



LACK OF AN EFFECT OF PROSTAGLANDIN INJECTION AT ESTRUS ONSET ON THE TIME OF OVULATION AND ON REPRODUCTIVE PERFORMANCE IN WEANED SOWS

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Received for publication: December 7, 2000

Accepted: May 15, 2001

ABSTRACT

We evaluated the effects of a PGF₂α analogue on time of ovulation and reproductive performance in multiparous Camborough sows (n=47). At onset of first post-weaning estrus, sows received either an intravulval injection of 3.75 mg of prostaglandin analogue (PGF) or, served as a non-injected control (CON). Beginning 24 h after the onset of estrus, transcutaneous ultrasonography was carried out every six h to determine time of ovulation. At 36, 54, and 72 h after the onset of estrus, blood samples were taken for progesterone analysis. Weaning-to-estrus (WEI), duration of estrus, ovulation rate and number of live embryos at d 28 of gestation were recorded. Treatment had no effect ($P > 0.05$) on any parameters measured. Duration of estrus classified as less or greater than the overall mean also had no effect ($P > 0.05$) on any of the parameters measured. Results indicate that treatment did not advance ovulation nor did it improve reproductive performance in sows. Overall, a negative correlation of WEI with the ovulation rate ($P = 0.0005$, $r = -0.54$) was established.

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Keywords: PGF₂α, ovulation, WEI

INTRODUCTION

Soede et al. (8) have shown that ovulation occurs at approximately $72 \pm 15\%$ of estrus duration in sows and optimal fertilization occurs when sows are bred between 0 and 24 hours before ovulation. Several reports have shown that insemination using boar seminal plasma is effective in advancing ovulation in sows. This effect was much more pronounced for sows exhibiting a long interval from estrus to ovulation (11,12). Estrogens present in boar seminal plasma cause the release of prostaglandins from the endometrium (1,2). There is also evidence

Acknowledgements

The authors gratefully acknowledge Janes Gollar, Artur Ceglieski, and Shirley Shostak for their help, Pig Improvement Ltd (Canada) for contributions to the provision of experimental animals, and Alberta Swine Genetics Corp. for the provision of semen.

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that prostaglandins may play a role in improving litter size and farrowing rate in herds with lower productivity, particularly herds with low conception rates and in which summer infertility and poor management are a problem (5,6). Peña et al. (5) showed that an intravulva injection of PGF₂alpha at insemination increased sow fertility. The mechanisms regulating these effects remain to be determined, but possibly include an improvement in sperm transport along the female reproductive tract, advancement of ovulation, or improvement in the fertilization of ova (5).

The experiment reported in this paper was designed to further elucidate these mechanisms, and the primary goals were to 1) to determine if PGF₂alpha is effective in improving ovulation rate and conception rate in a high producing herd, and 2) to determine whether the positive effects of PGF₂alpha treatment on sow fertility were mediated by an advance in the time of ovulation.

Additionally, the data accumulated in this trial was used to explore relationships between post-weaning characteristics and subsequent sow reproductive performance. Steverink et al. (9) have shown that litter size and farrowing rate are greatest when the WEI is between 2 and 4 d, then decrease when WEI increases from 5 to 7 d. Furthermore, several authors have reported a negative relationship between WEI and duration of estrus (8,9). Because these relationships exist, it was relevant to examine them as potentially confounding factors in this experiment.

MATERIALS AND METHODS

This study was completed at the University of Alberta Swine Research Unit and with approval by the Faculty Animal Policy and Welfare Committee. Forty-seven multiparous PIC (Pig Improvement Canada) Camborough sows were used in the study and weaned between 20 and 29 d of lactation. After weaning, sows were housed individually in the breeding room. From Day 3 after weaning, sows were allowed 10 min of fence-line contact twice a day (at 0800 and 2000) with one of two mature boars. The onset of standing heat was recorded as half the time between the time the sow first exhibited the standing reflex to the back pressure test by a technician and the time at the previous heat check at which the sow was not standing, and was in the presence of a boar. Sows continued to be heat checked until the end of standing heat to determine duration of estrus. The end of estrus was determined as half the time between the last time standing estrus was detected and the first time estrus was not detected. If sows did not show standing heat by Day 10 after weaning, they were removed from the trial. Sows were weighed and P2 backfat depth (Scanoprobe II, Scano, Ithaca, New York) was measured at farrowing and weaning. Sow weight was also recorded at standing heat. At the onset of standing heat, sows were assigned to treatment based on parity and weight at weaning. To test whether PGF₂alpha effectively advanced ovulation, sows received 0.5 mL of a PGF₂alpha analogue (7.5 mg/mL luprostiol, Prosolvin, Intervet, Boxmeer, Holland), which was given at the onset of standing heat (PGF), or served as a non-injected control (CON). The vulva was first cleaned and PGF injected into the external vulva at the junction of the vulva with the skin. To minimize trauma, a half-inch (12 mm) 20 gauge needle was used and directed medially and 20° to 30° cranially. From start of weaning until slaughter, sows were fed a gestation diet ad libitum that contained 13.4 percent crude protein, 3.1 Mcal/kg digestible energy, and 0.6 percent lysine. At breeding, sow feed was reduced to 2.0 kg/d. All sows had ad libitum access to water.

