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Prevention of embryonic death using different hormonal treatments in ewes

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Abstract: The purpose of this study was to evaluate different treatment protocols to prevent embryonic death in ewes. A total of 180 Akkaraman crossbred ewes and 10 healthy rams were used as material. The ewes were divided into 3 equal groups, with each of the 3 groups then separated into 3 subgroups. Ewes in estrus, determined with teaser rams, were exposed to mating. Three different treatment protocols of gonadotropin-releasing hormone (GnRH) analog (buserelin, intramuscularly) at a dose of 20 µg, vaginal sponges containing 30 mg fluorogestone acetate (FGA), and saline at a dose of 1 mL (control, intramuscularly) were applied on days 4, 12, and 16, respectively, for each subgroup after mating. No significant differences were observed in the pregnancy or multiple birth rates among any of the treatment groups. In the groups treated on days 4 and 12 after mating, the hormonal treatments gave lower rates of embryonic death compared to the control group ($P < 0.05$). In conclusion, the application of GnRH or FGA on days 4 and 12 after mating was found to be effective in preventing embryonic death in ewes.

Key words: Ewe, hormonal treatment, embryonic death

1. Introduction

Embryonic losses are observed within the first 3 weeks of pregnancy in ewes. The proportion of synchrony between the uterus and the developmental stage of the conceptus is one of the basic factors influencing fecundity. Embryonic losses are reported to be 20%–30% in ewes. Of this total loss, 70%–80% occurs between days 8 and 16 after insemination (1–3). The major reason for embryonic mortality is likely to be inadequate luteal function (4). To reduce embryonic losses and also to increase fertility, progesterone administration during early pregnancy has been applied to sheep and cattle (5–8). Progesterone usage after mating or insemination increases pregnancy rates and induces fetal growth. Additionally, progesterone administration may compensate for luteal insufficiency. The use of human chorionic gonadotropin (hCG) and gonadotropin-releasing hormone (GnRH) is an alternative method to reduce embryonic losses and hence to increase the pregnancy rate after different times of mating or insemination. The injection of GnRH between days 10 and 13 after mating has been shown to increase embryonic survival and pregnancy rate (5,7,8). These hormones can be used on different days of the cycle after mating, such as on days 4, 5, 11, and 12. Administration on day 12 after mating is suggested to be the critical period for maternal recognition of pregnancy in the ewe, because the corpus luteum starts to regress on day 12 in the estrous cycle. Hormonal treatments to increase progesterone production on day 12 after mating may prevent luteolysis of the corpus luteum and prostaglandin F2α ($PGF_{2\alpha}$) secretions (9).

The objective of this study was to determine the effects of different hormones applied after mating on the prevention of embryonic losses in ewes.

2. Materials and methods

This study was performed at a private farm in Konya during the sheep breeding season of 2007. A total of 180 Akkaraman crossbred ewes and 10 healthy rams were used as material. The ewes were divided into equal 3 groups, with each group then separated into 3 treatment subgroups. Each subgroup consisted of a total of 20 ewes. Ewes in estrus, determined with teaser rams, were exposed to mating. Mated ewes were then selected, and 3 different treatment protocols of GnRH analog (buserelin, intramuscularly) at a dose of 20 µg, vaginal sponge containing 30 mg fluorogestone acetate (FGA), and saline at a dose of 1 mL (control, intramuscularly) were applied on days 4, 12, and 16, respectively, for each subgroup

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after mating. Pregnancy and the number of embryos were detected on days 24, 30, and 36 after mating using real time B-mode ultrasound.

3. Results

As shown in the Table, no significant differences were observed in the pregnancy and multiple birth rates among any of the treatment groups. Hormonal treatments applied on days 4 and 12 after mating gave lower rates of embryonic death compared to the control group ($P < 0.05$). The rates of pregnancy, multiple births, and embryonic losses are illustrated in the Table.

3.1. Statistical analysis

The results of study were evaluated by the chi-square test. Differences with values of $P < 0.05$ were considered to be statistically significant. Statistical analyses were performed by using SPSS 11.5.

4. Discussion

The results of the present study indicated that the usage of GnRH and progesterone sponges on days 4 and 12 after mating did not improve the pregnancy rates but did reduce embryonic losses. Luteal function is the most important factor affecting embryo survival (10). Embryonic mortality is manifested as a constant loss of 20% to 30% of eggs in sheep. In an open breeding system, this situation may not present a problem, as nonpregnant ewes will ultimately return to service. Prolificacy may be lowered, however, if there is a significant partial loss of multiple ova with the female remaining pregnant. Embryo loss consists of fertilization failure, observable genetic damage, and idiopathic loss (11–13).

