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The Effect of Human Chorionic Gonadotropin on the Reproduction Performance in Lory Sheep Synchronized with Different Doses of Pregnant Mare Serum Gonadotrophin Outside the Breeding Season

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Abstract: Two experiments performed to determine the effects of different doses of PMSG and subsequent hCG treatment on the reproductive performance in estrus-induced mature Lory ewes. In first experiment 192 Lory anestrus ewes were divided into two groups and after synchronization with progestagen sponge (Fluorogestone acetate, 40 mg FGA) the ewes in first group (T₁) were injected 400IU PMSG and in second group (T₂) were injected 600 IU PMSG intramuscularly at sponge removal time. At insemination time (AI) time, ewes divided into 4 subgroups; T₁ and T₂h were injected 200 IU hCG and T₁C and T₂C were kept as the controls. In second experiment the effect of supplementing hCG at AI time or 12 days after AI were measured on the reproductive performance using 374 estrus-induced mature Lory ewes. After synchronization with progestagen sponge, all ewes were injected 400IU PMSG. The ewes then, were randomly divided into three groups: the ewes in (h₀) were injected 200 IU hCG at AI time, (h₁₂) were injected 200 IU hCG at day 12 after mating time and (C) were kept as the control group. Serums progesterone P4 concentrations were measured in days 12, 14 and 16 after AI in both experiments. The result of 1st experiment indicated that single lambs in T₁h subgroup had higher weight compared with T₁C subgroup at birth day (p<0.05). The prolificacy were higher in hCG treated groups compared with control (p<0.05). However, fertility did not differ significantly among subgroups. Mean weight of single lambs born was increased in T₁h compared with T₁C and T₁h subgroup had higher P4 concentration compared with T₁C subgroup (p<0.05). In Experiment 2; in comparison with control, the hCG increased prolificacy in h₀ treatment (p<0.05). Mean weight of lambs born was significantly increased in h₀ and h₁₂ groups compared with control. The hCG increased P4 concentration in h₀ and h₁₂ group and the h₁₂ had higher P4 concentration compare with other groups (p<0.05). It can be concluded that hCG injection at AI time increased progesterone concentrations and subsequent could improve reproductive performance in Lory ewes but there were no differences between the ewes treated with 400 or 600 IU PMSG.

Key words: Progestagen sponge, estrus synchronization, reproductive performance

INTRODUCTION

Treatment with intravaginal sponge impregnated with FGA for a period of 10-16 days and intramuscular injection of PMSG at intravaginal device removal, have been successfully used to improved the reproductive performance in ewes (Gomez-Brunet *et al.*, 2006).

Intramuscular administration of 400, 500 and 700 IU PMSG in sponge removal time increased the ratio of ovulation and twinning (Mehmet *et al.*, 2006). In sheep, 30-40% of fertilized eggs are lost during the first 3 weeks of pregnancy. One of the major causes of embryonic loss is likely to be the inadequate luteal function (Ashworth *et al.*, 1989).

Human Chorionic Gonadotropin (hCG), which is similar to LH in function, has been shown to increase luteal weight and endogenous synthesis of progesterone from the Corpus Luteum (CL) in sheep (Nephew *et al.*, 1994). The increase in P4 concentrations after hCG treatment suggests that hCG by its LH like activity may provide luteotrophic stimulation to CL. This luteotrophic stimulation may either be in the form of conversion of small luteal cells to large luteal cells which then secrete higher concentrations of progesterone or may even be due to an increase in the size of large luteal cells (Khan *et al.*, 2006). The beneficial effect of hCG administration on embryo survival may be through the stimulatory effect of hCG-induced progesterone on fetal growth, because it has been shown that progesterone supplementation increased subsequent fetal growth (Kleemaun *et al.*, 1994).

The hCG has been administered to ewes at different times during the cycle after AI or breeding in an attempt to reduce embryonic mortality and improve reproductive performance, but the effectiveness of these treatments has not been consistent between studies and the timing of such hormonal treatments also may seem to be important. The administration of hCG on the day of mating, 4, 5 and 12 days post mating have been reported by Peters (1996), Thatcher *et al.* (2001), Cam *et al.* (2002) and Khan *et al.* (2003).

Ishida *et al.* (1999) and Fukui *et al.* (2001) reported that the hCG treatment given at the early luteal phase increase the plasma P4 levels in hCG treated ewes, but this was not reflected in the pregnancy and lambing rates of the inseminated ewes. However, to improve the fertility, hCG would have to increase the fertilization rate and reduce the embryonic death rate or both.

