Conceptus, Progesterone, and Breed Effects on Uterine Protein Secretion in Swine[1,2]

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ABSTRACT: This experiment consisted of the following treatment-breed groups: 1) White crossbred gilts, 2) White crossbred gilts treated with progesterone (200 mg/d in corn oil given on d 2 and 3 after estrus), and 3) Chinese Meishan gilts. Pregnant and nonpregnant gilts (n = 3 to 6) from each treatment-breed combination were assigned to be slaughtered on d 10, 11, 12, 13, and 15. At slaughter each uterine horn was flushed with 20 mL of minimal essential medium. Uterine flushings were assayed for total protein, acid phosphatase, uteroferrin, retinol-binding protein, and oxytocin. Uterine flush total protein was increased by progesterone treatment, was unaffected by pregnancy status, and was less in Meishans. Similar patterns were found for retinol binding protein and uteroferrin, except that uteroferrin was greater in pregnant than in nonpregnant gilts. Oxytocin was greater in pregnant than in nonpregnant gilts, was not influenced by progesterone treatment, and was similar in Meishan and in White crossbred gilts. These results indicate that the conceptus does not influence secretion of either total protein or retinol binding protein during pregnancy and that the onset of secretion of these uterine proteins may be controlled by progesterone. The presence of the conceptus is associated with increased uteroferrin and oxytocin production. The decreased secretion of uterine proteins in Meishan gilts may partially explain the slower embryonic development that has been reported for this breed.

Key Words: Meishan, Uteroferrin, Retinol Binding Protein

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Introduction

Proteins, such as uteroferrin and retinol-binding protein (**RBP**), secreted by the uterus in swine constitute one mechanism by which vitamins and minerals are transported to the developing conceptus (Roberts and Bazer, 1988). Dramatic increases in the content of both proteins occur in pregnant and nonpregnant gilts between d 10 and 13 (Vallet et al., 1996). Lack of a difference between pregnant and nonpregnant gilts suggests that uterine protein secretion is primarily controlled by mechanism(s) other than conceptus estrogen. An alternative mechanism may be the duration that the uterus is exposed to progesterone (Vallet et al., 1996). If this hypothesis is true, increased uterine protein secretion should occur earlier in gilts treated with progesterone at 48 and 72 h after estrus. However, it is unclear from previous results whether the conceptus has an influence on secretion of uterine proteins on d 11 or 12 of pregnancy.

Litter size at farrowing is greater in Chinese Meishan pigs and is due in part to greater ovulation rate, greater embryonal survival, and greater uterine capacity (Ashworth et al., 1990; Wilmut et al., 1992; Christenson, 1993; Ford and Youngs, 1993; Galvin et al., 1993). Meishan gilts may achieve decreased embryonal loss and possibly increased uterine capacity via a decrease in the developmental rate of the conceptuses (Anderson et al., 1993; Ford and Youngs, 1993). The mechanism by which the Meishan uterus limits conceptus development is not known. One hypothesis is that some rate-limiting nutrients that are provided by the uterus are secreted at a slower rate in Meishans than in European breeds. Protein in the uterine flushings of Meishans was less than that of European breeds (Bazer et al., 1991; Ford and Youngs, 1993). A further comparison of uterine

[1]Names are necessary to report factually on available data; however, the USDA neither guarantees nor warrants the standard of the product, and the use of the same by USDA implies no approval of the product to the exclusion of others that may also be suitable.

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protein secretion in Meishan vs White crossbred pigs would be useful to more fully evaluate the possibility that uterine protein secretion may limit conceptus development.

Materials and Methods

The animal handling methods used were reviewed and approved by our institutional animal care committee. Adult Chinese Meishan and composite White crossbred (¼ Large White, ¼ Landrace, ¼ York, ¼ Chester White) gilts were observed for estrous behavior twice daily (0800 and 1600) and were assigned to be mated or remain nonpregnant after at least one estrous cycle of normal duration (17 to 23 d). One-half of the White crossbred gilts within each status received progesterone (200 mg/d in 4 mL of 90% corn oil/10% ethanol; Sigma Chemical Co., St. Louis, MO) 48 and 72 h after estrus was detected. This dose is similar to the dose required to maximize uterine protein secretion in ovariectomized gilts (Knight et al., 1974), and this treatment is unlikely to interfere with corpus luteum development (Sammelwitz et al., 1961). Mated gilts that did not become pregnant were dropped from the experiment. White crossbred, White crossbred progesterone-treated, and Meishan gilts (n = 3 to 6) of each status were slaughtered on d 10, 11, 12, 13, and 15 (for scheduling purposes, a gilt was considered d 0 if it was first detected in estrus in the morning or the previous afternoon). At slaughter, the uterus from each gilt was recovered within 20 min, and each uterine horn was flushed with 20 mL of minimum essential medium (**MEM**) containing one-tenth the normal amount of leucine. The uterine horns were then opened, and the endometrium was dissected from the myometrium. One endometrial sample per gilt was placed into 20 mL of MEM on ice for subsequent explant culture. Additional samples (~1 g) were frozen in liquid nitrogen for isolation of RNA. Endometrial tissue was cultured using conditions described previously (Vallet and Christenson, 1993). Uterine flushings were measured for total protein using the method of Lowry et al. (1951) with BSA as the standard. Acid phosphatase was measured in uterine flushings and endometrial culture medium as previously described (Vallet and Christensen, 1994). Endometrial culture medium was dialyzed against three changes of 10 m*M* Tris, .02% sodium azide, and pH 8.2, and an aliquot of the dialyzed medium was subjected to scintillation counting to determine the amount of nondialyzable radioactivity in each culture.

Retinol-Binding Protein, Uteroferrin, Oxytocin, and Estrogen Radioimmunoassays

Retinol-binding protein in uterine flushings and endometrial culture medium was measured using a validated assay as previously described (Vallet, 1994). Inter- and intraassay CV were 16.2 and 15.1%, respectively. Uteroferrin was measured using a validated assay as previously described (Cardenas et al., 1997). The intra- and interassay CV were 1.3 and 8.9%, respectively. Oxytocin was measured as previously described (Lutz et al., 1991); intra- and interassay CV were 8.5 and 13.2%. Finally, estradiol in uterine flushings was measured as previously described (Rozell and Keisler, 1990) in a single assay in which the intraassay CV was 10.2%.

Northern and Slot Blot Analysis

Total RNA was extracted from endometrial samples using the method of Chomczynski and Sacchi (1987). For Northern blotting, 10 μg of total RNA was electrophoresed on a 1.5% agarose-MOPS gel and transferred to nylon filters (Hybond; Amersham, Arlington Heights, IL). For slot blots, 10 or 30 μg of total RNA were loaded onto nylon filters using a manifold. Blots were then ultraviolet-crosslinked. For generation of probes and standards, a 191-bp fragment of RBP cDNA (base 251–442, Trout et al., 1991) and a 240-bp fragment of UF cDNA (base 328–567, Simmen et al., 1989) were subcloned into pBS2KS and PCRII vectors, respectively. Sense RNA was generated using SP6 (RBP) and T3 (UF) RNA polymerases. Standard curves of sense RNA were then constructed for RBP (2.4 to 10,000 pg) and UF (39 to 10,000 pg). Standards were included on each filter. The RBP and UF cDNA probes were generated by PCR in the presence of $\alpha[^{32}P]$-dCTP (300 Ci/mmol; Amersham) and unlabeled dGTP, dATP, and dTTP. Blots were prehybridized in Denhardt's solution (Sambrook et al., 1989) and then hybridized with 2×10^6 cpm radiolabeled cDNA in 10 mL of Denhardt's solution. Blots were then washed (2× SSC, .1% SDS, 42°C then .5× SSC, .1% SDS, 55°C; SSC = .15 *M* NaCl, .015 *M* Na citrate, pH 7) and exposed to x-ray film. Autoradiograms were then quantified using laser densitometry.

