Characterization of Ovarian Follicular Cysts and Associated Endocrine Profiles in Dairy Cows


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ABSTRACT

It is generally accepted that ovarian follicular cysts (cysts) are nonovulatory follicular structures that contribute to extended calving intervals. Follicle/cyst dynamics and the etiology of cysts are unclear. The present study was conducted to characterize follicle/cyst dynamics and to define endocrine changes (etiology) associated with cyst development. Thirty-two dairy cows were studied: controls (n = 8), cows with spontaneously occurring cysts (n = 14), and cows in which cysts were induced by exogenous steroid treatment (n = 12). Ovaries of cows were scanned daily by ultrasonography to record follicle/cyst dynamics. Blood was collected to determine endocrine changes associated with follicle/cyst life span. Three ovarian responses in cows with cysts were observed: persistence of cysts, turnover of cysts, or spontaneous recovery (self-recovered). Mean maximum size of cysts was larger (p < 0.05) than that of ovulatory follicles (2.80 ± 0.19 vs. 1.60 ± 0.05 cm). Mean interval from initial detection of follicle/cyst wave to detection of a new follicle/cyst wave in cows with cysts was longer (13.0 ± 1.1 days; p < 0.05) and more variable (6 to 26 days; p < 0.05) than in controls (8.5 ± 0.5 days and 6 to 14 days, respectively). Cysts grow at the same rate as follicles but continued to grow for a longer period of time. A transient increase in FSH preceded detection of all follicle/cyst waves. Concentrations of LH (p < 0.01) and estradiol-17β (p < 0.05) were greater in cows with cysts than in cows in ovulatory cycles around the time of wave detection and around the time of ovulation or at the time when cysts reached a diameter equivalent to that of follicles that ovulated. In conclusion, cysts and follicles are dynamic structures, with cysts having longer intervals between wave detections compared to the interval in cows with normal estrous cycles. Greater concentrations of estradiol-17β and LH, but not FSH, were associated with development and persistence of cysts.

INTRODUCTION

Ovarian follicular cysts (cysts) are nonovulatory follicular structures that contribute to an extended calving interval in dairy cattle [1]. Early work suggested that cysts remain as long as the condition persists, since cysts are nonovulatory [2]. This view of cyst persistence is contrary to our knowledge of the dynamic nature and turnover of ovarian follicles [3–5]. Ovulatory estrous cycles in cattle are characterized by the occurrence of two, three, or four waves of follicular growth. Keisler et al. [6] first suggested that cysts may regress and new follicular structures develop into other nonovulatory cysts. In a recent study, cysts were marked so that their fate and turnover could be monitored [7]. In some cases (3 of 23), cysts persisted for as long as 40 days. Additional follicular development accompanied the persistent structures. Most cysts (13 of 23), however, regressed or turned over and were replaced by new cysts. In 5 cows that resumed ovarian cyclicity (ovulated), none of the marked cysts ovulated. Although others have reported the occurrence of turnover, persistence, and spontaneous self-recovery of cysts in early postpartum dairy cows [7, 8], no reports have correlated endocrine changes with follicle/cyst dynamics.

Real-time ultrasonographic examination of ovaries has allowed individual follicles to be monitored on a daily basis in a noninvasive manner. Neither the turnover interval nor the endocrine factors associated with turnover, self-recovery, or persistence of cysts were known. Therefore, the objectives of this study were to 1) characterize growth and turnover rate of follicular structures that develop into cysts and subsequently self-recover and 2) characterize endocrine changes associated with persistence, turnover, or spontaneous recovery of cysts in cows.

MATERIALS AND METHODS

Animals

Thirty-two dairy cows were used and were assigned to groups as illustrated in Figure 1. Cysts were induced in 12 cows with twice-daily subcutaneous injections of estradiol-17β (15 mg) and progesterone (37.5 mg) for 7 days [7]. Spontaneously occurring cysts were diagnosed in 14 cows. Six cows exhibiting normal estrous cycles were administered 25 mg prostaglandin F2α (PGF2α) to synchronize estrus and were used as controls.

Ultrasonography and Blood Collection

Ovaries were scanned by ultrasonography using a real-time, B mode instrument equipped with a 7.5-MHz linear-array intrarectal transducer (Aloka 210; Corometrics Medical Systems, Wallingford, CT). Ultrasound examinations were performed by a single operator. Diameters of follicles/cysts and CL were measured to the nearest 0.5 mm by means of electronic calipers located on the ultrasound device.

