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MORPHOLOGY AND REPRODUCTIVITY PROFILING OF MALE SENDURO GOATS BASED ON AGE DIFFERENCES

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Supporting Information

ABSTRACT: Senduro goats, a local breed of meat and dairy goats from Indonesia, are recognized for their significance in improving goat breeding and preserving valuable genetic resources. However, limited information exists regarding the reproductive physiology of Senduro goats, which poses challenges to the development of breeding programs and the preservation of genetic resources. This study aimed to investigate the morphological and reproductive profiles of male Senduro goats at different ages, focusing on morphological characteristics, mating behavior, and sperm quality. Morphological characteristics are assessed through body length measurements, while mating behavior serves as an indicator of reproductive behavior. Macroscopic evaluations of sperm quality include assessments of color, viscosity, pH, and volume, while microscopic examinations encompass motility (mass and individual), viability, and spermatozoa membrane integrity. The results showed morphological similarities between juvenile and adult samples, with their testicular size being the only significant difference. Based on macroscopic and microscopic examinations, no significant differences were found between groups. From the results it was concluded that there were no distinct differences in morphological characteristics, mating behavior, and sperm quality between male Senduro goats in the juvenile and adult stages.

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INTRODUCTION

Goats are highly adaptable livestock species thriving in tropical regions (Knight and Garcia, 1997). In general term, among the goat breeds, Senduro goats hold a significant position as a local Indonesian breed (Hariyono and Endrawati, 2023). This particular local breed emerged from successful crossbreeding between *Jamunapari* goats from Etawah, India and Indonesian Bean goats. As part of preserving the indigenous genetic material and mitigating the risk of extinction, it is crucial to continually increase the population of Senduro goats. Several methods, including natural and artificial mating, can be used to bolster Senduro goat populations.

For successful breeding programs, it is imperative to gather information on the morphological and behavioral characteristics of male Senduro goats (Birhanie et al., 2019). Understanding reproductive behavior not only contributes to effective breeding but also aids in the evaluation of animal welfare (Zamiri et al., 2010). Additionally, a comprehensive understanding of the morphological features and behavioral patterns of male goats is crucial for breeding and cultivation programs, as these factors can be influenced by environmental conditions and the age of the animal (Dias et al., 2017). Behavior and morphology are often associated with the age of goats (Ambali et al., 2018). Studies on Sokoto Goats have shown differences in morphological characteristics and sperm quality with increasing age, where older goats tend to exhibit higher rates of sperm abnormalities (Akpa, et al., 2013). Evaluation of morphological traits is equally essential, as it shows a correlation between morphological characteristics and age in Boer goats (Abd-Allah et al., 2019). The age factor also plays a role in the production and reproduction capabilities of Senduro goats (Khandoker et al., 2018).

The reproductive performance of goats is influenced by age, nutrition, and seasonal factors (Đuričić et al., 2021; Ali et al., 2022). This kind of performance is closely associated with the apparent morphological characteristics. A previous study fconducted by Soutu et al. (2017) showed that testicular measurements, including length, width, and volume, are correlated with the age of male goats. These parameters can serve as a criterion for selecting exceptional young males. Age and nutritional status have also been found to influence the reproductive characteristics of sheep (Ptáček et al.,

2017). However, there is a paucity of reviews on the morphological and reproductive characteristics of Senduro goats, an indigenous breed in Indonesia.

Therefore, this study aims to observe male Senduro goats of various ages to investigate the relationship between age and morphological and reproductive characteristics.

MATERIALS AND METHODS

Ethical regulation

The implementation of this research previously received research ethics, namely 070-KEP-UB-2023.

Determination of animals testing groups

The study was conducted at the Senduro Goat Breeding Center in the Senduro district of the Lumajang Regency of East Java, Indonesia. Data were collected from a total of 18 male Senduro goats, which were divided into two groups, including juveniles (ages 8 months to 1.6 years) and adults (aged 2 to 4.25 years). Each group consisted of nine males.

