

found. The potency of these 2 preparations was nearly identical. These findings strongly indicate that the development of a refractory condition in prolonged treatments with preparations containing the thyrotropic principle is not due to an exhaustion of the thyroid or to the formation of an "antihormone."

### 8635 P

#### **Antibody Nature of Refractoriness to Injections of Hypophyseal Extracts Containing Thyrotropic Hormone.\***

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It has been shown in the preceding paper that the development of refractoriness to repeated injections of thyrotropic hormone is influenced by the type of extract in which the hormone is contained. This suggests that refractoriness is an immune response to foreign proteins and is not due to the formation of an "antihormone." If this is the explanation, preparations not inducing refractoriness would be less antigenic. This decrease in antigenicity has been tested and confirmed. No gland iodine determinations were done in this series. The extracts and methods used were the same as in the preceding paper.

A. *Effect of non-stimulating doses of the Na<sub>2</sub>SO<sub>4</sub> and the flavianate preparations of beef anterior hypophyses on the development of refractoriness in the thyroid.* Two series of 6 male guinea pigs each, weighing 250-350 gm., were used. Five of the first series were given 0.1 cc. of the Na<sub>2</sub>SO<sub>4</sub> preparation subcutaneously every other day for 32 days. They showed no rise in the BMR. Twenty times this amount (2 cc.) then given daily for 5 days caused no activation of the thyroid, as revealed by the BMR and biopsy specimens of thyroid. The previously untreated control showed a marked response to this large dosage. The second series was treated like the above except that the flavianate extract was used and the treatment lasted for 41 days (0.02 gm. dried gland equivalent was given at each injection). No elevation of BMR took place during this treatment. Twenty times the initial

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dosage (0.4 gm. dried gland equivalent) was then injected daily for 4 days. It caused marked thyroid stimulation in 3 of the 5 treated animals as well as in the control. The other 2 animals showed only slight thyroid stimulation.

B. *Absence of inhibitory substances in serum of animals treated with the flavianate preparation of beef anterior hypophysis.* Three rabbits and 2 sheep were used. One rabbit received daily intraperitoneal injections of one cc. of the  $\text{Na}_2\text{SO}_4$  preparation for 30 days and 2 rabbits 0.2 gm. equivalent of dried beef hypophysis as the flavianate daily for 50 days. One sheep was given 0.4 gm. dried gland equivalent of the flavianate intraperitoneally each day for 45 days and then 1.6 gm. equivalent for 46 days more. The other sheep was treated daily with the equivalent of 1.6 gm. dried hypophysis as a flavianate for 41 days. No inhibitory properties against the flavianate extract were found in the serum of either the sheep or the rabbits using the technic of Collip *et al.*<sup>1</sup> of injection of the serum and the extract into normal guinea pigs.

C. *Renewed thyroid stimulation by the flavianate preparation in guinea pigs refractory to the  $\text{Na}_2\text{SO}_4$  preparation.* Fourteen guinea pigs were made refractory to the  $\text{Na}_2\text{SO}_4$  preparation of beef hypophysis by daily injections for 35-45 days. Ten of these received one cc. daily and 4 received 0.1 cc. every other day. To test for refractoriness 2 cc. of the same preparation was then given daily for 4 days. No response to this increased dosage took place as revealed by the BMR and small thyroid biopsy specimens. Eleven of these animals were then injected daily with 0.5 gm. equivalent of the beef hypophyseal flavianate extract for 4 days, a dosage about equal in potency to 2 cc. of the  $\text{Na}_2\text{SO}_4$  preparation. Definite elevation of BMR and activation of the thyroid gland structurally occurred in 9 of the 11 animals. Two were not stimulated. As controls, the other 3 guinea pigs were continued on the 2 cc. dose of the  $\text{Na}_2\text{SO}_4$  preparation for 4 days after biopsy. These showed no thyroid activation or rise in the BMR at the termination of the treatment.

Four points indicate that as regards the thyrotropic principle the alleged "antihormone" formation is an immune response to proteins associated with the hormone. First, the development of the refractoriness depends on the type and not the potency of the thyrotropic preparation. Second, immunizing but non-stimulating

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<sup>1</sup> Collip, J. B., *J. Mt. Sinai Hosp.*, 1934, 1, 28; Collip, J. B. and Anderson, E. M., *Lancet*, 1934, 1, 76; Bachman, C., Collip, J. B. and Selye, H., *Proc. Soc. Exp. Biol. and Med.*, 1934, 32, 544.

doses of a certain preparation produce refractoriness as efficiently as do large doses, whereas another preparation—the flavianate—only incompletely immunizes in small doses. Third, the sera of animals chronically treated with the flavianate preparation show no inhibitory properties against it. Fourth, the thyroids of animals which have become refractory to the  $\text{Na}_2\text{SO}_4$  preparation are stimulated when the flavianate preparation is injected even though the extract is made from anterior hypophyses of the same species of animal—the ox.

The flavianate has some degree of antigenicity, as shown by partial immunization and by its partial interaction with antibodies against the other preparation used. This remaining antigenicity suggests that a species which forms antibodies more readily than the guinea pig, might become immunized to the flavianate extract and hence refractory to its injection. This is indicated by the work of Evans *et al.*<sup>2</sup>

### 8636 P

#### Cytology of the Monkey Endometrium Under Influence of Follicular and Corpus Luteum Hormones.\*

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The histologic features of the endometrium of *Macacus rhesus* monkeys under treatment with the ovarian hormones have already been described by one of us (Hisaw<sup>1</sup>). We have had the opportunity to make cytological studies involving the use of various special stains upon specimens from 4 uteri under the influence of the ovarian follicular hormone (oestrin) and upon specimens from 6 uteri under the influence of the corpus luteum hormone (corporin). The monkeys used were castrated immature or young adult females, and the dosage and duration of treatment were sufficient to produce the histologic pictures in the endometrium which have been described (Hisaw<sup>1</sup>) as characteristic of oestrin or of corporin stimulation.

<sup>2</sup> Evans, H. M., Pencharz, R. I. and Simpson, M. E., *Endoc.*, 1935, **19**, 509.

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<sup>1</sup> Hisaw, F. L., *Am. J. Ob. and Gyn.*, 1935, **29**, 638.