Prostaglandin secretion by endometrium of pregnant and cyclic cattle at Day 17 after oestrus in response to in-vitro heat stress*

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Summary. Bilateral perfusion devices were utilized to measure prostaglandin F-2 α (PGF) secretion by bovine endometrium in response to in-vitro heat stress. Tissues were collected at Day 17 after oestrus from cyclic (N = 4) and pregnant (N = 5) cows, placed into 3 perfusion devices, perfused (3 ml/10 min, Krebs–Ringer–bicarbonate [KRB]) for 5 h, and fractions were collected every 10 min. Endometrial tissues within each device were subjected to a different temperature and oxytocin (1 i.u./ml KRB) treatment sequence: (1) control–oxytocin: 1 h at 39°C; 2 h at 39°C, 0.5 h at 39°C with oxytocin, 0.5 h at 39°C and 1 h at 39°C; (2) heat–oxytocin: 1 h at 39°C, 2 h at 42°C, 0.5 h at 42°C with oxytocin, 0.5 h at 42°C and 1 h at 39°C; (3) heat–KRB: 1 h at 39°C, 2 h at 42°C, 0.5 h at 42°C, 0.5 h at 42°C and 1 h at 39°C. Regardless of reproductive status, heat stress induced a rapid increase ($P < 0.01$) in PGF secretion rates. Oxytocin induced an increase ($P < 0.01$) in PGF secretion for endometrium from cyclic cows regardless of temperature. Endometria from pregnant cows did not respond to oxytocin when perfused at 39°C. However, PGF secretion rates from endometrium of pregnant cows increased ($P < 0.01$) in response to oxytocin when perfused under heat stress conditions. Increased PGF secretion rates and responsiveness to oxytocin for endometria from pregnant cows in response to heat stress suggests that the ability of the conceptus to inhibit uterine PGF-2 α synthesis may be compromised during early pregnancy when cattle are subjected to high ambient temperatures.

Keywords: endometrium; heat stress; prostaglandin; cattle; oxytocin

Introduction

Environmental heat stress often results in transient infertility in cattle (Gwazdauskas *et al.*, 1973; Ingraham *et al.*, 1974; Thatcher, 1974; Badinga *et al.*, 1985). The early bovine embryo (Days 1–7 of development) is extremely sensitive to the harmful effects of maternal hyperthermia in that the frequency of non-viable, abnormal and retarded embryos is increased (Putney *et al.*, 1986). In addition, maternal heat stress between Days 8 and 16 after insemination reduces conceptus weight resulting in a trend towards increased pregnancy failure in beef cattle (Biggers *et al.*, 1987). In-vitro heat stress of conceptuses recovered at Day 17 of pregnancy induced a marked decrease in the synthesis and secretion of proteins into culture medium (Putney *et al.*, 1987). Since heat stress retards development and reduces protein synthetic capacity of conceptuses, it is possible that embryonic mortality may result, in part, from failure of conceptuses to produce adequate amounts of antiluteolytic biochemical signals necessary to maintain a functional corpus luteum (CL).

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There is considerable evidence that the uterine luteolysin in cattle is prostaglandin (PG) F-2 α (Lauderdale, 1974; Thatcher *et al.*, 1984). Exogenous administration of PGF-2 α will induce luteolysis (Lauderdale, 1974; Thatcher & Chenault, 1976) while passive immunization against PGF-2 α extends corpus luteum life-span (Fairclough *et al.*, 1981). Furthermore, increased PGF-2 α concentrations have been demonstrated in uterine flushings (Bartol *et al.*, 1981), uterine venous plasma (Nancarrow *et al.*, 1973) and in endometrial tissues (Thatcher *et al.*, 1984) before luteolysis. The pre-attachment bovine conceptus secretes an array of proteins that are believed to be involved in attenuating the sequence of events leading to luteolysis (Bartol *et al.*, 1985; Knickerbocker *et al.*, 1986a). These antiluteolytic proteins may act locally on the gravid uterine horn to alter the synthesis and release of PGF-2 α from the endometrium, possibly by stimulating the production of an intracellular endometrial inhibitor of prostaglandin synthesis (Gross *et al.*, 1988a).

