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*Article*

# Use of a Vaginally Administered Gel Containing the GnRH Agonist Triptorelin and Fixed Time and Single Artificial Insemination of Pigs under Commercial Conditions. Productive and Economic Impact

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**Abstract:** Fixed-time artificial insemination is an important technique in swine production that can improve reproductive efficiency and meat production quality by making better use of the genetic potential of breeding males and reducing the costs associated with double or multiple inseminations. Our goal was to evaluate the vaginal application of the GnRH agonist triptorelin acetate to synchronize post-weaning sows and facilitate implantation of a single fixed-time insemination. In a first experiment, the efficacy of treatment with the triptorelin in animals with or without signs of estrus was analyzed using a single insemination at a fixed time compared to the control following the standard insemination strategy. Farrowing rate was comparable between triptorelin and control groups (100 vs. 87.50%), but triptorelin without estrus had a lower rate (50%). Litter size did not differ between groups. Estradiol and progesterone levels at 96- and 120-hours post-weaning were similar in the control and triptorelin groups ( $P>0.05$ ). These results suggest that Triptorelin has the potential to synchronize ovulation in pigs without affecting post-weaning hormonal profiles. In a second experiment, the objective was to evaluate the productive and economic impact of implementing a treatment with triptorelin acetate 96 hours after weaning compared to the standard insemination protocol. Sows were grouped according to treatment (control vs. triptorelin) and estrus onset ( $\leq 5$  days and  $>5$  days after weaning considered as late estrus). Farrowing rate was lower in the control late estrus group than in the control and triptorelin groups and similar to the triptorelin late estrus group. No differences were found for litter size, live or dead piglets born ( $P>0.05$ ). We developed an estimation model to assess the cost/benefit of intravaginal triptorelin administration at 96 h post-weaning. The overall result is that the use of triptorelin increases the benefit per inseminated sow in the range of €15-20. This improvement is mainly related to an increase in the reproductive performance of the treated sows compared to the control sows and the reduction in the number of inseminations per sow. These results highlight the potential of triptorelin to optimize reproductive management in pigs, improving efficiency and economic viability.

**Keywords:** porcine reproduction; fertility; reproductive management; artificial insemination; reproductive outcomes

## 1. Introduction

Fixed-time artificial insemination is an important technique in swine production that can improve reproductive efficiency and meat production quality by making better use of the genetic potential of breeding males and reducing the costs associated with double or multiple inseminations [1].

An important step in the implementation of fix-time artificial insemination is the correct synchronization between ovulation and insemination time. In pigs, there is a high variability in the time between weaning and/or onset of estrus and ovulation [2–4]. Various hormonal protocols, including those using GnRH analogues, have been used to synchronize the timing of ovulation (reviewed in [1,5]).

Triptorelin is a GnRH analog that binds to receptors in the pituitary gland and stimulates the secretion of the hormones LH and FSH. This compound was first studied by Dr. Andrew Schally [6], who was awarded the 1977 Nobel Prize in Medicine “for their discoveries concerning the peptide hormone production of the brain” [7]. In the porcine species, several studies have evaluated the vaginal application of triptorelin acetate to synchronize post-weaning sows and facilitate implantation of a single fixed-time insemination [8,9]. Preliminary studies on the use of triptorelin defined the gel viscosity conditions for the vaginal administration [8] and determined that the dose of 200 µg triptorelin administered 96 h after weaning was the most effective treatment for inducing the ovulation [10]. A single AI given 22–26 h after the triptorelin treatment provides the best results [11], with ovulation occurring 40–48 h after treatment [8,11].

Based on this previous information, our goal was to evaluate the applicability of this method on a large scale under commercial conditions and to study it in a large number of animals. In a first experiment, the aim was to analyze the efficacy of the treatment with the GnRH agonist triptorelin in animals with or without signs of estrus with a single insemination at a fixed time compared to the control following the standard insemination strategy. A second experiment was designed to evaluate the effect of the day of estrus on the reproductive and economic effects of triptorelin compared to the standard insemination protocol. A third section was developed to estimate the economic and productive impact of applying this methodology.

## 2. Material and Methods

### *Animals*

The study was conducted on a commercial farm with 750 sows located in the Region of Murcia, in southeastern Spain. It is a breed-to-wean farm with a herd (Landrace x Large White) crossed with semen doses from Duroc boars. In this farm, after weaning, the sows were housed in individual crates with slatted floors for estrus detection and insemination until the day 28 of gestation, when they were in pens in groups of 24 sows. Approximately 1 week before the expected farrowing date, the sows were moved to farrowing rooms in individual crates. Information on parity and duration of lactation was recorded for each sow, and sows with parity 2–8 were maintained in lactation for an average of 26 days.

One group of sows was treated with an intravaginal application of gel containing the GnRH agonist triptorelin (200 µg triptorelin) (2 ml OvuGel®, Vetoquinol, Madrid, Spain) 96 h after weaning (TRIPTORELIN group), while other sows received no treatment (CONTROL group).

This study was developed in accordance with the Spanish Animal Protection Policy (RD 53/2013), which is in line with the European Union Directive 2010/63/EU on the protection of animals used in scientific experiments. This project was approved by the Ethics Committee of the University of Murcia (Project 34071).

