Serum Hormone Concentrations in Ruminants During Mammary Growth, Lactogenesis, and Lactation: A Review

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Abstract

Apparently the mammary gland grows in response to sustained high serum concentrations of estrogens and progesterone which are incurred during pregnancy in ruminants. Quantities of prolactin, growth hormone, and glucocorticoid in serum are probably sufficient to support mammary growth during pregnancy. In sheep and goats mammary growth may be stimulated by a lactogenic material of placental origin.

Increased mammary enzymatic activity during the weeks preceding parturition in cattle is temporally related to increased serum estrogen concentrations. Perhaps estrogens cause the marked increase in enzymatic activity that occurs at this time. The sharp decrease in progesterone that occurs 48 to 24 h preparrum in ruminants may allow final maturation of the mammary cell to one characteristic of lactation. If a progesterone repression of lactation occurs in ruminants, once the repression is overcome it is not reestablished to a noticeable degree by increased progesterone incurred during reoccurring estrous cycles or during concurrent pregnancy and lactation.

Increased serum corticoid and growth hormone at parturition may be involved in onset of milk secretion, but for now there is little evidence to support or refute this view. In contrast, the increase in prolactin at parturition apparently is involved in onset of milk secretion since inhibition of the preparrum prolactin surge will suppress onset of copious milk flow. In contrast, suppression of serum prolactin concentrations with ergocryptine during lactation does not decrease milk production in cattle. This does not mean that prolactin is not necessary for maintenance of lactation in ruminants but does suggest that serum prolactin levels are not indicative of only lactational events.

Introduction

This review will discuss changes in serum hormones during mammary growth, lactogenesis, and lactation. The topic is timely since hormone radioassays have been available long enough to generate data for discussion of concepts but no answers have been established. Perhaps now is the time to look critically at results which are available to determine the best route in charting future progress.

There are a few things to clear up at the outset.

1) Different investigators have measured hormone concentrations in blood, serum, or plasma, and no distinction is made between them here.
2) I would like to stress what a serum hormone concentration represents. It is necessarily the hormone concentration in a sample at a given time. As such it is a function of the rate hormone is entering the vascular system and the rate at which it is being cleared. As we investigate changing physiological states, changes in either input or output will affect hormone concentration in the serum.
3) When we measure serum concentrations of protein hormones by radioimmunoassay, we are actually measuring the immunoreactive protein. It has not been demonstrated that immunologically active protein hormones in serum are biologically active either in part or in toto. If the percent biological activity of immunoreactive protein hormones in serum varied with changing conditions, important physiological relationships could be missed.
4) Finally, the vascular system is serving the entire body and may serve as a pool from which organs withdraw hormones as needed to maintain cell function. Hormone induced physiological changes in target tissues need not be reflected in serum hormone changes unless the change in serum hormone concentration, per se, served as a specific trigger for
the event.

The reader should be familiar with the time course of mammary gland development to appreciate relationships between serum hormone concentration and mammary development. During pregnancy these changes are covert and will be reviewed briefly before the main discussion. Following parturition, the capacity of the mammary gland to produce milk is assessed conveniently by measuring milk yield.

**Mammary Development During Pregnancy**

**Proliferative changes.** Information is limited concerning proliferative changes in ruminant mammary glands. Development of the udder of cattle (39) and goats (19) up to mid-pregnancy involves deposition of the ductular tree, vascular elements, and connective tissue supports of the gland. Thereafter, true alveoli are deposited on the duct system, the alveoli increase in size and, eventually, acquire a honey-like secretion. Although the gross and histological structure of the mammary gland of the goat and cow appears developed prior to parturition, it is likely that changes within the secretory cell required for normal milk production continue up to parturition and perhaps into early lactation. These covert changes may be evaluated best by investigating changes in the enzymatic competency of the gland.

