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Corpus Luteum and Estrous Cycle Influence on Physiological Properties of Ewe Utero-Ovarian Arteries (38270)

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The results of several investigations reviewed by Del Campo and Ginther (1) have indicated in the cow, ewe, and sow that a functional and anatomical relationship exists between the uterus and ovaries with the utero-ovarian vasculature serving as the link between these organs. Whereas most of the studies have been concerned with the possible role of the utero-ovarian vein and artery in the unilateral luteolytic phenomenon demonstrated in these animals (2–4), very little has been reported regarding the smooth muscle physiology of these vessels throughout the estrous cycle. Mattner and Thorburn (5) indicate there is a threefold increase in ovarian blood flow coincident with the luteal phase of the cycle in sheep. They found similar cyclic variations in blood flow for ovaries with or without corpora lutea and suggested that the tone of the ovarian vessels might be under the control of circulating progesterone levels. In contrast, other investigators have shown the estrogens to have a vasodilating effect on the uterine vessels in sheep (6–9).

We have found no reports regarding further investigations of possible interrelationships between the hormonal state and vascular smooth muscle properties of the reproductive system; however, it has been demonstrated that the effects of certain vasoactive substances may be modified by varying sex steroid concentrations in the body. Lloyd (10) and Hettiaratchi and Pickford (11) have shown that progesterone can reduce the vasoconstrictor effects of vasopressin, oxytocin, and angiotensin as measured systemically in the rat, whereas Boxill and Brown (12) have reported that the pressor response to epinephrine is augmented in dogs when progesterone is present. Chronic treatment of guinea pigs with testosterone has been demonstrated to reduce or abolish contractile and pressor responses of the isolated, superfused seminal vesicles to angiotensin, barium chloride, tyramine, epinephrine, and norepinephrine (13).

The intent of this investigation was to examine the responsiveness of the utero-ovarian arteries of the ewe during two opposing hormonal stages of the estrous cycle to the biogenic amines, norepinephrine, epinephrine, serotonin and to sympathetic nerve stimulation. Estrus (Day 0) and the luteal phase (Day 13) of the estrous cycle were chosen as meeting this hormonal criterion, with estrogen being the predominant circulating ovarian steroid during the former and progesterone during the latter stage.

Materials and Methods. Crossbred ewes were examined for estrus twice daily using vasectomized rams, with the first day of detected estrus designated as Day 0 of the cycle. Out of 15 ewes sacrificed, 12 ewes bearing a corpus luteum in only one ovary were assigned to two experimental groups representing Day 0 and Day 13 of the estrous cycle, with each group having six animals. Each ewe was sacrificed by exsanguination, and the ovarian vasculature with its accompanying ovary was excised from each side of the uterus. Note was taken of which ovary, right or left, contained the corpus luteum from the previous cycle in Day 0 animals and for the present cycle in Day 13 animals. A 3.5 cm portion of ovarian artery and its associated vein was sectioned immediately distal to the bifurcation of these vessels into ovarian and uterine components. The artery was cannulated with polyethylene tubing at both ends and the tissue mounted in a perfusion chamber, the experimental procedure being an adaptation of the isolated perfused rabbit ear artery preparation of Steinsland *et al.* (14). Perfusion systems were assem-

bled in duplicate so as to permit simultaneous treatment and monitoring of two arterial segments. Krebs bicarbonate solution equilibrated with 5% carbon dioxide in oxygen at 37° was delivered by two Harvard infusion pumps to both preparations with a perfusion rate of 17 ml/min and superfusion rate of 10 ml/min. Changes in perfusion pressure arising from the changes in the resistance to flow through the arterial segments were measured with Statham pressure transducers and recorded in mm Hg by a Gould Brush 280 two channel recorder.

Periarterial sympathetic nerves were excited by field stimulation. Square wave pulses of 1 msec duration and supramaximal voltage (60–70 v) were applied through bipolar platinum electrodes located at each end of the perfusion chambers, and delivered from a Grass Model SD5 stimulator. Electrical stimulation was applied in 30 sec trains separated by 5 min intervals at frequencies ranging incrementally from 5 to 30 Hz.

A Harvard dual syringe infusion pump was used to administer drug solutions at a rate of 0.17 ml/min into the perfusion cannulae. The concentrations (M) of the infused drugs l-norepinephrine bitartrate, l-epinephrine bitartrate, and serotonin creatinine sulfate were calculated for the perfusion fluid.

Standard statistical techniques were used to calculate means and standard errors of the means for each set of data. Student's t tests for unpaired and paired data were utilized to determine the statistical significance of observed differences.

