

Comparative Ovarian and Pituitary Hormone Secretion in Pregnant Meishan and Yorkshire Gilts

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Summary and Implications

Chinese Meishan and Yorkshire were used to investigate mechanisms regulating the production and secretion of relaxin, progesterone, prolactin (PRL), and growth hormone (GH) during late pregnancy and lactation. Uterine surgical removal (hysterectomy) of nonpregnant gilts during the estrous cycle (day 8) extends luteal function to 150 days which is beyond the duration of normal pregnancy. Hysterectomy provides a useful model to examine shifts in hormone secretion at the time of expected parturition in gravid pigs. Blood samples were collected via an indwelling jugular cannula twice daily (0800 and 2000 hours) from days 90 to 120 and every 20 minutes within a 3-hour period on days 112 to 116. Relaxin and progesterone are hormones of ovarian origin, and PRL and GH are hormones secreted by the pituitary gland. Parturition occurred earlier (day 114) in Yorkshire than Meishan (day 115) gilts. The prepartum peak relaxin release occurred about 16 hours earlier in both breeds, thereafter relaxin dropped to basal levels during lactation. After hysterectomy, relaxin plasma levels were consistently greater in Meishan compared with Yorkshire gilts from days 110-118. A programmed peak relaxin release occurred one day earlier in Yorkshire compared with Meishan gilts. Following the relaxin peak, corpora lutea persisted in both breeds, but Meishan gilts continued to secrete consistently greater amounts of relaxin than Yorkshire gilts. Progesterone plasma levels remained higher longer in late pregnant Meishan compared with Yorkshire gilts. After hysterectomy, progesterone plasma levels were consistently higher in Meishan compared with Yorkshire gilts from days 101 to 118. Prolactin circulating concentration increased during late pregnancy and early lactation, but at a higher level in Yorkshire compared with Meishan gilts. Growth hormone blood levels increased only during late pregnancy and early lactation in both Meishan and Yorkshire gilts. These results indicate significant differences in the timing, and in some cases, magnitude of hormone secretion profiles in pregnant and hysterectomized Yorkshire and Meishan gilts.

Introduction

The importation of Chinese breeds of pigs has allowed us to evaluate their genetic and endocrine traits for reproduction, and compare these traits with those of our traditional breeds of pigs. Increased litter size in Chinese

breeds of pigs are of particular interest to livestock producers. Relaxin and progesterone production by corpora lutea in pigs occur during the estrous cycle, pregnancy, and after hysterectomy. Progesterone is required to maintain pregnancy, and relaxin is required for delivery of the piglets. Porcine antirelaxin serum given during late pregnancy delayed parturition and increased the duration of delivery of piglets and number of stillborns compared with phosphate buffer saline-treated controls. In pigs, prolactin (PRL) is luteotropic in mid-to late pregnancy. Growth hormone (GH) during lactation alters nutrient utilization in muscle and mammary tissue for milk synthesis.

The objective was to determine pituitary and ovarian hormones in Chinese Meishan and Yorkshire pigs during acute shifts in luteal function in different reproductive states. We investigated 1) relaxin, progesterone, estrone (E_1) and 17β -estradiol (E_2) secretion during late pregnancy and early lactation, and after hysterectomy, and 2) anterior pituitary gland secretion of PRL and GH during these stages.

Materials and Methods

Animals

Purebred Yorkshire gilts, averaging 130 ± 10 kg BW (\pm SE), that had exhibited at least one estrous cycle averaging 21 ± 1 day(s) (mean \pm SE) were used in this experiment. Seven pigs were bred on the first day of the second observed estrus, which was designated day 0. Nine unmated gilts were hysterectomized from days 6 to 8 after estrus. Meishan gilts exhibiting at least one estrous cycle averaging 21 ± 1 days (mean \pm SE) were used in this experiment. Six gilts were bred on the first day of the second observed estrus. Eleven unmated gilts were hysterectomized between days 6 to 8 after estrus.

Blood collection

Beginning on day 100 after estrus or mating, a blood sample was collected from the anterior vena cava. Blood (10 ml.) was collected in disposable borosilicate culture tubes (16 x 100 mm) containing .2 ml. of heparin sodium salt (100 IU/ml.; NBCO Biochemicals, Cleveland, OH). Plasma was decanted and stored at -20°C until required for hormone assay.

