

Effect of Administration of Lesireline Five Days After Artificial Insemination on Pregnancy Rates and Luteal Function in Cows

Cihan KAÇAR* Savaş YILDIZ** Şükrü Metin PANCARCI*
Mehmet KAYA*** Örsan GÜNGÖR* Umut Çağın ARI**

* Kafkas Üniversitesi Veteriner Fakültesi Doğum ve Jinekoloji AD, Kars-TÜRKİYE

** Kafkas Üniversitesi Veteriner Fakültesi Dölerme ve Suni Tohumlama AD, Kars-TÜRKİYE

*** Ondokuz Mayıs Üniversitesi Veteriner Fakültesi Fizyoloji AD, Samsun-TÜRKİYE

Yayın Kodu: 2007/20-A

Summary

The aim of this study was to increase pregnancy rates by administering lesireline five days after artificial insemination (AI). Five days after AI, lesireline acetate (2 ml) and physiologic saline solution (2 ml) were injected intramuscularly in Group I (n=29) and Group II (n=27), respectively. Blood samples were collected at AI (day 0) and 5, 10 and 20 days after AI to determine serum progesterone concentrations. Pregnancies were diagnosed via palpation per rectum at day 60 after AI. Pregnancy rates were 24.1% and 22.2% in lesireline (Group I) and control (Group II) groups, respectively, and these pregnancy rates did not significantly differ between groups ($p>0.05$). Serum progesterone concentrations ten days after AI were not significantly different between experimental and control groups ($p>0.05$). Although serum progesterone concentration in lesireline group (6.3 ± 0.9 ng/ml) was higher than those in control group (5.2 ± 1.0 ng/ml), it did not significantly differ between groups ($p>0.05$). In spite of the fact that administration of lesireline resulted in significant rise in progesterone levels in the middle of the cycle, pregnancy rates did not differ between groups.

Keywords: Cow, Lesireline, Progesterone, Pregnancy rate

İneklerde Suni Tohumlamadan Sonraki Beşinci Günde Lesireline Uygulamalarının Gebelik Oranı ve Luteal Fonksiyona Etkisi

Özet

Bu çalışmada, ineklerde suni tohumlamadan 5 gün sonra lesireline uygulaması yapılarak gebelik oranının artırılması amaçlandı. Suni tohumlamadan sonraki 5. günde Grup I (n:29) lesireline ml, Grup II (n:27) ise 2 ml fizyolojik NaCl çözeltisi intramuskuler yolla uygulandı. İneklerden tohumlama gününden (0. gün) itibaren, 5, 10 ve 20. günlerde kan alınarak serum progesteron değerleri belirlendi. Gebelik muayeneleri tohumlamayı izleyen 60. günde rektal palpasyon ile yapıldı. Gebelik oranları lesireline grubunda %24.1, kontrol grubunda ise %22.2 olarak tespit edildi. Gruplar arasında gebelik oranı bakımından istatistiksel yönden önemli bir fark saptanmadı ($p>0.05$). Suni tohumlamadan 10 gün sonra serum progesteron değerleri lesireline grubunda 6.3 ± 0.9 ng/ml ile kontrol grubuna göre (5.2 ± 1.0 ng/ml) daha yüksek bulunmasına rağmen istatistiksel yönden önemli bir fark belirlenmedi ($p>0.05$). Lesireline uygulaması ile siklusun ortasında progesteron seviyesinde önemli derecede yükselme olmasına rağmen gebelik oranlarını etkilemediği belirlendi.

Anahtar sözcükler: İnek, Lesireline, Progesteron, Gebelik oranı

İletişim (Correspondence)

Phone: +90474 2426800/1233

e-mail: cihan3000@hotmail.com

INTRODUCTION

GnRH is widely used to control of reproduction and to improve fertility¹. In many studies GnRH analogs were administered at artificial insemination (AI) to eliminate late ovulations and to increase ovulation rates^{2,3}. Moreover, many researchers attempted to obtain high pregnancy rates using GnRH between 5-14 days following AI during early pregnancy period³⁻⁶. Administration of Buserelin at mid-diestrus resulted in increase in FSH and LH release leading to elevated progesterone concentrations^{4,6,7}.

The aim of this study was to increase progesterone concentrations during early luteal period by means of GnRH injection at day five after AI. With administration of GnRH following AI, developing dominant follicles either ovulates and develops accessory corpus luteum (CL) or goes atresia. Mann and Lamming⁸ reported that rates of embryonic death in cows with lower progesterone concentrations were higher, and pregnancy rates were increased by enhanced progesterone concentrations following GnRH administrations.

In this way, progesterone concentrations could be increased due to GnRH administration⁶. Therefore, in this study it was aimed to eliminate embryonic losses due to low progesterone concentrations and to improve pregnancy rates by administering lesireline at day five after AI.

