Influence of Cortisone on Nervous Tissue, Adrenal and Thyroid. (20747)

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A recent investigation by Castor *et al.*(1) indicated that the administration of cortisone to rats induces chromatolysis and cytoplasmic vacuolation of thalamic and hypothalamic nerve cells. The present study was designed primarily to determine whether the changes described by this group could be substantiated and to investigate the effects of large doses of cortisone on the histology of the cat brain. Furthermore, observations were recorded relating to the effects of this hormone on the adrenal and thyroid glands of the cat.

Materials and Methods. A) Cats: The 9 normal cats used in this experiment were subdivided into 3 groups. Group I consisted of 2 animals which received cortisone acetate* for 10 days at a daily dosage of 50 mg per kg of body weight. The 4 animals in Group II were given 25mg of cortisone acetate per kg of body weight daily for 10 days. Group III was comprised of 3 control cats to which was administered a daily injection of the vehicle for 10 days. A tenth cat was given the hormone for 20 days at a dosage of 25 mg per kg of body weight. All the injections were subcutaneous. One week in advance of the first injection all the cats were started on a controlled daily diet of 100 g of horsemeat and 100 ml milk, and maintained on this regimen until the experiment was terminated. Each animal was weighed daily. Twenty-four hours after the last injection the cats were sacrificed, their heads perfused with 10% formalin and the brains removed. After further fixation, the brains were embedded in nitrocellulose. The thyroids and adrenals of these animals were also removed, fixed in 10% formalin and embedded in paraffin. Frontal serial sections of the brains were cut at 10 μ; every tenth section was stained with cresyl violet, and the next section with toluidine blue. The thyroids and adrenals were cut at 6 μ and stained with Gomori's trichrome stain. B) Rats: The brains of 44 young adult Sprague-Dawley rats were used in this study. Twelve animals were given daily subcutaneous injections of 5 mg of cortisone for 20 days and pair-fed with 12 uninjected controls. Another group of 10 animals was given 8 mg of the hormone for 10 days, and pair-fed with 10 uninjected controls. Twenty-four hours after the last injection the brains were removed, fixed in 10% formalin and embedded in paraffin. Frontal serial sections were cut at 10 μ and stained with toluidine blue. Insofar as possible, control and experimental sections of all the nervous tissue were matched area for area and the slides stained in pairs. A systematic study of the entire brain was undertaken. The hypothalamic nuclei examined were those listed by Rioch, Wislocki, and O'Leary(2), and Ingram(3).

Results. A) Nervous system: Microscopic examination of the cat and rat brains revealed no evidence of cytological changes, such as chromatolysis or vacuolization. There were also no indications of glial proliferation, hyperemia, hemorrhage or edema of the brain or meninges. B) Adrenals: The glands of the treated cats all displayed a marked atrophy of the zona fasciculata and a slight regression of the zona glomerulosa. There were no demonstrable changes in the zona reticularis. C) Thyroids: The glands of the control cats displayed medium sized or large follicles which contained, in general, large quantities of dense colloid. The epithelial lining of these follicles was cuboidal or lower. The thyroid follicles of the cortisone-treated groups were noticeably smaller and contained little or no colloid. The cells lining the follicles were definitely taller than those of the control group.

Discussion. A) Adrenals. The cortical atrophy observed in the cats given cortisone is consistent with the findings of others in the rat, and much evidence has accrued to indi-

* Cortisone (Cortone acetate Merck) was generously supplied by Dr. Charles A. Winter, Merck & Co., Rahway, N. J.

cate that this involution is due to a restricted output of ACTH by the anterior pituitary (4-6). B) *Thyroids.* The morphological picture of the thyroid in the cortisone-treated cats appears to be indicative of a higher than normal level of thyrotrophic activation. This is in agreement with the findings of Higgins *et al.*(7), and Halmi and Barker(8) in rats. C) Nervous system: Castor *et al.*(1) found that ACTH given to rats in 10 mg dosages per day for a period of 10 days resulted in consistent cytological changes which were restricted to the cells of the hypothalamic paraventricular nucleus. Animals treated with similar quantities of cortisone showed changes of a more widespread nature which involved several hypothalamic and thalamic nuclei. The conclusion reached was that the changes effected by ACTH might be interpreted as evidence of altered secretory activity within the cells of the paraventricular nucleus. However, the ACTH used in their experiment was contaminated with posterior lobe hormones, and it is possible that the latter were responsible for the alterations in the paraventricular nuclei (9). Furthermore, the rats of the Castor group that were treated with cortisone were in poor condition at autopsy whereas the animals used in the present experiment were in excellent condition. This tends to favor the interpretation considered by the authors that the morphological changes in the central nervous system after cortisone treatment might possibly be non-specific and due to the general deterioration of the animals rather than to a specific action of cortisone on the nervous system.

Summary. 1. We attempted to ascertain the effects of large doses of cortisone on the histology of the cat and rat brain. In addition, the adrenals and thyroids of the cats were examined. 2. No change was encountered in the nervous tissue of cortisone-treated animals. 3. The adrenals of the cortisone-treated cats demonstrated the compensatory atrophy which is believed to be brought about by a decreased discharge of ACTH from the anterior pituitary. 4. The thyroids of the treated cats presented histological signs of thyrotrophic activation.

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Large Scale Preparation of Liver Mitochondria with the Waring Blendor.* (20748)

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For studies on the isolation of oxidative enzymes from liver, it was desirable to have a method for the preparation of mitochondria from fairly large amounts of tissue. Methods employing pestle type of homogenizers(1-6) are not convenient for this purpose. The Waring blendor can be used to disintegrate larger amounts of tissue, and procedures utilizing this means of homogenization have been used to prepare mitochondria from skeletal muscle(7), heart(8), and liver(9). However,

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