**Enzymatic changes.** Lactose synthesis occurs exclusively in the mammary gland and the prepartum appearance of lactose in the gland may be considered the onset of milk production or lactogenesis. Lactose synthesis from glucose is catalyzed by five enzymes: hexokinase, phosphoglucomutase, UDP-glucose pyrophosphorylase, UDP-glucose-4-epimerase, and lactose synthetase. Each of these enzymes is in bovine mammary tissue by 30 days before parturition (67), and there is general agreement that activity increases 1.5 to 2.5 times from late gestation to early lactation (Fig. 1) in cows (67, 6). Maturation of the lactose biosynthetic pathway and synthesis of lactose appear to coincide. Lactose in urine of cows was low until days 8 to 6 prepartum and increased until parturition (99) and lactose in milk from cows subjected to prepartum milking increased from 10 days prepartum through parturition (76). Similarly, lactose was in plasma of goats 4 days prepartum and increased eight times by 12 h prepartum (57). In addition, bovine mammary tissue from cows at 7 days prepartum synthesized lactose in vitro (67). These results suggest that the enzymatic capacity to produce lactose appears in the bovine mammary gland between days 30 and 10 prepartum and increases with approaching parturition.

Development of the enzymatic capacity to synthesize fatty acids also should be an indication of lactogenesis since approximately 60 molar % of the fatty acids in ruminant milk are synthesized de novo in the mammary gland (70). Acetyl-CoA carboxylase, which is postulated to be the regulatory enzyme in fatty acid synthesis (36, 62, 85), is demonstrable in bovine mammary tissue homogenates at 7 but not 30 days prepartum, and the rate of conversion of C-acetate to fatty acids increased approximately four times from 30 to 7 days prepartum (67). Significant development of the fatty acid synthetic capacity of the gland occurs between 30 and 7 days prepartum. Further increases in the capacity to synthesize fatty acids through parturition and early lactation (67) indicate that bovine mammary tissue reaches full potential for fatty acid synthesis after parturition.

Acetyl-CoA used in synthesis of fatty acids in mammary tissue is synthesized by the cytoplasmic enzyme acetyl-CoA synthetase. Acetyl-CoA synthetase activity increases markedly during the final week of pregnancy and continues to increase during early lactation (Fig. 2).

Mammary uptake of blood lipid requires the initial hydrolysis of blood triglycerides by mammary gland lipoprotein lipase. This enzyme is prolactin inducible in rabbits (31) and appears to be extremely sensitive to changes in lactational status of laboratory animals (40, 62, 63, 72). Lipoprotein lipase
activity increases markedly in bovine mammary tissue across parturition (Fig. 2) with first evidence for these changes beginning 1 wk prepartum (83).

Therefore, investigations of changes in mammary enzymes during the periparturient period in cattle have shown that mammary preparation for the postpartum production of milk begins in advance of parturition. Exact timing of these events is not possible from published data, but these changes take place within the 3-wk preceding parturition and continue postpartum.

Serum Hormones: Pregnancy and Parturition

Considering the proliferative and enzymatic changes in the mammary gland during growth, lactogenesis, and lactation, we can now turn to a discussion of serum hormone concentrations during these same periods to evaluate possible endocrine control of these events.

Estrogens. Recent reports from several laboratories have described serum changes in total estrogens (42, 91), estrone (26, 33, 73, 88), and estradiol (26, 46, 88, 98) during pregnancy and parturition in cattle. Changes in serum estrone and estradiol during pregnancy are in Fig. 3. Most investigators agree that estrogens gradually increase in serum beginning at 3 to 5 wk prepartum followed by a precipitous increase during the last 2 wk of pregnancy and a rapid decrease to low concentrations during early lactation.

Average serum estradiol concentration in heifers when measured at 26 days prepartum (88) was approximately four times greater than the comparable average during days 18 to 75 days of pregnancy (98). Similarly, total estrogens averaged less than 5.0 pg/ml from days 12 through 39 of pregnancy in beef heifers and was greater than 500 pg/ml at 14 days prepartum in dairy cows (42). The difference in estrogen concentration is not likely due to breed but probably reflects a marked increase in serum estrogens from early to late pregnancy. In addition, estrone and estradiol concentrations in serum collected from cows, in the first, second, and third trimesters of pregnancy, reveal a gradual increase in these estrogens during gestation (58). Serum estrogen analysis confirms earlier investigations that demonstrated an increase in estrogenic activity in bovine urine and feces during pregnancy (review, 68). During pregnancy in cattle, serum estrogen concentration apparently stabilizes in the first trimester of pregnancy at somewhat less than the level of estrus, increases gradually through pregnancy until approximately 3 wk prepartum when the rate of increase is markedly accelerated.

