

## Ovulation Induced by PMS and HCG in Hypophysectomized Immature Rats<sup>1</sup> (36336)

SHAO-YAO YING<sup>2</sup> AND ROLAND K. MEYER

*Department of Zoology, University of Wisconsin, Madison, Wisconsin 53706*

The physiological role of gonadotropin has been investigated using hypophysectomized animals as a model. The experimental induction of ovarian follicular development and the accompanied luteinization of follicles and ovulation in many species can be achieved by homologous and heterologous gonadotropins. Gonadotropic hormones of nonpituitary origin have been very useful in studying folliculogenesis and the process of ovulation. PMS (pregnant mare's serum), appears to have predominantly FSH-like activity and also has some LH activity, is capable of stimulating the follicles (1-5). In immature rats, Ying and Meyer (6) have reported that ovulation occurred in 80% of immature rats treated with 3 and 30 IU of PMS, but treatment with 12 or 15 IU of PMS produced little, if any, ovulation. Although the induction of ovulation in hypophysectomized rats has been thoroughly studied (2-5, 7), the objective of these experiments was to determine whether various doses of PMS account for the biphasic dose-response observed in the intact immature rats.

*Materials and Methods.* Twenty-one-day-old female rats of Sprague-Dawley strain weighing between 45-50 g were received from the Badger Research Corp., Madison, WI. The animals were housed in an airconditioned room (75-80°F) under a controlled 14-hr light and 10-hr dark photoperiod. Rockland rat chow and tap water were supplied *ad libitum*.

All animals were hypophysectomized by

<sup>1</sup> Supported by Grant 63-505 from The Ford Foundation.

<sup>2</sup> Present Address: Laboratory of Human Reproduction and Reproductive Biology and Department of Anatomy, Harvard Medical School, Boston, MA 02115.

the parapharyngeal route between 5:00 and 7:00 AM on Day 22 and injected sc with 0, 3, 6, 12, 15, 25, or 30 IU of PMS (Equinex, Ayerst Laboratories, Chicago) in 0.25 ml of physiological saline between 7:30 and 8:00 AM on the same day. Five, 7.5, or 10 IU of HCG (International Hormones, Hicksville, L.I., NY) in 0.25 ml of physiological saline was given ip at 1:30 PM on Day 24. The animals were killed on Day 25. The dissected oviducts were compressed between 2 glass slides and ova were counted under a low power light microscope. The sella turcica of each rat was examined under magnification to determine if the entire pituitary had been removed. Rats in which pituitary fragments were found were not included in the results.

*Results.* Administration of 3, 12, 15, or 30 IU of PMS with or without 5 IU of HCG in a 54-hr interval did not cause ovulation. There was no difference between ovarian weights of animals treated with 3, 12, 15, or 30 IU of PMS only and those of animals treated with 3, 12, 15, or 30 IU of PMS plus 5 IU of HCG. In the group treated with 3 IU of PMS and 7.5 IU of HCG, 1 of 6 animals (16.7%) ovulated with an average of 6 ova; of the group treated with 3 IU of PMS and 10 IU of HCG, 3 of 6 rats (50%) ovulated with an average of 3 ova. Treatment with 6 IU of PMS and 10 IU of HCG produced an average of 20.83 ova in 85.7% of the animals. All animals in groups treated with 12, 15, 25, or 30 IU of PMS plus 7.5 or 10 IU of HCG ovulated with average numbers of 27.83-48.33 ova. In all cases the increase in ovarian weight was proportional to the amount of PMS administered.

*Discussion.* The induction of ovulation with 3 IU PMS in hypophysectomized rats was not as effective as that in intact rats.

TABLE I. Effect of Various Amounts of PMS and HCG on Ovulation in Hypophysectomized Immature Rats.