Thermal stress between Days 8 and 16 after insemination increased concentrations of 13,14-dihydro-15-keto PGF-2 α (PGFM), a metabolite of PGF-2 α , in the peripheral circulation of gilts (Wettemann *et al.*, 1984). Additionally, Lewis *et al.* (1984) reported that pre-partum heat stress increased the concentrations of PGFM during the post-partum period in dairy cattle. Increased concentrations of systemic PGFM may reflect greater uterine secretion of PGF-2 α in response to heat stress. The present study examined whether in-vitro heat stress would increase the secretion of PGF-2 α by perfused endometrial tissue from pregnant and cyclic cattle at Day 17 after oestrus. Alterations in uterine PGF-2 α secretion may exert pronounced effects on CL life-span and therefore maintenance of pregnancy.

Materials and Methods

Materials. Harvard infusion pump systems were purchased from Harvard Apparatus (Dover, MA, U.S.A.). Oxytocin and radioinert PGF-2 α were purchased from Sigma Chemical Company (St Louis, MO, U.S.A.). Radio-labelled [5,6,8,9,11,12,14,15-³H]PGF-2 α (sp. act. $\approx$ 160–180 Ci/mmol) was purchased from Amersham Corporation (Arlington Heights, IL, U.S.A.). Rabbit antiserum to PGF-2 α was provided by Dr T. G. Kennedy (University of Western Ontario, Canada).

Collection of uterine endometrium. Beef cattle (Angus or Brangus) were observed for behavioural oestrus and were mated by natural service (N = 5) or not mated (N = 4). Cattle were slaughtered on Day 17 after oestrus and reproductive tracts were recovered and flushed with Minimal Essential Medium (MEM, 40 ml) to collect conceptus tissue and confirm pregnancy. The uterine horn ipsilateral to the CL was opened longitudinally along the antimesometrial border and endometrium was dissected free from myometrium over a surface area of 6–7 cm². Each endometrial sample was placed into sterile Krebs'–Ringer bicarbonate solution (KRB) which was gassed with O₂:CO₂ (95:5 v/v), washed twice with fresh KRB, and dissected into three pieces of 2 cm² each.

In-vitro perfusion of endometrium. Each of the three endometrial samples was placed into a bilateral perfusion device which allows for separation of myometrial and luminal surfaces of endometrium so that each side can be perfused separately in a 0.5 ml hemi-chamber (Lacroix & Kann, 1983). Tissue was stretched and anchored into each device with the aid of 4 needles. Each chamber was placed into a water bath (39°C), attached to dual 60-ml syringes containing KRB, and perfused at a rate of 3 ml/10 min for 5 h using a Harvard infusion pump system. Fractions (3 ml) were collected every 10 min for a total of 30 fractions.

Each of the 3 endometrial perfusion preparations within each cow was subjected to a different temperature and oxytocin (1 i.u./ml KRB) treatment sequence: (1) control-oxytocin: 1 h at 39°C, 2 h at 39°C, 0.5 h at 39°C with oxytocin, 0.5 h at 39°C and 1 h at 39°C; (2) heat-oxytocin: 1 h at 39°C, 2 h at 42°C, 0.5 h at 42°C with oxytocin, 0.5 h at 42°C and 1 h at 39°C; (3) heat-KRB: 1 h at 39°C, 2 h at 42°C, 0.5 h at 42°C with no oxytocin, 0.5 h at 42°C and 1 h at 39°C. Temperature treatments were imposed on endometrium by placing perfusion devices in water baths maintained at 39 or 42°C and perfusing tissues with KRB maintained at 39 or 42°C.

Measurement of PGF-2 α . Fractions were analysed for PGF-2 α using a radioimmunoassay (RIA) procedure (Knickerbocker *et al.*, 1986b) modified to use an antibody characterized by Kennedy (1985) and validated by Gross *et al.* (1988b) for analysis of unextracted medium. Cross-reactivities of the PGF-2 α antiserum with other prostaglandins were 94% for PGF-1 α , 2.4% for PGE-2, <1% for 6-keto PGF-1 α , and <0.1% for PGFM, PGE-1 and arachidonic acid. Due to the high cross-reactivity with PGF-1 α , PGF-2 α measurements are defined as PGF. Inter- and intra-assay coefficients of variation were 12.2% and 9.7%, respectively.

Statistical analysis. Data were analysed by Least Squares Analysis of Variance using the General Linear Models procedure of the Statistical Analysis System (SAS, 1985). Prostaglandin secretion rates of luminal, myometrial or

luminal + myometrial sides of endometrium were analysed statistically for the following perfusion treatment periods: (1) 1 h before heat stress, (2) 2 h before oxytocin, (3) 0.5 h with or without oxytocin, (4) 0.5 h after oxytocin, and (5) 1 h after heat stress (Fig. 1). The means $\pm$ s.e. for each period, presented in Tables 2, 3 and 4, are means for the series of 10-min measurements made within each period. The statistical model is presented in Table 1, and analyses for total (luminal + myometrial sides of endometrium) PGF secretion for periods 2 and 3 are also presented in Table 1.

