

Alteration in uterine contractility in mares with experimentally induced placentitis

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Abstract

An experimental model of ascending placentitis was developed in the mare to characterize the uterine myoelectrical pattern in late gestation and determine how ascending placentitis altered this pattern. In experiment 1, myometrial electrical activity was analyzed during the early morning, late morning and evening hours in four mares in the last 15 days of gestation to identify patterns of activity. In experiment 2, nine mares received intra-cervical inoculations of *Streptococcus equi* subspecies *zooepidemicus*. Myoelectrical activity in the early morning and evening hours in these mares was compared with four control mares. In experiment 1, the number of spike burst clusters >30 s was greater in the evening than in the late morning hours ($P < 0.04$). Spike burst activity (number \times duration) of mares in experiment 1 was similar during day and night recordings until the last 6 days of gestation when it gradually increased each evening until parturition ($P < 0.05$). In experiment 2, control mares experienced a gradual increase in the number of small spike burst clusters in the last 6 days ($P = 0.008$) and an increase in large and small spike burst clusters in the evening hours in the last 4 days of gestation ($P = 0.03$). Mares with experimentally induced placentitis never exhibited a rise in spike burst clusters but had an increase in the mean duration and activity index of large spike burst clusters in the 4 days before parturition ($P < 0.04$). In conclusion, control mares had a progressive, reversible rise in myoelectrical activity at night in the week preceding parturition. This was not observed in mares with experimentally induced placentitis. They exhibited an increase in the intensity and duration of large spike burst clusters possibly in response to local inflammation.

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Introduction

Placentitis, the single most important cause of late term abortion and stillbirth in the mare, is highly associated with premature delivery (Platt 1975, Whitwell 1988, Giles *et al.* 1993, Hong *et al.* 1993). Unlike women, premature birth usually results in the delivery of a non-viable foal because final fetal maturation in the horse occurs only during the last 5 to 7 days of development *in utero* (Silver & Fowden 1994). Precocious fetal maturation may occur in response to placental infection if premature labor can be delayed. Consequently, pharmacological suppression of premature labor could significantly improve postnatal survival. A determination of the normal uterine contractile pattern in the late-gestational mare and how it is altered by placentitis is required to develop treatments to prevent premature birth.

Two models of uterine myoelectrical patterns in late gestation, the primate and ruminant, have been described. In the primate, there is a switch from low amplitude,

long-lasting epochs of electrical and contractile activity that occur throughout pregnancy, called contractures, to prominent, short-lived epochs accompanied by greater contractile force called contractions. The switch occurs around the onset of darkness several nights before delivery (Farber *et al.* 1997) and is reversible: it occurs over a number of nights before delivery with myoelectrical activity returning to low amplitude contractures during the day. The switch is also progressive because nightly contractions increase in frequency and amplitude as the dam nears parturition. In the ruminant, the uterus begins to contract in the last 6–24 h of gestation. Contractions increase in frequency and amplitude until the neonate is born (Harding *et al.* 1982). Relatively little is known about the uterine contractility pattern in the late-gestational mare. In a small study in which myoelectrical activity was analyzed for 2 h daily in two pony mares in late gestation, Haluska *et al.* (1987) observed that the change in the percent myoelectrical activity per hour was

a function of the frequency of the myoelectrical bursts, and frequency of spike bursts increased as parturition approached.

In women, intrauterine infection is highly associated with idiopathic preterm labor. Bacteria that infect maternal and fetal gestational tissues ascend from the maternal vagina (Romero & Mazor 1988). Infection of human gestational tissues activates decidual macrophages resulting in production of cytokines and arachidonic acid metabolites by decidual and chorion cells (Dudley & Trautman 1994). Production of interleukin (IL)-8 activates and recruits neutrophils into the affected tissues. These inflammatory processes result in prostaglandin production by amniotic cells and stimulation of myometrial cell contractility. In the primate, intra-amniotic inoculation with group B streptococcus results in sequential increase in amniotic fluid cytokines, prostaglandins, and uterine activity (Gravett *et al.* 1994). Pro-inflammatory cytokines rise as early as 6 h after inoculation with an increase in uterine contractility (measured by intrauterine pressure; IUP) 14–40 h later and delivery 55–72 h after inoculation. There are no studies in the mare on the interactions of bacterial placentitis, inflammation, and uterine contractions. The aims of the present study were to (1) develop a technique for measuring uterine myoelectrical activity in the mare in late gestation, (2) characterize the contractile pattern during late gestation, and (3) determine if ascending placentitis alters the normal contractile pattern. We hypothesized that the mare is like the primate and has a progressive, reversible increase in uterine contractions at night in the week that precedes parturition. We further hypothesized that ascending placentitis would be associated with an increase in uterine contractions resulting in premature birth of a foal.

Materials and Methods

Experimental animals

Fifteen pony mares were bred naturally or by artificial insemination in 1997 ($n = 4$), 1998 ($n = 3$), 1999 ($n = 6$), and 2000 ($n = 2$). Mares were maintained at pasture or in stalls after surgical instrumentation at the College of Veterinary Medicine, University of Florida. Pregnancy was confirmed by ultrasonographic examination of the reproductive tract. Mares received normal preventative health care as dictated by the Equine Research Protocol of the College. In addition, mares were vaccinated against viral abortion during the 5th, 7th, and 9th months of pregnancy as a prophylactic against possible abortion due to equine herpes virus 1. This project was approved by the Institutional Animal Care and Use Committee (A350).