There is a little information regarding synchronization efficiency and fertility induced by administration of hormones in Iranian Lory ewes during breeding and outside the breeding season. The purpose of this study was to investigate the effect of different doses of PMSG at sponge removal and the time of hCG administration on reproductive performance in Lory ewes outside of the breeding season.

MATERIALS AND METHODS

Two experiments were carried out in Iran, Lorestan, Pol dokhtar township, (latitude 43°-09°N), at 713.5 m above sea level during spring 2006. The ewes had access to the nearby stubble and low quality ranges for 8 h a day. Each ewe also received a diet on a daily basis consisting of 3 kg corn silage, 700 g chopped wheat straw, 200 g alfalfa hay and 150 g barley grain. The experiments started on 25 November, 2006. Estruses were induced by treating all ewes with an intravaginal sponge impregnated with synthetic progestagen (40 mg FGA, Intervet) for 13 days. Ewes were observed for estrus using teaser rams. Semen from 50 selected rams was collected using an artificial vagina and all of the ewes in estrous were inseminated civically 48 h after sponge removal with fresh semen which it was diluted with homogenized milk.

The number and weight of lambs born were recorded at lambing time. Blood samples were collected from jugular vein on days 12, 14 and 16 after AI from 20% of ewes chosen at random from each group. Blood samples were centrifuged to separate serum and were stored at -20°C until analyses for progesterone concentration.

Experiment 1

This experiment performed to determine the effects of PMSG treatments and subsequent hCG treatment on progestagen-synchronised Lory breed ewes. A total of 192 anestrus ewes, 4-5 years of age with a 35±4 kg body weight, were randomly divided into two groups and after synchronization with progestagen sponge, at sponge removal time, the ewes in first group (T₁) were injected (i.m.) with 400 IU PMSG and the ewes in second group (T₂) were injected (i.m.) with 600 IU PMSG. At insemination time (AI), each groups divided into 2 subgroups; T₁h and T₂h were injected 200 IU hCG and T₁C and T₂C were kept as the control groups.

Experiment 2

A total of 374 cyclic Lory ewes, 4-5 years of age with a 35 ± 4 kg body weight, were used in this experiment. In this experiment the effect of supplementing hCG at AI or 12 days after AI were measured on the reproductive performance. After synchronization with progestagen sponge the ewes were randomly divided into three groups; the ewes in (h_0) were injected 200 IU hCG at AI time, ewes in (h_{12}) were injected 200 IU hCG, 12 days after mating time and the ewes in (C) were kept as the control group.

Statistical Analysis

The experimental design for first experiment was a 2×2 factorial. Data for serums P4 analyzed by use of a repeated measure procedure. The means were compared using Duncan's multiple range tests when ANOVA indicated significant at $p < 0.05$). Prolificacy (No. of lambs born alive per ewe lambing), fertility (% ewes lambing per ewes mated) were assessed by Chi-squared analysis. The data for mean birth weight were analyzed by ANOVA test.

RESULTS

Experiment 1

The dosage of PMSG and time of hCG injection did not increase fertility and prolificacy in the subgroups (Table 1).

Mean weight of lambs born was significantly increased in T_1h compared to T_1C in single lambs ($p < 0.05$).

The mean serum P4 profiles of the ewes in the treatment groups are depicted in Table 2. The P4 concentration was decreased in days 14 and 16 in all groups, however in T_1C and T_2C the P4 concentrations were decreased significantly ($p < 0.05$). In day 16, T_1h has higher P4 concentration compared with T_1C and T_2C subgroups but not statistically significant ($p > 0.05$). Ewes in T_2h subgroup had higher P4 concentration in day 16 compare with controls ($P < 0.05$) (Table 2).

Experiment 2

The fertility and prolificacy was higher in h_0 compared with h_{12} and control. The hCG injection increased prolificacy in h_0 group which was statistically significant ($p < 0.05$) (Table 3). Mean weight of lambs born was significantly increased in h_0 and h_{12} groups compare with control group in single lambs (Table 3).

Table 1: Reproductive performance of ewes in 1st experiment

Treatments	T_1		T_2	
	T_1h	T_1C	T_2h	T_2C
No. of ewes	48	50	48	46
Fertility (%)	45.8	36	37.5	39.1
Prolificacy (%)	14.5 ^b	100 ^a	144 ^b	111 ^a
Mean birth weight (kg)				
Singles	4.2 \pm 0.13 ^b	3.5 \pm 0.21 ^a	3.8 \pm 0.15 ^{ab}	3.8 \pm 0.09 ^{ab}
Twins	2.3 \pm 0.33	-	2.7 \pm 0.2	2.2 \pm 0.12
Triplets	2.0 \pm 0.20	-	1.6 \pm 0.0	-

