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Superovulatory response of Boer goat does pre-treated with a GnRH-agonist during the natural breeding season

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Abstract

A trial was conducted to evaluate the effect of pre-treatment with a GnRH agonist (GnRHa) on the superovulatory response in Boer goats during the natural breeding season. Twenty-one does were synchronised with CIDR's for 17 days and superovulated with a total dose of 200 mg FSH/doe, administered i.m. in seven dosages, at 12 h intervals, starting 48 h prior to CIDR removal (the first dose being 50 mg and all the others being 30 mg each). The treatment group (n = 11) additionally received a GnRHa (Lucrin® 40 µg/day/doe) treatment for seven days, starting on day 7 of CIDR insertion, while the control group (n = 10) received only FSH. Laparoscopic inseminations with fresh diluted Boer goat semen were performed 36 h and 48 h following CIDR removal. Embryos were flushed six days following the second artificial insemination. The oestrous response (80 *vs*. 100%), onset (25.5 ± 7.4 h *vs*. 24.0 ± 7.4 h) and duration of the induced oestrous period (26.9 ± 4.0 *vs*. 19.6 ± 5.5 h) for the FSH and FSH/GnRHa treated groups, respectively, did not differ significantly. Similarly, no significant differences were recorded between groups in terms of the mean total number of structures, embryos and unfertilised ova flushed/doe. The pre-treatment with a GnRHa had no real advantage over the use of FSH alone, as it tended to reduce the number of transferable embryos (3.4 ± 2.7 *vs*. 9.3 ± 6.1, respectively) by increasing the total number of degenerated embryos (6.6 ± 4.2 *vs*. 1.7 ± 1.5, respectively) in Boer goats during the natural breeding season. Under these conditions treatment with GnRHa is not recommended.

Keywords: Does, oestrous synchronisation, MOET, embryo, Lucrin®
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Introduction

The South African Boer goat, due to its outstanding traits is very popular and enjoys a high demand in many countries. However, in most of these countries, the entry of live animals is prohibited due to health risks and the importation of embryos under strict regulatory conditions is the only viable option. Therefore multiple ovulation and embryo transfer (MOET), becomes an important tool to produce Boer goat embryos for exportation. However, currently a very important stage in MOET e.g. superovulation, lacks efficiency and repetitiveness. The major factor leading to variation in response to superovulation in small stock has been reported to be the follicular status at the onset of the superovulation (Gonzalez-Bulnes *et al.*, 2004). The presence of a large or dominant follicle at the onset of the superovulatory treatment has been reported to decrease the ovarian response in small ruminants (Rubianes & Menchaca, 2003). The ovulation rate can be significantly improved if a large number of small follicles are present in the ovaries during the absence of a dominant follicle (secreting LH), at the onset of superovulation (Cognie *et al.*, 2003; Gonzalez-Bulnes *et al.*, 2004). These ovarian conditions occur naturally when superovulation is initiated soon after ovulation – known as the 'Day 0 protocol' (Rubianes & Menchaca, 2003). In sheep, high ovulation rates, improved embryo quality and increased number of embryos recovered have been reported with the use of this protocol (Rubianes *et al.*, 1997). However, treatment in the absence of a dominant follicle is difficult to perform in practice due to the unpredictability of the ovarian follicular patterns (Driancourt, 2001). Another option would be the induction of a low blood LH concentration, in order to avoid dominance (Campbell *et al.*, 1995).

Low serum LH concentrations have been achieved via the administration of exogenous gonadotrophin-releasing hormone (GnRH) antagonists or agonists. The GnRH antagonists inhibit GnRH release for several hours after administration, by blocking the pituitary receptors. The GnRH agonists on the other hand, also produce a 'flare up' effect after administration and then over time downregulate the GnRH receptors,

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desensitising the anterior pituitary gland to endogenous GnRH and thus prevent the pulsatile release of LH (Cheung, *et al*., 2005). GnRH agonists have therefore been used as a pre-treatment before superovulation in MOET programmes with success in sheep (Cognie, 1999) and cattle (D'Occhio *et al.,* 1998).