Statistical Analysis

The number of hours from estrus to slaughter was calculated for each gilt in the experiment and then all data were analyzed by homogeneity of regression using the number of hours from estrus to slaughter as the dependent variable. The model included the effect of treatment-breed (White cross untreated, White cross progesterone-treated, and Meishan), status (pregnant or nonpregnant), the status × treatment-breed interaction, the linear and quadratic effects of hours from estrus to slaughter, and the interaction of treatment-breed, status, and treatment-breed × status with the linear and quadratic effects of hours from estrus to slaughter. All data except endometrial culture nondialyzable radioactivity and endometrial culture acid phosphatase were log transformed before

analysis because the variance of the observations increased with time and as the mean of each variable increased (scale effect).

Results

Analysis indicated that for total protein in the uterine flushings (Table 1), there was no difference between pregnant and nonpregnant gilts in the pattern of change in uterine protein content with time nor was there a difference between pregnant and nonpregnant gilts in the overall mean uterine protein content. There was also no difference between the patterns of change with time between treatments or breeds. However, there was an increase in the overall mean protein content ($P < .05$) for White crossbred progesterone-treated gilts compared with White crossbred untreated gilts (Table 2). Furthermore, the overall mean amount of protein in the uterine flushing was less ($P < .01$) for Meishan than for White crossbred untreated gilts.

There was no difference between pregnant and nonpregnant gilts in the pattern of change in intrauterine retinol-binding protein content (Table 3) with time examined, nor was there a difference between pregnant and nonpregnant gilts in overall mean uterine RBP. There was no difference in the pattern of change with time between White crossbred progesterone-treated and White crossbred untreated gilts. However, overall mean uterine RBP differed between White crossbred progesterone-treated and White crossbred untreated gilts ($P < .01$; Table 2). Examination of the results indicated that this difference was due primarily to increased RBP from 240 to 268 h (d 10 and 11) in progesterone-treated gilts compared with untreated gilts. We also note a difference between White crossbred untreated gilts and Meishan gilts in the pattern of change with time ($P < .05$) and in overall mean uterine RBP ($P < .01$). Examination of the data (Figure 1) indicated that this difference was due to less RBP in the uterine flushings of Meishan gilts than in White crossbred gilts.

For uterine flush total acid phosphatase (Table 4), there was no difference between pregnant and nonpregnant gilts in the pattern of change with time or overall mean total acid phosphatase. There was no difference in the pattern of change with time between White crossbred progesterone-treated and untreated gilts. However, there was an overall difference ($P < .01$) in acid phosphatase between White crossbred progesterone-treated and White crossbred untreated gilts. This difference was due to increased acid phosphatase primarily from 240 to 268 h (d 10 and 11) in progesterone-treated gilts compared with untreated gilts. Additionally, analysis indicated a difference in the pattern of change with time between breeds ($P < .01$; Figure 1) and an overall difference ($P < .01$). These differences were due primarily to lower acid phosphatase in Meishan gilts than in White crossbred untreated gilts from 240 to 291 h (d 10 to d 12). Acid phosphatase and uteroferrin contents were highly correlated ($r = .98$, $P < .01$).

Results of the analysis of the uteroferrin RIA data (Table 5) differed slightly from that of acid phosphatase (Table 4). We observed a difference in the pattern of change with time between pregnant and nonpregnant gilts ($P < .01$; Figure 2). This difference was due to increased uteroferrin in pregnant gilts from 256 to 291 h (d 11 and d 12). We found no

Table 1. Least squares means and number of observations (parentheses) for uterine flush total protein (mg) along with the range within one SEM using pooled variance from ANOVA obtained with log transformed data

	Hours from estrus to slaughter					
Treatment group	240–244	256–268	280–291	304–316	328–331	360–364
White crossbred, nonpregnant[a]	10.5 (3) 8.0–13.7	20.8 (4) 16.5–26.2	35.8 (5) 29.1–44.1	55.0 (5) 44.7–67.7	69.7 (1) 43.8–110.9	107.3 (4) 85.1–135.3
White crossbred, pregnant	12.1 (3) 9.3–15.8	16.4 (4) 13.0–20.7	44.1 (6) 36.5–53.3	48.1 (4) 38.1–60.7	42.4 (2) 30.5–58.9	116.3 (3) 89.0–151.9
White crossbred, progesterone-treated, nonpregnant	16.3 (3) 12.5–21.3	16.3 (5) 13.3–20.0	29.6 (3) 22.7–38.7	79.3 (4) 62.9–100.0	141.0 (1) 88.7–224.2	101.5 (3) 77.7–132.6
White crossbred, progesterone-treated, pregnant	19.6 (3) 15.0–25.6	27.9 (5) 22.7–34.4	47.6 (5) 38.7–58.6	86.7 (4) 68.7–109.3	75.9 (1) 47.7–120.7	118.4 (3) 90.7–154.6
Meishan, nonpregnant	12.1 (2) 8.7–16.8	17.3 (4) 13.7–21.8	28.3 (3) 21.7–37.0	19.7 (3) 15.1–25.7	68.2 (3) 52.2–89.1	102.7 (3) 78.6–134.1
Meishan, pregnant	11.5 (2) 8.3–16.0	10.2 (5) 8.3–13.3	18.3 (5) 14.9–22.5	23.4 (6) 19.4–28.3	24.1 (2) 17.4–33.5	65.4 (3) 54.1–85.4

[a]Analysis indicated an overall effect of progesterone treatment ($P < .05$) and breed ($P < .01$).

Table 2. Overall means along with the range within one SEM and number of observation (parentheses) for nonpregnant (NP), pregnant (P), control (C) White crossbred untreated (WC-C), progesterone-treated (WC-P4), and Meishan (ME) untreated gilts

Item	NP	P	WC-C	WC-P4	ME
Total protein, mg[ad]	46.5 (59) 45.2–47.9	43.5 (66) 42.3–44.7	46.1 (44) 44.6–47.6	57.0 (40) 55.0–59.0	31.8 (41) 30.7–32.9
Total retinol-binding protein, mg[bd]	5.4 (59) 4.6–6.3	5.2 (67) 4.5–6.0	5.7 (45) 4.8–6.8	7.5 (40) 6.2–9.1	2.5 (41) 2.1–3.0
Total acid phosphatase, μmol P_i/min[bd]	.40 (59) .35–.46	.47 (67) .41–.53	.44 (45) .38–.51	.61 (40) .52–.72	.25 (41) .21–.29
Total uteroferrin, mg[bd]	2.2 (59) 1.9–2.5	2.1 (65) 1.8–2.4	2.1 (44) 1.8–2.5	3.3 (39) 2.8–3.9	1.1 (41) .9–1.3
Total oxytocin, ng[f]	.3 (59) .1–.8	1.7 (65) .7–4.1	1.0 (44) .3–2.9	1.7 (39) .5–5.4	.4 (41) .1–1.2
Total estrogen, ng[c]	—	—	4.4 (20) 3.2–6.0	3.8 (20) 2.8–5.2	2.4 (23) 1.8–3.2
Endometrial nondialyzable radioactivity, dpm $\times 10^{-6}$	6.8 (59) ± .03	6.4 (66) ± .03	6.4 (44) ± .03	6.7 (41) ± .03	6.7 (40) ± .03
Endometrial acid phosphatase, μmol $P_i \cdot min^{-1} \cdot g^{-1}$ tissue	1.4 (59) ± .03	1.3 (67) ± .03	1.46 (44) ± .04	1.80 (41) ± .04	.70 (41) ± .04
Endometrial retinol-binding protein, μ/g tissue[d]	32.6 (59) 31.4–33.8	30.0 (66) 29.0–31.0	34.8 (43) 33.4–36.3	39.4 (41) 37.7–41.1	19.5 (41) 18.7–20.4
Retinol-binding protein mRNA, ng/μg tcRNA[bdf]	.56 (58) .46–.68	.82 (65) .68–1.0	.74 (44) .59–.93	.96 (40) .76–1.22	.37 (39) .29–.47
Uteroferrin mRNA, ng/μg tcRNA[bd]	.111 (58) .108–.114	.113 (65) .110–.116	.109 (44) .106–.113	.137 (40) .133–.142	.091 (39) .088–.094