During the immediate prepartum period serum estrogens increase markedly. Smith et al. (88) observed a linear increase in estradiol from 5 to 26 days before parturition. Estrone concentrations paralleled those of estradiol but were approximately eight times higher. Both steroids decreased rapidly to low levels immediately postpartum. Others have demonstrated marked increase in estrone (29, 46, 73), estradiol (29, 46), and total estrogens (42, 91) during the month preceding parturition, but estrogens do not always decline prepartum (42, 46). Estrone values for individual cows bled daily showed that time of the serum es-

Fig. 2. Changes in some bovine mammary enzymes involved in fatty acid metabolism during the periparturient period. Enzymatic activity at day 32 or 40 of lactation was arbitrarily set at 10 units. Data points marked by a: 0 are from Mellenberger (70); △ are from R. M. Cook, Michigan State University, personal communication; and □ are from Shirley, Morrow and Emery (83).

Fig. 3. Schematic representation of serum estrone and estradiol during gestation and parturition in cattle. Estradiol ———; Estrone ———.
trone decrease ranged from 1 to 5 days prepartum (73).

Information on serum estrogen changes during pregnancy and parturition in sheep and goats is preliminary. In goats, concentrations of serum estradiol 17α and estrone, as measured by radioimmunoassay, were high relative to cows at 25 days prepartum and remained unchanged until 24 h prepartum when they increased 2 to 3 times (92). Whether estrogen at day 25 prepartum represents an increase from earlier in pregnancy could not be determined. Estrogen fell precipitously with discharge of the fetal membranes. Quantitatively, changes in serum estrogens in pregnant ewes are markedly different from those of the goat and cow. Total estrogens were undetectable during the estrous cycle, less than 5 pg/ml until 31 days before parturition, and stabilized at 20 to 40 pg/ml until day 5 prepartum (13). Total serum estrogens (13, 14), estrone, and estradiol 17β averaged 20 pg/ml in serum at 48 h prepartum and rose to 880 pg/ml within a few hours of birth; the major increase occurred almost entirely within the last 12 h (92). Whether there are gradual shifts in serum estrogen earlier in pregnancy that might be related to changes in rate of mammary growth cannot be discerned from the available literature and will require investigations with frequent sampling and more animals.

Prepartum increase in serum estrogens in cows is temporally related to increased enzymatic activity in the mammary gland. Increased estrogens might induce enzyme activity in preparation for onset of milk secretion at parturition. However, specific experiments will have to be designed to test this hypothesis. It would be of interest to investigate changes in enzyme activities during the periparturient period in sheep and goats. If enzyme activity is induced by the prepartum increase in serum estrogens, then one might expect this increase to occur nearer parturition in these species since the major rise in estrogen occurs immediately prepartum.

Progestosterone. During the early stages of gestation, plasma progesterone percent were similar to those which characterize the luteal phase of the cow's estrous cycle (43, 75, 98). Serum progesterone measured during the first 75 days of pregnancy was maximum on days 18 through 20 in heifers and cows, respectively, declined about 20% during the next 4 to 5 days, then increased (98). This decrease at the time of estrus would have occurred if conception had not taken place (43). Whether the subsequent increase in progesterone comes from accessory luteal tissue resulting from ovulation, luteinization of follicles, or from reactivation of the original corpus luteum by a luteotropin is not known.

Between 90 and 150 days of gestation there is a decline in serum progesterone to relatively low concentrations followed by a secondary rise (74). This mid-gestation increase coincides with beginning lobulo-alveolar development (19), but establishing a relationship between the two will require additional studies.

