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SUMMARY

Lecirelin, a gonadotropin (GnRH) analogue, is a nonapeptide with a molecular structure differing from the decapeptide of the natural GnRH, owing to the substitution of glycine in position 10 with ethylamide and in position 6 with D-tert-leucine.

These structural changes ensure that lecirelin remains bound to the specific hypophyseal receptors for a longer time and consequently prolongs its duration of action. With this study, performed in a rural environment, the effect was studied of 50 mg administered at the time of insemination to evaluate the effects on the conception rate at first insemination. The study involved 203 cows of which 101 in the treated group (GT) and 102 in the control group (GC) and was performed in eighteen (18) different farms, taking into consideration the following parameters: the number of lactations, hormonal induction of estrus (IE) and the calving-1st insemination interval (IP/AI).

Assignment of the animals to one group or the other was done alternately; no administration was performed to any of the animals in the Control group. In the cows of the treated group (GT) the conception rate at first insemination was 50% (n=61) compared to the cows in the control group where it was 44% (n=65). The cows which ovulated spontaneously showed a higher conception rate at first insemination in both groups: both treated (GT) 67% (n=63) and control animals (GC) 55% (n=73). These results were more evident in the group of cows which went spontaneously into estrus, with 67% (n=36) for the treated group (GT) and 49% (n=41) for the control group (GC).

Regarding the calving-1st AI interval, the cows which had been inseminated prior to the 60th day postpartum, like those inseminated after the 80th day postpartum, showed a conception rate at first insemination as follows: with an interval < 60: GT 67% (n=18) and GC 56% (n=27); with an interval > 80: GT 39% (n=31) and GC 26% (n=23). The cows inseminated in the period between the 60th and 80th day postpartum showed a conception rate at first insemination of 52% in the experimental group (GT) and 50% in the control group (GC).

INTRODUCTION

The obtaining of good reproductive parameters constitutes an indispensable requisite to obtain good milk production. For some time it has been noted that the increasing demands for milk production produces, as a consequence, a decrease in the fertility

of the herd. In the USA, milk production from 1960 to 2000 rose by 5,000 kg and at the same time the calving-conception interval increased by approximately 30 days (Silva 2000). For some time a decrease in herd fertility has been noted (Bayer 2004). The mean percentage conception rate at first artificial insemination in England decreased to 40% (Royal et al., 2000) and similarly in the last 10 years in Holland it has gone from 55% to 45% (Jorritsma et al., 2000). The number of artificial inseminations per conception, in the USA, in the final 20 years, has gone from 1.8 to 3.0 (Lucy 2001). Improvements in reproductive parameters have been obtained with the use of targeted therapies to restore fertility (such as Prostaglandins and the Ovsynch method). An important objective in management consists of the induction of heat or of fixed time insemination. All these systems can be attained only through pharmacological treatment of the animal.

To improve the conception rate at the time of artificial insemination use was made of gonadotropin (GnRH) or its analogues, various studies being performed which gave different results.

The treatment with 100 mg of GnRH improved the conception rate by 20% (Lee et al., 1983, Stevenson et al., 1984, Morgan et al., 1993).

The cows treated pharmacologically showed a higher concentration of luteinising hormone (LH). However, it was not possible to demonstrate a relationship between the concentration of luteinising hormone (LH) and subsequent pregnancy (Lee et al., 1985). The higher pregnancy rates were found after administration of at least 250 mg of GnRH (Morgan 1993). Injections from 25 to 100 mg of fertirelin acetate or 10 mg of buserelin did not produce better results in inseminations (Chenault, 1990). Furthermore, no differences were found in heifers.

In the molecular structure of the nonapeptide lecirelin, glycine is substituted with ethylamide in position 10 and with D-tert-leucine in position 6.

This structural difference creates a more long-lasting bond for the specific hypophyseal receptors with a consequent prolongation of the action. Compared to d-pherelin, a single administration of lecirelin produced a greater induction of oestruses (Masiulis et al., 2003). Adopting the Ovsynch method, lecirelin showed itself to be as efficacious as buserelin (Baruselli et al., 2001).

The objective of the present study was to evaluate the effect of the administration of 50 mg of lecirelin at the moment of insemination to evaluate the effect on the conception rate.

MATERIALS AND METHODS

Together with the present work, a controlled study was performed to study the effects of lecirelin (a gonadotropin analogue) on the conception rate. 203 cows, originating from 18 different herds, were used, of which 134 adult cows (number of lactations > 1) and 69 heifers (number of lactations=0). The animals of the treated group (GT, 101 animals) were injected by the intramuscular route, immediately after the first insemination, with 50 mg of lecirelin in the form of lecirelin acetate (Dalmarelin, produced by FATRO, Ozzano Emilia – Italy and distributed by Selectavet, Dr. Otto

Fischer GmbH, Weyarn-Holzolling) while the animals in the control group (GC) remained untreated.