MATERIALS and METHODS

This study was conducted on 56 cows with different breeds (10 Simmental, 35 Brown Swiss cross bred, 10 Zavot cross bred and 1 local black cattle), ages (2 to 13 years old), postpartum days (30 to 360 days) and average milk yield of 7.9 kg. No abnormalities were detected in reproductive tract at AI in all experimental cows. Number of parity was ranged in 1 to 10. Body condition scores (BCS) of experimental cows were determined using 1 to 5 scale (1=skinny, 5=obese) as described by Edmonson⁹. Cows were randomly assigned into two groups. Lesireline acetate (Dalmarelin®, Vetaş, Istanbul, Turkey, Lesireline asetat, 25 µg; 2 ml; IM) and physiologic saline solution (2 ml) were given via intramuscular injection to cows in Group I

(n=29) and Group II (Control Group; n=27) 5 days after AI, respectively. Artificial inseminations were performed after visual observation of estrus. Blood samples were collected via jugular vein puncture at day 0 (AI), 5, 10 and 20 after AI in order to determine serum progesterone concentrations. Blood samples were centrifuged at 3000 rpm for 20 minutes and serum samples were stored in eppendorf tubes at -20 °C until progesterone concentrations were assayed. Serum progesterone values were determined using ELISA (Enzymimmunoassay) as described by Prakash et al.⁶. Pregnancies were diagnosed by palpation per rectum two months after AI.

Data were analyzed statistically by using Chi-square and t-test in SPSS 10.0 programme.

RESULTS

Body condition scores (BCS) of cows enrolled in this study were between 1.75 and 3.5. BCS of cows in Group I and Group II were ranged in 1.75-3.5 and 1.75-3.0; respectively. There were no significant differences between groups for BCS ($p>0.05$). There were no significant difference between pregnancy rates cows in lesireline and those in control groups for BCS ($p>0.05$). Pregnancy rates in cows with BCS from 1.75 to 2.25 and from 2.5 to 3.5 were 16.7% and 30.0%; respectively, and this difference was statistically significant ($p<0.01$, Figure 1).

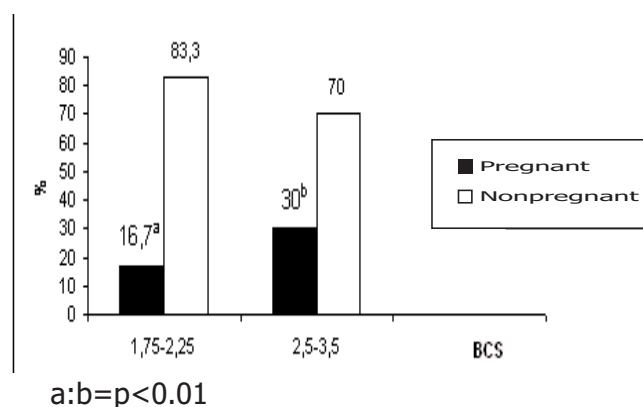


Fig 1. Pregnancy rates of cows classified based on BCS (1.75-2.25 and 2.5-3.5)

Şekil 1. İneklerde BCS (1.75-2.25 ve 2.5-3.5) grupları arasındaki gebelik oranı

Overall pregnancy rate was 23.2% (13/56) in this study. Pregnancy rates for lesireline and control groups were shown in Table 1. Pregnancy

rates for lesireline and control groups were 24.1% (7/29) and 22.2% (6/27); respectively *Table 1*. There was not significantly difference between groups for pregnancy rates ($p>0.05$).

Table 1. Pregnancy rates for lesireline and control groups
Tablo 1. Lesireline ve kontrol grupları arasındaki gebelik oranı

Group n (%)	Group I: Lesireline (n:29)	Group II: Control (n:27)
Pregnant n (%)	7 (24.1) ^a	6 (22.2) ^b
Nonpregnant n (%)	22 (75.9)	21 (77.8)

a:b= $p>0.05$

Following evaluation of progesterone concentrations, one cow in GnRH group and one cow in control group were excluded from study and their progesterone values were not considered because the values of progesterone of these cows were higher than 1 ng/ml at the insemination time. Average serum progesterone concentration at AI was 0.6 ± 0.1 ng/ml. Progesterone values five days after AI revealed that serum progesterone concentrations of six cows in control group and three cows in lesireline group were lower than 1 ng/ml which indicated ovulation failure in those cows. Among regularly cycling cows, serum progesterone levels ten days following AI were 6.3 ± 0.9 ng/ml and 5.2 ± 1.0 ng/ml in lesireline and control groups; respectively (*Figure 2*).