### *Oestrus Detection and Insemination*

Oestrus signs were assessed, and data recorded daily by trained personnel. A boar was placed in the aisle in front of the sows. A back pressure test was performed on each sow to determine if the immobilization response occurred. The first day of expression of the estrus sign was recorded, and the day and number of inseminations were used to calculate the interval between weaning and insemination (days).

All sows were inseminated with semen from the same group of boars. The insemination dose consisted of  $2.5 \times 10^9$  total spermatozoa in a total volume of 60 mL and was applied by experienced

technicians using a post-cervical artificial insemination catheter. The insemination protocol was adapted for each experiment, as described in the experimental design section.

At 22-25 days after the first insemination, a pregnancy detection ultrasound scan was performed to calculate the fertility rate and subsequently the farrowing rate and litter size by recording the number of piglets born, total, dead and alive.

*Hormone Analyses*

Plasma levels of progesterone (P4. ng/mL) and estradiol (E pg/mL) were evaluated in sows 96 hours after weaning and showing signs of oestrus, and 24 hours later. Blood samples were collected by direct venipuncture of the jugular vein using lithium heparin collection tubes (BD Vacutainer®, BD, Spain) and centrifuged at 1500 g for 15 min. Plasma was immediately separated and stored at -20 °C until hormone quantification.

Estradiol and progesterone were quantified using an electrochemiluminescence immunoassay (Elecsys® Estradiol III and Progesterone Assays), using a Cobas e 411 analyzer (Roche Diagnostics, Barcelona, Spain).

*Experimental Design*

Experiment 1. Use of Triptorelin in Sows 96 Hours after Weaning with Sign of Estrus

A group of 55 sows (parity 2-8) were weaned. Signs of estrus were checked 96 hours after weaning, 33 of them in open estrus were treated with triptorelin and inseminated with a single dose of semen 22-24 hours after treatment (TRIPTORELIN group). On the other hand, 16 sows in open estrus did not receive any treatment and were inseminated as usual on the farm with double insemination at 0 and 24 hours (Control group). Finally, a small group of 6 sows without signs of estrus was treated with triptorelin and inseminated at 22-24 hours after the treatment without estrus signs (TRIPTORELIN NO ESTROUS) (Table 1).

Plasma levels of progesterone (P4. ng/mL) and estradiol (E pg/mL) were evaluated on the day of estrus detection (96 hours after weaning) and 24 hours later.

For each experimental group, parity, lactation length, gestation length, farrowing rate, total piglets born, live and stillborn piglets per litter were evaluated.

**Table 1.** Experimental design experience 1. Treatment of the sows after weaning in the different experimental groups.

Group	N	Estrous 96 h post weaning	Triptorelin 96 h Post weaning	AI 96 h post weaning	Estrous 120 h post weaning	AI 120 h post weaning
CONTROL	16	+	-	+	+	+
TRIPTORELIN	33	+	+	-	+	+
TRIPTORELIN NO ESTRUS	6	-	+	-	-	+

Experiment 2. Effect of Day of the Onset of the Estrus on Reproductive and Economic Impact of Triptorelin Treatment

A total of 857 sows (parity 2-8) were used in this study. One group of sows (n=415) was treated vaginally with TRIPTORELIN 96 hours after weaning (TRIPTORELIN group), while 442 sows received no treatment (CONTROL group). The sows in each group were divided into two subgroups according to the onset of estrus. Triptorelin treated sows that showed signs of estrus before 120 hours after weaning were inseminated with a single dose of semen at 120 hours post-weaning (n=418). Those sows that showed signs of estrus later were inseminated at 0 and 24 hours after the onset of estrus and formed the late estrus triptorelin group (n=24).

All control sows were inseminated with double doses at 0 and 24 hours after onset of estrus, or with an additional semen dose at 48 hours if they were in estrus as usual on the farm. The control sows were divided into two subgroups of 390 and 25 sows, respectively, according to the day of onset of estrus, before or after 120 hours after weaning.

**Table 2.** Experimental design experiment 2. Treatment of the sows after weaning in the different experimental groups.

Group	N	Triptorelin 96 h after weaning	Estrus onset before 120 h after weaning	Artificial insemination
Control	390	no	yes	2-3 (0 and 24 h, 0,24,48 h) after onset sign estrus
Control late estrus	25	no	later	2-3 (0 and 24 h, 0,24,48 h) after onset sign estrus
Triptorelin	418	yes	yes	1 (24 h) with sign of estrus
Triptorelin late estrus	24	yes	later	2 (0.24 h) after onset sign estrus

