Serum Progesterone profile of pregnant Ettawa crossbred does at day-0, -21, -42, -63 and -84

Profil Progesterone serum kambing peranakan Ettawa bunting pada hari ke-0, -21, -42, -63 dan -84

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ABSTRACT

This study aimed to determine the progesterone concentrations of Ettawa crossbred does at day-0, -21, -42, -63, and -84 after mating. Five Ettawa crossbred does were injected with PGF₂α twice with an interval of 11 days for estrous synchronization followed by natural mating on day-3 after the second PGF₂α injection. Blood was collected through the jugular vein for the measurement of serum progesterone concentrations using ELISA, whereas pregnancy was diagnosed at 90 days after mating by abdominal palpation. The results showed that all does were on estrus on the third day after the second PGF₂α, three of them diagnosed to be pregnant following mating. The progesterone concentration at day-0 was 1.52 ± 0.33 ng/mL, increased (p <0.05) to 7.08 ± 1.02 and 5.50 ± 2.82 ng/mL at day-21 and day-42, and increased (p <0.05) again to 12.01 ± 5.30 and 12.04 ± 0.30 ng/mL at day 63 and day 84, respectively. There was no significant difference in progesterone concentrations between day 21 and day 42 and between day 63 and day 84. It could be concluded that there was a biphasic increase of progesterone concentrations of the pregnant Ettawa crossbred does, i.e. from day-0 to day-21/day-42 and from day-21/day-42 to day-63/day-84.

Keywords: Ettawa crossbred, estrus synchronization, post-mating, progesterone concentration

INTRODUCTION

Ettawa goat is a popular name in Indonesia for Jamnapari (or Jamunapari), a breed of goat originating from the Indian subcontinent (Susilowati et al., 2019). The Ettawa crossbred goat is a crossbreed between Ettawa and Kacang goats (Indonesian local goat) (Winaya et al., 2017). This breed is widely spread throughout Indonesia and has a similar appearance to an Ettawa goat with a smaller size. Ettawa crossbred goat is potential as one of the sources of animal protein obtained through both meat and milk. Ettawa crossbred goats could be categorized as superior dairy goats because they could produce milk on average 2 to 3 liters per day (Triswanto et al., 2019). Therefore, Ettawa crossbred goats are interesting to raise.

In the development of the livestock industry, reproductive management is vital to improve productivity and maintenance management, and good breeding. The reproductive cycle of livestock relates to various phenomena, including puberty and reproduction maturity, breeding season, estrous cycle, and postpartum reproduction activity (Hafez and Hafez, 2000). Progesterone

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concentration in puberty showed cyclic ovarian activity before the first estrus. Therefore, puberty could be interpreted as the progesterone concentration in blood samples exceeding one ng/mL (Estill, 2015). The estrous cycle is the rhythm of certain physiological functions of the gonadal system in animals after puberty (Lestari and Ismundiono, 2014) that could be divided into four periods, i.e. proestrus, estrus, metestrus, and diestrus. The reproductive system begins to prepare for ovulation in the proestrus phase (Dávila et al., 2017). The metestrus phase is showed by the sudden cessation of the estrus phase, and the diestrus phase, shows the development of corpus luteum and progesterone production (Ismundiono et al., 2010). During pregnancy, uterine growth and development are affected by increasing progesterone and estradiol concentrations (Anderson, 2003). Progesterone concentration could be measured in biological fluids such as blood and milk. A Low concentration of progesterone was found in non-pregnant animals, while pregnant animals showed higher concentration (Nepal et al., 2019). Progesterone is primarily produced by granulosa-lutein cells in the corpus luteum (Cable et al., 2021). Progesterone concentration in serum/plasma or milk could be used for determining the specific stage in the reproductive cycle of observed animals (Antanaitis et al., 2020).

Therefore, this study aims to determine the profile of serum progesterone, which can be useful information in increasing the reproductive efficiency of Ettawa crossbred does.

