Characteristics of libido and testosterone concentrations of Bali polled and horned Bulls

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Abstract. Bali cattle as one of Indonesia's native cattle have horns basically, both male and female. However, in its development, Bali cattle without horns have been found and known as polled. Bali polled is cattle whose horns do not grow naturally. The objective of this study was to determine the characteristics of libido and the concentration of testosterone in Bali polled and horned bulls. The samples were 8 individuals (4 polled and 4 horned) with an age between 3-8 years. Libido characteristics were measured by recording the time the bull first fondled the cow until ejaculate and analyzing the testosterone concentration using the enzyme-linked immunosorbent assay (ELISA) method. The collected data were analyzed by an independent sample T-Test. The results of this study indicated that the time required for a polled bull from fondling the female to ejaculation was longer (P<0.05) compared to horned (7.45±2.51 vs 2.42±0.60 minutes). While the testosterone concentration was not significantly different (P>0.05) 6.94±2.43 ng/mL vs 3.74±1.34 ng/mL although showed a higher tendency for the horned bull at 3.2 ng/mL. In conclusion, the polled bull took a longer time from fondling the female to ejaculation compared to horned, while the concentration of the hormone testosterone was equal. However, libido characteristics have a positive correlation with the testosterone concentration with a correlation coefficient of 0.805.

1. Introduction
Bali cattle as one of Indonesia's native cattle have horns basically, both male and female. However, in its development, have been found Bali cattle without horns and known as polled. Bali polled is cattle whose horns do not grow naturally. Bali polled cattle have advantages in terms of maintenance management. One of them is the ease of maintenance which is associated with the productivity of the meat, such as reducing the risk of injury to livestock caused by horned. It can prevent damage to the skin and bruising of the carcass. Therefore, the selection of Bali-pollled cattle is very important, especially in modern livestock management. Homozygous breed generation can reduce costs and time for dehorning and relieve stress for livestock [1]. However, one of the shortcomings of the Bali polled bull is to have a low libido during the semen collecting time. Limited information related to the development of Bali polled cattle, especially the characteristics of libido and testosterone concentration in bulls, is the basis for this study.

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Bulls must have reproductive requirements include high libido and the ability to serve/mate well. The libido or sexual ability of a bull is described by the desire to mate, which means that the male does not experience erection problems, trouble riding the female/teaser, or difficulty inserting the penis and ejaculating. The male libido is a very important aspect of reproductive function and needs to be considered. Even though the macroscopic and microscopic tests of spermatozoa are good but not followed by adequate libido or the desire to mate, the male fertility rate will decrease [2]. Further described by Rachmawati et al. [3], the level of libido and the process of spermatogenesis is controlled by the hormone testosterone. Testosterone has an important role in the reproductive process and it can improve the reproductive system in male animals. The testosterone regulates male characteristics, helps the development of the testes from the abdominal cavity into the scrotum during the fetal period, the development of primary and secondary sexual characteristics, and spermatogenesis [4]. The testosterone helps create optimal conditions for the process of spermatogenesis and spermatozoa transport into the male genital tract [5].

2. Materials and methods

2.1. Materials
Eight Bali bulls aged between 3-8 years which consists of 4 polled and 4 horned were used in this research.

2.2. Methods

2.2.1. Libido observation. Observation of libido was carried out using Bali cow as a teaser. Libido was measured by recording the time the bull begins to fondle the cow until ejaculation occurs [3,6].

2.2.2. Blood sampling. Blood sampling was taken from the jugular vein. First, the vein was pressed at the base of the neck until it is swollen (visible) then the area around which will be pierced cleaned using alcoholic cotton, a vacutainer needle was inserted at 30° upward into the vein. Blood collected using a vacuum tube with ± 3 mL. The blood plasma was separated by centrifuge technique at a speed of 2500 rpm for 5 minutes until it separated into 3 layers, plasma, buffy coat, and erythrocytes. Furthermore, blood plasma is collected in a microtube tube and stored at a temperature of -20 °C (until analyzed). Testosterone levels were measured using the enzyme-linked immunosorbent assay (ELISA) method [7].

2.2.3. Measurement of testosterone concentration. Testosterone levels were measured using the enzyme-linked immunosorbent assay (ELISA) method that was carried out by direct ELISA kit DRG Testosterone ELISA EIA-1559 [7].

2.2.4. Data analysis. The collected data will be analyzed using the Independent sample t-test.

3. Results and discussion

3.1. Characteristic of libido of Bali polled and horned bulls
The requirements for a male to become a superior bull include having high libido, serving/mating ability, serving/mating capability, and having high genetic transmitted ability [8]. Libido is a desire to mate caused by an increase of testosterone in bulls [9]. Libido is a very important aspect of reproductive function that needs to be considered to increase the male fertility rate. Libido or mating desire of males can be tested through the frequency of erections, mates, and ejaculates normally in a certain time unit [2]. The results of the observations of libido characteristics in Bali polled and horned bulls are presented in figure 1.
Figure 1 shows a significant difference (P<0.05) of the characteristics of libido that were measured by looking at the time from fondling the female-teaser to the ejaculation of the Bali polled bull that is compared to horned. The results showed that polled libido was lower than horned. This was evidenced by the time required for the polled to be longer than the horned starting from teasing the female-teaser to the ejaculation (7.45±2.51 vs 2.42±0.60 min). The results of this study differ from those that have reported that the libido of the Bali bull average 4.7 minutes and 4.5 minutes [10,11]. Furthermore, Saputra [12] stated that Bali bulls have more aggressive characteristics, differences in libido that depend on each breed of cattle. High and low libido are factors that can be used to indicate the quality of bulls and fertility rate.