[a]White cross untreated vs progesterone-treated differed ($P < .05$).
[b]White cross untreated vs progesterone-treated differed ($P < .01$).
[c]Meishan vs White crossbred differed ($P < .05$).
[d]Meishan vs White crossbred differed ($P < .01$).
[e]Nonpregnant vs pregnant differed ($P < .05$).
[f]Nonpregnant vs pregnant differed ($P < .01$).

Table 3. Least squares means and number of observations (parentheses) for uterine flush total retinol-binding protein (mg) along with the range within one SEM using the pooled variance from ANOVA obtained with log transformed data

	Hours from estrus to slaughter					
Treatment group	240–244	256–268	280–291	304–316	328–331	360–364
White crossbred, nonpregnant[a]	.0089 (3) .0047–.017	.30 (4) .17–.52	3.4 (5) 2.1–5.5	9.2 (5) 5.7–15.0	5.9 (1) 2.0–17.5	18.0 (4) 10.4–31.0
White crossbred, pregnant	.030 (3) .016–.056	.16 (4) .093–.28	4.5 (6) 2.9–7.0	7.0 (4) 4.1–12.1	9.3 (2) 4.3–20.1	13.4 (4) 8.6–23.1
White crossbred, progesterone-treated, nonpregnant	.089 (3) .047–.17	.28 (5) .17–.46	2.8 (3) 1.5–5.2	13.1 (4) 7.6–22.6	14.7 (1) 5.0–43.6	17.2 (3) 9.2–32.2
White crossbred, progesterone-treated, pregnant	.19 (3) .10–.36	1.9 (5) 1.2–3.1	4.1 (5) 2.5–6.7	16.2 (4) 9.4–27.9	7.1 (1) 2.4–21.1	16.4 (3) 8.8–30.7
Meishan, nonpregnant	.014 (2) .0065–.03	.025 (4) .015–.043	.19 (3) .10–.36	1.2 (3) .64–2.2	6.5 (3) 3.5–12.2	9.4 (3) 5.0–17.6
Meishan, pregnant	.007 (2) .003–.015	.0088 (5) .0054–.014	1.1 (5) .68–1.8	2.8 (6) 1.8–4.4	5.5 (2) 2.5–11.9	7.1 (3) 3.8–13.3

[a]The pattern of change with time differed between White crossbred and Meishan groups ($P < .05$). Overall differences between White crossbred progesterone-treated and untreated groups ($P < .01$) and between White crossbred untreated and Meishan groups ($P < .01$) were observed.

difference in the pattern of change with time between White crossbred progesterone-treated and untreated groups, but we detected an overall difference ($P < .01$) in uteroferrin. As for acid phosphatase, this was due to greater uteroferrin primarily from 240 to 268 h (d 10 and d 11) in progesterone-treated gilts. Finally, a difference in the pattern of change with time was observed between breeds ($P < .05$) and overall ($P < .01$). This difference was due to decreased uteroferrin in Meishan gilts (Figure 2). Taken together, these data indicate that progesterone treatment accelerates uteroferrin secretion and that uteroferrin secretion in Meishan gilts is less than in White crossbred gilts. The conceptus may or may not influence uteroferrin secretion.

No difference in the pattern of change with time in uterine flush estrogen (Table 6) was detected between White crossbred progesterone-treated and untreated gilts. Also, overall mean estrogen did not differ between progesterone-treated and untreated gilts. However, estrogen was considerably greater from 256 to 268 h (d 11) of pregnancy ($P < .05$) in progesterone-treated gilts than in untreated gilts, which suggests that the progesterone treatment accelerated

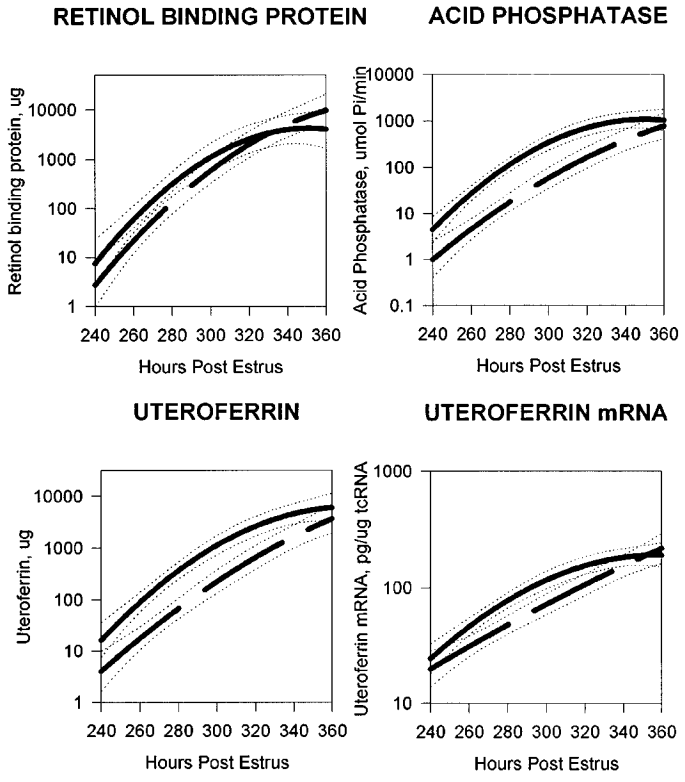

Figure 1. Regression lines for White crossbred untreated (solid lines) and Meishan (broken lines) gilts along with 95% confidence intervals are illustrated for uterine flush retinol-binding protein ($P < .05$), acid phosphatase ($P < .01$), uteroferrin ($P < .05$), and endometrial uteroferrin mRNA ($P < .05$). Note the log scale.

conceptus estrogen secretion. No difference in the least squares means for estrogen from 240 to 244 h (d 10) was detected. This suggests that in progesterone-treated gilts, increased uterine protein secretion preceded increased conceptus estrogen secretion. Our analysis indicated that no difference in the pattern of change with time of intrauterine estrogen content occurred between breeds; however, overall mean uterine estrogen content differed between breeds ($P < .05$; Table 2). Examination of the data indicated that this was due to less estrogen secretion in Meishan gilts.

Overall mean uterine oxytocin did not differ between White crossbred progesterone-treated and untreated gilts or between breeds (Table 7), nor did the pattern of change in intrauterine oxytocin content over time differ between White crossbred progesterone-treated and untreated gilts or between breeds. The pattern of change with time in intrauterine content of oxytocin differed between pregnant and nonpregnant gilts ($P = .05$; Figure 2), and examination of the data indicated that this was due to greater oxytocin content in pregnant gilts than in nonpregnant gilts from 256 to 291 h (d 11 and 12). Substantial oxytocin was observed in several of the nonpregnant gilts, and the intrauterine content of oxytocin was highly variable (root mean square error was 2.6 natural logs).

