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Monitoring Estradiol and Progesterone Content of Saanen Goats During Pregnancy

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ABSTRACT: Saanen is a specialized dairy goat breed originating from Switzerland, imported and encouraged to be raised in Vietnam. This article presents the results of a survey of estradiol and progesterone levels during pregnancy in Saanen goats at the Binh Duong Large Livestock Research and Development Center. After monitoring mating, goats were sampled for blood every 7 days until birth, at 6-8 am. Hormone levels were determined by electrochemiluminescence assay, with the Cobas E601 immunoassay system. The results showed that after 14 days of conception, progesterone levels gradually increased (3.21±0.36 ng/ml) and shifted to higher levels, peaking on day 70 of pregnancy (12.28±0.52 ng/ml) (in the 1-2 years old group); corresponding values were 3.18±0.75-11.88±0.92 ng/ml (in the 3-4 years old group). The high value range was maintained throughout pregnancy and decreased rapidly in the 14-21 days before birth in both age groups. Estradiol, maintained at low concentrations from 0.73±0.35 to 2.65±1.25 pg/ml (in the 1-2 years old group); from 0.98±0.48 to 3.27±1.95 pg/ml (in the 3-4 years old group) in the first 21 days of pregnancy and then gradually increased to above 10 pg/ml (in both age groups) on day 28. Estradiol continued to increase and reached a maximum level (205.48±32.81 pg/ml) in group 1 on day 126 and had an average value below 10 pg/ml within 1-2 days after giving birth. The results of this study help diagnose pregnancy, predict gestational age for effective reproductive management, and serve as a reliable reference when using assisted reproductive tools and strategies to improve reproductive performance in goats.

KEYWORDS: Estradiol, goat, pregnancy, progesterone, Saanen.

I. INTRODUCTION

Goat farming plays an important role in the economy. In recent years, goat farming has developed strongly, especially in the Southeast provinces of Vietnam, many large-scale goat farms have been established and developed. Reproduction is a major factor contributing to the increase in meat and milk production efficiency of female goats. Determining the levels of estradiol and progesterone hormones indicates the reproductive physiological status of livestock and is important in selecting, mating, and supporting effective reproduction. On the other hand, many variables such as nutrition, stress, temperature, climate conditions, disease, and age can affect the levels of reproductive hormones. Therefore, determining the reference values of these parameters for goat breeds being raised is very important. Due to this fact, many studies have examined reproductive hormone parameters for many goat breeds raised in different locations around the world. However, in Vietnam, there has been no research on these parameters for domestic and imported goat breeds currently being raised.

To accurately characterize the different phases and determine the normal progesterone profiles of the estrous cycle in goats, several studies have been conducted using radiometric methods or modified enzyme immunoassays (EIAs) to measure serum progesterone in female goats. Plasma progesterone, estrogen, LH, FSH and PRL variations were measured by RIA (Zamfirescu et al., 2009). Several studies have reported plasma progesterone concentrations during the estrous cycle in goats for different breeds (Kanuya et al., 2000; Marcin et al., 2004; Khanum et al., 2019). Khadiga et al., (2005) studied the changes in plasma progesterone concentrations during different phases of the estrous cycle in Damascus goats. Early pregnancy was determined through plasma progesterone concentrations, under semi-intensive rearing conditions. Blood samples were collected after the 21st day of mating to measure progesterone concentration by ELISA. Increased progesterone concentration (post-mating) in goats is considered one of the best indicators of pregnancy. This study suggests that the ELISA kit can be used to determine early pregnancy in goats (Yasin et al., 2019).



However, there is currently little information on changes in plasma estradiol and progesterone during pregnancy in goats compared to other farm animals, especially imported goat breeds. In this study, we monitored plasma estradiol and progesterone levels by electrochemiluminescence assay (ECLIA) to provide data on these two hormones in Saanen goats, as a reference for determining pregnancy and predicting gestational age. This study suggests that the hormone levels determined by ECLIA can be used to determine early pregnancy as well as determine gestational age and predict due date in goats. In reproductive management and breeding, the need for early pregnancy detection is increasing, so that the herd can be managed more effectively. On the other hand, decisions can be made in case of unsuccessful mating to minimize economic losses in livestock production.