A gradual decrease in serum progestins over the final weeks of gestation has been reported (84, 90), but statistical verification was not provided. Others (30, 87) have failed to observe a significant decrease at this time. In any event, the changes in serum progesterone that occur over the last 2 wk of gestation are small relative to the precipitous decrease (Fig. 4) that occurs 2 to 3 days prepartum (42, 74, 79, 88, 90).

The increase in plasma progesterone in goats following conception is comparable to the increase during the first part of a normal estrous cycle (93). Progesterone concentrations in plasma are maintained at approximately 3 ng/ml from day 8 to about day 60 of gestation (93) when a secondary increase to 5 ng/ml occurs. This is maintained until 2 days prepartum when progesterone declines precipitously (27, 92, 93).

Progesterone concentration during pregnancy in sheep differs from cow and goat because of substantial contribution of progesterone from the placenta-fetal unit (59). Progesterone concentrations in sheep plasma during the first trimester of pregnancy (to day 50) are

![Fig. 4. Serum progestin (expressed as progesterone) in 10 cows from 26 days before to 9 days after parturition (from Smith, Edgerton, Hafs and Convey, 88).](image-url)
comparable to those during the luteal phase of the estrous cycle (7, 89) which suggests that the corpus luteum is the major source of progesterone. Serum progesterone concentrations increase after day 50 (7, 89), and the increase is greater in ewes carrying twin lambs (89). Plasma progesterone concentration in sheep is greatest at 125 to 130 days of gestation (7, 92) and decreases gradually until parturition (7, 34, 92), indicating that endocrine changes heralding parturition occur well in advance of the event.

The precipitous decrease in serum progestins prior to parturition in ruminants is consistent with the popular view that progesterone represses lactation during pregnancy. Decrease in serum progesterone at parturition may allow epithelia of the mammary gland to respond to the lactogenic complex. In laboratory animals, removal of the ovaries on day 18 or 19 of pregnancy increased lactose synthesis in the mammary gland which was prevented by injecting 5 mg progesterone daily (102). Similar studies in ruminants are needed to investigate this possibility.

Corticoids. To my knowledge estimates of corticoid concentrations in serum samples collected throughout pregnancy in nonlactating ruminants are not yet available. However, the effects of pregnancy on serum corticoid concentration of cows which are simultaneously lactating and pregnant revealed no significant effect of gestation on serum corticoid concentration (55), but serum corticoid concentrations change substantially near parturition in cattle (88). Total corticoids (Fig. 5) in serum from heifers increased from an average of 5 ng/ml (day 26 to 1 prepartum) to 16.7 ng/ml at parturition, then decreased to 5 ng/ml by 12 h postpartum. Although the data in Fig. 5 indicate greater variation in serum glucocorticoids during the first 9 days postpartum than during the comparable prepartum period, average of serum glucocorticoids after parturition (5.6 ng/ml) was not different from that before parturition (5.0 ng/ml). Serum corticoids also increased at parturition in multiparous cows (3, 28) but not in ewes (8).

Although the physiological role of the surge of glucocorticoids at parturition is not known, several possible functions may be considered. Of primary interest here is a possible role in initiation of lactation. Exogenous glucocorticoids administered during early, mid-, or late pregnancy initiated milk secretion in heifers without immediately disturbing pregnancy (96). Similarly, synthetic adrenocorticotropic hormone (ACTH) infused into goats on day 125 of gestation caused a marked increase in udder size and had induced lactation within 6 days which coincided with parturition (92). These reports are of particular importance since they demonstrate that lactation can be induced with exogenous glucocorticoid or ACTH in the presence of high progesterone concentrations. Thus, if there is a progesterone block to lactogenesis in ruminants, apparently it can be overcome with a surge of corticoids. It also seems that once the progesterone block is removed, it is not reestablished since lactation continues in the absence of continued corticoid treatment. Corticoids released at milking may prevent progesterone repression of lactation (see below) or steroid receptors may differentiate from those characteristic of resting cells to ones characteristic of lactating mammary cells. Additional research will be required to investigate these suppositions.

