

Effects of GnRH (buserelin) Administration 12 Days After Artificial Insemination on the Pregnancy Rates of Cattle

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(Received November 26, 2002/Accepted June 9, 2003)

Summary : Gonadotropin-releasing hormone (GnRH ; buserelin) was administered 12 days after artificial insemination to 307 head of cattle bred by a stock-raising farmer in a public pasture and here at the Fuji Zootechnical station. Cattle were given doses of 6, 10 and 20 μg respectively and pregnancy rates were compared to and examined with those of a non-treated control population. For the control population, the pregnancy rate was 60.0%, while for the treated group which was administered 10 and 20 μg of buserelin, a high pregnancy rate of 72.3 and 68.6% respectively, resulted. After the group was divided into two, a short division (less than 120 days) group and long division (120 days or more) group, the rates were examined. For the short division group, the 6, 10 and 20 μg groups showed pregnancy rates higher than the 59.0% in the control population. For the long division group, the control population rate of 63.2% was low in comparison to that of the 6 μg group. However, it was high in the 10 and 20 μg groups. Moreover, in comparison with the 10 and 20 μg group, the conception rate for one-time administration also tended to be high in the control population at 59.2%.

We examined administration frequency by dividing the group into one-time and two-time groups and found the rate to be high in the 10 μg group, at 73.3%. In the 10 and 20 μg groups, the pregnancy rate after two-time administration was high when compared with the 66.7% in the control population, and especially high in the 20 μg group at 81.0%. In addition, a high pregnancy rate was obtained due to effects of administration in cattle with medical treatment history at 57.1, 36.4 and 52.9% in comparison to the 25.0% in the control population. The above results indicate that the pregnancy rate improved with buserelin administration 12 days after artificial insemination in cattle and that the effect of a 10 μg buserelin dose was equivalent to a 20 μg dose. Of special note is the effectiveness of buserelin administration on cattle including repeat breeders etc., which had pregnancy difficulties.

Key Words : cattle, artificial insemination, GnRH (buserelin), pregnancy rate, non-pregnant condition period

Introduction

By administering gonadotropin-releasing hormone (GnRH) 11~14 days after artificial insemination it is possible to delay the regression of corpus which originally existed in the highest-ranking follicle appearing when luteinizing hormone (LH) is discharged, according to hypothesis, during the ovulation phase or closure. In this period the growth of the embryo is temporarily stimulated and the possibility of early extinction of the embryo appears to decrease. It is thought that it

maintains the blood progesterone concentration at a high level and in addition effectively improves the conception chance for yellow body dysfunction. Therefore, the basis of this hypothesis is that the conception rate improves in some tests when a GnRH pharmaceutical preparation is administered 11~14 days after artificial insemination¹⁻⁶⁾. However the effect cannot be differently and sufficiently examined.

Recently, the effects of GnRH and prostaglandin F₂ α or synchronization of estrus ovulation which jointly uses the analogue, periods of artificial insemination etc,

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have been developed and studied⁷⁾. However, by using this method, there is no obvious estrus sign as the pregnancy rate for the past 6 years has been carelessly reported at a high of 51%⁸⁾. Although chemical administrations such as hormone treatment are not carried out with insemination, the economic merits are generally large if an improvement in conception rates can come about with a single dose of buserelin or in simple operations of repeated doses. Previously, the authors administered GnRH (buserelin) to cattle simultaneously with artificial insemination and the pregnancy rate increased 7.6% to 9.2%. It was reported that the effect of the buserelin administration on cattle is especially hard to note⁹⁾. The effect of buserelin administration 12 days after artificial insemination on pregnancy rates in cattle was examined.

Materials and Methods

Holstein and Japanese black cows (a total of 307 head) bred by a stock-raising farmer, at a public pasture and at the Fuji Zootechnical station were used. Trial medicine GnRH Acetic acid buserelin 4.2 μ g (buserelin 4 μ g: the following buserelin, Germany Hoechst Co.) were used in buserelin (Estomal[®]: Shionogi) which was the material Estomal note of 1.5 (6 μ g as a buserelin), 2.5 (10 μ g) and 5.0 ml (20 μ g) were treated after artificial insemination on day 12, and a control population was untreated. Medication was injected 12 days after artificial insemination into the muscles of 74 cows receiving 6 μ g buserelin, and 83 and 70 cows receiving 10 μ g and 20 μ g doses respectively. Eighty cows were non-treated as control population. However re-treatment was carried out 12 days after artificial insemination for cows that did not become pregnant. The following results were similar to those of a previous report of individual cow subjects while a significant test on the pregnancy rate was administered similarly to the one in the previous report⁹⁾.