II. MATERIALS AND METHODS

A. Animals, breeding, care and housing

Thirty-four female Saanen goats (n=18, 1-2 years old; n=16, 3-4 years old), selected on the basis of known history, healthy based on clinical monitoring, and successful mating. The goats were raised under semi-intensive conditions at the Center for Research and Development of Large Livestock, Lai Hung Commune, Bau Bang District, Binh Duong Province. All goats were identified by ear tag numbers. Vaccination and deworming of the goats were carried out according to the center's schedule.

Goats are raised semi-intensively, with a density of 2 m²/head. The barn is designed in the style of a stilt house, with a wooden floor 1m above the cement floor and a roof made of corrugated iron. The goats are grazed during the day and fed in the barn when they return to the barn. The goats are fed 2 meals/day and night, including 1 breakfast (around 7-8am) and 1 dinner (around 4-5pm), including green elephant grass, mixed bran (De Heus), leaves (binh linh, or jackfruit leaves, soapberry). On average, each goat gets 2-4 kg of grass, 0.5-0.8 kg of bran, 0.5 kg of leaves/day. Dry straw is rolled up and placed on a trough for the goats to eat at night. Drinking water is clean water, put in a clean trough placed in the barn for the goats to drink freely. The water trough is cleaned daily and the water is changed once a day. The barn is washed with a water sprayer daily. Sanitation and disinfection are carried out every 2 weeks. The disinfectant solution used is BESTAQUAM-SR with ingredients: didecyl dimethyl ammonium bromide, mixed at a ratio of 1/400.

B. Sample collection and analysis

- After monitoring for mating, goats are sampled once every 7 days until birth, at 6-8 am. The day of mating is considered day 1 of pregnancy.

- Collect 2 ml of venous blood properly, the sample is quickly put into an anticoagulant tube (EDTA-K2), label the sample tube, shake gently, preserve and transfer to the laboratory, centrifuge the blood sample. Centrifuge the sample for 15 minutes at 1,000 x g at 4° C within 60 minutes after collection. After centrifugation, collect 1 mL of extracted plasma into a 1.5 mL eppendorf vial and store the sample at -20°C. Do not repeat the freeze/thaw cycles.

- Analysis: Thaw once, leave the specimen, standard, control at room temperature (20-25°C) and shake well before testing. To avoid any influence on the results, the specimens, standards and controls are analyzed within 2 hours. The amount of P4 and E2 is determined by electrochemiluminescence testing, with the Cobas E601 immunoassay system - Roche Diagnostics (Switzerland). The testing procedure follows the manufacturer's KIT instructions.

C. Data Analysis: Data was expressed as mean (Mean ± SD). Analysis of ANOVA and Post hoc test with Tukey-Kramer test to evaluate the difference between groups (P<0.05). Statistical parameters were processed by MS-Excel 2020 software.

III. RESULTS AND DISCUSSION

The results of monitoring the estradiol and progesterone levels of Saanen goats during pregnancy according to 2 age groups are presented in Table 1 and Table 2. Figure 1 and Figure 2 show the change charts of the 2 hormones estradiol (E2) and progesterone (P4) of 2 individuals F8SA (16-month-old Saanen female goat, group 1: 1-2 years old) and F2OSA (32-month-old Saanen female goat, group 2: 3-4 years old).

Tables 1 and 2 show that, after 14 days of conception, the progesterone level gradually increased (3.21±0.36 ng/ml) and moved to a higher level, peaking on day 70 of pregnancy (12.28±0.52 ng/ml) (in the 1-2 years old group); The corresponding values were 3.18±0.75-11.88±0.92 ng/ml (in the 3-4 years old group). The high value range was maintained throughout pregnancy and decreased rapidly during the 14-21 days before delivery, reaching a minimum of 0.94 ng/ml (in group 1) and 1.05 ng/ml (in group 2) at the end of pregnancy (Tables 1, 2, Figures 1, 2, 3). The maximum progesterone levels during pregnancy were 12.83 ng/ml (in group 1) and 13.93 ng/ml (in group 1).

