals were sacrificed by ether, the heads removed for dental caries evaluation by methods previously described(7) and the salivary glands and thyroid glands fixed for histologic study.

Results. The results of these experiments are seen in Table I. The injection of radioiodine results in a significant increase in the incidence of dental caries and confirms previous studies(3). Similarly, when thyroxine was injected into non-thyroidectomized rats the incidence of dental caries was significantly decreased. When both thyroxine and testosterone were injected into non-thyroidectomized rats the dental caries incidence was essentially the same as the non-thyroidectomized group which received thyroxine.

The injection of thyroxine into thyroidectomized rats produces a significant decrease in the caries rate and results in the same incidence of dental caries as found in the con-

† The composition of diet used was as follows: yellow corn grits, 52.7%; ground yellow whole corn, 11.3%; powdered whole milk, 30.0%; alfalfa, 4.8%; iodized sodium chloride, 1.0% and irradiated yeast, 0.2%.

trol group. Testosterone injection does not decrease the caries incidence to the same degree as thyroxine in thyroidectomized rats, but does result in significantly less dental caries than found in the non-hormone injected radiothyroidectomized animals. The injection of both thyroxine and testosterone into radiothyroidectomized rats results in approximately the same incidence in caries as found in the controls.

The thyroid glands of the animals which received radioactive iodine exhibited the expected microscopic findings of severe fibrosis with loss of recognizable follicles in most instances. The submaxillary salivary glands reacted to the thyroid disturbance in a manner similar to that following the administration of propylthiouracil. The granular tubules were markedly diminished in size and exhibited severe loss of granules in those animals which had been radiothyroidectomized. The administration of thyroxine alone or in combination with testosterone to radiothyroidectomized rats returned the structure of the submaxillary glands to that comparable to the control animals. Testosterone alone did not exhibit this ability. It may be concluded, therefore, that thyroxine itself or one of its metabolic products is responsible for maintenance of the submaxillary gland granular tubules, directly or indirectly.

Discussion. The relationship between the thyroid gland and the dental caries incidence is clearly shown in this study. When thyroxine is injected into non-thyroidectomized rats a significant decrease in the caries incidence results while radioiodide causes a significant increase in the incidence of dental caries. Thus, the amount of thyroxine used in this study and desiccated thyroid fed as a component of the diet used in previous studies appear to have about the same degree of anticariogenicity. The simultaneous administration of thyroxine and testosterone does not appear to have a synergistic effect on caries, and results in about the same decrease in dental caries as found by the injection of thyroxine alone.

The marked anticariogenic effect of thyroxine is best manifested in the radiothyroidectomized group. In this instance the administration of thyroxine results in a marked decrease in the dental caries experience. Testosterone also significantly decreases the caries experience in radiothyroidectomized rats, suggesting that both of these hormones are necessary for maintaining the normal salivary gland function in the rat. These studies seem to convincingly implicate the thyroid hormone in maintaining the function of the salivary glands and in turn directly affecting the incidence of dental caries in the rat.

Summary. The activity of the thyroid gland appears to be directly related to the incidence of dental caries in the rat, for thyroxine injected significantly decreases the caries rate and radiothyroidectomy significantly increases it. The thyroid hormone appears to be primarily involved since the increased caries rate resulting from radiothyroidectomy is directly counteracted by the administration of thyroxine. Testosterone similarly appears to play a role in this mechanism, since the dental caries rate is also significantly reduced when it is injected into radiothyroidectomized animals even in the absence of thyroxine.

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Received August 27, 1956. P.S.E.B.M., 1956, v93.