Alternatively, changes in maternal serum glucocorticoids may be involved in initiation of parturition since exogenous glucocorticoids will cause parturition in sheep and cattle when administered during late pregnancy (1, 2). Another possibility is that the precipitous increase in serum corticoids near parturition results from neural stimuli associated with labor. Uterine and abdominal contractions are increased at 2 days prior to delivery in cattle (37).

Prolactin. The concentration of prolactin in serum or plasma of cattle is influenced by season (56, 78); serum prolactin is highest during the warm months. Accordingly, seasonal effects must be considered when interpreting prolactin changes during prolonged physiological events such as pregnancy or lactation, especially when animals conceive during the same period. This was demonstrated by Koprowski and Tucker (56), who reported that serum prolac-
tin of nonlactating heifers which conceived during 2 mo, appeared to increase during gestation (Fig. 6). Serum prolactin during the first 90 days of gestation averaged 9 ng/ml but increased to 76 ng/ml by 270 days of pregnancy. To determine whether these changes were seasonal, each of six heifers in the first or third trimester of pregnancy was bled on the same day. Serum prolactin was quantitated and averaged 10 ng/ml for each group. From these data serum prolactin appears to be unchanged in heifers during the major part of gestation (Fig. 6). Similarly, serum prolactin did not change significantly during pregnancy in lactating cows when means of prolactin were adjusted for stage of lactation and season (Fig. 7; 59). Baseline prolactin of pregnant-lactating cows were significantly lower than comparable values for open-lactating cows. Perhaps lactogenic material of placental origin distinct from radioimmunoassayable prolactin may inhibit release of pituitary prolactin during pregnancy. This hypothesis seems tenable in view of recent evidence for a placental lactogen in goats (12; see below).

Serum prolactin changes in cows during the periparturient period are consistent (48, 51, 78; Fig. 7). Approximately 5 days before parturition, serum or plasma prolactin concentration increased to a peak at 24 h prepartum, then decreased linearly to approximate values characteristic of pregnancy by 48 h postpartum (Fig. 7).

In view of reports describing increased serum prolactin following acute stress in cows (71, 95) the possibility that the partum increase in serum prolactin results from pain associated with delivery must be considered. However, the rise in serum prolactin precedes by several days onset of propageative and coordinated uterine contractions (37, 45). Evidence that the prolactin surge at parturition importantly influences onset of lactation in cattle has been demonstrated by virtue of its elimination. Thus, the ergot alkaloid 2-Br-a-ergocryptine-methane-sulfonate which effectively reduces serum prolactin concentrations in cattle (53, 86) when administered during the periparturient period in cows, depressed onset of lactation (80).

Changes in serum prolactin during pregnancy in sheep (Fig. 8) are difficult to assess from the literature. Arai and Lee (4) measured prolactin concentration in sera of sheep which were collected at infrequent intervals during pregnancy and observed relatively low prolactin concentrations during mid-pregnancy. Sampling more frequently, Davis et al. (24) observed a stabilization of serum prolactin in sheep at a low level (2.4 to 6.0 ng/ml) from wk 7 through wk 16 of gestation; and a grad-

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**Fig. 6.** Serum prolactin in heifers during pregnancy. On December 8, 1970 each of six cows in the first and third trimester of pregnancy was bled and serum prolactin quantitated. The means of this analysis are indicated. From Koprowski and Tucker (56).

**Fig. 7.** Schematic representation of serum prolactin during gestation and parturition in cows. The arrows indicate that values are not absolute but rather vary with season.

**Fig. 8.** Schematic representation of serum prolactin during pregnancy and lactation in sheep and goats. Fractions indicate the number of pregnant goats having lactogenic activity greater than 100 ng/ml ovine prolactin equivalents as determined by an in vitro bioassay (from Buttle, Forsyth and Knaggs, 12).
ual rise so that prolactin averaged 25 ng/ml at 1 wk prepartum.

A rapid increase in serum prolactin in ewes 3 days preceding parturition has been reported (24). Highest serum prolactin was on the day of parturition. Prolactin then decreased during early lactation to values significantly greater than those characteristic of late pregnancy. These authors did not indicate whether the lambs were kept with the ewes. Others (32, 65) have reported increased serum prolactin at parturition in ewes, but limited sampling schedules and animals precluded drawing conclusions as to magnitude of the increase relative to comparable pregnancy values or time course of response.