Table 1. Least squares analyses of variance of total (luminal + myometrial surfaces) endometrial PGF secretion rates for thermoneutral vs heat stress (period 2; 2 h at 39 or 42°C) and oxytocin (period 3; 0.5 h at 39°C with oxytocin, or 42°C with or without oxytocin) treatments

Sources of variation ^d	Perfusion treatment periods			
	2		2	
	df	MS $\times 10^{-4}$	df	MS $\times 10^{-4}$
Status ^a	1	2487.9*	1	1827.0**
Cow (status)	7	448.2**	7	146.9**
Treatment ^b	2	1957.8**	2	776.9*
Status $\times$ treatment	2	6.4	2	32.4
Cow (status) $\times$ treatment	10	16.6	10	176.0
Time ^c	11	104.1**	2	48.3
Status $\times$ time	11	27.2*	2	16.5
Treatment $\times$ time	22	30.1**	4	30.8
Status $\times$ treatment $\times$ time	22	15.1	3	35.4
Residual	162	13.1	22	37.2

^aStatus of endometrium: cyclic vs pregnant.

^bPerfusion at 39°C + oxytocin, 42°C + oxytocin or 42°C without oxytocin.

^cPerfusion for 10-min time periods.

^ddf = degrees of freedom; MS = mean squares; * $P < 0.05$, ** $P < 0.01$.

Results

Endometrial tissues remained viable throughout perfusion as indicated by sustained PGF release into KRB solution throughout the duration of perfusion (Fig. 1). Mean secretion rates of total (luminal + myometrial surfaces) PGF when KRB was perfused across tissues maintained at thermoneutral temperatures (treatment period 1) were 46% lower ($P < 0.01$) for endometrium from pregnant compared to cyclic cows (678 ± 34 vs 1282 ± 128 pg/10 min; Table 2).

Endometrial secretion of PGF occurred primarily from the luminal surface of tissues for each reproductive status, but the magnitude of the difference between sides was less ($P < 0.01$) for pregnant (181 pg/10 min) than for cyclic (482 pg/10 min) cows (Tables 3 and 4). Mean PGF secretion rates by tissues maintained at thermoneutral temperature (treatment period 1) were 77% higher ($P < 0.05$) for luminal compared to myometrial surfaces of endometrium from pregnant cows (427 ± 14 vs 241 ± 19 pg/10 min) and 120% higher ($P < 0.01$) for cyclic cows (884 ± 82 vs 402 ± 38 pg/10 min). Although myometrial surfaces secreted less PGF than did luminal surfaces, PGF secretion responses to temperature and oxytocin treatments were parallel between sides (Tables 3 and 4). Therefore, PGF responses detected in luminal and myometrial KRB perfusates were combined to assess total PGF secretion rates by endometrium in response to heat stress and oxytocin challenge (Fig. 1; Tables 1 and 2).

Fig. 1. Total (luminal + myometrial surfaces) secretion rate (pg/10 ml KRB) of PGF by endometrium, of (a) pregnant and (b) cyclic cows, perfused at 39°C with an oxytocin challenge (—) or at 42°C with (---) or without (---) an oxytocin challenge. Perfusion periods analysed were: (1) 1 h at 39°C, (2) 2 h at 39 or 42°C, (3) 0.5 h at 39°C with oxytocin or 42°C with or without oxytocin, (4) 0.5 h at 39 or 42°C, and (5) 1 h at 39°C.

Table 2. Total (luminal + myometrial surfaces) endometrial secretion rates (mean ± s.e.) of PGF (pg/10 min) into KRB during perfusion at the following treatment periods: (1) 1 h at 39°C, (2) 2 h at 39 or 42°C, (3) 0.5 h at 39°C with oxytocin or 42°C with or without oxytocin, (4) 0.5 h at 39 or 42°C, and (5) 1 h at 39°C