Surgical instrumentation and post-surgical management

Mares were fitted with myometrial electrodes between days of gestation (dGa) 235 and 289. Twelve hours before

surgery, they were moved into a stall and provided with free choice water. Mares were administered 20 000 units potassium penicillin G (Marsam Pharmaceuticals, Inc., Cherry Hill, NJ, USA)/kg body weight, 6 mg gentamicin (Schering-Plough, Kenilworth, NJ, USA)/kg body weight i.v. and 1 mg flunixin meglumin (Schering-Plough)/kg body weight i.v. through an indwelling jugular catheter 2 h before anesthesia was induced. Altrenogest, an oral progestin was also given (0.0088 mg/kg of body weight; DPT Laboratories, San Antonio, TX, USA) for 7 days. Antibiotics and flunixin meglumin were given at the appropriate intervals for 4–6 days after surgery, depending on the mare's response to instrumentation.

Thirty minutes before induction of anesthesia, mares were premedicated with xylazine (Bayer Corporation, Shawnee Mission, KA, USA) at 0.4 mg/kg i.v. General anesthesia was induced using a combination of guaifenesin (Ft Dodge Animal Health, Ft Dodge, IA, USA) given to effect and ketamine (Ft Dodge Animal Health) at 2.0 mg/kg i.v. In 1998 and 1999, general anesthesia was maintained using halothane in oxygen. In 2000 and 2001, general anesthesia was maintained using i.v. administration of 50 g guaifenesin, 1 g ketamine, and 500 mg xylazine in 1 litre 5% dextrose solution (Baxter Health Care Corporation, Deerfield, IL, USA) given to effect in a slow intravenous drip. These mares were ventilated with oxygen only.

Following anesthetic induction, the mare was positioned on a surgical table in lateral recumbency with the side of the non-gravid uterine horn down. A paramedian incision was made approximately 10 cm off midline from midway in the costal arch and directed caudoventrally towards the inside fold of the flank. The uterus was exteriorized and electrodes were sutured on the dorso-lateral surface of the pregnant horn below the attachment of the broad ligament. Two types of myometrial electrodes were used during the 4-year study. In 1998–2000, five bi-polar Ag–AgCl electrodes constructed as described previously (Lester *et al.* 1992) were implanted. In 2001, electrodes consisted of three sets of paired wires that were passed with an 18 gauge needle through the myometrium (Gravett *et al.* 1994). A bi-polar electrode was made by placing the paired wires 3 mm apart. Electrodes exited the abdomen through the flank region. In 1998–2000, wires exited the body wall through the flank whereas in 2001 the wires were tunneled subcutaneously from the flank to the withers. Changes in electrode type and site of exit were made to decrease granulation tissue formation at the exit site due to excessive movement and the large size of the wires.

Experimental design

Experiment 1

Four pony mares were surgically implanted with five myometrial electrodes between dGa 280 and 285 to determine the pattern of myoelectrical activity in late gestation.

Myometrial electrical activity was recorded daily from 0530 to 0830 h, 1000 to 1200 h and 1630 to 2230 h beginning on dGa 310 until parturition. Studies were conducted in July and August 1998. The number and duration of large spike burst clusters per hour (>30 s) from the last 15 days of gestation were analyzed. Amplified raw data (Fig. 1) were filtered using a band-pass finite impulse response set between 10 and 70 Hz to isolate high-frequency spike burst activity. Raw, analog-filtered data were then analyzed visually.

A large spike burst was defined as myoelectrical activity that appeared on 50% or more of the active electrode channels for more than 30 s. The start of a spike burst was defined as a rise in activity at least two times the height of baseline. The end of a spike burst was defined as a drop in myoelectrical activity below two times the height of the baseline for more than 16 s.

Experiment 2

Thirteen pony mares were surgically fitted with three to five uterine myometrial electrodes between dGa 235 and 289. Recording of myometrial electrical activity began 7–14 days after instrumentation. Four of the 13 mares

served as controls while nine mares received intra-cervical inoculations of *Streptococcus equi* subspecies *zoepidemicus*. Uterine activity was recorded daily in control mares from dGa 310 until parturition. In mares with ascending placentitis, activity was recorded daily from 3 days before inoculation until parturition. In 1998–2000, uterine activity was monitored daily from 0530 to 0830 h and from 1730 to 2230 h ($n = 11$; four control mares and seven mares with experimental placentitis). In 2001, myoelectrical activity was recorded only in the evening (two mares with experimental placentitis) because data analyzed from the previous 3 years indicated that changes in activity occurred in the evening hours. The hours monitored were reduced because of cost.

Bacterial inoculation

A single stock solution of 1×10^9 colony forming units (CFU)/ml *Streptococcus equi* subspecies *zoepidemicus* was divided into 2.5 ml aliquots, stored in vials at -70°C and used for all studies. It was obtained from a clinical case of equine endometritis submitted to the Microbiology Laboratory of the College of Veterinary Medicine, University of Florida. An inoculum was prepared by diluting the

