Effect of repeated low doses of GnRH analogue (buserelin) on fertility performance of dairy cows with anovulation type I

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Abstract

The aim of this study was to evaluate the fertility response of dairy cows with anovulation type I on repeated low doses of GnRH agonist buserelin. The study was conducted on 83 anovulatory and 60 cyclic Polish Holstein Friesian cows. Anovulation type I was defined as small ovaries with follicles of ≤ 5 mm in diameter and without corpus luteum on two examinations in a 7-10 day interval between 50–60 days after parturition. Cows from the experimental group (n=58) received 0.4 µg of buserelin i.m. once a day for 5 consecutive days. Cows from the negative control group (n = 25) received saline. Sixty cyclic cows receiving no treatment served as positive controls. Intervals from calving to estrus and from calving to conception, pregnancy rate 30-35 days and 260 days after AI, and pregnancy loss were calculated.

The anovulatory cows had a substantially prolonged calving to conception interval, decreased pregnancy rate and increased pregnancy loss and culling rate compared to cyclic herd mates. The average calving to conception interval was significantly (p<0.05) shorter in treated cows compared to non-treated anovulatory cows (153.7 days vs 209.3 days). In conclusion, repeated low doses of GnRH analogue buserelin led to a significant shortening of calving to conception interval. More clinical trials are needed to determine the practical usefulness of this method for the treatment of anovulation type I in dairy cows.

Keywords: anovulation type I, cows, fertility, repeated doses of GnRH

Introduction

Postpartum anovulation is considered to be one of the main causes of infertility in dairy cattle (Santos et al. 2009, Walsh et al. 2011). In lactating dairy cows, the interval from calving to first ovulation typically lasts 2 to 4 weeks (Darwash et al. 1997, McCoy et al. 2006, Crowe et al. 2008). The lack of ovarian cyclicity affects 25% of dairy cows within 60 days after calving, but in some herds the incidence of anovulation can reach up to 40% (Walsh et al. 2007, Santos et al. 2009). Recently, anovulation has been classified into three types (I, II and III) on the basis of functional states of follicular development. Type I is characterized by the...
growth of follicles to emergence without further deviation or establishment of a dominant follicle (Wiltbank et al. 2002, Peter et al. 2009). Previously, such cows were clinically classified as having inactive ovaries. Ovarian inactivity with poor follicular growth and low progesterone level until day 50-60 pp was also referred to as true anoestrus, acycia or ovarian afunction (Opsomer et al. 1996, Mwaanga and Janowski 2000).

The pathophysiology of anovulation type I is not well understood. It is presumed that this condition may result from a suppression of GnRH and gonadotropins pulse frequency (Roche and Diskin 2001, Peter et al. 2009, Crowe et al. 2014). Risk factors for this condition are severe negative energy balance, BCS loss, parity, periparturient disorders, season of calving and stress factors (Walsh et al. 2007, Dubuc et al. 2012, Monteiro et al. 2021). Anovulation type I can be diagnosed by ultrasound or determination of progesterone concentration at an interval of 7-10 days (Gümen et al. 2003, Stevenson et al. 2006). Management of anovulatory dairy cows consists, in the first instance, of correcting the negative energy balance (Beam and Butler 1999, Peter et al. 2009, Crowe et al. 2014). Hormonal treatments were used in many studies with variable effectiveness to stimulate ovarian follicular growth and induce ovulation in acyclic cows. Intravaginal progesterone inserts (PRID – progesterone-releasing intravaginal device and CIDR – controlled internal drug releasing device) are recommended for the therapy of non-cyclic postpartum cows (McDougal et al. 2004, Yaniz et al. 2004). The increased circulatory concentration of progesterone suppresses GnRH and gonadotropins release and causes their storage. Removal of the progesterone insert produces a surge of GnRH, followed by FSH and LH release with subsequent resumption of ovarian cyclicity (Zerbe et al. 1999). Several studies showed that treatment of anestrous cows using intravaginal progesterone inserts alone or in combination with equine chorionic gonadotrophin (eCG) reduced the interval to first estrus and to conception compared with untreated controls (Mwaanga et al. 2004, Bryan et al. 2013, Shephard 2013, de Graff and Grimard 2018), but one study did not show this (Rhodes et al. 2003). Progesterone intravaginal devices may also be included in estrus synchronisation protocols for acyclic cows (Stevenson et al. 2006; Chebel et al. 2010, McDougall 2010, Colazo et al. 2013).