Groups	Perifusion treatment period				
	1 ^a	2 ^b	3 ^b	4 ^{a,c}	5 ^a
Pregnant (N = 5)					
39°C-oxytocin	857 ± 87	609 ± 33	530 ± 43	541 ± 54	505 ± 50
42°C-oxytocin	618 ± 43	1572 ± 96	2300 ± 265	1005 ± 153	581 ± 51
42°C-KRB	560 ± 34	1465 ± 114	870 ± 87	885 ± 67	523 ± 35
Cyclic (N = 4)					
39°C-oxytocin	1337 ± 148	1323 ± 100	2352 ± 300	1483 ± 159	1166 ± 86
42°C-oxytocin	1316 ± 156	2323 ± 153	3024 ± 344	1731 ± 177	1104 ± 107
42°C-KRB	1193 ± 105	2124 ± 165	2040 ± 261	1595 ± 128	1199 ± 69

^a*P* < 0.01 for status and cow (status).
^bSee Table 1 for statistical analyses and differences.
^c*P* < 0.01 for treatment.

Before the application of heat stress (treatment period 1), PGF secretion rates by endometria within each reproductive status were similar among treatment groups (Table 2). Elevation of perfusion temperature from 39 to 42°C stimulated PGF secretory capacity of endometrium regardless of reproductive status (Table 2). Overall secretion rates of PGF during the first 2 h of heat stress (treatment period 2) were 68% greater ($P < 0.01$) for cyclic (2223 ± 96 vs 1323 ± 100 pg/10 min) and 149% greater ($P < 0.01$) for pregnant (1518 ± 65 vs 609 ± 33 pg/10 min) endometrium. Heat stress induced a rapid (within 10 min) release of PGF by endometrium from pregnant and cyclic cows. However, the response was not sustained by pregnant endometrium, and declined precipitously within 1 h after initiation of thermal stress (Fig. 1).

Table 3. Secretion rates (mean $\pm$ s.e.) of PGF (pg/10 min) into KRB by luminal surfaces of endometrium perfused during the following treatment periods: (1) 1 h at 39°C, (2) 2 h at 39 or 42°C, (3) 0.5 h at 39°C with oxytocin or 42°C with or without oxytocin, (4) 0.5 h at 39 or 42°C, and (5) 1 h at 39°C

Groups	Perifusion treatment period				
	1 ^a	2 ^b	3 ^c	4 ^d	5 ^e
Pregnant (N = 5)					
39°C-oxytocin	478 $\pm$ 42	386 $\pm$ 18	343 $\pm$ 33	362 $\pm$ 35	337 $\pm$ 22
42°C-oxytocin	435 $\pm$ 29	1080 $\pm$ 55	1332 $\pm$ 97	613 $\pm$ 67	391 $\pm$ 31
42°C-KRB	354 $\pm$ 32	869 $\pm$ 69	563 $\pm$ 77	537 $\pm$ 58	327 $\pm$ 33
Cyclic (N = 4)					
39°C-oxytocin	896 $\pm$ 101	955 $\pm$ 62	1594 $\pm$ 222	1088 $\pm$ 118	879 $\pm$ 77
42°C-oxytocin	948 $\pm$ 93	1410 $\pm$ 78	1952 $\pm$ 127	1144 $\pm$ 83	914 $\pm$ 93
42°C-KRB	809 $\pm$ 78	1362 $\pm$ 99	973 $\pm$ 81	1043 $\pm$ 129	907 $\pm$ 78

^a $P < 0.01$ for status and cow (status).

^b $P < 0.05$ for status and $P < 0.01$ for cow (status), treatment, cow (status) $\times$ treatment and time and treatment $\times$ time.

^c $P < 0.01$ for status and treatment and $P < 0.05$ for cow (status).

^d $P < 0.01$ for status, cow (status) and treatment.

^e $P < 0.01$ for status and cow (status).

Table 4. Secretion rates (mean $\pm$ s.e.) of PGF (pg/10 min) into KRB by myometrial surfaces of endometrium perfused during the following treatment periods: (1) 1 h at 39°C, (2) 2 h at 39 or 42°C, (3) 0.5 h at 39°C with oxytocin or 42°C with or without oxytocin, (4) 0.5 h at 39 or 42°C, and (5) 1 h at 39°C

