BIOLOGY OF REPRODUCTION 50, 239–247 (1994)

Luteal Function: The Estrous Cycle and Early Pregnancy

G.D. NISWENDER,[1] J.L. JUENGEL, W.J. MCGUIRE, C.J. BELFIORE,[2] and M.C. WILTBANK[3]

Animal Reproduction & Biotechnology Laboratory, Colorado State University, Fort Collins, Colorado 80523

ABSTRACT

A number of morphological and biochemical changes occur as the cells of the recently ovulated follicle luteinize and develop into a functional CL. There are two distinct steroidogenic luteal cell types that appear to differentiate from thecal and granulosal cells in the follicle. The control of progesterone secretion is quite different in the two cell types. Prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) is the primary luteolytic hormone in most mammals. $PGF_{2\alpha}$ appears to exert its antisteroidogenic actions via activation of the protein kinase C system, while its cytotoxic effects appear to be mediated via a dramatic increase in intracellular levels of free calcium. The mechanisms involved in maternal recognition of pregnancy are very diverse between species and may involve direct luteotropic stimulation of the CL, reduced uterine secretion of $PGF_{2\alpha}$, and/or inhibition of actions of $PGF_{2\alpha}$ at the level of the CL.

INTRODUCTION

The corpus luteum (CL) is a transient endocrine organ required for normal pregnancy in mammals. The first report of the biological significance of this gland was published in 1903 by Frankel [1], who demonstrated that pregnancy was terminated in rabbits after removal of the CL. Subsequent purification and crystallization of progesterone were accomplished in 1934 by four groups [2–5]. Perhaps it is of evolutionary significance that the simplest steroidogenic pathway is that for biosynthesis of progesterone, which plays such a key role in successful reproduction.

The biosynthetic pathway for progesterone is depicted in Figure 1 for a generic luteal cell. In most cases, cholesterol utilized as substrate is obtained from high or low density lipoproteins (HDL, LDL) rather than synthesized de novo from acetate [6, 7]. Uptake of LDL occurs through classic receptor-mediated endocytosis [8], whereas uptake from HDL involves binding to specific membrane binding sites and shuttle of cholesterol into the cell by an unknown mechanism [9]. Cholesterol from the various sources can then be utilized for steroid synthesis or can be incorporated into cholesterol esters by acyl CoA cholesterol acyltransferase (ACAT) and stored as lipid droplets (reviewed in [10]). As luteal progesterone secretion increases during the luteal phase of the reproductive cycle, lipoprotein binding sites increase on luteal cells [11, 12].

Release of cholesterol from cholesterol esters is dependent on a neutral cholesterol esterase (also known as hormone-sensitive lipase). Activity of this enzyme is regulated by phosphorylation of two serine residues. Cyclic AMP-dependent protein kinase A (PKA) causes phosphorylation of one serine residue and activation of the enzyme, whereas Ca^{2+}/calmodulin-dependent protein kinase phosphorylates the other serine residue and prevents activation of the enzyme (reviewed in [10]).

The rate-limiting step in progesterone biosynthesis is cleavage of the side chain of cholesterol. This process involves transport of cholesterol from cytoplasm to the mitochondria and from the outer to the inner mitochondrial membrane, the site of side-chain cleavage. Transfer of cholesterol to the mitochondria appears to involve the cytoskeleton (reviewed in [13]). Cholesterol must also be transported from the outer mitochondrial to the inner mitochondrial membrane, where cytochrome P450 side-chain cleavage enzyme ($P450_{scc}$) is localized. This transport may be mediated by many factors, including steroidogenesis activator peptide, sterol carrier protein 2, endozepines/benzodiazepines, and lipoxygenase metabolites (reviewed in [10]).

Three proteins are involved in conversion of cholesterol to pregnenolone: adrenodoxin, adrenodoxin reductase, and cytochrome $P450_{scc}$. Messenger mRNAs for these proteins are regulated similarly; thus mRNA for $P450_{scc}$ is often used to monitor transcription of genes encoding the enzymes in this complex [14, 15]. Conversion of pregnenolone to progesterone is catalyzed by 3β-hydroxysteroid dehydrogenase, Δ^5,Δ^4 isomerase (3-βHSD) (reviewed in [16]).

LUTEINIZATION AND FORMATION OF THE CL

The preovulatory surge of LH sets in motion a series of morphological and biochemical changes resulting in reorganization of follicular cells into the CL. The basement membrane between the theca interna and membrana granulosa begins to break down, blood vessels invade the follicular antral space, and an extensive vascular network develops. During luteinization, there is significant hypertrophy and hyperplasia of thecal cells [17], which migrate into the previous follicular cavity and become dispersed among luteinizing granulosal cells. Granulosal cells accumulate smooth endoplasmic reticulum, mitochondria become rounded with tubulovesicular cristae, and glycogen-containing granules accumulate [18]. Mitotic activity occurs in thecal cells after

[1]Correspondence.

[2]Current address: Department of Molecular Biology, University of Wyoming, Laramie, WY 82071.

[3]Current address: Department of Dairy Science, University of Wisconsin, Madison, WI 53706.

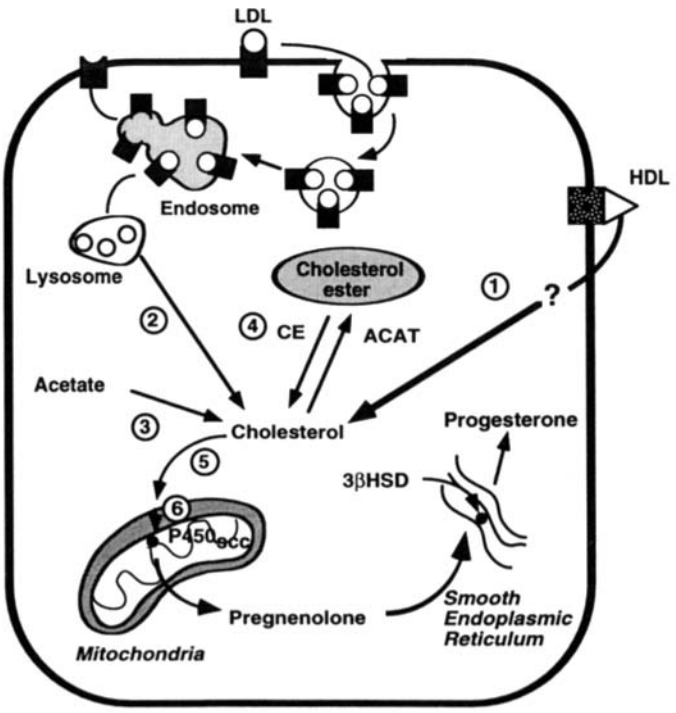

FIG. 1. Pathway for progesterone biosynthesis in a generic luteal cell. Four sources of cholesterol can be utilized for substrate and include cholesterol derived from HDL (1) or LDL (2) from the blood, synthesis of cholesterol from acetate (3) and hydrolysis of cholesterol esters (4). The free cholesterol is transported to the mitochondria (5) apparently with cytoskeletal involvement. The transport of cholesterol from the outer to the inner mitochondria membrane (6) appears to be a key mechanism increased by trophic hormone stimulation or decreased by protein kinase C activation.

ovulation, but there does not appear to be significant cell division in luteinizing granulosal cells [19].

There are also numerous biochemical changes associated with the process of luteinization. After the LH surge, but prior to ovulation, there is a temporary decrease in mRNA for $P450_{scc}$ and 3β-HSD [20]. This is followed by increases in mRNA and enzyme activity for $P450_{scc}$ and 3β-HSD after ovulation and during luteal formation. The activity of cholesterol esterase [21], cytochrome $P450_{scc}$ [22–24], and 3β-HSD [23–26] increases as the CL becomes fully functional. In most species there are decreases in androgen and estrogen production as follicular cells luteinize. Levels of mRNA and protein for 17α-hydroxylase cytochrome P450, which catalyzes conversion of pregnenolone or progesterone to androgen, are abundant in preovulatory follicles but are low in CL of cattle [14, 22] and rats [27]. Levels of mRNA and protein for aromatase cytochrome P450 enzyme decrease rapidly after the LH surge in several species [28, 29]; however, in other species such as the human and the rat, aromatase activity is present in the CL.

