Effects of Ether and Barbiturates on Serum LH Concentrations in Overiectomized Hamsters[1] (37549)

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In the female hamster serum LH levels are significantly elevated within 3.5 hr after gonadectomy (1). In contrast, serum LH increases much more slowly following ovariectomy in the adult rat (2). The rapid changes in circulating LH level, observed by Goldman and Porter (1) in ovariectomized hamsters, can probably be explained as equivalent to the periodic pulsatile discharges of LH from the pituitary of the ovariectomized rat (3). Anesthetic doses of pentobarbital lowered serum LH levels in etherized ovariectomized rats apparently by its action on the central nervous system (4). The present experiment examines the effects of barbiturates and ether on serum LH concentrations in ovariectomized hamsters. The doses of barbiturates and levels of ether anesthesia were those routinely used in our laboratory.

Materials and Methods. Adult female hamsters (*Mesocricetus auratus*) were maintained on a 14:10 hr lighting schedule with the lights on from 0500 to 1900. In Expt I the animals were anesthetized with ether and either ovariectomized or sham ovariectomized on diestrus (the day before proestrus). Blood was collected by heart puncture under light ether anesthesia at hourly and daily intervals thereafter. Recovery from this anesthesia occurred within 3 min. The blood was allowed to clot for 30 min at room temperature and serum was collected following centrifugation. Samples were stored at −20° until assayed for LH as previously described (5). All samples were assayed in duplicate and the values were averaged. LH is expressed in terms of NIAMD-RP-1 which has a biological potency equivalent to 0.03 × NIH-LH-S1.

In the second experiment, the hamsters were ovariectomized under ether anesthesia at least 3 wk before they were used experimentally. Pentobarbital sodium (8.0 mg/100 g ip), phenobarbital sodium (10 mg/100 g sc) and ether (60 min deep anesthesia) were administered to separate groups of animals immediately after initial blood samples had been withdrawn under light ether anesthesia. Subsequent samples were taken at 30, 60, 120 and 240 min under light ether anesthesia. Those animals treated with pentobarbital and phenobarbital were exposed to ether during blood collection for the same length of time as the noncannulated controls. Animals under deep ether anesthesia (0–60 min) recovered within 10 min after the ether cones had been removed at 60 min, and subsequent samples at 120 and 240 min were taken under light ether anesthesia. In one of the two untreated control groups, blood was collected via indwelling right atrial cannulae without ether anesthesia. In Expt II the blood was treated and assayed exactly as in the first experiment. Student's *t* test was used to analyze the data.

Results. Expt I. As summarized in Table I, within 4 hr after ovariectomy (ovx) there

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TABLE I. Serum LH Concentrations Following Ovariectomy and Sham Ovariectomy in the Hamster.[a]

After ovariectomy (hr)	Serum LH (ng/ml ± SE)	
	Ovariectomized	Sham ovariectomized
0	33 ± 2 (4)[b]	34 ± 11 (4)
1	17 ± 6 (3)	6 ± 1 (4)
2	55 ± 29 (4)	5 ± 1 (4)
4	400 ± 172 (3)	14 ± 5 (4)
8	102 ± 56 (4)	17 ± 4 (4)
24	161 ± 25 (6)	15 ± 1 (4)

[a] Surgery was performed on day 2 (diestrus) of the cycle.

[b] Number of animals.

was a significant increase in serum LH compared with concentrations in either pre-ovx or 4 hr sham ovx controls ($p < 0.05$). However, by 8 hr postcastration, the serum LH level had declined and was not significantly higher than either control value. By 24 hr post-ovx serum LH was again elevated ($p < 0.01$) over control values, and it continued to rise to a plateau of 400 to 600 ng/ml at 3 to 4 wk (Table II). Serum LH was extremely variable in individual samples from long-term (> 3 wk) ovx hamsters, with values ranging from 182 to 1760 ng/ml.

Expt II. As seen in Fig. 1 there was no significant change in serum LH concentration in either of the ovx control groups during the 4 hr period of the experiment. However, serum LH at 240 min in the cannulated group was greater than in the other control group in which blood was collected by heart puncture ($p < 0.05$). Deep ether anesthesia (0 to 60 min) resulted in a significant increase ($p < 0.05$) in serum LH above the ovx control value at 30 min, *i.e.*, 30 min before the end of exposure to ether, but there was no difference from controls at later times (Fig. 1). Both phenobarbital and pentobarbital lowered serum LH, but the depression caused by pentobarbital was significant only at the 60 min collection. At 120 min serum LH in the pentobarbital group was significantly ($p < 0.025$) higher than in the phenobarbital-treated animals. The depressing effect of phenobarbital on serum LH concentration was significant by 30 min ($p < 0.05$) and remained so during the rest of the experiment.

Discussion. The data in Table I showing an increase in serum LH within 4 hr after ovariectomy agree with observations of Goldman and Porter (1) who found LH to be elevated by 3.5 hr after ovariectomy. However, the rise was temporary and by 8 hr following ovariectomy serum LH was not statistically greater than in sham controls (Table I). By 24 hr postcastration, serum LH was again significantly elevated ($p < 0.05$) above controls, and it increased to a mean of 400 to 600 ng/ml by 3 to 4 wk (Table II). The transient increase in LH at 4 hr after castration has been suggested to be a result of the sudden withdrawal of estrogen (1) which was presumably elevated at the time ovariectomy was performed (6). The extreme variability in serum LH values found in long-term castrated hamsters suggests that this species might have a pulsatile-type release of pituitary LH similar to that reported for the rat (3).