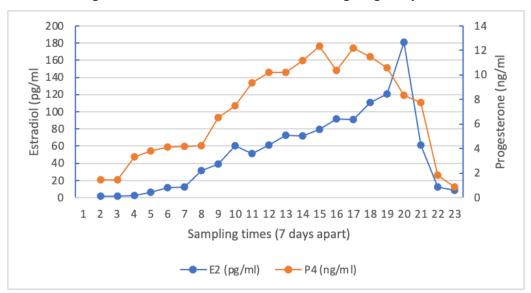


Figure 1. Changes in hormone levels of F8SA goat (16 months old) during pregnancy

Regarding the estradiol profile, after conception, it remained at low concentrations from 0.73±0.35 to 2.65±1.25 pg/ml (in the 1-2 years old group); from 0.98±0.48 to 3.27±1.95 pg/ml (in the 3-4 years old group) in the first 21 days and then gradually increased to levels above 10 pg/ml (in both age groups) on day 28. Estradiol continued to increase and reached a maximum level (205.48±32.81 pg/ml) in group 1 on day 126 and decreased again to a baseline value below 10 pg/ml within 1-2 days after birth. Similarly, the change in estradiol hormone in group 2, after reaching the peak on day 126 (175.84±24.74 pg/ml) also gradually decreased, reaching a minimum of 16.68±7.43 pg/ml at the end of pregnancy.

This result is also consistent with studies in other goat breeds. As observed in other studies, progesterone concentration remained at the basal level throughout the estrus period. In Damascus goats, progesterone levels in the luteal phase ranged from 2.6 to 5.4 ng/ml, which was similar to that in dwarf goats (Khadiga et al., 2005). In studies on native East African goats, the mean plasma progesterone concentration ranged from 2.6 to 10.8 ng/ml from conception to mid-pregnancy (Kanuya et al., 2000). The overall increase in progesterone levels during pregnancy and the decline towards the pre-partum and parturition period, observed in Dwarf goats, is similar to other breeds (Khanum et al., 2008).

The pre-partum decline in progesterone levels correlates with the onset of parturition. Gaafar et al. (2005) reported that in Damascus goats, estradiol and estrone sulphate concentrations were inversely correlated with progesterone (r = -0.902; p < 0.01) during the last 9 days of gestation. In the case of Saanen goats in this study, progesterone showed a decreasing trend from day 126 of gestation, while estradiol levels gradually increased and peaked at day 133 of gestation.

	ESTRADIOL (pg/ml)			PROGESTERONE (ng/ml)		
Day	Mean ±SD	MIN	MAX	Mean ±SD	MIN	MAX
1	0.73±0.35	0.25	1.58	1.02±0,49	0.16	1.27
7	1.25±0.66	0.42	1.95	1.47±0.54	1.25	2.25
14	1.47±0.73	0.57	2,78	3.21±0.36	2.56	3.36
21	2.65±1.25	1.26	4.69	3.66±0.38	3.53	3.67
28	10.47±0.91	8.95	11.34	3.16±0.46	2.38	3.54
35	14.85±2.04	12.36	17.58	3.65±0.54	2.87	4.52
42	34.13±5.32	26.24	45.39	4.74±1.32	3.72	6.86
49	47.27±5.46	36.95	63.71	6.22±1.56	3.28	7.32
56	55.83±3.66	45.28	72.84	9.51±2.32	5.82	11.81
63	58.94±5.57	41.22	83.91	11.42±1.36	8.36	12.65
70	61.05±4.42	51.56	85.29	12.28±0.52	11.35	12.66
77	67.56±4.14	53.72	91.58	11.53±0.38	10.47	12.72
84	73.17±5.65	60.81	94.93	11.46±0.69	10.26	12.56
91	81.67±6.15	68.58	102.57	11.52±0.86	10.28	12.54

Table 1. Hormone index of Saanen goats aged 1-2 years (n=18) during pregnancy

Monitoring Estradiol and Progesterone Content of Saanen Goats During Pregnancy							
	98	90.85±7.18	79.56	112.68	11.94±1.57	9.65	12.83
	105	92.43±8.62	82.53	131.52	10.73±1.27	10.37	11.72
	112	99.83±9.73	87.35	126.53	11.26±0.95	9.36	11.56
	119	125.75±18.86	99.18	136.67	8.97±0.72	7.56	9.27
	126	205.48±32.81	150.81	240.58	8.68±0.36	7.76	9.25
	133	145.82±33.65	87.56	181.29	4.65±3.26	2.45	8.73
	140	86.45±26.34	56.54	167.43	1.24±0.35	0.87	1.69
_	147	19.83±6.65	8.13	21.85	0.94±0.41	0.84	1.25

It is not possible to determine with certainty whether the increase in estrogens preceded the decrease in progesterone levels. When compared with many studies it can be concluded that Bach Thao goats exhibit progesterone and estradiol profiles comparable to other breeds and have a short gestation period with multiple parturition being common. Reproductive management, using reproductive aids and improved nutritional strategies can further improve reproductive efficiency in goat.

