African Journal of Biotechnology Vol. 9(10), pp. 1523-1527, 8 March, 2010
Available online at http://www.academicjournals.org/AJB
ISSN 1684–5315 © 2010 Academic Journals

Full Length Research Paper

Follicular and ovulatory responses of the right versus left ovaries to eCG treatment in Shall Iranian ewes

Hamed Kermani Moakhar[1], Hamid Kohram[1,2]*, Reza Salehi[1] and Ahmad Zare Shahneh[1]

[1]Department of Animal Science, Faculty College of Agriculture and Natural Resources, University of Tehran, Karaj, Iran.
[2]Depertment of Clinical Sciences, Faculty of Veterinary Medicine, Shahid Chamran University, Ahvaz, Iran.

Accepted 11 February, 2010

The present study considers the right versus the left ovarian responses to the different doses of eCG treatment in ewe. The estrous cycles of 32 adult Shall Iranian ewes were synchronized by CIDR during breeding period. At the time of CIDR removal, 0 (n = 8), 450 (n = 8), 550 (n = 8) and 650 (n = 8) IU of eCG (Intervet, Booxmer, Netherlands) was administered to ewes. The number of large follicles (> 3 mm, LF) and CL in the right and left ovaries was monitored by transrectal ultrasonography on the day of estrus and 6 days later. In another analysing view, all ewes (n = 32) were set into 3 groups. Following superovulation, groups A and D ewes had at least one extra large follicle and CL, respectively, in their right ovary than that of the left one. Group B and E had the opposite position and group C and F had equal LF in their ovaries. The data were analysed by GLM procedure of SAS. The mean number of LF in the right (2.35 ± 0.14) was higher (P < 0.001) in comparison to the left (1.42 ± 0.14) ovaries in all treatment groups. Also, the mean number of LF in the right was higher (P < 0.05) than that in the left ovaries between every treatment group. In addition, the mean number of CL in the right (1.92 ± 0.47) was higher (P < 0.05) in comparison to the left (1.33 ± 0.25) ovaries in all treatment groups. Furthermore, in the other analysis, we observed 62.5% of the ewes were in group A (n = 20) which was a higher (P < 0.001) percentage compared to group B (n = 4, 12.5%) and group C (n = 8, 25%). Also, it is observed that 44% of the ewes were set into group D (n = 16) (the right ovary had at least one CL extra in comparison with the left ovary) which was a higher (P < 0.05) percentage compared to groups E and F; and that both groups had equal number of ewes. The results suggest that the greater responses of follicular development occur in the right ovaries after eCG treatment in Shall Iranian ewes.

Key words: Ewe, Shall, follicle, CIDR, eCG.

INTRODUCTION

Equine chorionic gonadotrophin (eCG) increases ovulation rate in the ewe by recruiting small ovarian follicles, increasing the growth rate of antral follicles and altering ratios of large follicle size-classes at estrus. However, eCG does not appear to rescue follicles from atresia in the ewe (Driancourt et al., 1992). eCG causes early oocyte activation (Moor et al., 1985) and follicles exposed to eCG synthesize larger amounts of progesterone (Driancourt et al., 1992).

There is evidence that showed that administration of eCG in the time of CIDR removal is the most common, in order to increase the superovulation responses in ewes (Boscos et al., 2002; Menegatos et al., 2003; Barrett et al., 2004). However, low doses of eCG (300 - 500 IU) has been combined with CIDR treatment which probably has little effect on ovulation rate but reduces the interval to the onset of estrus when compared with ewes fully administered with CIDR (Barrett et al., 2004; Botha et al., 1975; Gordon, 1971). In contrast, increasing doses of eCG can enhance the ovulation rate (Robinson, 1951).

In most ewe breeds, ovulation is performed in more than 2 or 3 of the ovulatory follicles during estrous cycle. Ovulation rate in ewes enhances with increasing age. It is showed that ovulation rate in right ovary (53.4%) was

*Corresponding author. E-mail: kohram@can.ut.ac.ir. Tel: +98-261-2248082. Fax: +98-261-2246752.