The pattern of change with time ($P = .05$; Figure 2) for endometrial culture nondialyzable radioactivity (Table 8) differed between pregnant and nonpregnant gilts. Data indicated that this was due to greater nondialyzable radioactivity secreted by the endometrium from nonpregnant gilts primarily from 240 to 268 h. There was no difference in the pattern of change with time between White crossbred progesterone-treated and untreated gilts or between breed groups, nor did the overall mean nondialyzable radioactivity differ between treatment or breed groups.

There were no differences in the pattern of change with time between status (pregnant vs nonpregnant), progesterone treatment (White crossbred progesterone-treated vs untreated), or breed (Meishan vs White crossbred) groups for retinol-binding protein secreted into culture (Table 9). Furthermore, overall mean RBP in culture did not differ between status or progesterone treatment groups. However, overall mean RBP in endometrial culture differed between breeds ($P < .01$; Table 2). Data indicated that this was due to less RBP secretion by the endometrium from Meishan gilts than from White crossbred untreated gilts.

We observed no differences in the pattern of change with time between status, treatment, or breed groups for acid phosphatase secreted into culture (Table 10). Likewise, overall mean acid phosphatase did not differ with pregnancy status. However, overall mean acid phosphatase differed between White crossbred

Table 4. Least squares means and number of observations (parentheses) for uterine flush total acid phosphatase (μmole P_i/min) along with the range within one SEM using pooled variance from ANOVA obtained with log transformed data

	Hours from estrus to slaughter					
Treatment group	240–244	256–268	280–291	304–316	328–331	360–364
White crossbred, nonpregnant[a]	.0031 (3) .0018–.0053	.066 (4) .041–.11	.24 (5) .16–.37	.43 (5) .28–.66	.48 (1) .19–1.2	1.6 (4) 1.0–2.6
White crossbred, pregnant	.0066 (3) .0038–.011	.027 (4) .017–.043	.65 (6) .44–.96	.51 (4) .32–.82	.43 (2) .22–.84	.85 (3) .53–1.4
White crossbred, progesterone-treated, nonpregnant	.031 (3) .018–.053	.033 (5) .022–.050	.34 (3) .20–.59	.60 (4) .37–.96	1.18 (1) .46–3.0	1.2 (3) .70–2.1
White crossbred, progesterone-treated, pregnant	.061 (3) .035–.11	.19 (5) .12–.29	.58 (5) .38–.88	1.9 (4) 1.2–3.0	.98 (1) .38–2.5	.88 (3) .51–1.5
Meishan, nonpregnant	.0038 (2) .0020–.0074	.0085 (4) .0053–.014	.025 (3) .015–.043	.067 (3) .039–.12	.78 (3) .45–1.3	1.0 (3) .58–1.72
Meishan, pregnant	.0019 (2) .00098–.0037	.0014 (5) .00092–.0021	.079 (5) .052–.12	.18 (6) .12–.26	.38 (2) .20–.74	.69 (3) .40–1.2

[a]The pattern of change with time differed between White crossbred untreated and Meishan groups ($P < .01$). Overall differences between White crossbred progesterone-treated and untreated ($P < .01$) and between White crossbred untreated and Meishan groups ($P < .01$) were observed.

progesterone-treated and untreated gilts ($P < .01$) and between breeds ($P < .01$; Table 2). Data indicated that the differences were due to greater acid phosphatase in cultures from progesterone-treated gilts than from untreated gilts and less acid phosphatase in cultures from Meishan gilts than from White crossbred gilts.

A representative Northern blot for RBP and uteroferrin mRNA is shown in Figure 3. Analysis indicated no effect of breed or pregnancy status on the pattern of change with time for endometrial RBP mRNA concentrations (Table 11). However, the pattern of change (Figure 4) differed between White crossbred progesterone-treated and untreated gilts ($P < .05$). Overall mean endometrial mRNA concentrations for RBP were greater ($P < .01$; Table 2) in pregnant than in nonpregnant gilts. Overall mean RBP mRNA also differed ($P < .01$) between White

Table 5. Least squares means and number of observations (parentheses) for uterine flush total uteroferrin (mg) along with the range within one SEM using pooled variance from ANOVA obtained with log transformed data

	Hours from estrus to slaughter					
Treatment group	240–244	256–268	280–291	304–316	328–331	360–364
White crossbred, nonpregnant[a]	.016 (3) .0092–.028	.21 (4) .13–.34	.77 (5) .50–1.2	.82 (5) .53–1.3	.91 (1) .35–2.4	9.8 (4) 1.8–15.8
White crossbred, pregnant	.025 (2) .013–.049	.082 (4) .051–.13	2.7 (6) 1.8–4.0	3.3 (4) 2.0–5.3	1.6 (2) .81–3.1	5.2 (3): 3.0–9.0
White crossbred, progesterone-treated, nonpregnant	.15 (3) .086–.26	.15 (5) .098–.23	.57 (3) .33–.99	5.6 (4) 3.5–9.0	3.8 (1) 1.5–9.9	10.3 (3) 5.9–17.9
White crossbred, progesterone-treated, pregnant	.16 (3) .092–.28	.84 (5) .55–1.3	2.6 (5) 1.7–4.0	7.4 (4) 4.6–11.9	6.1 (1) 3.8–15.9	3.6 (2) 1.8–7.1
Meishan, nonpregnant	.011 (2) .0056–.022	.033 (4) .020–.053	.068 (3) .040–.12	.33 (3) .19–.57	3.0 (3) 1.7–5.2	6.1 (3) 3.5–10.6
Meishan, pregnant	.0073 (2) .0037–.014	.0074 (5) .0048–.011	.29 (5) .19–.44	.73 (6) .49–1.08	1.6 (2) .81–3.1	2.7 (3) 1.6–4.7

[a]The pattern of change with time differed between pregnant and nonpregnant groups ($P < .01$) and between White crossbred untreated and Meishan groups ($P < .05$). Overall differences between White crossbred progesterone-treated and untreated ($P < .01$) and between White crossbred untreated and Meishan groups were also observed ($P < .01$).

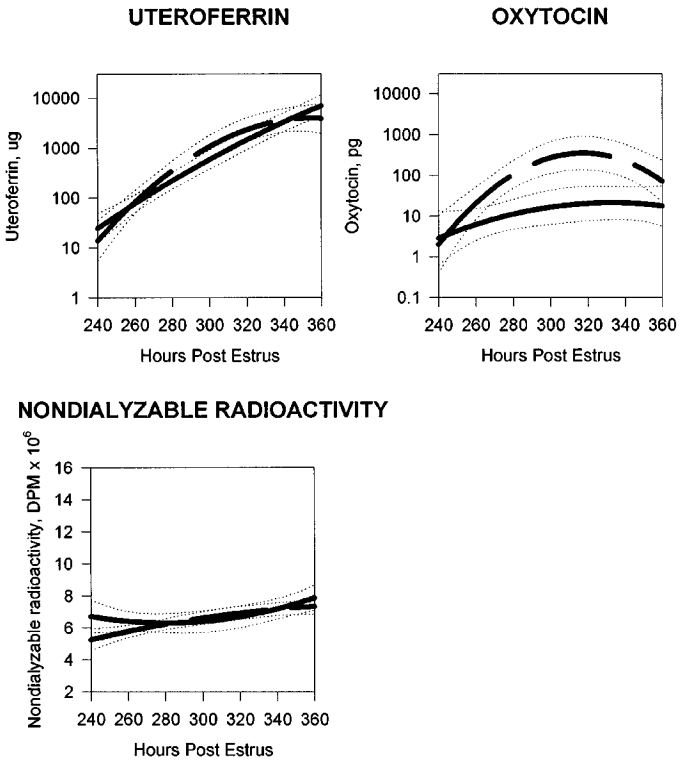

Figure 2. Regression lines for nonpregnant (solid line) and pregnant (broken line) gilts along with 95% confidence intervals are illustrated for uterine flush uteroferrin ($P < .01$) and oxytocin ($P < .05$) and for endometrial culture nondialyzable radioactivity ($P < .05$). Note the log scale.

crossbred progesterone-treated and untreated gilts and between breeds ($P < .01$; Table 2). These differences were due to greater RBP mRNA in White cross progesterone-treated gilts and less RBP mRNA concentrations in the endometrium from Meishan gilts than from White crossbred untreated gilts (Table 11).

