# B M R and Thyroxine Secretion Rate in Salivariectomized Female Rats.* (22239)

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The salivary glands have been known for many years to play a role in iodine metabolism by secreting (excreting?) blood iodide as a component of saliva. Recently a function of the salivary glands in iodine metabolism has been reported by Fawcett and Kirkwood(1.2) in the rat, namely to deiodinate diiodotyrosine and monoiodotyrosine, the latter by virtue of the presence of an enzyme, tyrosine iodinase. These workers also reported that the rapid deiodination of intravenously injected diiodotyrosine which occurs in normal rats was prevented (up to 60 minutes) in rats previously salivariectomized. Consequently the suggestion has been made (1) that not only is the blood level of thyroxine in the rat maintained by the rate of thyroxine synthesis in the thyroid gland but also by the rate of destruction of thyroxine and/or its precursors, monoiodotyrosine and diiodotyrosine wherever this may occur.

The purpose of the experiments reported herein was to determine the effect of salivariectomy on thyroid function as shown by the BMR and thyroxine secretion rate; *viz.* do the salivary glands play an essential role in extrathyroidal thyroxine metabolism or can their function be dispensed with without seriously disturbing thyroxine metabolism?

*Procedure.* Young adult Sprague-Dawley female rats, fed Purina Laboratory Chow and water *ad lib.*, were used in these 2 experiments. In the *first experiment,* 8 female rats were salivariectomized under pentabarbital (Nembutal) anesthesia and allowed to recover; 8 other normal females served as control animals. The individual body weight range for these 16 animals was 100-125 g. The BMR's of these rats were determined from $O_2$ consumption data obtained by using a closed, large glass desiccator with a water manometer and an attached 10 cc syringe filled with air. $CO_2$ was absorbed by NaOH (Ascarite). The pressure inside the desiccator was kept relatively constant by replacing periodically the oxygen consumed with air from the syringe. Constant temperature was maintained by use of a water bath at room temperature. The BMR's used in the final calculation for the individual rats were averages of at least 2 determinations. In the *second experiment,* thyroxine secretion rates of 23 salivariectomized and 25 intact rats were calculated by comparing thyroid gland weights at autopsy of a control group of rats having received no thiouracil with 3 groups of animals having received .1% thiouracil† in their drinking water for 2 weeks; in addition, 2 of the latter 3 groups were injected daily with 3 $\gamma$ and 6 $\gamma$ respectively of thyroxine‡ subcutaneously. The thyroxine crystals were dissolved in distilled water with just enough 0.1 N NaOH added to make them soluble. The thyroxine solution was prepared weekly and stored in a refrigerator between injections. The body weight range of these animals was 150-180 g.

*Results.* The results of calculating the BMR's of normal rats and salivariectomized rats, 4-40 days postoperatively, are shown in

TABLE I. Comparison of BMR's of Salivariectomized and Intact Rats. Differences between experimental and normal groups are not statistically significant using the t-test.

| | | Mean BMR (Cal./M²/hr) | | |
|---|---|---|---|---|
| | | Days postoperative | | |
| Rats | No. | 4-6 | 18-20 | 38-40 |
| Salivariectomized | 8 | 44.4 ± 5.6 | 44.1 ± 6.2 | 40.1 ± 5.0 |
| Normal | 4 | 47.1 ± 6.9 | 46.6 ± 3.1 | |
| " | 4 | | | 41.2 ± 5.5 |

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† Thanks are due Lederle Laboratories for a generous supply of thiouracil.

‡ Crystalline thyroxine was supplied through the courtesy of G. S. Reed of the Squibb Institute for Medical Research. This material contained approximately 90% l-thyroxine and 10% d-thyroxine.

TABLE II. Data for Determination of Daily Thyroxine Secretion Rate in Rats. Differences in thyroid weights between respective normal and experimental groups are not statistically significant using the t-test.

| Groups | Thiourea | Daily thyroxine dose, γ | No. animals | Mean initial body wt, g | Mean body wt at autopsy, g | Mean thyroid wt, mg % |
|---|---|---|---|---|---|---|
| Normal | No | 0 | 7 | 161 | 183 | 6.08 ± .7 |
| | Yes | 0 | 6 | 166 | 185 | 15.4 ± 1.3 |
| | " | 3 | 6 | 159 | 175 | 10.7 ± 2.1 |
| | " | 6 | 6 | 164 | 182 | 7.2 ± .8 |
| Experimental | No | 0 | 5 | 156 | 180 | 6.14 ± .7 |
| | Yes | 0 | 6 | 153 | 169 | 16.6 ± 2.2 |
| | " | 3 | 6 | 153 | 172 | 9.78 ± 2.9 |
| | " | 6 | 6 | 151 | 167 | 5.9 ± 1.9 |

Table I. In spite of the fact that at the time of each determination the mean BMR of the control animals was higher than that of the experimental animals, this difference is not statistically significant when analysed with the t-test.