Groups	Perifusion treatment period				
	1 ^a	2 ^b	3 ^c	4 ^d	5 ^e
Pregnant (N = 5)					
39°C-oxytocin	325 $\pm$ 43	223 $\pm$ 15	232 $\pm$ 15	213 $\pm$ 21	200 $\pm$ 15
42°C-oxytocin	192 $\pm$ 14	494 $\pm$ 41	863 $\pm$ 151	404 $\pm$ 68	205 $\pm$ 11
42°C-KRB	208 $\pm$ 16	597 $\pm$ 45	351 $\pm$ 51	337 $\pm$ 56	203 $\pm$ 12
Cyclic (N = 4)					
39°C-oxytocin	434 $\pm$ 47	368 $\pm$ 17	800 $\pm$ 101	393 $\pm$ 51	304 $\pm$ 14
42°C-oxytocin	417 $\pm$ 63	914 $\pm$ 76	1172 $\pm$ 199	683 $\pm$ 108	381 $\pm$ 3
42°C-KRB	356 $\pm$ 37	763 $\pm$ 66	960 $\pm$ 148	389 $\pm$ 35	293 $\pm$ 33

^a $P < 0.01$ for status and cow (status).

^b $P < 0.05$ for status, time and status $\times$ time and $P < 0.01$ for cow (status), treatment and cow (status) $\times$ treatment.

^c $P < 0.01$ for status, cow (status) and treatment.

^d $P < 0.05$ for status and $P < 0.01$ for cow (status) and treatment.

^e $P < 0.01$ for status and $P < 0.05$ for cow (status).

Perfusion of KRB supplemented with oxytocin (treatment period 3) induced a rapid 164% increase ($P < 0.01$) in secretion of PGF by heat-stressed pregnant endometrium (2300 ± 265 vs 870 ± 87 pg/10 min). In contrast, pregnant endometrium perfused at 39°C did not respond to oxytocin treatment with increased PGF release (Table 2). Cyclic endometrium, regardless of perfusion temperature, increased the rate of PGF secretion in response to oxytocin challenge; a 48% increase ($P < 0.05$) in PGF secretion by heat-stressed tissues (3024 ± 344 vs 2040 ± 261 pg/10 min) and a 77% increase ($P < 0.05$) by thermoneutral tissues (2353 ± 300 vs 1323 ± 100 pg/10 min) was detected. At the conclusion of oxytocin perfusion, secretion rates of PGF declined rapidly and returned to pre-oxytocin rates of secretion within 30 min of perfusion with KRB (treatment period 4; Table 2, Fig. 1).

At the conclusion of heat stress, a reduction in incubation temperature from 42 to 39°C (treatment period 5) reduced ($P < 0.01$) endometrial prostaglandin secretion rates. Overall mean secretion rates of PGF into KRB by tissues within each reproductive status were similar among treatment groups in period 5 (Fig. 1, Table 2).

Discussion

Establishment of pregnancy in cattle is dependent upon interactions between antiluteolytic and luteolytic factors produced by the periattachment conceptus and uterine endometrium, respectively. Maintenance of luteal function in pregnant cattle is associated with diminished secretion of PGF- 2α during the expected period of luteolysis (Thatcher *et al.*, 1984; Gross *et al.*, 1988b). Likewise, the release of PGF- 2α from the uterus in response to oestrogen (Thatcher *et al.*, 1984) and oxytocin (Lafrance & Goff, 1985) is attenuated during early pregnancy. Results from the present study using endometrial perfusions confirm that endometrial secretion of PGF is reduced during early pregnancy in cattle.

Heat stress induced a rapid increase in endometrial secretion rates of PGF by luminal and myometrial surfaces for pregnant and cyclic cows. Increased PGF synthetic capacity of endometrium exposed to heat stress may be due to heat-induced alterations in endometrial cellular membranes resulting in increased mobilization of substrate for prostaglandin biosynthesis. The primary cellular site for the action of heat damage on tissues is membranes (Bowler *et al.*, 1973; Hahn, 1982). Exposure to high ambient temperature is accompanied by alterations in membrane lipid composition (Anderson & Parker, 1982) as well as increases in membrane fluidity, phospholipase activity and phosphoinositide turnover (Calderwood *et al.*, 1987). Heat-induced increases in the turnover of membrane phospholipids and release of fatty acids, such as arachidonic acid, may provide substrates for prostaglandin synthesis (Flint *et al.*, 1986). Arachidonic acid, the principal substrate for PGF- 2α synthesis, is present in large quantities within the bovine endometrium before the time of increased endometrial PGF-2 production and luteolysis (Hansel *et al.*, 1975; Shemesh & Hansel, 1975). Furthermore, exogenous arachidonic acid will stimulate prostaglandin production by pregnant and cyclic endometrial tissues *in vitro* (Thatcher *et al.*, 1984). Elevated secretion rate of PGF by heat-stressed pregnant endometrium was not sustained throughout the duration of heat-stress perfusion, but declined precipitously within 1 h after initiation of thermal stress. This decrease in PGF secretion may be due, in part, to a depletion of the intracellular pool of arachidonic acid in endometrial cells of heat-stressed pregnant cows. Perfusion of heat-stressed endometrium of 2 pregnant cows with KRB supplemented with arachidonic acid ($50 \mu\text{g/ml}$ KRB) resulted in sustained increases in PGF release throughout the period of heat stress (D. J. Putney, T. S. Gross & W. W. Thatcher, unpublished data).

