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EFFECT OF ESTRUS SYNCHRONIZATION PROTOCOLS INCLUDING $PGF_{2\alpha}$ AND GnRH ON FERTILITY PARAMETERS IN HAIR GOATS DURING BREEDING SEASON

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ABSTRACT

The present study was performed to determine the effect of $PGF_{2\alpha}$ and GnRH injections in different time intervals on fertility in Hair goats during breeding season. Totally, 80 primiparous Hair goats were divided into four groups (n=20 in each) according to the body condition scores of the animals. The animals in groups were treated as follows; first group (Ovsynch), GnRH on day 0, $PGF_{2\alpha}$ on day 7 and GnRH on day 9; second group (2PG-G), $PGF_{2\alpha}$ on day 0, $PGF_{2\alpha}$ on day 7 and GnRH on day 9; third group (PG-G), $PGF_{2\alpha}$ on day 7 and GnRH on day 9; fourth group (G), GnRH on day 9. Bucks were included into the groups on day 10-14 and insemination was performed during standing heat. Blood samples were collected to determine possible embryonic losses on day 21 for further analysis of serum progesterone concentrations. Transabdominal ultrasonography was performed to detect pregnancy on day 40-45. It was determined that in Ovsynch, 2PG-G, PG-G and G groups, estrous rate was 100%, 95%, 95% and 85%; pregnancy rate was 85%, 95%, 95% and 85%; kidding rate was 100%, 78.9%, 94.7% and 88.2%; twinning rate was 41.2%, 40%, 22.2%, 20%; litter size was 1.41, 1.40, 1.22, 1.20, respectively. However, these parameters did not show any significant difference between groups ($P > 0.05$). In conclusion, it is suggested that all treatment procedures may be used for estrous synchronization in Hair goats during breeding season due to the satisfactory results.

Keywords: Goat, Fertility, Synchronization, Ovsynch, GnRH, $PGF_{2\alpha}$.

INTRODUCTION

Estrous synchronization can be managed by different programs including single or combined use of hormones such as progestagens, estrogens, melatonin, $PGF_{2\alpha}$, PMSG, hCG, GnRH in breeding, transition or anestrous seasons in goats (Alaçam, 2005; Titi *et al*., 2010; Alkan *et al*., 2012).

During breeding season, progesterone impregnated intra-vaginal sponge treatment combined with PMSG is commonly used (Holtz, 2005). However, the vaginitis is the expected side effect of intravaginal sponge treatment together with the vaginal discharge following the removal of the device (Ataman and Akoz, 2006). It has been reported that repeated progesterone/ PMSG treatments may cause anti-PMSG antibody and hence, infertility (Baril *et al.*, 1992; Roy *et al.*, 1999; Redmer *et al.*, 2000). The high concentrations of anti-PMSG antibody delay the standing heat and LH peak. Therefore, hCG and GnRH has been recently used as an alternative to PMSG (Alaçam *et al*., 1999). On the other hand, adding GnRH injection(s) to progesterone based protocols improve the success of estrous synchronization programs but do not increase the fertility (Saribay *et al*., 2012).

The development of more practical and/or economic synchronization programs to minimalize the possible disadvantages of protocols in small ruminant breeding still maintains its importance for researchers and breeders (Holtz *et al.*, 2008; Alkan *et al*., 2012).

Nowadays, the Ovsynch protocol improved for cattle breeding is began to be used in sheep and goat breeding (Deligiannis *et al*., 2005; Amiridis *et al*., 2006; Holtz *et al*., 2008; Ashmawy, 2011; Riaz *et al.*, 2012) to synchronize the estrous and ovulation (Pursley *et al*., 1995).

First GnRH injection used in Ovsynch protocol initiates the release of FSH and LH from anterior pituitary. If the ovulation occurs following the first GnRH of Ovsynch, follicular wave emergence may be synchronized. A follicle on the ovary grows and becomes a dominant follicle and the progesterone concentration is decreased by regression corpus luteum after $PGF_{2\alpha}$ on day 7. Follicular growth continues and preovulatory follicle grows. The second GnRH injection on day 9 re-initiates the realising of LH from pituitary and ovulation of preovulatory follicle is achieved 24-32 hours later. Therefore, timed artificial insemination can be performed 16-20 hours after this injection (Pursley *et al.*, 1997; Geary *et al.*, 1998; Stevenson *et al.*, 1999).