Twelve and 24 h after the onset of standing heat sows were inseminated using pooled Dalland semen at a dose of 3.0×10^9 morphologically normal sperm (Alberta Swine Genetics Cooperation, Leduc, Alberta, Canada) per insemination. Semen was no older than 3 d from the collection date. Sows were allowed fenceline contact with 1 of 2 boars during insemination.

Transcutaneous ultrasound of the right ovary using a Pie Medical Scanner 200, model 41480 (Can Medical, Kingston, Ontario, Canada) with a 5.0 to 7.5 MHz multiple scan transducer was performed every 6 h beginning 24 h after the onset of estrus, until ovulation was detected. The time of ovulation was recorded as half the time between the last detection of follicles and their subsequent disappearance. At 36, 54, and 72 h after the onset of standing heat a 2.5 mL sample of blood was taken from an ear vein. For the majority of occasions, the blood sample was taken without restraint while the sows were exhibiting the standing reflex; however, when necessary a brief period of nose-snare restraint was used to collect the blood sample. Previous experience from our laboratory has determined that the stress associated with nose-snare restraint does not influence progesterone concentrations. These samples were subsequently analyzed for progesterone. After breeding, sows were moved to individual stalls in the gestation room. Twenty-eight days after insemination, sows were transported to a local abattoir for slaughter and recovery of reproductive tracts. The number of corpora lutea on each ovary was counted to determine ovulation rate, and the uterus was dissected to determine total number of live embryos, chorioallantoic fluid volume, and embryo length.

Statistical Analysis

Differences between control and prostaglandin-treated sows for duration of estrus, ovulation rate, number of viable embryos and embryonic survival at Day 30 of gestation were analyzed using the SAS GLM procedure (7). For all these variables, treatment and WEI were included in the model. Lactation length was used as a covariate since it varied between sows. Because not all sows were slaughtered at Day 30, due to restrictions at the abattoir, day was used as a covariate for chorioallantoic fluid volume and embryo length. To determine whether different durations of estrus affected the response to treatment, sows were grouped on the basis of a mean duration of estrus less (Group 1) or greater than (Group 2) the overall mean. As progesterone concentration data were not normally distributed, the data were log-transformed prior to analysis. Data were examined using repeated measures analysis of variance (7) to determine overall treatment, group, and the interaction effects and the treatment and group over time by the use of orthogonal contrasts. Differences between groups for progesterone concentration, ovulation rate, number of live embryos and embryo survival were analyzed using the SAS GLM procedure (7).

RESULTS

Of the 47 sows that were allocated to treatment at weaning, seven were removed from the trial: five sows did not return to estrus within 10 d of weaning, one sow was lame, and one sow was detected as non-pregnant at Day 30 of gestation. Data from these animals were not included in the analysis. The remaining 40 sows were divided evenly among treatment and the stratified allocation to treatment resulted in no difference in parity (mean \pm SD) (4.3 ± 1.9 vs 3.8 ± 1.8) or weaning weight (250.1 ± 36.0 vs 240.1 ± 36.3) between CON and PGF sows.

The effect of an intravulval injection of PGF at the detection of estrus on the duration of estrus, ovulation rate, number of live embryos and embryonic survival are shown in Table 1. Treatment had no effect ($P > 0.05$) on any of these parameters. Due to an accident involving the probe of the ultrasound machine, and due to the fact we were unable to replace the probe before the completion of the trial, ultrasound data were only obtained for 13 sows on trial. However, the results obtained for the 13 sows were not compromised by the accident and the results are considered accurate.