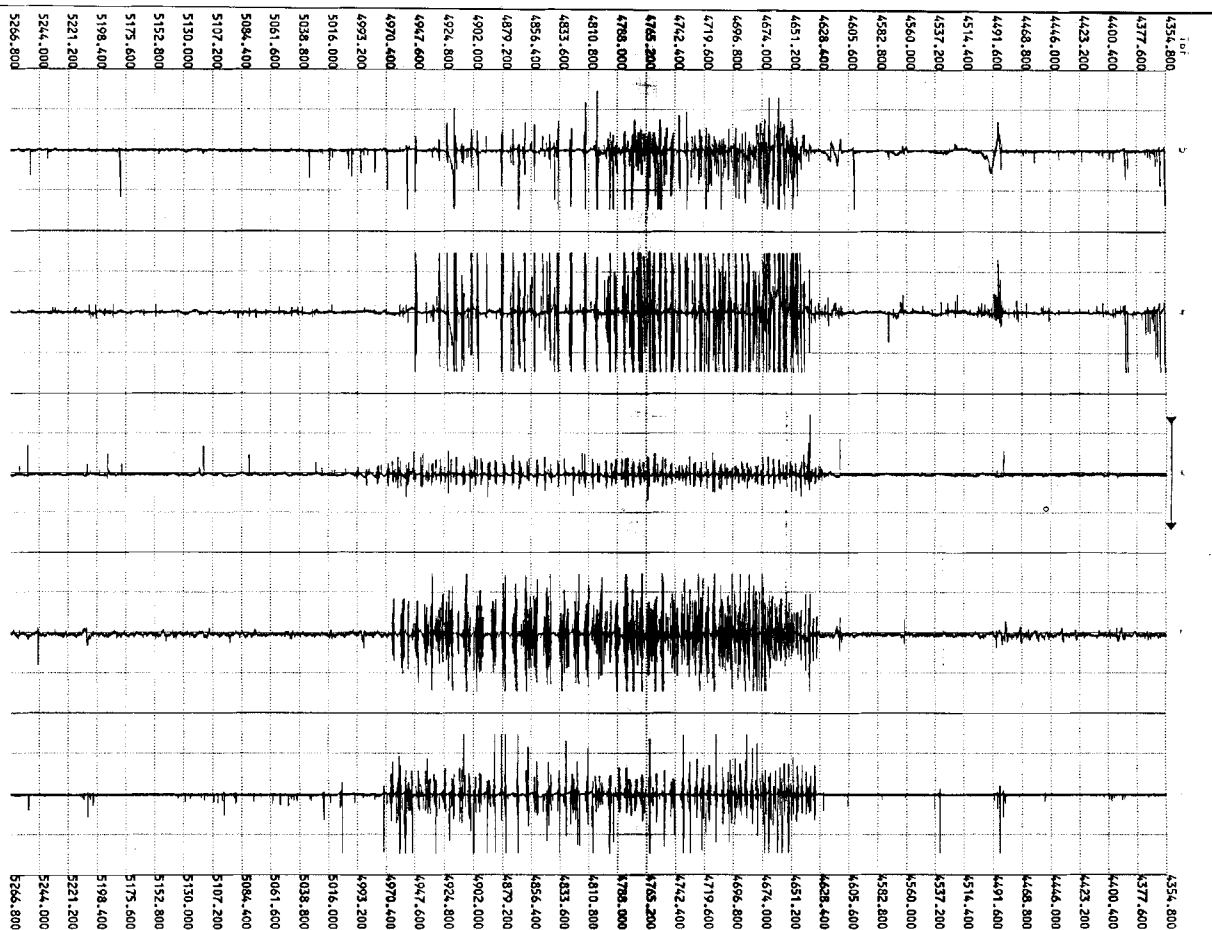


Figure 1 Example of a single spike burst cluster (compressed raw data).

stock solution with sterile saline to yield 1×10^8 CFU in a volume of 1 ml. In the first two mares, the dose needed to induce a chronic placentitis was not known. Accordingly, one mare received 3×10^{10} CFU while the other mare received 1×10^6 CFU *Streptococcus equi* subspecies *zooepidemicus*. The first mare delivered a dead foal 4 days after inoculation. Mare 2 (1×10^6 CFU) exhibited no clinical signs (vaginal discharge, mammary development) for 14 days after inoculation so re-inoculation was performed with 1×10^8 CFU on day 15. Within 24 h of the second inoculation, the mare developed a vaginal discharge. The remaining seven mares received a single dose of 1×10^8 CFU of *Streptococcus equi* subspecies *zooepidemicus*.

Mares were inoculated between dGa 255 and 295. Eight of nine mares received intrauterine inoculations between dGa 277 and 295 (mean dGa 287). Mare 2 described above was inoculated on dGa 255 and dGa 270. The inoculation procedure was as follows. The tail of the mare was wrapped, and the perineum washed thoroughly with an iodine-based soap. A sterile artificial insemination pipette was introduced into the vagina by an operator whose hand and arm were covered with a sterile glove and sleeve. The cervix was located, a portion of the cervical plug removed and the inoculum deposited approximately 2 cm into the cervix. A second syringe with 6 ml air was used to clear the pipette of remaining inoculum before it was withdrawn.

Myoelectrical data acquisition and analysis

Ponies were conditioned to the recording environment before surgery. Mares stood in stocks or were tied in box stalls during the recordings. They were fed free choice hay, had continual access to water and were given a 12% protein sweet feed every 8 h. Myoelectrical activity was recorded using a Grass instrument amplifier with a low frequency cut-off of 5.3 Hz and a high-frequency cut-off of 100 Hz. After amplification, the analog signal was digitized analog-to-digital conversion (ADC) board; Data Translation, Marlboro, MA, USA) at a rate of 100 samples/s per channel (Codal, Dataq Instruments, Akron, OH, USA) and stored on electromagnetic media for future analysis. A channel file from each electrode was digitally filtered with a bandpass Finite Impulse Response digital filter (Grass-Telefactor Instruments, West Warwick, RI, USA) using the window method. Filtered data were integrated into 10-s blocks (1000 data points) and blocked into 60-min periods to quantify clusters of spike bursts. The data were analyzed using a customized spike burst recognition program written in the Fortran language (Lester *et al.* 1992). Pattern recognition was verified by visual analysis of raw data plots.