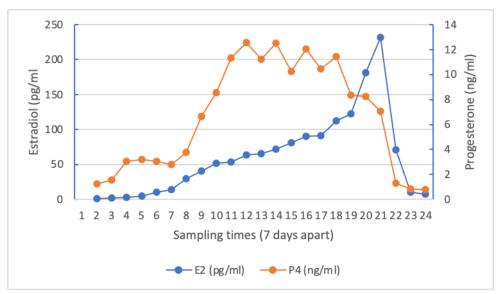


Figure 2. Changes in hormone levels of F18SA goat (32 months old) during pregnancy

In the study by Yasin et al. (2019) on Kamori goats (Pakistan), the progesterone concentration level on the 21st day after parturition was measured and recorded as 2.42 ± 1.37 , 2.57 ± 1.21 , 2.71 ± 1.06 ng/ml in the pregnant goat group, while a low concentration of 1.27 ± 0.98 ng/ml was recorded in the non-pregnant goats. The results recorded from the present study are in agreement with the results reported by Duggal et al., (2001), M Islam et al., (2014). It was reported that the plasma progesterone concentration was more than 2.2 ng/ml, when the samples were collected after the 3rd week after parturition and was considered as a good indication of pregnancy. From day 21 post-mating an increase in progesterone levels of 2.3 ng/ml has been reported in pregnant goats (Alwan et al., 2010; Aly et al., 2002). These reports are consistent with our findings in Boer and Boer goats. When a female becomes pregnant, progesterone becomes the dominant hormone and remains dominant until the onset of parturition (Matsas, 2007). Increased levels of progesterone in the blood have been reported as a useful technique for diagnosing pregnancy by hormonal analysis (Gonazelz et al., 2001). Progesterone may be a key factor for systemic hormonal signaling of maternal or fetal origin (Duggal et al., 2001; Aly et al., 2002). Furthermore, the results reported by Alwan et al. (2010) are also consistent with those reported in the present study. Progesterone concentrations reached up to 2.9 \pm 2.5 and 3.34 \pm 2.3 ng/ml in pregnant individuals, respectively. Progesterone concentrations during the early days of pregnancy were not different from those reported in the present study in Saanen goats.

Thus, an increase in progesterone concentration is a sign of pregnancy and is better predictive of pregnancy. On the other hand, non-pregnancy can be diagnosed when progesterone levels are low (less than 1 ng/ml) around day 21 after mating. The results of the study on non-pregnant goats were based on the progesterone concentrations reported in Black Bengal goats (MIslam et al., 2014). During this period, progesterone levels were very low (1.1 ng/ml) and the goats were considered non-pregnant. Low progesterone concentrations in maternal blood after mating can predict that the goat is not pregnant. Measuring progesterone at a specific time after breeding is a useful management tool to determine pregnancy at an early stage. It is

evident that during pregnancy, progesterone levels in the blood increase to a certain level (Aly et al., 2002; Gonazelz et al., 2004).

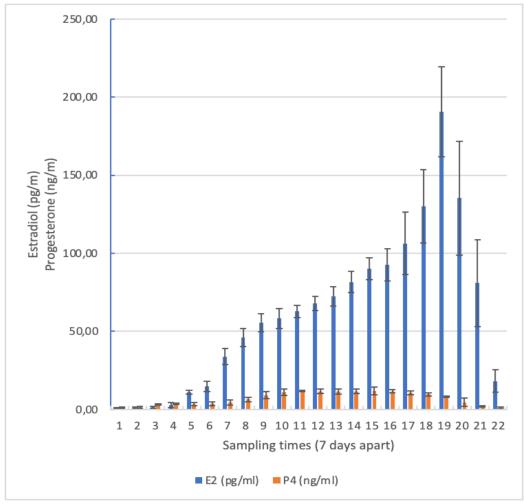


Figure 3. Changes in E2 and P4 hormone levels (average of both age groups) of Saanen goats during pregnancy