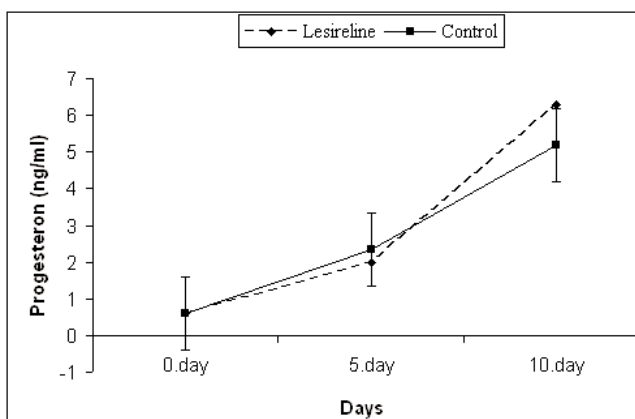


Fig 2. Serum progesterone values five days after AI in lesireline and control group

Grafik 2. Lesireline ve kontrol grubunda 5. günden sonraki serum progesteron konsantrasyonu

Among pregnant and non-pregnant cows,

serum progesterone concentration in lesirelin group was higher than that in control group ten days following AI, however, this difference was not statistically significant ($p>0.05$) *Table.2*. Also, no difference was found between lesireline and control groups for serum progesterone concentrations at day 20 after AI among pregnant cows ($p>0.05$).

Table 2. Serum progesterone concentrations (ng/ml) 10 and 20 days after AI

Tablo 2. Suni tohumlamadan sonraki 10. ve 20. günlerdeki progesteron (ng/ml) konsantrasyonu

Days	n=29	Pregnant (n=7)	Non-pregnant (n=22)	n=27	pregnant (n=6)	Non-pregnant (n=21)
10	$6.3\pm 0.9^{a,b}$	$9.6\pm 2.7^{c,d}$	5.3 ± 0.9	$5.2\pm 1.0^{a,b}$	$10.9\pm 3.2^{c,d}$	4.1 ± 0.9
20	2.9 ± 0.9	2.8 ± 1.1	2.9 ± 1.2	3.9 ± 1.1	5.6 ± 1.2	3.4 ± 1.5

^{a,b,c}The values with different superscripts are significantly different within row (a:b= $p>0.05$; c:d= $p>0.05$).

DISCUSSION

In many studies, administration of GnRH analogs with different dosages (10, 25, 100 and 250 μ g) prior to AI, at AI and within 25 days following AI resulted in pregnancy rates between 39.2% and 81.3%^{3,4,10-12}. Although pregnancy rates were lower in control groups (25.4 to 65.4%) than GnRH administered groups in those studies, statistically significant difference in pregnancy rates were reported in a few studies^{5,13}. Moreover, many studies revealed that there was no significant difference following GnRH application^{4,10,12-16}. Similarly, no difference was found for pregnancy rates between groups in our study ($p>0.05$).

Clindworth¹¹ achieved the highest pregnancy rates following the first service in cows with BCS of 3.0, and they reported lower pregnancy rates in cows with BCS lower or higher than score of 3. Smith¹⁷ and Villa-Godoy¹⁸ reported the significant relationship between BCS and fertility. Likewise, significant ($p<0.01$) positive effect of BCS on pregnancy rates was found in present study.

In a study, administration of GnRH twelve days following AI resulted in elevation of progesterone concentrations within only several hours¹⁹. In the same study, pregnancy rate was increased 5.8% without statistical significance

following GnRH administration. In another study, after administration of GnRH at day 5 following AI, a significant ($p < 0.05$) rise in serum progesterone concentrations was reported ten days after AI. GnRH injection causes differentiation of luteal cells during early luteal development⁷. LH release is stimulated leading to increase in differentiation of small luteal cells to large luteal cells following GnRH administration. It has been reported that large luteal cells release progesterone 20 times higher than small luteal cells²⁰. Schmitt²¹ determined that the progesterone levels increased due to the development of accessory corpus luteum following GnRH and hCG treatments at day 5 after AI, although there were no significance between pregnancy rates. Same researchers also showed that the treatment mentioned above caused a new follicular wave and luteotropic effect on the present corpus luteum. Similarly, in our study differences in serum progesterone concentrations between lesireline and control groups were not significant ($p > 0.05$) ten days following AI.

In conclusion, administration of lesireline five days after AI increased serum progesterone concentrations ten days after AI in spite of no differences in pregnancy rates. Moreover, this study revealed the importance of BCS to increase pregnancy rates in cattle.