After ovulation, receptors for FSH and LH on granulosal cells are down-regulated due to internalization of occupied receptors and reduced expression of genes encoding the receptors [30–32]. Receptors for LH increase as the CL forms

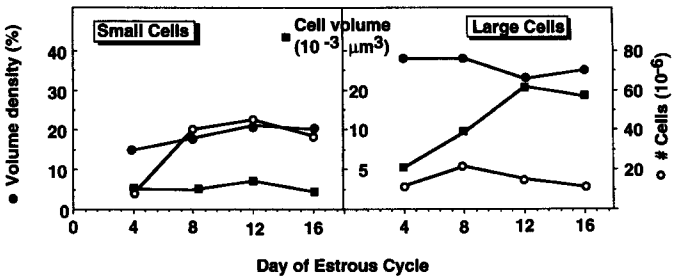

FIG. 2. The volume density (solid circles) of the corpus luteum occupied by small and large luteal cells, changes in cell volume (solid squares) and cell number (open circles) throughout the estrous cycle in ewes. Adapted from Farin et al. [37].

[33], while receptors for FSH are present in CL from only a few species, e.g., hamsters [34] and cows [35]. In a study of rats, there was enhanced expression of the gene encoding the LH receptor as the CL developed, apparently due to the stimulatory effects of prolactin [31].

LUTEAL PHASE OF THE ESTROUS CYCLE

The majority of the parenchyma of the CL consists of steroidogenic cells referred to as luteal cells. Support cells account for approximately 20% of the volume and include vascular elements (endothelial cells, pericytes), macrophages, smooth muscle cells, and fibroblasts [36, 37]. There are at least two morphologically and biochemically distinct steroidogenic luteal cell types in the ewe [38, 39], cow [40, 41], pig [42], rat [43, 44], rabbit [45], monkey [46], and human [47]. The most obvious difference between the two steroidogenic cell types is size, leading to their designation as small and large luteal cells. Ovine small luteal cells are 12–22 μm in diameter, are usually spindle-shaped, and contain an abundance of smooth endoplasmic reticulum, numerous mitochondria, and lipid droplets within the cytoplasm. In contrast, ovine large luteal cells are 22–50 μm in diameter, are spherical in shape, and contain numerous mitochondria, abundant smooth endoplasmic reticulum, stacks of rough endoplasmic reticulum, and electron-dense secretory granules. In some species, large luteal cells also contain lipid droplets. The secretory granules in large cells have been shown to contain oxytocin [48] or relaxin [49] and are released by exocytosis [48]. These granules may also contain growth factors, depending upon the reproductive state and species.

Morphometric analyses have provided reliable data regarding numbers of steroidogenic luteal cells in the ewe [37, 50, 51] and cow [51]. Number, volume, and volume density of ovine small and large luteal cells throughout the estrous cycle are depicted in Figure 2. The number of small cells increases approximately 5-fold with little change in cell volume, while large cells increase in size with little change in number. The net result is that the volume of the CL occupied by each cell type (volume density) remains relatively constant [37, 50].

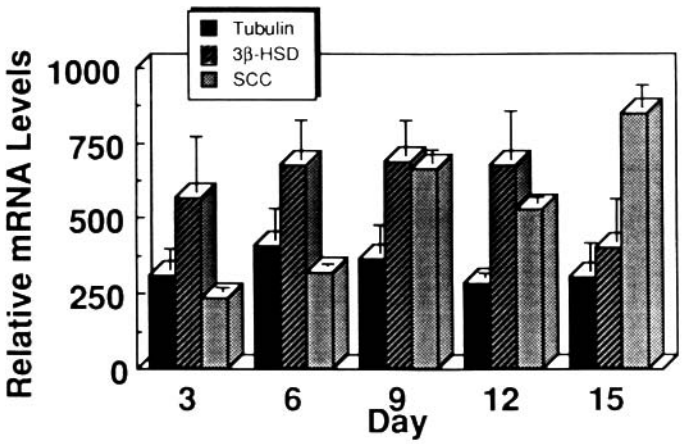

FIG. 3. Relative levels of mRNA encoding tubulin, 3β-HSD and cytochrome $P450_{scc}$ in ovine luteal tissue (densitometric units/20 μg total RNA) during the estrous cycle. Adapted from Hawkins et al. [26] and Belfiore et al. [61].

The principal hormone that stimulates progesterone production by the CL is LH [18]. In a study of ovine CL, the number of LH receptors did not reach maximum until the midluteal phase (Day 10) of the estrous cycle [33]. Similar observations have been made in monkeys [52] and humans [53]. Levels of mRNA encoding LH receptors in monkey CL are also higher during the midluteal than in the early luteal phase of the cycle [54]. In rat CL, mRNA encoding the receptor for LH returns to amounts similar to those seen in the preovulatory follicle by Day 4 of pregnancy [31]. Luteinization of rat granulosal cells is also associated with an increase in prolactin receptors [55], which are essential for normal luteal function (reviewed in [56]).

There is evidence that unstimulated large luteal cells secrete progesterone at a higher rate (2–40 fold) than small luteal cells [38, 41, 42, 44, 46, 57]. Small luteal cells respond to maximally effective doses of LH with a large increase (up to 40-fold) in secretion of progesterone, while LH has little or no effect on large luteal cells [38, 39, 41, 42]. In normally cycling animals, a similar number of receptors for LH has been observed on large and small luteal cells in the ewe [58], cow [59], and rat [44]. Large luteal cells produce over 80% of the progesterone secreted by the CL during the midluteal phase of the estrous cycle [60].

There is evidence that during the ovine estrous cycle, luteal 3β-HSD mRNA is maximally expressed by Day 3 and remains relatively constant through Day 12 [26], while maximum expression of $P450_{scc}$ mRNA does not occur until the midluteal phase [61]. This suggests that these enzymes are differentially regulated in ovine luteal cells (Fig. 3). Differential regulation of these messages also appears to occur in rats [62]. Removal of LH support in monkeys was reported to cause dramatic down-regulation of mRNA for both $P450_{scc}$ and 3β-HSD [63]. Interestingly, removal of prolactin support decreased luteal mRNA encoding for $P450_{scc}$ and the enzyme itself in pregnant rats [64].

The mechanism whereby LH stimulates secretion of progesterone from small luteal cells involves formation of cAMP, activation of PKA, and subsequently increased progesterone production ([65]; Fig. 4). Generation of cAMP and activation