Different superscripts in Rows differ significantly, a, b: $p < 0.05$

Table 2: Mean serum P4 concentrations (ng mL⁻¹) of the ewes in treatment groups (Experiment 1)

Days after AI	T_1		T_2	
	T_1h	T_1C	T_2h	T_2C
12	4.2	3.8 ^b	5.9	5.6 ^b
14	3.8	2.7 ^{ab}	5.6	3.3 ^{ab}
16	3.6 ^{ab}	2.0 ^a	5.6 ^b	2.1 ^a

Different superscripts in rows and columns differ significantly, a, b: $p < 0.05$

Table 3: Reproductive performance of treatments ewes (Experiment 2)

Treatments	h ₀	H ₁₂	C
No. of ewes	128	121	125
Fertility (%)	50	47.9	35.2
Prolificacy (%)	160 ^a	129 ^{ab}	118 ^a
Mean birth weight (kg)			
Singles	3.75±0.14 ^b	3.70±0.12 ^b	3.20±0.12 ^a
Twins	2.25±0.08	1.87±0.08	2.32±0.1
Triples	2±0.09	2.22±0.16	1.94±0.10

Different superscripts in rows differ significantly; a, b: p<0.05

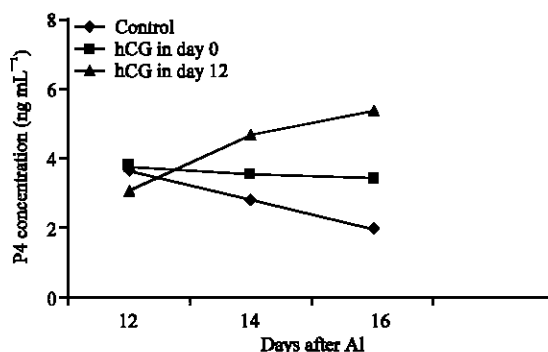


Fig. 1: Mean P4 concentration in treatment groups (Experiment 2)

The mean serum P4 concentration of the ewes in the hCG treated and control groups are depicted in Fig. 1. The hCG treatment increased P4 concentration in days 14 and 16 in treated groups compared with control and it was higher significantly in h₁₂ (p<0.05).

DISCUSSION

It has been well-known for some time that treatment with PMSG increases both ovulation rate and proportionally, luteal progesterone production in FGA-synchronised ewes. Khan *et al.* (2006) reported that Gonadotrophin supplementation during early pregnancy can reduce embryonic losses. The injection of PMSG immediately after progestagen sponge removal may produce an increase in the rate of ovulation (Romano *et al.*, 1997; Maxwell *et al.*, 1993). The PMSG increased conception rate and litter size, but the response is highly variable and leads to embryonic losses. In earlier study we reported that there was no significant difference between ewes received 300, 450 and 600 IU of PMSG upon progestagen withdrawal (Hojabri *et al.*, 2007).

In present study, there were no differences between the ewes treated with 400 or 600 IU PMSG (p>0.05) but the doses of 400 IU compared to 600 IU of PMSG were found to be less effective to induce multiple births (p>0.05). This might be because the dose used in this study was not sufficient to stimulate additional follicular development in native Lory breed used in this experiment. Intramuscular administration of 400, 500 and 700 IU PMSG in sponge removal time increased the ratio of ovulation and twinning (Mehmet *et al.*, 2006).

One of the main purposes of this study was to determine whether a single injection of hCG given to estrus-induced ewes at the time of insemination or 12 day after insemination could increase the secretion of P4 and subsequent fertility efficiency (p<0.05).

The results of this study showed that in synchronized and artificially inseminated Lory ewes, the injection of 200 IU hCG at the time of cervical AI or 12 days after AI can improve overall fertility

or prolificacy significantly and the fertility and prolificacy was higher in h_0 compare with h_{12} ($p < 0.05$). Therefore, the hCG injection at AI time was more effective to improve reproductive performance of Lory ewes compare with ewes were injected 200 hCG, 12 days after AI ($p < 0.05$). The results of present study are in agreement with other reports i.e., Kittot *et al.* (1983) reported that hCG administration before the time of maternal recognition of pregnancy increased pregnancy rate in lactating, seasonal anestrus ewes. There are also studies reporting that hCG administration improved reproductive performance in sheep during the breeding season (MacMillan *et al.*, 1986; Cam *et al.*, 2002).