Spike burst clusters were divided into greater than 30 s (large clusters) and less than 30 s in length (small clusters). The beginning and end of a spike burst cluster was determined by analyzing graphed integrated data visually

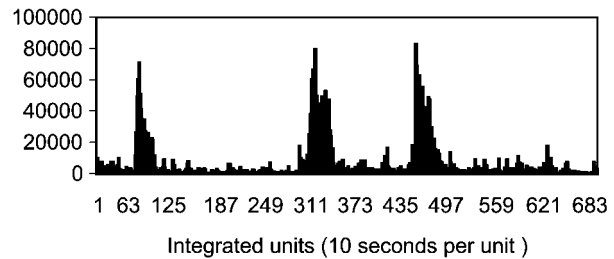


Figure 2 Sample of filtered, smoothed myoelectrical data integrated into 10-s blocks. Three clusters of spike burst are seen in this chart. Low cut-off value was set at 15 000 and high cut-off at 45 000. Each bar represents 10 s of uterine myoelectrical data.

(Fig. 2). A low cut-off point (baseline activity) and a high cut-off point (the minimum activity that a burst had to reach to be considered a spike burst cluster) were identified visually from each recording file. These values were added to the computer program and the recording file was then analyzed.

The number and duration of small and large spike burst clusters and the activity index were measured daily until foaling. The activity index was defined as the relative intensity of a cluster. It was calculated by the computer program by dividing the total integrated value of the cluster by its duration.

Statistical analysis

Experiment 1

The number and duration of spike burst clusters >30 s and the spike burst activity within a day (duration \times frequency) was analyzed by repeated measures analysis of variance using the Mixed procedure of SAS (SAS Institute Inc., Cary, NC, USA). The mathematical model included effects of mare, day, time of day, day \times time of day interaction. The interaction between day and spike burst activity (number of spike bursts clusters >30 s \times duration of bursts expressed as a percentage of time within a recording session) was calculated by regression analysis. Significance was set at $P < 0.05$.

Experiment 2

Uterine myoelectrical activity was analyzed by least squares analysis of variance using the General Linear Models Procedure of SAS (SAS Institute Inc.). The variables analyzed were number of spike burst clusters per hour, activity index, and mean duration of each cluster. Variables were further divided into bursts greater than 30 s and bursts less than 30 s. Mares were divided into three groups: control mares (controls; $n = 4$), mares that aborted within 7 days of inoculation (acute placentitis; $n = 5$), and mares that delivered foals 15 or more days after inoculation (chronic placentitis; $n = 4$). Three data sets were analyzed: (1) data from the last 10 days of gestation from control mares, (2) data from mares with placentitis, and (3) data collected in the evening hours only from all

mares. Data used from mares with experimental placentitis included 3 days of baseline uterine activity before inoculation, all days recorded in mares with acute placentitis and the last 10 days of activity before delivery in mares with chronic placentitis. Data from the last 8 h of gestation were not included in any analysis as not all mares were monitored in the last 8 h and recording during active labor would skew data.

The mathematical model for analysis of data from control mares included the effect of year, mare (nested within year), day prepartum, mare \times day prepartum interaction, time of day, time of day \times mare interaction and residual error. Data from mares with placentitis were analyzed with the model presented above, and with a second model that used days around inoculation (day -3 to $+3$; 0 = inoculation) instead of day prepartum. Analysis of the data set consisting of evening hours monitored in control mares and mares with placentitis involved a similar mathematical model with the inclusion of treatment (control or infected) and interactions with treatment \times day prepartum. Significance was set at $P \leq 0.05$. Data are presented as least squares means (LSM) \pm S.E.M.

Results

Experiment 1

All mares delivered viable foals between dGa 318 and 331. Mares had recurring clusters of spike bursts that were comprised of a series of short (<5 s) spike bursts grouped over periods of 2–8 min. There was a day \times time of day interaction on the number of spike bursts >30 s/h ($P = 0.04$). The number of spike burst clusters between 1000 and 1200 h was less than that recorded in the evening hours for the 15 days preceding parturition. Beginning 9 days before and continuing to parturition, the number of spike bursts recorded in the early and late morning hours was less than that recorded in the evening session (Fig. 3). Duration of spike bursts tended to decrease as the mare neared parturition (day \times time of day interaction $P = 0.09$). Spike burst activity (expressed as percent time in activity during a recording session) was similar during day and night recordings until the last week of gestation. Seven to eight days before delivery, spike burst activity began to increase in the early evening hours and then increased steadily in the evening as parturition approached (Fig. 4; $P < 0.05$ – time \times day interaction).

Experiment 2

Three of four control mares delivered viable foals between dGa 317 and 340. The fourth mare experienced a dystocia on dGa 340 and the foal died shortly after birth. Time from inoculation to delivery in mares with experimentally induced placentitis varied from 4 to 30 days. Foals were delivered between dGa 284 and 314. One foal born on dGa 313 was viable and lived with minimal care.

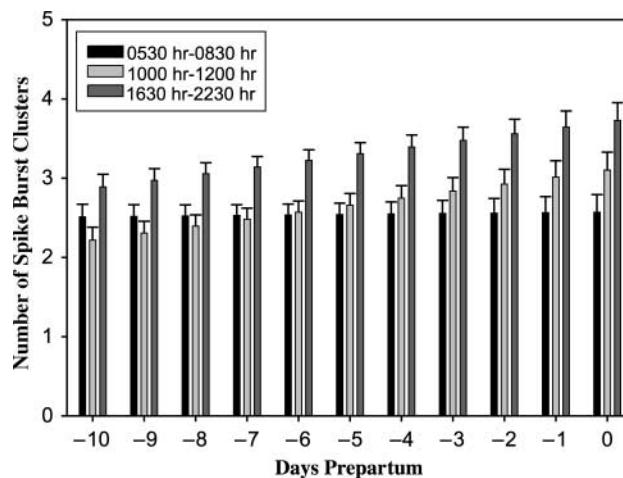


Figure 3 Number of spike burst clusters >30 s/h in control mares from experiment 1. Results are expressed as LSM \pm S.E.M. The number of spike burst clusters in the morning hours was less than that recorded in the evening sessions ($P < 0.01$) beginning on day 9 before parturition.