MATERIALS AND METHODS

This study was approved ethically by Animal Care and Use Committee of the Faculty of Veterinary Medicine, Universitas Airlangga, with ethical clearance No.2.KE.143. 07.2019. The does used in this study were reared at the Technical Implementation Unit of Livestock Breeding and Forage, Singosari, Malang. Five, 4-5-year-old healthy Ettawa crossbred does weighing 20-30 kg, body condition score (BCS) 2-3 (scale 1-5), and a minimum parity of one, had their estrus synchronized using 7.5 mg intramuscular injection with 11 days interval (Siregar et al., 2013). Estrus was detected visually, followed by natural mating to an Ettawa crossbred buck 12-18 hours after the onset of the observed second estrus. Blood samples were collected through the jugular vein five times with 21 days intervals, i.e., day 0 (estrus), day 21, 42, 63, and 84 after mating.

The measurement of serum progesterone concentration was conducted using Enzyme-Linked Immunosorbsent Assay (ELISA) (He, 2013; Gan and Patel, 2013) with Progesterone ELISA Kit of Fine Test (China) at the Institute of Tropical Disease, Universitas Airlangga. Pregnancy diagnosis was conducted using abdominal palpation 90 days post-mating. Progesterone concentrations at days 0, 21, 42, 63, and 84 of Ettawa crossbred does were analyzed with General model repeated measure at 95% level of significance using SPSS v.23 (Singh et al., 2013).

RESULTS

The five does which had their estrus synchronized showed a 100% (5/5) estrus rate, three days onset of estrus, and 60% (3/5) pregnancy rate after natural mating (Tabel 1). Serum progesterone concentrations of the pregnant does at 0, 21, 42, 63, and 84 days after mating is presented in Figure 1. There were two phases of significant increases (p <0.05) of progesterone concentration from day 0 to day-21/day-42 and from day-21/day-42 to day-63/day-84.

| Table 1 The results of estrus synchronization and pregnancy diagnosis of Ettawa crossbred does |
|---------------------------------|----------------|----------------|
| does # | estrus observed | onset of estrus | pregnancy diagnosis |
|-------|----------------|----------------|---------------------|
| 318   | +              | 3              | -                   |
| 105   | +              | 3              | +                   |
| 125   | +              | 3              | +                   |
| G010  | +              | 3              | -                   |
| 324   | +              | 3              | +                   |

Onset of estrus was counted from the second PGF2α injection; pregnancy was diagnosed by abdominal palpation 90 days after mating.
Figure 1 Serum progesterone concentrations (ng/mL) of pregnant Ettawa crossbred does at day-0, -21, -42, -63, and -84 days (from mating).

DISCUSSION

Three days after the second PGF2α injection, does was observed showing signs of estrus such as the redness and swelling around vulva, tail wagging, and mounted the other goats, followed by a thick and clear discharge from the vulva (Triagil et al., 2020). All of the estrus synchronized does were observed for their estrus after twice PGF2α injection. This result was better than twice PGF2α injection on Kacang that resulting in an 85-95% estrus rate (Susilowati et al., 2020).

Hormonal treatments for synchronization of estrus were essential for improving the goats’ reproductive efficiency and genetic progress (Luo et al., 2019). Synchronization of estrus is useful for insemination on the same day (Sen and Onder, 2016) and control the estrus behavior and lambing rate (Almadaly et al., 2016). Prostaglandin (PG) F2α injection was used for regressing corpus luteum (CL). Double PGF2α injections are commonly used for estrus synchronization in ewes (Hashemi and Safdarian, 2017). Prostaglandins are mainly administered intramuscularly (Simões et al., 2015). Double injection of prostaglandin, administered 11 days apart, resulted in higher estrus (Omontese et al., 2016).

The serum progesterone concentration on day 0 was 1.52 ± 0.33 that was lower than 2.1 ± 0.5 ng/mL, the basal concentration during estrus in goats (Socheh et al., 2019; Susilowati et al., 2020).