Libido is influenced by several factors including genetics and breeds, male to female ratio, type of male and mating method, social relations between males, age of males, sexual experience of males, climate/thermal environment, nutrition, various causes of stress-related to relocation, differences in genotype and phenotype, topography and group distribution, and temperament in the new environment [13]. The reaction of the male to mount the female is related to the libido in the male after being stimulated by the female. Libido is related to reaction time, which is the time (seconds) it takes for males to approach the teaser to false mounting [14-17]. The high libido of bulls is due not only to genes but also to good maintenance management. Toelihere [18] stated that the factors that influence libido can come from outside or within the livestock. The ability of a bull to mount a female is influenced by the age factor. The older the bull results in the malfunctioning of the limbs caused by dislocation/fracture (cracks) and osteoarthritis of the hind legs or spine.

3.2. Testosterone concentration of Bali polled and horned bulls

The hormone that affects male reproduction is the androgen hormone, testosterone. Bull fertility can be seen from high or low levels of testosterone in the blood. The role of testosterone can be seen in the process of spermatozoa formation, stimulating the growth of accessory glands and stimulating male character, and also plays a role in causing libido. Libido is a common event in male cattle, mounting a female for sexual activity. This condition explains the reproductive performance of bulls and it can be seen whether a male is suitable or not used as a superior seed or a breeder male for future breeding purposes. The results of measuring the testosterone concentration in polled and horned bulls are presented in figure 2.

![Figure 1. Characteristic of libido of Bali polled and horned bulls. Different superscripts show significant differences (P<0.05)](image)
Figure 2 showed that the mean concentrations of testosterone in polled and horned bulls were not different, 6.94±2.43 vs 3.74±1.34 ng/mL. However, it showed a higher tendency for horned of 3.20 ng/mL, compared to polled bulls. The results of this study indicated that the testosterone concentration of polled is lower than previously reported by Ihsan [19] that the concentration of the testosterone that can cause libido is 6.70±0.20 ng/mL. Nevertheless, polled bulls still showed libido even though it took a longer time, 7.45±2.51 min, starting from fondling the female/teaser until the occurrence of ejaculation. Furthermore, Rasyid et al. [20] reported that the concentration of testosterone in PO bulls was 7.0 ± 0.8 ng/mL, and Simmental was 6.1±0.4 ng/mL.

Steroid concentration per blood volume is very different depending on the individual and the conditions/time in the same livestock. Hormone levels in the blood depend not only on secretion and metabolism but also on the age of the cattle, season, sampling frequency, sampling conditions, and sensitivity and specifications of the assay. Steroid hormones in males are androgens that stimulate the growth and function of secondary reproductive organs, the development of specific sex characters that can be used as a basis for hormone testing [19].

Physiologically, testosterone is released fluctuatively due to the negative feedback mechanism caused by stimulation from outside. Luteinizing hormone (LH) is a hormone produced by the anterior pituitary which functions to stimulate interstitial cells (Leydig cells) to secrete the hormone testosterone [21]. The desire to mate or libido greatly determines the concentration of testosterone, while male sexual behavior depends on genetic, environmental, nutritional, hormonal, sensory acuity, ages, and sex experience [22]. Furthermore, Astuti et al. [23] reported that testosterone secretion fluctuates diurnal and has functions to induce libido. The fluctuation in the hormone testosterone occurs due to the feedback mechanism of the LH from the pituitary and GnRH from the hypothalamus. The testes within 24 hours will release testosterone pulses of 12 to 24 times so that it will create a daily pattern with certain fluctuations.

3.3. Correlation between characteristics of libido and testosterone concentrations

The results of this study indicate that there is a positive correlation between libido characteristics and the concentration of the hormone testosterone in polled and horned bulls with a correlation coefficient of 0.805. This showed that the high concentration of testosterone in the blood plasma can expedite the time required for the bull, starting from fondling the female/teaser to ejaculating. The same thing has been previously reported by Rachmawati et al. [3] that there is a positive correlation between the concentrations of testosterone on the level of libido, the higher the concentration of testosterone, the higher the level of libido. It is further explained that libido is influenced by the concentration of testosterone [24,25].

The correlation between libido characteristics and the concentration of hormone testosterone shown in the results of this study is in line with Rachmadi [26] that the testosterone hormone derived from the
testes plays a role in stimulating growth and maintaining the continuity of the genital glands function to produce semen plasma at the ejaculation process, stimulating the male desire to mate (libido). The same thing was explained by Brinkmann [27] that the hormone testosterone also plays a role in the male’s ability to erect and ejaculate. The mechanism of testosterone in reaching the target cell and then carrying out its role in influencing male sexual activity is through passive diffusion.

4. Conclusion
Characteristics of libido in Bali polled bull required a longer time starting from fondling the cow/teaser to ejaculate compared to horned. Meanwhile, the testosterone concentration of polled was not different with horned, although it showed a tendency to be higher 3.20 ng/mL in the horned bull. However, there is a positive correlation between testosterone concentration and libido characteristics with a correlation coefficient of 0.805.

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