The results of this study are also similar to a recent publication on plasma hormone levels of two breeds of goats Bach Thao and Boer (Nguyen and Nguyen, 2022). The results of this study showed that, after conception, progesterone levels were high (3.02±0.34 – 12.20±0.50 ng/ml in Boer goats; 3.26±0.47-11.88±0.92 ng/ml in Bach Thao goats); the high value range was maintained throughout the gestation period and decreased rapidly in the 19 days before parturition, reaching 0.87-0.94 ng/ml on the day of parturition. The average maximum progesterone levels during pregnancy were 12.20±0.50 ng/ml in Boer goats and 11.88±0.92 ng/ml in Bach Thao goats. Estradiol remained at lower concentrations ranging from 0.96±0.44 to 1.42±0.68 pg/ml (in Bach Thao goats) during the first 14 days; from 0.81±0.46 to 2.62±1.28 pg/ml (in Boer goats) during the first 21 days and then gradually increased to levels above 10 pg/ml (in both goat breeds) on day 28. Estradiol continued to increase and reached a maximum of 204.48±36.68 pg/ml in Boer goats on day 126 and decreased below 10 pg/ml within 1-2 days after parturition (Nguyen and Nguyen, 2022).

Interestingly, we also observed that progesterone concentrations on days 7 and 14 after insemination showed significant differences between the goats that conceived and those that did not conceive. We can detect pregnancy as early as possible by using the ECLIA method to estimate progesterone on day 14, even on day 7-10 after insemination, while it can only be confirmed on day 20 by ultrasound (Hussein, 2010). From the study, it was found that progesterone levels that started to increase after 1 week of insemination and were higher on the day of insemination (day 0) may have a higher probability of conception. Currently, there is no report on the relationship between progesterone levels on the day of insemination and conception rate after synchronization procedure in goats. Therefore, further research is needed to confirm whether progesterone levels on the day of mating may be an important factor determining the success of insemination.

	ESTRADIOL (pg	ESTRADIOL (pg/ml)			PROGESTERONE (ng/ml)		
Day	Mean ±SD	MIN	MAX	Mean ±SD	MIN	MAX	
1	0.98±0.48	0.77	1.46	1.22±0.46	0.24	1.87	
7	1.15±0.34	0.56	2.18	1.28±0,56	0.77	2.49	
14	1.46±0.69	0.67	2.66	3.18±0.75	1.76	3.96	
21	3.27±1.95	1.38	5.97	3.71±0.72	2.92	4.87	
28	11.72±1.76	7.36	11.46	3.46±1.63	2.31	4.88	
35	14.35±4.56	10.61	25.54	3.54±1.71	2.44	3.89	
42	33.68±4.68	27.52	41.83	3.86±1.95	2.41	6.35	
49	44.84±6.25	38.66	51.78	6.43±1.32	4.29	7.86	
56	55.35±8,02	41.25	61,45	8.78±2.51	6.45	11.58	
63	57.53±7.15	40.93	60,87	10.56±2.63	7.25	12.84	
70	64.54±3.32	52.75	64,45	11.61±3.65	9.26	13.18	
77	68.28±5.09	63.26	72,44	11.72±2.18	9.21	14.11	
84	71.78±6.95	62.43	81,16	11.71±2,49	9.35	12.82	
91	81.56±7.43	70.96	83,99	11.63±2.24	10.22	13.93	
98	89.26±6.84	84.28	91,32	11.64±3.32	9.88	13.59	
105	92.65±11.82	81.57	134.81	12.37±0.92	10.52	12.18	
112	112.91±30.47	81.12	171.58	9.96±1.23	8.36	11.56	
119	134.55±27.83	98.67	183.88	10.36±1.24	8.87	11.65	
126	175.84±24.74	146.46	215.66	7.68±1.60	8.53	9.56	
133	124.76±38.96	60.28	198.73	4.49±2.17	2.65	7.75	
140	75.34±29.35	43.68	235.28	2.61±0.65	0.74	2.24	
147	16.68±7.43	5.92	22.26	1.05±0.73	0.85	1.35	

Table 2. Hormone index of Saanen g	nats aged 3-4 years	(n=16) during pregnancy
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IV. CONCLUSION

Saanen goats exhibited progesterone and estradiol profiles during pregnancy that were comparable to those observed in other goat breeds worldwide, with differences between the two age groups studied. ECLIA monitoring of progesterone and estradiol can be used to determine pregnancy, estimate gestational age and due date. Monitoring of hormone dynamics helps in effective reproductive management, and the results of this study serve as a reliable reference when using reproductive aids and improved nutritional strategies that can further improve reproductive efficiency in goats.

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