



## Effects of Bull Age on the Fresh and Frozen Semen Quality of Aceh Cattle

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**Abstract** | Age is one of the essential factors determining the reproductive potency of the bull because it is related to the growth and development of livestock organs, including their reproductive organs. The purpose of the present study was to evaluate the characteristics of fresh and frozen semen quality of Aceh cattle at different ages. Three Aceh bulls were involved in this study. The semen was collected from each bull aged 6 to 10 years and the fresh and frozen semen quality was evaluated directly. The results showed that the bull age significantly affects semen volume, pH, sperm concentration, total sperm, total motile sperm, and frozen semen production of Aceh cattle ( $p < 0.05$ ). However, it was found that there was no significant change in the sperm motility, before freezing, post-thawing, and during recovery across age ( $p > 0.05$ ). Semen volume, total sperm, total motile sperm, and frozen semen production increased in bulls from 6 to 7 years of age and then gradually reduced from 7 to 10 years of age. It could be concluded that the optimum semen productivity of Aceh cattle is when they are 7 years old.

**Keywords** | Artificial insemination, Frozen semen production, Indonesian local cattle, Reproductive performance, Total sperm output

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## INTRODUCTION

The increase in the Indonesian population has resulted in increased demand for food, including beef. This is also supported by the increasing knowledge and public awareness of the consumption of balanced nutrition. However, the increase in beef consumption is not proportional to the increase in livestock production. According to the CADIS (2020), the total beef production in Indonesia is 402.22 thousand tons, but the demand for beef reaches 623.64 thousand tons. Therefore, efforts are needed to increase beef production so that it can meet the national demand.

One type of local Indonesian cattle that can be used as a

meat producer is Aceh cattle. Aceh cattle is one of the local Indonesian cattle that endemic in Aceh Province (MARI, 2011). Aceh cattle have resistance to a harsh environment such as feed and water crises, parasitic diseases, and high temperatures. In addition, Aceh cattle have good productivity, produce 49-51% carcasses, and produce meat that has a smoother and denser tissue (MARI, 2011). However, the development of Aceh cattle is still constrained by the traditional breeding and maintenance system (Sofyan et al., 2020), so there is a need for technological innovations that can be used to support their production.

Artificial insemination is one of the reproductive technologies that is carried out to accelerate the increase in the cattle population effectively and efficiently. Semen quali-

ty can highly influence the success of artificial insemination (Isnaini et al., 2021a). Before going through further processing, the semen should be evaluated macroscopically and microscopically. Macroscopic semen evaluation includes volume, color, pH, and consistency (Iswati et al., 2018). Microscopic semen evaluation includes mass motility, individual motility, spermatozoa concentration, before freezing motility, and post thawing motility (Susilawati et al., 2018). Semen that can be processed into liquid and frozen semen should have good quality and comes from superior bulls.

Superior bulls should have good body and reproductive performances. Factors that determine semen quality include age, season, temperature, libido, feed, semen collection method, and collection frequency (Isnaini et al., 2021b). Age is related to the growth and development of livestock organs including their reproductive organs, which is associated with the reproductive potency of the bull (Snoj et al., 2013). The purpose of this study was to evaluate the fresh and frozen semen quality of Aceh cattle at different ages. This research is very important to be carried out to determine the age of the male that produces the best quality semen in order to improve reproductive efficiency.

## MATERIALS AND METHODS

### ETHICAL APPROVAL

The procedures in this study were performed based on the institutional animal care of the Faculty of Animal Science, Universitas Brawijaya, Indonesia. This study was also as part of routine semen collection in the Lembang Artificial Insemination Center, which was held under SNI ISO 9001:2015.

### LOCATION

This experiment was performed at the Lembang Artificial Insemination Center, Bandung, Indonesia. The location is at 6°S and 107°E with an altitude of 1,241 m above sea level. The ambient temperature ranged from 22.9 to 24.3°C, while relative humidity ranged from 67.2 to 81.9%.