The aim of this study was to evaluate the response of Boer goat does to superovulation and embryo recovery rates following pre-treatment with a GnRH agonist (Lucrin®), during the natural breeding season.

Materials and Methods

Twenty-one mature Boer goat does were used in this trial, conducted during the natural breeding season (autumn). Does were kept in open pens and fed maintenance pellets and lucerne hay. All does were synchronised for oestrus with the aid of controlled internal drug release dispensers (CIDR; Phamacia & Upjohn, Auckland, New Zealand) for 17 days and superovulated using FSH (Folltropin®-V). A total dose of 200 mg FSH/doe was administered in seven i.m. injections, at 12 h intervals, starting 48 h prior to CIDR removal (the first dose being 50 mg and all others 30 mg each). Does in the treatment group (n = 11) additionally received a GnRH agonist (GnRHa - Leuprolide, Lucrin®, NL, Ch) (40 µg/day/doe) administered as 2 s.c. injections per day for seven days (Padula & Macmillan, 2005), starting on day seven of CIDR insertion. The control does (n = 10) received no GnRHa. Oestrous behaviour was detected three times daily at eight hour intervals following CIDR withdrawal with the aid of teaser bucks to determine the oestrous response, onset and duration of the induced oestrus. Fixed time laparoscopic inseminations with fresh diluted semen (150 x 10^6 sperm/mL) were performed 36 h and 48 h following CIDR withdrawal.

On day 6 following the second artificial insemination, embryos were surgically recovered under general anaesthesia. Laparoscopic evaluation of the ovaries was performed prior to flushing and does with no *corpora lutea* (CL's) and abnormal CL's, as classified by Espinosa-Marquez *et al.* (2004), were not flushed. Does were deprived of feed and water 24 h before embryo recovery. All recovered structures were evaluated and classified as unfertilised ova, degenerated and transferable embryos (Lindner & Wright, 1983, Nuti *et al*., 1987). Data on onset and duration of the induced oestrus were analysed using the Chi-square test and the total number of structures recovered, unfertilised ova and embryos collected were analysed using ANOVA procedures of SAS (1999).

Results and Discussions

All does superovulated with FSH/GnRHa demonstrated oestrous signs following synchronisation, while only 20% (two does) of those from the control group (FSH alone) failed to show signs of oestrus. There was thus, no significant difference between the two groups regarding the oestrous response. These responses are similar to previous reports in goats synchronised with either progestagen sponges or CIDR and superovulated with FSH (Pendleton *et al.*, 1992; Espinosa-Marquez *et al.*, 2004). The time from CIDR removal to the onset of oestrus (25.5 ± 7.4 and 24.0 ± 7.4 for FSH and FSH/GnRHa, respectively) was in line with data on goats superovulated with a FSH/GnRH antagonist (Baril *et al.*, 1996), but shorter when compared to the 32.0±3.5 h recorded in different goat breeds and the 42.0±18.0 h in Boer goats synchronised with CIDR and superovulated (Pendleton *et al.*, 1992; Greyling *et al.*, 2002). The duration of the induced oestrous period (26.9 ± 4.0 and 19.6 ± 5.5 for control and FSH/GnRHa groups, respectively) was similar, but shorter than the 34 ± 7.2 h and 40.8 ± 0.4 h reported in Boer (Greyling *et al.*, 2002) and Angora (Armstrong *et al*., 1983) goats superovulated with FSH.

The laparoscopic inspection prior to flushing revealed that 38.1% of the does (five from the treated and three from the control group) had abnormal CL's (small ovulation points, pale to pink in colour and avascular), while 23.8% of the does (four control and one treated) had no CL. Therefore, none of these does were flushed. Only 38.1% of the does (five treated and three controls) had normal CL's (highly vascular and bright red in appearance) and these goats were flushed. Based on ovarian inspection, the FSH/GnRHa treatment performed better as more animals were eligible to flushing than the control group. A slightly higher percentage (47%) of abnormal CL's has been recorded in Murciana goats superovulated with FSH (Pintado *et al.*, 1998). A high incidence of abnormal CL's has also been reported to occur in PMSG treated goats, compared to FSH treated animals (Pendleton *et al.*, 1992).

