Progesterone level of normal cycling and repeat breeding Ongole grade cows

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Abstract. The present study was carried out to compare plasma progesterone level of normal cycling and repeat breeding Ongole grade cows. On the beginning of the luteal phase, blood samples were collected from 15 fertile and 15 repeat breeding Ongole grade cows. Progesterone hormone level was measured using progesterone hormone kit (DRG, Germany). The data were analyzed using independent sample t-test to determine the differences of progesterone level in repeated breeding and fertile cows. The finding of the study showed significant different progesterone level of between repeat breeding and fertile cows. Repeat breeding cows were found to have lower levels of progesterone level (1.17 ± 0.39 ng/ml) than that of fertile cows (3.07 ± 0.33 ng/ml). Finally, progesterone level of repeat breeding Ongole grade cows was lower than fertile cows. Low level of progesterone can have some effects on reproductive problems as repeat breeding.

1. Introduction
Reproduction process is closely related to the hormonal system and its mechanism, including the relationship between hypothalamic-pituitary hormones (gonadotropin-releasing hormone, follicle-stimulating hormone, and luteinizing hormone), ovarian hormones (estrogen, progesterone), and uterine hormone (prostaglandin) [1]. Ovarian hormones, estrogen, and progesterone have main role in reproduction. During pregnancies, progesterone, and estradiol have significant role on the growth and development of uterus and mammary glands. Low concentration of progesterone in luteal phase can cause embryonic demise and reduce pregnancy rate. This condition would lead to repeat breeding in female. Late formation corpus luteum (CL) either alone or in combination with a decrease in progesterone secretion during the luteal phase has been identified as one of the main causes of repeat breeding [2]. For that, repeat breeding can be defined as condition where a female breed failed to conceive after mated three times or more with a fertile male breed, without any abnormalities observed [3].

Since, repeat breeding can cause economic loss, reduce dairy production, and increase calving interval therefore, high rates of repeat breeding become problem cattle breeding and needs an immediate response. Accurate information regarding reproductive hormones in repeat breeding becomes a necessary information to observe. There were many studies on aspects of the reproduction
of Ongole grade cows, however progesterone profile on repeat breeding was very limited. For that, this study aimed to measure plasma progesterone level during luteal phase of repeat breeding cow and compare it to fertile cow. Moreover, from this study it could be the a way to use progesterone hormone level as marker in clinical diagnosis of repeat breeding.

2. Materials and methods

2.1 Materials
Thirty (30) heads of Ongole grade cows with body condition scores ranging from 2.0 to 3.0 were equally divided into repeat breeder and fertile groups cow [4]. The animals were maintained by smallholder farmers in several areas of Sleman District, Yogyakarta, Indonesia. The cows were fed with forage (parra grass, native field grass, and rice straw) twice a day: in the morning and evening, concentrates (wheat pollard and rice bran) in the afternoon and ad libitum of water.

2.2. Methods

2.2.1 Blood sampling. Blood samples were taken once in the early luteal phase during two oestrus cycles. Ten milliliters of blood were collected from the caudal vein of repeat breeder and fertile cows into anticoagulant-coated tubes. The tubes were transported in ice container to the laboratory followed with centrifuged at 2000 rpm for 20 minutes. Blood plasma then stored at -20°C for further analysis.

2.2.2. Progesterone analyses. Blood plasma samples were analyzed for hormones using a commercial enzyme-linked immunosorbent assay progesterone kit (DRG, Germany).

2.3. Data analysis. Progesterone levels were expressed as mean ± standard deviation and analyzed by using independent sample t-test to compare means between fertile and repeat breeder groups.

3. Results and discussion
The mean plasma concentration of progesterone in the luteal phase of fertile cows and repeat breeding cows were presented in Figure 1.

![Figure 1](image)

**Figure 1.** Progesterone level of fertile and repeat breeding Ongole grade cows in the early luteal phase (ng/ml)
The research showed that there was a significant difference in progesterone level between fertile and repeat breeding group (p<0.05). The level of progesterone on fertile cows was higher than that of the repeat breeding cows. The low level of progesterone during the luteal phase caused embryonic demise and pregnancy failure, leading to a repeat breeding.

Progesterone was produced by corpus luteum and crucial in maintaining pregnancies. Corpus luteum also played a major role in regulating the endometrial secretion, which was necessary to stimulate and mediate changes in conception growth and differentiation throughout the early pregnancy of ruminants [5]. Several studies had reported the relation between increase of systemic progesterone and acceleration of conception extension. The combination of in vivo and in vitro experiments found that progesterone had indirect effects toward the conceptus extension, and mediated through progesterone-induced effects in endometrial [6]. Progesterone was necessary to successful ovulation in the ovary and for multi-faceted oviduct in mammal’s reproduction [7]. The effect was mediated by the progesterone receptor (PGR), which was highly expressed in the ovary, specifically granulosa cells of preovulatory follicles in response to a surge in Luteinizing Hormone (LH) that occurred just before ovulation, and in the oviduct, especially epithelial cells and muscle cells. The low levels of progesterone in repeat breeding cows were considered to be related to the low level of lipids, given that progesterone was synthesized from lipids. The low level of lipids contributed to the low energy status and reduced the presence of precursors for synthesizing all steroids in the body, such as adrenal cortex hormones, sex hormones, vitamin D, and bile acids [8].

In addition, lipids, especially cholesterol were very important for the biosynthesis of androstenedione, progesterone, and estrogen by granulosa cells under the influence of LH. This hormone stimulates the production of DHEAS by granulosa cells; pregnenolone was then converted to androstenedione by theca cells. Androstenedione returned to granulosa cells and was converted to estrone and estradiol. The lack of lipids prolonged the anestrus period, resulted in repeated breeding and decreased the amount of ovulation [9, 10]. Other researchers reported that total cholesterol levels were lower in repeat breeding cows compared to fertile cows [11, 12].

4. Conclusion
Progesterone level of repeat breeding Ongole grade cows was lower than fertile cows. Low level of progesterone can have some effects on reproductive problems as repeat breeding.

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