Morphological observations

Morphological observations of the goats included height, body length, body circumference, face shape, body shape, leg shape, presence or absence of horns, tail shape, hair color, hair distribution, presence or absence of beard, beard length, testicular length, testicular width, testicular size, and reproductive behavior. The testicular size was determined by measuring the length and diameter of the testicle using tape (Varghese et al., 2019). Meanwhile, testicle length was measured from the caudal part to the attachment point at the end of the scrotum. The circumference of the scrotum was evaluated to determine the diameter of the testicle. Length-measuring instruments were used for the morphological observations.

Observation of reproductive behavior

Reproductive behavior was observed based on libido behavior during mating or when collecting sperm using artificial vaginal techniques. The observed reproductive behaviors included the time of erection, frequency of mounting for ejaculation, and ejaculation time.

Fresh semen collection

After stimulation with a doe, sperm was collected using an artificial vagina. A total of 18 sperm samples were successfully collected from each goat and immediately placed in a water bath at 37°C. The collected sperm samples were then subjected to macroscopic and microscopic examinations. Macroscopic observations included assessing volume, color, consistency, pH, and spermatozoa concentration. Meanwhile, the microscopic analysis involved evaluating spermatozoa motility, viability, and integrity of sperm membranes.

Macroscopic observation

About two different studies independently conducted observations of fresh sperm volume, color, and consistency. pH was determined by using pH paper (Sigma-Aldrich), whereas spermatozoa concentration was determined using a counting chamber method. To perform the analysis, semen was diluted with Natrium Chloride (NaCl) solution, added to a haemocytometer, and observed under a light microscope at 400x magnification (Olympus).

Microscopic observation

Observations of spermatozoa motility

Both mass and individual motility of spermatozoa were assessed (Hayati et al., 2019). For mass motility evaluation, 20 µL of fresh sperm was added to a glass slide and observed under a light microscope (Olympus) at 40x magnification. Individual motility observations were performed by placing 10 µL of sperm on a glass object, followed by examination under a microscope at 400x magnification.

Observations of Spermatozoa Viability

Viability observations were conducted using Eosin-negrosin staining (Merck, Germany). About 10 microliters (μ L) of sperm was placed on a glass object, followed by the addition of 20 μ L of Eosin-nigrosin dye. The mixture was homogenized and allowed to rest for 30 seconds (Kamal et al., 2022). Subsequently, the sample-dye mixture was added to the glass slide by gently pushing it through the edge of the glass using another glass object at a 45° angle. The sample was allowed to dry at room temperature. After drying, observations were made under a light microscope at 400x magnification. The viability of spermatozoa was then determined by counting 200 cells in one field of view. Dead spermatozoa were stained purple, while live spermatozoa remained colorless (Srivastava et al., 2017; Ducha et al., 2020).

Observations of Spermatozoa Integrity

Spermatozoa membrane integrity was evaluated using the Hypo Osmotic Swallen Test (HOST). The HOST solution consisted of 0.9 g of fructose (Merck, Germany) and 0.49 g of sodium citrate fructose (Merck, Germany) dissolved in 100 ml of distilled water (Khan et al., 2017). About 100 microliters (µI) of fresh sperm was added to 1 ml of the HOST solution and incubated for 60 minutes at 37°C. To assess sperm swelling, 15 µl of the well-mixed sample was placed on a

warmed slide (37°C), covered with a coverslip, and observed under a light microscope at 40x magnification. Swollen sperm cells with intact membranes were considered normal and indicative of fertilization potential. About 300 sperm were counted per slide, and the percentage of swollen sperm was calculated (Jamali et al., 2019).