Results

1. Pregnancy rate of all cow subjects

The pregnancy rates of the group administered buserelin 12 days after artificial insemination and of the control population are shown in Table 1.

The pregnancy rates in the 6, 10 and 20 μ g groups were 58.1% (43/74), 72.3% (60/83) and 68.6% (48/70) respectively. The pregnancy rates of the 10 and 20 μ g dose group were higher than the 60.0% (48/80) of the control population. For the 6 μ g buserelin group, lower rate was obtained than the 10 and 20 μ g group, and the differences were not significant among each group.

2. Pregnancy rate according to differences in the non-pregnant condition period

The test cows were divided into two groups, short division (less than 120 days) and long division (120 days or more), differences in the pregnancy rates with regard to the non-pregnant condition period are shown in Fig. 1.

The pregnancy rates in the non-pregnant condition period were 62.2% (28/45), 73.6% (39/53) and 68.3% (28/41) and when compared to differences in Fig. 1. Those of the non-pregnant condition period are lower in the 10 μ g, and 20 μ g dose group. The result is higher than the 59.0% (36/61) of the control population. The 10 and 20 μ g groups tended to be especially high. Moreover, in the conception rates for the long division group, the pregnancy rate in groups administered with buserelin of 6 μ g, 51.7% (15/29), is lower than the 63.2% (12/19) of the control population. However, the rates of 70.0% (21/30) and 69.0% (20/29) were observed in 10 and 20 μ g groups, and higher than that of the control population with no significant difference.

3. Pregnancy rate according to the number of medication times

Cattle which became pregnant when administered buserelin one or two times were divided into two groups. The difference in pregnancy rates is shown in Table 2.

Table 1 Pregnancy rate with buserelin medication 12 days after artificial insemination

Dose. μ g	No. of cattle inseminated	No. of cattle pregnant	Pregnancy rate (%)
0	80	48	60.0
6	74	43	58.1
10	83	60	72.3
20	70	48	68.6

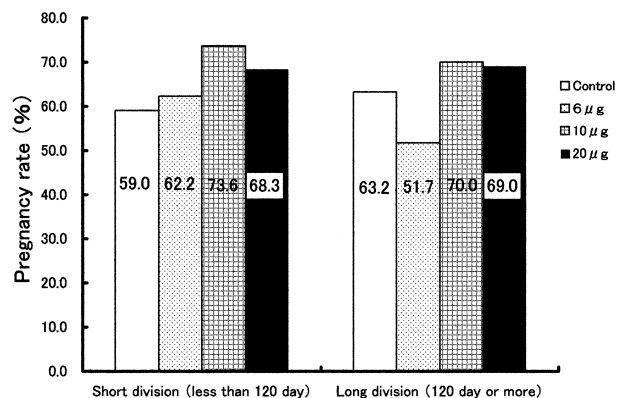


Fig. 1 Pregnancy rate according to difference during the non-pregnant condition period

Table 2 Pregnancy rate according to difference in the number of times of medication

No. of administration times	Dose. μ g	No. of cattle inseminated	No. of cattle pregnant	Pregnancy rate (%)
1	0	71	42	59.2
	6	57	33	57.9
	10	60	44	73.3
	20	49	31	63.3
2	0	9	6	66.7
	6	17	10	58.8
	10	23	16	69.6
	20	21	17	81.0

The pregnancy rate differs according to differences in administered doses. Rates were 57.9% (33/57) at 6 μ g, 73.3% (44/60) at 10 μ g and 63.3% (31/49) at 20 μ g in the administered group respectively. For the 6 μ g group, the rate was a little lower than the 59.2% (42/71) of the control population, and in the 10 and 20 μ g group, it tended to be high, especially in the 10 μ g group. With a 6 μ g dose, the pregnancy rates for twice administered subjects, in the 6 μ g group and in the 20 μ g group are lower than the 66.7% (6/9) of the control population at 58.8% (10/17), and the pregnancy rate of 69.6% (16/23) and 81.0% (17/21) observed in administered group of 10 and 20 μ g respectively. Also, in the 20 μ g group, a high pregnancy rate is shown, with no significant difference in either interval.