Intermittent (pulsatile) injections of GnRH at 1 to 4 h intervals to mimic endogenous patterns of gonadotropins secretion were reported to be successful in some studies (Edwards et al. 1983, Vorstermans and Walton 1985, Spicer et al. 1986, D’Occhio et al. 1989, Bishop and Wettemann 1993, Vizcarrra et al. 1997); however, this technique is very difficult to use in practice.

Hussein et al. (1992) administered repeat injections of GnRH twice weekly for 6 weeks in anestrous dairy cow with low progesterone levels. However, there were no significant differences between treated and control cows in the number of days from calving to first observed estrus or the number of days open.

In a previous study we showed that repeated low doses of GnRH analogue buserelin (0.4 µg i.m.) once a day for 5 consecutive days stimulated the development of ovarian follicles in anovulatory dairy cows with follicle growth to emergence (Barański et al. 2022). The aim of the present study was to evaluate the effect of this treatment regime on fertility performance in dairy cows with anovulation type I.

Materials and Methods

The study was conducted on 83 anovulatory and 60 cyclic Polish Holstein Friesian cows from four dairy herds in North-East Poland. Herd sizes ranged from 60 to 100 milking cows. The cows were housed in loose housing barns and fed total mixed ration based on grass silage, maize silage and concentrate according to their requirements. The average milk yield was 8000 l per year. Cows were examined by ultrasound using Honda 1500 scanner with a 5 MHz linear transducer twice in a 7-10 day interval between 50-60 days after parturition. Anovulation type I was diagnosed if small ovaries with follicles of ≤ 5 mm in diameter and without corpus luteum were found on both examinations. The cows were divided into three groups. Cows from the experimental group (n=58) received 0.4 µg of buserelin (Receptal, MSD, Poland) i.m. once a day for 5 days. Cows from the negative control group (n=25) received saline. Sixty cyclic cows that showed estrus before 60 days postpartum served as positive controls. Cows included into the study were 4 to 6 year old and showed no clinical symptoms of endometritis, lameness and mastitis. After detection of estrus the cows were artificially inseminated (AI). Pregnancy was diagnosed using ultrasonography between days 30 and 35 after insemination. The cows diagnosed pregnant were re-examined on day 260 ± 3 d after AI.

The following reproductive performances were calculated for the treated and control cows: intervals from
Effect of repeated low doses of GnRH analogue (buserelin) ...

Table 1. Reproductive performance and culling rates in anovulatory treated, non-treated and cyclic cows.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Anovulatory treated (n=58)</th>
<th>Anovulatory non-treated (n=25)</th>
<th>Cyclic (n = 60)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Interval, calving to estrus (mean ± SD)</td>
<td>100.1 ± 24.7a</td>
<td>133.5 ± 61.8b</td>
<td>50.4 ± 7.2c</td>
</tr>
<tr>
<td>Interval, calving to conception (mean ± SD)</td>
<td>153.7 ± 67.3a</td>
<td>209.25 ± 85.6b</td>
<td>109.0± 23.5c</td>
</tr>
<tr>
<td>Pregnancy rate 30-35 days after AI (%)</td>
<td>63.8 (37)</td>
<td>56.0 (14)b</td>
<td>75.0 (45)b</td>
</tr>
<tr>
<td>Services per conception</td>
<td>2.2</td>
<td>2.7</td>
<td>1.9</td>
</tr>
<tr>
<td>Pregnancy rate 260 days after AI (%)</td>
<td>51.7 (30)a</td>
<td>40.0 (10)b</td>
<td>68.3 (41)b</td>
</tr>
<tr>
<td>Pregnancy loss (%)</td>
<td>18.9 (7)</td>
<td>28.6 (4)</td>
<td>8.9 (4)</td>
</tr>
<tr>
<td>Culling rate (%)</td>
<td>17.2 (10)</td>
<td>28.0 (7)a</td>
<td>6.7 (4)b</td>
</tr>
</tbody>
</table>

Different superscript letters indicate statistical significance at p<0.05.

