6. Scherr, G. H., *Science,* 1952, v116, 685.
7. Munck, A., Scott, J. F., Engel, L. L., *Biochem. et Biophys. Acta,* 1957, v26, 397.
8. McIlwain, H., Rodnight, R., *Biochem. J.,* 1949, v45, 337.
9. Abood, L. G., Romanchek, L., *Ann. N. Y. Acad. Sci.,* 1957, v66, 812.
10. Woolley, D. H., *Science,* 1958, v128, 1277.
11. Green, D. E., Academic Press, N. Y., 1958.
12. McIlwain, H., *Nature,* 1949, v163, 641.
13. Civen, M., Knox, W. E., *Science,* 1959, v129, 1672.

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Effect of Desiccated Thyroid and Thiouracil on Dental Caries Experience of Adult Rats.* (25235)

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Many previous studies(1-7) from these laboratories have shown that feeding desiccated thyroid to rats reduces the incidence of dental caries and administration of either I^{131} or propylthiouracil increases the caries experience. In all previous studies, the thyroid drugs were administered to weanling rats and continued for periods varying from 90 to 140 days. Since previous experience has shown that the proteolytic activity of the secretions of salivary glands of control animals increases with age, but that the effect of propylthiouracil on proteolytic activity is independent of age(8), it was considered of importance to study the relationship between thyroid activity and dental caries in adult animals. Since the proteolytic activity of the salivary glands increases with age with continued administration of desiccated thyroid(8), such information may be helpful in explaining the mechanism of action of the thyroid gland on the caries experience of the rat.

Method. A total of 120 weanling male Sprague-Dawley strain rats was divided equally into 3 groups according to body weight. All of the animals were housed in pairs in raised screen cages and placed on a low caries producing diet (this diet would produce about a mean of 1.5 carious lesions in 100 days) consisting of (in g %): granulated sugar, 72; vitamin test casein, 18; cottonseed oil, 5; vitamin mixture, 11; salts, 4; plus 20 drops of Percomorph oil per kilo of diet. After 100 days (123 days of age) they were divided into 3 groups: Group I animals received a stock corn cariogenic diet consisting of (in g %): yellow corn grits, 52.7; ground yellow whole corn, 11.3; powdered whole milk, 30; alfalfa, 4.8; iodized sodium chloride, 1; and, irradiated yeast, 0.2. The animals in Group II received the same diet to which 0.1% proplythiouracil had been added, and those in Group III the same diet as those in Group I except that it contained 250 mg of desiccated thyroid per kilo of diet. All of the animals received fluoride-low (F = 0.05 mg/ml) drinking water *ad libitum* and remained on their respective diets for an additional 100 days. They were then sacrificed, their heads prepared for dental caries evaluation by methods previously described(9), and thyroid and salivary glands removed for histologic examination.

Results. The data pertaining to body weight gain of the animals and the effect of thyroid drugs on the caries experience are seen in Table I. As previously reported, administration of propylthiouracil is associated with a failure to gain weight. In this study the final body weight of the control group had a mean value of 333 g while that of the animals receiving thiouracil was 214 g. In previous studies the animals receiving desiccated thyroid similarly failed to gain as much weight as the controls, due undoubtedly to their high rate of metabolism. In this study, however, amount of desiccated thyroid was adjusted so that their weight gain was comparable to the controls.

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TABLE I. Effect of Desiccated Thyroid or Propylthiouracil on Body Weight and Dental Caries Experience of Adult Rats.

Group	No. of rats	Final body wt (g)	Dental caries experience: Mean No. of lesions	p*	% rats with caries	Mean severity index of lesions
I (control)	16	333	2.4 ± .09†		81	2.00
II (thiouracil)	28	214	5.5 ± .08	.01	95	2.41
III (thyroid)	28	330	2.4 ± .05		60	1.71

* Calculated on basis of T-test. † Stand. dev.

The incidence of dental caries is also shown in Table I. Controls had a mean caries experience of 2.4 lesions, those receiving propylthiouracil 5.5, and those which received the desiccated thyroid 2.4. The group receiving thiouracil is significantly ($p = 0.01$) different from the controls. The lesions of animals receiving thiouracil were the largest, and those receiving the desiccated thyroid were the smallest.

Discussion. The control animals had an extremely low incidence of dental caries even though they received a highly cariogenic diet for 100 days. This finding is in keeping with our previous findings(10) that rats must be placed on the cariogenic diet before 30 days of age in order to produce a high caries incidence. More attention should be given to those factors which affect the physical or chemical nature of the erupted tooth making the dentition less susceptible to experimentally produced caries as the animals increase in age. It may also be important to consider what environmental factors are changed as well as the changes in the salivary tissues as the animals increase in age, since it has been shown that both salivary flow, viscosity, and pH are a function of the age of the animal(11).

The cariogenic effect of propylthiouracil, as demonstrated so repeatedly in weanling animals, is evident even in adult rats. The mechanism responsible for modifying salivary gland function is, therefore, still operable in adults, thus suggesting that the effect of the propylthiouracil on salivary tissue is not dependent on age. Not only is propylthiouracil associated with an increase in mean number of lesions, but its cariogenic effect is shown also by a higher number of animals with caries as well as by a higher severity index of the lesions.

