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Failure of Hebin to Cause Ovulation in Toads.*

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Hebin, the maturity hormone of pregnancy urine, has been shown by Mazer and Goldstein,[1] and Friedman[2] to produce ovulation in rats, mice, and rabbits. The authors wished to determine if hebin also acts as an inductor of ovulation in toads.

Preparation and Standardization of Hebin. The Wallen-Lawrence and Van Dyke[3] method of preparation of hebin was modified by concentrating the urine under reduced pressure at a temperature below 35°C. and separating the precipitates by filtering instead of centrifuging. In standardization, the equivalent of 0.3 cc. original urine caused the opening of the vagina in each of 3 immature 30-day old female rats; 2 of which showed oestrus cycles and ripe ovarian follicles, but only one of these 2 had an enlarged uterus. This amount was taken as a rat unit. Larger amounts of the preparation showed activity in every case.

Injection of Hebin into toads. Three sexually mature *Bufo americanus* females were placed with males and the former given hebin injections into the dorsal lymph sac. Daily dosages of 1, 2, and 3 rat units, respectively, were used for a period of 5 days. No ovulation occurred during this time. During the next 5 days, injections of 0.5, 1.0, and 1.5 rat units, respectively, were made every 6 hours; making a total of 15, 30, and 45 rat units used. No ovulation occurred during the next 4, 7, and 7 days respectively. The failure to obtain ovulation might be due to one of more possibilities (1) use of immature females, (2) an over-dosage causing degeneration of the ovary, (3) too small of a dosage, and (4) that hebin does not contain the active maturity principle of the anterior lobe of the hypophysis of toads. To test these possibilities, 2 hypophyses from other toads were implanted daily into the above females and ovulation was obtained in all cases on the second day. Since 45 rat units is considered a very high dosage for an animal of this nature,

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[1] Mazer, C., and Goldstein, L., W. B. Saunders Co., Philadelphia, 1932.

[2] Friedman, M. H., *Am. J. Physiol.*, 1932, **99**, 332.

[3] Wallen-Lawrence, Z., and Van Dyke, H. B., *J. Pharm. and Exp. Therap.*, 1931, **43**, 93.

it appears that hebin does not contain the active maturity principle of the anterior lobe of the hypophysis in toads.

Thinking that it might be possible to obtain ovulation in a time of greater sexual activity, both hebin and oestrin were used. Therefore, 4 toads were injected simultaneously with hebin and oestrin into separate lateral lymph sacs with doses of 1, 2, 3, and 4 rat units of hebin and 10, 20, 30, and 40 rat units of oestrin over a period of 4 days; injections being made every 6 hours. No ovulation occurred.

It therefore appears that neither hebin alone nor hebin with oestrin is capable of inducing ovulation in toads. Reichert and his associates,[4] Evans and his coworkers,[5] and Iimuro and Murata[6] have suggested that hebin is not the hormone identical with the gonad stimulating hormone of the hypophysis. Our work with toads substantiates this view.

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A Simple Method for Detecting Abnormal Hearts.

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On the basis of a pulse-ratio method Tuttle and Wells[1] showed quite conclusively that in the case of the normal heart the increase in rate caused by exercise is directly proportional to the intensity of the exercise and that the form of the curve representing the relationship between heart rate and exercise intensity is rectilinear. It occurred to us that if abnormal hearts did not conform to this principle a simple method for detecting them is provided.

By using the pulse-ratio technique data were collected from 32 University freshmen reported as having abnormal hearts. The physicians' report of these cases at the time of examination was as

[4] Reichert, F. L., Pencharz, R. I., Simpson, M. E., Meyer, K., and Evans, H. M., PROC. SOC. EXP. BIOL. AND MED., 1931, **28**, 843.

[5] Evans, H. M., Meyer, K., and Simpson, M. E., PROC. SOC. EXP. BIOL. AND MED., 1931, **28**, 845.

[6] Iimuro, Syozo and Murata, Miyakita., *Trans. Japan. Path. Soc.*, 1931, **21**, 156. (From *Chem. Abst.*)

[1] Tuttle, W. W., and Wells, Geo., *Arbietsphysiol.*, 1931, **4**, 519.