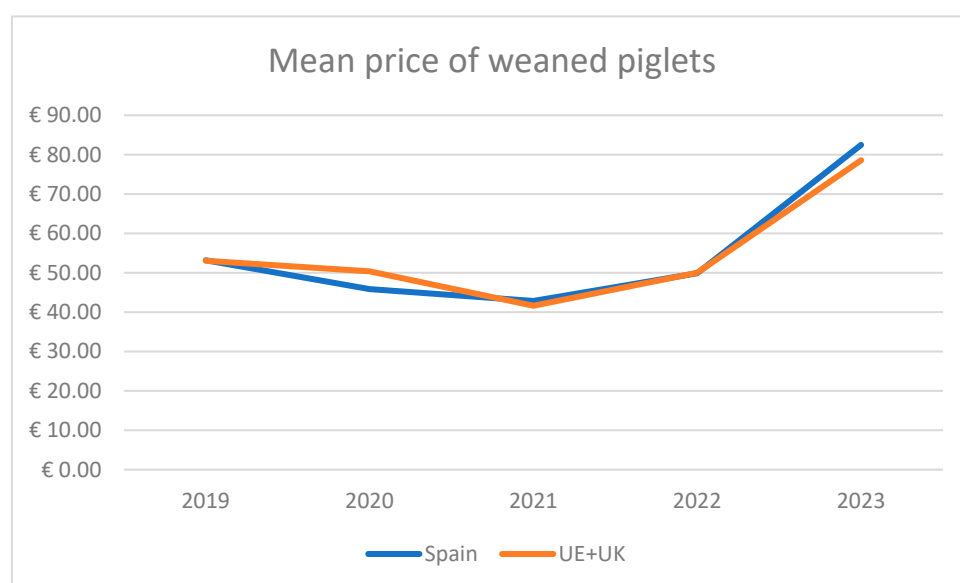
For each experimental group, parity, lactation length, days after weaning to estrus onset, weaning insemination interval (days), number of inseminations per sow, pregnancy rate, gestation length, farrowing rate, day of week of farrowing, total piglets born, live and stillborn piglets per litter were evaluated.

Experiment 3. Economic Impact Estimation

A cost-benefit model was developed based on the reproductive data from experiment 2 (farrowing rate, live piglets born, number of AI) for each experimental group. The mean number of viable piglets born per inseminated sow (farrowing rate \* live piglets born) was calculated. To estimate the number of piglets that would complete the growing phase and produce 20 kg commercial piglets, a mortality rate of 14% during lactation and 7% mortality rate after weaning was used for all groups. Although the mortality rate during lactation ranges from 10-25% of live born piglets according to different studies [12–14], and the mortality rate after weaning ranges from 4.1 to 7.5% (reviewed by [15]).

To calculate the income from the sale of 20 kg commercial piglets, it is necessary to evaluate the prices in the EU in the last 5 years. According to the Directorate-General for Agriculture and Rural Development (European Commission), the prices for piglets in Spain and EU+UK were as shown in Figure 1.





**Figure 1.** Prices for 20 kg piglets in Spain and EU+UK. Directorate-General for Agriculture and Rural Development (European Commission),.

#### *Calculation of AI Costs*

In calculating the costs associated with AI, we must first consider the cost of heat detection in the sow, which includes the cost of personnel and the use of a boar. In this case, the costs associated with the heat detection process are the same for all groups because, according to Experiment 1, it is necessary to check for estrus in both the triptorelin and control groups.

The cost of the insemination itself includes the cost of the personnel applying the dose of semen, the cost of the dose of semen and the cost of the material necessary for its application: cleaning of the sow, insemination catheter and additional material. In this sense, different authors have estimated these costs under different scenarios, so [16] estimated the cost of AI at \$10.93/insemination, including the sperm dose (\$7) and the cost of the procedure (operator time, catheter and other materials (\$3.93). While [17] calculated the cost at \$7.17/insemination, with seminal dose cost of \$6.00, catheter \$0.17, average labor cost \$1.00 per sow.

In our production conditions in Europe, the average cost of the semen dose was 4-6 €/dose, and the costs associated with insemination mainly depend on the cost of labor, which in Europe ranged from 15.5 €/hour in Spain to 27.9 €/hour in the Netherlands [18]. The time required for insemination is close to 5 min for cervical insemination and 2.5 min for post-cervical insemination [19], although in our conditions the estimated time for post-cervical insemination is closer to 1.5 min. While the cost of the catheter ranges from €0.15 for cervical insemination to €0.60 for post-cervical insemination [20]. Considering all the previous information, to explore the application under different scenarios, we will simulate the cost-benefit of applying the time-fixed protocol when the total cost per insemination is 6, 8, 10 and/or 12 €. These different insemination costs will cover all possible situations in European pig production.

On the other hand, we calculated the cost of application of triptorelin that includes the cost of the compound (5-5.50 €/sow) and the time to clean the vulva and apply the treatment (0.5-1 minute/sow, 15.5-27.9 €/hour) with an estimated cost in the range of 0.129-0.465 €/sow. Thus, the total cost of the treatment could range from 5.13€ to 5.97€ per sow according to the different scenarios. To facilitate the estimation of the economic impact we have used a mean cost of 5.5 € per sow.

#### *Statistical Analysis*

Results are expressed as mean  $\pm$  SEM. Normality of samples was assessed by the Shapiro-Wilk or Kolmogorov-Smirnov test, depending on the sample size. Because the parameters did not follow

a normal distribution, they were analyzed using the nonparametric Kruskal-Wallis test, which is a nonparametric analog of a one-factor analysis of variance with treatment groups as the main variable. When the Kruskal-Wallis test showed a significant effect, all pairwise multiple comparisons were evaluated. Differences were considered statistically significant at  $p < 0.05$ . Data were processed using IBM SPSS software (version 28.0.1.1), and graphs of the data were generated using Orange data mining software (version 3.36.2).