Radioimmunoassays

Relaxin concentration was quantified with a double antibody RIA using [^{125}I] monotyrosylated porcine relaxin and R6 antibody from B.G. Steinetz and by procedures we described previously. The assay sensitivity was 40 pg./tube.

Plasma progesterone was extracted with a benzene-hexane mixture. Plasma extracts were assayed for progesterone with a modification of the RIA procedure previously described using a fully characterized antibody (GDN antiprogestosterone-11 BSA 337). Sensitivity of progesterone RIA was 50 pg./tube.

Prolactin concentration in plasma was quantified with a double-antibody homologous RIA we described previously. The radioiodinated pPRL was prepared with the chloramine-

T method. Sensitivity of the assay was $.34 \pm .15$ ng./ml.

Growth hormone concentration was determined in duplicate 200- μ l. aliquots of plasma with a double antibody homologous RIA. Sensitivity of the assay was 230 ± 32 pg./ml.

Concentration of E_1 and 17β - E_2 were quantified by a procedure described previously. An aliquot of .05 to 1.0 ml. serum was brought to a volume of 2 ml. by adding sterile water, extracting three times with benzene, and dried. The estrogens were submitted to RIA by using fully characterized antiserum S-310 5/G. Assay sensitivity, defined as E_1 or 17β - E_2 standard that yielded 95% of radioactive counts in buffer control tubes, was about 2 pg.

Statistical analyses

Experimental units in this study were the individual gilts, and they were assigned to treatments at random. Data were analyzed with a split-plot analysis for reproductive state, time, and reproductive state x time. A one-way analysis of variance and a student's *t*-test for continuous variables were used for comparisons between treatment groups. Values are presented as mean \pm standard error.

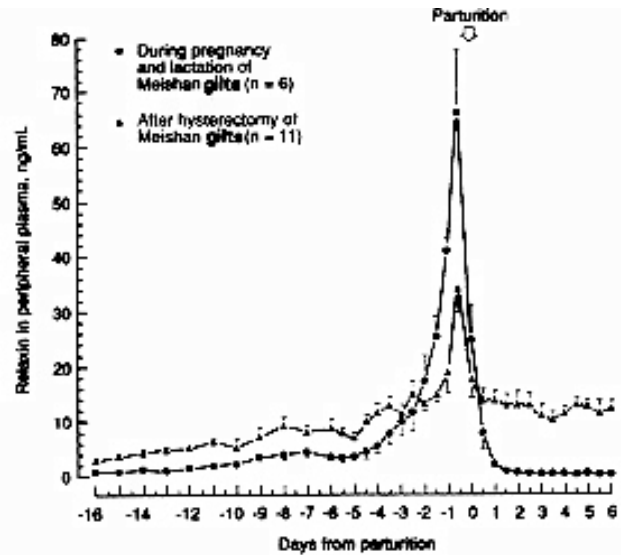
Results and Discussion

Parturition occurred on day 114 in Yorkshire and day 115 in Meishan gilts. Plasma relaxin concentrations were measured daily from days 99 to 120 in mated and hysterectomized Meishan gilts (Figure 1). The day of parturition, or day 115, in both groups was designated day 0 in Figures 1, 2, 3, and 4. Relaxin concentrations remained consistently greater (6 ± 1.0 ng./ml.; mean \pm SE) in hysterectomized than in pregnant gilts ($2 \pm .4$ ng./ml.) from days 99 to 109 (equivalent to days -16 to -6, $P < .05$). A gradual increase in relaxin concentrations began on day 111 in pregnant pigs and led to a sharp increase ($P < .01$) on days 112 to 114 (equivalent to days -2 to 0 in Figure 1). Relaxin peaked at 34 ± 4.2 ng./ml. in hysterectomized gilts compared with 66 ± 10.9 ng./ml. in pregnant gilts ($P < .05$); the relaxin peaks occurred on day 114 in both groups.