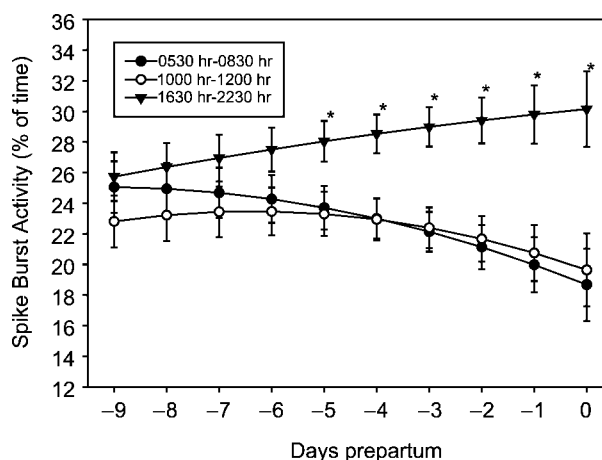


Figure 4 Diurnal effects on uterine myoelectrical activity in the late-gestation mare. Spike burst activity increased steadily over the last 6 days of gestation, increasing steadily until parturition ($P < 0.05$).

The remaining eight foals were either born dead or were euthanized. Five mares aborted between 4 and 7 days after inoculation (day 4, one mare; day 5, two mares; day 7, two mares) and were referred to as having acute placentitis. Four mares foaled 15–30 days after inoculation (days 15, 22, 27, and 30). The mare that delivered 30 days after inoculation was inoculated twice and delivered 15 days after the second inoculation. The mare that delivered 22 days after inoculation delivered a live, viable precociously mature foal. Placentitis was confirmed by culture of the fetal stomach contents in dead foals and by gross and histopathological examination of the placenta. *Streptococcus equi* subspecies *zooepidemicus* was recovered from fetal fluids in all dead foals. All mares had gross and histological inflammatory changes consistent with

ascending placentitis in the cervical star region of the placenta (Stawicki 2001).

Control mares experienced a gradual increase in the number of small spike burst clusters in the last 6 days of gestation ($P = 0.008$). Small spike burst clusters rose from 1.51 ± 1.24 at 10 days before parturition to 15.72 ± 2.59 (LSM \pm S.E.M.) on the day of parturition (excluding the last 8 h before delivery; Fig. 5). The number of large and small spike burst clusters was greater in the evening as compared with the morning in the last 4 days of gestation ($P = 0.03$). Total number of spike burst clusters per hour rose from 6.06 ± 2.69 at 10 days before parturition to 28.42 ± 5.63 on the day of parturition (excluding the last 8 h before delivery). The activity index of spike burst clusters did not vary as mares neared parturition and mean duration of spike bursts remained constant.

Treatment affected the total number of clusters ($P < 0.05$; Fig. 6) and the number of small clusters ($P = 0.005$) but did not significantly affect the number of large clusters. There was a significant interaction between treatment and day prepartum on the number of small clusters per hour ($P < 0.0001$) and total number of

clusters ($P = 0.04$). In contrast to the controls, mares with experimentally induced placentitis did not experience a rise in the number of small or large spike burst clusters per hour before delivery. However, there was a treatment \times day interaction in the last 4 days prepartum on mean duration ($P = 0.035$; Fig. 7) and activity index of large spike burst clusters ($P = 0.01$; Fig. 8) in mares with placentitis. Both variables increased as the mare neared parturition. There were no differences in myoelectrical patterns between mares that developed acute or chronic placentitis. The number of small clusters decreased below baseline for 3 days after inoculation in mares with acute and chronic placentitis ($P = 0.02$; Fig. 9).

Discussion

The present findings suggest that uterine contractility during late gestation in the mare is similar to that in the rhesus macaque (Ducsay *et al.* 1983), baboon (Morgan *et al.* 1992), and woman (Main *et al.* 1991). All of these species exhibit a diurnal rhythm in the number of uterine contractions with a marked clustering during the