In this study, the effect of $PGF_{2\alpha}$ and GnRH injections performed at different time intervals on fertility parameters was evaluated during breeding season in Hair goats.

MATERIALS AND METHODS

Experimental site: The study was conducted in Nigde province, Turkey (37° 25′ - 38° 58′ north latitude, 33° 10′- 35° 25′ east longitude and 1229 m altitude) on October-December 2012 in Hair goats during breeding season. A total of 80 multiparous Hair goat aged between 2 and 4 years, weighing 35-45 kg and eight bucks aged between 2 and 5 which were used for mating were included to the study.

Housing and Feeding: During the experiments, animals were pastured in the daytime and housed in semi-opened barns. No additive feeding regimen was used for goats before or after mating, whereas bucks were fed by additive 1 kg/day/head barley meal. Clean water and licking block were administered *ad libitum*.

Groups and Treatment: Body condition scoring (BCS; 1 to 5) was recorded at the first day of experiments and the goats were divided into four groups (n = 20 in each) followed as Ovsynch (BCS, 1.87), 2PG-GH (BCS, 1.98), PG-GH (BCS, 1.85) and GH (BCS, 1.86) based on BCS. The goats were treated by the synchronization protocols as indicated in Table 1. The treatments were started regardless of the day of estrous cycle and the first day of the treatments was considered as Day 0.

Table 1. The synchronization protocol schedule applied to the goats in groups.

Groups	Day 0	Day 7	Day 9	Day 10-14
Ovsynch	GnRH*	$PGF_{2\alpha}$**	GnRH	Mating
2PG-GH	$PGF_{2\alpha}$	$PGF_{2\alpha}$	GnRH	
PG-GH	-------	$PGF_{2\alpha}$	GnRH	
GH	-------	-------	GnRH	

* Buserelin acetate, 4 µg, intramuscular, Receptal, Intervet.
** Dinoprost tromethamine, 5 mg, intramuscular, Dinolytic, Pfizer.

Observation of Standing Heat and Insemination: Standing heat was determined at early in the morning (06.00-07.00 am) and late in the evening (06.00-07.00 pm) by one hour observation using bucks. The bucks were separated from the goats until the end of protocols for mating and then allowed to mate on day 10-14 of experiment (for 5 days).

Blood Sampling and Ultrasonography: Blood samples of mated goats were collected into anticoagulant vacutainer tubes via jugular venepuncture 21 days after mating. Samples were transferred to laboratory at 4 °C and centrifuged at 3000 rpm for 10 minutes. Sera were transferred in Eppendorf tubes and were kept in a freezer at −20°C for later estimation of P_4 concentrations. Serum P_4 concentrations were determined by electrochemiluminiscence immunoassay (ECLIA) using a commercial test kit (Elecsys Progesterone II, Roche Diagnostics GmbH, Germany) in an immunologic test analyser (cobas e 601, Roche Diagnostics GmbH, Germany). Transabdominal ultrasonography (Pie Medical 100 Falco Vet Model 7.5 MHz probe, Netherlands) was performed to confirm pregnancy 40-45 days after mating.

Fertility Parameters: The parameters for standing heat, pregnancy, birth, single kidding, twinning and kid yields were calculated as described below.

Estrous rate; (number of standing heat/ number of synchronized goats) x 100

Pregnancy rate; (number of pregnant goat/number of synchronized goats) x 100

Kidding rate; (number of goat having birth/number of pregnant goats) x 100

Single kidding rate; (number of goat with single kid/ number of goat having birth) x 100

Twinning rate; (number of goat with twin kids/number of goat having birth) x 100

Litter size; number of kid born/number of goat having birth

Fecundity; number of kid born/number of goats synchronized

Statistical analysis: Chi-square test was conducted to analyse of fertility parameters, whereas BCS was evaluated by variance analyse. All statistical analysis was performed with the SPSS software for windows (15.0). Statistical significance was declared at $P<0.05$.