The progesterone concentration of Ettawa crossbred goat at day-21 was 7.08 ± 1.02 ng/mL that was lower than the progesterone concentration of Kacang goats on day-22 (15.45 ± 3.94 to 16.58 ± 2.85 ng/mL) (Susilowati et al., 2020). Meanwhile, progesterone concentrations on day-42 was in accordance with Devalal et al. (2018) who reported that progesterone concentrations at day 45 post-service of the goat was 6.27 ± 0.45 ng/mL. The progesterone concentrations at day-63 and day-84 were 12.01 ± 5.30 and 12.04 ± 0.30 ng/mL (p >0.05). Singh et al. (2019) reported that during mid-pregnancy (day-50 to day-100), progesterone concentrations were relatively stable in around 12-14 ng/mL. Progesterone concentration in ewes was significantly influenced by the number of corpora lutea found in the ovaries (Hennebold, 2018). The pattern of progesterone concentrations during pregnancy was increased; it is low from mating and increases noticeably by the 3rd week and remains high until the 19th week (Hussain, 2015). Progesterone concentration during pregnancy in single kid bearing does was lower than those in twin kid bearing does (Madan et al., 2020).

Pregnancy after artificial insemination is the best indicator of the reproductive potential of sires (Moura and Memili, 2016). Serum progesterone is indicative of pregnancy after an estrus cycle of insemination. Blood plasma concentrations >1 ng/mL between days 18–22 in parous goats indicate pregnancy. The accuracy for the diagnosis of pregnancy is 75-86%, while the accuracy for the diagnosis of non-pregnancy is 90-100%. The non-return to oestrus in a cycle is a very early pregnancy diagnosis after the mating (Karadaev, 2015). Islam et al. (2014) reported progesterone concentrations of 1.5 ng/mL, 6.6-9.3 ng/mL, and 14-15.5 ng/mL respectively on 25-30, 60-70, and 90-92 days of pregnancy. However, a high concentration of progesterone has not proven pregnancy because it could not differentiate between mid-cycle, true or false pregnancy (Devalal et al., 2018). On the other hand, the doe may not be diagnosed as pregnant for having low progesterone concentration (1ng/mL) around day-20 to day-24 after mating. Previous research in Kamori goats reported that pregnancy reliably could be diagnosed at day-21 after breeding using ELISA, and the concentration was 2.71 ± 1.06 ng/mL (Kunbhar et al., 2019).
There are some possible causes for the increased and decreased progesterone concentration, such as pseudopregnancy, nutrition during pregnancy, BCS, and the number of corpus luteum. Progesterone concentrations in pseudopregnancy decline over time, eventually resulting in the spontaneous evacuation of the uterus and similar to those observed during normal pregnancy (serum concentrations greater than one ng/mL) (Menzies, 2019). Nutrition during pregnancy also affects embryo survival and fetal programming of adult performance (Herring et al., 2018). Underfeeding is thought to act on progesterone delivery to the uterus, which is essential for developing the placenta and fetus (Cable et al., 2021).

Body Condition Scoring is an essential tool for livestock managers, feeding programs, reproduction, and welfare of the animals. The scoring was based on the amount of muscle and fat around the vertebrae. Scoring is performed in goats using a BCS ranging from 1.0 to 5.0 (Widiyono et al., 2020). Ideally, the BCS of goats was 2.56 ± 0.13 for a higher pregnancy rate (Nawito et al., 2015). BCS directly affects hypothalamic activity and GnRH secretion, but not pituitary sensitivity to GnRH (Sejian et al., 2010). These effects on reproductive performance are also mediated through changes in ovarian hormones or hypothalamic-pituitary sensitivity to ovarian hormones (Marques et al., 2018).

CONCLUSION

There was a two phase (biphasic) increase in progesterone concentrations of pregnant Ettawa crossbred does, from day-0 to day-21/day-42 and from day-21/day42 to day-63/day-84.