Analysis indicated no effect of status or progesterone treatment on the pattern of change with time in endometrial uteroferrin mRNA (Table 12). The pattern of change with time and overall mean for uteroferrin mRNA differed between breeds ($P < .05$, .01, respectively; Figure 2), and this was primarily due to lower uteroferrin mRNA concentrations from 256 to 316 h (d 11 to d 13) in Meishan than in White crossbred pigs. Overall means for White crossbred progesterone-treated and untreated gilts differed ($P < .01$), primarily as a result of greater uteroferrin mRNA concentrations in progesterone-treated gilts from 240 h to 291 h. No difference in overall uteroferrin mRNA concentrations between pregnant and nonpregnant gilts was detected.

Discussion

These data indicate the relative importance of progesterone timing and of the conceptus on the secretion of proteins by the swine uterus during pregnancy. Neither total intrauterine protein content nor total intrauterine RBP content were influenced by the presence of the conceptus, despite the prevailing hypothesis that uterine protein secretion is influenced by conceptus estrogen secretion. Instead, these data suggest that the onset of secretion of total protein and RBP is controlled by progesterone timing, because administration of progesterone on d 2 and 3 accelerated the changes in uterine secretion of these proteins. Uteroferrin secretion increases dramatically from d 10 to 15 in pregnant and nonpregnant gilts, and early progesterone treatment accelerates this increase. This suggests that progesterone timing is also likely to be the dominant influence over uteroferrin secretion. Uteroferrin may also be influenced by the presence of the conceptus because secretion of uteroferrin measured by RIA differed between pregnant and nonpregnant gilts. Oxytocin, by contrast, seems to be influenced only by the conceptus and not by progesterone timing. Finally, these data suggest that secretion of estrogen by the conceptus is also acceler-

Table 6. Least squares means and number of observations (parentheses) for uterine flush estrogen (ng) along with the range within one SEM using pooled variance from ANOVA obtained with log transformed data

	Hours from estrus to slaughter					
Treatment group	240–244	256–268	280–291	304–316	328–331	360–364
White crossbred, pregnant[a]	.090 (2) .041–.20	.29 (4) .17–.50	8.8 (6) 5.6–13.8	8.1 (3) 4.3–15.3	3.3 (2) 1.5–7.2	1.7 (3) .90–3.2
White crossbred, progesterone-treated, pregnant	.14 (3) .074–.26	3.3 (5) 2.0–5.4	8.0 (4) 4.6–13.9	2.1 (4) 1.2–3.6	3.4 (1) 1.1–10.2	2.5 (2) 1.2–5.4
Meishan, pregnant	.14 (2) .064–.30	.12 (5) .073–.20	2.8 (5) 1.7–4.6	5.1 (6) 3.3–8.0	3.2 (2) 1.5–7.0	1.8 (3) .95–3.4

[a]Estrogen differed between White crossbred progesterone-treated and untreated gilts on d 11. An overall effect of breed ($P < .05$) was also detected.

Table 7. Least squares means and number of observations (parentheses) for total uterine oxytocin (pg) along with the range within one SEM using the pooled variance from ANOVA obtained with log transformed data

	Hours from estrus to slaughter					
Treatment group	240–244	256–268	280–291	304–316	328–331	360–364
White crossbred, nonpregnant[a]	26.7 (3) 5.9–119.8	60.5 (4) 16.5–222.0	38.2 (5) 12.0–121.9	1,472.3 (5) 462–4697	4.1 (1) .3–55.2	1,646.8 (4) 448.8–6,043.6
White crossbred, pregnant	5.4 (2) .9–34.0	46.2 (4) 12.6–169.5	1,188.8 (6) 412–3,431	4,424.8 (4) 1,206–16,236	3,219.4 (2) 511.5–20,271	105.4 (3) 23.5–473
White crossbred, progesterone-treated, nonpregnant	8.8 (3) 2.0–39.4	18.8 (5) 5.99–60.0	305.6 (3) 68.2–1,369.6	114.4 (4) 31.2–419.8	1,992.8 (1) 148–26,831	200.7 (3) 44.8–899.5
White crossbred, progesterone-treated, pregnant	16.2 (3) 3.6–72.6	562.7 (5) 176–1,795	12,395.4 (5) 3,885.7–39,540	58.6 (4) 16.0–215.0	11.0 (1) .8–148.1	407.9 (2) 64.8–2,568.4
Meishan, nonpregnant	2.8 (2) .44–17.6	11.9 (4) 3.2–43.7	13.6 (3) 3.0–61.0	94.1 (3) 21.0–421.7	64.5 (3) 14.4–289.1	28.5 (3) 6.4–127.7
Meishan, pregnant	3.3 (2) .52–20.8	4.2 (5) 1.32–13.4	1,498.9 (5) 470.0–4,781.4	1,574.6 (6) 545.5–4,544.9	443.1 (2) 70.4–2,790.0	108.3 (3) 24.2–485.4

[a]An effect of pregnancy status on the pattern of change with time was present ($P < .05$) along with an overall effect of status ($P < .01$).

ated by early progesterone treatment, and the increase in uterine protein secretion precedes the change in conceptus estrogen secretion. This result suggests the interesting possibility that uterine protein secretion may control conceptus estrogen secretion. Relevant to this concept, data indicate that Meishan gilts undergo a more gradual increase in both uterine protein secretion and estrogen secretion. Thus, a more gradual increase in uterine protein secretion may decrease the negative impact that faster developing blastocysts may have on their slower developing littermate blastocysts by limiting the ability of the developing conceptus to secrete estrogen.

Of the four previous studies comparing pregnant and nonpregnant gilts available (Geisert et al., 1982; Zavy et al., 1984; Bazer et al., 1991; Vallet et al., 1996), only one study (Bazer et al., 1991) indicates greater intrauterine protein content in pregnant than in nonpregnant gilts. In the current study, twice-daily estrus detection and regression analysis using the number of hours from estrus to slaughter were used to decrease the variability associated with inaccuracies in these two events and to improve our ability at detecting differences. Gilts observed only once a day for estrus vary as much as 24 h in the actual timing of the onset of estrus. Our data indicate that, between d

Table 8. Number of observations (parentheses) and least squares means ± SEM from ANOVA for endometrial culture nondialyzable radioactivity (dpm × 10^{-6}/g tissue)