Table 1. Conception rate, ovulation rate, live embryos, embryonic survival, estrus to ovulation interval and the percentage of ovulation of estrus duration (mean \pm SD) for sows grouped by treatment.

	CON (n = 20)		PGF (n=20)		P
	Mean	SD	Mean	SD	
Duration of estrus (h)	58.8	16.0	63.0	15.5	0.53
Ovulation rate (n)	21.1	4.3	21.6	4.0	0.72
Total Live embryos (n)	14.5	4.6	14.3	5.5	0.88
Embryonic survival (%)	68.2	16.1	66.2	21.8	0.86
Embryo length (mm)	23.0	3.9	23.4	3.6	0.71
Chorioallantoic fluid volume (mL)	166.5	72.0	172.6	60.8	0.65
Estrus to ovulation interval (h)	43.0 ^a	4.9	46.3 ^b	6.9	0.52
Ovulation / estrus duration (%)	78.3 ^a	13.5	81.9 ^b	13.0	0.88
Conception rate (%)	100	-	95	-	0.32

^a n=6

^b n=7

Table 2 shows ovulation rate, number of live embryos and embryonic survival at Day 30 of gestation for sows with a duration of estrus equal to or less than 60 h (Group 1; average = 49.1 ± 9.7 h) and greater than 60 h (Group 2; average = 75.3 ± 6.9 h). Group had no effect on any of these parameters ($P > 0.05$). Also, the treatment x group interaction was not significant, indicating that PGF2alpha was not effective in increasing ovulation rate, number of live embryos or embryo survival even in sows with a short or long duration of estrus.

The overall progesterone concentration in CON and PGF gilts 36, 54 and 72 h after the detection of the onset of estrus (Figure 1a) was not different between PGF and CON sows ($P = 0.20$) and concentrations at 36 h after onset of estrus differed between treatments ($P < 0.05$). For

Table 2. Number of live embryos and embryonic survival of gilts with a short (Group 1) and long (Group 2) duration of estrus

	Group 1		Group 2		P	
	Mean	SD	Mean	SD	Group	Trt x group
Ovulation rate (n)	20.4	4.2	22.3	4.5	0.20	0.76
Live embryos (n)	14.1	14.8	14.8	5.4	0.70	0.80
Embryo survival (%)	68.5	19.0	65.9	19.5	0.69	0.68
Embryo length (mm)	22.0	4.2	24.8	2.3	0.29	0.54
Chorioallantoic fluid volume (mL)	153.2	68.5	189.5	56.7	0.65	0.69
Estrus to ovulation interval (h)	21.7	5.0	24.1	3.5	0.81	0.18

sows grouped on the basis of duration of estrus, the relationship between progesterone concentration and time is shown in Figure 1b, and overall, progesterone concentrations differed among groups (mean \pm SD), Group 1: 0.60 ± 0.42 ; Group 2: -0.017 ± 0.50 ; $P = 0.0006$.

WEI was negatively associated with ovulation rate (Figure 2a), number of live embryos (Figure 2b) and duration of estrus (Figure 2c). One sow was considered as an outlier and removed from the analysis because her WEI was greater than two standard deviations from the mean. However, even when including data from this sow the same relationships existed: ovulation Rate = $30.5 - 0.10$ WEI (h), $r = -0.38$, $P = 0.02$; duration of estrus = $888.7 - 0.29$ WEI (h), $r = -0.32$, $P = 0.05$; and embryo survival = 0.84 (WEI) -0.0017 ; $r = -0.16$; $P = 0.34$.

DISCUSSION

Our study shows that PGF2alpha given at the onset of standing heat had no effect on ovulation rate, number of live embryos, embryo survival, chorioallantoic fluid volume or embryo length at d 30, or conception rate. Peña et al. (5) reported that a PGF2alpha injection at insemination improved farrowing rate (78.5 vs 54.4) and litter size (10.8 vs 9.1) in sows during the low fertility summer season, and addition of PGF2alpha to semen doses improved conception rate and litter size (7,11). However, in these studies farrowing rates (54.4%) and conception rates (75%) in control sows were low and the 98% conception rate in control sows in our study virtually removed any possibility of demonstrating the efficacy of PGF2alpha treatment.