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REFERENCES

Almadaly E, Ashour M, El-Kon I, Heleil B, Fattouh E. 2016. Efficacy of various synchronization protocols on the estrus behavior, lambing rate, and prolificacy in Rahmani Egyptian Ewes during the non-breeding season. Asian J Anim Vet Adv. 11: 34-43.

Anderson ST, Bindon BM, Hillard MA, O’Shea T. 1998. Increased ovulation rate in Merino ewes immunized against small synthetic peptide fragments of the inhibin alfa sub unit. Reprod Fertil Dev. 10: 421-31.

Antanaitis R, Malašauskienė D, Televičius M, Juozaitienė V, Žilinskas H, Baumgartner W. 2020. Dynamic Changes in Progesterone Concentration in Cows’ Milk Determined by the At-Line Milk Analysis System Herd Navigator™. Sensors (Basel). 20: 5020.

Cable JK, Grider MH. 2021. Physiology, Progesterone. In: StatPearls. StatPearls Publishing, Treasure Island, Florida. [Updated 2021 May 9]. https://www.ncbi.nlm.nih.gov/books/NBK558960/

Dávila FS, González ASB, Barragán HB. 2017. Reproduction in Goats. In: Kukovics S. (Ed). Goat Science. InTech, Rijeka, Croatia. 87-105.

Devalal K, Joseph M, Kurien MO, Aravind A, Ajithkuma S. 2019. Serum progesterone and oestradiol profile in pseudopregnant and pregnant goats below 3 months post-service. J Vet Anim Sci. 50: 63-7.

Estill CT. 2015. Initiation of puberty in heifers. In: Hopper RM (ed). Bovine Reproduction. 1st Ed. John Wiley & Sons, Inc. Starkville, Mississippi. USA. 195-200.

Gan SD, Patel KR. 2013. Enzyme immunoassay and enzyme linked immunosorbent assay. J Invest Dermatol. 133: 1-3.

Hafez B, Hafez ESE. 2000. Reproductive cycles. In: Hafez B, Hafez ESE (Eds). Reproduction in farm animals. 7th Ed. Lippincott Williams & Wilkins, Philadelphia, USA. 55-67

Hashemi M, Safdarian M. 2017. Efficiency of different methods of estrus synchronization followed by fixed-time artificial
insemination in Persian downy does. Anim Reprod. 14: 413-7.

He J. 2013. Practical guide to ELISA development. In: Wild D (Ed) The Immunoassay Handbook. 4th Ed. Elsevier Science, Amsterdam, The Netherlands. 381-95.

Hennebold JD. 2018. Corpus Luteum. In: Spencer TE, Flaws JA (Eds). Encyclopedia of Reproduction, vol 2: Female Reproduction. 2nd Ed. Elsevier. 99-105.

Herring CM, Bazer FW, Johnson GA, Wu G. 2018. Impacts of maternal dietary protein intake on fetal survival, growth, and development. Exp Biol Med (Maywood). 243: 525-33.

Hussain SO. 2015. Serum progesterone analysis for monitoring estrus, pregnancy and parturition in Shami goats. AL-Qadisiya J Vet Med Sci. 14: 69-73.

Islam MM, Kizaki K, Takahashi T, Khanom JS, Debnath S, Khandelwal R, Morley JE, New M, Purnell J, Sahay R, Singer F, Stratakis CA, Trence DL, Wilson DP (Eds). Endotext. MDText.com, Inc. South Dartmouth, Massachusetts. [Updated 2018 Jun 19]. https://www.ncbi.nlm.nih.gov/books/NBK279070/

Menzies P. 2019. Pseudopregnancy in goats - an important cause of subfertility. Vet Rec. 184: 767-9.

Moura AA, Memeli E. 2016. Functional aspects of seminal plasma and sperm proteins and their potential as molecular markers of fertility. Anim Reprod 13: 191-9.