### 3. Results

#### *Experiment 1. Use of Triptorelin in Sows 96 Hours after Weaning with Sign of Oestrus*

The experimental groups were homogeneous in terms of previous lactation duration (Table 3,  $P=0.664$ ). Regarding the mean parity, no differences were found between the TRIPTORELIN and CONTROL groups, but the TRIPTOLERIN without estrus group was lower than the others (Table 4,  $P<0.05$ ). Farrowing was similar between the triptorelin and control groups (100 vs. 87.50%), but triptorelin without estrous had a lower farrowing rate (50%,  $P<0.001$ ). There was no difference in litter size and gestation length between the 3 groups (Table 4.  $P>0.05$ ).

**Table 3.** Characteristics of sows in the control and triptorelin groups. Parity and previous lactation length in days. Mean $\pm$ sem (n).

	n	Parity	Previous lactation Length
CONTROL	16	4,13 $\pm$ 0.26 <sup>a</sup>	27,31 $\pm$ 1.23
TRIPTORELIN	33	3,88 $\pm$ 0.19 <sup>a</sup>	27,61 $\pm$ 1.25
TRIPTORELIN NO ESTRUS	6	3,00 $\pm$ 0.26 <sup>b</sup>	25,67 $\pm$ 1.76
P value		0.062	0.664

**Table 4.** Characteristics of sows in the control and triptorelin groups. Mean $\pm$ sem (n).

	n	Farrowing rate (%)	Gestation length (days)	Live born	Dead born	Total piglets born
CONTROL	14	87.50 $\pm$ 8.54 <sup>a</sup>	117,14 $\pm$ 0,29	18.07 $\pm$ 0.68	1,14 $\pm$ 0.25	19,21 $\pm$ 0.67
TRIPTORELIN	33	100 <sup>a</sup>	116. $\pm$ 30 $\pm$ 0.26	16.30 $\pm$ 0.75	1,91 $\pm$ 0.52	18,21 $\pm$ 0.69
TRIPTORELIN NO ESTRUS	3	50 $\pm$ 22.36 <sup>b</sup>	117 $\pm$ 1.73	14.33 $\pm$ 1.86	1,67 $\pm$ 1.67	16,00 $\pm$ 1.53
P value		<0.001	0.220	0.195	0.777	0.269

At 96 hours and 24 hours after weaning, estradiol and progesterone levels were similar between the CONTROL and TRIPTORELIN groups. In both groups, estradiol levels decreased, and blood progesterone levels increased 24 hours after the onset of estrus, resulting in a decrease in the estradiol/progesterone ratio with a similar pattern (Table 5,  $P>0.05$ ).

**Table 5.** Estrogen (pg/ml) and Progesterone(ng/mL) in blood serum from sows 96 and 120 hours after weaning treated with (TRIPTORELIN) or not with Treptorelin acetate (CONTROL). Mean $\pm$ sem.

	Estrogen 96h after weaning	Estrogen 120 h after weaning	Progesterone 96h after weaning	Progesterone 120 h after weaning	Ratio E/P 96h after weaning	Ratio E/P 120 h after weaning
Control (n=15)	21.06 $\pm$ 3.38	8.81 $\pm$ 1.77	0.16 $\pm$ 0.03	0.26 $\pm$ 0.06	161.95 $\pm$ 25.53	97.98 $\pm$ 39.63

TRIPTORELIN (n=14)	28.12±4.17	7.09±1.90	0.22±0.06	0.33±0.09	206.83±49.33	98±12.6
p-value	0.278	0.531	0.306	0.486	0.417	0.207

Experiment 2. Effect of Day of Estrus Onset on Reproductive and Economic Impact of Triptorelin Treatment

The experimental groups were homogeneous in terms of previous lactation length (Table 6, P=0.190). No differences were found between the TRIPTORELIN and CONTROL groups with respect to mean parity, but the TRIPTORELIN late estrus group was lower than the previous groups and similar to the late estrus control group (Table 6, P=0.009). The late estrus groups contained a higher proportion of sows with parity 2 (triptorelin late estrus 37.5%, control late estrus 24%) compared to the groups with estrus before 120h after weaning (control 11.8% and triptorelin 14.8%) (chi-square p=0.04), these last groups showed a more homogeneous distribution of sows in parity 2-8 (Figure 2, violin plots, P<0.04).

Table 6. Characteristics of sows in the control and triptorelin groups. Mean±sem.

	Parity (n)	Previous lactation length (n)
CONTROL	5.21±0.10 <sup>a</sup> (390)	25.96±0.33 (389)
CONTROL late estrus	4.80±0.47 <sup>ab</sup> (25)	26.56±1.03 (25)
TRIPTORELIN	5.11±0.10 <sup>a</sup> (418)	26.11±0.25 (418)
TRIPTORELIN late estrus	3.79±0.39 <sup>b</sup> (24)	27.63±1.14 (24)
P value	0.009	0.190