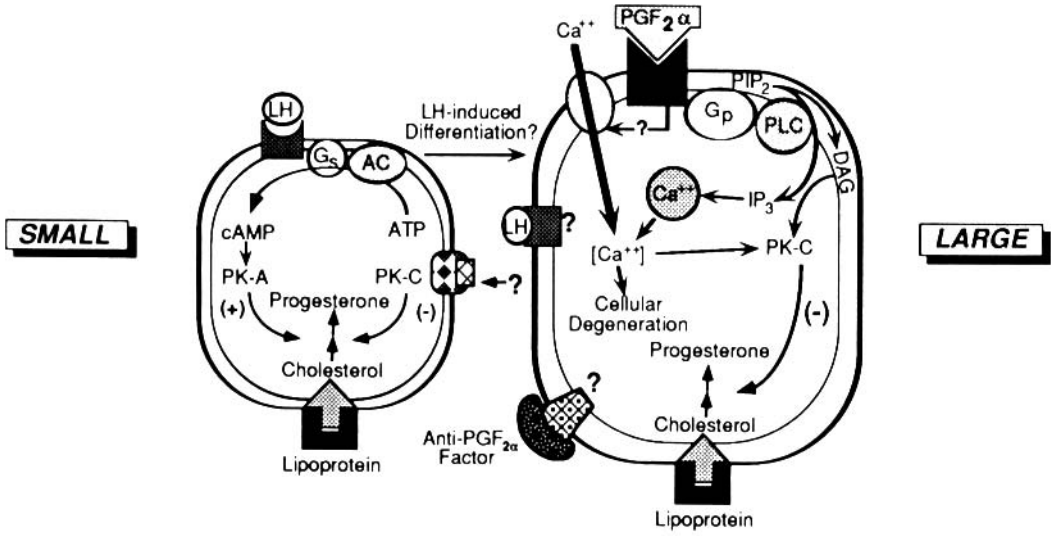

FIG. 4. Current model of the regulation of progesterone secretion from the two types of ovine luteal cells. LH activates protein kinase A (PK-A) and stimulates secretion of progesterone from small but not large luteal cells. Activation of PK-C inhibits secretion of progesterone from LH-stimulated small luteal cells and from large luteal cells. $PGF_{2\alpha}$ activates PK-C in large cells but it is not clear what activates PK-C in small cells. $PGF_{2\alpha}$ also stimulates influx of Ca^{++} in large cells which appears to induce changes associated with cellular degeneration. An anti-$PGF_{2\alpha}$ factor prevents the anti-steroidogenic and cytotoxic effects of $PGF_{2\alpha}$ in large cells through some unknown cellular mechanism. How LH exerts its trophic effect on large cells or how $PGF_{2\alpha}$ exerts cytotoxic effects on small cells is not known at the present time. Adapted from Wiltbank et al. [104].

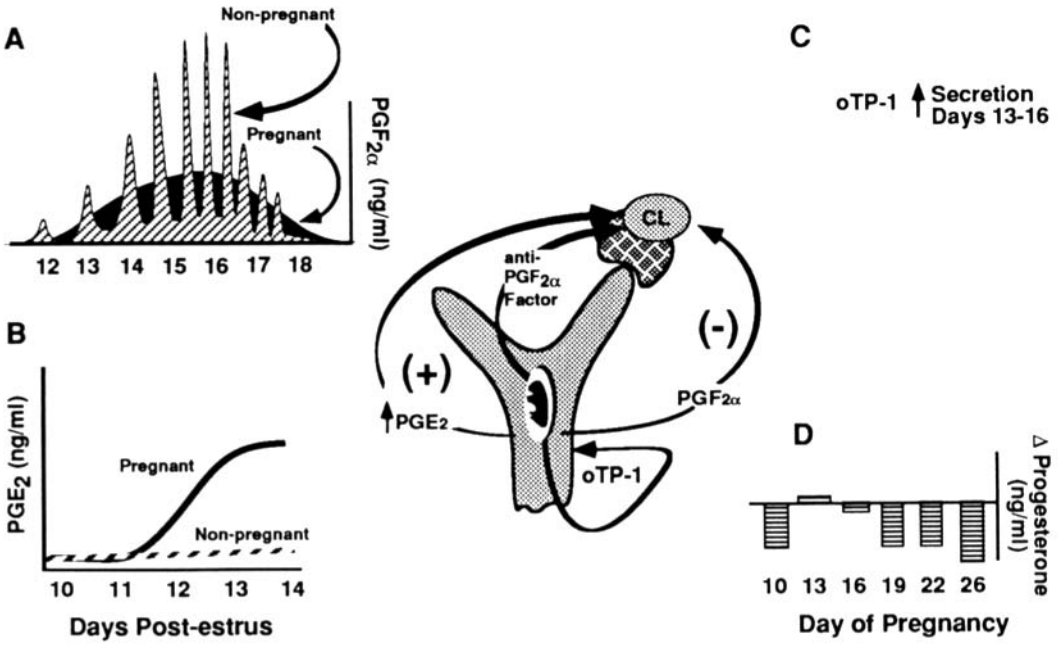

FIG. 5. Current model for maternal recognition of pregnancy in the ewe. A) High amplitude pulsatile secretions of $PGF_{2\alpha}$ do not occur during pregnancy, although basal levels are higher than in ewes during the late luteal phase of the estrous cycle. This change in the pattern of $PGF_{2\alpha}$ secretion appears to be due to oTP-1 secretion from the embryo. B) During the time of maternal recognition, uterine secretion of PGE_2 is increased. C) Secretion of oTP-1 is increased during maternal recognition of pregnancy. D) The corpus luteum of ewes on Days 13 and 16 of pregnancy is resistant to the luteolytic actions of $PGF_{2\alpha}$ (4 mg/58 kg) but is not resistant on Days 10, 19, 22, or 26 of pregnancy. An anti-$PGF_{2\alpha}$ factor is produced by the embryo which makes the corpus luteum of pregnancy resistant to $PGF_{2\alpha}$.

of the PKA system stimulates cholesterol esterase activity [21, 66] and may enhance transport of cholesterol to the inner mitochondrial membrane [66, 67]. Large and small luteal cells have similar amounts of PKA activity [68]; but treatment of large cells with cAMP, cholera toxin, or LH does not enhance secretion of progesterone [69]. Activation of the protein kinase C (PKC) enzyme system with phorbol-12 myristate-13 acetate (PMA) decreases progesterone production [68]. The identity of the factor(s) that stimulates PKC activity in small luteal cells is not known.

In the monkey, it has been shown that both small and large luteal cells isolated from tissue collected early in the menstrual cycle respond to hCG, dbcAMP, or PGE_2 with increased progesterone secretion; but responsiveness in small luteal cells was lost when cells were collected past the early luteal phase [46, 70]. Incubation of small or large luteal cells with $PGF_{2\alpha}$ causes an increased production of inositol phosphates (IP) and induces a transient rise in intercellular Ca^{2+} concentrations. Percentages of cells responding increase as the CL ages [71].

Rat large and small luteal cells also respond to LH [44], forskolin, and dbcAMP stimulation with an increase in progesterone secretion [57]. Large luteal cells contain more $P450_{scc}$ and sterol carrier protein-2, suggesting a higher rate of cholesterol transport and conversion to pregnenolone than in small cells [72]. Large but not small luteal cells also express mRNA for IGF-I and IGF-I receptor and respond to IGF-1 with an increase in progesterone secretion [73].

Receptors for steroid hormones are also present in most CL. Estrogen receptors have been localized in sheep [74] and rats [75]; and in rats and rabbits it is well established that estrogens are involved in maintaining and enhancing luteal progesterone secretion [56, 77]. CL of monkeys contain androgen [78] and progesterone receptors [76], but their function has not been defined. While the positive effect of estradiol on progesterone production in vivo in CL from pregnant rats is clear, conflicting data have been reported in vitro. Nelson et al. [44] reported no effect on basal progesterone production by estradiol, whereas Tekpetey and Armstrong [57] reported an inhibitory effect of estradiol on progesterone secretion. That the dose of estradiol varied greatly between these two experiments perhaps explains the differing results.

LUTEAL REGRESSION

If pregnancy does not occur, it is essential that the CL regress, allowing initiation of a new reproductive cycle. Two processes are involved in loss of luteal function at the end of the cycle. First, there is decreased secretion of progesterone followed by loss of luteal tissue, or luteolysis. At the onset of luteal regression, there is a precipitous decline in concentrations of progesterone in serum [33, 79, 80] fol-

lowed by loss of luteal weight [81, 82]. Morphological changes during luteal regression include accumulation of lipid droplets in the cytoplasm of luteal cells, degeneration of capillaries, and an increase in the number of primary lysosomes [83]. As luteal regression continues, there is an eventual decrease in the number of steroidogenic luteal cells [37, 82].

Prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) of uterine origin is the primary luteolytic agent in domestic farm animals and most rodents (reviewed in [84]). In monkeys, infusion of $PGF_{2\alpha}$ directly into the CL causes premature luteal regression [85]. In most rodents and in ruminants and pigs, the luteolytic action of uterine $PGF_{2\alpha}$ appears to be a local effect, since removal of the uterine horn ipsilateral to the CL prevents luteal regression whereas removal of the contralateral horn has no effect on luteal life span (reviewed in [86]). In primates, the uterus does not appear to be necessary for normal luteal regression [87]; therefore, intraluteal production of $PGF_{2\alpha}$ may be important in modulating luteal life span. An interaction between $PGF_{2\alpha}$ and estrogens appears to be important for normal luteolysis in monkeys [88, 89] and ewes [90, 91]. The role of estrogens may be to regulate luteal levels of receptors for $PGF_{2\alpha}$.

