The Effect of Post-Mating Diclofenac Sodium Injections on The Pregnancy Rate of The Lactating Goats Synchronized by Buck Effect at The Initiation of Breeding Season

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SUMMARY

This research was carried out to determine the effect of Diclofenac sodium injections at 15-16th days post mating on the pregnancy rate of the lactating goats. A total of 107 Damascus goats were synchronized by buck effect. The 54 goats between 3-6 years of age which were showing estrus and mated between the 9-15th days after the induction of buck to the herd constituted the material of the study. The goats were randomly assigned to two groups as diclofenac sodium (DFS) and control. Diclofenac sodium was injected to DFS group (n=27) intramuscularly with a dose of 2.5 mg/kg in the mornings of the 15-16th days postmating with 24 hours intervals twicely. Placebo was injected to control group at the same days and hours. The ultrasonographic examination performed at the 50th day post insemination revealed that while 77.8% (21/27) of the goats was determined to be pregnant in DFS group, the %51.9 (14/27) of the goats was found to be pregnant in control group. The pregnancy rates of the DFS and control groups were found to be statistically different (p<0.05). It was concluded that Diclofenac sodium injections at 15 and16th days post mating to lactating goats at the initiation of breeding season improved the pregnancy rates.

Key Words   Goat, Diclofenac Sodium, Fertility

Üreme Sezonu Başlangıcında Teke Etkisi İle Senkronize Edilen Laktasyondaki Keçilerde Aşım Sonrasında Yapılan Diklofenak Sodyum Enjeksiyonlarının Gebelik Oranı Üzerine Etkisi

ÖZET

Çalışma; aşım sezonu başlangıcında teke etkisi ile östrüsleri uyarılan keçilere, aşım sonrası 15-16. günlerde diklofenak sodyum enjeksiyonlarının gebelik oranı üzerindeki etkisinin belirlenmesi amacıyla yapıldı. Toplam 107 baş Şami Keçisi teke etkisi ile senkronize edildi. Yaşları 3-6 arasında değişen, tekelerin sürüye katılmasından 9-15 gün arasında östrüs gösteren ve aşım yaptırılan 54 baş keçi araştırmanın hayvan materyalini oluşturdu. Keçiler Diklofenak sodyum (DFS) ve kontrol olmak üzere rastgele 2 eşit gruba ayrıldı. DFS grubuna diklofenak sodyum aşımlardan sonraki 15-16. günlerin sabahında 24 saat ara ile, iki kez, 2.5 mg/kg dozda kas içi enjekte edildi. Kontrol grubuna ise aynı gün ve saatlerde plasebo enjeksiyonu yapıldı. 50. günde yapılan ulrasonografik muayenede diklofenak sodyum grubunda gebelik oranı %77.8 (21/27), kontrol grubunda ise %51.9 (14/27)olarak bulundu. DFS ve Kontrol gruplarının gebelik oranları istatistik olarak farklı bulundu (p<0.05). Sonuç olarak laktasyondaki keçilerde aşım sezonu başlangıcında aşım sonrası 15-16. günlerde yapılan diklofenak sodyum enjeksiyonlarının gebelik oranlarını artırdığı kanaatine varıldı.

Anahtar Kelimeler   Keçi, Diklofenak Sodium, Fertilite

INTRODUCTION

In small ruminants, early embryonic loss is the major item of postfertilization losses also is a significant reason of economic loss in animal production and agriculture. During the first 3 weeks of pregnancy, 30–40% of fertilized eggs are lost in sheep and goats (Nancarrow 1994; Cam and Kuran 2004). Of this total loss, 70–80% occurs between days 8 and 16 after insemination (Sreenan et al. 1996).

In ruminants, secretion of IFN-τ by the conceptus is responsible for the maintenance of corpus luteum function and establishment of pregnancy (Guillomot et al. 1998; Ealy et al. 2004). In the goat, it was demonstrated that the trophoblast secreted two different isotypes of IFN-τ until day 17 of gestation (Somi 2003). IFN-τ, produced by the conceptus at the time of pregnancy recognition may have a direct action on endometrial cells by; preventing the appearance of functional estrogen and oxytocin receptor or blocking pulsatile release of $PGF_{2\alpha}$ in ruminants. At the time of pregnancy recognition, the endometrial release of pulsatile $PGF_{2\alpha}$, must be blocked (Asselin et al. 1997).

The nonsteroidal anti-inflammatory drugs (NSAIDs) are chemically diverse group of agents that has similar pharmacologic properties and are widely used to control pain and inflammation. Diclofenac sodium, a NSAID, is a nonselective cyclic oxygenase (COX) inhibitor and may exert its analgesic effect as a result of prostaglandin

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synthesis inhibition (EMEA 2004). It was reported that the $PGF_{2\alpha}$ synthesis could be delayed by the administration of NSAID's (Asselin et al. 1997). Güzeloğlu et al. (2007) reported that such a delay would provide an extra time for a slowly developing but viable embryo to secrete sufficient IFN- τ to inhibit the luteolitic secretion of $PGF_{2\alpha}$. NSAID's such as flunixin meglumine, was used at embryo transfer in cows (Scenna et al. 2005), in heifers (Guzeloglu et al. 2007) and in repeat breeding heifers (Dogruer et al. 2008) to increase the pregnancy rates also stated to increase the pregnancy rates. No literature was encountered on the effects of both NSAID's and DFS on fertility in goats.