	Hours from estrus to slaughter					
Treatment group	240–244	256–268	280–291	304–316	328–331	360–364
White crossbred, nonpregnant[a]	6.4 ± .8 (3)	5.6 ± .7 (4)	6.4 ± .7 (4)	7.0 ± .7 (5)	6.9 ± 1.5 (1)	8.5 ± .7 (4)
White crossbred, pregnant	5.0 ± .8 (3)	5.2 ± .7 (4)	6.6 ± .6 (6)	7.0 ± .7 (4)	5.1 ± 1.0 (2)	7.2 ± .7 (4)
White crossbred, progesterone-treated, nonpregnant	5.1 ± .8 (3)	7.1 ± .6 (6)	6.2 ± .8 (3)	6.4 ± .7 (4)	7.2 ± 1.5 (1)	7.4 ± .8 (3)
White crossbred, progesterone-treated, pregnant	5.6 ± .8 (3)	5.4 ± .7 (5)	6.9 ± .7 (5)	7.5 ± .7 (4)	5.3 ± 1.5 (1)	8.7 ± .8 (3)
Meishan, nonpregnant	9.3 ± 1.0 (2)	6.2 ± .7 (4)	5.8 ± .8 (3)	6.7 ± .8 (3)	6.4 ± .8 (3)	9.0 ± .8 (3)
Meishan, pregnant	5.6 ± 1.0 (2)	6.3 ± .7 (5)	5.9 ± .7 (5)	6.9 ± .7 (5)	6.6 ± 1.0 (2)	7.1 ± .8 (3)

[a]Pattern of change with time differed between pregnancy status groups ($P < .05$).

Table 9. Least squares means and number of observations (parentheses) for endometrial culture retinol-binding protein (μg/g tissue) along with the range within one SEM using the pooled variance from ANOVA obtained with log transformed data

	Hours from estrus to slaughter					
Treatment group	240–244	256–268	280–291	304–316	328–331	360–364
White crossbred, nonpregnant[a]	19.1 (3) 14.1–25.9	17.9 (4) 13.7–23.3	30.8 (4) 23.6–40.1	55.0 (5) 43.4–69.7	48.2 (1) 28.4–81.9	61.4 (4) 47.1–80.0
White crossbred, pregnant	19.7 (3) 14.5–26.7	21.2 (4) 16.3–27.6	28.8 (6) 23.2–35.7	35.1 (4) 26.9–45.8	43.0 (2) 29.6–62.6	51.5 (3) 37.9–69.9
White crossbred, progesterone-treated, nonpregnant	17.2 (3) 12.7–23.4	21.2 (6) 17.1–26.3	33.4 (3) 24.6–45.4	47.5 (4) 36.4–61.9	81.5 (1) 48.0–138.5	71.4 (3) 52.6–97.0
White crossbred, progesterone-treated, pregnant	16.5 (3) 12.2–22.4	25.6 (5) 20.2–32.4	31.5 (5) 24.9–39.9	52.7 (4) 40.4–68.7	61.4 (1) 36.1–104.3	61.2 (3) 45.1–83.1
Meishan, nonpregnant	12.8 (2) 8.8–18.6	9.0 (4) 6.9–11.7	14.3 (3) 10.5–19.4	22.9 (3) 16.9–31.1	21.6 (3) 15.9–29.3	43.2 (3) 31.8–58.7
Meishan, pregnant	11.1 (2) 7.6–16.2	8.3 (5) 6.5–10.5	18.9 (5) 14.9–24.0	21.5 (6) 17.3–26.7	27.1 (2) 18.6–39.4	36.2 (3) 26.7–49.2

[a]An overall effect of breed ($P < .01$) was detected.

10 to 12 of the estrous cycle or pregnancy, RBP concentrations increase an order of magnitude in each 24-h period. Gilts observed in estrus in the afternoon were clearly more like gilts from the same morning; thus, once-a-day estrus detection probably increases the variability of observations because some animals will be up to 24 h ahead of others when they are assigned to the same day. Even with twice-daily estrus detection, variability among animals was high and was likely due in part to the remaining variability in onset of estrus and in part to variability in the timing of events of the cycle and pregnancy from the onset of estrus onward. Previous reports have attempted to account for this variability by adjusting the data according to changes in conceptus development (i.e., spherical, tubular, and filamentous blastocysts; Geisert et al., 1982). Although this strategy is useful for demonstrating that changes in conceptus development occur coincidentally with changes in uterine protein secretion, no correction is available for nonpregnant gilts, and, once such corrections are made for pregnant gilts, the corrected data are no longer directly comparable to data from nonpregnant gilts.

Our results at first seem to differ from the results of Xie et al. (1990). Those researchers transferred more- or less-advanced blastocysts into the ligated uterine

Table 10. Number of observations (parentheses) and least squares means ± SEM from ANOVA for endometrial culture acid phosphatase (μmol P_i/g tissue)

	Hours from estrus to slaughter					
Treatment group	240–244	256–268	280–291	304–316	328–331	360–364
White crossbred, nonpregnant[a]	1.06 ± .28 (3)	1.27 ± .25 (4)	1.58 ± .25 (4)	1.94 ± .22 (5)	1.49 ± .49 (1)	1.06 ± .25 (4)
White crossbred, pregnant	1.11 ± .28 (3)	1.06 ± .25 (4)	1.72 ± .20 (6)	2.10 ± .25 (4)	1.33 ± .35 (2)	1.35 ± .25 (4)
White crossbred, progesterone-treated, nonpregnant	1.84 ± .28 (3)	1.58 ± .20 (6)	1.98 ± .28 (3)	2.06 ± .25 (4)	2.02 ± .49 (1)	2.17 ± .28 (3)
White crossbred, progesterone-treated, pregnant	.93 ± .28 (3)	2.10 ± .22 (5)	1.81 ± .22 (5)	1.7 ± .25 (4)	2.12 ± .49 (1)	1.51 ± .28 (3)
Meishan, nonpregnant	.68 ± .35 (2)	.46 ± .25 (4)	.76 ± .28 (3)	.84 ± .28 (3)	.84 ± .28 (3)	1.12 ± .28 (3)
Meishan, pregnant	.65 ± .35 (2)	.311 ± .22 (5)	.87 ± .22 (5)	.73 ± .20 (6)	.80 ± .35 (2)	.70 ± .28 (3)

[a]Overall effects of treatment ($P < .01$) and breed ($P < .01$) were detected.

horns of gilts, which allowed a comparison of uterine protein secretion within the same gilt in the presence of more- or less-developed blastocysts. They found twice the amount of acid phosphatase and 55% more protein in the presence of advanced blastocysts than in the presence of slower developing blastocysts, although the difference in protein was only marginally significant ($P < .1$). The authors concluded from these results that the conceptus influences protein secretion. Our results also suggested that uteroferrin is increased twofold to threefold by the presence of the conceptus. Thus, our data agree with the data of Xie et al. (1990). The previous authors did not measure any other proteins. Furthermore, our results indicate that uteroferrin content eventually increases to levels that are similar between pregnant and nonpregnant gilts by d 15 and are greater than 200 times the uteroferrin content on d 10. Because no conceptus is present in nonpregnant gilts, the increase in uteroferrin in nonpregnant gilts dramatically illustrates the relative contributions of the conceptus and of other mechanisms in controlling uteroferrin secretion.