The CL is not always maintained for its normal duration but may instead undergo premature luteal regression. This short luteal phase has been studied extensively in beef cattle; it occurs at puberty [92] and during the transition from postpartum anestrus to cyclicity [93]. Evidence is accumulating that the CL regresses early as a result of a premature release of $PGF_{2\alpha}$ from the uterus [94].

Prostaglandin $F_{2\alpha}$ appears to have multiple biological actions, all of which have a negative effect on luteal function. Early investigators proposed that luteolysis might be the result of reduced luteal blood flow [95]. The CL is a highly vascularized gland that receives over 80% of the ovarian blood supply [83]. Injection of $PGF_{2\alpha}$ into rats does not decrease ovarian blood flow [96], but $PGF_{2\alpha}$ causes a rapid reduction in luteal blood flow in ewes [83]. It remains unclear whether reduced luteal blood flow is a major factor in the initiation of luteal regression or simply a symptom of luteolysis.

It has been reported that numbers of receptors for LH decrease after treatment with $PGF_{2\alpha}$ in rats [97] and ewes [81]. However, in the latter study, the decrease in number of receptors did not occur until after a significant decrease in concentrations of progesterone in serum. In rats, administration of $PGF_{2\alpha}$ decreased plasma membrane fluidity and increased superoxide radical formation [98, 99]. Thus, in the rat an initial site affected by $PGF_{2\alpha}$ may be the plasma membrane.

The antisteroidogenic effects of $PGF_{2\alpha}$ appear to be mediated through the PKC second messenger system (Fig. 4). $PGF_{2\alpha}$ activates phospholipase C [100, 101], which causes hydrolysis of membrane phosphatidylinositol 4,5-bisphosphate (PIP_2) to inositol-1,4,5-triphosphate (IP_3) and 1,2-diacylglycerol (DAG) [102]. Diacylglycerol increases the affinity of PKC for calcium and IP_3 releases calcium from intracellular stores, resulting in an increase in free intracellular calcium concentrations [103] and activation of PKC. In the rhesus monkey, treatment of dispersed luteal cells with $PGF_{2\alpha}$ results in an increase in PIP_2 hydrolysis [71]. In populations of purified large ovine luteal cells, the addition of $PGF_{2\alpha}$ results in activation of PKC [104]. Pharmacological activation of PKC reduces progesterone production from large ovine luteal cells [104–106] and isolated rat luteal cells [107]. The acute antisteroidogenic effects of PKC do not appear to be exerted directly on the activity of any steroidogenic enzyme; rather they appear to inhibit cholesterol transport to cytochrome $P450_{scc}$ [66]. However, activation of PKC may have long-term effects on steroidogenic enzymes. $PGF_{2\alpha}$ has been found to reduce steady-state levels of mRNA encoding 3β-HSD [26]. Interestingly, pharmacologic activation of PKC has no detrimental effects on cell viability [104, 106]. Similar effects have been noted after PMA treatment of ewes in vivo [108]. The second messenger system implicated in mediating the luteolytic effects of $PGF_{2\alpha}$ is free intracellular calcium. Treatment of ovine large luteal cells with $PGF_2\alpha$ increases free intracellular calcium concentrations [104]. Large luteal cells are unable to equilibrate this increased calcium; ovine luteal cells cultured in the presence of A23187 died in culture [106]. Treatment of dispersed rhesus monkey luteal cells with $PGF_{2\alpha}$ also caused a rapid but transient increase in concentrations of free intracellular calcium in both cell types [71].

Ovine luteal cells appear to undergo apoptosis during $PGF_{2\alpha}$ induced luteolysis [109], and cleavage of DNA into characteristic oligonucleosome-size fragments by endonucleases appears to be a common mechanism in the apoptotic process [110]. The developmental expression of a calcium/magnesium-dependent endonuclease has been demonstrated in both granulosa and luteal cells in the rat [111]. In the cow and ewe, $PGF_{2\alpha}$ treatment results in oligonucleosome formation similar to that seen in other cases of apoptosis [112, 113]. This effect is not due to activation of PKC, since infusion of PMA into the ovarian artery of ewes has been found to decrease serum levels of progesterone without oligonucleosome formation [113].

In summary, the mechanisms involved in luteal regression in the different species are complex and varied; but evidence is accumulating that the antisteroidogenic actions of $PGF_{2\alpha}$ are mediated through activation of the PKC pathway while the luteolytic actions of $PGF_{2\alpha}$ are most likely to be manifested through the process of apoptosis, with increases in concentrations of free intracellular calcium being the signal for induction of this process.

EARLY PREGNANCY

Normal pregnancy depends upon the early embryo's signaling its presence to the maternal system, a process termed

maternal recognition of pregnancy. Hormones involved in this signaling differ between species, as do the mechanisms by which these signals maintain the CL. In species in which secretion of $PGF_{2\alpha}$ from the uterus is the primary signal for luteolysis, an obvious potential strategy for blocking luteolysis is the inhibition of $PGF_{2\alpha}$ secretion. In cattle there appears to be inhibition of both basal and oxytocin- or estradiol-stimulated $PGF_{2\alpha}$ secretion in pregnant animals, possibly due to secretion of an endometrial prostaglandin inhibitor [114]. In sheep, there is an inhibition of pulsatile $PGF_{2\alpha}$ secretion but an apparent increase in basal $PGF_{2\alpha}$ secretion (Fig. 5) [115]. Alterations in $PGF_{2\alpha}$ production in ruminants are probably due to secretion of embryonic interferon, also termed interferon tau or ovine or bovine trophoblast protein-1 (see reviews [116–119]). There is also evidence from sheep that embryo-derived platelet-activating factor may play a role in suppressing uterine $PGF_{2\alpha}$ secretion [120].

In pigs there appears to be an alteration in $PGF_{2\alpha}$ secretion such that $PGF_{2\alpha}$ is secreted into the uterine lumen rather than into the bloodstream [118]. This change in $PGF_{2\alpha}$ secretion appears to be due to secretion of estradiol by the early pig embryo. It appears that estradiol-induced calcium cycling and synergistic effects of prolactin may be involved in this redirection of $PGF_{2\alpha}$ secretion [118].

Another potential mechanism for maintaining the CL is inhibition of the action of secreted $PGF_{2\alpha}$ at the level of the luteal cell. In sheep, injections of low doses of $PGF_{2\alpha}$ will induce luteolysis in nonpregnant but not in pregnant animals (Fig. 5) [121, 122]. Wiepz et al. [123] found that the number of receptors for $PGF_{2\alpha}$ was similar in pregnant and nonpregnant animals during the period of maternal recognition of pregnancy. In addition, responsiveness of large luteal cells to $PGF_{2\alpha}$ was not different between pregnant and nonpregnant animals [124]. Thus, it appears that the pregnant uterus secretes a factor that reduces the luteolytic effects of $PGF_{2\alpha}$ injections.

The factor from the pregnant uterus may be PGE_2, since this hormone inhibits the $PGF_{2\alpha}$-induced decrease in progesterone synthesis and since treatments with PGE_2 appear to lengthen the life span of the CL in sheep [125], cattle [126], and primates [127]. However, definitive studies on the role of PGE_2 in maternal recognition of pregnancy have not yet been performed. Another substance that may be important during maternal recognition of pregnancy in sheep is a luteal protective protein(s), secreted by the early ovine embryo, that appears to antagonize the action of $PGF_{2\alpha}$ (Fig. 5) [128].

In primates, the secretion of chorionic gonadotropin appears to be the central factor in the maintenance of the CL during early pregnancy (reviewed in [18]). This hormone has been found to stimulate luteal progesterone secretion both in vivo and in vitro and may inhibit intraluteal production of $PGF_{2\alpha}$, allowing sustained luteal function [129].