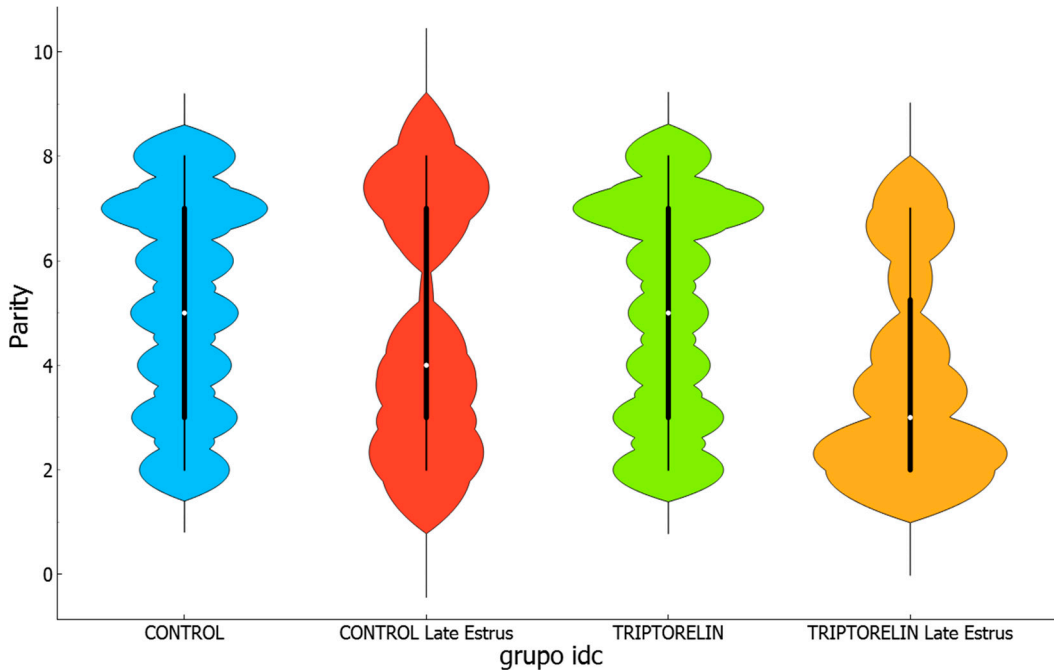


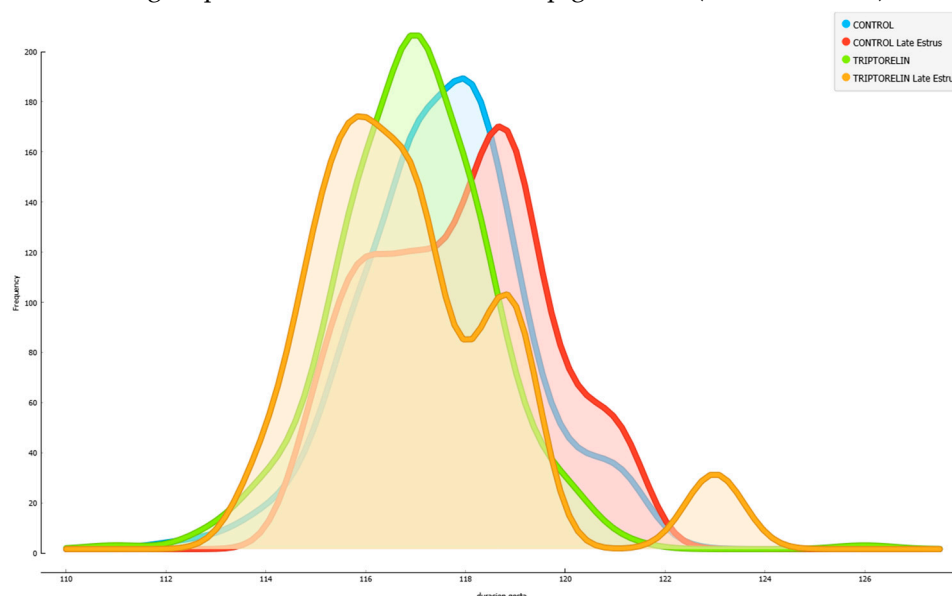
Figure 2. Violin plot of asymmetric distribution of parity of sows from different experimental groups. Chi-square P=0.04.

The day of estrus onset was similar in the TRIPTORELIN and control groups but was significantly higher in the control late estrus group and even higher in the TRIPTORELIN late estrus group (Table 7, P<0.001). According to the different insemination protocol per group, the interval between weaning and first insemination and the number of inseminations were significantly different (Table 7, P<0.001). The weaning-to-first insemination interval was shorter in the control group than in the triptorelin group, and both were shorter than in the control and triptorelin-late estrus groups (Table 7, P<0.001). In the control group (normal and late estrus), 85.8% (361/421) of the sows were



inseminated twice, while 14.3% (60/421) were inseminated for 3 days, so the mean number of inseminations per sow was 2.12-2.14. This implied the use of a higher number of semen doses than in the triptorelin groups, which used 1 and 2 AI per sow (Table 7,  $P<0.001$ ).

There were no differences in pregnancy rates (Table 7,  $P=0.296$ ). Gestation length was shorter in the Triptorelin group than in either control group, whereas Triptorelin late estrus was intermediate (Table 7 and Figure 3,  $P<0.001$ ). Farrowing rate was lower in the control late estrus group than in the control and triptorelin groups and similar to the triptorelin late estrus group (Table 7). No differences were found between groups for litter size, live or dead piglets born (Table 8,  $P>0.05$ ).