Dose of HCG	Dose of PMS	Average body wt (g)	No. rats ovul. (%)	Average ovarian wt (mg) $\pm$ SE	Average no. ova/ovul. rat $\pm$ SE
—	3 IU	46.80	0/5 ( 0)	9.97 $\pm$ 1.61	—
—	12 IU	46.80	0/5 ( 0)	30.32 $\pm$ 1.77	—
—	15 IU	44.60	0/5 ( 0)	42.67 $\pm$ 3.68	—
—	30 IU	47.00	0/6 ( 0)	93.30 $\pm$ 6.54	—
5 IU	3 IU	45.40	0/5 ( 0)	10.80 $\pm$ 0.89	—
5 IU	12 IU	44.20	0/5 ( 0)	39.25 $\pm$ 1.45	—
5 IU	15 IU	43.80	0/5 ( 0)	46.24 $\pm$ 5.02	—
5 IU	30 IU	44.00	0/5 ( 0)	93.00 $\pm$ 10.0	—
7.5 IU	3 IU	46.60	1/6 ( 17)	12.32 $\pm$ 0.66	6.00
7.5 IU	12 IU	48.50	6/6 (100)	45.38 $\pm$ 5.10	28.67 $\pm$ 9.00
7.5 IU	15 IU	48.60	6/6 (100)	59.95 $\pm$ 4.07	40.00 $\pm$ 3.79
7.5 IU	30 IU	46.00	6/6 (100)	121.60 $\pm$ 12.4	43.67 $\pm$ 3.79
10 IU	—	50.50	0/6 ( 0)	12.05 $\pm$ 0.94	—
10 IU	3 IU	49.80	3/6 ( 50)	20.63 $\pm$ 1.00	3.00 $\pm$ 0.41
10 IU	6 IU	48.00	6/7 ( 86)	36.84 $\pm$ 9.90	20.83 $\pm$ 9.57
10 IU	12 IU	48.50	6/6 (100)	54.72 $\pm$ 10.0	27.83 $\pm$ 6.45
10 IU	15 IU	48.83	6/6 (100)	61.07 $\pm$ 4.65	48.33 $\pm$ 3.50
10 IU	25 IU	49.83	6/6 (100)	99.30 $\pm$ 4.56	43.17 $\pm$ 4.13
10 IU	30 IU	48.80	5/5 (100)	111.57 $\pm$ 1.43	43.00 $\pm$ 10.2

Even with the addition of HCG, only 17 or 50% of animals ovulated 6 or 3 ova, respectively. But higher doses of PMS (12 to 30 IU) alone were capable of developing adequate follicles responding to exogenous gonadotropin. The response of mature follicles to 7.5 or 10 IU of HCG with subsequent ovulation observed here agrees with an earlier observation that an adequate amount of ovulating hormone is necessary for ovulation in both intact and hypophysectomized rats (8, 9–11). Under the conditions of this study, HCG appeared to provide the hormonal condition necessary for the rupture of the mature follicles. In intact animals treated with 15 IU of PMS, 0.5–2.0 IU of HCG was adequate to induce ovulation (9). Our results showed that higher doses of HCG were needed for the ovaries of hypophysectomized rats to respond. This coincided with the observations that ovulation and *corpora lutea* formation can be induced relatively easier in intact rats than in hypophysectomized animals (7, 12–15).

Although our failure to induce ovulation with low doses of PMS alone in hypophysectomized rats agrees with findings of many

previous investigators (2–5), the induction of ovulation with very high doses of PMS in hypophysectomized rats (1, 2) has been reported. The half-life of PMS in the plasma has been estimated to be 26 hr in the rat (16). Therefore, the PMS circulating 56 hr after its administration probably served as ovulating hormone and consequently caused ovulation.

*Summary.* Ovulation was produced on Day 25 in immature rats hypophysectomized just prior to the administration of from 3 to 30 IU of PMS on Day 22 plus injection of 7.5 or 10 IU of HCG in the afternoon of Day 24. However, the incidence of ovulation did not show the biphasic dose-response as reported previously in intact immature rats treated with the same amounts of PMS. Administration of PMS alone or PMS plus 5 IU HCG did not induce ovulation in hypophysectomized immature rats.

1. Williams, P. C., *J. Endoc.* **4**, 137 (1945).
2. Carter, F., Woods, M. C., and Simpson, M. E., in "Control of Ovulation" (C. A. Villee, ed.), p. 1. Pergamon Press Inc., New York. (1961).
3. Greep, R. O., VanDyke, H. B., and Chow, R. F., *Endocrinology* **30**, 635 (1942).

4. Rowlands, I. W., and Williams, P. C., J. Endocrinol. **3**, 310 (1943).
5. Velardo, J. T., Science **131**, 356 (1960).
6. Ying, S. Y., and Meyer, R. K., Proc. Soc. Exp. Biol. Med. **130**, 40 (1969).
7. Malven, P. V., and Sawyer, C. H., Endocrinology **78**, 1259 (1966).
8. Lostroh, A. J., and Johnson, R. E., Endocrinology **79**, 991 (1966).
9. McCormack, C. E., and Meyer, R. K., Proc. Soc. Exp. Biol. Med. **110**, 343 (1962).
10. McCormack, C. E., and Meyer, R. K., Gen. Comp. Endocrinol. **3**, 300 (1963).
11. Zarrow, M. X., and Quinn, D. L., J. Endocrinol. **26**, 181 (1963).
12. Noble, R. L., Rowlands, I. W., Warwick, M. H., and Williams, P. C., J. Endocrinol. **1**, 22 (1939).
13. Rowlands, I. W., and Williams, P. C., J. Endocrinol. **2**, 380 (1940).
14. Rowlands, I. W., and Williams, D. C., J. Endocrinol. **3**, 310 (1943).
15. Smith, P. E., J. Anat. **45**, 205 (1930).
16. Parlow, A. F., in "Human Pituitary Gonadotropins" (C. Albert, ed.), p. 300. Charles C. Thomas, Springfield, Ill. (1961).

---

Received Nov. 3, 1971. P.S.E.B.M., 1972, Vol. 139.