RESULTS

Fertility parameters obtained in groups are given at Table 2. Accordingly, the estrous (100%, 95%, 95% and 85%), pregnancy (85%, 95%, 95% and 85%), kidding (100%, 78.9%, 94.7% and 88.2%), twinning (41.2%, 40%, 22.2% and 20%) and litter size (1.41, 1.40, 1.22 and 1.20) rates between Ovsynch, 2PG-GH, PG-GH and GH groups did not show any significant difference.

Blood progesterone concentrations of Ovsynch, 2PG-GH, PG-GH and GH groups were 7.744, 7.631, 6,918 and 9.758 ng/ml, respectively. The progesterone concentrations detected 21 days after mating did not differ between groups. However, no embryonic loss was detected in groups, when the blood progesterone concentrations and transabdominal ultrasonography findings compared.

Table 2. Estrous, pregnancy, birth, single kidding, twinning and kid yield rates obtained in Ovsynch, 2PG-GH, PG-GH and GH groups.

Parameters	Ovsynch		2PG-GH		PG-GH		GH		P
Estrous rate (%)	100	(20/20)	95	(19/20)	95	(19/20)	85	(17/20)	0.317
Pregnancy rate (%)	85	(17/20)	95	(19/20)	95	(19/20)	85	(17/20)	0.565
Kidding rate (%)	100	(17/17)	78.9	(15/19)	94.7	(18/19)	88.2	(15/17)	0.640
Single kidding rate (%)	58.8	(10/17)	60	(9/15)	77.8	(14/18)	80	(12/15)	0.677
Twinning rate (%)	41.2	(7/17)	40	(6/15)	22.2	(4/18)	20	(3/15)	0.677
Litter size	1.41	(24/17)	1.40	(21/15)	1.22	(22/18)	1.20	(18/15)	0.377
Fecundity	1.20	(24/20)	1.05	(21/20)	1.10	(22/20)	0.90	(18/20)	0.501

DISCUSSION

It has been reported that Ovsynch protocol has promising results (Deligiannis *et al*., 2005; Amiridis *et al*., 2006) and a low cost protocol in sheep and goat (Ataman and Akoz, 2006). Ovsynch protocol can be used for synchronization of ovulations and for decreasing the lambing intervals in sheep during breeding season (Ashmawy, 2011; Ashmawy, 2012). Alnimer *et al.*, (2005), reported that the combination of GnRH and PGF2α injections and double PGF2α injections 10 days interval had similar pregnancy rates in sheep during breeding season. In other study, it was stated that GnRH+PGF2α injections positively influenced the estrous and pregnancy rates in sheep synchronized by progesterone during anestrus season (Husein and Kridli, 2003). The combination of GnRH+$PGF_{2\alpha}$ and $PGF_{2\alpha}$+$PGF_{2\alpha}$ injections have 93.7% vs 86.6% estrous rate, 85.7% vs 84.6% pregnancy rate, 83.3% vs 81.8% kidding rate and 1.70 vs 1.66 kid yield in sheep during breeding season (Ataman and Akoz, 2006). Majdi *et al.*, (2014), reported that fertility parameters obtained in Nubian goats following the 11 days apart double intramuscular injections of 125µ cloprostenol on days postpartum 45, 60 and 90 were low and the day of treatment did not show any significant difference.

In addition, pregnancy rates following artificial insemination in sheep synchronized by combination of GnRH+ $PGF_{2\alpha}$ are 50%, whereas the pregnancy rate can be increased till 90-95% by natural mating (Deligiannis *et al*., 2005; Amiridis *et al*., 2006). It has been reported that P4+PGF+hCG is the best option to induce and synchronize estrous as well as ovulation regarding the administration of GnRH during the natural anestrus season in goats (González-Álvarez *et al*., 2016), Acar *et al.*, (2013) reported that use of norgestomet implants and FGA sponges in combination with eCG and PGF2α were effective for estrous response, onset of estrous, fecundity, prolificacy and fertility in nulliparous Saanen does at an age of 7-9 months under local conditions at the end of the transition period.