In the rat there are dramatic increases in serum progesterone concentrations as well as in luteal weight between Days 10 and 16 of gestation [130]. Decidual and trophoblastic tissues both secrete a prolactin-like luteotrophin important for stimulating luteal function [56]. This luteotrophin maintains LH receptor numbers and the capacity of luteal cells to secrete estradiol. Prolactin (or prolactin-like compounds) is sufficient to maintain luteal function for 10 days after ovulation; however, later increases in luteal weight and progesterone secretion require both prolactin and intraluteal estradiol [56, 131]. Thus, pregnancy-associated increases in luteal function in the rat are due to a synergistic action of prolactin-like luteotrophins and estradiol.

Estradiol also has a key role in maintenance of the CL in rabbits [132]. Although there is not a dramatic stimulation of progesterone secretion during pregnancy, there is an approximate doubling in luteal life span (30 vs. 15 days) as compared to pseudopregnancy [77]. This extended life span of the CL during pregnancy appears to be due to an unidentified embryonic factor, probably of placental origin, that interacts with estradiol to maintain luteal function during pregnancy [132].

Although successful maternal recognition of pregnancy is essential to the survival of an embryo and ultimately of a species, the mechanisms for maternal recognition of pregnancy do not appear to be well conserved during mammalian evolution. Maintenance of the CL of pregnancy is due to a surprisingly diverse group of hormones and mechanisms in different species. In general, these diverse mechanisms are focused on preventing regression of the CL either by stimulating luteal function or by inhibiting $PGF_{2\alpha}$ secretion or action. Questions remain on the identity of certain factors involved in maternal recognition of pregnancy as well as on the intracellular mechanisms for many of the hormones involved in this process.

ACKNOWLEDGMENTS

This research was supported by NIH grant HD11590, and a number of trainees who contributed to this research were supported by NIH Training Grant HD07031. Dr. Gregory Wiepz developed Figure 5. The authors also wish to thank Kathy Thomas and Carol Moeller for production of graphs and typing.

REFERENCES

1. Frankel L. Die function des corpus luteum. Arch Gynaekol 1903; 68:438–443.
2. Allen WM, Wintersteiner O. Crystalline progestin. Science 1934; 80:190–191.
3. Butenandt A, Westphal V. Zur isoliersungund charakteryerung des corpus-luteum hormones. Bev Chem Ges Frankfurt 1934; 63:1440–1442.
4. Hartmann M, Wettstein A. Zur kenntnis der corpus luteum hormone (Z. Mitteilung). Helv Chim Acta 1934; 17:878–882.
5. Slotta KH, Ruschig H, Fells E. Reindarstellung der hormone aus dem corpus luteum. Ber Dtsch Chem Ges 1934; A67:1270–1273.
6. Andersen JM, Dietschy JM. Relative importance of high and low density lipoproteins in the regulation of cholesterol synthesis in the adrenal gland, ovary, and testis of the rat. J Biol Chem 1978; 253:9024–9032.
7. Azhar S, Menon KMJ. Receptor-mediated gonadotropin action in the ovary. Rat luteal cells preferentially utilize and are acutely dependent upon the plasma lipoprotein-supplied sterols in gonadotropin-stimulated steroid production. J Biol Chem 1981; 256:6548–6555.