Progesterone is essential for pregnancy maintenance (14) and one of the important functions of the blastocyst is to ensure that the uterine luteolytic mechanism is counteracted (15). Progesterone and estrogen determine the proper function of the uterus in preparation for embryo development and implantation (16–18). Steroid imbalance may induce asynchrony between the embryo and uterus, i.e. alteration of embryonic growth rates relative to uterine timing (19–21). This could be viewed as a mechanism to reestablish synchrony, but generally asynchrony results in embryonic mortality (10,21). FGA and GnRH administration in the present study reduced the embryonic death rate through enhanced luteal function. These effects of the GnRH and FGA on embryonic survival are in agreement with the previous findings of Beck et al. (5) and Drew and Peters (22) in cattle but contradicted with the findings of Beck et al. (23), who did not detect any stimulatory effect of GnRH/FGA on the plasma progesterone in sheep. Dutt (24) strongly suggested that variability in progesterone concentration through estrus, and the timing and extent of the luteal rise, account for a proportion of embryonic mortality. Progesterone is necessary at a certain level after mating, as ewes with lower progesterone levels suffer greater embryo loss (25). Low periovulatory progesterone secretion may result in abnormal uterine function or poorly developed oocytes, and low steroid secretion may originate from follicles rapidly undergoing atresia (26).

Days 12 and 13 after mating are the critical period of maternal recognition of pregnancy, which coincides with the beginning of the regression of the corpus luteum in the estrous cycle (9). Progesterone provided by GnRH and FGA on day 12 may increase interferon-T production (27), which in turn prevents luteolysis by preventing $PGF_{2\alpha}$ secretion (9). Luteolysis may also be prevented by GnRH administration, because GnRH will ovulate or luteinize any developing follicles and hence decrease the estradiol production of the follicles. The decrease in estradiol production and increase in progesterone prevent the synthesis of $PGF_{2\alpha}$, providing pregnancy maintenance (28). The GnRH and FGA treatments may have increased plasma progesterone levels, which may in turn stimulate the secretion of embryonic substances from

Table. Effect of different hormone administrations applied after mating on the rates of pregnancy, multiple births, and embryonic death.

Day	Group	Pregnancy rate (%)	Multiple birth rate (%)	Embryonic death rate (%)
Day 4	GnRH	85	41.2	17.6[a]
	FGA	90	33.3	5.5[a]
	Control	90	33.3	38.8[b]
Day 12	GnRH	90	50.0	5.6[a]
	FGA	90	38.8	5.6[a]
	Control	95	26.3	31.6[b]
Day 16	GnRH	85	29.4	29.4
	FGA	90	38.8	22.2
	Control	80	37.5	31.2

a, b, c: Different superscripts within the same column show significant differences ($P < 0.05$).

the uterus. Wilmut et al. (29) demonstrated that a uterine environment advanced by treatment with the exogenous progesterone accelerated conceptus development. Fluctuations in the concentration of plasma progesterone appear to have implications for embryo survival (25). In this study, on postmating day 12, hormonal treatments such as GnRH and FGA decreased the embryonic death rate. In conclusion, the application of GnRH or FGA on days 4 and 12 after mating was found to be effective in preventing embryonic death in ewes.