Results. Figure 1 depicts the vasoconstrictor responses elicited in sections of utero-ovarian arteries by the biogenic amines, norepinephrine, epinephrine, and serotonin. A typical dose response relationship was demonstrated for each drug; however, transducer limitations prevented determination of the maximally effective dose for each of the drugs. These results suggest a tendency for arteries removed from ewes on Day 13 of the estrous cycle to be more responsive to

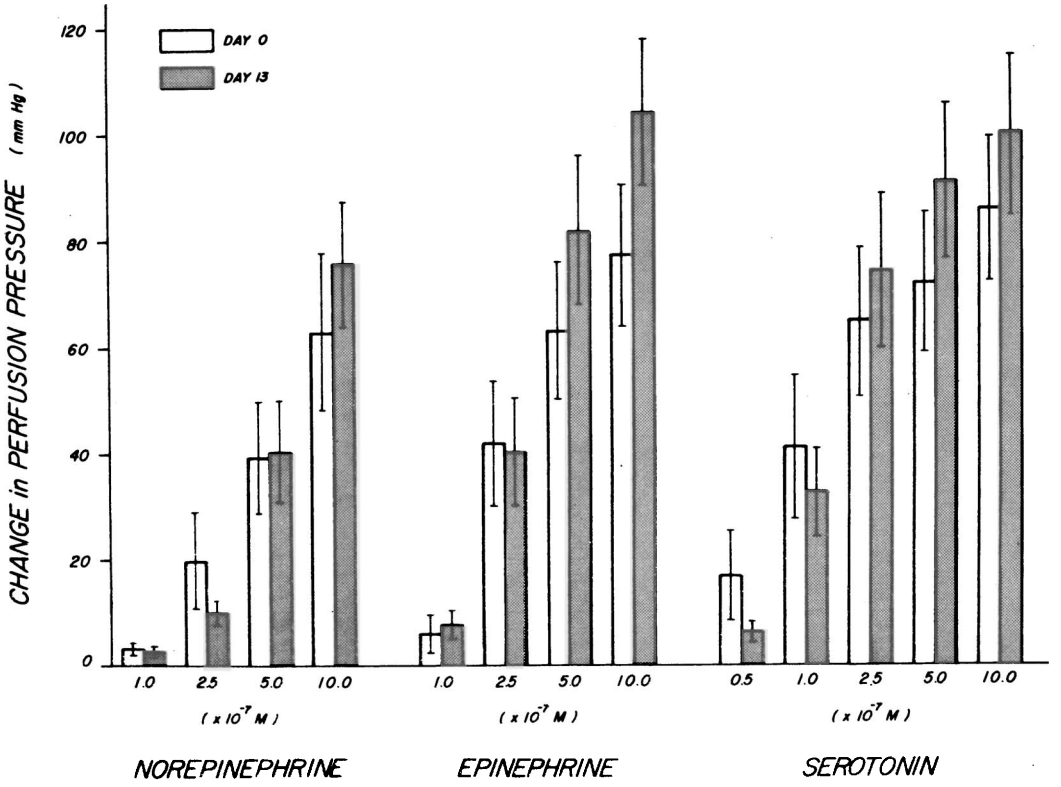

FIG 1. Vasoconstrictor responses of utero-ovarian arteries from ewes sacrificed on Day 0 (open bars) and Day 13 (shaded bars) of the estrous cycle to various doses of norepinephrine, epinephrine, and serotonin. Each bar represents the mean changes in perfusion pressure (mm Hg) ± SE for 12 arteries (two samples per animal in each group).