In cows in which cysts were induced, ultrasonography
was performed twice weekly for detection of follicular growth after termination of steroid treatment. After follicular growth was detected, ultrasonographic examination of the ovaries was conducted daily. Initiation of a follicle/cyst wave was determined by detection of the follicles in a cohort. Follicles 5 mm or greater in diameter could be progressively monitored on a daily basis. Follicles were observed from initiation of detection (5 mm) until they were no longer detectable (< 5 mm). Spontaneously occurring cysts were initially detected via rectal palpation and confirmed by ultrasonography in cows during routine weekly herd health examinations at the University of Missouri Dairy Farm. Therefore, all spontaneously occurring cysts could have been in existence for at least a week before cows were placed on the study. Verification of a cyst was based on the finding of a follicular structure 2.0 cm or greater in diameter in the absence of a CL. Ovaries of cows with cysts (spontaneously occurring) and ovaries of cows exhibiting ovulatory estrous cycles (controls) were scanned daily by ultrasonography during the experimental period.

Scanning was terminated in cows with cysts after they resumed cyclicity (ovulation; self-recovery) or in cows that exhibited at least two cyst waves or persistence of a single cyst for at least 30 days. Observations for control and some self-recovery cows (n = 8) were recorded from the time of PGF2α-induced estrus or of the estrus associated with spontaneous recovery until the subsequent estrus and ovulation. In other cases in the self-recovery group, observation was terminated after ovulation and early CL development.

Three blood samples were collected at 15-min intervals twice daily during the experimental period, and serum was stored at −20°C until concentrations of FSH, LH, estradiol-17β, and progesterone were determined.

**Endocrine Analyses**

One serum sample from the morning sampling period was analyzed by RIA for serum concentrations of progesterone [9] and estradiol-17β [10]. The intra- and interassay coefficients of variation (CV) for progesterone and estradiol-17β were 13.7% and 12.6%, and 12.3% and 19.8%, respectively. Two serum samples, one from each morning and evening, were analyzed for concentrations of FSH [11] using NIAMDD-oFSH-RP-1 as the reference preparation (r = 0.99; [12]). The intra- and interassay CVs for FSH were 9.5% and 14.4%, respectively. All samples were analyzed for LH [13] by means of RAO.LH TEA #35 antibody (J.J. Reaves, Washington State University, Pullman, WA) 34 (r = 0.99; [14]). The intra- and interassay CVs for LH were 6.0% and 10.7%, respectively.

**Statistical Analyses**

Student's t-tests for unequal observations were used for comparisons between follicle/cyst size, estrous cycle length, and interval between detection of follicle/cyst waves. Data from cysts were used in the analysis only if a follicle/cyst plateaued in growth (diameter did not increase more than 0.15 cm for 3 consecutive days).

Endocrine data were examined using the Statistical Analysis System General Linear Model procedures [15] for a factorial split plot over time ANOVA for repeated measures [16]. Two separate analyses were performed. In the first, endocrine measures were compared among control, self-recovery, and cyst groups during the period preceding (−4 to 0 days) and following (0 to +4 days) follicle/cyst wave detection. Day of follicle/cyst wave detection was designated as Day 0. The second analysis consisted of congruent data examined relative to the day on which ovulation occurred in the control and self-recovery groups and relative to the day (Day 0) on which cysts reached 1.60 cm in diameter in the cyst group. The size of 1.60 cm was chosen because this was the average largest diameter of ovulatory follicles in cows that ovulated. The period analyzed included that preceding (−4 to 0 days) and following (0 to +4 days) time of attainment of mean ovulatory follicle size (Day 0). Additional comparisons were made with dominant nonovulatory follicles in the self-recovery and control groups. Day 0 for nonovulatory follicles was based on the day that dominant nonovulatory follicles from each cohort reached 1.60 cm in diameter or maximum size. Treatment groups, time, cows, and follicular/cyst waves were the main effects in both analyses. A preovulatory surge of LH was presumed to have occurred if a consecutive increase or decrease in LH was observed in the three morning or evening samples with at least one measurement exceeding 5 ng/ml [17]. When concentrations of LH were in an ascending or descending mode and greater than 5.0 ng/ml, the values were omitted from both analyses to prevent confounding effects of the preovulatory LH surge in the analysis.