Therefore this research was conducted to determine the effects of DFS injections approximately at the time of maternal recognition on the pregnancy rates of lactating goats.

MATERIALS and METHODS

The research was carried out in July of 2007 in Hatay that is located in eastern Mediterranean region of Turkey which is in 35° 52´ and 37° 04´ North latitude and 35° 40´ and 36° 35´ east longitude. During the study the average annual temperature was 35 °C during the day, and 25 ºC during the night. The area receives an average annual precipitation of 570-1160 mm^3.

Buck effect was used for induction and synchronization of estrus in the goats. At the beginning of the research the bucks were isolated from the goats for 28 days. The bucks were fed with commercial feeds and lentil straws and the bucks also received daily 250 gr additional barley. The water and mineral salts were also ad libitum for the bucks. After introducing the bucks to the herd the estrous of the goats were observed and recorded throughout the day. The goats which were detected to be in estrous were mated with five fertile bucks and recorded.

A total of 107 Damascus goats were used for synchronization by buck effect. Of these fifty-four, healthy, multi-parous, dairy, Damascus goats constituted the animal material of the study. The ages of the goats were ranging between 3 to 6. These goats showed the signs of estrus and mated between the 9-15th days after the induction of buck to the herd. Remaining 53 goats were excluded from the study. The goats were hand milked daily in the mornings and the goats were suckling the kids after the milking. The goats were fed in the natural pasture during the days and were given commercial feeds and lentil straws in the evenings. The goats in the research received 250 gr additional barley per animal 15 days before and 15 days after the matings. The water and mineral salt were ad libitum during the study.

Diclofenac sodium (REUFLOGIN;VETAS) was injected to DFS group (n=27) intramuscularly with a dose of 2.5 mg/kg LW in the mornings of the 15-16th days postmating, with an interval of 24 hours, twicely. Placebo was injected to control group at the same days and hours post mating.

The pregnancy diagnosis of both DFS and control groups was performed with a B-mode sector array 5-7.5 mHz ultrasonography via abdominal wall 55 days after mating. The difference of the pregnancy rates among the groups was detected with chi-square test in SPSS 11.0 packet programme.

RESULTS

The pregnancy rates were detected as 77.8% (21/27) and 51.9% (14/27) in the DFS and control groups respectively (Fig 1). The pregnancy rates of the DFS and control groups were found to be statistically different (p<0.05).
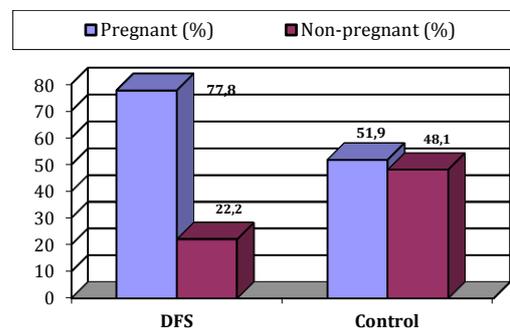

Fig. 1. The pregnancy rates of DFS and Control groups

DISCUSSION and CONCLUSION

Embryonic loss is an important reason of economic decrement in goat production. In small ruminants the ratio of embryonic loss varies between 8.9 to 20.1 (Regassa et al. 2007; Sarıbay and Erdem 2007) in ewes, and averaged 10.8 -11.0 % in goats (Engeland et al. 1998, Anwar and Ahmad 1999). A portion of these losses is thought to occur as a result of asynchrony between the conceptus and uterine endometrium and/or inadequate production of the antiluteolytic signal (interferon tau (IFN-τ)) by conceptus, (Martinod et al. 1991). Studies carried out for prevention of embryonic loses including both postmating GnRH (Alaçam et al. 1999; Cam and Kuran 2004) and hCG administrations (Fonseca et al. 2005) aimed sustaining high progesterone levels in goats. These studies revealed that serum progesterone levels remained high in both GnRH and hCG treated goats. Although Alaçam et al. (1999) reported that administration of GnRH at 12th day postmating increased both progesterone levels and fertility in Angora goats, Fonseca et al., (2005) stated that mean plasma progesterone concentration was higher in the hCG-treated does than in the control animals but this did not cause an increase in pregnancy rate. High plasma progesterone concentration at the onset of pregnancy did not seem to have a beneficial effect on the establishment of pregnancy. The exposure of the endometrium to progesterone for 10 to 12 days, as occurs during diestrus, not only prepares the uterus for establishment of pregnancy, but also activates mechanisms for endometrial production of luteolytic PGF in the event that pregnancy is not established (Morgan et al. 1993). Progesterone increases phospholipid stores (Spencer et al. 1996) and prostaglandin synthase activity necessary for conversion of arachidonic acid to $PGF_{2\alpha}$. Elevated uterine luminal concentrations of $PGF_{2\alpha}$ is pointed out to be negatively associated with embryo quality and pregnancy rates (Scenna 2005). In this study it was aimed to diminish the embryonic loss by diclofenac sodium administrations on day 15-16 postmating with 24 hours interval with the help of COX inhibition also temporarily suppressing the intrauterine $PGF_{2\alpha}$. In this study the pregnancy rate was found to be 77.8% and 51.9% in DFS and control groups respectively. This result is thought to be the result of reduced uterine luminal $PGF_{2\alpha}$ levels because of prostaglandin inhibition by DFS.

