7666 C

Effect of Salt Treatment on Certain Changes Following Adrenalectomy.

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It has been shown[1, 2, 3] that the loss of sodium chloride and possibly other inorganic ions contributes to the development of the severe symptoms which follow adrenalectomy, and that the administration of these ions will partially or completely prevent the development of these symptoms.

Since disturbances of the oestrous cycle have repeatedly been studied in adrenalectomized animals,[4, 5, 6] and since various authors came to the conclusion that cortical extracts have a specific influence on the sex organs,[6, 7, 8] it seemed of importance to establish whether the administration of salt would correct the disturbance of the cycle, as our preliminary experiments seemed to indicate.[9]

Forty-six normally cyclic rats were adrenalectomized. Fifteen of these were untreated, and went into a dioestrus which lasted until death. The remaining 31 were given the Rubin salt mixture.[3] Of the 18 animals maintained in good health by salt, 14 remained normally oestric; the animals which died in spite of treatment were nearly all dioestric until death. The completeness of the operation was assured by the death which followed soon after the removal of the salt, and by the absence of any cortical tissue at autopsy.

In general, then, it seems to be true that where the salt mixture maintains the life and weight of the adrenalectomized animal, oestrus appears at normal intervals. In this connection we would like to emphasize that the salt mixture maintained only 18 of the 31 animals treated.

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1 Loeb, R. F., Atchley, D. W., Benedict, E. M., and Leland, J., *J. Exp. Med.*, 1933, **57**, 775.

2 Harrop, G. A., Soffer, A. J., Ellsworth, R., and Trescher, J. H., *J. Exp. Med.*, 1933, **58**, 1.

3 Rubin, M. I., and Krick, E. T., PROC. SOC. EXP. BIOL. AND MED., 1933, **31**, 228.

4 Martin, S. J., *Am. J. Physiol.*, 1932, **100**, 180.

5 Wyman, L. C., *Am. J. Physiol.*, 1928, **86**, 528.

6 Corey, E. L., and Britton, S. W., PROC. SOC. EXP. BIOL. AND MED., 1933, **30**, 592.

7 Corey, E. L., *Am. J. Physiol.*, 1933, **105**, 24.

8 Migliavacca, A., *Fol. Gynaec.* (Genova), 1932, **29**, 453.

9 Kutz, R. L., McKeown, T., and Selye, H., *Royal Society of Canada*, May, 1934.

As an incidental finding we observed that unusually large quantities of a greenish yellow pigment, giving the Prussian Blue reaction, were deposited in the spleens of all adrenalectomized animals, treated or untreated. Similar depositions have also been observed in the spleens of hypophysectomized animals. The decrease in number of eosinophiles in the hypophysis which usually follows adrenalectomy was not influenced by the salt treatment.

From these experiments we conclude that the dioestrus following removal of the adrenals is probably not the result of the absence of any specific sex hormone found in the adrenal glands, but rather of the general ill-health of the animal.

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Oxidation of Cystine Sulfur to Sulfate by Ferric Chloride.

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The oxidation of thiol-imidazole sulfur to sulfate by $FeCl_3$ is frequently used in the conversion of thiol-imidazole compounds to imidazoles. One would assume from the literature that cystine sulfur is not similarly oxidized by this reagent.[1] This suggested the utilization of this reaction as a means of testing for the possible presence of a thiol-imidazole compound in insulin. So far only about 70% of the sulfur of insulin has been accounted for. The highly specific Sullivan reaction indicates the presence of about 8.4% cystine, whereas if all the sulfur of insulin (3.2%) were present as cystine, 12.0% should be present.

To test the feasibility of this approach experiments with cystine were first undertaken. These experiments showed contrary to our expectations that cystine sulfur was quite readily oxidized to sulfate by $FeCl_3$. Twenty mg. of cystine heated with 2.5 cc. of 6.5% $FeCl_3$ for 1 hour in the boiling water bath gave a positive test for sulfate. Under the same conditions no sulfate was formed from cysteic acid.

Other experiments on cystine were conducted in which the compound was heated in $FeCl_3$ (6.5%) with HCl varying in concentration from N/10 to 10%. All samples showed appreciable amounts of sulfate after being heated for an hour. The amount of oxidation

[1] Barger, G., and Ewins, A. J., *J. Chem. Soc.*, 1911, **99**, 2336.