EVALUATION OF SEMEN QUALITY AND ESTIMATION OF SEMEN REPEATABILITY VALUE OF MADURA BULLS AT LEMBANG ARTIFICIAL INSEMINATION CENTER

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Abstract
Madura bulls are one of the local bulls in Indonesia whose semen is used for artificial insemination. This study aims to evaluate the quality of semen and the repeatability value of semen from Madura bulls at the Lembang Artificial Insemination Center. A total of 1,402 semen collection data from three Madura bulls from 2014-2020 were used for analysis. The data were analyzed descriptively to evaluate semen quality consisting of semen volume, sperm motility and sperm concentration. Repeatability value was estimated using analysis of variance and intra-class correlation. The results showed that the average volume, motility, and concentration were 5.32 ± 1.50 ml; 68.10 ± 7.07 %; and $1,044.27 \pm 277.63$ million/ml. The semen volume repeatability value was high, motility was low, and concentration was medium.
Keywords: semen quality, repeatability, Madura bull

EVALUASI KUALITAS SEMEN DAN PENDUGAAN NILAI RIPITABILITAS SEMEN PADA PEJANTAN SAPI MADURA DI BALAI INSEMINASI BUATAN LEMBANG

Abstrak
Sapi Madura merupakan salah satu sapi pejantan lokal di Indonesia yang digunakan semennya untuk Inseminasi Buatan. Penelitian ini bertujuan untuk mengevaluasi kualitas semen dan nilai ripitabilitas semen dari sapi Madura di Balai Inseminasi Buatan Lembang. Sebanyak 1.402 data penampungan semen dari tiga ekor pejantan sapi Madura dari tahun 2014-2020 digunakan untuk analisis. Data dianalisis secara deskriptif untuk mengevaluasi kualitas semen yang terdiri dari volume semen, motilitas sperma, dan konsentrasi sperma. Nilai ripitabilitas diduga menggunakan analisis ragam dan korelasi intra-kelas. Hasil penelitian menunjukkan rata-rata volume, motilitas, dan konsentrasi adalah 5,32±1,50 ml; 68,10±7,07 %; dan 1.044,27±277,63 juta/ml. Nilai ripitabilitas volume semen termasuk kategori tinggi, motilitas termasuk rendah, dan konsentrasi termasuk sedang.
Kata kunci: kualitas semen, ripitabilias, sapi Madura

INTRODUCTION

Artificial Insemination (AI) bulls are bulls that have gone through selection and meet certain requirements to be used as frozen semen producing livestock. AI bulls must have the ability to produce good quality semen. Semen quality can be assessed by observing the components of semen assessment, including volume, motility, and concentration.

The good quality of fresh semen is one of the determinants of the quality of frozen semen that will be produced. The fresh semen produced must meet the requirements to be further processed into frozen semen which still meets the requirements during post-thawing

(Butler, et al., 2021). These quality requirements in Indonesia are regulated by the National Standardization Agency as stated in SNI 4869-1:2017 concerning Frozen Semen Part 1: Bull.

Apart from prioritizing quality aspects, semen production must also maintain its sustainability. In other words, AI bulls must be able to continuously maintain their ability to produce high-quality semen.

One of the livestock breeds used as AI bull is the Madura bull. Madura bulls are a type of local Indonesian bull known for their ability to adapt to environmental conditions, such as resistance to weather and disease (Widi et al.,

2014; Hartatik et al., 2010). This cattle breed is designated as a national cattle breed through Decree of the Minister of Agriculture Number 3735/Kpts/HK.040/11/2010 concerning the establishment of the Madura cattle breed in 2010. Madura cattle are a cross between *Bos javanicus* and *Bos indicus*, a cross between Bali cattle and Zebu cattle from India in Madura (Sutarno & Setiawan, 2015) which weigh up to 226-338 kg when they are 2-3 years old (Hartatik, et al., 2010).

Madura cattle, apart from being used for their power strength and meat, are related to the traditions of the Madura people because they are used in the *karapan* (bull race contest) and *sonok* (cow beauty contest due to its good body appearance) traditions. The genetic purity of Madura bulls can continue to be maintained through this tradition. Good quality of Madura bulls' semen is also important to increase the number of Madura bull population.

The ability of Madura bulls to repeat good quality semen production can be estimated by calculating the repeatability value. Repeatability comes from the words repeat and ability, and has the meaning of 'the ability to repeat'. Repeatability is an important number used to determine the accuracy of measurements and the strength of the phenotype (Nakagawa & Schielzeth, 2010). Repeatability value in quantitative genetics is used to explain the proportion of variation in a trait observed among individuals and within individuals. The variance component from repeated observations of an animal trait can be broken down into within animal variation (within groups or classes) and among animal variation (among groups or classes) (Wolak, et al., 2012).