Comparisons were made through use of linear regression analyses performed on circulating concentrations of LH and estradiol-17β prior to the time of spontaneous recovery or cyst wave detection in the self-recovery group and the cyst group, respectively. The self-recovery group was divided further into two groups based on concentrations of progesterone: 1) < 1 ng/ml progesterone prior to detection of the ovulatory follicle (n = 5) and 2) > 1 ng/ml progesterone prior to detection of the ovulatory follicle (n = 13). Comparisons were made beginning from the initial observation of each cow to the day the ovulatory follicle was detected in the self-recovery group and from initiation of the study to day of detection of the last cyst observed in each cow in the cyst group.

**RESULTS**

**General**

Of 12 cows with steroid-induced cysts, 7 spontaneously ovulated (self-recovered) during the experimental period.
The remaining 5 cows displayed either turnover or persistence of cysts throughout the experimental period. Of these 5 cows, 2 displayed persistent cysts during the experimental period. Of the 14 cows with spontaneously occurring cysts, 11 ovulated a new follicular structure. The remaining 3 cows displayed either turnover or persistence of follicles/cysts. One of these cows displayed persistent cysts throughout the experimental period. Follicle/cyst turnover was not observed prior to self-recovery in either group.

**Ovarian Structures**

**Follicle/cyst dynamics.** The first cows that spontaneously ovulated in both the steroid-induced cyst (n = 4) and spontaneously occurring cyst (n = 4) groups were monitored from self-recovery ovulation to subsequent ovulation. Similar ovarian structures were observed during estrous cycles in the controls (n = 6). Therefore, data from ovulatory estrous cycles in control (n = 6) and self-recovered (steroid-induced, n = 4; spontaneously occurring cysts, n = 4) cows were combined (Fig. 1). Eight of these 14 cows displayed 3 waves of follicular growth during the estrous cycle following ovulation (example in Fig. 2). The remaining 6 cows had 2 waves of follicular growth. Initiation of growth of waves of follicles (> 5 mm) was detected on Days 1.7, 9.5, and 16.9 during estrous cycles with 3 waves of follicular growth and on Days 1.3 and 12.0 during estrous cycles with 2 waves of follicular growth. Estrous cycle length for cows with 3 follicular waves tended to be longer (p = 0.09; 23.3 ± 1.3 days) than for cows having estrous cycles with 2 waves of follicular growth (20.8 ± 0.5 days).

**Follicular structures in cows with cysts.** Thirty-one observations of cyst turnover were obtained from 8 cows. Of 31 follicle/cyst waves, 28 were monitored to completion of the follicle/cyst growth plateau. Mean cyst size and time interval between detection of follicle/cyst waves were similar in cows with steroid-induced and spontaneously occurring cysts; therefore, these data were combined. Mean maximum size of cysts was 2.80 ± 0.19 cm in diameter, which was larger (p < 0.05) than the mean maximum diameter (1.60 ± 0.05 cm) of ovulatory follicles (Table 1). Of 28 cysts, 27 were larger than the average diameter of ovulatory follicles. The single remaining cyst was 1.50 cm in diameter.

Mean length of time from initial detection of follicle/cyst waves to detection of a new follicle/cyst wave in cows with cysts was longer (p < 0.05; 13.0 ± 1.1 days) and was more variable (range 6–26 days) than in controls (8.5 ± 0.5 days; range 6–14 days). Of 28 intervals in time between detection of a follicle/cyst wave and subsequent detection of a new follicle/cyst wave in the cyst group, 21 were longer (15.1 ± 0.9 days; p < 0.05) than the average interval in time between the detection of a follicular wave and subsequent detection of a new follicular wave in the control and self-recovery groups. The remaining 7 time intervals between follicle/cyst waves tended to be shorter (6.6 ± 0.5 days; p = 0.10) in the cyst group than the average interval between detection of follicular waves in the control and self-recovery groups. Cysts took 7.2 ± 0.3 days from wave detection to reach a comparative mean ovulatory size of 1.60 cm—an interval similar to the interval in time from day of detection of ovulatory follicles to ovulation (6.9 ± 0.4 days). Cysts ultimately reached their maximum size 12.7 ± 0.8 days from initial detection of a follicle/cyst wave.

