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Gonadotropic Activity of the Pituitaries of Horses.*

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There is a quantitative difference in the relative gonadotropic capacity of the pituitaries of various types of animals. The pituitaries of hogs and sheep contain much more of the gonad stimulating substances than do the pituitaries of beef.[1] According to Wallen-Lawrence and Van Dyke[2] similarly prepared extracts of beef and sheep glands showed the former to have less than one-tenth the activity of the latter.

During a series of studies of horse pituitaries collected at various times of the sexual cycle, and at different periods in pregnancy, we have compared their gonad stimulating activity with that of the pituitaries of other animals. The horse glands have considerably more gonadotropic activity than those of sheep, which have been the chief source of material for comparison.

The material was prepared by grinding the fresh glands, desiccating them in 2 volumes of acetone 3 times, completely drying in a warm current of air, and powdering.† A typical experiment is as follows: A total subcutaneous injection of 12.5 mg. of the dried powder administered twice daily over a period of 5 days to 21-22 day old rats produced ovaries weighing 97.4, 64.6, 35.0, 51.2, 35.4, 79.3, and 51.1 mg., average 59.1 mg.; 25 mg. total injection resulted in ovarian weight of 192, 107, 263.4, average 187.5 mg.; and 50 mg. produced ovaries 324.3, 369.2, 250.7, and 177.9 mg. respectively, averaging 280.5 mg.

Ovaries of rats injected with 50 mg. of dried sheep pituitary powder weighed 35-50 mg., while the ovaries of normal untreated controls weighed 12-14 mg. It is thus evident that the concentration of the sex stimulating hormones in horse pituitaries is approximately 4 times that of similarly prepared sheep material.

Furthermore, the effect of horse pituitary powder on the ovaries

* Aided by a grant from the National Research Council, Committee on Problems of Sex, and the University of Wisconsin Research Fund, administered by Frederick L. Hisaw.

[1] Bugbee, E. P., Simond, A. E., and Grimes, H. M., *Endocrinol.*, 1931, **15**, 41.

[2] Wallen-Lawrence, Zonja, and Van Dyke, H. B., *J. Pharm. and Therap.*, 1931, **43**, 93.

† The horse pituitaries were kindly furnished by the Chappel Bros., Inc., of Rockford, Ill., through the courtesy of Dr. A. E. Meyer.

of immature rats appears to differ from that of sheep pituitary powder in that horse pituitaries seem to produce a greater follicular stimulation as compared to luteinization than those of similar preparations from sheep. This difference is especially noticeable in the case of castrate male horses. Pituitaries from 6 castrate males were prepared in powder form as described above. When this material was injected in saline emulsion in doses varying from 3 to 25 mg. the ovaries of the test rats showed predominantly follicular development, even in cases where the ovaries had increased in size as much as 210 mg. Slight luteinization was noticeable in only 2 cases out of 16 rats so injected. It seems therefore, that the pituitaries of these 6 castrate horses contained primarily follicular stimulating activity with very little luteinizing potentiality. This is in sharp contrast to pituitary powder from sheep which when injected in saline emulsion, in doses sufficient to produce ovaries of similar size, stimulated luteinization in every case.

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A Technique for Rendering Tissues Blood-free.

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(Introduced by R. S. Hubbard.)

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The problem of rendering the tissues of a small laboratory animal like the rat, blood-free, must be solved if this animal is used in the study of tissue iron metabolism. In order that a more nearly correct iron content of a tissue may be obtained it will not suffice merely to correct for the iron contained in the blood of such a tissue, nor will the bleeding of the animal through decapitation or through severing a large artery render the tissues blood-free. The solution of this problem, we believe, rests in the development of a perfusion technique whereby the blood may be washed out of the living animal. A method adapted from Whipple's[1] procedure we wish to report here.

The method requires the services of 2 individuals. The equipment includes an ordinary dissecting kit, a ligature needle, a No. 23 hypodermic needle attached by means of a piece of rubber tubing

[1] Whipple, G. H., *Am. J. Physiol.*, 1926, **76**, 693.