Semen repeatability can be estimated using a certain calculation formula based on production records. Calculation of repeatability in statistics refers to the Intra-class Correlation Coefficient (ICC). This calculation is used to determine correlation within one data class. The ICC concept was first introduced by R. A. Fisher (Bobak, et al., 2018; Liljequist, et al., 2019). The repeatability value resulting from this calculation can be used as a consideration in selecting bulls based on their reproductive ability.

Evaluating the semen quality of Madura bulls used for AI and estimating the repeatability of semen quality is important to determine the accuracy of the implemented selection program. Such evaluations can also increase and streamline the utilization of bulls in the frozen semen supply program for Madura bull at insemination centers, such as the Lembang Artificial Insemination Center.

MATERIAL AND METHOD

The material used in this study was 1,402 semen collection data from 3 Madura bulls at Lembang Artificial Insemination Center. Data were obtained from 7 years (January 2014-December 2020) of semen quality record. Semen data analysis began when the bull were 2 and 3.5 years old. Semen collection was carried out 1 to 2 times a week depending on the temporary environment that influenced the collection. The average number of ejaculates per bull was 467 data.

The quality of semen observed in this study was in the form of semen volume, namely the amount of semen obtained from each semen collection; sperm motility (%) namely the percentage of sperm that move forward; and sperm concentration (million/ml), namely the number of spermatozoa in one unit volume.

The data analysis for evaluating semen quality was carried out using descriptive statistics, while for estimating repeatability values used one-way analysis of variance and intra-class correlation. Calculation of descriptive statistics and analysis of variance used IBM SPSS Statistics 26 software.

The mathematical model used is as follows (Indrijani, 2018):

$$Y_{ij} = \mu + \alpha_{ij} + \varepsilon_{ij}$$

Remarks:

Y_{ij} = semen quality value of the ith bull from the jth collection

μ = average semen quality

α_{ij} = influence of the ith bull

ε_{ij} = Uncontrolled environmental influence on the ith bull of the jth collection

In the meantime, estimation of the repeatability value (r) used the intra-class correlation formula as follows:

$$r = \frac{\sigma_b^2}{\sigma_b^2 + \sigma_w^2}$$

Remarks:

r = repeatability

σ_b^2 = variance among bulls

σ_w^2 = variance within bull

The standard error calculation to determine the accuracy of data calculations is as follows:

$$Se_{(r)} = \sqrt{\frac{2(1-r)^2[1+(k-1)r]}{k(k-1)(n-1)}}$$

Remarks:
$Se_{(r)}$ = standard error of repeatability
r = repeatability
n = number of bulls
k = number of production records observed; In this case, because the number of records for each bull is not the same, the formula

$$k = \frac{1}{n-1}\left(m - \frac{\sum m_i^2}{m}\right)$$

was used in which m is the total amount of data analyzed from all bulls, and m_i is the total amount of data from each individual bull.

RESULTS AND DISCUSSION

Semen Evaluation

The semen evaluation results of Madura bulls are presented in Table 1. The semen volume obtained was 1.75-8.50 ml with an average of 5.32 ± 1.50 ml which is higher than three different studies on Madura bulls, namely 3.00 ± 0.38 ml (Romadhoni, et al., 2014); 4.86 ± 1.46 ml (Rahmawati, et al., 2015); and 4.57 ± 0.61 ml (Yekti, et al., 2018). The bulls used in this study had high semen production in quantity. Larger semen volumes, in relation to frozen semen, can increase the number of cows that can be inseminated.

The average sperm motility of Madura bulls in this study was $68.10 \pm 7.07\%$. This value is lower than those reported in two other studies on Madura bull: 73.00 ± 2.58 % (Romadhoni, et al., 2014) and 70.00 ± 0.00 % (Yekti, et al., 2018). However, this result is higher than 65.03 ± 11.39 % reported by Rahmawati, et al. (2015). Fresh semen must have a minimum motility of 70% to be processed into frozen semen (BSN, 2017). The semen motility data in this study had a minimum of 40% and a maximum of 80%. In relation to frozen semen, the higher the percentage of sperm motility, the more sperm can swim forward to fertilize the ovum.

The average concentration of Madura bulls' sperm in this study was $1,044.27 \pm 277.63$ million/ml. This value is smaller than three different studies on Madura bulls, namely $1,814.60 \pm 297.40$ million/mL (Romadhoni, et al., 2014); $1,084.02 \pm 392.13$ million/mL (Rahmawati, et al., 2015); and $1,068.57 \pm 70.81$ million/mL (Yekti, et al., 2018). However, the average value in the results of this study is still within the normal range for bull sperm concentration, namely 800-2,000 million/ml (Garner & Hafez, 2000). Higher concentration of sperm, in relation to frozen semen, may increase the number of doses that will be produced.