There was a sustained increase in relaxin over a 3-day period in pregnant gilts that resulted in the peak release that occurred on day 114, whereas 4 days of increase in relaxin concentrations preceded the relaxin peak in hysterectomized gilts (Figure 1). By day 1, postpartum relaxin concentration decreased from a peak of 66 ± 10.9 to $1 \pm .3$ ng./ml. and remained low until day 120. In hysterectomized gilts, relaxin decreased sharply from a peak of 34 ± 4.2 to 13 ± 3.2 ng./ml. within 1 day but remained consistently greater ($P < .05$) than in lactating pigs until day 120. A gradual increase in relaxin plasma concentrations began about day 110 and led to a sharp increase with peak relaxin at day 113 in Yorkshire and day 114 in Meishan gilts. Thereafter, relaxin decreased abruptly to basal levels in both breeds. Relaxin secretion profiles from representative Yorkshire and Meishan gilts revealed no evidence of a consistent episodic relaxin release during pregnancy and early lactation. After hysterectomy of unmated gilts at day 8 of estrous cycle, corpora lutea were maintained beyond 140 days on both breeds. From days 101 to 112, relaxin plasma concentration increased steadily in Meishan and Yorkshire gilts. A programmed relaxin peak release occurred on day 113 in

Yorkshire and day 114 in Meishan gilts. Thereafter, relaxin levels remained consistently greater in Meishan compared with Yorkshire gilts.

Figure 1. Relaxin concentrations in peripheral plasma of Meishan gilts during pregnancy and lactation compared with those in unmated gilts hysterectomized on days 6 to 8 after estrus. The open arrow indicates the day of parturition (day 0) or the day after peak hormone concentration in hysterectomized pigs. The number of gilts in each group is indicated in parentheses. Values are mean \pm SE.



Peripheral plasma concentrations of progesterone in hysterectomized gilts were greater ($P < .05$) than in pregnant gilts (29 ± 3.3 and 18 ± 1.9 ng./ml., respectively) from days 99 to 109 (equivalent to days -16 to -6 in Figure 2). Thereafter, in hysterectomized gilts, progesterone decreased by half from days 109 to 115 and remained at 16 ng./ml. until day 120. In contrast, progesterone concentration decreased sharply from 18 ± 1.9 ng./ml. 6 days before parturition to basal values of $< .5$ ng./ml. ($P < .05$) after parturition and remained basal until day 120. Progesterone plasma concentration remained higher longer in Meishan compared with Yorkshire gilts in late pregnancy. After hysterectomy, progesterone remained consistently greater than seen during pregnancy and lactation from days 101 to 118. However, at the time of expected parturition (day 114 or 115) progesterone decreased about half in both breeds of hysterectomized animals. Progesterone levels were consistently greater in hysterectomized Meishan compared with Yorkshire gilts from days 101 to 118.

Figure 2. Progesterone concentrations in peripheral plasma of Meishan gilts during pregnancy and lactation compared with those in unmated gilts hysterectomized on days 6 to 8 after estrus. The open arrow indicates the day of parturition (day 0) or the day after relaxin surge in hysterectomized gilts. The number of gilts in each group is shown in parentheses. Values are mean \pm SE.