8. Brown MS, Goldstein JL. A receptor-mediated pathway for cholesterol homeostasis. Science 1986; 232:34–47.
9. Schuler LA, Langenberg KK, Gwynne JT, Strauss III JF. High density lipoprotein utilization by dispersed rat luteal cells. Biochim Biophys Acta 1981; 664:583–601.
10. Jefcoate CR, McNamara BC, Artemenko I, Yamazaki T. Regulation of cholesterol movement to mitochondrial cytochrome $P450_{scc}$ in steroid hormone synthesis. J Steroid Biochem Mol Biol 1992; 43:751–767.
11. Rajkumar K, Couture RL, Murphy BD. Binding of high-density lipoproteins to luteal membranes: the role of prolactin, luteinizing hormone, and circulation lipoproteins. Biol Reprod 1985; 32:546–555.
12. Brannian JD, Shiigi SM, Stouffer RL. Gonadotropin surge increases fluorescent-tagged low-density lipoprotein uptake by macaque granulosa cells from preovulatory follicles. Biol Reprod 1992; 47:355–360.
13. Hall PF, Almahbobi G. The role of the cytoskeleton in the regulation of steroidogenesis. J Steroid Biochem Mol Biol 1992; 43:769–777.
14. Rodgers RJ, Waterman MR, Simpson ER. Levels of messenger ribonucleic acid encoding cholesterol side-chain cleavage cytochrome P-450, 17α-hydroxylase cytochrome P-450, adrenoxin, and low density lipoprotein receptor in bovine follicles and corpora lutea throughout the ovarian cycle. Mol Endocrinol 1987; 1:274–279.
15. Golos TG, Miller WL, Strauss III JF. Human chorionic gonadotropin and 8-bromo cyclic adenosine monophosphate promote an acute increase in cytochrome $P450_{scc}$ and adrenodoxin messenger RNAs in cultured human granulosa cells by a cycloheximide-insensitive mechanism. J Clin Invest 1987; 80:896–899.
16. Labrie F, Simard J, Luv-The V, Belanger A, Pelletier G. Structure, function and tissue-specific gene expression of 3β-hydroxysteroid dehydrogenase/5-ene-4-ene isomerase enzymes in classical and peripheral intracrine steroidogenic tissues. J Steroid Biochem Mol Biol 1992; 43:805–826.
17. O'Shea JD, Cran DG, Hay MF. Fate of the theca interna following ovulation in the ewe. Cell Tissue Res 1980; 210:305–319.
18. Niswender GD, Nett TM. The corpus luteum and its control. In: Knobil E, Neill J (eds.), The Physiology of Reproduction. New York: Raven Press, 1988: 489–525.
19. McClellan MC, Diekman MA, Abel JH, Niswender GD. Luteinizing hormone, progesterone and the morphological development of normal and superovulated corpora lutea in sheep. Cell Tissue Res 1975; 164:291–307.
20. Voss AK, Fortune JE. Levels of messenger ribonucleic acid for cholesterol side chain cleavage cytochrome P-450 and 3-β hydroxysteroid dehydrogenase in bovine preovulatory follicles decreases after the luteinizing hormone surge. Endocrinology 1993; 132:888–894.
21. Caffrey JL, Fletcher PW, Diekman MA, O'Callaghan PL, Niswender GD. The activity of ovine luteal cholesterol esterase during several experimental conditions. Biol Reprod 1979; 21:601–608.
22. Rodgers RJ, Waterman MR, Simpson ER. Cytochromes P-450_{scc}, P-$450_{scc17\alpha}$, adrenoxin, and reduced nicotinamide adenine dinucleotide phosphate-cytochrome P-450 reductase in bovine follicles and corpora lutea. Endocrinology 1986; 118:1366–1374.
23. Richards JS, Jahnsen T, Hedin L, Lifka J, Ratoosh S, Durica JM, Goldring NB. Ovarian follicular development from physiology to molecular biology. Recent Prog Horm Res 1987; 43:231–276.
24. Bassett SG, Little-Ihrig LL, Mason JI, Zeleznick AJ. Expression of messenger ribonucleic acids that encode for 3β-hydroxysteroid dehydrogenase and cholesterol side-chain cleavage enzymes throughout the luteal phase of the macaque menstrual cycle. J Clin Endocrinol & Metab 1991; 72:362–366.
25. Couet J, Martel C, Dupont E, Luu-The V, Sirard M-A, Zhao H-F, Pelletier G, Labrie F. Changes in 3β-hydroxysteroid dehydrogenase/Δ^5-Δ^4 isomerase messenger ribonucleic acid, activity and protein levels during the estrous cycle in the bovine ovary. Endocrinology 1990; 127:2141–2148.
26. Hawkins DE, Belfiore CJ, Kile JP, Niswender GD. Regulation of mRNA encoding 3β-hydroxysteroid dehydrogenase/Δ^5-Δ^4-isomerase (3β-HSD) in the ovine corpus luteum. Biol Reprod 1993; 48:1185–1190.
27. Hedin L, Rodgers RJ, Simpson ER, Richards JS. Changes in content of cytochrome $P450_{17\alpha}$, cytochrome $P450_{scc}$, and 3-hydroxy-3-methylglutaryl CoA reductase in developing rat ovarian follicles and corpora lutea: correlation with theca cell steroidogenesis. Biol Reprod 1987; 37:211–223.
28. Hickey GJ, Chem S, Besman MJ, Shively JE, Hall PF, Gaddy-Kurten D, Richards JS. Hormonal regulation, tissue distribution, and content of aromatase cytochrome P450 messenger ribonucleic acid and enzyme in rat ovarian follicles and corpora lutea: relationship to estradiol biosynthesis. Endocrinology 1988; 122:1426–1436.
29. Doody KJ, Lorence MC, Mason JI, Simpson ER. Expression of messenger ribonucleic acid species encoding steroidogenic enzymes in human follicles and corpora lutea throughout the menstrual cycle. J Clin Endocrinol & Metab 1990; 70:1041–1045.
30. LaPolt PS, Oikawa M, Jai X-C, Dargan C, Hseuh AJW. Gonadotropin-induced up- and down-regulation of rat ovarian LH receptor message levels during follicular growth, ovulation and luteinization. Endocrinology 1990; 126:3277–3279.
31. Segaloff DL, Wang H, Richards JS. Hormonal regulation of luteinizing hormone/chorionic gonadotropin receptor mRNA in rat ovarian cells during follicular development and luteinization. Mol Endocrinol 1990; 4:1856–1865.
32. Nakamura K, Minegishi T, Takakura Y, Miyamoto K, Hasegawa Y, Ibuki Y, Igarashi M. Hormonal regulation of gonadotropin receptor mRNA in rat ovary during follicular growth and luteinization. Mol Cell Endocrinol 1991; 82:259–263.
33. Diekman MA, O'Callaghan P, Nett TM, Niswender GD. Validation of methods and quantification of luteal receptors for LH throughout the estrous cycle and early pregnancy in ewes. Biol Reprod 1978; 19:999–1009.
34. Oxberry BS, Greenwald GW. An autoradiographic study of the binding of ^{125}I-labeled follicle stimulating hormone, human chorionic gonadotropin and prolactin to the hamster ovary throughout the estrous cycle. Biol Reprod 1982; 27:505–516.
35. Manns JG, Niswender GD, Braden T. FSH receptors in the bovine corpus luteum. Theriogenology 1984; 22:321–328.
36. Rodgers RJ, O'Shea JD, Bruce NW. Morphometric analysis of the cellular composition of the ovine corpus luteum. J Anat 1984; 138:757–769.
37. Farin CE, Moeller CL, Sawyer HR, Gamboni R, Niswender GD. Morphometric analysis of cell types in the ovine corpus luteum throughout the estrous cycle. Biol Reprod 1986; 35:1299–1308.
38. Fitz TA, Mayan MH, Sawyer HR, Niswender GD. Characterization of two steroidogenic cell types in the ovine corpus luteum. Biol Reprod 1982; 27:703–711.
39. Rodgers RJ, O'Shea JD. Purification, morphology, and progesterone production and content of three cell types isolated from the corpus luteum of the sheep. Aust J Biol Sci 1982; 35:441–445.
40. Ursely J, Leymarie P. Varying response to luteinizing hormone of two luteal cell types isolated from bovine corpus luteum. J Endocrinol 1979; 83:303–310.
41. Koos RD, Hansel W. The large and small cells of the bovine corpus luteum: ultrastructural and functional differences. In: Schwartz NB, Hunzicker-Dunn M (eds.), Dynamics of Ovarian Function. New York: Raven Press; 1981: 197–203.
42. Lemon M, Loir M. Steroid release *in vitro* by two luteal cell types in the corpus luteum of the pregnant sow. J Endocrinol 1977; 72:351–359.
43. Miyauchi F, Midgley AR Jr. Morphologically and functionally distinct subpopulations of steroidogenic cells in corpora lutea during pregnancy in rats. Endocrinol Jpn 1990; 37:649–663.
44. Nelson SE, McLean MP, Jayatilak PG, Gibori G. Isolation, characterization, and culture of cell subpopulations forming the pregnant rat corpus luteum. Endocrinology 1992; 130:954–966.
45. Hoyer PB, Keyes PL, Niswender GD. Size distribution and hormonal responsiveness of dispersed rabbit luteal cells during pseudopregnancy. Biol Reprod 1986; 34:905–910.
46. Hild-Petito SA, Shiigi SM, Stouffer RL. Isolation and characterization of cell subpopulations from the monkey corpus luteum of the menstrual cycle. Biol Reprod 1989; 40:1075–1085.
47. Ohara A, Mori T, Taii S, Ban C, Narimoto K. Functional differentiation in steroidogenesis of two types of luteal cells isolated from mature human corpora lutea of menstrual cycle. J Clin Endocrinol & Metab 1987; 65:1192–1200.
48. Sawyer HR, Abel JH Jr, McClellan MC, Schmitz M, Niswender GD. Secretory granules and progesterone secretion by ovine corpora lutea *in vitro*. Endocrinology 1979; 104:476–486.
49. Fields PA. Ultrastructural localization of relaxin in corpora lutea of pregnant, pseudopregnant and cycling gilts using porcine relaxin antiserum and goat anti-rabbit IgG-colloidal gold. Biol Reprod 1984; 30(suppl):116A.
50. O'Shea JD, Rodgers RJ, Wright PJ. Cellular composition of the sheep corpus luteum in the mid- and late luteal phases of the oestrous cycle. J Reprod Fertil 1986; 76:685–691.
51. O'Shea JD, Rodgers RJ, D'Occhio MJ. Cellular composition of the cyclic corpus luteum of the cow. J Reprod Fertil 1989; 85:483–487.
52. Cameron JL, Stouffer RL. Gonadotropin receptors of the primate corpus luteum. II Changes in available luteinizing hormone- and chorionic gonadotropin-binding sites in macaque luteal membranes during the non-fertile menstrual cycle. Endocrinology 1982; 110:2068–2073.
53. Lee CY, Coulan CB, Jiang NS, Ryan RJ. Receptors for human luteinizing hormone in human corpora lutea tissue. J Clin Endocrinol & Metab 1973; 36:148–152.
54. Ravindranath N, Little-Ihrig LL, Zeleznick AJ. Characterization of the levels of messenger ribonucleic acid that encode for luteinizing hormone receptor dur-