The inclusion of animals in the treated group (GT) and in the control group (GC) was performed following an alternate order. Pregnancy was ascertained through ultrasonography performed 28 days after insemination. The animals which were already pregnant before the 28 days were excluded from the study. In 67 animals, after verifying the presence of the corpus luteum, luteolysis was induced with administration of (+)-cloprostenol (a PGF α analogue).

The conception rate at insemination in the groups is considered dependent on administration of lecorelin (LC), the administration of hormones for the induction of estrus and on the calving-1st insemination interval: IP/AI < 60 days, IP/AI between 60-79 days and IP/AI \geq 80 days. To determine the group statistics, the Chi-square test was used.

RESULTS

In the treated group, the conception rate at first insemination was 55%, depending on the treatment with lecorelin, the induction of heat or the calving-1st insemination interval, while in the control group it was 51% (Fig. 1). The heifers in the Control group showed a higher conception rate at first insemination compared to the cows (63% compared to 44%). The conception rate at first insemination in the group of treated heifers and the untreated group was 65% and 63%, respectively.

In the cows of the treated group, the conception rate at 1st insemination was 6% higher than in the untreated group (50% against 44%) (Fig. 2).

Fig. 1: Pregnancy rate at first insemination following administration of lecorelin at the moment of insemination in the treated group (GT) compared to the control group (GC).

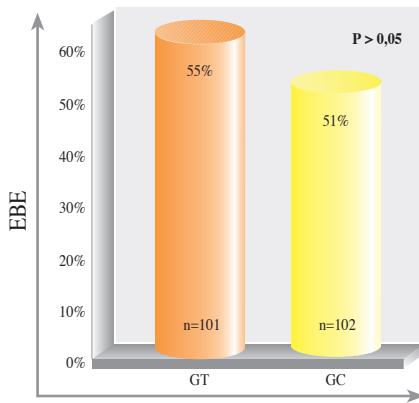


Fig. 2: Pregnancy rate at first insemination following administration of lecorelin at the time of insemination in the treated group (GT) compared to the control group (GC) in relation to the number of lactations.

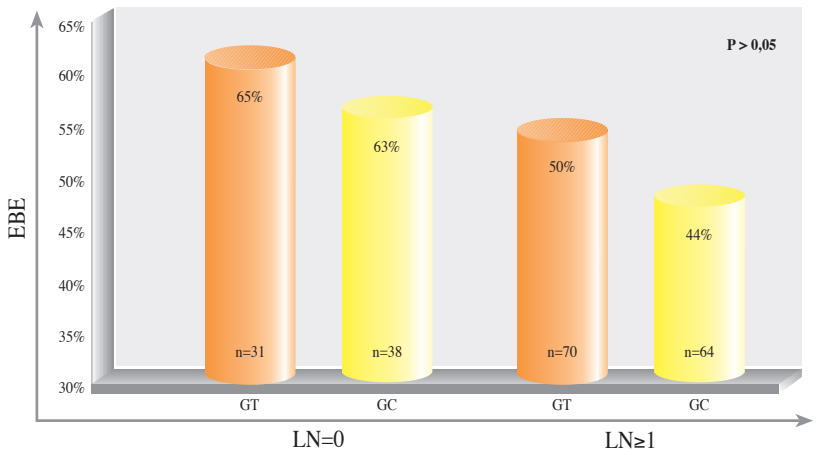


Fig. 3: Pregnancy rate at first insemination following administration of lecorelin at the time of insemination in the treated group (GT) compared to the control group (GC) in relation to previous hormonal induction of estrus.

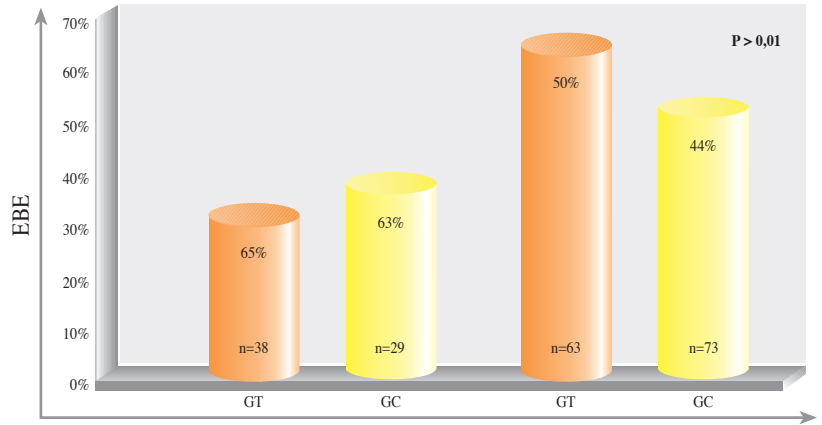
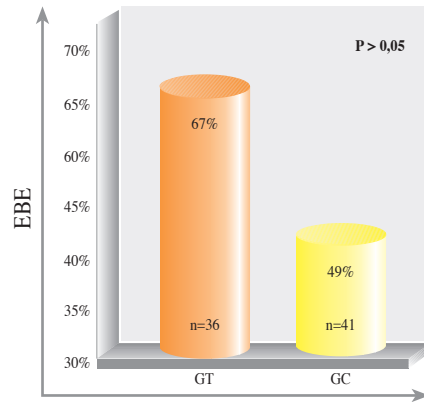