### ANIMALS AND MANAGEMENT

Three Aceh bulls from Lembang Artificial Insemination Center (Bandung, Indonesia) were involved in this study. The bulls were kept in an open-sided housing. The daily feed for each bull consisted of 40 kg of fresh forage (elephant grass), 1 kg of hay (African star grass), 3 kg of concentrate supplement, and 0.3 kg of a mung bean sprout. The concentrate had 16% crude protein and 65% total digestible nutrient. The bulls had free access to drinking water during experimental periods.

## SEMEN COLLECTION AND EVALUATION

The semen was collected from each bull during the age of six to 10 years using an artificial vagina. The fresh and frozen semen quality was evaluated with the following details. The evaluation of semen volume was done using a scaled tube apparatus (Kusumawati et al., 2017). Semen pH was evaluated using litmus paper. The evaluation of sperm concentration was done using a spectrophotometer (Minitube, Germany) (Isnaini et al., 2021c). To evaluate the total sperm, semen volume was multiplied by sperm concentration (Isnaini et al., 2021c). The evaluation of sperm motility was done under a phase-contrast microscope (Olympus, Japan) (Ratnawati et al., 2018). To evaluate the total motile sperm, total sperm was multiplied by sperm motility (Isnaini et al., 2021c). Evaluation of before freezing and post-thawing sperm motilities were done under a light microscope (Olympus, Japan) (Ratnawati et al., 2018). Semen with approved quality were filled into a mini straw (0.25 ml).

### STATISTICAL ANALYSIS

Data of fresh and frozen semen production were analyzed using analysis of variance. Semen quality traits with statistically significant differences ( $p < 0.05$ ) were further evaluated using Duncan post-hoc test. All data analyses were conducted using the statistical software of IBM SPSS Statistics 22.

## RESULTS AND DISCUSSION

Table 1 presented the fresh semen quality of Aceh cattle at different ages. The results showed that the bull age significantly affected semen volume and semen pH ( $p < 0.05$ ). Semen volume increased from 6 to 7 years old and then gradually reduced from 7 to 10 years old. The semen pH was increased along with the advancing age. Moreover, sperm concentration, total sperm, and total motile sperm of Aceh cattle were also affected by the bull age ( $p < 0.05$ ). The sperm concentration from 7 years old onward was lower than at 6 years old. In terms of total sperm and total motile sperm, these variables increased from 6 to 7 years old and then gradually reduced from 7 to 10 years old. However, it was found that sperm motility was equal across cattle of different ages ( $p > 0.05$ ). This study clearly indicated that the peak semen volume of Aceh cattle was reached at 7 years old. In agreement with the current study, Snoj et al. (2013) observed that the peak semen volume of Limousin cattle was at 7 years old and subsequently reduced after this point. Similarly, in a study by Argiris et al. (2018), it was also found that the semen volume of Holstein bulls increased up to 7 years old and subsequently decreased thereafter. However, Nugraha et al. (2019) found that the semen volume of Bali bull was increased from 6 to 10 years old. However, in a study conducted by Briliansyah et al. (2020),

**Table 1:** Fresh semen quality of Aceh cattle at varying ages in Lembang Artificial Insemination Center

Bull age	SV	SpH	SC	TS	SM	TMS
6 years	4.98 <sup>b</sup>	6.57 <sup>a</sup>	1.16 <sup>c</sup>	5.69 <sup>cd</sup>	68.68	3.93 <sup>cd</sup>
7 years	6.27 <sup>c</sup>	6.65 <sup>bc</sup>	1.03 <sup>b</sup>	6.46 <sup>d</sup>	69.00	4.47 <sup>d</sup>
8 years	4.60 <sup>b</sup>	6.63 <sup>b</sup>	1.04 <sup>b</sup>	4.92 <sup>bc</sup>	67.68	3.42 <sup>bc</sup>
9 years	4.73 <sup>b</sup>	6.69 <sup>c</sup>	0.88 <sup>a</sup>	4.11 <sup>ab</sup>	68.60	2.83 <sup>ab</sup>
10 years	3.54 <sup>a</sup>	6.70 <sup>c</sup>	1.07 <sup>bc</sup>	3.73 <sup>a</sup>	67.44	2.53 <sup>a</sup>
SEM	0.107	0.008	0.016	0.140	0.333	0.100
<i>p</i> value	0.001	0.001	0.001	0.001	0.557	0.001