Radioimmunoassayable serum prolactin of goats was uniformly low (≈ 2.5 ng/ml) from conception through wk 12 of pregnancy and then increased to an average exceeding 200 ng/ml by wk 21 of pregnancy (12). Although radioimmunoassayable prolactin activity was low during mid-pregnancy, serum prolactin lactogenic activity as determined by bioassay (33) exceeded 100 ng/ml ovine (NIH-P-S-6) equivalents (Fig. 8). Mammogenic activity was in all samples tested from wk 16 through parturition and was also demonstrable in cotyledon tissue. These results raise the interesting possibility that placental lactogen is in the goat and plays an important role in mammary growth. The fact that radioimmunoassay and bioassay results agree for serum samples from goats at times other than pregnancy suggests that this lactogenic activity is unique to the pregnant animal. Appearance of lactogenic activity at mid-pregnancy coincides with the time when there is increased mammary proliferation in this species (19). In view of these discoveries in goats, it will be important to determine whether a placental lactogen exists in cows and sheep.

Prolactin changes at parturition in goats parallel those reported for cows (51). In contrast to the bovine (48, 51, 78) serum prolactin of sheep (24) and goats (41, 51) does not decrease following parturition (Fig. 8). The difference may be due to varied nursing intensity and frequency since restricting suckling caused a rapid decrease in serum prolactin in goats (51).

Growth hormone. There is little information concerning serum and pituitary growth hormone concentrations during pregnancy and lactation in ruminants. Ingalls et al. (48) measured growth hormone in the serum of heifers from 26 days before to 26 days after parturition (Fig. 9). Serum growth hormone concentration increased gradually from 9 to .5 days prepartum, was markedly increased at parturition and .5 days postpartum, then decreased linearly to values characteristic of the immediate prepartum period by day 4 postpartum. Similarly, prepartum serum growth hormone increased in three ewes, but the onset of this increase ranged from 1 to 10 days before parturition (9).

What stimulates increased pituitary growth hormone release at parturition in ruminants? Release may have resulted from stimuli associated with birth of the young. Although serum growth hormone concentration is less influenced by physical stimuli than is serum prolactin concentration (95), growth hormone is increased by severe stress associated with surgery (82). Alternatively, changing serum estrogens may cause increased serum growth hormone since Trenkle (94) reported a significant increase in serum growth hormone concentration in animals fed diethylstilbestrol. But correlations within day between estradiol or estrone and serum growth hormone were not significant (48), and the increase in growth hormone in ewes precedes the prepartum increase of serum estrogens (92).

Whatever the stimulus to enhance release of growth hormone at parturition, growth hormone is increased in serum at this time and its physiological role should be considered. Growth hormone is required for normal lactation in ruminants (66) and is galactopoietic in cows (47, 60, 101), sheep (52), and goats (20). Exogenous growth hormone when administered to heifers from 9 days prepartum to 16 days postpartum increased milk produc-
tion, and the effect lasted throughout lactation (81). Brumby (10) observed enhanced milk production in identical twins treated with 50 mg growth hormone from 14 days before to 14 days after calving, but the response was transient and undetectable by 7 days after withdrawal of growth hormone treatment. Therefore, a surge of growth hormone at parturition would, at the very least, benefit onset of milk production.

To limit our discussion to mammary physiology at a time when the animal’s body metabolism shifts from pregnancy to lactation is risky indeed. At this time there is imposed on the animal increased metabolic demands for energy and protein. Accordingly, it is possible that the increase in serum growth hormone concentration at parturition is associated with this change in body metabolism to meet the demand for these nutrients.