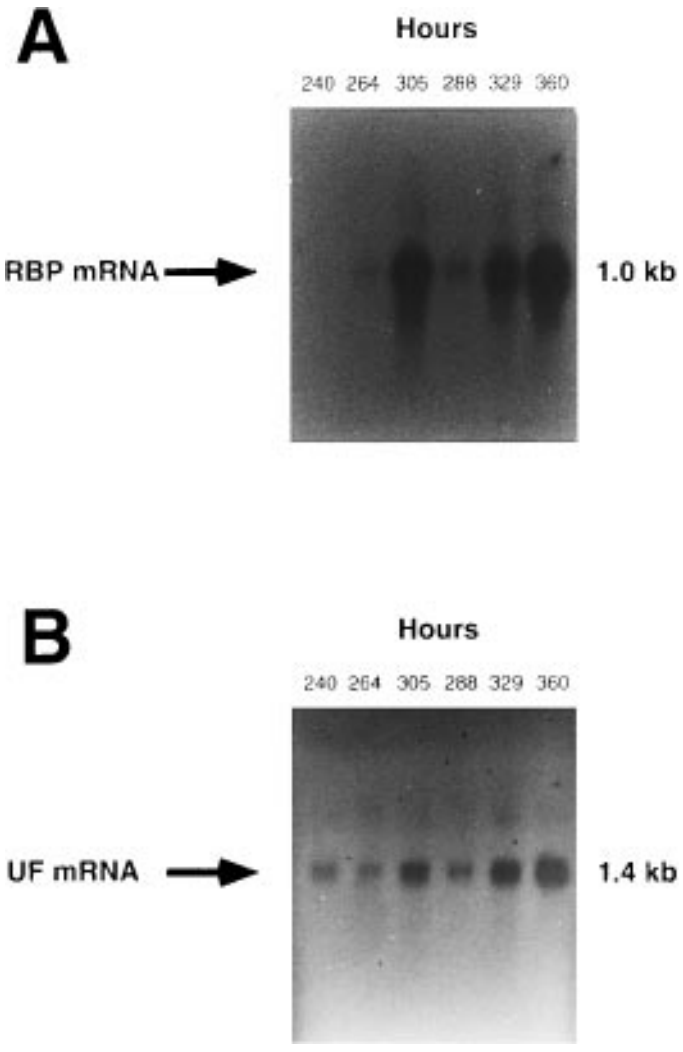

Figure 3. Representative Northern blots for endometrial (A) retinol-binding protein and (B) uteroferrin mRNA are shown for Meishan nonpregnant gilts at various times after estrus.

The mRNA measurements, which provide information on the control of transcription of RBP and uteroferrin, provide information on only one aspect of the overall mechanism that controls secretion of these proteins. The results indicate that progesterone treatment on d 2 and 3 accelerated the increase in mRNA levels for both proteins, and that mRNA levels were less for both proteins in Meishan gilts than in White crossbred gilts. Thus, the changes in intrauterine content of RBP and uteroferrin caused by progesterone treatment and the differences in intrauterine content between breeds may be due at least in part to changes in transcription of these genes. In contrast, the effect of pregnancy status on mRNA levels for each protein was different from the effect of status on the intrauterine content of each protein. Retinol-binding protein mRNA levels were increased in pregnant gilts, but this was not reflected in changes in intrauterine protein content. Uteroferrin mRNA levels were unaffected by pregnancy status even though intrauterine content of uteroferrin may be greater in pregnant gilts on d 11 and 12. One explanation for this discrepancy is that factors other than transcription may have an influence on intrauterine content of each protein. Other factors likely to be important that could also be influenced by the presence of the conceptus, progesterone treatment, or breed include the rate of degradation or translation of each mRNA and the rates of secretion and degradation of each protein within the intrauterine environment. Further studies are needed to determine which factors exercise the most control over the final amount of retinol-binding protein, uteroferrin, and other proteins known to be secreted by the pig uterus.

The influence of progesterone on uterine protein secretion is well established (Chen et al., 1975; Adams et al., 1981; Simmen et al., 1991; Trout et al., 1992). The idea that uterine protein secretion is influenced by progesterone timing is novel and may suggest a role for progesterone and progesterone receptor changes in the onset of protein secretion. Progesterone receptors are reported to disappear from endometrial lumenal and glandular epithelial cells by d 10 of the estrous cycle or pregnancy (Geisert et al., 1994). This roughly corresponds to the time when injected estrogen is capable of stimulating early onset of protein secretion. It is tempting to speculate that early progesterone treatment accelerates the disappearance of progesterone receptors and that the disappearance of progesterone receptors could initiate a cascade of events with the end result of increased uterine protein secretion.

Experiments indicate that conceptus estrogen secretion and uterine protein secretion are strongly associated (Geisert et al., 1982; Trout et al., 1992). Three explanations of this association are possible: 1)

conceptus estrogen stimulates uterine protein secretion, 2) the two phenomena occur coincidentally, possibly controlled by a third factor, but do not influence each other, or 3) uterine protein secretion stimulates conceptus estrogen secretion. The absence of an effect of the conceptus on total protein and retinol-binding protein combined with the increase in uteroferrin that occurs in nonpregnant gilts is strong evidence against the first hypothesis. In support of the second hypothesis, there is currently no evidence for or against a third factor that could control both uterine protein secretion and conceptus estrogen secretion. However, an argument supporting the third hypothesis can be made. Results of this study suggest that early progesterone treatment increased uterine protein secretion and estrogen secretion, and the increase in protein secretion (on d 10) preceded the increase in conceptus estrogen secretion (on d 11). Because these proteins transport vitamins and minerals that are essential for conceptus development, changes in the rate of their delivery might be expected to influence the rate of conceptus development. Also, many growth factors are known components of the uterus during this period of pregnancy (Letcher et al., 1989; Brigstock et al., 1990; Kim et al., 1995), and these growth factors are likely to influence the rate of conceptus development. Furthermore, IGF-I treatment of filamentous porcine blastocysts increased cytochrome P450 aromatase gene expression (Green et al., 1995). Taken together, these data support the hypothesis that increased secretion of proteins by the uterus could control conceptus estrogen secretion via numerous interrelated mechanisms.

The hypothesis that uterine protein secretion influences conceptus estrogen secretion suggests that modification of uterine protein secretion should influence conceptus development and conceptus estrogen secretion. Results of this study and the results of others regarding protein secretion and conceptus development and estrogen secretion in Meishan pigs are consistent with this hypothesis. The conceptus estrogen data of this study indicate that uterine protein secretion and conceptus estrogen secretion are both lower in Meishan pigs, which agrees with previous reports (Anderson et al., 1993; Ford and Youngs, 1993). It has also been reported that Meishan conceptuses develop more slowly (Youngs et al., 1993) and elongate at a decreased number of cells compared with white European breeds (Rivera et al., 1996), which results in smaller embryos and placentae of lighter weight at d 30 of gestation (Christenson, 1993). It has been further hypothesized that this slower development and decreased estrogen secretion are at least partly responsible for the increased fertility of Meishan pigs (Anderson et al., 1993; Ford and Youngs, 1993). Conceptus estrogens are known to alter prostaglandin metabolism associated with maternal recognition of pregnancy as well as alter the flow of ions and other low-molecular-weight substances (Bazer et al., 1984). These estrogen-induced changes are thought to alter the intrauterine environment so that slower developing conceptuses are lost. Uteroferrin itself, which the present study suggests may be modulated by the presence of the conceptus, has been shown to be potentially toxic to developing embryos because of its ability to catalyze lipid peroxidation (Vallet, 1995). Elongation at a smaller number of cells may limit the extent of elongation, thus each conceptus takes up less space within the

Table 11. Least squares means and number of observations (parentheses) for endometrial retinol-binding protein mRNA along with the range within one SEM using the pooled variance from ANOVA obtained with log transformed data