It has been assumed that Ovsynch protocol is an alternative to vaginal sponge treatments, since the Ovsynch protocol as compared to vaginal sponge treatment has high results for kidding (58% vs 46%) and fecundity rates (1.86 vs 1.83) in goats during the breeding season (Holtz *et al.*, 2008) Nur *et al.*, (2013) found that progesterone based Co-synch vs Ovsynch protocols had 92% vs 84% estrous rate, 38% vs 24% pregnancy, kidding rates 1.4 vs 1.2, respectively in Saanen goats. The authors (Nur *et al.*, 2013) also suggested that Co-synch model was more advantageous than Ovsynch protocol due to its low labour cost. Estrous rate as well as pregnancy and kidding rates obtained in this study were higher than those Nur *et al.*, (2013) reported, whereas fecundity rates were similar. Bowdridge *et al.*,(2013), observed that NCSynch-TA protocol (PGF injection on Day 1, GnRH injection on Day 7 and second PGF injection 7 days later and timed artificial insemination after 72 h) showed 88%, 53%, 51% for estrous, pregnancy and kidding rates, respectively in Boer and crossbreed-Boer goats during breeding season. It was seen that estrous, pregnancy and kidding rates detected in this study were higher than the results of Bowdridge *et al.*, 2013. Titi *et al.*,(2010) reported that control, S (progesterone impregnated intravaginal sponge and eCG), GP (GnRH and $PGF_{2\alpha}$) and GSP (GnRH, progesterone impregnated intravaginal sponge and $PGF_{2\alpha}$) groups showed 50%, 87%, 73% and 87% for kidding rate and 1.8, 1.5, 1.9 and 2.0 for fecundity rate, respectively in Damascus goats and it was concluded that progesterone impregnated intravaginal sponge treatment combined with GnRH and $PGF_{2\alpha}$ injections might improve the success of estrous synchronization and fertility. Another study (Riaz *et al.*, 2012) in which Ovsynch protocol and ten days apart double injection of $PGF_{2\alpha}$ were compared in goats during breeding season has showed similar pregnancy rate (60% vs 78%) and fecundity rate (1.5 vs 1.7). Therefore, the authors assumed that Ovsynch protocol might be used instead of other protocols which based on the using of steroids (Riaz *et al.*, 2012). In this study, estrous, pregnancy, kidding, twinning and litter size in Ovsynch group were 100%, 85.0%, 41.2% and 1.41, respectively. Pregnancy rate was higher than those reported by Holtz *et al.*, (2008) and Riaz*et al.*, (2012), whereas mean litter size was lower. It is suggested that

the discrepancies may be due to lower twinning rate of Hair goats.

Estrous, pregnancy, kidding, twinning and fecundity rates did not show any significant difference in 2PG-GH, PG-GH and GH groups. In this study, pregnancy rates observed in Ovsynch, 2PG-GH, PG-GH and GH groups were higher than those reported by Riaz *et al.,* (2012), while fecundity was lower. It was found that estrous, pregnancy, kidding and fecundity rates were similar with other reports (Deligiannis *et al.,* 2005; Amiridis *et al.,* 2006; Ataman and Akoz, 2006). Discrepancies above may be caused by some factors such as genotype, housing and management. However, higher pregnancy rate in the combination of GnRH+$PGF_{2\alpha}$ injections may be associated with the occurrence of GnRH induced ovulation (Deligiannis *et al.,* 2005; Alkan *et al.,* 2012).

It is known that luteal regression starts around the day 12-13 in non-pregnant goats and the concentrations of progesterone decrease, whereas pregnant goats display higher progesterone concentrations 5-8 ng/ml on average on day 13 (Homeida and Cooke, 1982). The injection of gonadorelin causes not only luteotrophic support but also prevents the luteolytic mechanism by decreasing of plasma estradiol 17β concentration and hence prohibits the rapid decreased of progesterone (Alaçam *et al.,* 1999). It is suggested that the higher progesterone concentrations detected in pregnant goats might be due to the GnRH injections including in all groups.

Conclusion: Satisfactory synchronization and fertility parameters were obtained by using Ovsynch, 2PG-GH, PG-GH and GH protocols in Hair goats during breeding season. It is concluded that the using of Ovsynch and 2PG-GH protocols have more benefits to attain higher twinning rate and fecundity.

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