**Serum Hormones and Milk Secretion**

**Ovarian hormones.** The placental and ovarian hormones, in the presence of pituitary hormones, provide for growth of the mammary gland during pregnancy. However, these hormones are decreased in serum to very low concentrations either just before or following parturition. Withdrawal of progesterone and probably estrogens also removes the block to lactation, and copious milk secretion results. Milk secretion is probably maintained via hormones released in response to milking or by continued removal of milk from the mammary gland. It is unlikely that changes in serum estrogen and progesterone concentrations, which occur with recurrent estrous cycles, significantly influence lactation in cattle. Thus, ovariec-tomy during lactation does not alter milk yield (17), and notwithstanding the popular view of dairymen, there is no experimental evidence for decreased milk production at estrus (16). However, in the pregnant, lactating cow, the major increase in serum estrogens that occurs in late pregnancy (see above) may be inhibitory to milk synthesis. During a normal lactation involving a concurrent pregnancy, milk yields decline progressively and solids-not-fat increases from the second trimester of pregnancy at a rate exceeding that of the nonpregnant cow (5). Estradiol benzoate administered in quantities greater than 25 mg can mimic effects of pregnancy on milk yield and composition.

Results of many investigations of the effects of estrogens on established lactation in cows and goats may be found in the literature (17, 66: review). In general, exogenous estrogens administered in low doses improve or enrich the composition of milk and increase fat and solids-not-fat, while higher doses of estrogens suppress milk production.

Although resumption of estrous activity does not influence lactation per se, there is evidence that the converse is true, i.e. suckling or milking intensity will influence onset of estrous cycles after parturition in ewes (61) and cows (100).

**Corticoids.** During early lactation, serum corticoid concentrations in heifers (88) and cows (28) are comparable to prepartum concentrations. Total corticoid concentration, measured in serum from cows during lactation, was variable but changed little as lactation advanced (55). The shift from pregnancy to lactation is not accompanied by noticeable changes in serum corticoids at rest. It is conceivable that lactational requirements for corticoids are satisfied by corticoids released at milking (25, 69, 87, 97). The coordinated release of corticoids and prolactin, and perhaps thyroid stimulating hormone, that follows milking, may be required for maintenance of lactation. This view is supported by evidence that correlations between serum prolactin or corticoids and milk yield in lactating cows were higher for hormones measured immediately post-milking than for those measured 1 to 2 h before or 1 h after milking (55).

**Prolactin.** Prolactin plays an important role in mammary physiology of laboratory animals (17, 66). It is likely that it influences lactation in ruminants. Prolactin is apparently important, among other hormones, in reestablishing lactation in goats hypophysectomized by surgery (20, 21) or proton beam irradiation (35), and a significant relationship has been established for cows between serum prolactin and milk yield (56). Serum was collected at 2 to 4 h before, immediately after, and 1 h after milking. Milk production paralleled serum prolactin measured immediately after milking (Fig. 10). Correlations between serum prolactin measured in serum collected immediately after and 1 h after milking were .36 and .19. Whether this relationship is cause and effect has not been established. Preliminary results (64) demonstrate a significant negative relationship between the concentration of immunoreactive prolactin in milk and milk yield.

An increase in serum prolactin following suckling or milking could be predicted from measurement of pituitary prolactin in laboratory animals. Prolactin is released in response to the milking stimulus in bovine (49, 50, 77, 95), ovine (32) females, and does (11, 49,
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50). Because prolactin released at milking in rats directly influences rate of mammary refill-
ing (38), it is logical that prolactin released at milking or nursing in ruminants is responsible, at least in part, for maintenance of established lactation in ruminants. However, suppression of prolactin with ergocryptine does not inhibit milk production (53, 86) although resting prolactin is reduced to approximately 1 ng/ml and prolactin release at milking is inhibited (Fig. 11). Reconciling these results with the view that prolactin released at milking is necessary for continued milk production is difficult. However, results should not be inter- preted as evidence that prolactin is not re-
quired for maintenance of lactation in cows. Restoration of milk yield to prehypophysec-
tomy amounts in goats was attained with daily injections of approximately 14 mg sheep pro-
lactin daily (20, 21). For a blood to milk ratio of 400:1, the mammary gland of a cow pro-
ducing 25 kg milk and having serum prolactin of 1 ng/ml would be exposed to approximately 20 mg prolactin daily which may be adequate to sustain milk secretion. Alternatively, it is conceivable that prolactin released at a given milking may not be expressed in terms of milk production until sometime later. This would be conceivable if prolactin maintained mam-
mary enzymes having a slow turnover rate or the mammary cells themselves. That milk pro-
duction can continue in ruminants for variable periods without prolactin is indicated by studies in goats wherein milk production did not decrease following prolactin withdrawal (18).