Abbreviations: eCG, Equine chorionic gonadotrophin; **LF,** large follicle; **CL,** corpus luteum; **FSH,** follicle stimulating hormone; **CIDR,** progesterone-releasing intravaginal device.

significantly more than the left ovary (46.6%) (Scaramuzzi and Downing, 1997). Also, it was suggested that unequal functions occur between the right and left ovaries in various species including ewes (Casida et al., 1966). Casida et al. (1966) reported that the right ovary produces more corpus luteum (CL) comparing to the left ovary in ewes. Naqvi and Guliany (1998) showed that the right ovary has a greater number of CL and large follicles in response to eCG or Follicle Stimulating Hormone (FSH) treatment. However in heifers, it was reported that there is an effect of the day relative to initiation of FSH treatment on all follicular categories responding positively to superovulation and there is no effect of side (left or right ovary) or of CL diameter (ipsilateral or contralateral) (Purwantara et al., 1993).

The present study was conducted to consider the right versus the left ovarian responses (that is the mean number of follicles > 3 mm in diameter at estrus and the number of CL 6 days later) to the different doses of eCG treatment.

MATERIALS AND METHODS

Animals and management

The experiment was conducted in breeding seasons (September - November). The institute farm is located in Saveh city at 50.19 ± 21° E, latitude of 35.01 ± 13° N and at an altitude of 1250 m above mean sea level in semi-arid tract of the country. The annual rainfall ranges from 220 mm with an erratic distribution throughout the year. Thirty two adult Shall Iranian ewes, between 2 and 4 years of age and body weight 69 kg (range 60 - 80) were housed in conditions of natural daylight and temperature and were in good body condition throughout the observational period. The ewes were provided with water *ad libitum* and were fed a live weight maintenance ration during breeding period.

Estrous synchronization and superovulatory program

Estrous cycles of the animals were synchronized by CIDR (EAZI-BREED™ CIDR®; progesterone). The day of CIDR insertion was considered as the starting point of the experiment (day 0). At the time of CIDR removal (day 14), the ewes were randomly divided equally (n = 8) among four superovulation groups: 0 (control), 450 (G1), 550 (G2) and 650 (G3) IU of eCG (Intergonan ®, Intervet, Toenisvorst, Netherlands) was administered to ewes. The animals were observed for estrus behaviour using a vasectomised rams.

Ultrasound examination

The right and left ovaries were monitored by transrectal ultrasonography (Piemedical, Falco100; Holland, with an 8 MHz transducer) on days 16 (day of estrus) for observation of the large follicles (LF) and 22 for observation of the number of CL. During examination the animals were held in a standing position. Scanning of both ovaries was recorded using a MP4 player (Marshal X720, China). Follicles > 3 mm in diameter were identified, measured and mapped to their location using printed images of both ovaries.

In another analysing view, all ewes (n = 32) were set into 3 groups on day 16. Following superovulation, group A ewes had at least one extra large follicle in their right ovary in compare to their left ovary at estrus. Group B had the opposite position in their ovaries and group C had equal LF in both ovaries. Similarly all ewes were divided into 3 groups on day 22. Group D ewes had at least one extra CL in their right ovary in compare to their left ovary. Group E had the opposite position in their ovaries and group F had equal CL in both ovaries.

Statistical analysis

An analysis of variance was used for the comparison between treatment groups regarding the mean number of follicles and the mean number of CL. When a significant difference was recorded, the Duncan's multiple range test was used to determine how the means differed. The SAS-program was used in the analyses.

RESULTS

All ewes showed estrus behaviour and ovulation until 48 and 82 h after the end of the synchronization treatment, respectively. The mean number of large follicles (> 3 mm in diameter) in the right (2.35 ± 0.14) was higher ($P < 0.001$) in comparison to the left (1.42 ± 0.14) ovaries in all treatment groups. Also, the mean number of LF in the right was higher ($P < 0.05$) than in the left ovaries within treatment groups (Figure 1). In addition, the mean number of CL in the right (1.92 ± 0.47) was higher ($P < 0.05$) in comparison to the left (1.33 ± 0.25) ovaries in all treatment groups. However, the mean number of CL in the right ovaries was not significantly different from the left ovaries within treatment groups (Figure 2).