Semen Repeatability

Repeatability is a measure of the accuracy of measurement and strength of the phenotype from repeated observations. The estimated repeatability value indicates the repeatability of an observed trait. In this study, estimating the repeatability value of Madura bulls aimed to assess the ability of their semen at the Lembang Artificial Insemination Center to consistently produce semen with good quantity and quality. This value can also help describe the relationship between semen production in the initial months or years and the following production over their lifespan.

The estimated repeatability values of Madura bulls' semen from the three parameters studied are presented in Table 2. The estimated repeatability values for volume, motility, and concentration in this study are lower than the study of Prihatin, et al (2017) which also calculated repeatability values for the same breed of Madura bulls at the Singosari Artificial Insemination Center (BBIB).

Table 1. Semen quality of Madura bulls at Lembang Artificial Insemination Center

Semen Quality	Number of Ejaculates	Minimum Value	Maximum Value	Average	Standard Deviation
Volume (ml)	1,402	1.75	8.50	5.32	1.50
Motility (%)	1,402	40.00	80.00	68.10	7.07
Concentration (million/ml)	1,402	255.00	1,800.00	1,044.27	277.63

Table 2. Variance components and repeatability estimates for semen quality of Madura bulls at Lembang Artificial Insemination Center

Semen Quality	Number of Ejaculates	σ_b^2	σ_w^2	Repeatability Estimates	Standard Error
Volume	1,402	0.74	1.75	0.30	0.02
Motility	1,402	4.20	47.27	0.08	0.01
Concentration	1,402	19,908.53	63,949.33	0.24	0.02

The estimated value of volume repeatability of Madura bull semen in this study was 0.30; still lower than the study results of Prihatin, et al. (2017) in the same breed, namely 0.376. Likewise, when compared with volume repeatability values of other breed. Sitanggang (2018) reported results of higher volume repeatability for PO bulls, namely 0.480. Similar results were also reported by Supraptono, et al. (2018) and Boujenane & Boussaq (2013) in FH bulls, namely 0.423 and 0.411 respectively.

The estimated value of Madura bull sperm motility repeatability in this study was 0.08; still higher than the results of Sitanggang's (2018) study on PO bulls, namely 0.010. However, the results of this study are still lower when compared with the results of study in the same breeds, namely 0.567 (Prihatin, et al., 2017), as well as in other breed, in this case FH, at 0.296 (Supraptono, et al., 2018) and 0.257 (Boujenane & Boussaq, 2013).

The estimated repeatability value of Madura bull sperm concentration in this study was 0.24; still higher than the results of study by Boujenane & Boussaq (2013) on FH bulls, namely 0.175. However, the results of this study are still lower when compared to the results of study on the same breed, namely 0.445 (Prihatin, et al., 2017), as well as other study on FH breeds, namely 0.505 (Supraptono, et al., 2018) and study on PO bulls at 0.420 (Sitanggang, 2018).

The results of repeatability calculations for volume, motility, and concentration of semen in this study were 0.30; 0.08; and 0.24. This shows that the Madura bull breed at Lembang Artificial Insemination Center tends to have the same performance for semen volume due to its high repeatability estimates. The estimated repeatability values of Madura bulls' semen production are respectively classified into the high, low, and medium categories. The higher repeatability value indicates that the diversity of semen production is strongly influenced by genetic factors and permanent environmental variations. On the other hand, the lower the estimated repeatability value, the more temporary environmental variations play a significant role in the production of Madura bulls' semen.

The temporary environment in this study can be in the form of maintenance management at the Lembang Artificial Insemination Center including feeding, implementation of collection including collecting officers and collection intervals, and the influence of seasons. With regard to the influence of seasons, Bhakat et al., (2009) affirms this by suggesting that the dry season, in general, has a bad influence on overall semen quality. Dry season affects the reproductive process by causing reduced feed intake and inhibiting the release or response to important reproductive hormones in spermatogenesis. The collection officer's knowledge of reproductive physiology can also have an influence because limited knowledge can limit the results of good semen collection with normal morphology (Schenk, 2018).

Differences in the estimated value of Madura bulls repeatability at the Lembang Artificial Insemination Center with other studies on the same breed or different breeds of bull are common. They are thought to be caused by differences in the methods used, differences in environmental conditions, and maintenance conditions and management.

Based on the results of this study, Lembang Artificial Insemination Center is expected to continue to maintain the use of Madura bulls with good genetic so that the high repeatability value of semen volume can be maintained in the future. On the other hand, Lembang Artificial Insemination Center is expected to minimize the influence of temporary environmental factors by improving good maintenance management, which can improve the quality of semen motility and concentration.

CONCLUSION

Semen quality of Madura bulls at the Lembang Artificial Insemination Center is in the normal and good category, with a volume of 5.32 ± 1.50 ml; motility 68.10 ± 7.07 %; and concentration 1,044.27 ± 277.63 million/ml. The estimated semen volume repeatability value is high (0.30 ± 0.02), motility is low (0.08 ± 0.01), and concentration is medium (0.24 ± 0.02).

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