4. Effect of buserelin administration on subjects with medical treatment history

The pregnancy rate of the buserelin-administrated group of subjects with prior treatment history for reproductive disorders by herd, and the rate of the control population are shown in Table 3.

The pregnancy rates in groups given buserelin doses of 6, 10 and 20 μ g were 57.1% (4/7), 36.4% (4/11) and 52.9% (9/17) respectively. They were higher than that of the control population (25.0% (1/4)).

Discussion

It is thought that the effects of the buserelin administration 12 days after artificial insemination causes the activation and maintenance of the lustrum function by LH emission, a series of phenomenon of estrogen actinia by ovulation induction of the superior follicle after artificial insemination^{3,10}. On the other hand, estrogen actinia reversely suppresses the increase in the oxytocin receptor concentration of the uterus film and stops autolysis by a control in production of PGF2 α , which is the autolysin factor¹¹. Furthermore, it produces a signal material (cattle trophoblast protein: bTP-1) which informs to the mother with a pregnancy signal

Table 3 Buserelin medication effect on individuals with previous medical treatment history

Dose. μ g	No. of cattle inseminated	No. of cattle pregnant	Pregnancy rate (%)
0	4	1	25.0
6	7	4	57.1
10	11	4	36.4
20	17	9	52.9

from around fertilization post-15th embryo of lute in and this material stimulates the control or the lustrum function with respect to autolysis, maintaining pregnancy¹⁰.

By delaying the regression of corpus lustrum, buserelin administration prevents the extinction of this early embryo and it is believed that this gives it time to reach a size at which the embryo can produce bTP-1. In addition, the lustrum function is activated, pregnancy is established and maintained and conception chances improve.

In the present test, the conception rates in the 10 μ g and 20 μ g buserelin group were compared with the control population and were found to be respectively higher, 12.3~8.6% than that of control population. Therefore buserelin was shown to improve the pregnancy rate. In the 6 μ g group, it was slightly lower than the control population. From this fact, it was thought that GnRH was necessary to an extent for the activation and maintenance of the lustrum function than for ovulation promotion. When the non-pregnant condition period was divided into the short division group (less than 120 days) and long division group (120 days or more), and pregnancy rates examined in the short groups, the pregnancy rates for the 6 μ g group in the short group were higher than that of the control population, and were higher for the 10 μ g and 20 μ g groups than the control population regardless of the non-pregnant condition period. Moreover, in the long group, the rate was low. For the long group, it is thought that administration of 6 μ g buserelin to improve the preg-

nancy rates is not possible since the conception rate for the control population was high. Since a smaller number of subjects was examined in the long group compared to the short group, differences in the effect of buserelin during the non-pregnant condition period were not clear. This was especially true when the control population pregnancy rates were examined and compared to the 10 μ g group in one-time administration, when the group was divided into those that became pregnant after one-time buserelin administration and those that became pregnant after two-time administration. In the 20 μ g group, a slightly high rate was obtained and the pregnancy rate was higher than the control population. In the 6 μ g group, it was slightly lower than that in the control population. For the 20 μ g and 10 μ g groups, the rates were higher than that of the control population, because for two-time administration, there were a smaller number of subjects in the control population, and because the pregnancy rate was high in the 6 μ g group, it was reversely lowered. We could not determine this fact in this experiment. It seemed that there was no clear difference between administration frequency and the non-pregnant condition period in indicating buserelin as a factor which influences the conception chances. The number of subjects between the time of artificial insemination and buserelin administration were small.

This all indicates that the effect of buserelin administration 12 days after artificial insemination on the pregnancy rate in cattle is great, and that a 10 μ g dose is equally effective as a 20 μ g one. Buserelin administration was especially notable in its remarkable effect on cattle (including repeat breeder, etc.) whose conception chances are low.