The mean number of large follicles in the right ovaries of groups that was administrated eCG was higher ($P < 0.05$) in comparison to the right ovaries in control group (Table 1). In addition, the mean number of CL in the right ovaries of only group G3 was higher ($P < 0.05$) in comparison to the right ovaries in control group (Table 1). On the other hand, the mean number of large follicles and CL in the left ovaries of groups G1, G2 and G3 was not significantly different from the left ovaries of control group.

Furthermore, in the other analysis, we observed 62.5% of the ewes were in group A (n = 20) which was a higher ($P < 0.001$) percentage compared to group B (n = 4, 12.5%) and group C (n = 8, 25%). Also, 44% of the ewes were set into group D (n = 16) (the right ovary had at least one CL extra in comparison the left ovary) which was a higher ($P < 0.05$) percentage compared to group E and group F that both groups had the number of equal ewes (Figure 3).

DISCUSSION

All ewes treated with eCG showed estrus behaviour (acceptance of the male and mucus discharge) and ovulation (disappearance of large follicles and observation of CL) after the end of the synchronization treatment. The addition of low-dose eCG (300 - 500 IU) to progestogen

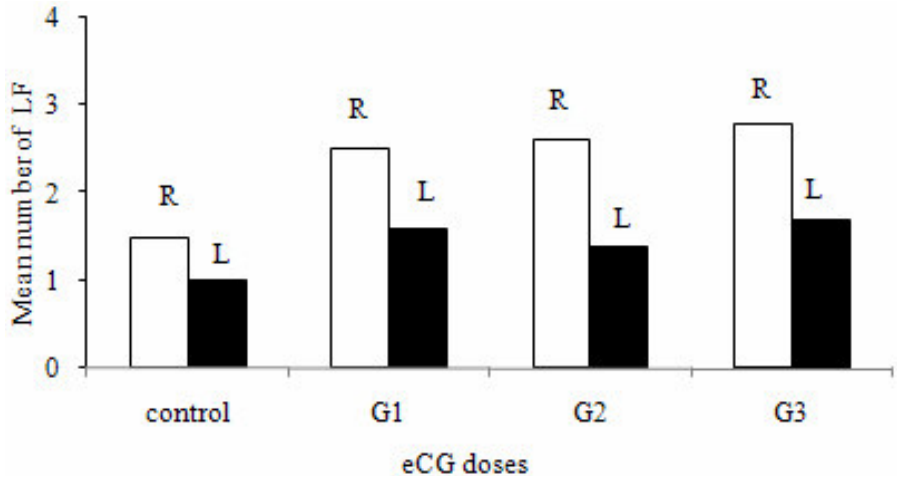

Figure 1. The mean number of large follicles (> 3 mm) in the right (R) and left (L) ovaries at estrus using various doses of eCG treatment.