Experimental data used in determining the daily thyroxine secretion rate of salivariectomized and intact rats are shown in Table II. Differences between the corresponding normal and experimental groups receiving otherwise similar treatment are statistically insignificant when analyzed with the t-test. By plotting the thyroid gland weights from Table II against different groups of treated animals as shown in Fig. 1, an estimation of mean daily thyroxine secretion is obtained. For the control group of rats this value is approximately 6.6 γ thyroxine secreted per rat per day and for the experimental group. 5.7 γ/rat/day. Because l-thyroxine is the more active if not *the* active form of thyroxine(4), the true secretion rate of l-thyroxine would be 90% of these values or 5.9 γ and 5.1 γ/rat/day respectively. Converting these figures to a weight basis, the values become approximately 3.4 and 2.9 γ l-thyroxine secreted/100 g rat/day.

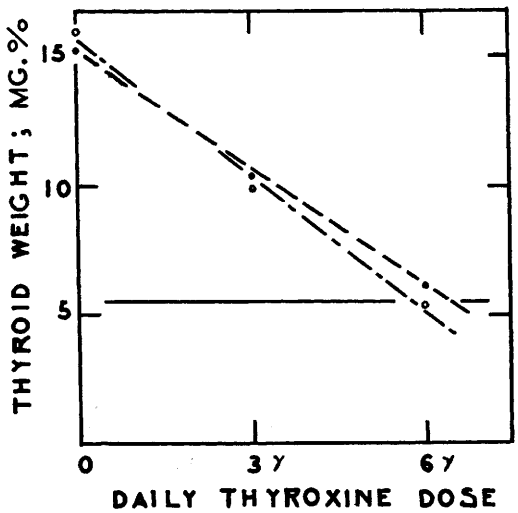


FIG. 1. Plotted data from which daily thyroxine secretion rate is calculated. ———, all rats receiving no thiouracil; – – – –, normal rats receiving thiouracil; — · — · —, salivariectomized rats receiving thiouracil.

*Discussion.* As a result of their experiments, Fawcett and Kirkwood(1) concluded that salivary glands of the rat are capable of acting as "thyroids-in-reverse" by degrading thyroxine into its constituent parts. This degradation by the salivary glands presumably occurs in steps, one of which(2) they have identified, namely, the deiodination of monoiodotyrosine under the control of tyrosine iodinase. This enzyme is reported to be comparatively abundant in the 3 salivary glands of the rat, particularly in the submaxillary and parotid glands. These 2 glands in the rat have greater concentrations of tyrosine iodinase than the thyroid gland where this enzyme catalyzes one step in thyroxine synthesis. In addition to the above action in the salivary glands, it has been reported that the normally rapid deiodination of intravenously injected diiodotyrosine does not occur in salivariectomized rats(1), at least within the space of 1 hour.

From the results of the present experiments it is concluded that the role of the rat's salivary glands in controlling thyroxine levels in

the body by fostering the degradation of thyroxine and/or its breakdown products is not an indispensable role under normal circumstances because the salivary glands can be removed without causing a statistically significant change in thyroxine metabolism as revealed by the BMR and thyroxine secretion rate.

Perhaps after salivariectomy, control of thyroxine level through degradation is assumed by other tissues at a slower but nonethe-less equally effective rate. The kidney and stomach have been reported to possess tyrosine iodinase in low concentrations(2). Evidence is accumulating to show that many tissues can deiodinate thyroxine with formation of triiodothyronine as for example the kidney in which the process appears to be controlled by an adaptive enzymatic mechanism(3).

On the other hand, an important point in determining the comprehensive role of the salivary glands in thyroxine metabolism still remains to be demonstrated; *i.e.*, their participation in the degradation of thyroxine itself to an extent relatively greater than that of other peripheral tissues which are metabolically stimulated by thyroxine and thus "utilize" thyroxine in their normal metabolism. If the salivary glands do not destroy thyroxine to a relatively greater extent than other tissues do, it is evident that a salivariectomy will probably not result in a recognizable change in BMR or thyroxine secretion rate.

Obviously more experimental data need to be available before an adequate appraisal of the normal role of the salivary glands in thyroxine metabolism can be made, however, in view of the present finding, it appears that this role is nonessential or adaptive.

*Summary.* Salivariectomy performed in young adult female rats produced no statistically significant effect on BMR or thyroxine secretion rate. It is concluded that rat salivary glands play a nonessential or adaptive role in thyroxine production and metabolism.

NOTE: After this paper was submitted for publication, two other papers(5,6) reported similar conclusions reached by different methods.

The technical assistance of Miss Gwendolyn Slover is gratefully acknowledged.

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## Serological Response in Chickens to Beta-Propiolactone Treated Newcastle Disease Virus. (22240)

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In a previous study(1) it was demonstrated that Newcastle disease virus (NDV) was rendered noninfectious for chickens by treatment with 0.025% beta-propiolactone (BPL). The vaccinated birds developed no symptoms of Newcastle disease while 97.4% unvaccinated ones developed symptoms or died from the disease. Hemagglutination inhibition tests performed on paired sera from the vaccinated group showed a significant rise in titer.

The present communication presents additional information about the antibody response in the vaccinated group.

*Materials and methods.* White Leghorn chickens were used. All birds were from the same apparently disease-free flock. Beta-propiolactone was supplied by Dr. T. L. Gresham, B. F. Goodrich Research Center, Brecks-