SYMPOSIUM: ESTRUS, NEW DEVICES, AND MONITORING

Endocrine and Neural Control of Estrus in Dairy Cows

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ABSTRACT

During proestrus, gonadotropins induce final follicular maturation, resulting in increased secretion of estradiol. Estradiol, in the relative absence of progesterone, acts on the hypothalamus to induce estrous behavior. The mean duration of estrus is 12 to 16 h and ranges from 3 to 28 h. The effects of estradiol appear to be "all or none". That is, once a threshold of estradiol is achieved, estrus is induced, and additional amounts of estradiol above threshold do not further enhance the estrous response (duration and intensity of estrus). Also, progesterone can block the estrus-inducing actions of estradiol. In addition, prior exposure to progesterone does not potentiate the estrus-inducing actions of estradiol except in the early postpartum period. In dairy cows, the first postpartum ovulation is often "silent". In other words, ovulation is not preceded by estrous behavior. High levels of estradiol during late gestation apparently induce a refractory state such that the brain cannot respond to the estrus-inducing actions of estradiol at the first postpartum ovulation. Progesterone can "reset" the brain, allowing it to respond to subsequent estradiol exposure. In the case of the postpartum cow, the corpus luteum formed after the first ovulation provides the progesterone that resets the brain. As a consequence, the second postpartum ovulation is preceded by estrous behavior. Finally, stress (or injection of ACTH) has been shown to delay, shorten, or inhibit completely the expression of estrus in the presence of estrus-inducing concentrations of estradiol. In summary, estrus is induced by estradiol in the absence of progesterone, progesterone is inhibitory to estrus, and situations exist in which estrus may be absent prior to ovulation. (Key words: estrus, estrous behavior, hormones, neural control)

INTRODUCTION

Understanding of the endocrine and neural factors that control estrus facilitates the construction of strategies aimed at improving the detection of estrous behavior. A concept map on estrus in cattle has been assembled (Figure 1) and should be referred to frequently during the reading of this paper to assist comprehension of the concepts included in the map. Interested readers seeking further information are referred to excellent reviews on estrous behavior and detection of estrus in cattle (1) and on reproductive physiology and endocrinology of cattle (27, 29). The objective of this paper is to focus on endocrine and neural factors that are responsible for the induction of estrus in cattle. Recent information on inhibitory influences on estrus are included, and citations have been limited largely to cattle but some supporting contributions on other domestic animals are included. Because data concerning reproductive behavior in rodents often conflicts with data from domestic animals, that voluminous research area has been ignored.

ROLE OF ESTRADIOL

During proestrus, a preovulatory follicle develops on one of the two ovaries. Under the influence of pituitary gonadotropins, this follicle secretes large amounts of estrogens, which, in turn, cause plasma concentrations of the primary estrogen, estradiol, to reach peak levels (17, 18, 33, 55, 56). The onset of estrous behavior (estrus, heat) and the surge release of
LH are coincident with this peak in plasma estradiol (11, 28, 42, 51, 66).

Estrus is induced by action of estradiol upon the hypothalamus (Figure 1) in the relative absence of progesterone (8, 12). The induction of estrus by estradiol has been demonstrated in ovariectomized cattle (5, 10, 35, 41, 59), horses (3), sheep (22, 23), and pigs (25).

Immunization of cattle (43) and sheep (24) against estradiol inhibited the expression of estrous behavior in spite of the presence of estrus-inducing serum concentrations of estradiol. Apparently, the antibodies directed against estradiol effectively neutralized the hormone. Because the antibodies specifically bind estradiol and no other hormones, this evidence lends excellent support to the hypothesis that estradiol is indeed the endogenous hormone that induces estrus. In addition, low concentrations of estradiol following prostaglandin administration in cattle treated with human chorionic gonadotropin failed to induce estrus (7).

The effects of estradiol in inducing estrus appear to be "all or none" (Figure 1). That is, once the concentration of plasma estradiol is sufficient to induce estrus, additional amounts have no further stimulatory influence on expression of the behavior. However, additional amounts may be required for reproductive tract function, such as secretion of specific proteins required to nourish the early embryo. This all-or-none effect has been demonstrated both in intact (13, 28, 66) and ovariectomized cattle (14, 52). In addition, when low concentrations of estradiol are administered and <50% of the ovariectomized cattle are induced into estrus, those cows in estrus display a frequency of various behaviors similar to those of cows receiving markedly higher dosages (14).

![Concept map of estrus in dairy cattle. MBH = Medial basal hypothalamus, CL = corpus luteum, and CRF = corticotropin-releasing hormone.](image)
As a point of interest, body weight of the ovariectomized cow can apparently be ignored when estrus is induced by estradiol injection. In previous studies conducted (2, 14, 15, 30), .5 mg of the hormone induced estrus in cows ranging from 450 to 820 kg in body weight. Most other researchers also use an absolute dose of estradiol rather than a dosage based upon body weight of the animal.