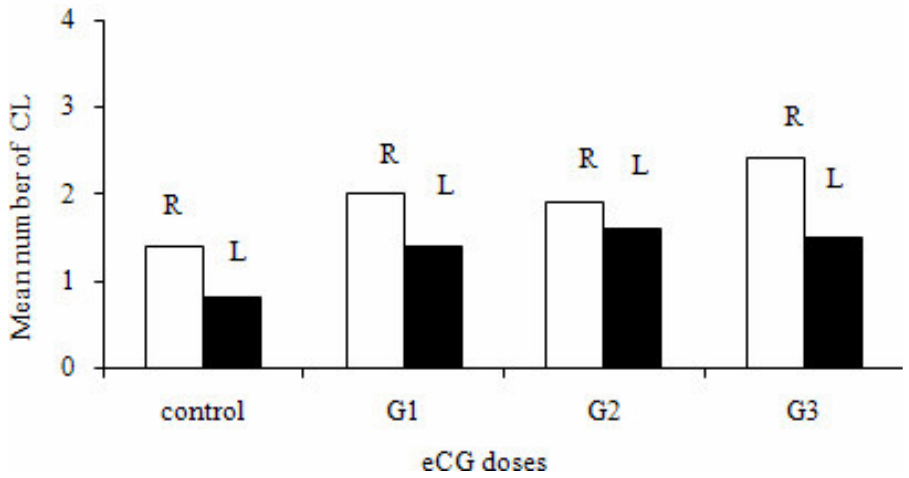

Figure 2. The mean

CIDR treatment can cause advancement of estrus and ovulation and a small increase in ovulation rate in ewes (Ainsworth et al., 1977). The timing of estrus and ovulation in eCG-treated ewes in our study was similar to that in other studies (Boland et al., 1978; Gordon, 1997). The preovulatory follicles (> 3 mm in diameter) are the source of estradiol hormone (Barrett et al., 2004), which probably resulted in appearance of estrus and ovulation in all of experimental groups.

We were able to detect ovaries with reasonable frequency by rectal ultrasonography in Iranian Shall ewes during standing position that is more comfortable for the ewes than dorsal recumbency (Riesenberg et al., 2001). Overall, significantly more ovaries were observed after superovulation with low doses of eCG than control group ($P < 0.05$). This finding could not be explained either by a higher number or by a smaller diameter of follicles, suggesting (Riesenberg et al., 2001) that the ovaries themselves were enlarged after eCG stimulation. Moreover, using of MP4 for recording, reviewing and drawing each ultrasonogeraphy in this experiment was much easier and faster in comparison to other experiment that was usually performed by a set of video.

In the present study, the administration of different doses of eCG at CIDR withdrawal increased the mean number of large follicles and the ovulation rate. This is in accordance with results of other authors, although the mechanism by which eCG increased the mean number of large follicles and ovulation rate is not yet clear (Driancourt et al., 1991). There are several possibilities, namely an increased rate of entry of follicles into the > 2 mm size category, the prevention of the occurrence of natural atresia or the reversal of the atretic process (Bister et al., 1999; Mandiki et al., 2000).

The mean number of LF (> 3 mm follicles) and CL in the right ovaries was higher ($P < 0.05$) in comparison to

Table 1. Effect of different doses of eCG on the mean (±S.E.M.) number of large follicles and CL on the right versus left ovaries following synchronization of estrus in ewe.

Parameter	Control	G1	G2	G3
Number of LF on the right ovaries	1.5 ± 0.25[1]	2.5 ± 0.25[2]	2.6 ± 0.25[2]	2.8 ± 0.25[2]
Number of CL on the right ovaries	1.4 ± 0.18[1]	2.0 ± 0.18[1]	1.9 ± 0.18[1]	2.4 ± 0.18[2]
Number of LF on the left ovaries	1.0 ± 0.12[1]	1.6 ± 0.12[1]	1.4 ± 0.12[1]	1.7 ± 0.12[1]
Number of CL on the left ovaries	0.8 ± 0.11[1]	1.4 ± 0.11[1]	1.6 ± 0.11[1]	1.5 ± 0.11[1]

Values with different superscripts (1 and 2) in the same row differ significantly (P < 0.05).