ing the luteal phase of the primate menstrual cycle. J Clin Endocrinol & Metab 1992; 74:779–785.
55. Richards JS, Williams JJ. Luteal cell receptor content for prolactin (PRL) and luteinizing hormone (LH): regulation by LH and PRL. Endocrinology 1976; 99:1571–1581.
56. Gibori G, Khan I, Warshaw ML, McLean MP, Puryear TK, Nelson S, Durkee TJ, Azhar S, Steinschneider A, Rao MC. Placental-derived regulators and the complex control of luteal cell function. Recent Prog Horm Res 1988; 44:377–425.
57. Tekpetey FR, Armstrong DT. Steroidogenic response of rat and pig luteal cells to estradiol-17β and catecholestrogens in vitro. Biol Reprod 1991; 45:498–505.
58. Harrison LM, Kenny N, Niswender GD. Progesterone production, LH-receptors, and oxytocin secretion by ovine luteal cell types on days 6, 10 and 15 of the oestrous cycle and day 25 of pregnancy. J Reprod Fertil 1987; 79:539–548.
59. Chegini N, Lei ZM, Rao ChV, Hansel W. Cellular distribution and cycle phase dependency of gonadotropin and eicosanoid binding sites in bovine corpora lutea. Biol Reprod 1991; 45:506–513.
60. Niswender GD, Schwall RH, Fitz TA, Farin CE, Sawyer HR. Regulation of luteal function in domestic ruminants: new concepts. Recent Prog Horm Res 1985; 41:101–151.
61. Belfiore CJ, Hawkins DE, Niswender GD. Regulation of the mRNA encoding cytochrome P450 side-chain cleavage ($P450_{scc}$) in the ovine corpus luteum (CL). Proc West Sec Anim Sci 1992; 43:55–58.
62. Kaynard AH, Periman LM, Simard J, Melner MH. Ovarian 3-β hydroxysteroid dehydrogenase and sulfated glycoprotein-2 gene expression are differentially regulated by the induction of ovulation, pseudopregnancy and luteolysis in the immature rat. Endocrinology 1992; 130:2192–2200.
63. Ravindranath N, Little-Ihrig L, Benyo DF, Zeleznick AJ. Role of luteinizing hormone in the expression of cholesterol side-chain cleavage cytochrome P450 and 3-β hydroxysteroid dehydrogenase in the primate corpus luteum. Endocrinology 1992: 131:2065–2070.
64. Hickey GJ, Oonk RB, Hall PF, Richards JS. Aromatase cytochrome P450 and cholesterol side-chain cleavage cytochrome P450 in corpora lutea of pregnant rats: diverse regulation by peptide and steroid hormones. Endocrinology 1989; 125:1673–1682.
65. Hoyer PB, Niswender GD. Adenosine 3′,5′-monophosphate-binding capacity in small and large ovine luteal cells. Endocrinology 1986; 119:1822–1829.
66. Wiltbank MC, Belfiore CJ, Niswender GD. Steroidogenic enzyme activity after acute activation of protein kinase (PK) A and PKC in ovine small and large luteal cells. Mol Cell Endocrinol 1993; (in press).
67. Ghosh DK, Dunham WR, Sands RH, Menon KMJ. Regulation of cholesterol side-chain cleavage enzyme activity by gonadotropin in rat corpus luteum. Endocrinology 1987; 121:21–27.
68. Wiltbank MC, Knickerbocker JJ, Niswender GD. Regulation of the corpus luteum by protein kinase C. I. Phosphorylation activity and steroidogenic action in large and small ovine luteal cells. Biol Reprod 1989; 40:1194–1200.
69. Hoyer PB, Fitz TA, Niswender GD. Hormone-independent activation of adenylate cyclase in large steroidogenic ovine luteal cells does not result in increased progesterone secretion. Endocrinology 1984: 114:604–608.
70. Brannian JD, Stouffer RL. Progesterone production by monkey luteal cell subpopulations at different stages of the menstrual cycle: changes in agonist responsiveness. Biol Reprod 1991; 44:141–149.
71. Houmard BS, Guan Z, Stokes BT, Ottobre JS. Activation of the phosphatidylinositol pathway in the primate corpus luteum by prostaglandin $F_{2\alpha}$. Endocrinology 1992; 131:743–748.
72. McLean MP, Nelson SE, Billheimer JT, Gibori G. Differential capacity for cholesterol transport and processing in large and small rat luteal cells. Endocrinology 1992; 131:2203–2212.
73. Parmer TG, Roberts CT Jr, LeRoith D, Adashi EY, Khan I, Solan N, Nelson S, Zelberstein M, Gibori G. Expression, action, and steroidal regulation of insulin-like growth factor-I (IGF-I) and IGF-I receptor in the rat corpus luteum: their differential role in the two cell populations forming the corpus luteum. Endocrinology 1991; 129:2924–2932.
74. Glass JD, Fitz TA, Niswender GD. Cytosolic receptor for estradiol in the corpus luteum of the ewe: variation throughout the estrous cycle and distribution between large and small steroidogenic cell types. Biol Reprod 1984; 31:967–974.
75. Richards JS. Estradiol receptor content in rat granulosa cells during follicular development: modification by estradiol and gonadotrophins. Endocrinology 1975; 97:1174–1184.
76. Hild-Petito S, Stouffer RL, Brenner RM. Immunocytochemical localization of estradiol and progesterone receptors in the monkey ovary throughout the menstrual cycle. Endocrinology 1988; 123:2896–2905.
77. Keyes PL, Gadsby JE, Yuh K-CM, Bill II CH. The corpus luteum. In: Greep RD (ed.), International Review of Physiology, vol. 27. Baltimore: University Park Press; 1983: 57–98.
78. Hild-Petito S, West NB, Brenner RM, Stouffer RL. Localization of androgen receptor in the follicle and corpus luteum of the primate ovary during the menstrual cycle. Biol Reprod 1991; 44:561–568.
79. Butcher RL, Collins WE, Fugo NW. Plasma concentrations of LH, FSH, prolactin, progesterone and estradiol-17β throughout the 4-day estrous cycle of the rat. Endocrinology 1974; 96:1704–1708.
80. Baird DT. The ovary. In: Austin CR, Short RV (eds.), Reproduction in Mammals, second edition. Cambridge, England: Cambridge University Press; 1984: 91–114.
81. Diekman MA, O'Callaghan P, Nett TM, Niswender GD. Effect of prostaglandin $F_{2\alpha}$ on the numbers of LH receptors in ovine corpora lutea. Biol Reprod 1978; 19:1010–1013.
82. Braden TD, Gamboni F, Niswender GD. Effects of prostaglandin $F_2\alpha$-induced luteolysis on the populations of cells in the ovine corpus luteum. Biol Reprod 1988; 39:245–253.
83. Nett TM, McClellan MA, Niswender GD. Effects of prostaglandins on the ovine corpus luteum: blood flow, secretion of progesterone and morphology. Biol Reprod 1976; 15:66–78.
84. Knickerbocker JJ, Wiltbank MC, Niswender GD. Mechanisms of luteolysis in domestic livestock. Domest Anim Endocrinol 1988; 5:91–107.
85. Auletta FJ, Kamps DL, Pories S, Bisset J, Gibson M. An intracorpus luteum site for the luteolytic action of prostaglandin $F_{2\alpha}$ in the rhesus monkey. Prostaglandins 1984; 27:285–298.
86. Ginther OJ. Internal regulation of physiological processes through local venoarterial pathways: a review. J Anim Sci 1974; 39:550–564.
87. Neill JD, Johansson EDB, Knobil E. Failure of hysterectomy to influence the normal pattern of cyclic progesterone secretion in the rhesus monkey. Endocrinology 1969; 84:464–465.
88. Karsch FJ, Sutton GP. An intra-ovarian site for the luteolytic action of estrogen in the rhesus monkey. Endocrinology 1971; 98:553–561.
89. Auletta FJ, Agins H, Scommegna A. Prostaglandin F mediation of the inhibitory effect of estrogen on the corpus luteum of the rhesus monkey. Endocrinology 1978; 103:1183–1189.
90. Karsch FJ, Noveroske JW, Roche JF, Norton HW, Nalbandov AV. Maintenance of ovine corpora lutea in the absence of ovarian follicles. Endocrinology 1970; 87:1228–1236.
91. Gengenbach DR, Hixon JE, Hansel W. A luteolytic interaction between estradiol and prostaglandin $F_{2\alpha}$ in hysterectomized ewes. Biol Reprod 1977; 16:571–579.
92. Gonzales-Padilla E, Niswender GD, Wiltbank JN. Puberty in beef heifers: II. Effect of injections of progesterone and estradiol-17β on serum LH, FSH, and ovarian activity. J Anim Sci 1975; 40:1105–1109.
93. Odde KG, Wood HS, Kiracofe GH, McKee RM, Kittock RJ. Short estrous cycles and associated serum progesterone levels in beef cows. Theriogenology 1980; 14:105–110.
94. Zollers WG Jr, Garverick HA, Youngquist RS, Ottobre JS, Silcox RW, Copelin JP, Smith MF. In vitro secretion of prostaglandins from endometrium of postpartum beef cows expected to have short or normal luteal phases. Biol Reprod 1991; 44:522–526.
95. Pharriss BB, Cornette JC, Gutnecht GD. Vascular control of luteal steroidogenesis. J Reprod Fertil Suppl 1970; 10:97–103.
96. Behrman HR, Luborsky-Moore JL, Pang CY, Wright K, Dorflinger LJ. Mechanisms of $PGF_{2\alpha}$ action in functional luteolysis. In: Channing CP, Marsh J, Sadler WA (eds.), Ovarian Follicular and Corpus Luteum Function, Advances in Experimental Medicine and Biology. New York: Plenum; Vol. 112: 557–571.
97. Hichens M, Grinwich DL, Behrman HR. $PGF_{2\alpha}$-induced loss of corpus luteum gonadotrophin receptors. Prostaglandins 1974; 7:449–458.
98. Carlson JC, Buhr MM, Riley JC. Plasma membrane changes during corpus luteum regression. Can J Physiol Pharmacol 1989; 67:957–961.
99. Sawada M, Carlson JC. Rapid plasma membrane changes in superoxide radical formation, fluidity, and phospholipase A_2 activity in the corpus luteum of the rat during induction of luteolysis. Endocrinology 1991; 128:2992–2998.
100. Davis JS, Weakland LL, Weiland DA, Farese RV, West LA. Prostaglandin $F_{2\alpha}$ stimulates phosphatidylinositol 4,5-bisphosphate hydrolysis and mobilizes intracellular Ca^{2+} in bovine luteal cells. Proc Natl Acad Sci USA 1987; 84:3728–3732.
101. Veldhuis JD. Prostaglandin $F_{2\alpha}$ initiates phosphatidyl inositol hydrolysis and membrane translocation of protein kinase C in swine ovarian cells. Biochem Biophys Res Commun 1987; 149:112–117.
102. Berridge MJ. Inositol triphosphate and diacylglycerol: two interacting second messengers. Annu Rev Biochem 1987; 56:159–193.
103. Nishizuka Y. Studies and perspectives of protein kinase C. Science 1986; 233:305–311.
104. Wiltbank MC, Diskin MG, Niswender GD. Differential actions of second messenger systems in the corpus luteum. J Reprod Fertil Suppl 1991; 43:65–75.