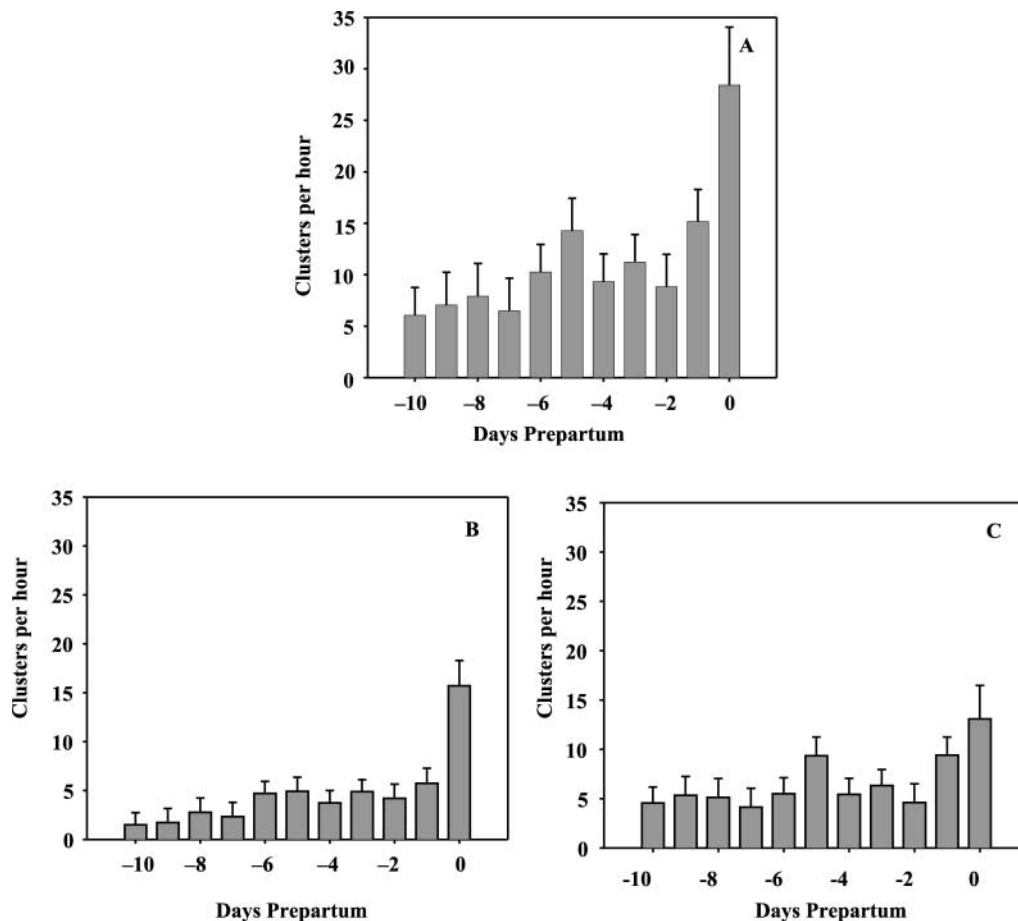


Figure 5 Number of spike burst clusters per hour in the last 10 days of gestation in control mares in experiment 2 expressed as LSM \pm S.E.M. (morning and evening hours combined). (A) Total clusters, (B) small clusters, and (C) large clusters. Total number of clusters tended to be affected by day ($P = 0.09$) while small clusters began to increase 6 days before parturition ($P = 0.008$). The number of large clusters per hour did not change in the last 10 days.

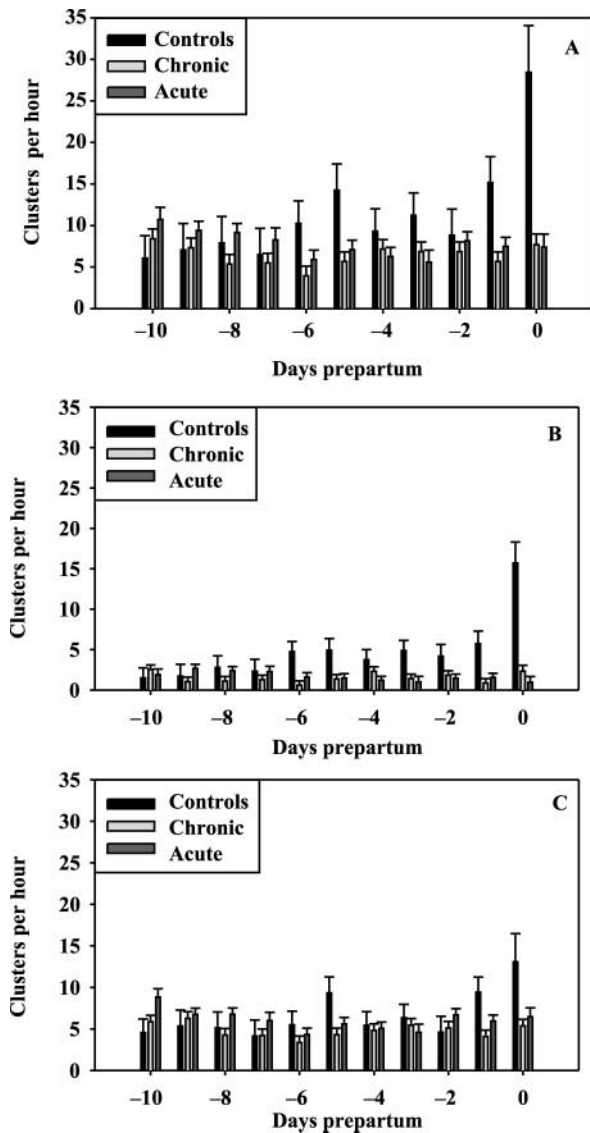


Figure 6 Number of spike burst clusters per hour in control mares and in mares with experimentally induced ascending placentitis during the last 10 days of gestation. Results are LSM \pm S.E.M. (A) Total clusters, (B) small clusters, and (C) large clusters. Treatment affected the total number of clusters ($P = 0.05$) and the number of small clusters, but not the number of large clusters. There was a treatment \times day interaction for the total number of clusters ($P = 0.002$) and the number of small clusters per hour ($P < 0.0001$). Total clusters per hour began to increase 6 days before parturition in control mares as a result of an increase in the small clusters ($P < 0.0001$). Total, small, or large spike burst clusters did not change in mares with placentitis.

night-time hours as parturition approaches. In women, 67.4% of uterine contractions occur in the hours from 2000 to 0800 h. Near term, daytime contractions in women (0900–1500 h) reach a maximum of three to five contractions per hour, whereas during evening monitoring periods (2100–0300 h) the peak is as high as eight to ten contractions hourly (Moore 1995). In the rhesus monkey in late gestation, the contractile pattern peaks between