**Figure 3.** Kernel density distribution of the pregnancy length (in days) of sows from different experimental groups.

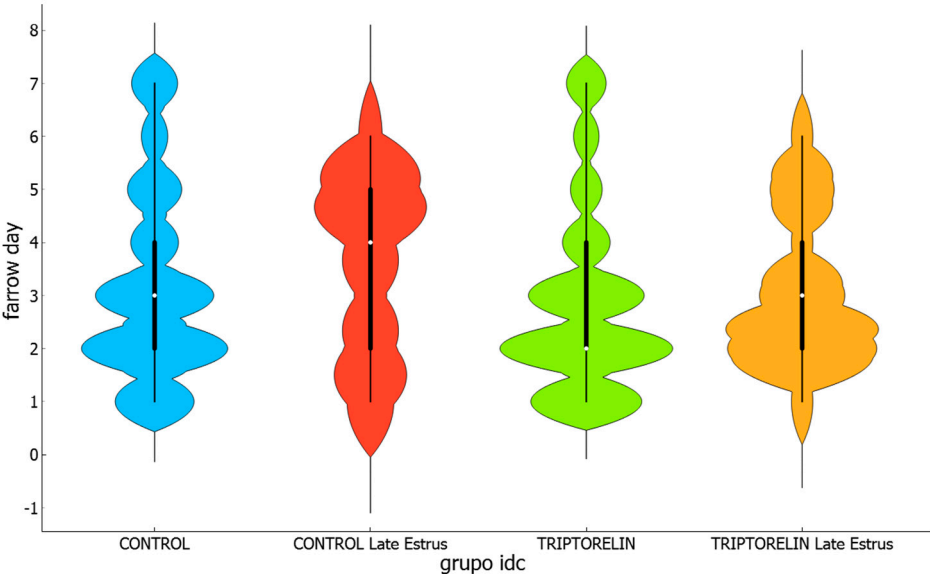
**Table 7.** Characteristics of sows in the control and triptorelin groups. Mean $\pm$ sem.

	Days start oestrus	Weaning insemination interval (days)	Number of AI	Pregnancy rate (%)	Gestation length	Farrowing rate
Control	4.50 $\pm$ 0.03 <sup>a</sup> (388)	4.50 $\pm$ 0.03 <sup>a</sup> (388)	2.14 $\pm$ 0.02 <sup>a</sup> (395)	92.04 $\pm$ 1.35 (402)	117.60 $\pm$ 0.09 <sup>a</sup> (315)	85.83 $\pm$ 1.82 (367) <sup>a</sup>
Control late estrus	6.31 $\pm$ 1.21 <sup>b</sup> (26)	6.31 $\pm$ 1.21 <sup>b</sup> (26)	2.12 $\pm$ 0.06 <sup>a</sup> (26)	88.46 $\pm$ 6.39 (26)	117.92 $\pm$ 0.51 <sup>a</sup> (12)	66.67 $\pm$ 11.43 (18) <sup>b</sup>
Triptorelin	4.52 $\pm$ 0.25 <sup>a</sup> (421)	5.00 <sup>c</sup> (421)	1.00 <sup>b</sup> (421)	93.16 $\pm$ 1.23 (424)	116.97 $\pm$ 0.08 <sup>b</sup> (341)	86.40 $\pm$ 1.72 (397) <sup>a</sup>
Triptorelin late estrus	8.09 $\pm$ 0.85 <sup>c</sup> (23)	8.09 $\pm$ 0.85 <sup>b</sup> (23)	2.00 <sup>a</sup> (24)	83.33 $\pm$ 7.77 (24)	117.00 $\pm$ 0.52 <sup>ab</sup> (17)	77.27 $\pm$ 9.14 (22) <sup>ab</sup>
P value	<0.001	<0.001	<0.001	0.296	<0.001	0.085

**Table 8.** Characteristics of sows in the control and triptorelin groups. Mean $\pm$ sem.

	n	Live born	Dead born	Total piglets born
CONTROL	315	15.42 $\pm$ 0.21	1.78 $\pm$ 0.08	17.19 $\pm$ 0.20
Control late	12	14.58 $\pm$ 1.41	2 $\pm$ 0.28	16.58 $\pm$ 1.61
TRIPTORELIN	341	15.60 $\pm$ 0.22	1.75 $\pm$ 0.07	17.36 $\pm$ 0.22
TRIPTORELIN late OESTRUS	17	14.65 $\pm$ 0.82	1.94 $\pm$ 0.33	16.59 $\pm$ 0.85
P value		0.602	0.543	0.717

The distribution of farrows during the days of the week was similar in all groups (chi-squared,  $P=0.107$ ). The proportion of farrows on the weekend was similar in the CONTROL (14%) and TRIPTORELIN (12%) groups, while this proportion was lower in the CONTROL late estrus (8.3%) and TRIPTORELIN late estrus (5.9%) groups (Figure 3). Interestingly, in the control group, 209/315 (66.35%) of the farrowing is concentrated in the first 3 days of the week, while in the triptorelin group this proportion increased to 73.47% (252/343, chi-square  $p < 0.05$ . Figure 3).



**Figure 4.** Violin plot of symmetric distribution of farrowing day (Monday 1, Sunday 7) of sows from different experimental groups. Chi-square  $P=0.107$ .