105. Conley AJ, Ford SP. Effects of TPA, A23187, and prostaglandin $F_{2\alpha}$ on progesterone synthesis by dispersed ovine luteal cells. Biol Reprod 1989; 40:1224–1230.
106. Hoyer PB, Marion SL. Influence of agents that affect intracellular calcium regulation on progesterone secretion in large and small luteal cells of the sheep. J Reprod Fertil 1989; 86:445–455.
107. Baum MS, Rosberg S. A phorbol ester, phorbol 12-myristate 13-acetate, and a calcium ionophore, A23187, can mimic the luteolytic effect of prostaglandin $F_{2\alpha}$ in isolated rat luteal cells. Endocrinology 1987; 120:1019–1026.
108. McGuire WJ, Hawkins DE, Niswender GD. Activation of protein kinase (PK)-C inhibits progesterone production in vivo. Biol Reprod 1992; 46(suppl):84.
109. Sawyer HR, Niswender KD, Braden TD, Niswender GD. Nuclear changes in ovine luteal cells in response to $PGF_{2\alpha}$. Domest Anim Endocrinol 1990; 7:229–238.
110. Compton MM, Haskill JS, Cidlowski JA. Analysis of glucocorticoid actions on rat thymocyte deoxyribonucleic acid by fluorescence-activated flow cytometry. Endocrinology 1988; 122:2158–2164.
111. Zeleznik AJ, Ihrig LL, Basset SG. Developmental expression of a Ca^{++}/Mg^{++}-dependent endonuclease activity in rat granulosa and luteal cells. Endocrinology 1989; 125:2218–2220.
112. Juengel JL, Gaverick HA, Johnson AL, Youngquist RS, Smith MF. Apoptosis during luteal regression in cattle. Endocrinology 1993; 132:249–254.
113. McGuire WJ, Niswender GD. Oligonucleosome formation during $PGF_{2\alpha}$-induced luteolysis or luteal protein kinase C (PKC) activation. Biol Reprod 1993; 48(suppl):101.
114. Thatcher WW, Danet-Desnoyers G, Wetzels C. Regulation of bovine endometrial prostaglandin secretion and the role of bovine trophoblast protein-1 complex. Reprod Fertil Dev 1992; 4:329–334.
115. Inskeep EK, Murdoch WJ. Relation of ovarian functions to uterine and ovarian secretion of prostaglandins during the estrous cycle and early pregnancy in the ewe and cow. Int Rev Physiol 1980; 22:325–356.
116. Roberts RM, Leaman DW, Cross JC. Role of interferons in maternal recognition of pregnancy in ruminants. Proc Soc Exp Biol Med 1992; 200:7–18.
117. Flint APF, Parkinson TJ, Stewart HJ, Vallet JL, Lamming GE. Molecular biology of trophoblast interferons and studies of their effects in vivo. J Reprod Fertil Suppl 1991; 43:13–25.
118. Bazer FW, Mirando MA, Ott TL, Harney JP, Dubois DH, Schalue TK, Pontzer CH, Hostetler C, Johnson HM, Ogle T. Roles of ovine trophoblast protein-1 and oestradiol/prolactin in the establishment of pregnancy in sheep and pigs. Reprod Fertil Dev 1992; 4:335–340.
119. Salamonsen LA, Cherny RA, Findlay JK. In-vitro studies of the effects of interferons on endometrial metabolism in sheep. J Reprod Fertil Suppl 1991; 43:27–38.
120. Battye KM, O'Neill C, Evans G. Platelet activating factor (PAF) suppresses oxytocin-stimulated phosphatidylinositol turnover in ovine endometrium. Proc Endocr Soc Aust 1991; 34:21.
121. Nancarrow CD, Evison BM, Connell PJ. Effect of embryos on luteolysis and termination of early pregnancy in sheep with cloprostenol. Biol Reprod 1982; 26:263–269.
122. Silvia WJ, Niswender GD. Maintenance of the corpus luteum of early pregnancy in the ewe. IV. Changes in luteal sensitivity to prostaglandin $F_{2\alpha}$ throughout early pregnancy. J Anim Sci 1986; 63:1201–1207.
123. Wiepz GJ, Wiltbank MC, Nett TM, Niswender GD, Sawyer HR. Receptors for prostaglandins $F_{2\alpha}$ and E_2 in ovine corpora lutea during maternal recognition of pregnancy. Biol Reprod 1992; 47:984–991.
124. Wiltbank MC, Wiepz GJ, Knickerbocker JJ, Braden TD, Sawyer HR, Mayan MH, Niswender GD. Cellular regulation of corpus luteum function during maternal recognition of pregnancy. Reprod Fertil Dev 1992; 4:341–347.
125. Inskeep EK, Smutny WJ, Butcher RL, Pexton JE. Effects of intrafollicular injections of prostaglandins in non-pregnant and pregnant ewes. J Anim Sci 1975; 41:1098–1104.
126. Gimenez T, Henricks DM. Prolongation of the luteal phase by prostaglandin E_2 during the estrous cycle in the cow. A preliminary report. Theriogenology 1983; 19:693–700.
127. Zelinski-Wooten MB, Stouffer RL. Intraluteal infusions of prostaglandins of the E, D, I, and A series prevent $PGF_{2\alpha}$-induced, but not spontaneous, luteal regression in rhesus monkeys. Biol Reprod 1990; 43:507–516.
128. Wiltbank MC, Wiepz GJ, Knickerbocker JJ, Belfiore C, Niswender GD. Proteins secreted from the early ovine conceptus block the action of prostaglandin $F_{2\alpha}$ on large luteal cells. Biol Reprod 1992; 46:475–482.
129. Houmard BS, Ottobre JS. Progesterone and prostaglandin production by primate luteal cells collected at various stages of the luteal phase: modulation by calcium ionophore. Biol Reprod 1989; 45:560–565.
130. Meyer GT, Bruce NW. The cellular pattern of corpus luteal growth during pregnancy in the rat. Anat Rec 1979; 193:823–830.
131. Keyes PL, Possley RM, Brabec RK. The roles of prolactin and testosterone in the development and function of granulosa lutein tissue in the rat. Biol Reprod 1987; 37:699–707.
132. Gadsby JE. Control of corpus luteum function in the pregnant rabbit. J Reprod Fertil Suppl 1989; 37:45–54.