There is far more prolactin in serum than is re-
quired to maintain milk secretion, and serum prolactin levels probably are not reflective of mammary gland function only. Other physio-
logical roles for this hormone should be consid-
ered.

It is also difficult to rationalize the wide va-
riety of stimuli which will increase serum pro-
lactin and yet hypothesize that prolactin is only involved in mammary physiology. Often the magnitude of prolactin release which follows exterceptive stimuli is more marked than comparable changes during lactation or milking. I think it appropriate to discuss these stim-
uli because many of us tend to see prolactin with tunnel vision with a mammary gland at the end of the tunnel.

Stressful stimuli such as pain, restraint, and emotional disturbances cause prolactin release in ruminant animals (11, 50, 71, 77, 95). So does sexual excitement in bulls (16), rams, and ewes (11). Serum prolactin concentration in cows fluctuates with a circadian periodicity with a doubling from nadir to peak (54). In addition, serum prolactin is influenced by sea-
son (56, 77).

Changes in metabolic events also alter serum prolactin concentrations. Thus, resting prolac-
tin is decreased during fasting, and prolactin release at milking is inhibited (11). Prolactin increases in blood after feeding (11) and infusion of large quantities of amino acids cause an increase in serum prolactin concentration which is independent of glucose changes in sheep (22) and cattle (44). The possibility that changes in serum hormone under different physiological conditions may be due, at least in part, to changes in metabolic clearance rate has also been advanced (23).

These changes in plasma prolactin that fol-
low the variety of stimuli listed above indicate a more fundamental metabolic role for pro-
lactin than involvement in mammary physiology.

Fig. 10. Average prolactin concentrations during lactation in serum collected 2 to 4 h before (△△), 5 min after (●●●●), and 1 h after (○○○○) milking. Average daily milk yields are plotted as [] from Koprowski and Tucker (56).

Fig. 11. Serum prolactin and growth hormone (GH) in control (○) or ergocryp-
tine (▲) treated cows, from Smith, Beck, Convey, and Tucker (86).
and lactation. Investigations designed to evaluate alternative physiological roles for prolactin in both male and female are needed.

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References

(22) Davis, S. L. 1972. Plasma levels of prolactin, growth hormone and insulin in sheep following the infusion of arginine, leucine and phenylalanine. Endocrinology 91:549.


(88) Smith, V. G., L. A. Edgerton, H. D. Hafs,


**Hormonal Effects on Mammary Cytology**

The mammary epithelial cycle of an adult can be divided into four morphological stages of development and regression. Adult development of the epithelium begins with cellular proliferation producing the major outline of the mammary gland microanatomy in the form of small ducts, terminal end buds, and finally alveoli. Proliferation begins during puberty and is completed during pregnancy. The second stage of adult development is the morphological differentiation of the epithelia of terminal ducts and alveoli into secretory cells. This change occurs during late pregnancy and extends to immediately after parturition. The third stage of the adult mammary epithelial cycle begins when cellular activity is directed toward milk synthesis and cell growth diminishes. It starts shortly after parturition and predominates through lactation. The fourth stage, involution, is the regression of secretory cells which results in atrophy of the gland. Involution starts early in lactation and becomes most pronounced after extended lactation or cessation of suckling or milking. After involution the cycle is reset by the subsequent pregnancy. Control of cell proliferation and differentiation will have the greatest ramifications in applied milk production, control of mastitis and breast cancer, and artificial control of lactation.

Because many hormones alter mammary function, initial studies seeking the single hormone or several hormones responsible for mammosogenesis led to confusion. The work of Lyons and colleagues (30) is a major demonstration of the interacting roles of ovarian and adrenal steroids with pituitary hormones in...