*Experiment 3. Economic IMPACT estimation*

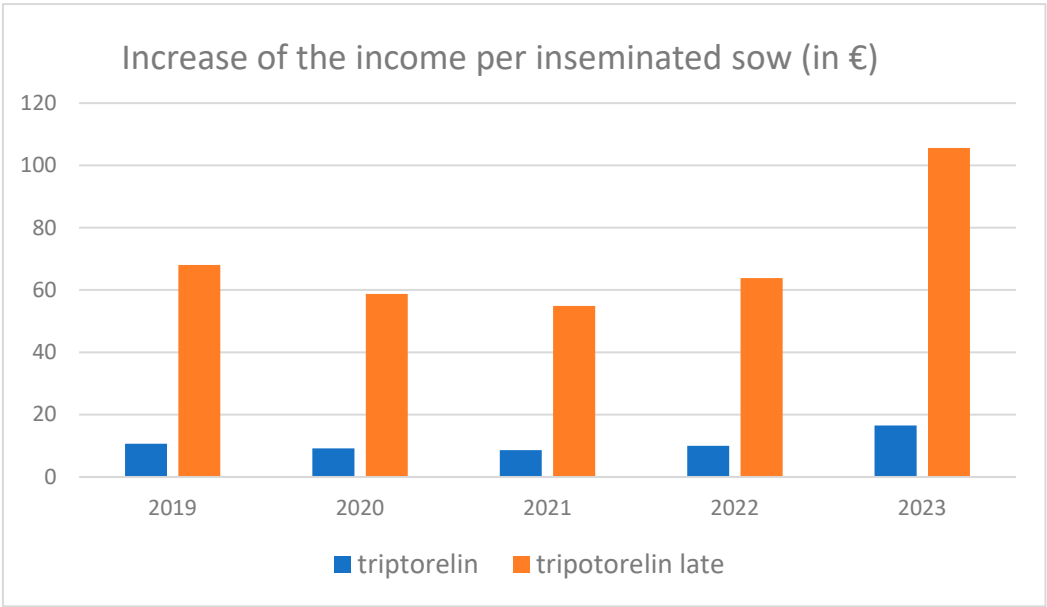
The estimated economic model created and based on the reproductive results of the use of triptorelin compared to control groups, showed that the use of triptorelin imply an increase in the number of commercial 20 kg piglets produced per inseminated sow estimated in 0.20 units when estrus is present before 5 days after weaning (10.58 vs 10.78; Table 9), or 1.28 units when estrus occurs after 5 days after weaning (7.77 vs 9.06; Table 9).

**Table 9.** Estimation of commercial 20 kg piglets produced per inseminated sow according to the treatment with or without triptorelin and the onset of the estrus.

	Farrowing rate (%)	Alive born	Alive born* farrowing g=Alive born/inseminated sow	Weaned/inseminated sow (86 % survival)	Commercial 20 kg piglets/inseminated sow (93% survival)
CONTROL	85,83	15,42	13,23	11,38	10,58
Control late estrus	66,67	14,58	9,72	8,36	7,77
TRIPTORELIN	86,4	15,6	13,48	11,59	10,78
TRIPTORELIN late estrus	77,27	14,65	11,32	9,74	9,06

Considering the prices for the sale of commercial 20 kg piglets in Spain during the period 2019-2023, the use of triptorelin was associated with an increase in income per inseminated sow, ranging

from €8.57 to €16.49 for sows in normal estrus (mean value € 10,97) and from €54.86 to €105.56 per inseminated sow for sows in late estrus (mean value € 70,20 (Figure 5).



**Figure 5.** Increase in the income from the sale of commercial 20 kg piglets derived from the use of triptorelin in comparison to control group, in sow with normal onset of estrus before 5 days or later.

If the cost of AI is estimated to be €6, €8, €10 or €12, and the estimated cost of the triptorelin treatment of 5.5 €/sow, the benefit of using triptorelin is a cost reduction ranged from 1.34 to 8.18 € per sow, when estrus onset was 5 days after weaning or before, and it was higher when the cost of insemination was higher. When the onset of the estrus was later than 5 days, the use of triptorelin increased the reproductive cost in a range of 4.06 to 4.78 € (Table 10)

**Table 10.** Estimation of the reproductive cost per sow treated with triptorelin in comparison to control group, in sow with normal onset of estrus or late estrus.

Group	No AI	Triptorelin cost €	Reproductive cost (6€/AI)	Reproductive cost (8€/AI)	Reproductive cost (10€/AI)	Reproductive cost (12€/AI)	Increase incomes
Control	2,14	0	12.84	17,12	21,40	25,68	
Control late estrus	2,12	0	12.72	16,96	21,20	25,44	
Triptorelin	1	5,5	11.5 (-1.34€)	13,5 (-3.62 €)	15,50 (-5.9 €)	17,5 (-8.18 €)	+10.97 €
Triptorelin late estrus	2	5,5	17.5 (+4.78€)	21,5 (+4.54 €)	25,50 (+4.30€)	29,5 (+4.06 €)	+70.20 €

When the two approaches are added together, increased incomes from the sale of piglets and reduction in th reproduction costs, the use of triptorelin in normal estrus represents an economic improvement in the range of €12.31-19.15 per treated sow, while in late estrus it represents a benefit of between €65.42 and €66.14 per treated sow. As the proportion of late estrus sows was 3.67% (12/327) in the control group and 4.75% (17/358) in the triptorelin group, the overall result is that the use of triptorelin increases the benefit per inseminated sow in the range of €15-20.