The anticariogenic effect of desiccated thyroid is not present in the adult rats. This may be due to the fact that such a low dosage level of desiccated thyroid was required not to offset body weight gain of the animals. In earlier studies much higher levels were used. Furthermore, the most dramatic anticariogenic effect of administering desiccated thyroid or thyroxin results when these drugs are given to animals whose thyroid activity had been previously diminished by either I^{131} or propylthiouracil. This certainly is in keeping with the hypothesis that administering thyroxin or desiccated thyroid to an animal with "normal" thyroid function should not result in such dramatic dental changes as would be found in an animal with hypothyroidism. However, even at the low dosage levels of desiccated thyroid used in this study, one can see evidence of the anticariogenic effect of the thyroid drug. For example, the number of rats with caries is much less as is the severity of the lesion.

Histologic examination of the thyroid glands of animals receiving propylthiouracil revealed the typical extreme hyperplasia of the gland with increased cellularity and lack of colloid formation. In contrast, the animals receiving thyroid exhibited small thyroid glands with follicles lined by flattened cells and filled with colloid.

The salivary glands of animals in this study did not differ from those in previous studies on younger animals. The granular tubules of the submaxillary glands in animals receiving propylthiouracil were extremely small and nearly devoid of granules while those receiving thyroid were extremely large and choked with granules. No changes were noted in either the parotid or sublingual glands of either group.

From histologic evidence alone, there does not appear to be significant difference in reaction of either the thyroid gland or the submaxillary gland to desiccated thyroid or to propylthiouracil between young weanling or older adult animals.

Conclusions. The effect of feeding either desiccated thyroid or propylthiouracil was studied to determine if the former drug resulted in an anticariogenic effect and the latter produced a cariogenic effect in adult rats to the same degree as in weanling animals. Propylthiouracil resulted in significantly more caries than were found in weanling animals, but desiccated thyroid did not reduce the caries experience. Similar histologic changes were found in both thyroid and salivary glands in the adult rats as were found previously in weanlings.

1. Muhler, J. C., Shafer, W. G., *Science,* 1954, v119, 667.
2. ———, PROC. SOC. EXP. BIOL. AND MED., 1955, v88, 191.
3. Bixler, D., Muhler, J. C., Shafer, W. G., *J.A.D.A.,* 1956, v53, 667.
4. Shafer, W. G., Muhler, J. C., *J. Dent. Res.,* 1956, v35, 1956.
5. Muhler, J. C., Shafer, W. G., *ibid.,* 1957, v36, 1957.
6. Bixler, D., Muhler, J. C., *ibid.,* 1957, v36, 304.
7. Bixler, D., Muhler, J. C., Shafer, W. G., *ibid.,* 1957, v36, 571.
8. Clark, P. G., Shafer, W. G., Muhler, J. C., *ibid.,* 1957, v36, 403.
9. Muhler, J. C., Nebergall, W. H., Day, H. G., *ibid.,* 1954, v33, 33.
10. Muhler, J. C., *ibid.,* 1954, v33, 245.
11. Shafer, W. G., Clark, P. G., Bixler, D., Muhler, J. C., *ibid.,* 1958, v37, 848.

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Effects of High Pressures on Influenza Viruses.* (25236)

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Various viruses have been subjected to high compression pressures of 30,000 to 120,000 lb/in^2 (2000 to 8000 atmospheres) to test the effect of these pressures on infectivity, but the results were complicated by temperature increases accompanying the pressure rise. Since pressure was generated by a hydraulic press operated at room temperature, increase in heat of the virus suspension in the press was unavoidable. Using the hydraulic press apparatus Basset(1,2) studied a number of viruses including 2 plant agents (tobacco mosaic and tobacco mecrosis), 6 animal (vaccinia, herpes, rabies, yellow fever, Rous sarcoma, and equine encephalomyelitis) and staphylococcus bacteriophage. The most susceptible to inactivation was the bacteriophage which was inactivated at 30,000 lb/in^2 and the most resistant was tobacco mosaic inactivated at 12,000 lb/in^2. All of the above studies were reported prior to 1941 so that much modern data regarding measurements of infectivity were not available. An ingenious device recently developed by Arthur D. Little produces pressures of over 30,000 lb/in^2 at below freezing temperatures. The method depends on freezing and expansion of water in a steel cell. The sample to be tested is placed inside the cylinder which is then filled with water. The cell is sealed and placed in a deep freeze at various freezing temperatures. As the water freezes it expands against the contracting steel and internal compression pressures result which can be measured by a strain gauge. Using this device Curtis *et al.*(3) studied avian erythroblastosis and mouse hepatitis (Nelson) viruses and found inactivation complete at 30,000 and 25,000 lb/in^2 respectively. Experiments to determine minimum pressure were not reported. None of the previous studies of the effects of pressure on viruses have included influenza virus. Since this virus is represented by many diverse strains all of which have 2 distinct and easily

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