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Acute Inhibition of Thyroid Function in the Rat Produced by Rabbit Antibody to Beef Thyrotrophic Hormone.*† (27727)

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Rabbit antibody produced in response to injection of bovine thyrotrophic hormone (TSH) effectively neutralizes the thyroid stimulating activity of injected rat pituitary extract(1). Since the anti-beef TSH antibody interferes with the effects of exogenous rat TSH, it seemed likely that the antibody could also inactivate circulating endogenous hormone in this animal. An analogous experiment, the production of acute diabetes mellitus in the rat has already been accomplished by Wright(2) using an anti-beef insulin antibody produced in the guinea pig. In the following experiment thyroid function has been measured continuously in the rat by the *in vivo* counting of neck radioactivity following injection of radioactive iodine; the effects of injection of antibody were then tested during the course of a thyroid I^{131} release curve. An abstract of this work has been published(1).

Methods. Antibody to bovine TSH was produced in rabbits by the method of Werner *et al.*(3). Adult male New Zealand rabbits were injected in multiple intracutaneous sites with an emulsion of Thytropar (Armour Laboratories) in Freund's adjuvant. Three series of injections of 3 units each were given at 10-day intervals. The initial injection was with the complete adjuvant; later injections used incomplete adjuvant. Plasma was harvested 10 days after the last injection and at intervals after booster injections of one unit and was stored in the frozen state. Thyroid I^{131} release curves were determined by serial neck counts of groups of adult male rats injected with 40 μc of radioiodine 24 hours prior to the initial neck count(4).

Results. (Fig. 1). Following a control period in which initial I^{131} release rates were determined one group of 6 rats was injected intraperitoneally once a day for 3 days with plasma from TSH-immunized rabbits. Each animal was given a total of 8 ml. Another group of 9 animals was given 8 ml of normal rabbit plasma. In the antibody treated groups there was prompt cessation of I^{131} release persisting for at least 6 days. Although there is a suggestion from the curve that return towards normal function had begun about 6 days after injection, it is possible that inhibition was present even on the seventh day. To determine when the antibody induced thyroid inhibition became significant statistically, comparisons by the t test were made for successive sets of neck counts following injection: at 16 hours post injection, P value was $<.1$; at 35 hours, the mean neck counts were significantly different ($P<.01$) and remained so for each succeeding set of determinations ($P<.01$ to $<.001$). Even though significant differences in mean neck counts were not apparent until 35 hours after antibody injection, it is possible to estimate roughly the time of onset of antibody effect by extrapolation of the post-injection curve back to the pre-injection curve. The point of intersection falls 90 minutes after injection.

Discussion. Acute inhibition of thyroid I^{131} release in the rat follows injection of rab-

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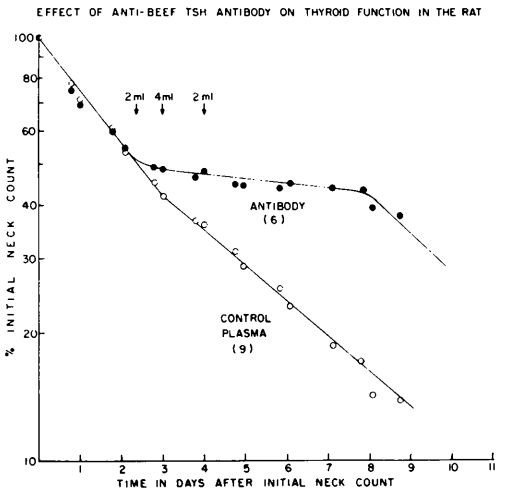

FIG. 1. Effect of anti-beef TSH antibody (rabbit) on thyroid function in the rat. Illustrated here is the course of thyroidal I^{131} release as measured by external neck counting in groups of rats. The marked inhibitory effects of antibody injections are contrasted with the minimal effects due to normal rabbit plasma. The curve has been drawn to suggest that return to normal thyroid function has begun by the eighth day. There are insufficient data however to establish this point with certainty, and it is possible that the last 2 determinations in the antibody treated group fall on a straight line drawn between the third and ninth day.

bit antibody to beef TSH. Cross reactivity of the antibody, recognized by a number of workers(5-7) is thus confirmed. The material is potent enough to inhibit endogenous TSH action. A similar effect on endogenous thyroid function in the rat has also been described by Baschieri *et al.*(8) who showed that injection of 0.1 ml of immune rabbit plasma every other day for 10 days led to significant inhibition of thyroid I^{131} uptake and to decrease in thyroid gland size. The minimum effective dose of the anti-plasma has not been determined; following *in vitro* incubation 0.005 of antiplasma effectively neutralizes 1.0 mU of USP standard TSH as determined by bioassay in the mouse.

Of particular interest is the time course of the thyroid response to the anti-plasma injection. It closely resembles that seen following thyroxine injection in the rabbit(9) and the rat(10), as well as that produced by hypophysectomy in these species(10,11). These results further confirm the fact that the thyroid mechanisms responsible for release of thyroid hormone from the gland are dependent from minute to minute upon the local concentration of thyrotrophic hormone.

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Test for Metabolic Attack on Tris(Hydroxymethyl)Aminomethane and α-Hydroxymethylserine in the Rat.* (27728)

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Tris(hydroxymethyl)aminomethane† is under investigation as a therapeutic alkalinizing agent (see bibliography in reference 1). The fact that this substance has appreciable affinity for liver alcohol dehydrogenase, and is apparently slowly oxidized by it(2), raises

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† Tris is used here for tris(hydroxymethyl)aminomethane.