The h_{12} is more effective on P4 concentration in 2nd experiment and in day 16 was higher compared with control ($p < 0.05$). Nephew *et al.* (1994) also reported that hCG administration on day 11 post mating increases pregnancy rate in sheep. They showed that hCG increased interferon secretion, luteal weights and conceptus length determined on day 13 of pregnancy. The effect of the hCG on pregnancy rate and fetal weights could be attributed to its effects on progesterone production and uterine secretions which were embryotrophic. This may result in a stronger signal for maternal recognition of pregnancy from embryos in hormonal treatment groups which would degenerate otherwise. The hCG administration has also been reported to increase the number of CL (Beck *et al.*, 1998) and plasma progesterone concentration.

A trend toward higher fertility rates using hCG treatment at AI or 12 day after AI time has also been observed in cows on farms where reproductive efficiency was low (Nakao *et al.*, 1983), as well as in heat-stressed dairy cows (Willard *et al.*, 2003).

These results are in contrast with reports given by Khan *et al.* (2003), in which hCG given on day of mating not increased the pregnancy rate and litter size of ewe lambs. These results are similar to that previously reported by Gomez-Brunet *et al.* (2006). These results is also similar with earlier studies in cattle (Swanson and Young, 1990) and sheep (Zamiri and Hosseini, 1998), in which treatment of hCG on the day of AI or mating was not effective in improving fertility significantly.

As inadequate levels of progesterone during the early and mid-luteal phases of the estrous cycle has been related with decreased fertility due to abnormal development of embryos and early embryonic death (Ashworth *et al.*, 1987). To the contrary Ishida *et al.* (1999) and Fukui *et al.* (2001) reported hCG treatment given at the early luteal phase also to increase the plasma P4 levels in hCG treated ewes, but this was not reflected in the pregnancy and lambing rates of the Lory inseminated ewes.

It is noteworthy that the birth weights of single lambs were higher in T_h group which is agree with Mehmet and Kuran (2003). It impossible that hormonal treatments used in the present study prevented the mortality of the twin embryos by stimulating the fetal growth.

When evaluating changes in P4 concentrations in control groups, it should be noted that the corpus luteum regressed between days 14 and 16 in these two control groups and plasma P4 levels on days 12, 14 and 16 were slightly higher in the hCG-treated ewes and no significant differences were found in prolificacy between hCG-treated ewes. These results were agreement with earlier studies in ewes (Zamiri and Hosseini, 1998; Khan *et al.*, 2003), in which hCG treatment given at time of insemination caused an increase the number of lambs born per ewe lambing as a result of an increase in ovulation rate which it is disagree with Mehmet *et al.* (2006) and Gomez-Brunet *et al.* (2006). These differences could possibly be due to the different protocols used, but it is also probable that other factors such as breed, management systems, nutritional and physiological status, could also have affected the response of animals.

It can be concluded that the hCG injection increased progesterone concentration and subsequent improved reproductive performance in Lory ewes but there were no differences between the ewes treated with 400 or 600 IU PMSG. Likewise 200 IU hCG injection at AI time was more effective to improve reproductive performance of treated Lory ewes compare with ewes were injected 200 hCG, 12 days after AI.