Even though we did not have ultrasound data to identify when ovulation occurred in all sows, we believe that the difference in progesterone concentration at 36 h after the detection of onset of estrus between treatments was probably related to variation in time of ovulation and estrus duration. Neither our ultrasound data, nor the progesterone data, are consistent with the hypothesis that PGFa treatment advanced ovulation.

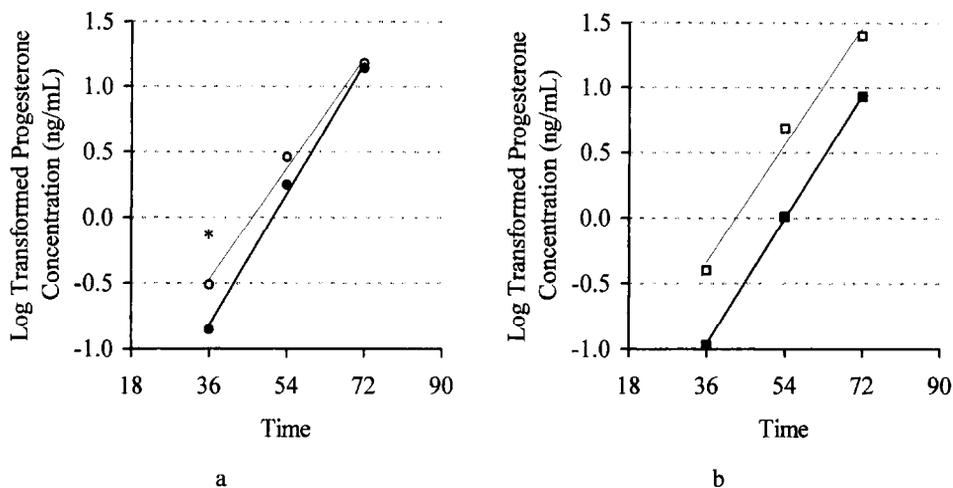


Figure 1. a) Progesterone concentrations at 36, 54 and 72 hours after the onset of estrus (time) in CON (closed circles) and PGF (open circles) sows. * $P < 0.05$ for differences between treatment within time of sampling. b) Progesterone concentrations in Group1 and Group2 sows over time; Group1 (open squares) Group2 (closed squares).

Waberski et al. (11) has reported that ovulation inducing factors may be more beneficial for sows with a longer duration of estrus. It would be predicted that sows with a longer duration of estrus would ovulate relatively later than sows with a short duration of estrus, thus explaining the difference in progesterone concentration between Group1 and Group2 sows. Soede et al.(8) have shown that optimal fertilization occurs 0 to 24 h prior to ovulation and that the percentage of sows with 90% normal embryos decreases with intervals greater than 24 h before ovulation. Therefore, insemination at fixed times after the detection of the onset of estrus may result in sows with a long duration of estrus being inappropriately inseminated. Thus, to determine whether PGF2alpha was effective in advancing ovulation, treatment was given at the onset of standing estrus. If PGF2alpha was effective in advancing ovulation in a long duration of estrus, we would have predicted that PGF sows would have an improved number of live embryos and embryo survival compared to CON sows, owing to the fact more sows would fall into the optimal period for insemination. However, when the effectiveness of a PGF2alpha injection was determined for sows grouped as having a longer duration of estrus, our results still indicate that prostaglandin was ineffective in improving ovulation rate, number of live embryos or embryo survival.

In the context of differences to previous studies, it is possible that the positive effects reported by Peña et al. (5) were not due to advanced ovulation but to enhanced sperm transport in the female reproductive tract. The experiment of Peña et al. (5) differed from our own in three important elements: 1) there was no boar stimulation at insemination, 2) sows were given a prostaglandin injection at insemination rather than the onset of estrus and, 3) summer infertility