REFERENCES

1. **Stola R. und Leidl W:** Einsatz von Hormonen in der Reproduktion der Haustiere. *Tierärztliche Umsch*, 45, 154-163, 1990.
2. **Bostedt H, Sellinger M, Schuler G:** Ovulations- und Graviditätsergebnis nach Anwendung eines GnRH- oder β -Blocker-Präparates prä inseminationem beim Milchrind. *Tierärztliche Umsch*, 50, 311-316, 1995.
3. **Heuwieser E, Guard CL, Ferguson JD, Foote RH, Mansfeld R:** Zum Einfluss von Gonadotropin Releasing Hormon (GnRH) und Analogen auf die Konzeptionsrate beim Rind Eine Kritische Literaturübersicht. *Dtsch Tierärztl Wschr*, 97, 430-433, 1990.
4. **Anderson GA, Malmo J:** Pregnancy rates of cows given synthetic gonadotrophin-releasing hormone at the time of service. *Aus Vet J*, 62, 222-224, 1985.
5. **Kroworsch S, Krochmal W, Stolla R:** Einfluss einer einmaligen GnRH-Injektion am 5. Tag inseminationem auf die Trächtigkeitsrate beim Rind. *Tierärztliche Prax*, 28, 84-87, 2000.
6. **Prakash BS, Meyer HHD, Schallenberger E, Van De Wiel DVM:** Development of sensitive enzymimmunoassay for progesterone determination in unextracted bovine plasma using the second antibody techniques. *J Steroid Biochem*, 28 (6), 623-627, 1987.
7. **Thatcher WW, Drost M, Savio JD, Macmillan KL, Entwistle KW, Schmitt EJ, De La Sota RL, Morris GR:** New clinical uses of GnRH and its analogues in cattle. *Anim Reprod Sci*, 33, 27-49, 1993.
8. **Mann GE, Lamming GE:** Progesterone inhibition of the development of the luteolytic signal in cows. *J Reprod Fertil*, 104, 1-5, 1995.
9. **Edmonson AJ, Lean IJ, Weaver LD, Farver T, Webster G:** A body condition scoring chart for Holstein Dairy Cows. *J Dairy Sci*, 72, 68-78, 1989.
10. **Chenault J:** Effect of fertilerin acetate or busserelin on conception rate at first or second insemination in lactating dairy cows. *J Dairy Sci*, 73, 633-638, 1990.
11. **Klindworth HP, Hoedemaker M, Burfeindt D, Heilkenbrinker T:** Ovulationssynchronization (Ovsynch) in hochleistenden Milchviehherden: I. Fruchtbarkeitsparameter, Body Condition Score und Plasmaprogesteronkonzentration. *Dtsch Tierärztl Wschr*, 108, 11-19, 2001.
12. **Pennington JA, Hill DL, Callahan CJ, Brown CM, Brown, D:** Effect of preinsemination injection of gonadotropin-releasing hormone on reproductive performance of dairy cattle. *Bov Practitioner*, 20, 14-16, 1985.
13. **Macmillan KL, Taufu VK, Day AM:** Effects of an agonist of gonadotropin-releasing hormone (buserelin) in cattle. III. Pregnancy rates after a postinsemination injection during metöstrüs or diöstrüs. *Anim Reprod Sci*, 11, 1-10, 1986.
14. **Lewis GS, Caldwell DW, Rexroad CE, Dowlen HH, Owen JR:** Effect of gonadotropin-releasing hormone and human chorionic gonadotropin on pregnancy rate in dairy cattle. *J Dairy Sci*, 73, 66-72, 1990.
15. **Lucy ML, Stevenson JS:** Gonadotropin-releasing hormone at estrus: Luteinizing hormone, estradiol, and progesterone during the periestrual and post-insemination periods in dairy cattle. *Biol Reprod*, 35, 300-311, 1986.
16. **Stevenson JS, Phatak AP, Rettmer I:** Postinsemination administration of Receptal: Follicular dynamics, duration of cycle, hormonal responses and pregnancy rates. *J Dairy Sci*, 76, 2536-2547, 1993.
17. **Smith RD:** Interrelationships between energy balance and postpartum reproductive function in dairy cattle. *J Dairy Sci*, 72, 767-783, 1989.
18. **Villa-Godoy A, Hughes TL, Emery RS, Enright WJ, Schmidt MK, Stanisewski EP, Zinn SA, Fogwell RL:** Effect of energy balance and body condition on luteal function in heifers. *J Anim Sci*, 63 (1): 369, 1986.
19. **Bostedt H, Okyere K:** Auswirkung einer singulären GnRH-Injektion am 12. Tag post inseminationem auf die periphere LH- und Progesteronkonzentration bei wiederholt besamten Kühen. *Tierärztliche Umsch*, 43, 412-429, 1988.
20. **Niswender GD, Schwall RH, Fitz TA:** Regulation of luteal function in domestic ruminants: New concepts. *Recent Prog Horm Res*, 41, 101-105, 1985.
21. **Schmitt EJ, Diaz T, Barros CM, De La Sota RL, Drost M, Fredriksson EW, Staples CR, Thorner R, Thatcher WW:** Differential response of the luteal phase and fertility in cattle following ovulation of the first-wave follicle with human chorionic gonadotropin or an agonist of gonadotropin-releasing hormone *J Anim Sci*, 74(5), 1074-83, 1996.