References

1. Bolet, G.: Timing and extent of embryonic mortality in pigs, sheep and goats; genetic variability. In: Sreenan J.M., Diskin, M.G., Eds. Embryonic Mortality in Farm Animals. Martinus Nijhoff, The Hague, 1986; 12–43.
2. Michels, H., Vanmontfort, D., Dewil, E., Decuypere, E.: Genetic variation of prenatal survival in relation to ovulation rate in sheep: a review. Small Rumin. Res., 1998; 29: 129–142.
3. Jainudeen, M.R., Hafez, E.S.E.: Reproduction failure in females. In: Hafez, E.S.E., Ed. Reproduction in Farm Animals. 6th ed., Lea & Febiger, Philadelphia, 1993; 261–286.
4. Nancarrow, C.D.: Embryonic mortality in the ewe and doe. In: Zavy, M.T., Geisart, R.D., Eds. Embryonic Mortality in Domestic Species. CRC Press, Boca Raton, FL, USA, 1994; 79–97.
5. Beck, N.F.G., Peters, A.R., Williams, S.P.: The effect of GnRH agonist (buserelin) treatment on day 12 post-mating on the reproductive performance of ewes. Anim. Prod., 1994; 58: 243–247.
6. Thatcher, W.W., Moreira, F., Santos, J.E.P., Mattos, R.C., Lopes, F.L., Pancarci, S.M., Risco, C.A.: Effects of hormonal treatments on reproductive performance and embryo production. Theriogenology, 2001; 55: 75–89.
7. Cam, M.A., Kuran, M., Yildiz, S., Selcuk, E.: Fetal growth and reproductive performance in ewes administrated GnRH agonist on day 12 post-mating. Anim. Reprod. Sci., 2002; 72: 73–82.
8. Cam, M.A., Kuran, M.: GnRH agonist treatment on day 12 post-mating to improve reproductive performance in goats. Small Rum. Res., 2004; 52: 169–172.
9. Bazer, F.W., Ott, T.L., Spencer, T.E.: Maternal recognition of pregnancy: comparative aspects: a review. Placenta, 1998; 12: 375–386.
10. Wilmut, I., Sales, D.I., Ashworth, C.J.: Maternal and embryonic factors associated with prenatal loss in mammals. J. Reprod. Fertil., 1986; 76: 851–864.
11. Willingham, T., Shelton, M., Thompson, P.: An assessment of reproductive wastage in sheep. Theriogenology, 1986; 26: 179–188.
12. Thatcher, W.W., Staples, C.R., Danet-Desnoyers, G., Oldick, B., Schmitt, E.P.: Embryo health and mortality in sheep and cattle. J. Anim. Sci., 1994; 72: 16–30.
13. Kleemann, D.O., Walker, S.K.: Fertility in South Australian commercial Merino flocks: sources of reproductive wastage. Theriogenology, 2005; 63: 2075–2088.
14. Foote, W.D., Gooch, L.D., Pope, A.L., Casida, L.E.: The maintenance of early pregnancy the ovariectomized ewe by injection of ovarian hormones. J. Anim. Sci., 1957; 16: 986–989.
15. Bazer, F.W.: Establishment of pregnancy in sheep and pigs. Reprod. Fertil. Dev., 1989; 1: 237–242.
16. Bindon, B.M.: The role of progesterone in implantation in the sheep. Aust. J. Biol. Sci., 1971; 24: 149–158.
17. Miller, B.G., Moore, N.W.: Effect of progesterone and oestradiol on RNA and protein metabolism in the genital tract and on survival of embryos in the ovariectomized ewe. Aust. J. Biol. Sci., 1976; 29: 565–573.
18. Miller, B.G., Moore, N.W., Murphy, L., Stone, G.M.: Early pregnancy in the ewe: effects of oestradiol and progesterone on uterine metabolism and embryo survival. Aust. J. Biol. Sci., 1977; 30: 279–288.
19. Lawson, R.A.S., Cahill, L.P.: Modification of the embryo-maternal relationship in ewes by progesterone treatment early in the oestrous cycle. J. Reprod. Fertil., 1983; 67: 473–475.
20. Lawson, R.A.S., Parr, R.A., Cahill, L.P.: Evidence for maternal control of blastocyst growth after asynchronous transfer of embryos to the uterus of the ewe. J. Reprod. Fertil., 1983; 67: 477–483.
21. Pope, W.F.: Uterine asynchrony: a cause of embryonic loss. Biol. Reprod., 1988; 39: 999–1003.
22. Drew, S.B., Peters, A.R.: Effect of buserelin on pregnancy rates in dairy cows. Vet. Rec. 1994; 134: 267–269.
23. Beck, N.F.G., Jones, M., Davies, B., Mann, G.E., Peters, A.R.: The effect of GnRH analogue (buserelin) treatment on day 12 post-mating on ovarian structure and plasma progesterone and oestradiol concentration in ewes. Anim. Sci., 1996; 63: 407–412.
24. Dutt, R.H.: Fertility rate and embryonic death loss in ewe early in the breeding season. J. Anim. Sci., 1954; 11: 464–473.
25. Ashworth, C.J., Sales, D.L., Wilmut, I.: Patterns of progesterone secretion and embryonic survival during repeated pregnancies in Damline ewes. Proceedings of the 10th International Congress Animal Reproduction and Artificial Insemination, 1984; 2: 74.1–74.3.
26. Kittok, R.J., Stellflug, J.N., Lowry, S.R.: Enhanced progesterone and pregnancy rate after gonadotropin administration in lactating ewes. J. Anim. Sci., 1983; 56: 652–655.
27. Thatcher, W.W., Meyer, M.D., Danet-Desnoyers, G.: Maternal recognition of pregnancy. J. Reprod. Fertil., 1995; 49: 15–28.
28. Khan, T.H., Beck, N.F.G., Mann, G.E., Khalid, M.: Effect of post-mating GnRH analogue (buserelin) treatment on $PGF_{2\alpha}$ release in ewes and ewe lambs. Anim. Reprod. Sci., 2006; 95: 107–115.
29. Wilmut, I., Sales, D.I., Ashworth, C.J.: Physiological criteria for embryo mortality: is asynchrony between embryo and ewe a significant factor? Genet. Reprod. Sheep, 1985; 275–289.