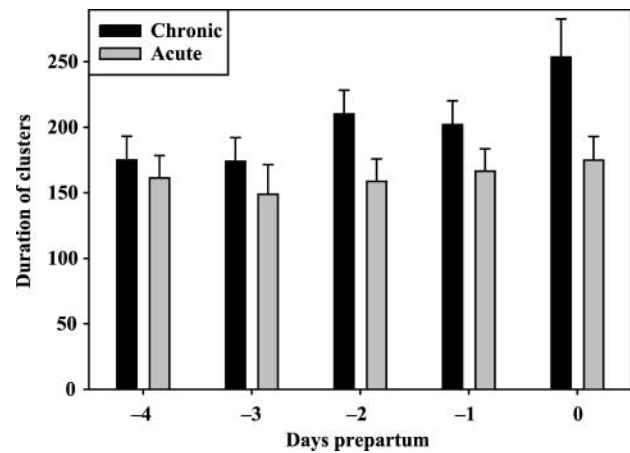


Figure 7 Duration of large spike burst clusters in mares with experimentally induced placentitis during the last 4 days of gestation expressed as LSM \pm S.E.M. Duration of the large clusters increased with impending parturition ($P = 0.035$).

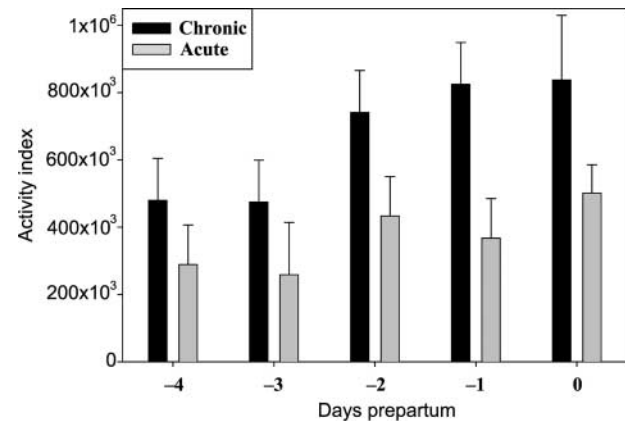


Figure 8 Activity index of large spike burst clusters in mares with experimentally induced placentitis during the last 4 days of gestation expressed as LSM \pm S.E.M. Intensity of the large clusters increased with impending parturition ($P = 0.01$).

2300 and 0100 h, with a low from 0800 to 1400 h (Ducsay *et al.* 1983). In the present study, control mares from experiment 1 exhibited spike burst clusters for approximately 25% of the time of each recording session (11 h recording daily) until 6 days before delivery when evening activity increased to approximately 30% of the recording time. These findings are in agreement with Haluska *et al.* (1987) who found that the myometrium is electrically active 30% of the time in the mare in late gestation.

Uterine activity in women and primates has been classified into two categories: contractures and contractions. Contractures are epochs of myometrial activity relatively long in duration that generate little change in IUP, while contractions are much shorter in duration and are associated with increases in IUP (Morgan *et al.* 1992). In the primate, contractures are seen throughout pregnancy, occur at a frequency of two to six per hour, and last for roughly

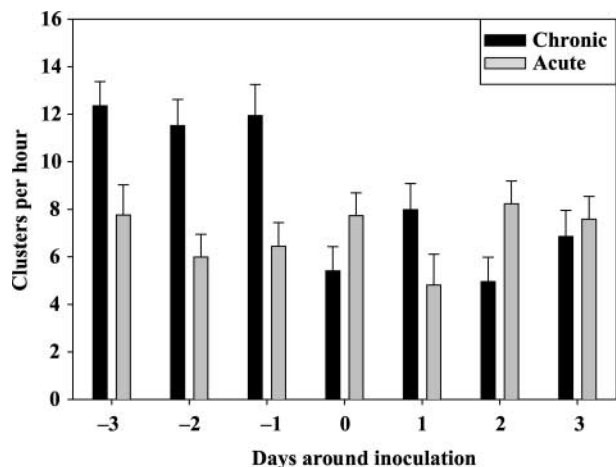


Figure 9 LSM \pm S.E.M. for the total clusters of spike burst in mares with experimentally induced placentitis 3 days before and after inoculation. Total clusters per hour decreased for 3 days following inoculation ($P = 0.02$).

3–5 min (Farber *et al.* 1997). As parturition nears, there is a progressive switch from contractures to contractions, which is centered around the onset of darkness (Morgan *et al.* 1992). The contractions, which generally last less than 1 min, can occur as frequently as 30 times per hour (Farber *et al.* 1997). The ruminant does not display the progressive shift in uterine myoelectrical activity that is observed in the primate, although activity can still be grouped into contractures and contractions. In the pygmy goat, contractures that are 6–7 min in length occur every 45–74 min (Taverne & Scheerboom 1985). In the cow, contractions begin to appear independently of the contractures (Burton *et al.* 1987) beginning 18 h before parturition and continue through delivery of the offspring.

The mare also appears to experience contractures and contractions, although definitive conclusions cannot be made without measurements of IUP. The small clusters of spike bursts (<30 s) appeared to correspond to contractions while large clusters of activity (>30 s) correspond to contractures. In the control mares, the duration of large clusters (putative contractures) ranged from 2.54 ± 0.40 to 4.04 ± 0.53 min (LSM \pm S.E.M.), and the number per hour varied little as gestation progressed. Small clusters of activity ranged in duration from 8.25 ± 1.54 to 13.92 ± 3.22 s and increased in the last 6 days of gestation in control mares. Thus, it appears that patterns of uterine activity in mares carrying normal pregnancies are similar to those of the primate in the days preceding parturition.