Luteal regression is delayed or prevented by inhibiting synthesis of $PGF_{2\alpha}$ with non-steroidal anti-inflammatory drugs or by immunizing animals against $PGF_{2\alpha}$ (Silvia 1999). Güzeloğlu et al. (2007) reported that poorly developed embryos could be viable but slower to develop, so the process for maternal recognition of pregnancy induced by IFN were not initiated at the appropriate time and also stated that flunixin meglumine could have exerted an inhibitory effect on $PGF_{2\alpha}$ in heifers. In the current study, DFS which is another NSAID, may have exerted the same effect on $PGF_{2\alpha}$ and gained time for prospectively poor developed embryos in goats.

Early embryonic deaths occur on the days of maintenance of the corpus luteum. The critical period for embryo recognition in goats occurs around Day 15 of pregnancy (Weise et al. 1993). Gnatek et al. (1989) demonstrated that a conceptus must be present in the uterus between Days 15 and 17 to prolong CL life span in goats. Owing to the critical maternal recognition days cited in the literatures, the diclofenac sodium applications were carried out on days 15-16 of pregnancy in the current study.

The estrous synchronization of small ruminants include hormonal (progestogens, PMSG, hCG, hMG, FSH or GnRH) and non-hormonal treatments such as male effect (Chemineau 1987, Mellado et al. 2000). The introduction of bucks into the herd induces synchronous ovulations in the following days. The contact with males causes, an immediate increase in the number and the amplitude of LH pulses, which induces the appearance of an LH pre-ovulatory surge to start ovulation in the goats. The first induced ovulations are silent (i.e. not associated with oestrous behavior) in 40% of the does, and are followed by a short luteal phase, of 5-days duration, in 75% of the does. Later on, oestrous and ovarian cycles are restored (Chemineau 1987). In the current study at the initiation of the breeding season it was observed that the goats were successfully synchronized by buck effect.

The NSAID's were used to increase the pregnancy rates in cattle practice. Scenna et al. (2005) found that overall pregnancy rates of cows receiving flunixin meglumine improved pregnancy rate compared to control group. Pregnancy rates following transfer of good quality embryos did not differ among treatments. However, pregnancy rates of fair quality embryos were higher in animals receiving flunixin meglumine than the controls. Guzeloglu et al. (2007) obtained a higher pregnancy rates in heifers treated with flunixine meglumine than the controls. Also Doğruer et al. (2007) obtained a higher pregnancy rates in repeat breeding heifers treated with flunixine meglumine than the controls. Scenna et al. (2005) stated that uterine release of $PGF_{2\alpha}$ is elevated following embryo transfer and administration of a $PGF_{2\alpha}$ synthesis inhibitor at the time of embryo transfer improved pregnancy rates in cows. DFS belongs to non-steroidal anti-inflammatory drugs more specifically to the phenyl acetic acid derivates. As all NSAID's DFS shows the anti-inflammatory, analgesic and anti-pyretic effects due the decrease of prostaglandins by non-selective COX inhibition (EMEA 2003). The 77.8 % pregnancy rate obtained in DFS group versus 51.9 % pregnancy rate in control showed that similar effects could be obtained by DFS in goats. Also the chance of being pregnant by injection of DFS in goats increases nearly 1.5 times higher than the control group.

Diclofenac is intended for treatment in cattle and swine as an anti-inflammatory agent at a dose of 2.5 mg/kg by intramuscular route (EMEA 2003). In the recent study the drug was used to increase the fertility and pregnancy rates. The drug is eliminated in sheep in 2-3 hours for all routes of administration and the elimination of the drug is especially via enterohepatic circulation (Altaher et al. 2006). Although this drug is not provided for use in animals from which milk is produced for human consumption, no radiomateric residues were detected form milk (EMEA 2003). It has been shown that 24 h after the end of a 1-day infusion of 150 mg of diclofenac sodium following cesarean section, no drug was found in the colostrum (Ostensen and Musby 1985). Diclofenac is preferred in this research for undetected milk residues, for it is used in lactating goats.

It was concluded that Diclofenac sodium injections at 15 and16th days post mating to lactating goats at the initiation of breeding season improved the pregnancy rates.

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