Fig. 4: Pregnancy rate at first insemination following administration of lecorelin at the time of insemination in the treated group (GT) compared to the control group (GC) in cows with spontaneous estrus.

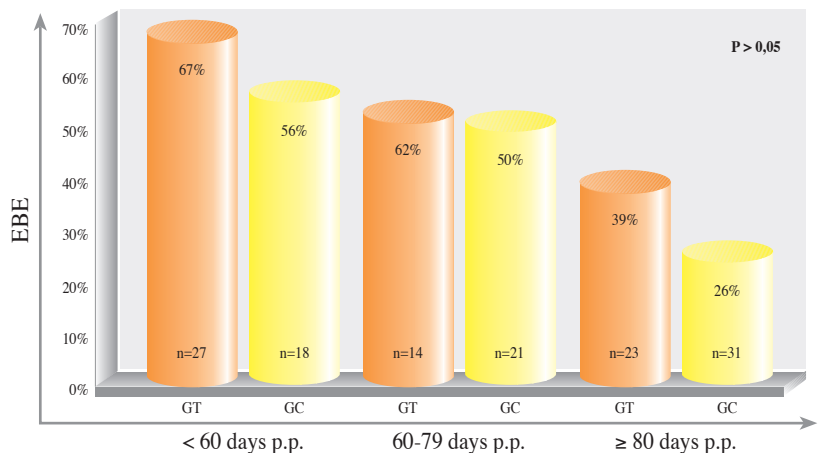


The animals in which heat had been induced showed a low conception rate at first insemination, both in the treated group and in the untreated group (Fig. 3). The difference within the treated group was statistically significant ($P < 0.001$). Treated animals in which heat had not been induced showed a higher conception rate at insemination compared to the untreated group (67% against 55%), while animals in which heat had been induced showed a lower conception rate at insemination compared to the untreated group (34% against 41%).

Cows with spontaneous estrus showed a conception rate at insemination of 49% in the untreated group and 67% in the treated group (Fig. 4).

In the treated and untreated cows, the conception rate at first insemination was influenced by the duration of the calving-1st insemination interval (Fig. 5). In the untreated group it was lower: 56% (IP/AI < 60 days), 50% (IP/AI between 60-79 days) and 26% (IP/AI ≥ 80 days) respectively; in the treated group, however, the figures were 67%, 56% and 39%.

Fig. 5: Pregnancy rate at first insemination following administration of lecorelin at the time of insemination in the treated group (GT) compared to the control group (GC) in cows with spontaneous heat in relation to the calving-1st heat interval.



The differences between groups IP/AI < 60 days, IP/AI between 60-79 days and IP/AI ≥ 80 days, untreated, were statistically significant ($p < 0.05$). The animals which showed different durations of the calving-1st insemination interval, belonging to the treated group, showed a higher conception rate at first insemination compared to the untreated group.

DISCUSSION

Following the increased metabolic requirements due to milk production, the pregnancy rate in cows decreases drastically. On average, the conception rates at first insemination were satisfactory, over 55% (de Kruif et al., 1998). The present study, performed in a rural environment, was conducted using 203 cows to study the positive effects of Lecirelin (Dalmarelin) in improving the conception rate at first insemination. The results highlighted a tendency to improvement in the first insemination in cows with more than one lactation.

These results were more evident in cows which came in oestrus spontaneously. In heifers at their first lactation, no differences were found. These results are in accordance with those of Lee et al. (1983), who, after administration of 100 µg GnRH at the time of insemination of the heifers, did not find higher pregnancy rates.

An improvement in the conception rate in the cows following administration of lecorelin was linked with an increased release of LH, which in itself can shorten the time between insemination and ovulation and which translates into an improvement in the conception rate (Lee et al., 1985). Further and more evident manifestations of heat, when this was induced through hormones, have been described in the literature.