Data analysis

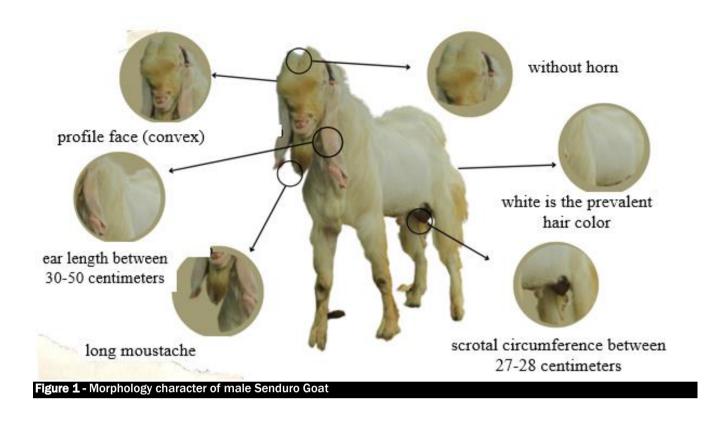
Most of the obtained data were analyzed descriptively, including calculating averages, sums, and standard deviations and analyzing qualitative data. Quantitative data from macroscopic and microscopic observations of fresh sperm were further analyzed using the Mann-Whitney U Test (IBM SPSS 23) to determine statistical differences between the juvenile and adult age groups.

RESULTS AND DISCUSSION

Male Senduro goats generally exhibit a straight, convex face shape, white hair, a beard, and horns (in some males). Long hair is predominantly observed on the head, neck, chest, front and hind legs, beard, buttocks, and a straight and short tail (Table 1 and Figure 1).

These results align with the previous observations made by Ciptadi et al. (2019), who reported similar morphological characteristics in male Senduro goats, including a straight body with a convex face shape, white hair, a beard, and the presence of horns in most males. Additionally, dominant long hair was noted on the head, neck, chest, front and hind legs, beard, buttocks, and a straight and short tail (Ciptadi et al., 2019). Senduro goats share morphological similarities with the Etawah Peranakan goats. As an Etawah crossbreed, Senduro goats exhibit a convex face, long ears, black and white hair, horns, and thicker and longer hair on the neck and legs (Susilorini et al., 2020). These similarities can be attributed to the fact that Senduro goats are the result of crossbreeding between Etawah Peranakan goats, Kacang goats, and Jawarandu goats (Susilorini et al., 2020). Based on SNI 2018 data, Senduro goats can be characterized by white fur, a convex face, downward-hanging ears, the presence or absence of horns, a long beard in males, and a lack of beard in females. Furthermore, males exhibit longer body hair on the neck and hips, with long body hairs being more prominent and short tails. Male Senduro goats also display a larger body size compared to their female counterparts (Figure 2). Importantly, there were no significant morphological differences observed between adolescent and adult age groups (Table 2). These results align with the study conducted by Abd-Allah et al. (2019), who reported no variations in the morphological characteristics of Boer goats across different ages (Salman et al., 2019). This is also in line with the research of Sesay et al. (2022), that there are no significant differences in the morphology of West African Dwarf goats at various ages. No significant difference was observed in the reproductive behavior between adults and juvenile Senduro goats (Table 3). The analysis using the Mann-Whitney test also showed no significant differences between young and juvenile goats, indicating that age does not affect reproductive behavior. These results are supported by the study conducted by Suyadi et al. (2021), which found no variation in reproductive behavior among male Boer goats aged between 11 and 25 months (Suyadi et al., 2021). Furthermore, these results align with the observation of Hafizuddin et al. (2021), which showed no variation in libixdo levels among Etawah crossbreeds, both in juveniles and adults, with an average libido level of 3 times (Hafizuddin et al., 2021). Observation of the reproductive profile and its relation to the age of livestock, is very important to determine the breeding strategy and management of goats in an effort to increase the expected population (Parvathi et al., 2020).