Three cows had cysts that remained present for the duration of the period studied (persistence; Fig. 1). The persistent cysts remained the largest structure on the ovaries in two of three cases, and mean maximum size in the three cows was 4.20 ± 0.8 cm. Although these cysts were present, additional follicle/cyst growth and development continued. The interval between detection of a follicle/cyst wave and a new follicle/cyst wave in cows with persistent cysts was 12.6 ± 1.9 days. In one case, however, no other follicle/cyst growth was detectable for 25 days (Fig. 3). In this cow (cow 800), follicle/cyst growth and development resumed after 25 days as evidenced by follicle/cyst waves.

The first cows that spontaneously ovulated on Days 1.7, 9.5, and 16.9 during estrous cycles with 3 waves of follicular growth and on Days 1.3 and 12.0 during estrous cycles with 2 waves of follicular growth. Estrous cycle length for cows with 3 follicular waves tended to be longer (p = 0.09; 23.3 ± 1.3 days) than for cows having estrous cycles with 2 waves of follicular growth (20.8 ± 0.5 days).

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of follicles/cysts of the waves occurring concurrently with the persistent cyst in cow 800 ranged from 1.50 to 3.20 cm. The persistent cyst was finally replaced on Day 60 of observation by another cyst that reached 5.60 cm in diameter.

In cows that spontaneously recovered, cysts were 2.80 ± 0.13 cm at the time the follicle destined to ovulate was first detected (5 mm). Ovulation occurred an average of 7.5 days after detection of the ovulatory follicle in the self-recovery group. Cysts decreased \( (p < 0.05) \) in size from 2.80 ± 0.13 cm to 2.28 ± 0.17 cm during this period (i.e., from initial detection of the preovulatory follicle to ovulation). Nine of 18 ovulations that occurred during the period of spontaneous recovery in the self-recovery group were double ovulations as compared to 3 double ovulations of 20 ovulations in the controls. In addition, in 3 of 8 cows that self-recovered, cysts remained present during the estrous cycles following self-recovery. Cysts in these cows ranged from 0.8 cm to 2.0 cm in diameter at the end of the experimental period.

**Endocrine Profiles**

**Initiation of follicle/cyst waves.** Detection of new waves of follicle and/or cyst growth was preceded by a transient increase \( (p = 0.08) \) in concentration of FSH from 0.26 ± 0.01 ng/ml (beginning 2.5 days prior to detection of a follicle/cyst wave) to a maximum concentration of 0.41 ± 0.01 ng/ml at 0.5 days before detection of the wave (examples shown in Figs. 2 and 3). FSH then returned to a basal concentration of 0.28 ± 0.01 ng/ml 1 day after detection of the follicle/cyst wave. It is noteworthy that there were no transient increases in FSH for a period of 24 days in cow 800, which had the large cyst that persisted for an extended period of time (Fig. 3).

**Preceding (-4 to 0 days) and subsequent (0 to +4 days) to time of follicular/cyst wave detection.** No difference in concentrations of LH, FSH, and estradiol-17β was detected within cows with nonovulatory (luteal phase) or ovulatory follicles around the time of wave detection (from Day -4 to Day +4) within the control and self-recovery groups. Therefore, data from nonovulatory and ovulatory waves within each group were combined.

Mean concentration of LH was greater \( (p < 0.01) \) in the cyst group (1.07 ± 0.01 ng/ml) than in the self-recovery (0.68 ± 0.10 ng/ml) group during the period around (-4 to +4 days) follicular/cyst wave detection (Fig. 4). Concentration of LH in the self-recovery group was greater \( (p < 0.01) \) than in the control group (0.47 ± 0.01 ng/ml). No difference in the profile of LH over time was detected among groups. Mean concentration of FSH and the profile over time were not different among groups (Fig. 4). Mean concentration of estradiol-17β around the time of follicle/cyst wave detection in the cyst group (7.5 ± 0.4 pg/ml) tended \( (p = 0.13) \) to be greater than in the self-recovery group (5.3 ± 0.3 pg/ml). Control (3.69 ± 0.41 pg/ml) and self-recovery groups did not differ in mean concentration of estradiol-17β. No difference was detected in the estradiol-17β profile over time among groups around the time of wave detection (Fig. 4).