![Fig. 1. Pregnancy rates 30-35 days and 260 days after AI (%) in anovulatory treated, non-treated and cyclic cows. Different superscript letters indicate statistical significance at p<0.05.](image)

calving to estrus and from calving to conception, pregnancy rate 30-35 days after AI, pregnancy rate 260 days after AI and pregnancy loss. Pregnancy loss was defined as the percentage of non-pregnant cows 260 days after AI diagnosed 30-35 days after AI as pregnant.

**Statistical analysis**

The data were analysed using the Mann-Whitney test and GraphPad Prism version 9.00 (GraphPad Software, San Diego, CA, USA). The level of significance was considered as p<0.05.

**Results**

Fertility performance of anovulatory and cyclic cows is presented in Table 1. The average length of calving to conception interval was significantly (p<0.05) greater in non-treated cows with anovulation type I than in cyclic cows (209.3 days vs 109.0 days). The pregnancy rates 30-35 days after AI and 260 days after AI were significantly (p<0.05) lower in anovulatory non-treated cows compared to cyclic cows, 56.0% vs 75.0% and 44.0% vs 68.3%, respectively (Fig. 1). The average number of services per conception was 2.7 in anovulatory non-treated cows and 1.9 in cyclic cows. There was a tendency towards higher pregnancy loss in anovulatory non-treated cows than in cyclic cows (28.6% vs 18.3%). The culling rate was significantly (p<0.05) higher in non-treated anovulatory cows compared to cyclic cows (28.0% vs 6.7%).
The average intervals of calving to estrus and calving to conception were significantly (p<0.05) shorter in anovulatory cows treated with repeated low doses of GnRH analogue buserelin compared to non-treated anovulatory cows (100.1 days vs 133.5 days and 153.7 days vs 209.3 days, respectively) (Fig. 2). There was no significant difference (p>0.05) in the pregnancy rates, pregnancy loss and culling rate between treated and untreated anovulatory cows (Fig. 3).

**Discussion**

The average intervals of calving to estrus and calving to conception were significantly (p<0.05) shorter in anovulatory cows treated with repeated low doses of GnRH analogue buserelin compared to non-treated anovulatory cows (100.1 days vs 133.5 days and 153.7 days vs 209.3 days, respectively) (Fig. 2). There was no significant difference (p>0.05) in the pregnancy rates, pregnancy loss and culling rate between treated and untreated anovulatory cows (Fig. 3).

Compared to cyclic cows, the length of calving to conception interval was significantly (p<0.05) greater and the pregnancy rate 260 days after AI significantly (p<0.05) lower in treated anovulatory cows (153.7 days vs 109.0 days and 51.7% vs 68.3%, respectively).
Effect of repeated low doses of GnRH analogue (buserelin) ...

In the study of Hussein et al. (1992). It seems that repeated low doses of GnRH analogue buserelin (0.4 µg i.m.) once a day for 5 days may be useful for the treatment of anovulation type I in dairy cows.

However, despite treatment, fertility in cows with anovulation type I was lower than in cyclic cows. This underlines the importance of preventive strategies for anovulation type I. The strategy to counteract the negative effects of anovulation on fertility is based on appropriate nutrition in the dry period and early lactation, early recognition and treatment of postpartum diseases and minimizing stress factors (Wiltbank et al. 2002, Rhodes et al. 2003, Peter et al. 2009).

In conclusion, anovulation type I had detrimental effects on reproductive performance in dairy cows. Repeated low doses of GnRH analogue buserelin once a day for 5 days led to a significant shortening of the calving to estrus and calving to conception intervals and numerically, but not significantly, increased the pregnancy rate. More clinical trials are needed to determine the practical usefulness of this method for the treatment of anovulation type I in dairy cows.

References


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