ROLE OF PROGESTERONE

Serum concentrations of progesterone are very low during proestrus and estrus (34, 42, 63), which is a necessary prerequisite to the expression of estrus because progesterone is clearly inhibitory to estrous behavior (3, 16, 21, 25). As with the actions of estradiol, progesterone appears to have an "all-or-none" effect on the inhibition of estrus (Figure 1). That is, once progesterone concentrations increase to a threshold level, estrus is inhibited even when estrus-inducing concentrations of estradiol exist (16, 21, 50, 64). In other words, actions of progesterone have priority over those of estradiol in controlling estrous behavior. Progesterone administered near the onset of estrus has also been shown to prevent the preovulatory surge of LH (40).

There is disagreement in the literature in regard to the role of prior exposure to progesterone facilitating estradiol's estrus-inducing actions. Some researchers have found enhancement of estradiol actions when a progesterone pretreatment is used (3, 21, 46), but others have not (2, 10, 16, 25, 64). The majority of recently conducted studies on cattle have shown that pretreatment with progesterone does not potentiate the actions of estradiol, but recent studies on sheep have shown that pretreatment with progesterone does potentiate actions of estradiol. In sheep, the estrous cycle can be shortened (to 8 d) by prostaglandin injections up to eight consecutive times without causing any alterations in estrous behavior or ovarian function (44). Hence, the length of progesterone exposure required to enhance estradiol's actions in this species is considerably shorter than the length of a normal luteal phase. No comprehensive studies have been conducted in estradiol-treated cattle to evaluate the role of pretreatment with progesterone and its influence on other physiological factors, such as sperm transport and secretions of the entire reproductive tract.

ROLE OF OTHER HORMONES

Testosterone alone can induce estrous behavior in cattle, but large amounts are required (100 to 400 mg), and the response is never as pronounced as when using estradiol (35). The actions of testosterone may be related to its metabolism to estradiol in the body. When testosterone was given concurrently with amounts of estradiol too low to induce estrus, no potentiation was noted, and no animals expressed estrus (49). Similarly, when testosterone was given concurrently with estrus-inducing concentrations of estradiol, no potentiation or inhibition was noted (2). Testosterone alone can also induce a shortened period of estrous behavior in sheep, and the hormone does not appear to facilitate or inhibit actions of estradiol when both are given concurrently (22).

Two additional androgens have been evaluated in cattle for their ability to induce estrus. Androstenedione (which can be aromatized to estradiol) and dihydrotestosterone (which cannot be aromatized to estradiol) were unable to induce estrous behavior in ovariectomized cattle (35).

Cortisol (up to 200 mg), when given concurrently with estrus-inducing amounts of estradiol, was unable to alter any estrous trait (15). In the same study, however, 4 mg of dexamethasone (a synthetic glucocorticoid) reduced the percentage of heifers in estrus. The inhibitory effect of 4 mg of dexamethasone was confirmed in a subsequent study by the same researchers (2). Dexamethasone also inhibited estrus in horses (4), sheep (20), and swine (26). Infusion of cortisol for 90 h to proestrous heifers completely blocked the LH surge and estrous behavior without altering plasma estradiol concentrations (6). Repeated stress, which elevated total corticosteroid concentrations during the follicular phase, prevented the surge release of LH in some heifers but did not interfere with the expression of estrus (61).

Administration of ACTH (800 units total) to heifers over a 3.5-d period, during proestrus, delayed the LH surge and the onset of estrus.
Standing estrus was shortened (50% of controls) after ACTH administration ceased. Also, in that study, plasma estradiol concentrations were decreased, but plasma cortisol and progesterone were increased during ACTH treatment. In a study that used only one ACTH injection (320 units) during proestrus, ACTH delayed the onset and shortened the duration of estrus in heifers (30). However, peak serum estradiol concentrations were not altered, but peak serum progesterone and cortisol concentrations were fourfold higher than controls. In the same study, an injection of 320 units of ACTH prevented estrus in the majority of estradiol-treated, ovariectomized cows. Again, peak estradiol concentrations were not altered, but progesterone and cortisol concentrations were elevated.

Treatment with GnRH was not able to potentiate the actions of estradiol (2, 14) even when estradiol was administered in subthreshold amounts (amounts not able to induce estrus in the majority of ovariectomized cattle). In sheep, elevated oxytocin in the hypothalamus has been hypothesized to mediate the decrease in length of sexual receptivity that occurs after repeated intromissions (36). The hypothalamic concentrations of the monoamines (norepinephrine, dopamine, and serotonin) in cattle did not change between the onset of estrus and 48 h later (62).

ESTRUS WITH OVULATION

During the normal estrous cycle of cattle, the onset of estrus precedes ovulation by 24 to 30 h. The duration of estrus is usually 12 to 16 h, but individual variation is great, and the effective range is from 3 to 28 h (1, 32). As a result, ovulation occurs approximately 12 h after the termination of estrus. Subsequent to ovulation, the corpus luteum forms and secretes progesterone during the luteal phase of the estrous cycle. After reaching puberty, cattle continue to have estrous cycles that are 21 d in length unless they are pregnant, and then all cycles cease until after parturition. The following two sections document situations in which estrus and ovulation are not always associated together as described in this paragraph.

