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OESTRUS SYNCHRONISATION AND THE USE OF BOVINE FOLLICULAR FLUID TO INCREASE REPRODUCTIVE EFFICIENCY IN ETTAWA CROSSBREED GOATS

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Abstracts

Twenty mature Ettawa crossbreed goats, one-and-a-half to two years of age and weighing 23 to 30 kg were used in these experiments. Oestrus was synchronised in 20 Ettawa crossbreed goats with two injections of 6.25 mg prostaglandin given 11 days apart. They were randomised into two equal groups; group I received 3 ml charcoal-extracted bovine follicular fluid daily at 06.00 and 18.00 on the 12th to 15th day of the synchronised oestrus cycle and group II received an equal volume of normal saline at the same times. Luteolysis was induced 48 hours after the treatments began by a single injection of 6.25 mg prostaglandin. The onset of oestrus was detected three times a day. In the goats of group I, oestrus occurred 52 ± 12.6 hours after the injection of prostaglandin, and in the goats of group II after 58 ± 7.8 hours. The mean duration of oestrus did not differ significantly between groups 32.8 ± 5.2 hours in group I compare with 34.6 ± 2.3 hours in group II. The early pregnancy diagnosis by measuring plasma progesterone concentration at 40 days following AI indicated that in group I, does had higher pregnancy rate (80%) than the does of group II (60%). However, only 6 does (60%) in group I and 2 does (20% in group II finally deliver kids. These finding suggest that the use of charcoal-extracted bovine follicular fluid could be used for increasing pregnancy rate and enhance the twining rate in Ettawa crossbreed goats.

Key words: Oestrus synchronisation, Superovulation, Prostaglandin, Goats

Introduction

Ettawa crossbreed goats are one of the domestic livestock that has great potential and could perhaps be highly profitable enterprise in Indonesia. This is due to the fact that the goats have good reproductive performance, they are valued for milk and meat productions, the demand for animal protein was high and the capital investment was quite low. Unfortunately, in most small-scale dairying at village level were used traditional management systems, so that its reproductive efficiency is relatively low (Setiadi *et al.*, 1995). Artificial breeding technology such as oestrus synchronisation, superovulation and artificial insemination (AI) could be used to

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increase reproductive efficiency in goats. It is clear, from many reports that oestrus can be effectively synchronised with prostaglandin. An injection of prostaglandin will induce luteal regression. Superovulation is a technique to stimulate ovaries at the end of cycle to produce a number of eggs beyond natural ability. Artificial Insemination using frozen good quality semen can be used to improve the genetic materials in goats.

Follicular fluid contains a variety of substances including steroid and protein hormones, plasma proteins, steroid-binding proteins, enzymes, mucopolysaccharides and non-steroidal ovarian factors (inhibin). Inhibin is mainly produced by the granulose cells of the ovaries, and its major physiological role is specific suppression of secretion of follicle stimulating hormone (FSH) from the anterior pituitary gland (Guilbault *et al.*, 1993; Taya *et al.*, 1991). Cessation of the treatment with follicular fluid results in a rebound release of FSH in larger quantities and leads to several follicles reaching the stage of ovulation. Kumar *et al* (1998) reported that the used of buffalo follicular fluid at 12 hours intervals for 4 days in the luteal phase of the cycle increase 40 % in ovulation rate.

Bovine follicular fluid, which is known as waste of the abbatoir, has a great potential for induce superovulation in animals. The reasons for the study were to explore the potential of bovine follicular fluid on the pregnancy rate and twining rate in Ettawa crossbreed goats.

Materials and Methods

Twenty mature Ettawa crossbreed goats, one-and-a-half to two years of age and weighing 23 to 30 kg were used in these experiments. Animals, which had exhibited at least two consecutive cycles of normal length (20 to 21 days), were included in the experiment. The animals then maintained in a one groups and keep in the one house of each. They fed fresh grass, leave and concentrate in the proportion of 40%, 30% and 30%. The dried fed were given 5% of the body weight. The concentrate was given one time a day in the morning while fresh grass was given twice a day in the morning and in the afternoon. Water provided *ad libitum*.

Oestrus was synchronised in 20 Ettawa crossbreed goats with two injections of 6.25 mg prostaglandin (Lutalyse™, Pharmacia & Upjohn, and NSW) given 11 days apart. They were randomised into two equal groups; group I received 3 ml charcoal-extracted cow follicular fluid daily at 06.00 and 18.00 on the 12th to 15th day of the synchronised oestrus cycle and group II received an equal volume of normal saline at the same times. In addition, all the goats received an intramuscular injection of 6.25 mg prostaglandin one day after the last treatments with follicular fluid or normal saline. Observations for oestrus were made tree time a day.

Bovine ovaries were collected from an abattoir and transported to the laboratory on ice. Follicular fluid was aspirated from follicles of all sizes on the ovarian surface

using needle (22 g) and syringe. In order to remove the steroids, activated charcoal (Polypharm, Germany) was added at 5mg/ml to the fluid and stirred at room temperature for one hour in a glass beaker. The mixture was centrifuged at 12,000 g for one hour at 4°C. The supernatant was decanted, and samples were stored at –20 °C until use.