Table 1 - Morphology male Senduro goats between juvenile and adult			
Parameters	Juvenile (8 Months - 1.6 Years)	Adults (2 - 4.25 Years)	
Shape of face	Dominantly convex	Dominantly convex	
The shape of the body	Dominantly proportionally straight	Dominantly proportionally straight	
Shape of leg	Dominantly proportionally	Dominantly proportionally	
The presence of a horn	The majority have a horn, but some have not	The majority do not have horns; some have	
Shape of tail	Dominantly straight and short	Dominantly straight and short	
Hair color	Dominantly white	Dominantly white	
Hair distribution	Dominantly long in the head, neck, chest, front and back leg, beard, and buttocks	Dominantly long in the head, neck, chest, front and back leg, beard, and buttocks	



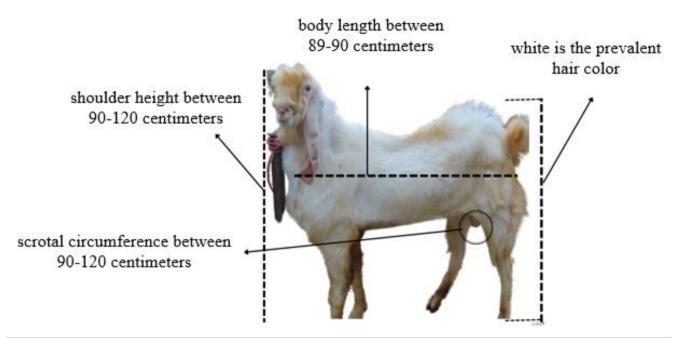


Figure 2 - Body posture of male Senduro goat

Table 2 - Statistical morphology between juvenile and adult Senduro goat

Parameters	Juvenile (8 Months - 1.6 Years)	Adults (2 - 4.25 Years)	Significant
Height (kg)	86.77±2.89 ^{bc}	97.22±2.43 ^{bc}	0.903
Body length (cm)	86±5.07 ^b	95.44±3.32 ^b	0.044
Length of beard (cm)	7.11±2.43 ^{bc}	15±2.42 ^{bc}	0.661
Circumference of the scrotum (cm)	26.11±0.99 ^{ab}	27.88±0.63 ^{ab}	0.052
Length of the scrotum (cm)	14.22±1.13 ª	17.11±0.26ª	0.023
Chest size (cm)	90.22±2.28 ^{bc}	98.33±2.35 ^{bc}	0.521

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Table 3 - Reproductive behaviour of adult and juvenile Senduro goats			
Parameters	Juvenile (8 Months - 1.6 Years)	Adults (2-4.25 Years)	Significance level
Erection Time (Seconds)	98.88±25.46	172.44±65.58	0.198
Number of false mountings: the frequency with which males do Mounting for ejaculation (Times)	2.55±0.44	2.8±0.38	0.495
Ejaculation time: the length of time it takes to ejaculate (times)	4.44±0.86	3.33±1.66	0.108

Table 4 - Macroscopic observations of juvenile and adult Senduro goats			
Parameters	Juvenile (8 Months- 1.6 Years)	Adults (2-4.25 Years)	Significance level
рН	6.78±0.10	6.8±0.11	0.876
Volume (uL)	1000±172.40	911±107.29	0.109
Colour	Milky white	Milky white	-
Consistency	Thick	Thick	-
Smell	Fishy Odour	Fishy Odour	-

Table 5 - Microscopic observations of juvenile and adult Senduro goats

Parameters	Juvenile (8 Months- 1.6 Years)	Adults (2-4.25 Years)	Significance level
Mass motility	++++	++++	-
Individual motility	70.941±1.34 ^a	72.246±2.07 ^a	0.593
Viability	74.141±0.97ª	76.480±2.583ª	0.198
Membrane integrity	75.153±0.81ª	77.341±2.84ª	0.146
Concentration	1.26 x 10 ⁸ a	1.27 x 10 ^{8 a}	0.197