All of the agents tested here for their effects on serum LH in ovx hamsters have blocked ovulation in the proestrous hamster when administered over a period of 6 hr (7). In the present study both pentobarbital and phenobarbital depressed serum LH while ether initially stimulated LH release in the

TABLE II. Changes in Serum LH Concentration Following Ovariectomy in the Hamster (Days 1–55).

After ovariectomy (days)	No. of animals	Serum LH (ng/ml ± SE)
1	6	161 ± 25
3	6	204 ± 60
5	13	216 ± 57
10	7	340 ± 76
15	7	250 ± 60
20	7	398 ± 112
25	7	366 ± 83
30	7	487 ± 55
35	6	576 ± 160
40	5	947 ± 298
45	5	624 ± 286
50	4	614 ± 93
55	3	435 ± 153

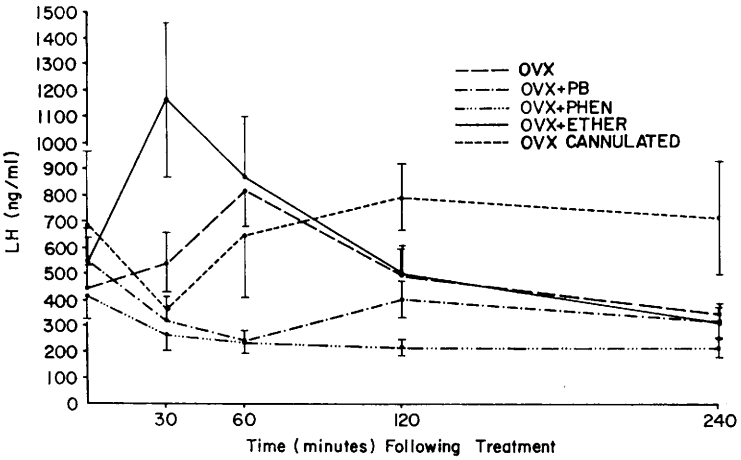

FIG. 1. Serum LH levels in ovariectomized hamsters after pentobarbital (PB) (8.0 mg/100 g), phenobarbital (PHEN) (10 mg/100 g) and ether (0 to 60 min) treatment. Controls include untreated and cannulated (right atrium) ovariectomized hamsters. Each point on the graph represents the mean (± SE) LH value for eight hamsters except in the cannulated group, which represents the mean of four hamsters.

ovx hamster. Another laboratory has recently reported a decrease in serum LH following pentobarbital administration in etherized ovx rats (4) and has suggested that this depressing effect on serum LH is exerted via the central nervous system (4). They also noted an initial elevation in serum LH following exposure to ether (4). Although ether is a rapidly acting anesthetic it is also a very potent stressing agent, and the initial elevation in serum LH is probably a response to stress in the hamster as in the rat (4). More prolonged exposure to ether or repeated dosages of pentobarbital might have depressed serum LH to the level achieved with phenobarbital. Neither the 1-hr exposure to ether nor the single injection of pentobarbital, although deeply anesthetizing the animals for more than 1 hr, would have blocked the proestrous surge of LH in the hamster (7).

In a previous communication, we reported that either pentobarbital or ether would block ovulation in the hamster but only if anesthesia was maintained for at least 6 hr on the afternoon of proestrus (7). If less than 6 hr, the LH surge was delayed for a period of time depending on the length of anesthesia, but not completely blocked. The single injection of phenobarbital at a dosage which does not cause the hamster to lose its righting reflex was effective here in lowering serum LH for long periods in the ovx animal and in blocking the ovulatory surge in the intact hamster (7). Although phenobarbital depressive action on LH release appears to be exerted at the level of the central nervous system (8) it is not anesthesia *per se* which inhibits pituitary LH release either in the ovx hamster in which the basic circulating levels of LH are high or in the proestrous animal with low serum concentrations of LH prior to the ovulatory surge (8).

It is of interest that in the long-term ovx hamster, in which the feedback action of ovarian steroids is no longer a factor in controlling LH release, the output of LH is still under nervous control and is responsive both to the stressing action of ether and the depressing action of nonanesthetic dosages of phenobarbital.

Summary. Serum LH showed a transient increase 4 hr after ovariectomy in the hamster but it declined by 8 hr. By 24 hr serum LH was again significantly elevated and it increased to a plateau of 400 to 600 ng/ml at 3 to 4 wk.

Doses of pentobarbital and phenobarbital that are commonly used in experimental procedures in the hamster depressed LH levels while ether anesthesia resulted in an eleva-

tion of this hormone in the blood. Deep anesthesia accompanied pentobarbital temporary suppression of serum LH. However, phenobarbital, which did not anesthetize the animals, had a much more pronounced depressive effect on serum LH levels. Therefore, the effects of these drugs on circulating LH do not appear to be related to their anesthetic action.

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