**Preceding (-4 to 0 days) and subsequent (0 to +4 days) to ovulation or at time of equivalent follicle/cyst size (1.60 cm).** Mean concentration of LH was greater \( (p < 0.01) \) preceding and subsequent to the time cyst(s) reached 1.60 cm (cyst group; 1.13 ± 0.08 ng/ml) as compared to the similar time interval around ovulation in the self-recovery (0.74 ± 0.09 ng/ml) group or the time at which dominant nonovulatory follicles (luteal phase) of the self-recovery (0.58 ± 0.10 ng/ml) group reached 1.60 cm (Figs. 5 and 6, respectively). Concentrations of LH tended \( (p = 0.06) \) to be greater in the self-recovery group around the time of ovulation than in the control group (0.48 ± 0.10 ng/ml).

A difference \( (p < 0.01) \) in the profile of LH over time
FIG. 3. **Top**) Example of persistence and turnover of follicles/cysts during the period of observation from cow 800. Each unique symbol connected by a contiguous line represents the dynamics of a single follicle. **Middle**) Concentrations of FSH (line) and LH (bars) relative to the day of observation. **Bottom**) Concentrations of estradiol-17β (stars) and progesterone (ovals) relative to the day of observation. First follicle/cyst wave (Day 0) was detected 10 days after the end of steroid injections used to induce cysts.
was detected around the time that cysts reached the same diameter as the average maximum diameter recorded for ovulatory follicles (1.60 cm). Concentrations of LH increased beginning at Days -2 to -1.5, peaked at Day 0, and returned to basal concentrations at Day 0.5 in cows around the time of ovulation in the self-recovery and control groups, respectively. All cows in both self-recovery and control groups had concentrations of LH around the time of estrus (n = 38) ranging from either 3.5 to 5.0 ng/ml (n = 10) or 5.1 to 10.0 ng/ml (n = 17) or greater than 10 ng/ml (n = 11) followed by ovulation within 36 h. This increase is representative of the increase in LH that occurs prior to ovulation. In contrast, an increase in LH was not detected around the time of equivalent ovulatory follicle size in cows with cysts.

A transient increase in FSH (p < 0.05) was detected after ovulation in the self-recovery and control groups but not in the cyst group after cysts reached an equivalent ovulatory size (Fig. 5). The increase in concentration of FSH would be representative of the increase associated with initiation of the first wave of follicular development in the subsequent estrous cycle of the self-recovery and control groups. Mean concentrations of FSH were similar in cows with cysts, control cows, and self-recovery cows during nonovulatory waves (luteal phase; Fig. 6).

Prior to and subsequent to the time of ovulation, concentrations of estradiol-17B were similar in control (3.36 ± 0.23 pg/ml) and self-recovery (5.08 ± 0.23 pg/ml) groups (Fig. 5). Concentration of estradiol-17B tended (p = 0.13)
was detected in the profile of estradiol-17β over time. Concentrations of estradiol-17β in cows with cysts (7.92 ± 0.95 pg/ml) were greater (p < 0.05) than in control cows (3.86 ± 0.34 pg/ml) and self-recovery groups. A difference (p < 0.05) was detected in the profile of estradiol-17β over time. Concentrations of estradiol-17β in control and self-recovery groups tended (p = 0.12) to decrease from Day -4 to Day 3 and remained constant to the end of the analysis period (Day 4). In contrast, concentrations of estradiol-17β in the cyst group increased (p < 0.01) from Day -4 to Day 0 (time of equivalent follicle cyst size; Fig. 6).

FIG. 6. Concentrations of LH (top; SEM = 0.10), FSH (middle; SEM = 0.04), and estradiol-17β (bottom; SEM = 1.32) shown relative to when nonovulatory follicles or cysts reached a diameter equal to the average diameter of follicles that ovulated (1.60 cm). Day 0 denotes the day of equivalent follicle/cyst size. Control = triangles (n = 6); self-recovery = squares (n = 6); cysts = stars (n = 8).

to be greater in cows with cysts (7.92 ± 0.95 pg/ml) than in the self-recovery cows.