	Hours from estrus to slaughter					
Treatment group	240–244	256–268	280–291	304–316	328–331	360–364
White crossbred, nonpregnant[a]	.017 (3) .0086–.033	.088 (4) .049–.16	.43 (5) .25–.73	.83 (5) .49–1.4	1.2 (1) .37–3.9	1.6 (4) .89–2.9
White crossbred, pregnant	.042 (3) .021–.083	.099 (4) .055–.18	1.1 (6) .68–1.8	1.4 (4) .78–2.5	1.2 (2) .52–2.8	1.7 (4) .94–3.1
White crossbred, progesterone-treated, nonpregnant	.043 (2) .019–.099	.10 (6) .062–.16	.39 (3) .20–.77	1.1 (4) .61–2.0	1.4 (1) .43–4.5	1.7 (3) .86–2.5
White crossbred, progesterone-treated, pregnant	.13 (3) .066–.26	.55 (5) .33–.93	1.1 (5) .65–1.9	2.0 (4) 1.1–3.6	1.9 (1) .59–6.2	1.6 (3) .81–3.2
Meishan, nonpregnant	.027 (2) .012–.062	.017 (4) .0094–.031	.16 (3) .081–.32	.23 (3) .12–.45	.53 (3) .27–1.04	1.2 (3) .61–2.4
Meishan, pregnant	.037 (2) .016–.085	.049 (5) .029–.083	.40 (5) .24–.68	.62 (6) .38–1.0	1.0 (2) .44–2.3	.65 (2) .28–1.49

[a]The pattern of change with time differed between White crossbred progesterone-treated and untreated gilts ($P < .05$). Overall effects of status ($P < .01$), progesterone treatment ($P < .01$), and breed ($P < .01$) were also observed.

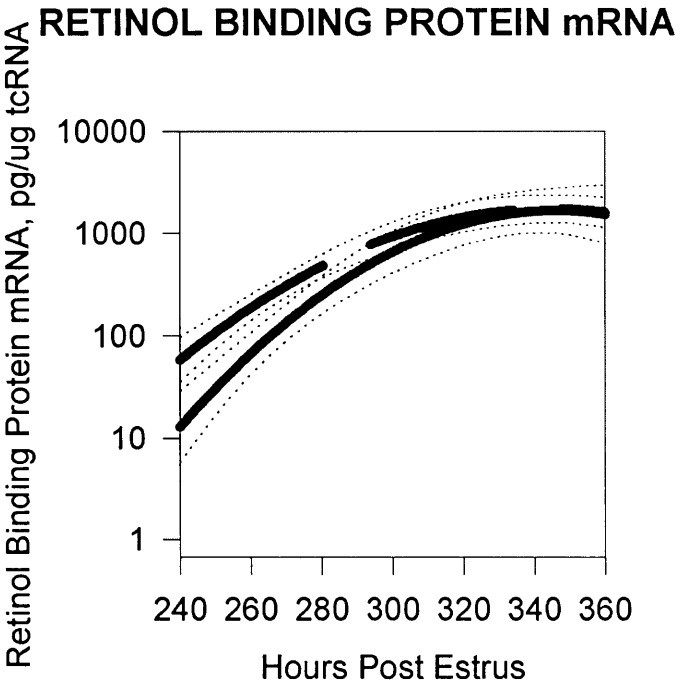

Figure 4. Regression lines for White crossbred progesterone-treated (broken line) and untreated (solid line) gilts along with 95% confidence intervals are illustrated for endometrial retinol-binding protein mRNA ($P < .05$).

intrauterine environment. This would also result in decreased conceptus loss via limitations in uterine capacity. Collectively, these results suggest that a more gradual increase in protein secretion may be associated with improved fertility. This possibility is currently under investigation.

Even though large amounts of oxytocin are present in the intrauterine environment of pregnant and nonpregnant pigs, the function of this oxytocin is not currently known. Data from this study indicate that oxytocin is greater in pregnant than in nonpregnant gilts, but concentrations were highly variable among animals. One possible explanation for this variability is that secretion from the endometrium is episodic and that some pigs were sampled during an episode and others were sampled between episodes. Given this scenario, the difference between pregnant and nonpregnant pigs could be due to more episodes, greater secretion during an episode or greater duration of each episode, which would thus increase the chances of sampling the individual gilts during an episode. Oxytocin receptors are present on the endometrial cells of gilts (Okano et al., 1996) and stimulation of these receptors with oxytocin stimulates the release of $PGF_{2\alpha}$ (Carnahan et al., 1996). This is similar to the control of $PGF_{2\alpha}$ during luteolysis in ruminants (Vallet et al., 1990; Lamming and Mann, 1995), in which the source of oxytocin is the corpus luteum (Flint et al., 1990). Luteal release of oxytocin can be caused by $PGF_{2\alpha}$ and a positive feedback loop initiated by subluteolytic amounts of oxytocin from the neurohypophysis has been suggested to be the mechanism that controls release of $PGF_{2\alpha}$ during luteolysis. It is tempting to speculate that in pigs, the endometrium has become a site of oxytocin formation. Higher oxytocin during pregnancy might serve to downregulate $PGF_{2\alpha}$ secretion or, alternatively, cause most of the prostaglandin to be released into the uterine lumen, which possibly would explain the endocrine-

Table 12. Least squares means (ng/μg tcRNA) and number of observations (parentheses) for endometrial uteroferrin in mRNA along with the range within one SEM using the pooled variance from ANOVA obtained with log transformed data

	Hours from estrus to slaughter					
Treatment group	240–244	256–268	280–291	304–316	328–331	360–364
White crossbred, nonpregnant[a]	.022 (3) .017–.028	.056 (4) .045–.070	.097 (5) .079–.117	.139 (5) .114–.170	.160 (1) .103–.251	.199 (4) .159–.248
White crossbred, pregnant	.033 (3) .026–.043	.036 (4) .029–.045	.136 (6) .113–.163	.174 (4) .139–.218	.095 (2) .070–.131	.187 (4) .149–.233
White crossbred, progesterone-treated, nonpregnant	.035 (2) .027–.045	.043 (6) .035–.051	.094 (3) .073–.122	.153 (4) .122–.191	.238 (1) .153–.372	.319 (3) .246–.411
White crossbred, progesterone-treated, pregnant	.036 (3) .028–.047	.089 (5) .073–.109	.165 (5) .135–.201	.202 (4) .162–.253	.220 (1) .141–.343	.161 (3) .125–.208
Meishan, nonpregnant	.030 (2) .022–.041	.037 (4) .030–.047	.041 (3) .032–.053	.069 (3) .054–.090	.166 (3) .128–.214	.284 (3) .220–.368
Meishan, pregnant	.022 (2) .016–.030	.023 (5) .019–.028	.071 (5) .058–.086	.105 (6) .087–.126	.182 (2) .133–.249	.139 (2) .102–.191

[a]The pattern of change with time differed ($P < .05$) between breeds. An overall effect of progesterone treatment ($P < .01$) and breed ($P < .01$) was also observed.

exocrine theory for inhibition of luteolysis in pigs (Bazer and Thatcher, 1977). Further work is needed to elucidate the role of endometrial oxytocin secretion.

Implications

Results indicate that intrauterine total protein and total retinol-binding protein are unaffected by the presence of the conceptus and instead seem to be influenced by progesterone timing. Uteroferrin may be influenced by progesterone timing and the presence of the conceptus. Oxytocin is controlled only by the conceptus and is unaffected by progesterone timing. Estrogen secretion by the conceptus was advanced by early progesterone treatment, but increased uterine protein secretion preceded the increase in estrogen secretion. Intrauterine content of total protein, retinol-binding protein, uteroferrin, and estrogen, which were lower in Meishan gilts than in White crossbred gilts, may play a role in the increased fertility of the Meishan breed. Collectively, the results are consistent with the hypothesis that progesterone timing controls the onset of increased uterine protein secretion and this in turn may influence the onset and extent of conceptus estrogen secretion.

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