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DETERMINATION OF EARLY PREGNANCY THROUGH PLASMA PROGESTERONE CONCENTRATION IN KAMORI GOAT

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ABSTRACT

An early pregnancy was determined through plasma progesterone concentration in Kamori goat, under semi-intensive management condition. Thirty adult normal cycling Kamori goats were selected and divided in to three equal (N-10) treatment groups and synchronized with different estrus synchronization protocols. Goats in group A were given progesterone on day 0 and PGF2α with GnRH on day 10^{th} , goats in group B were given PGF2α on day-0 and repeat dose of PGF2α with GnRH on day 10^{th}, whereas goats in group C served as control. All the goats were observed for estrus induction. Goats showing estrus were allowed for breeding, natural breeding was practiced using an intact breeding bucks. The blood samples were collected after day 21^{st} of post-breeding to measure progesterone concentration level with ELISA Reader (Beck-Man Coulter Inc.AD-430) using Progesterone kit (Accu-Bind®, Monobind Inc. USA). The mean (±SEM) progesterone concentration level was recorded as 2.42±1.37, 2.57±1.21, 2.71±1.06 in group A, B, and C, respectively in pregnant and 1.27±0.98 ng/ml in non-pregnant goats. No significant difference (*P*> 0.05) was found among the goat groups A, B and C. Increased progesterone concentration level (post-breeding) in goats was found to be one of the best indicators of pregnancy. This study suggests that the ELISA kit method could be used for early pregnancy determination in Kamori goats.

Keywords: ELISA, goat, Kamori, pregnancy, progesterone

INTRODUCTION

The tropical and sub-tropical climate provides a unique habitant for goat raising. Conducive climate, dense vegetation and rural socio-economic structures are facilitating the goat population in the region. The increased ratio of poverty and unemployment compels under privileged group of people towards the goat farming. Goats are manly reared for meat, milk and hair (Iqbal *et al*., 2008; Kunbhar *et al*., 2016). In Pakistan, continuous increasing trend has been recorded in goat population since last decade. The province of Sindh is bestowed with very fine goat breeds including Kamori goat (Devendra, 2007; Iqbal *et al*., 2008). Kamori is dairy goat breed of Sindh province and famous due to its beauty, high milk yield and heavy body weight (Issani and Baloch, 1996; Iqbal *et al*., 2008; Kunbhar *et al*., 2016). In tropical countries like Pakistan, the breeding season is extended round the year. In extensive management and breeding out of season, increased the need to detect pregnancy at early dates, so the herd can be managed more efficiently. On the other hand decisions can be made for culling or rebreeding unsuccessful bred does to minimize economic losses, incurred because of reduced milk production and kid crop (Al-Merestani *et al.,* 2003; Ameer, 2010). The pregnancy diagnosis techniques in goats include ultrasonography (Bukar *et al.,* 2012; Karadaev, 2015) and measuring of progesterone hormone (Abecia *et al.*, 2012; Ahmed *et al*., 2014; Najafi *et al*., 2014). Progesterone concentration is a good predictor tool to be used to detect pregnancy. As for the pregnancy detection is concerned, no information in hormonal analysis for pregnancy detection is available for goats particularly Kamori breed of Sindh. Therefore we conducted this study to detect pregnancy through the analysis of plasma progesterone concentration with Enzyme Linkage Immuno Sorbent Assay (ELISA) Kit method and to validate this kit in Kamori goat.

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MATERIALS AND METHODS

Management of animals

The study was conducted to determine the pregnancy through plasma progesterone concentration in blood. Thirty adult Kamori goats were selected on the basis of known history, farm record, normal cycling, non-pregnant and recently parturited. The goats were maintained under semi intensive management condition at Kamori goat farm. The goat flock was allowed for grazing in daytime and stall-feeding was practiced on return of flock in to the sheds. The seasonal green fodders were made available according to the season. The concentrates ration (barley, cotton seed cake and wheat bran) at the rate of 250 g were given/day/animal and common salt blocks were placed in mangers for licking. The clean water was provided ad-libitumin the sheds. All goats were identified with ear tag numbers. Vaccination and deworming of goat flocks were performed as per designed schedule.

Estrus synchronization of goats

The goats were synchronized using estrus synchronization hormonal protocols. The goats were divided into three equal (N-10) treatment groups. Goats of group A received progesterone (Pregtone, Selmore, Pakistan) IM on day 0 and PGF2α (Dalmazine, FATRO, Italy) along-with GnRH (Dalmarelin, FATRO, Italy) IM both on day 10th, goats in group B were given PGF2α IM on day-0 and repeat dose of PGF2α in combination with GnRH IM on day 10th, whereas goats of group C were given no any treatment and served as control. All the goats were observed closely for estrus. Goats showed estrus were allowed for natural breeding using an intact breeding bucks. The date of breeding was considered as day 0 of gestation.

Collection of blood samples

Blood samples were collected with 5 ml sterilized syringe in EDTA test tubes, from right jugular vein of goat after 3rd week (day 21st) post-breeding. After collection, blood samples were centrifuged at 3000 x rpm for 10 minutes. The serum was separated from blood cells and filled in capped plastic tubes (1.5 ml Eppendorf tubes) and stored at -20°C till hormonal analysis. The plasma progesterone concentration was analyzed with Enzyme Linked Immuno Sorbent Assay (ELISA) Reader (Beck-Man Coulter Inc. AD-430) with progesterone kit (Accu-bind®, Monobind Inc. USA) as per manufactures instructions.