When data were aligned relative to the time when follicle/cyst sizes reached 1.60 cm, concentrations of estradiol-17β in cows with cysts (7.92 ± 0.95 pg/ml) were greater (p < 0.05) than in cows when noovulatory follicles were missed (5.47 ± 0.21 pg/ml) group (Fig. 6). Concentrations of estradiol-17β did not differ between the control (3.86 ± 0.34 pg/ml) and self-recovery groups. A difference (p < 0.05) was detected in the profile of estradiol-17β over time. Concentrations of estradiol-17β in control and self-recovery groups tended (p = 0.12) to decrease from Day -4 to Day 0. In contrast, concentrations of estradiol-17β in the cyst group increased (p < 0.01) from Day -4 to Day 0 (time of equivalent follicle cyst size; Fig. 6).

DISCUSSION

A transient increase in FSH prior to the emergence of a follicular wave was detected in cows with normal estrous cycles (control and self-recovery) in the present study. This report is consistent with that of Adams et al. [18], who concluded that a transient increase in FSH preceded the emergence of a follicular wave. In the present study, a transient increase in FSH preceded the emergence of follicle/cyst waves in cows with cysts. Although intervals between follicle/cyst waves were highly variable compared to intervals between detection of follicular waves in control cows, an increase in concentration of FSH was detected only before the detection of a follicle/cyst wave in the self-recovery and cyst groups. Factors synthesized by the dominant follicle may have prevented the secretion of FSH and subsequently the development of new follicle/cyst waves. Proteinaceous fractions of follicular fluid, when infused into cyclic heifers, have been found to prevent the recruitment of follicles that normally occur during the first 5 days of the estrous cycle [19]. Turzillo and Fortune [20] reported that suppression of the secondary FSH surge with bovine follicular fluid delayed initiation of the first follicular wave. Also, injection of follicular fluid delayed estrus after injection of PGF2α in heifers [21]. Growing follicles produce inhibin; this in turn decreases secretion of FSH [22]. It is likely that the dominant follicle produces the majority of inhibin, which then would deter recruitment and selection of an additional cohort of follicles [23]. In cows with cysts, loss of dominance of the cyst and subsequent recruitment of a new wave of follicular growth may be associated with advanced stages of atresia of the granulosa and loss of at least partial function and secretory capacity of cysts. However, abnormal secretion of FSH is not likely to be associated with development of cysts, since no difference in the mean concentration or profile of FSH was found among the groups (cystic, control, self-recovery) in the present study. Similarly, Cook et al. [17] reported that mean concentrations, pulse frequency, and pulse amplitude of FSH did not differ between cows with cysts and control cows.

Earlier work from this laboratory characterized three phenomena that occur in cows with cysts: 1) turnover, 2) persistence, or 3) spontaneous recovery of ovarian cyclicity [7]. Similar observations were made in the present study. In the previous study [7], the interval between detection of two follicle/cyst waves or the endocrine profiles associated with the morphological changes of follicles/cysts were not determined. In the present study, it was determined that the interval from initial follicle/cyst wave detection to subse-
quent detection of a new follicle/cyst wave was longer in cows with cysts as compared to the intervals between follicular waves in cows exhibiting normal estrous cycles. Growth rates of ovulatory follicles and cysts were similar on the basis of number of days from wave detection to ovulation or equivalent ovulatory size (1.60 cm) for cysts. Follicles from cows with normal estrous cycles either ovulated or underwent atresia and another wave of follicles was recruited with a different follicle becoming dominant. However, cysts continued to increase in size for an additional 5.5 days and became significantly larger than the average ovulatory follicle.

In cows with normal estrous cycles, low concentrations of progesterone after natural luteolysis (insertion of a progesterone-releasing device [CIDR] maintained concentrations between 0.9 and 2.1 ng/ml) promoted prolonged development and dominance of follicles, and additional follicular waves were inhibited [24]. Cows that maintained concentrations of progesterone near luteal phase levels had additional waves of follicular growth during the artificially lengthened luteal phase. Although no difference was detected in basal concentrations of LH during the treatment period, increased LH pulse frequency (25; when serum progesterone concentrations are low) could be responsible for supporting continuous follicle growth [24]. Although sampling frequency in the present study was not sufficient to allow study of LH pulsatility, serum progesterone was low, indicative of a high-frequency mode of LH secretion. In addition, basal concentrations of LH were greater in cows with cysts than in self-recovery or control groups during the time around follicle/cyst wave detection or the time of ovulation/equivalent cyst size. Cook et al. [17] determined that mean concentration, pulse frequency, and pulse amplitude of LH were greater in cows with cysts than in controls. Such an increase in mean concentration and pulse frequency of LH could promote follicular steroidogenesis and be responsible for the greater concentration of estradiol-17β in the cyst group.