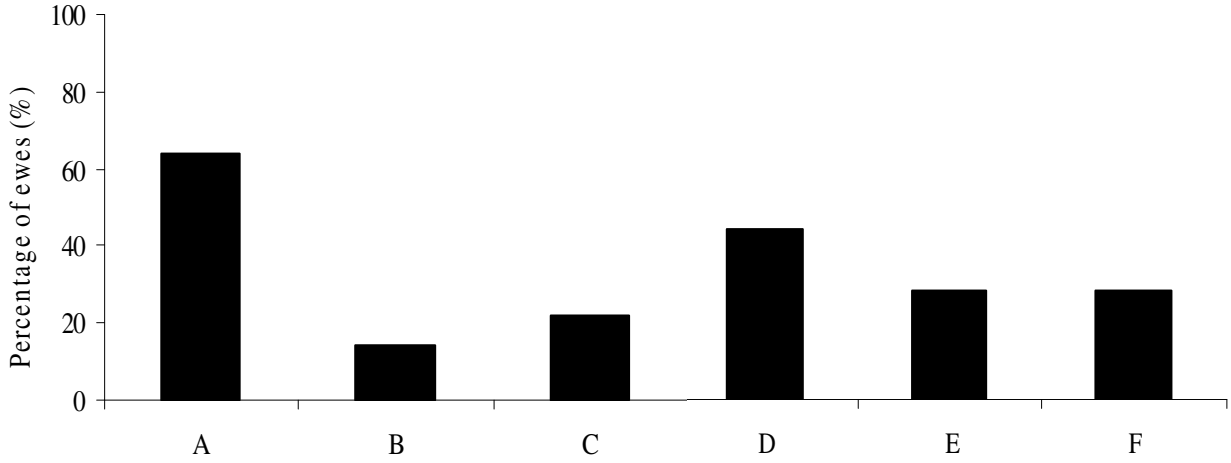

Figure 3. Percentage of ewes in groups A, B and C is relation to the present of large follicles and percentage of ewes in groups D, E and F is relation to the present of CL on ovaries.

the left ovaries in all treatment groups. Also, the mean number of LF in the right ovaries was higher (P < 0.05) than in the left ovaries within each treatment groups (Figure 1). These results are in agreement with the observations of the experiment carried out on ovaries after injection of different doses of eCG (Casida et al., 1966; Scaramuzzi and Downing, 1997; Martinez et al., 2007). In addition, the right ovary had a tendency to produce more CL than the left one (Figure 2). These differences have been reported previously (Mufti et al., 1997; Scaramuzzi and Downing, 1997) for ewes that received eCG after CIDR removal in spite of other ewes that FSH was administered. However, Naqvi and Guliany (1998) showed that the right ovary has a greater number of CL and large follicles in response to eCG or FSH treatment. In another study by Ali (2007), it was suggested that ovulations were distributed equally between the right and left ovaries after injection of eCG. Also during estous cycle, the number of the largest follicles and CL located in the left versus the right ovary did not differ significantly among wave categories (Ali, 2007).

In the other analysis, 62.5 and 44% of the ewes had at least one extra large follicle and CL, respectively, in their right ovary in comparison to their left ovary. Martinez et al. (2007) also reported that after injection of high doses of eCG the large follicles was present on the right ovaries in a slightly greater percentage than on the left ovaries (54.5% compared with 45.5%). Although, they reported that no relationship was found between the locations of the largest follicles of the different waves or between the location of the CL and the largest follicles during estrous cycle. Controversial observations were reported in heifers and sheep (McKenzie and Terrill, 1937; Hutchinson et al., 1966; Dufour et al., 1972).

In conclusion, the superovulatory responses examined in terms of the mean numbers of large follicles at estrus and ovulations 6 days later in the right versus the left ovaries in this experiment were similar to those previously reported. The tendency for ovulation to take place more often in the right or left ovary is not previously proven. However, in this experiment and several previous studies it has been shown that activity of the right ovary is more than the left one. In consequent, in several investigations it was observed that the number of large follicle and the number of CL in response to super-ovulation especially, after injection of eCG in the right ovary is higher than the left ovary.

ACKNOWLEDGEMENTS

Thanks are extended to Dr. A. Monfared for granting

private research animal farm and Mr. Bakhshi for the care of experimental animals.

REFERENCES

Ainsworth L, Hackett AJ, Heaney DP, Langford GA, Peters HF (1977). A multidisciplinary approach to the development of controlled breeding and intensive production systems for sheep. USA: Management of reproduction in sheep and goats symposium; pp. 101-108.

Ali A (2007). Effect of time of eCG administration on follicular response and reproductive performance of FGA-treated Ossimi ewes. Small Rumin. Res. 72: 33-37.

Barrett DMW, Bartlewski BA, Symington A, Rawlngs NC (2004). Ultrasound and endocrine evaluation of the ovarian response to a single dose of 500 IU eCG following a 12-day treatment with progestagen-releasing intravaginal sponges in the breeding and non-breeding season in ewes. Theriogenology, 61: 311-327.

Bister JL, Noel B, Perrad B, Mandiki SN, Mbayahaga J, Paquay R (1999). Control of ovarian follicles activity in the ewe. Domest. Anim. Endocrinol. 17: 315-328.