ESTRUS WITHOUT OVULATION

In heifers, instances of nonpuberal estrus have been reported (48, 53). This condition is characterized as behavioral estrus not followed by ovulation and subsequent formation of the corpus luteum. Because this condition may affect up to 60% of the heifers, caution should be exercised when puberty (usually considered the first estrus with ovulation) is determined in groups of animals. Fortunately, nonpuberal estrus usually involves the first behavioral estrus noted in heifers near puberty and not heifers of breeding age. Interestingly, most heifers that have the first behavioral estrus followed by ovulation and corpus luteum formation experienced an elevation in plasma progesterone concentrations prior to their first estrus (53). Nonpuberal estrus has also been reported in sheep (19).

Treatment of ovariectomized cattle with Syncro-Mate B® to synchronize estrus has been reported to induce estrus in about 50% of the cattle (45). This result is obviously a case of estrus without ovulation. This research was initiated to determine causes of reduced fertility after treatment of normal cattle with the product. Therefore, cattle in estrus after treatment with Syncro-Mate B® may either be in true estrus (which will be followed by an ovulation) or they may be in false estrus (which will not be followed by an ovulation). Adequate residual estradiol is very likely to be present at the time of the norgestomet ear implant removal (9 d after the 5 mg estradiol valerate injection) to induce estrus (6, 38). This notion is supported by the research of Vynckier et al. (65), which reported that plasma concentrations of estradiol remain elevated for as long as 9.5 d after injection of 10 mg estradiol cypionate.

OVULATION WITHOUT ESTRUS

After parturition, cows enter a period of postpartum anestrus. The end of the anestrus is marked by the first postpartum ovulation. In dairy cattle, the first postpartum ovulation is often not associated with estrous behavior. Hence, the term “silent” ovulation is used to describe this event. The reported incidence of silent ovulation at the first postpartum ovulation ranges from 50 to 94% (37, 39, 54, 57); the rate is dependent upon frequency of detection of estrus. At the second and subsequent ovulations, the majority of cattle are detected in estrus.
A current theory on silent ovulations (39) implicates the involvement of high concentrations of estradiol at the end of pregnancy. These high concentrations of estradiol (31, 58) are postulated to induce a state of refractoriness (at the hypothalamus) to the estrus-inducing concentrations of estradiol present at the first postpartum ovulation. Perhaps other steroids may also play a role in inducing the refractory state (47). Progesterone released during the first luteal phase (that follows the silent ovulation) then removes the refractory state (Figure 1) so that the second ovulation is associated with estrus. This theory is based on the work of Carrick and Shelton (10) that used high estradiol concentrations to induce a refractory state (Figure 1) in ovariectomized heifers and reported that progesterone was able to restore the estrous response to normal estrus-inducing concentrations of estradiol. Kyle et al. (39) have attempted to improve the incidence of estrous behavior at the first postpartum ovulation by treating cattle with progesterone soon after calving (and before return to cyclic ovarian function). Progesterone treatment tended to increase the percentage of cattle detected in estrus at the first postpartum ovulation. However, using a fabricated intravaginal device that released progesterone, we were unable to increase serum concentrations of progesterone for the planned 5-d treatment period. For progesterone treatment to be effective at this time, the author suggests that luteal progesterone concentrations be maintained for approximately 5 d.

Other theories may also be used to explain silent ovulations. For example, no studies have documented the amount of estradiol produced at the first, second, and third postpartum ovulations. Decreased estradiol production at the first postpartum ovulation (compared with subsequent ovulations) could account for the absence of estrous behavior. Another possibility is that the length of estrus is shorter at the first postpartum ovulation and, therefore, is detected less frequently. Still another possibility is that the hypothalamus is less sensitive to estradiol at the first postpartum ovulation and increases in sensitivity during the postpartum interval. One study has evaluated this subject by injecting a constant amount of estradiol into ovariectomized lactating cows at six different times during the postpartum period and found no difference in the estrous response (9). This result would argue against a change in sensitivity of the hypothalamus during the postpartum interval.

**SUMMARY**

Estradiol, in the relative absence of progesterone, acts on the hypothalamus to induce estrous behavior. The estrus-inducing actions of estradiol appear to have an "all-or-none" effect. That is, once a sufficient concentration of plasma estradiol is achieved to induce estrus, additional amounts have no further stimulatory influence on expression of the behavior. Progesterone is clearly inhibitory to estrous behavior, and actions of progesterone have priority over those of estradiol. Stress (modeled with injections of ACTH) has been shown to delay, to shorten, or to inhibit completely the expression of estrus in the presence of estrus-inducing concentrations of estradiol. The majority of dairy cows have a silent first postpartum ovulation. Progesterone, secreted during the first postpartum luteal phase, removes the refractory state responsible for the silent ovulation allowing the second (and subsequent) postpartum ovulation to be associated with estrus.

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