Statistical analysis

The data were analyzed using, Students Edition of Statistics (SXW-Version 8.1) to calculate the means and standard errors of means.

RESULTS AND DISCUSSION

The present study was conducted to determine the pregnancy on the basis of plasma progesterone concentration. In current study the progesterone concentration level on day 21st of post breeding was measured and recorded as 2.42±1.37, 2.57±1.21, 2.71±1.06 ng/ml in group A, B and C, respectively in pregnant goats, whereas a low concentration of 1.27±0.98 ng/ml was recorded in non-pregnant goats. A non-significant difference *(P*> 0.05*)* was found among the treatment groups A, B and C. However; all these three groups were significantly (*P*< 0.05) different from the non-pregnant goats for progesterone concentration in does (Table 1).

Table 1. Progesterone concentration level in pregnant Kamori goat

Group	Treatment protocols	Goats observed (N)	Goats induced estrus and served (N)	Mean (±SEM) P-4 concentration (ng/ml) day 21st post breeding	
A	Progesterone+ GnRh+PGF2α	10	10	2.42±1.37^{a}	
B	PGF2α +GnRH + PGF2α	10	10	2.57± 1.21^{a}	
C	Control	10	02	Pregnant	2.71±1.06^{a}
				Non-pregnant	1.27±0.98^{b}

The results recorded from current study were in accordance to the results reported by Gaafar *et al.* (2005); Khanum *et al.* (2008); Najafi *et al*. (2014). They reported that the plasma progesterone concentration was more than 2.2 ng/ml, when samples were collected after 3rd week post breeding and considered as good indicator of pregnancy. From day 21 post breeding and then after an increased level in progesterone concentration was reported as 2.3 ng/ml in pregnant ewes and does (Duggal *et al.*, 2001; Gonzalez *et al.,* 2004; Najafi *et al*., 2014). Their reported are in agreements with the results of our study on Kamori goat. Once a doe becomes pregnant the progesterone becomes the key hormone and remains dominant till initiation of parturition (Islam *et al.*, 2014; Karadaev, 2015). An increased blood progesterone hormonal level was recorded through useful technique to diagnose pregnancy by hormonal analysis (Gaafar *et al.,* 2005; Islam

et al., 2014). In contrast to that the non-pregnant goats may be reliably identified by their low progesterone level (Najafi *et al*., 2014; Karadaev, 2015). Progesterone could be a prime for systemic hormonal signal of maternal or fetal origin (Gonazelz *et al.*, 2004; Najafi *et al*., 2014). Furthermore the results reported by Alwan *et al.* (2010) were also in accordance to the results recorded in the current study. They reported that the progesterone concentration was reached up to 2.9±2.5 and 3.34±2.3 ng/ml in pregnant ewes and does, respectively. The progesterone concentration in early days of pregnancy were not different from the results recorded in current study in Kamori goat.

In the present study the overall conception rate was recorded as 90.91% in goats. It was further noticed that the goats conceived 100% in group A and 80% in group B, while in group C only 20% goats were able to conceive. The significant difference was found among the goat groups. The better conception rate was recorded in treatment groups A as compared to group B and C in Kamori goat (Table 2).

Table 2. Conception/ pregnancy rate in various groups of Kamori goat

Groups	Treatment Protocols	Observation (N)	Goats served (N)	Conception/ Pregnancy% (N)	
A	Progesterone + GnRH + PGF2α	10	10	10	100[a]
B	PGF2α+GnRH + PGF2α	10	10	08	80[b]
C	Control	10	02	02	20[c]
Over all		30	22	20	90.91

The findings recorded in current study for conception and pregnancy percentage on the basis of progesterone concentration were in close agreements to the results reported by Ahmed *et al.* (2014). They reported the pregnancy rate as 91% in beetle goats. The similar trend was also reported by Ameer, (2010). They reported the pregnancy rate was 85-88% in goats. Whereas results reported by Alwan *et al.* (2010) and Najafi *et al*. (2014) were not too far from our results. However; an increased level of progesterone concentration is the indication of pregnancy and it is better to predict that the doe is pregnant. On the other hand doe may be diagnosed non-pregnant having low progesterone level (1ng/ml) around day 20 to 24th post -breeding.

The results of current study for non-pregnant does on the basis of progesterone concentration are in accordance to the results reported by Aly *et al.* (2002) and Islam *et al.* (2014). During this period they reported the progesterone level was very low (1.1 ng/ml) and goats were considered as non-pregnant (Matsas, 2007; Najafi *et al*., 2014). Low progesterone concentrations in maternal blood of post breeding could be predicted that the goat is non-pregnant. The progesterone measurement at specific time after post-breeding is a useful management tool to determine pregnancy at an early date (Debnath, 2012). It is the evidence that in pregnancy, the progesterone level in the blood increases to certain level (Gonzalez *et al*., 2004; Gaafar *et al.,* 2005).

CONCLUSION

It is concluded that pregnancy reliably can be diagnosed at Day 21 after breeding with ELISA kit method in Kamori goats. When the date of breeding is known, only a single blood sample is required for ELISA test to differentiate pregnant from non-pregnant goats. ELISA kit method is more reliable, economical and has validation and could be used for early pregnancy diagnosis in Kamori goat too.

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