The mean total number of structures and the embryo recovery rates are set out in Table 1.

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Table 1 Mean (± s.d.) total number of structures and embryo recovery rates in Boer goats superovulated with FSH or FSH plus a GnRH agonist (Lucrin®)

	Overall	FSH	FSH/GnRHa
No. of goats	8	3	5
Embryo and ova recovery/donor	14.3 ± 5.8	15.0 ± 6.2	13.8 ± 5.5
Embryos/donor	11.0 ± 5.7	11.0 ± 7.5	11.0 ± 4.1
Fertilised ova /collected ova (%)	74.4 ± 26.6	72.4 ± 36.7	75.6 ± 19.8
Unfertilised ova/donor	3.9 ± 4.2	4.0 ± 4.5	3.8 ± 4.0
Degenerated embryos/donor	4.8 ± 3.5	1.7 ± 1.5	6.6 ± 4.2
Transferable embryos/donor	5.6 ± 4.2	9.3 ± 6.1	3.4 ± 2.7
Transferable embryos /fertilised ova (%)	39.7 ± 25.1	61.2 ± 26.6	26.8 ± 24.4

No significant differences were recorded between groups (FSH and FSH/GnRH)

The mean total number of structures recovered did not differ significantly between the two groups, but the overall mean (14.3 ± 5.8) was higher than the 7 ± 1.42, 7.1 ± 1.4 and 8.3 ± 2.36 reported in other goat breeds superovulated with FSH (Pendleton *et al.*, 1992; Selvaraju *et al.*, 2003; Senthil Kumar *et al.*, 2003), This may be due to the effect of breed on the ovarian response to superovulation, indicating that Boer goats may respond better to the superovulatory treatment, compared to other breeds. The mean number of embryos and the fertilisation rates obtained in this study were also similar in both treatments. In a similar trial where goats were superovulated with FSH and pre-treated with a GnRH antagonist (with similar activity as a GnRH agonist), the fertilisation rate was lower in goats pre-treated with the GnRH antagonist (Cognie *et al.*, 2003). The overall embryo recovery rate (74.4 ± 26.6%) obtained in this trial was lower compared to the 94% previously reported in Boer goats by Greyling *et al.* (2002). The overall mean number of embryos recovered/doe (11.0 ± 507) was also comparable to the findings of Gonzalez-Bulnes *et al.* (2003) in Murciano-Granadiana goats (11.3 ± 0.5) and Nowshari *et al.* (1995) in Boer goats (10.6 ± 1.3). The mean number of unfertilised ova did not differ significantly between the two treatments (4.0 ± 4.5 and 3.8 ± 4 for FSH and FSH/GnRHa, respectively) although, this mean was higher than 0.3 ± 0.21 recorded in Tellicherry goats superovulated with the same FSH product as utilised in the present trial (Senthil Kumar *et al.*, 2003). The mean number of degenerated embryos in the FSH/GnRHa group (6.6 ± 4.2) tended to be higher than in the FSH group (1.7 ± 1.5) although the difference was not significant. This may be attributed to the low number of does eventually flushed. The mean number of transferable embryos and the embryo recovery rates tended to be lower in the treatment group, but did not differ significantly from the control group. Similar tendencies have been reported in goats treated with FSH/GnRH antagonist compared to FSH alone (2.3 *vs.* 8.1 and 47.5% *vs.* 74.0%, respectively; Cognie *et al.*, 2003). The low number of transferable embryos obtained in this trial can be attributed to the high number of degenerated embryos. Nevertheless, these results are comparable to other studies in goats (Pendleton *et al.*, 1992; Senthil Kumar *et al.*, 2003).

Conclusions

The oestrous response in Boer goats does not seem to be affected by the GnRH agonist pre-treatment in FSH superovulated goats during their natural breeding season. However, its use demonstrated no advantage in terms of the embryo yield and instead, tended to reduce the number of transferable embryos by increasing the number of degenerated embryos. Under these conditions treatment with GnRHa is not recommended.

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