Macroscopic and microscopic observations showed no significant differences. The macroscopic test involved evaluating volume, aroma, viscosity, color, and pH. Juvenile Senduro goats exhibited a pH value of 6.78±0.10, sperm volume of 1000±172.40 uL, milky white semen color, thick consistency, and a characteristic fishy odor of sperm (Table 4). Similarly, adult male goats displayed a pH value of 6.8±0.11 and sperm volume of 911±107.29 uL, along with milky white semen color, thick consistency, and a characteristic fishy odor of sperm. The macroscopic examination of spermatozoa showed no significant differences. Hafizuddin et al. (2021) reported that sperm quality in Anglo-Nubian goats, Etawah goats, and Ampera goats was not significantly affected by age or social interaction (Hafizuddin et al., 2021). In addition to macroscopic observations, microscopic evaluations of the sperm of Senduro goats were performed.

Subsequently, the microscopic evaluation was conducted to assess sperm quality based on parameters such as motility (movement), viability, and membrane integrity. The results showed that fresh semen from both juvenile and adult Senduro goats exhibited mass motility of ++++, indicating highly progressive and concentrated movement of spermatozoa colonies (Table 5). The average percentage of individual motility ± standard deviation (SD) in juvenile groups was 70.941±1.34, while adult goats had an average individual motility of 72.246±2.07. The analysis results showed that male age had no significant effect on the motility of fresh semen in both juvenile and adult groups. However, the average motility of individual spermatozoa was higher in adult goats. These values fell within the normal range, as reported by Syarifuddin et al. (2022), who indicated that the motility of fresh semen of Etawah ranged from 70-75% (Syarifuddin et al., 2022). The best viability of fresh semen was observed in the male group aged 2 - 4.25 years, with an average viability of 74.141±0.97, while juvenile goats had an average viability of 76.480±2.583. Similarly, the best spermatozoa membrane integrity was observed in the male group aged 2 - 4.25 years, with an average of 77.341±2.84, whereas juvenile goats exhibited membrane integrity of 75.153±0.81. The concentration of spermatozoa in Senduro goats showed no significant difference between the juvenile and adult groups, with an average concentration of 1.26 x 10⁸ in juvenile goats and 1.27 x 10⁸ in adult goats. These results are consistent with the study of Souto et al., 2017, which also reported no significant difference in spermatozoa concentrations (Souto et al., 2017). Although the sperm quality was evident, it was found to be higher at a more mature age. This is in line with the results of Lacuesta et al. (2015), who observed an increase in spermatozoa quality with age until adulthood (Lacuesta et al., 2015). Furthermore, Nishimura et al. (2000) showed an improvement in sperm quality as Tokara goats transitioned from juvenile to adult age.

CONCLUSION

In conclusion, the results of this study showed that there were no significant differences in terms of morphology, reproductive behavior, and sperm quality between adult and juvenile male Senduro goats. However, the data suggested that the age range of 2 to 3.5 years was optimal for producing spermatozoa with good quality. Based on the results of

this research, it can be used as a guideline in selecting male Senduro goats that are ready to mate based on morphological and reproductive characteristics.

DECLARATIONS

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Data availability

The datasets used and/or analysed during the current study available from the corresponding author on reasonable request.

Authors' contribution

All authors played a role in the research and writing of this article. N. Ducha, N.N. Yusof, N. Samsulrizal, E.R. Damiman played a role in designing the research. The research carried out in the field and laboratory was carried out by N. Ducha, L. Lisdiana, G. Trimulyono, F.I. Muhaimin, A. Fudhaili, G.R.A. Pramesti., J.D. Rahayu. Data analysis was carried out by F.I.Muhaimin, A. Fudhaili, N.N.Yusof, N.Samsulrizal, E.R.Dasiman, G.R.A. Article writing was carried out by all authors.

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Consent to publish

The authors agree to the publication of this manuscript.

Competing interests

The authors declare no competing interest

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