Evaluation of the frozen-thawed semen was performing using the method described by Evans and Maxwell (1987). Artificial insemination was done at the time of oestrus or 56 hours after the injection of prostaglandin with double dose of frozen-thawed semen.

Pregnancy diagnosis were perform based on the clinical sign such as the enlargement of the abdomen and non return to oestrus after AI, also the increase of the concentration of plasma progesterone >1 ng/ml (Robertson and Sarda, 1971; Susmel and Piasentier, 1992). Plasma progesterone were measure using RIA as previously describe by Junaidi *et al.* (2000). The does were diagnosed as positive pregnant when the plasma progesterone >1 ng/ml. The pregnant does were allowed to proceed to term.

The statistical software package statistic version 4.1 (© 1994, Analytical Software) will be used for performing the analysis of all the experiment data. Data are presented as the mean (SEM).

Results and Discussion

The average interval from the injection of prostaglandin (PG) to the onset of oestrus did not significantly difference (P>0.05) between group (52 ± 12.6) hours in group I compare with (58 ± 7.8) hours in group II (Table 1). This high degree of synchrony achieved in this study indicated that does receive the PG injection at the stage when the corpus luteum was responsive. This finding is consistent with observations reported previously (Godfrey *et al.*, 1999; Kusina *et al.*, 2000). This response of synchrony achieved in this study can facilitate easy management of breeding does both during pregnancy and at parturition.

Table 1. The comparison of the onset of oestrus and the duration of oestrus between group I and group II after injection with prostaglandin (mean ± SE)

Description	Group I	Group II
Number of does	10	10
Onset of oestrus (hours)	53.4 ± 4.5^{a}	57.2 ± 8.1^{b}
Duration of oestrus (hours)	32.8 ± 5.2^{c}	34.6 ± 2.3^{d}

The values with same superscript in the same row differ significantly (P<0.05)

The use of bovine follicular fluid (BFF) has been reported delayed the onset of oestrus in goats (Miller *et al.*, 1979; Agarwal *et al.*, 1995; Kumar *et al*, 1998). In our study, there were no significantly differences in the onset of oestrus among treated and control does. The duration of treatment with BFF and the stage of the cycle when the treatment is initiated appear to play a critical role in the display of oestrus. When the administration of BFF ceases either before or within 24 hours of the induction of luteolysis, ovulation occurs at the normal time. However, treatment of does with BFF in the late follicular phase for more than 24 hours after the induction of luteolysis results in a delay in ovulation (Miller *et al*, 1979). In this study, the administration of BFF ceased 24 hours before the induction of luteolysis. It would be therefore appear that administration of BFF can be used to induce superovulation in goats when given at the luteal stage of the cycles.

The mean duration of oestrus did not differ significantly between groups 32.8 ± 5.2 hours in group I compare with 34.6 ± 2.3 hours in group II (Table 1). The results are similar to those reported by Kumar *et al.* (1998).

Assessment of the quality of frozen-thawed semen used in this study showed that the motility was 48 ± 7.9%, the concentrations were 25.1 ± 4.7 x 10^6 and the abnormality was 7.5 ± 2.3%. The quality of frozen-thawed semen found in this study was minimal prerequisite to AI in goats as recommended by Evans and Maxwell (1989).

Early pregnancy diagnosis in this study was based on plasma progesterone concentration, the concentration > 1 ng/ml 40 days after artificial insemination (AI) was judge as positive pregnant. Based on the plasma progesterone concentration indicated that 80 % does in group I were pregnant while does in group II indicated 60 % were pregnant. However, only 6 does in group I (60%) each deliver twin kids and only 2 does (20 %) in group II give single and twin kids (Tabel 2). There are larger number of animals incorrectly judged as positive, this may due to the presence of basal progesterone levels higher than usually found in the breed or may due to the different assay protocol used by different investigators.

Table 2. Reproductive efficiency of Ettawa crossbreed goats following synchronisation using prostaglandin and after treated with bovine follicular fluid

Description	Group I	Group II
Number of does	10	10
No. does bred (AI)	10	10
Positive pregnant (P> ng/ml 40 days after AI)	8	6
No. of does kidding	6	2
Twinning rate	6/6	½

The pregnancy rates in the does treated with BFF were higher than the control group, indicating the BFF increased ovulation rate in treated group. This finding

confirmed the previous observations of Kumar *et al.* (1998), who reported a 40% increase in ovulation rate in goats which received buffalo follicular fluid during luteal stage of the cycle. Miller and Martin (1993) reported there is an increase in ovulation rate up to 30% on the sheep received bovine follicular fluid. This study suggested that treatment with BFF induced an increase in FSH secretion that induce more ovulations and increase pregnancy rate and twining rate in the treated animals in this experiment.

It can be concluded that oestrus synchronisation in Ettawa crossbreed goats can be induced using single injection of prostaglandin on the luteal phase of the cycles. The use of bovine follicular fluid at 12 hours interval for 4 days can increase pregnancy rate and enhance the twining rate in the Ettawa crossbreed goats

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