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REFERENCES

- Ashworth, C.J., I. Wilmut, A.J. Springbett and R. Webb, 1987. Effect of an inhibitor of 3-hydroxysteroid dehydrogenase on progesterone secretion and embryo survival in sheep. *J. Endocrinol.*, 112: 205-213.
- Ashworth, C.J., D.I. Sales and I. Wilmut, 1989. Evidence of an association between the survival of embryos and periovulatory plasma progesterone concentration in the ewe. *J. Reprod. Fertil.*, 87: 23-32.
- Beck, N.F.G., A. Green, T.H. Khan and M. Khalid, 1998. The effects of buserelin (GnRH analogue) or hCG treatment on luteal weight and embryonic development during early pregnancy in the ewe. *J. Reprod. Fertil. Abstr., Series*, 21: 95-95.
- Cam, M.A., M. Kuran, S. Yildiz and E. Selcuk, 2002. Fetal growth and reproductive performance in ewes administered GnRH agonist on day 12 post-mating. *Anim. Reprod. Sci.*, 72: 73-82.
- Fukui, Y., R. Itagaki, N. Ishida and M. Okada, 2001. Effect of different HCG treatments on fertility of estrus-induced and artificially inseminated ewes during the non-breeding season. *J. Reprod. Dev.*, 47: 189-195.
- Gomez-Brunet, A., J. Santiago-Moreno, G.J. Montoro, P. Pons and A. Gonzalez-Bulnes *et al.*, 2006. Reproductive performance and progesterone secretion in estrus-induced Manchega ewes treated with hCG at the time of AI. *Anim. Reprod. Sci.*, 71: 117-122.
- Hojabri, F., M.M. Moeini and Hajarani, 2007. Estrus synchronization in ewes with different intravaginal devices and PMSG treatments. 28th Malaysian Society of Animal Production, May 29-31, pp: 107-109.
- Ishida, N., M. Okada, K. Sebata, M. Minato and Y. Fukui, 1999. Effects of GnRH and hCG treatment for enhancing corpus luteum function to increase lambing rate of ewes artificially inseminated during the non-breeding season. *J. Reprod. Dev.*, 45: 73-79.
- Khan, T.H., P.M. Hastie, N.F.G. Beck and M. Khalid, 2003. HCG treatment on day of mating improves embryo viability and fertility in ewe lambs. *Anim. Reprod. Sci.*, 76: 81-89.
- Khan, T.H., N. Beck and M. Khalid, 2006. The effects of GnRH analogue (buserelin) or hCG(Chorulon) on Day 12 of pregnancy on ovarian function, plasma hormone concentrations, conceptus growth and placentation in ewes and ewe lambs. *Anim. Reprod. Sci.*, 102: 247-257.
- Kittot, R.J., J.N. Stellflug and S.R. Lowry, 1983. Enhanced progesterone and pregnancy rate after gonadotropin administration in lactating ewes. *J. Anim. Sci.*, 56: 652-655.
- Kleemann, D.O., S.K. Walker and R.F. Seemark, 1994. Enhanced fetal growth in sheep administered progesterone during the first three days of pregnancy. *J. Reprod. Fertil.*, 102: 411-417.
- MacMillan, K.L., V.K. Taufan and A.M. Day, 1986. Effects of an agonist of gonadotrophin releasing hormone (Buserelin) in cattle 3. Pregnancy rates after a post-insemination injection during metoestrus or dioestrus. *Anim. Reprod. Sci.*, 11: 1-10.
- Maxwell, W.M.C., G. Evans, S.L. Rhodes, M.A. Hillard and B.M. Bindon, 1993. Fertility of superovulated ewes after intrauterine or oviducal insemination with low numbers of fresh or frozen-thawed spermatozoa. *Reprod. Fertil. Dev.*, 5: 57-63.

- Mehmet, A.C. and M. Kuran, 2003. Effects of a single injection of hCG or GnRH agonist on day 12 post mating on fetal growth and reproductive performance of sheep. *Anim. Reprod. Sci. Res.*, 80: 81-90.
- Mehmet, A., B. Bulbuli, M. Bozkurt and S. Dere, 2006. Induction of multiple births in Akkaraman cross-bred sheep synchronized with short duration and different dose of progesterone treatment combined with PMSG outside the breeding seasons. *Bull. Vet. Inst. Pulway.*, 50: 97-100.
- Nakao, T., S. Narita, K. Tanaka, H. Horn and J. Shirakawa *et al.*, 1983. Improvement of first-service pregnancy rate in cows with gonadotropin-releasing hormone analogue. *Theriogenology*, 20: 111-119.
- Nephew, K.P., H. C'Ardenas, K.E. McClure, T.L. Ott and F.W. Bazer *et al.*, 1994. Effects of administration of human gonadotropin or progesterone before maternal recognition of pregnancy on blastocyst development and pregnancy in sheep. *J. Anim. Sci.*, 72: 453-458.
- Peters, A.R., 1996. Embryo mortality in the cow. *Anim. Breed. Abstr.*, 64: 587-598.
- Romano, J.E., E. Rodas, A. Ferreira, I. Lago and A. Benech, 1997. Effects of progestagen, PMSG and artificial insemination time on fertility and prolificacy in Corriedale ewes. *Small Rum. Res.*, 23: 157-162.
- Swanson, L.V. and A.J. Young, 1990. Failure of gonadotropin-releasing hormone or human chorionic gonadotropin to enhance the fertility of repeat-breeder cows when administered at the time of insemination. *Theriogenology*, 34: 955-963.
- Thatcher, W.W., F. Moreira, J.E.P. Santos, R.C. Mattos and F.L. Lopes *et al.*, 2001. Effects of hormonal treatments on reproductive performance and embryo production. *Theriogenology*, 55: 75-89.
- Willard, S., S. Gandy, S. Bowers, K. Graves and A. Elias *et al.*, 2003. The effects of GnRH administration postinsemination on serum concentrations of progesterone and pregnancy rates in dairy cattle exposed to mid summer heat stress. *Theriogenology*, 59: 1799-1810.
- Zamiri, M.J. and M. Hosseini, 1998. Effects of human chorionic gonadotropin (hCG) and phenobarbital on the reproductive performance of fat-tailed Ghezel ewes. *Small Rum. Res.*, 30: 157-161.