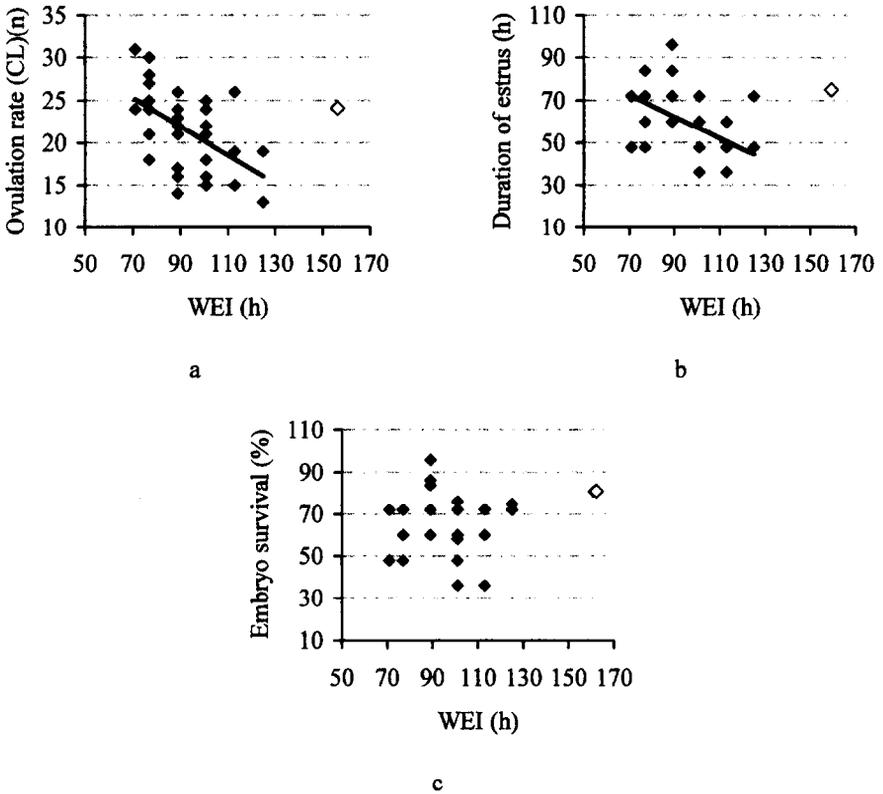


Figure 2. Relationships of WEI and different reproductive characteristics: a) Ovulation Rate = $37.12 - 0.17 (\text{WEI})$; $r = -0.54$, $P = 0.0005$; b) Estrus duration = $113.3 - 0.56 (\text{WEI})$; $r = -0.49$, $P = 0.002$; c) Embryo survival = $0.84 - 0.0017 (\text{WEI})$; $r = -0.12$, $P = 0.44$. Closed diamonds represent data used in the analysis, open diamond was flagged as an outlier and removed from analysis.

was a problem. Boar stimulation results in a release of oxytocin that subsequently results in a rise of prostaglandin, both enhancing muscle contractions. Sows that do not receive boar stimulation will not profit from the beneficial effects of enhanced sperm transport and, thus, treatment with prostaglandin at the time of insemination may improve sperm transport in the female reproductive tract and improve fertility. Also, the farm on which our trial was undertaken was intensively managed, had few reproductive problems, and did not suffer from summer infertility. Under conditions of lower productivity PGF2alpha may have exerted an effect.

We cannot rule out the possibility that an injection of PGF2alpha had a negative effect on early corpus luteum activity. It is well known that PGF2alpha is involved in the regulation of corpus luteum function, acting to inhibit progesterone production and induce luteolysis (4).

However, pooled progesterone concentration, ovulation rate, number of live embryos, embryo survival, chorioallantoic fluid volume or embryo length were not different between treatment, which leads us to believe if early luteal function did differ between PGF and CON sows, the difference in progesterone concentration did not have long-term effects.

Litter size and farrowing rate have shown to be influenced by the day of return to estrus after weaning (5,12). Several authors have reported a negative relationship between WEI and duration of estrus (5,12) and our data confirms these results. In previously unpublished data, Steverink et al. (12) reported that ovulation rate decreased from 21.6 to 19.7 oocytes when WEI increased from 3 to 6 d. Because no relationship between WEI and embryo survival ($r = -0.12$, $P = 0.44$) was detected, our data provide the first published evidence demonstrating that lower ovulation rate ($r = -0.54$, $P = 0.0005$) associated with increasing WEI probably contributes to lower litter size.

In conclusion, we found no benefit of a PGF $_{2\alpha}$ injection in sows with already acceptable reproductive performance, suggesting that the PGF $_{2\alpha}$ injection may only be a useful breeding tool on farms where summer infertility or poor management strategies are a problem. PGF $_{2\alpha}$ did not affect the overall progesterone concentration and thus did not appear to effect the time of ovulation of sows. A negative relationship existed between increasing WEI with ovulation rate and no relationship existed between increasing WEI with embryo survival at d 30, thus providing a mechanism to explain why a decrease in WEI is associated with lower litter size.

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