SV: Semen volume (mL), SpH: Semen pH, SC: Sperm concentration (x 10<sup>9</sup> sperm/mL), TS: Total sperm (x 10<sup>9</sup> sperm), SM: Sperm motility (%), TMS: Total motile sperm (x 10<sup>9</sup> sperm)

<sup>abcd</sup>different superscripts within a column indicate significant differences (*p* < 0.05)

**Table 2:** Frozen semen quality of Aceh cattle at varying age in Lembang Artificial Insemination Center

Bull age	BFSM	PTSM	RRSM	FSP
6 years	51.06	43.30	61.52	234 <sup>c</sup>
7 years	51.57	44.84	63.80	282 <sup>d</sup>
8 years	50.56	43.21	61.53	206 <sup>bc</sup>
9 years	53.80	42.64	60.91	163 <sup>ab</sup>
10 years	49.70	42.78	61.11	136 <sup>a</sup>
SEM	0.542	0.313	0.447	7.684
<i>p</i> value	0.279	0.237	0.309	0.001

BFSM: Before freezing sperm motility (%), PTSM: Post-thawing sperm motility (%), RRSM: Recovery rate of sperm motility (%), FSP: Frozen semen production (straw/ejaculate)

<sup>abcd</sup>different superscripts within a column indicate significant differences (*p* < 0.05)

it was found that the semen volume of Madura bulls was increased up to 12 years old. These discrepancies probably indicated that the peak semen volume may be varied across the breed.

The peak semen volume at 7 years old is probably related to the development of testis and accessory glands as well as increased activity of the hypothalamic-pituitary-testicular axis (Murphy et al., 2018). The higher semen volume at this age point was then subsequently followed by the higher total sperm. In line with this finding, Snoj et al. (2013) also reported that the higher semen volume of Limousin cattle subsequently provided higher total sperm. The increase in total sperm consequently could increase the total motile sperm. On the other hand, the reduction of semen volume at an older age is probably because of the onset of senile changes (Bhakat et al., 2011), which may reduce the capacity of semen volume secretion and followed by the reduction of other semen parameters. In a study by Baharun et al. (2021), it was found that the testosterone concentration of bulls aged 8-10 years was markedly lower, compared to those at 4-5 and 6-7 years. The reduction of testosterone concentration will also be followed by the reduction of semen volume (Al-Murshidi, 2018; Kandiel and El Khawagah, 2018; Hafizuddin et al., 2020).

Table 2 showed the frozen semen quality of Aceh cattle at varying ages. The results showed that the before freezing, post-thawing, and recovery rate of sperm motility of Aceh cattle were similar across ages (*p* > 0.05). However, the frozen semen production of Aceh cattle varied among ages (*p* < 0.05). From 6 to 7 years old, frozen semen production of Aceh cattle was increased and then gradually reduced from 7 to 10 years old. The higher frozen semen production at 7 years old was probably related to the higher total sperm at this age.

## CONCLUSIONS

The results of the current study showed that the fresh and frozen semen quality of Aceh cattle increased from 6 to 7 years of age and then gradually reduced from 7 to 10 years of age. Therefore, it could be concluded that the optimum semen productivity of Aceh cattle is reached at 7 years old. It is suggested that more samples from cattle of different ages can provide precise results.

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The authors declare that they have no competing interests.

## AUTHORS CONTRIBUTION

All authors equally contributed and approved the manuscript.

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