4. Discussion

In modern swine production, optimizing reproductive efficiency is a cornerstone of a sustainable and profitable business [21]. A key aspect of this optimization is to synchronize ovulation in sows to

ensure a more predictable breeding cycle and maximize resource utilization. Among the many methods available, the use of triptorelin, a potent synthetic analogue of gonadotropin-releasing hormone (GnRH), has emerged as a promising tool for synchronizing ovulation in pigs [9,22]. The strategic use of triptorelin could offer several advantages in swine reproduction. First, it facilitates tighter control of breeding schedules, allowing producers to better coordinate mating and farrowing times. Second, by synchronizing ovulation, triptorelin promotes more uniform litters. This results in improved litter size and consistent piglet quality. In addition, Triptorelin can help optimize the use of AI by ensuring that ovulation occurs within a defined timeframe, improving AI success rates and ultimately increasing genetic progress within swine herds.

In the first experiment, the efficacy of triptorelin application was tested in sows in estrus 96 hours after weaning and inseminated with a single dose 22-24 hours later. Reproductive results were similar to those obtained with the normal double insemination system. However, when triptorelin was administered to sows without obvious signs of estrus and a fixed-time insemination system was used, the results were very poor, limiting the use of a strict fixed-time insemination system for application to all sows, as previously reported by [23,24]. In this experiment, we confirmed that the application of the GnRH analog treatment induced a hormonal pattern similar to that of the control group, with a decrease in estradiol levels 24 hours after the onset of estrous signs and a slight increase in progesterone levels [8]. The importance of the timing of insemination is related to the need to maintain viable sperm in the fallopian tubes prior to ovulation [25]. This could be achieved by insemination 24 hours prior to ovulation, with an optimal time between 8-16 hours prior to ovulation as previously reported [26,27]. The use of GnRH analogues facilitates the synchronization of ovulation in most treated sows up to 40-48 h after treatment [8,11,24]. This means that a single insemination 22-24 h after the onset of estrus will be effective, as we observed in this study.

In the second experiment we evaluated the effect of day of onset of estrus on the reproductive and economic impact of triptorelin treatment. In these experiments, the animals that showed signs of estrus beyond day 5 after weaning, are characterized by being animals of a lower parity, especially the increase of this type of animals is significant in the animals of the second parity. These animals with delayed onset on estrus showed lower reproductive performances as previously was reported [3]. It would be necessary to monitor this type of animals and to take specific measures, such as changes in the duration and feeding in lactation, to allow a return to estrus in a better body condition, facilitating a better reproductive result.

In both experiments, no differences were observed in the farrowing rate (except for the late estrus control group), the number of live and dead pigs born, or the total number of pigs born per litter in the triptorelin and control groups. These results confirm the technical feasibility of GnRH agonist treatment as previously reported in several studies [8,28].

It is important to make efficient use of boars in AI protocols; this is achieved by reducing the number of sperm per insemination dose and increasing the number of semen doses per boar. This achievement maximizes the spread of genetic advances in the reproductive nucleus. [29]. At the same time, it is necessary to accurately evaluate the costs associated with both the reproductive strategies (labor time, materials and products) [30] and the reproductive outcomes [16,31], as well as possible additional benefits such as the reduction in the use of semen doses associated with a better use of the productive potential of boars with high genetic value [17,31] in order to make decisions for the implementation of fixed time insemination systems, as has been done previously in beef cattle [32].

An estimation model was developed to evaluate the cost/benefit of intravaginal triptorelin administration at 96 h post-weaning. In the case of sows with estrus onset earlier than day 5, the economic improvement is positive in the range of 15-20 €/treated sow, while in the case of sows with late estrus presentation, the economic improvement is higher and close to 65 €/treated sow. This improvement is mainly related to an increase in the reproductive performance of the treated sows compared to the control sows. This estimation model can also consider that different groups have different lactation mortality (which was not measured), or that synchronizing deliveries reduces weekend care costs and reduces mortality in attended deliveries.

Another interesting consequence of the use of this GnRH agonist is the reduction of gestation length variability. In this sense, we observed a shorter gestation and greater farrowing synchrony in animals treated with triptorelin compared to multiple AI, as has also been reported [22]. This fact could facilitate the organization of farrowing supervision and assistance during parturition to reduce mortality during the perinatal period [33], which has a great impact on the productive and economic performance of the farm [34].

In conclusion, triptorelin administration effectively induced estrus and synchronized ovulation, particularly within 96 hours of weaning when clear signs of estrus were detected, resulting in a reduction in the number of artificial inseminations with similar reproductive outcomes. While no significant differences in pregnancy rates or litter size were observed, sows with late estrus onset had lower reproductive performance. However, triptorelin treatment resulted in economic benefits by increasing income per inseminated sow, particularly in late estrus presentation. These results highlight the potential of triptorelin to optimize reproductive management in pigs, improving efficiency and economic viability.

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