Boland MP, Lemainque F, Gordon I (1978). Comparison of lambing outcome in ewes after synchronization of oestrus by progestagen or prostaglandin treatment. J. Agric. Sci. Camb 91: 765-766.

Boscos CM, Samartzi FC, Dellis S, Rogge A, Stefanakis A, Krambovitis E (2002). Use of progestagen-gonadotrophin treatment in estrus synchronization of sheep. Theriogenology, 58: 1261-1272.

Botha HK, Van Niekerk CH, Pagel RFE (1975). Influence of synchronization of the oestrous period, PMSG administration and flushing on oestrus and conception of South African mutton merinos. J. Anim. Sci. 5: 231-233.

Casida LE, Woody CO, Pope AL (1966). Inequality in function of the right and left ovaries and uterine horns of the Ewe. J. Anim. Sci. 25: 1169-1171

Driancourt MA, Webb R, Fry RC (1991). Does follicular dominance occur in ewes? J. Reprod. Fertil. 93: 63-70.

Driancourt MA, Fry RC (1992). Effect of superovulation with pFSH or PMSG on growth and maturation of the ovulatory follicles in sheep. Anim. Reprod. Sci. 27: 279-292.

Dufour JJ, Ginther OJ, Casida LE (1972). Intraovarian relationship between corpora lutea and ovarian follicles in ewes. J. Vet. Res. 33: 1445-1451.

Gordon I (1997). Controlled Reproduction in Sheep and Goats. CAB International, UK.

Hutchinson JSM, Robertson HA (1966). The growth of the follicle and corpus luteum in the ovary of the sheep. Res. Vet. Sci. 7: 17-24.

Martinez JA, Sanchez MT, Cordero JL, Mendoza GD, Garcia CM, Garcia M (2007). Ovarian follicular dynamics after cauterization of the dominant follicle in anestrous ewes. Anim. Reprod. Sci. 98: 225-232.

Mandiki SN, Noel B, Bister JL, Peeters R, Beerlandt G, Decuypere E, Visscher A, Suess R, Haulfuss KH, Paquay R (2000). Pre-ovulatory follicular characteristics and ovulation rates in different breed crosses, carriers or non-carriers of the Booroola or Cambridge fecundity gene. Anim. Reprod. Sci. 63: 77-88.

McKenzie FF, Terrill CE (1937). Estrus, ovulation and related phenomenon in the ewe. Res. Bull. Mo. Ag Exper Stat.

Menegatos J, Chadio S, Kalogiannis T, Kouskoura T, Kouimtzis S (2003). Endocrine events during the periestrous period and the subsequent estrous cycle in ewes after estrus synchronization. Theriogenology, 59: 1533-1543.

Moor RM, Osborn JC, Crosby IM (1985). Gonadotrophin-induced abnormalities in sheep oocytes after superovulation. J. Reprod. Fert. 74: 167-172.

Mufti AM, Wani GM, Wani NA, Mir MM, Khan MZ (1997). Superovulatory response in Corriedale sheep during different months of the breeding season. Small Rumin. Res. 25: 181-184.

Naqvi SMK, Gulyani R (1998). The effect of gonadotropin realising and follicle stimulating hormone in conjunction with pregnant mare serum gonadotiopin on the superovulation response in crossbred sheep in India. Trop. Anim. Health Prod. 30: 369-376.

Purwantara B, Schmidt M, Greve T, Callesen H (1993). Follicular dynamics prior to and during superovulation in heifers. Theriogenology, 40: 913-921.

Riesenberg S, Meinecke TS, Meinecke B (2001). Ultrasonic study of follicular dynamics following superovulation in German Merino ewes. Theriogenology, 55: 847-865.

Robinson TJ (1951). The augmentation of fertility by gonadotrophin treatment of the ewe in the normal breeding season. J. Agric. Sci. Camb 41: 6-38.

Scaramuzzi RJ, Downing JA (1997). The distribution of ovulations from the ovaries of Merino and Border Leicester × Merino ewes and its effect on the survival of their embryos. Anim. Reprod. Sci. 47: 327-336.