This realisation is attributed to the fact that normally the more animals that are synchronised, the more they mutually show symptoms of heat (Roelofs et al., 2005). In this study, estrus was induced only in a few individual animals, so the effect was not noted.

However, the animals of the treated group and those of the control group, in which heat had been induced, showed less success at first insemination, with a longer calving-1st conception interval. This contrasts with what is reported in the literature, which speaks of a greater success of insemination with a calving-1st conception interval increased to the hundredth day postpartum (Lotthammer and Wittkowski, 1993).

It can be stated that the cows which, up to 80 days postpartum, had not gone spontaneously into heat, were subjected to a number of inductions of heat, so the problem of animals linked to an increased calving-1st insemination interval increased in any case.

The positive effects of the administration of Lecirelin on success at first insemination were demonstrated with inseminated cows, both with or without induction of heat. In conclusion, the results of the present study have demonstrated that the success rate at first insemination can be improved administering 50 µg lecorelin at the time of insemination, both in cows which came on oestrus spontaneously and in cows which showed a particularly short or particularly long calving-first heat interval.

Bibliography

- BARUSELLI, P. S., R. AMARAL, F. B. BARUFI, R. VALENTIM and M. O. MARQUES: Lecirelin and Buserelin (Gonadotropin releasing hormone agonists) are equally effective for fixed time insemination in buffalo. *Braz. J. Vet. Res. Anim. Sci* 38, 142–145 (2001).
- CHENAULT, J. R.: Effect of fertirelin acetate or buserelin on conception rate at first or second insemination in lactating dairy cows. *J. Dairy Sci.* 73, 633–638 (1990).
- DE KRUIF, A., M. HOEDEMAEKER and R. MANSFELD: Tierärztliche Bestandsbetreuung beim Milchrind. Stuttgart, Enke (1998).
- JORRITSMA, R., H. JORRITSMA, Y. H. SCHUKKEN and G. H. WENTINK: Relationships between fatty liver and fertility and some periparturient diseases in commercial dutch dairy herds. *Theriogenology* 54, 1065–1074 (2000).
- LEE, C. N., E. MAURICE, R. L. AX, J. A. PENNINGTON, W. F. HOFFMAN and M. D. BROWN: Efficacy of gonadotropin-releasing hormone administered at the time of artificial insemination of heifers and post-partum and repeat breeder dairy cows. *Am. J. Vet.* 44, 2160–2163 (1983).
- LEE, C. N., J. K. CRITSER and R. L. AX: Changes of luteinizing hormone and progesterone for dairy cows after gonadotropin-releasing hormone at first post-partum breeding. *J. Dairy Sci.* 68, 1463–1470 (1985).
- LANDESKURATORIUM DER ERZEUGERRINGE FÜR TIERISCHE VEREDELUNG IN BAYERN E.V. (LKV): Leistungs- und Qualitätsprüfung in der Rinderzucht in Bayern 2004 – Ergebnisse und Auswertungen. München (2004).
- LOTTHAMMER, K. H., and G. WITKOWSKI: Fruchtbarkeit und Gesundheit der Rinder. Verlag Eugen Ulmer (1993).
- LUCY, M. C.: Reproductive Loss in High-Producing Dairy Cattle: Where Will It End? *J. Dairy Sci.* 84, 1277–1293 (2001).
- MASIULIS, M., H. ZILINSKAS and V. RISKEVICIENE: Follicular growth dynamics. Application of preparations DALMARELIN (Lecirelin) and DEIPHERELIN® (Gonavet® 50) for estrus stimulation in cows. *Vet. IR Zootech.* 23, 10–18 (2003).
- MORGAN, W. F., and I. J. LEAN: Gonadotropin-releasing hormone treatment in cattle: a meta-analysis of the effects on conception at the time of insemination. *Aust. Vet. J.* 70, 205–209 (1993).
- ROELOFS, J. B., F. J. van EERDENBURG, N. M. SOEDE and B. KEMP: Various behavioral signs of estrus and their relationship with time of ovulation in dairy cattle. *Theriogenology* 15, 1366–1377 (2005).
- ROYAL, M., G. E. MANN and A. P. F. FLINT: Strategies for reversing the trend towards subfertility in dairy cattle. *The Veterinary Journal* 160, 53–60 (2000).
- SILVA, J. W.: Addressing the decline in reproductive performance of lactating dairy cows: a researcher's perspective. www.vetsci.org/publish/articles/000043/article.pdf (2003).
- STEVENSON, J. S., M. K. SCHMIDT, and E. P. CALL: Gonadotropin-releasing hormone and conception of Holsteins. *J. Dairy Sci.* 67, 140–145 (1984).

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