Nawito MF, Mahmoud KGM, Kandiel MMM, Ahmed YF, Sosa ASA. 2015. Effect of reproductive status on body condition score, progesterone concentration and trace minerals in sheep and goats reared in South Sinai, Egypt. Afr J Biotechnol. 14: 3001-5.

Nepal S, Subedi D, Kaphle K. 2019. Pregnancy Diagnosis with Progesterone ELISA Kit in Farm Animals, Its Accuracy and Application. Nepal Vet J. 36: 111-7.

Omontese BO, Rekwot PI, Ate IU, Ayo JO, Kwu MU, Rwuaan JS, Nwannennal AI, Mustapha RA, Bello AA. 2016. An update on oestrus synchronization of goats in Nigeria. Asian Pac J Reprod. 5: 96–101.

Sejian V, Maurya VP, Naqvi SMK, Kumar D, Joshi A. 2010. Effect of induced body condition score differences on physiological response, productive and reproductive performance of Malpura ewes kept in a hot, semi-arid environment. J Anim Physiol Anim Nutr (Berl). 94: 154-61.

Sen U, Onder H. 2016. The effect of estrus synchronization programs on parturition time and some reproductive characteristics of Saanen goats. J Appl Anim Res 44: 376-9.

Simões J. 2015. Recent advances on the synchronization of ovulation in goats, out
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of season, for more sustainable production. Asian Pac J Reprod. 4: 157-65.
Singh SP, Ramachandran N, Sharma N, Goel AK, Gururaj K, Kharche SD. 2019. Temporal changes in plasma profile of pregnancy-associated glycoprotein, progesterone and estrone sulfate associated with fetal number during early- and mid-pregnancy in goats. Anim Reprod Sci. 205:115-25.
Singh V, Rana RK, Singhal R. 2013. Analysis of repeated measurement data in the clinical trials. J Ayurveda Integr Med. 4: 77–81.
Siregar TN, Siregar IK, Armansyah T, Syafruddin, Sayuti A, Hamdani. 2013. Tampilan Reproduksi Kambing Lokal Hasil Induksi Superovulasi dengan Ekstrak Pituitary Sapi. J Vet 14: 91-8.
Socheh M, Saleh DM, Purbojo SW, Setyaningrum A. 2019. Concentration of Estrogen and Progesterone during Estrus and the 14th Day of Mating in the Javanese Thin-Tailed Ewes. The 1st Animal Science and Food Technology Conference (AnSTC) 2019. IOP Conf. Series: Earth and Environmental Science 372: 012021. IOP Publishing. 1-8.
Susilowati S, Triana IN, Sardjito T, Suprayogi TW, Wurlina W, Mustofa I. 2020. Effect of Simmental bull seminal plasma protein in egg yolk-citrate extender on Kacang buck semen fertility. Cryobiology 97: 20-7.
Susilowati S, Triana IN, Wurlina W, Arimbi A, Srianjo P, Mustofa I. 2019. Addition of L-arginine in skim milk extender maintains goat spermatozoa quality in chilled temperature for five days. Vet World 12: 1784-9.
Triagil AL, Ismudiono I, Setiawan B, Lestari TD, Samik A, Utomo B. 2020. Relationship between the value of the estrous detector measurement result and serum progesterone level in Ettawa crossbred goats after estrous synchronization. Ovozoa 9: 82-5.
Triswanto, Ngadiyono N, Sumadi. 2019. Reproductive performance of crossbreed goats Ettawah in kulon progo regency. Agrinova 2: 1-8.
Widiyono I, Sarmin S, Yanuartono Y. 2020. Influence of body condition score on the metabolic and reproductive status of adult female Kacang goats. J Appl Anim Res. 48: 201-6.
Winay A, Prihartini I, Ramadhan SW, Adhim ATF, Rico MJI. 2017. Linear Body Measurement of Indonesian Etawah Crossbred Goat [Capra aegagrus hircus (Linnaeus, 1758)] and Its Relationship with Milk Production Ability. Proceedings of the Pakistan Academy of Sciences: B. Life Environ Sci. 54: 301-9.