An increase in the concentration of LH, representing the LH surge, was not detected at any time in cows that continued to have cysts, while an increase in LH preceding ovulation in control and self-recovery cows was observed in all cases. Rozell and Keisler [10] reported that the rate of increase in serum concentrations of estradiol-17β controls the LH surge in ewes. Estradiol-17β in all observed ovulations in control and self-recovery cows increased prior to ovulation. A preovulatory-like surge of LH was detected after concentrations of estradiol-17β increased and before ovulation occurred in control and self-recovery groups.

An elevated circulating concentration of estradiol-17β has been reported to reduce pituitary content of LH via negative feedback effects at the hypothalamus and pituitary [26]. In the present study, as well as that of Cook et al. [17], cows with cysts had greater peripheral concentrations of LH than controls. Also, concentrations of estradiol-17β were greater in the cyst group in the present study. This suggests that the negative feedback effects of estradiol-17β may not be functioning properly in cows with cysts. The increase in secretion of LH in cows with cysts may be stimulating an increase in estradiol-17β, thus creating a cascade effect. Alternatively, it is possible that the cyst is not producing some substance (produced by a normal dominant follicle) and thus lacks the feedback mechanism.

In another study [27], when estradiol benzoate was administered to cows with cysts, some cows released LH in response to the challenge, but the peak LH release was delayed compared to that in controls. Kesler et al. [28] reported that a GnRH-induced LH release in cows with cysts was not positively correlated with circulating levels of estradiol-17β as is normally found in postpartum cows. Although the pituitary of cows with cysts may release LH in response to exogenous GnRH or estradiol-17β, hypothalamic and/or pituitary function appears to be altered. In a previous study [17], GnRH content in the hypothalamus proper was lower, yet tended to be greater in the median eminence in cows with cysts than in control cows. This may partly explain an increase in LH during follicle/cyst development (increased GnRH in the median eminence) and lack of a preovulatory surge of LH (decreased GnRH in the hypothalamus). Perhaps the self-recovery group was reacquiring negative feedback sensitivity to estradiol-17β, since LH and estradiol-17β appeared to be intermediate in concentrations between the cyst and control groups, thus causing the concentration of LH to decline. Also, concentrations of GnRH in the hypothalamus may have increased concomitantly with reacquired sensitivity to estradiol-17β. This would decrease basal concentrations of LH but still allow concentrations of estradiol-17β from a dominant follicle to elicit an LH surge.

In conclusion, cysts are generally dynamic structures, as evidenced by the replacement of follicles/cysts with different follicles/cysts (turnover). However, cysts may also persist and remain the functional and morphologically dominant structure for extended periods of time. The interval between detection of follicle/cyst waves was longer in cows with cysts, but it was also more variable in length than in control cows. Concentrations of FSH were similar among all groups, and a transient increase in FSH was detected prior to detection of follicle/cyst waves in the control, self-recovery, and cyst groups. However, concentrations of estradiol-17β and LH were greater in cows with cysts around the time of follicle/cyst wave detection and the time of ovulatory follicle size/equivalent cyst size as compared to the values in controls. This could indicate that disruption of the hypothalamic-pituitary-ovarian axis involves interrelationships between LH and estradiol-17β but not FSH. An increased sensitivity of the hypothalamus to positive feedback of estradiol-17β may be associated with the elevated concentrations of LH due to increased release of GnRH. Cysts
that underwent spontaneous recovery produced intermediate concentrations of estradiol-17\(\beta\) and LH in most cases.

ACKNOWLEDGMENTS

The authors gratefully acknowledge Dr. J.J. Reeves, Washington State University, for LH antiserum; Dr. L.E. Reichert, Jr., Albany Medical College, for purified LH for iodination; and NIADDK for purified preparations of FSH for iodination and LH and FSH for standards.

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