Regulation of the patterns of myoelectrical activity in the primate and ruminant is associated with increases in maternal plasma estrogen and oxytocin. Rising estrogen during late pregnancy in rhesus monkeys plays a supportive role in establishing nocturnal uterine activity which is mediated by maternal oxytocin. Estrogen stimulates an increase in oxytocin production (Zhang *et al.* 1991), an

increase in oxytocin receptor availability and uterine sensitivity (Soloff *et al.* 1982, Honnebier *et al.* 1989), and increased prostaglandin synthesis (Zhang *et al.* 1991). The rise in maternal plasma estrogen concentrations in late pregnancy in the human (Buster *et al.* 1979), in the rhesus monkey (Novy & Walsh 1983), and in the sheep (Challis 1971) is associated with an increase in uterine activity. In the pregnant sheep, maternal plasma estrogen concentration rises abruptly in the last 24 h of pregnancy (Challis 1971) and is associated with a single switch from contractures to contractions about 6 h before delivery. In contrast, maternal plasma concentrations of estrogen in pregnant rhesus monkeys increase over several days and are associated with a nightly reversible, progressive increase in contractions in the last week of gestation. Maternal oxytocin regulates the episodes of nocturnal uterine activity in the primate as the increase in contractile activity in the early evening hours is correlated with elevated concentrations of oxytocin in maternal plasma and abolished by infusion of oxytocin antagonists (Honnebier *et al.* 1989, Hirst *et al.* 1993). Furthermore, infusion of androstenedione, an estrogen precursor, to rhesus monkeys for 24 h in late gestation maintains elevated estrogen concentrations in maternal plasma for the full 24-h period; however, the switch from contractures to contractions occurs only at night (Figuerola *et al.* 1989). In the mare, the rise in myoelectrical activity in the last 6 days of gestation corresponds with changes in the secretion pattern of plasma estradiol-17 β . Maternal estradiol-17 β appears to be released in pulses in late gestation. These pulses are most prominent at night, with the greatest difference between day and night hours during the 6 days preceding parturition (O'Donnell *et al.* 2003). The pattern of release of oxytocin in the last week of gestation in the mare is not known.

The mechanisms by which labor and delivery are regulated in spontaneous parturition and in infection may be entirely different (Challis & Smith 2001). Mares with experimentally induced placentitis did not exhibit a rise in small burst clusters (putative contractions) in the last week of gestation as did control mares. They exhibited an increase in the duration and intensity of the large spike bursts (putative contractures) in the 4 days preceding parturition. This increase may be associated with a rise in IUP resulting in cervical relaxation and dilation and eventual delivery. In the primate, experimentally induced chorioamnionitis is also associated with contracture type activity and conversion to progressive, reversible contractions are rarely seen. It is hypothesized that the contractile pattern and premature delivery seen in the experimental model of chorioamnionitis in rhesus monkeys is due to increased concentrations of prostaglandins and cytokines in amniotic fluid associated with infection (Gravett *et al.* 1994). Increases in IL-1, IL-6, tumor necrosis factor (TNF)- α , and high concentrations of prostaglandins E₂ and F₂ α have been found in the amniotic fluid of women with intra-amniotic infection and preterm labor (Romero *et al.* 1988, 1989a,b, 1990, Hillier *et al.* 1993). Bacteria and

bacterial products stimulate the production of IL-1, IL-6, and TNF- α by macrophages and decidual cells (Casey *et al.* 1989, Romero *et al.* 1989c, 1990). Explants of amnion and deciduas release prostaglandins in response to stimulation by these cytokines (Romero *et al.* 1989b,d, Mitchell *et al.* 1991, Pollard & Mitchell 1996, Saji *et al.* 2000). We have reported that mares with experimental placentitis have greater mRNA expression of pro-inflammatory cytokines in placental tissues at delivery and have higher concentrations of prostaglandins F and E in allantoic fluids in the last 48 h of gestation than do control mares (LeBlanc *et al.* 2002). An interaction between infection, cytokines and locally produced prostaglandins may contribute to premature delivery in the mare with ascending placentitis.

Several mares began foaling while myoelectrical activity was being recorded or foaled immediately after the recording session ended. Recording sessions within 8 h of parturition were not included in the statistical model because they included activity that would be considered first stage labor. Visual assessment of myoelectrical data files that were recorded in the last 8 h of gestation showed long periods of decreased activity followed by very short, rhythmic bursts of activity. This supports the findings of Haluska *et al.* (1987) that there is a decrease in uterine activity 2–4 h before foaling. This period of quiescence is thought to be the time when the foal rotates from the dorso-pubic position to the dorso-sacral position.

Myoelectrical activity in mares decreased below baseline for 3 days after intra-cervical inoculation with *S. equi* subspecies *zoepidemicus* and then returned to pre-inoculation values. The decrease in myoelectrical activity was mirrored by a decrease in the plasma PGFM concentration following inoculation (Stawicki 2001). It is speculated that up-regulation of IL-1 receptor antagonist may be associated with these findings (Romero *et al.* 1989a).

In conclusion, normal parturition in the mare appears to be similar to that of the primate in that uterine myoelectrical activity increases progressively at night during the last week of gestation. This pattern of activity is not seen in mares with ascending placentitis before delivery indicating that the mechanisms that control parturition and delivery may differ between normal delivery and infection-induced situations.

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