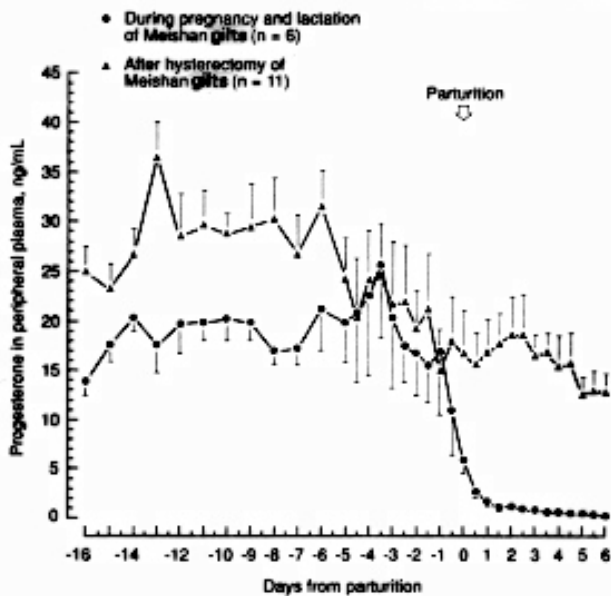
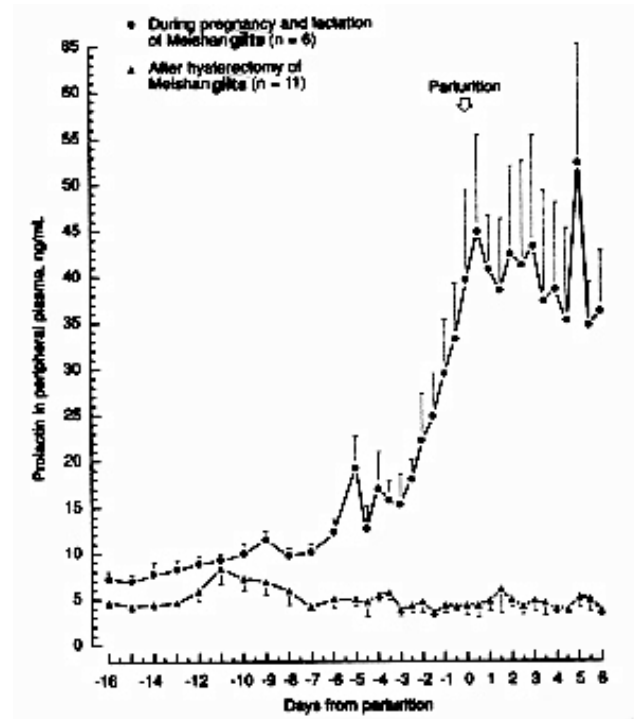


Figure 3. Prolactin concentrations in peripheral plasma of Meishan gilts during pregnancy and lactation compared with those in unmated gilts hysterectomized on days 6 to 8 after estrus. Day 0 is the day of parturition in pregnant pigs and the day after peak relaxin release in hysterectomized pigs. The number of gilts in each group is indicated in parentheses. Values are mean \pm SE.



Prolactin concentration in peripheral plasma of hysterectomized gilts remained low ($4 \pm .8$ ng./ml.) throughout the period of the study; whereas in pregnant gilts PRL increased steadily from $16 \pm .8$ on days 99 to 39 ± 10 ng./ml. at parturition (day 114) and remained increased during lactation (Figure 3). Prolactin plasma concentration remained similar after hysterectomy of unmated Meishan and Yorkshire gilts from days 101 to 118. During late pregnancy, prolactin plasma levels increased, and were consistently greater in Yorkshire compared with Meishan gilts from days 111 to 116 of pregnancy/lactation. There were occasional episodes of pulsatile PRL release during pregnancy and lactation.

Growth hormone concentrations (Figure 4) were similar in hysterectomized and pregnant gilts from days 99 to 114; however, after day 14, GH concentrations were consistently greater ($P < .05$) in lactating ($2.6 \pm .6$ ng./ml.) than in

hysterectomized ($1.0 \pm .3$ ng./ml.) gilts. GH plasma concentrations were similar in hysterectomized and pregnant Meishan and Yorkshire gilts from days 101 to 114; however, after day 114, GH levels were consistently greater in lactating dams than in hysterectomized dams. There were episodes of GH release detected in pregnant and hysterectomized pigs, but no consistent profile for either reproductive state. E_1 and 17β - E_2 increased markedly to peak blood concentrations by day 108 (parturition $114.6 \pm .4$ days), dropped precipitously after parturition, and remained consistently low during lactation. After hysterectomy of unmated gilts, E_1 and 17β - E_2 remained consistently low from days 12 to 168. Modest increases in E_1 and 17β - E_2 from days 132 to 168 coincided with increased follicular growth, but none of the gilts returned to estrus during this period.

Figure 4. Growth hormone concentrations in peripheral plasma of Meishan gilts during pregnancy and lactation compared with those in unmated gilts hysterectomized on days 6 to 8 after estrus. The day of parturition or the day after peak relaxin profiles in hysterectomized gilts is designated day 0. The number of gilts in each group is indicated in parentheses. Values are mean \pm SE.