Similar increases in uterine PGF- 2α secretion in response to heat stress *in vitro* by endometrial explants from cows at Day 17 of pregnancy have been reported (Putney *et al.*, 1987). Furthermore, in-vivo heat stress of gilts between Days 8 and 16 of pregnancy increased concentrations of PGFM in the peripheral circulation and compromised luteal function, as indicated by reduced plasma concentrations of progesterone during Days 13 through 19 (Hoagland & Wettemann, 1984;

Wettemann *et al.*, 1984). In cattle, pre-partum heat stress increases peripheral concentrations of PGFM and reduces CL diameter *post-partum* (Lewis *et al.*, 1984). Decreased concentrations of progesterone in plasma of gilts and CL size in cattle may be due to heat-induced partial regression of the CL in stressed animals, which may be attributed to greater uterine secretion of PGF-2 α (reflected by greater concentrations of systemic PGFM) in heat stress than in thermoneutral animals. Since maintenance of luteal function in cattle is associated with decreased endometrial PGF-2 α secretion, it is likely that increases in endometrial prostaglandin secretion rate in response to heat stress may compromise CL function, initiate partial or complete luteal regression and contribute to pregnancy failure.

Endometrium from pregnant cattle, when perfused at thermoneutral temperature, was unable to respond to oxytocin with an increased secretion of PGF, whereas endometria from cyclic cattle responded with an increase in the secretion of PGF. These in-vitro perfusion data are in agreement with previous in-vivo and in-vitro data which demonstrate an attenuated increase in secretion of prostaglandins in response to oxytocin during early pregnancy in cattle (Lafrance & Goff, 1985; Gross *et al.*, 1988b). However, oxytocin induced a rapid increase in luminal and myometrial secretion rates of PGF by endometrium from heat-stressed pregnant cows as well as tissues of cyclic cows, regardless of perfusion temperature. Such a rapid induction of endometrial prostaglandin secretion by oxytocin has been noted previously *in vivo* for cyclic cattle (Oyedipe *et al.*, 1984; Lafrance & Goff, 1985). *In vitro*, heat stress perturbed the physiological difference between responses of pregnant and cyclic endometrial tissues to oxytocin. Oxytocin-induced secretion rates of PGF for pregnant endometrium exposed to heat stress were similar to those by cyclic endometrium perfused at thermoneutral temperatures.

A heat stress-induced responsiveness of pregnant endometrium to oxytocin may be due in part to increased availability of oxytocin receptors because of alterations in membrane fluidity, or to activation of intracellular signal transduction associated with oxytocin binding to its receptor on the endometrial cell membrane. In particular, oxytocin has been reported to stimulate endometrial prostaglandin synthesis by increasing the hydrolysis of membrane-bound phosphoinositides to diacylglycerol and inositol phosphates with subsequent release of arachidonic acid from diacylglycerol (Flint *et al.*, 1986). Similarly, heat stress has been reported to stimulate membrane phospholipase activity and phosphoinositide turnover (Calderwood *et al.*, 1987) as well as activating cAMP production (Calderwood *et al.*, 1985). Hence, heat stress may potentiate oxytocin-induced prostaglandin synthesis by activating oxytocin receptor-induced prostaglandin synthesis by activating oxytocin receptor-mediated second messengers such as inositol phosphates and diacylglycerol.

Alternatively, heat stress may potentiate oxytocin-induced synthesis of endometrial PGF-2 α by altering factors which regulate prostaglandin synthesis during early pregnancy. One possibility is that heat stress alters activity of an endogenous intracellular endometrial inhibitor of prostaglandin synthesis, the activity of which is normally increased during early pregnancy (Basu & Kindahl, 1987; Gross *et al.*, 1988a). A heat stress-induced increase in PGF-2 α secretion of pregnant endometrium in response to oxytocin may be a consequence of a decrease in the activity of the prostaglandin inhibitor. Regardless of the mechanism, the sequence of events leading to luteolysis and termination of pregnancy may be initiated by heat stress.

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