References

- 1) STEVENSON, J.S., CALL, E.P., SCOPY, R.K. and PHATAK, A.P., 1990. Double insemination and gonadotropin-releasing hormone treatment of repeat-breeding dairy cattle. *J. Dairy Sci.*, **73**, 1766-1772.
- 2) STEVENSON, J.S. and MEE, M.O., 1991. Pregnancy rates of Holstein cows after postinsemination treatment with a progesterone-releasing intravaginal device. *J. Dairy Sci.*, **74**, 3849-3856.
- 3) MACMILLAN, K.L., TAUFU, V.K. and DAY, A.M., 1986. Effects of an agonist of gonadotrophin releasing hormone (buserelin) in cattle. III. Pregnancy rates after a post-insemination injection during met estrus or diestrus. *Anim. Reprod. Sci.*, **11**, 1-10.
- 4) LESLIE, K.E., BOSU, W.T., LISSEMORE, K. and KELTON, D., 1984. The effect of gonadotropin-releasing hormone administration four days after insemination on first-service conception rates and corpus lustrum function in dairy cows. *Can. J. Vet. Res.*, **50**, 184-188.
- 5) HUMBLLOT, P. and THIBIER, M., 1981. Effect of gonadotrophin releasing hormone (GnRH) treatment during the midlevel phase in repeat breeder cows. A preliminary report. *Theriogenology*, **16**, 375-378.
- 6) RETTMER, I., STEVENSON, J.S. and LARRY, R.C., 1992. Pregnancy rates in beef cattle after administering a GnRH agonist 11 to 14 days after insemination. *J. Anim. Sci.*, **70**, 7-12.
- 7) PURSLEY, J.R., MEE, M.O. and WILTBANIC, M.C., 1995. Synchronization of ovulation in dairy cows using PGF2 α and GnRH. *Theriogenology*, **44**, 915-923.
- 8) YAMADA, K., 2001. The novel reproductive technologies for improvement of reproductive performance in dairy cows. *J. Reprod. Dev.*, **47**, j39-j45.
- 9) MONJI, Y., SATO, M., KUWAYAMA, T. and DOMEKI, I., 2002. Effect of gonadotropin-releasing hormone administration during artificial insemination on the pregnancy rates in cattle. *Jour. Agri. Sci., Tokyo Univ. of Agric.*, **47**, 182-186.
- 10) THATCHER, W.W., MACMILLAN, K.L., HANSEN, P.J. and DROST, M., 1989. Concepts for regulation of corpus luteum function by the concepts and ovarian follicles to improve fertility. *Theriogenology*, **31**, 149-164.
- 11) MANN, G.E. and LAMMING, G.E., 1992. Steroid hormone manipulation in control of autolysis in cattle. *J. Reprod. Fertil. Abstract series*, **9**, 23.

人工授精後 12 日目の GnRH 製剤（ブセレリン） 投与が牛の受胎率に及ぼす影響

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（平成 14 年 11 月 26 日受付/平成 15 年 6 月 9 日受理）

要約：畜産農家、公共の牧場および本学富士畜産農場で飼育されている雌牛 307 頭に人工授精後 12 日目に性腺刺激ホルモン放出ホルモン（GnRH：ブセレリン）を 6, 10, 20 μ g 投与し、受胎率について無処置対照群のそれと比較検討した。その結果、ブセレリン投与群の受胎率は対照群の 60.0% に比べ 10 および 20 μ g 投与群ではそれぞれ 72.3 および 68.6% と高い値が得られた。次いで、空胎期間の短い群（120 日未満）と長い群（120 日以上）の 2 群に分けて検討したところ、短い群の受胎率は対照群の 59.0% に比べいずれの投与群とも高い値が得られ、長い群では対照群の 63.2% に比べ 6 μ g 投与群で 51.7% と低い値を示した。しかし、10 および 20 μ g 投与群で 70.0 および 69.0% といずれも高い傾向がみられた。また、投与回数を 1 回と 2 回に分けて検討したところ、1 回投与での受胎率は、対照群の 59.2% に比べ 10 および 20 μ g 投与群では高い傾向が認められ、特に 10 μ g 投与群では 73.3% と高い値であった。2 回投与での受胎率は対照群の 66.7% に比べると 10 および 20 μ g 投与群では高く、特に 20 μ g 投与群では 81.0% と高い値であった。さらに、治療歴を有する牛群への投与効果については、対照群の 25.0% に比べ各投与群で 57.1, 36.4, 52.9% と高い受胎率が得られた。

以上の成績より、牛において人工授精 12 日目のブセレリン投与は、受胎率を向上させることが示唆され、ブセレリン 10 μ g 投与量での効果が 20 μ g 投与と同等あるいはそれ以上であった。特にリピートブリーダーなどを含む受胎しにくい牛に対するブセレリン投与効果が高いことが示唆された。

キーワード：牛、人工授精後 12 日目、ブセレリン、受胎率、空胎期間

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