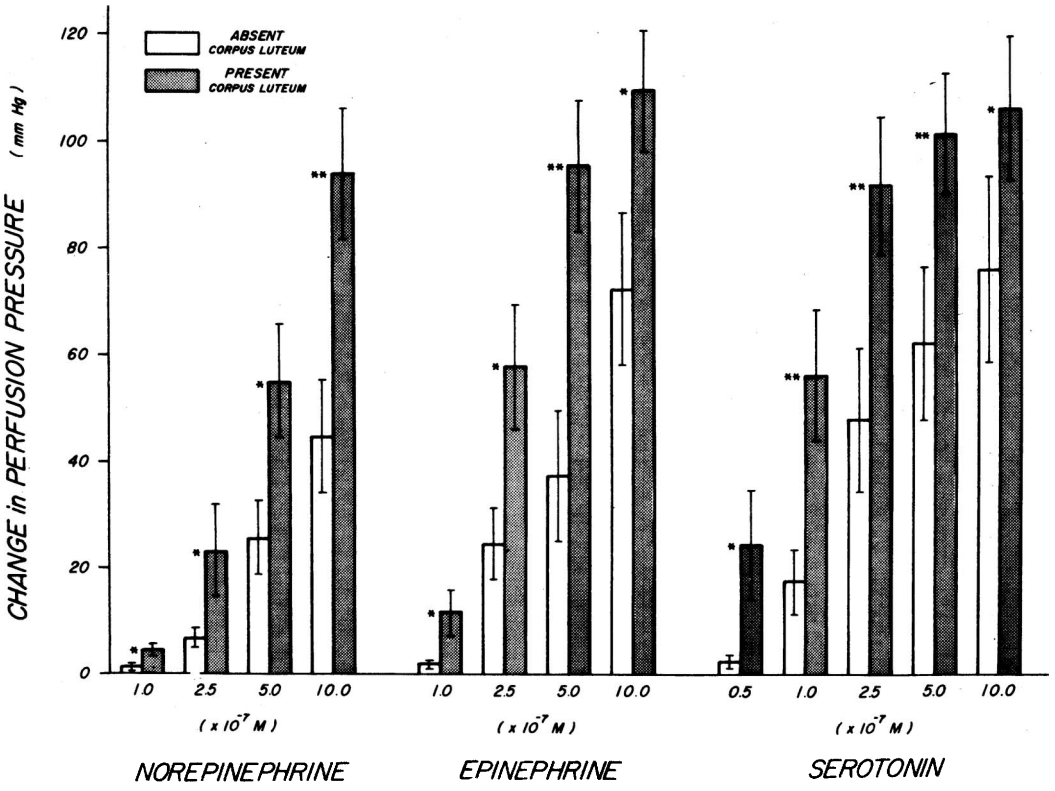

FIG. 2. Vasoconstrictor responses of utero-ovarian arteries from the side adjacent to the ovary without a corpus luteum (open bars) and from the side with the ovary bearing a corpus luteum (shaded bars) to various doses of norepinephrine, epinephrine, and serotonin. Each bar represents the mean changes in perfusion pressure (mm Hg) ± SE for 12 arteries. $^{**}P < 0.01$; $^{*}P < 0.05$.

the biogenic amines than are arterial segments from ewes sacrificed on Day 0 of their cycle. Further evaluation of these data reveal that the magnitudes of vasoconstrictor responses for arteries taken from the same animal were noticeably different for most of the ewes, suggesting that presence of the corpus luteum might be of importance. For all six ewes sacrificed on Day 13 of the estrous cycle, there was a correlation between corpus luteum location and the side from which the more responsive artery was sectioned, whereas in the Day 0 group, there were two of six ewes in which the more responsive artery came from the ovary not bearing the corpus luteum of the previous cycle. Quantitative vasoconstrictor response data for the utero-ovarian arteries classified according to corpus luteum location are presented in Fig. 2. Changes in perfusion pressure produced by each of the three biogenic amines were significantly greater ($P < 0.05$) for the arteries removed from the side adjacent to ovaries having corpora lutea.

Analysis of baseline perfusion pressure recorded for each artery following a one-hour initial equilibration period showed this pressure to be significantly higher ($P < 0.01$) for the arteries sectioned from the luteal side. Mean equilibrated baseline pressure for Day 0 and Day 13 arteries associated with the corpus luteum was 68.0 ± 2.1 mm Hg compared with that of 62.8 ± 1.7 mm Hg recorded for the arteries adjacent to ovaries not bearing corpora lutea. When compared on Day 0 and Day 13, disregarding corpus luteum location, the mean initial perfusion pressures at the arteries were 64.4 ± 2.2 mm Hg and 66.4 ± 1.9 mm Hg, respectively.

Preliminary data for sympathetic nerve stimulation with bipolar electrodes agrees with vascular responsiveness data for the biogenic amines. For the arteries tested from the Day 13 group, greater vasoconstrictor responses were elicited from the vessels removed from the corpus luteum side of the ewe. The association between luteal body and arteries responding with greater

vasoconstriction was not well defined for ewe from the Day 0 group. Comparison of the magnitude of responses of arteries grouped as Day 0 and Day 13 was inconclusive.

Discussion. Contrary to the findings of Mattner and Thorburn (5), the results of this investigation strongly suggest that the corpus luteum exerts some influence on the vascular properties of at least the utero-ovarian artery serving the ovary which bears it. The data indicate a greater responsiveness of this vessel to biogenic amines and sympathetic nerve stimulation, and the baseline tone of this particular artery is greater than that of its counterpart. Our findings suggest that at some point near Day 0, the influence of the previous corpus luteum on the associated utero-ovarian artery diminishes, whereas on Day 13 of the cycle there is definite luteal influence.

The mechanism by which the corpus luteum appears to exert a local effect on the artery serving it is undefined. It is, perhaps, another example of a counter-current transfer mechanism which is currently being argued to exist for the luteolytic process in these animals (4). The luteal body may release some substance into the ovarian vein, and this substance might then be transferred to the utero-ovarian artery at some distal point via a counter-current mechanism. Anatomically, such a process is feasible.

Another finding which deserves consideration is that of the tendency for arteries removed from ewes on Day 13 of the estrous cycle to be more responsive to norepinephrine, epinephrine, and serotonin. This agrees in some respects with the findings reported by Boxill and Brown (12). While these data collected on this day did not show the differences to be statistically significant, this might be the result of the small sample size as well as within animal variation associated with the corpus luteum.

Summary. An association between corpora lutea and physiological responses of the vascular smooth muscle of the utero-ovarian arteries was demonstrated. Arteries serving the ovaries bearing corpora lutea responded to norepinephrine, epinephrine, and serotonin with significantly greater changes in perfusion pressure than did the arteries associated with ovaries not having a luteal body. Vasoconstrictor responsiveness to periarterial sympathetic nerve stimulation agreed with these findings. In addition, baseline perfusion pressures were elevated for the arteries from the side adjacent to the ovary with the corpus luteum compared to values for the arteries from the other ovary. The arteries removed during the luteal phase (Day 13) of the estrous cycle, however, did not elicit significantly greater responses to vasoactive biogenic amines than the arteries removed at the time of ovulation (Day 0).

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