The real potential of semen production of Bali Bull: over year observation at Singosari National Artificial Insemination Center (SNAIC), Singosari-Indonesia

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Abstract. Increased reproduction is closely related to increased bull productivity. The high productivity of these bulls will increase the amount of straw. The productivity capability of Bali bull is essential in producing straw. High productivity will result in high straw production as well. So the research aims to determine the productivity of Bali bull at SNAIC in the last year. 31 Bali bulls aged 4 – 11 years were used in this study at SNAIC. Total 1.273 ejaculations collection from these during one year. The parameters used in this study include the number of successful holding of each bull, total productivity, and estimation of straw production in one year. From 31 Bali Bulls, the ranking was grouped into two based on values of sperm productivity. The two groups were analyzed using an unpaired T-test. The results showed that the pH, volume, individual motility, concentration, and abnormality were significantly different ($P < 0.05$) between the high and low groups. The percentage of successful shelters, total TSM, and the number of straws produced in one year in the high productivity group were higher than the low productivity group. High productivity groups are the best with high productivity potential and potential as a source of semen.

1 Introduction

Bali cattle (*Bos sondaicus*) is one of the local cattle breeds with the highest population among other local cattle in Indonesia. Bali cattle are resistant to parasites and tropical environments in Indonesia and have better reproductive efficiency than other local cattle [1]. Bali cattle have been established through the Decree of the Minister of Agriculture No. 325/Kpts/OT.140/1/2010, as native Indonesian cattle and have an essential role in the national meat supply [2].

Superior bulls have been selected based on their lineage, production, and reproductive capabilities [3]. Good semen must be above the minimum standard of fresh semen to be processed into frozen semen. The minimum motility value is 70% [4], while the maximum

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abnormality is 20%, the volume ranges from 7–10 ml with a spermatozoa concentration of 1000–1500 x 10^6/ml [5, 6]. Bull fertility is influenced by age, race, body condition, feed, ejaculation ability, and environment [7].

Efforts to develop and preserve Bali bull from all aspects need to be carried out sustainably. One crucial aspect of doing is to improve the quality of reproduction. Increased reproduction is closely related to spermatozoa fertility and increased bull productivity. Reproduction is an essential step in the management system that impacts the economic aspect [8]. The high productivity of these bulls will increase the amount of frozen semen production. The government regulation of the minister of agriculture No. 10/Permentan/PK.210/3/2016 in section 4, namely the provision of frozen semen for domestically produced ruminants, must be 60% from native and/or local ruminants. This is an excellent opportunity for Bali cattle if they produce higher productivity [9].

On the other hand, the quality of Bali cattle semen varies highly. For example, the motility value of Bali cattle in several studies includes 62.07% ; 70.00%; 80.00%; 89.62% [10 - 13].

The productivity capability of Bali cattle is essential in producing frozen semen. Productivity is closely related to the value of volume, individual motility, and concentration of spermatozoa. Higher productivity will result in high straw production as well. So the research aims to determine the productivity of Bali cattle at BBIB Singosari in the last year.

2 Materials and methods

2.1 Animals

The study was carried out at Singosari National Artificial Insemination Center (SNAIC), Singosari-Indonesia (SNI.ISO 90001:2015). A total of 31 Bali bulls aged 4–11 years were used in this study. Total 1.273 ejaculations collection of these during one year. The bulls were reared under similar management based on the standard procedure in SNAIC.

2.2 Semen Collection and Evaluation

Semen collection was conducted once a week every morning. Semen was collected using an artificial vagina as a Standard Operating Procedure (SOP) of the SNAIC. The artificial vagina type is Double-Walled type. Semen quality data were determined in laboratories through direct observation after semen collection. The microscopic evaluations were individual motility, sperm concentration, and viability. The macroscopic evaluations were volume, pH, and color.

The volume of semen (ml) is determined directly after the semen is stored on the scale of the tube used to hold the semen [5]. Individual motility (%) was observed with a microscope with 400x magnification at a constant temperature using a cover glass, then determined the proportion (percentage) of progressively moving spermatozoa. The sperm concentration was observed by using a spectrophotometer [14]. Sperm viability used the eosin nigrosine staining method. The head of living spermatozoa will remain transparent and the dead spermatozoa will be red in color [15].

2.3 Data Classification

The total motile spermatozoa from each of Bali cattle for one year were calculated. The Total Spermatozoa Motil (TSM) value obtained becomes a reference for determining the rank of bull. Based on the productivity of Bali bulls in a year is obtained were divided into two groups. The top three bulls became the high group, and the lowest three bulls became the low group.

2.4 Statistical Analysis

From 31 Bali Bulls, the ranking was grouped into two based on values of sperm productivity. The two groups were analyzed using an unpaired T-test and were processed by using SPSS 24. Data were presented with the mean ± standard deviation (SD). Bull productivity was assessed from the Total Spermatozoa Motile using the following formula:

$$Total\ Spermatozoa\ Motile = volume \times concentration \times individual\ motility \quad (1)$$

Standard Operational Procedures (SOP) at SNAIC in a dose of 1 straw or 0.25 ml total spermatozoa concentration are $25x10^6$. So, the estimated number of frozen semen straws produced in one year is calculated by the formula:

$$Number\ of\ straws = Total\ Spermatozoa\ Motile\ /\ 25 \times 10^6 \quad (2)$$

3 Results and discussion

Macroscopic examination (Table 1) showed that the pH and semen volume were significantly different ($P < 0.05$) between the high and low groups. The highest pH was in the low group, while the high volume was in the high group. The color of the semen of the high group was milk-white, while the low group was clear white. The range of semen volume ranged from 4.07-8.79 ml. Suyadi *et al.* [16] stated that the semen volume in Bali bulls was constant from 4.7 to 4.9 ml/ejaculate. Semen volume increases with age [14]. This is because increasing age will cause the scrotal circumference to grow so that the volume of semen increases.

Table 1. Characteristics of Bali bull semen in the two productivity groups

Parameter	Productivity Group	
	High	**Low**
N ejaculate	141	67
Color	Milk White	Clear White
pH	$6,49 \pm 0,40^a$	$6,62 \pm 0,22^b$
Semen Volume (mL)	$8,79 \pm 1,68^b$	$4,07 \pm 1,15^a$
Individual Motility (%)	$82,32 \pm 6,94^b$	$79,17 \pm 9,15^a$
Sperm Concentration (10^6 / mL)	$1320,21 \pm 292,95^b$	$757,49 \pm 369,54^a$
Sperm Abnormality (%)	$4,40 \pm 2,31^a$	$10,37 \pm 6,38^b$

Different superscripts within line indicate significant differences (P<0,05)

The results (Table 1) showed that microscopic examination (individual motility and concentration) of the high group was significantly ($P < 0.05$) higher than that of the low group. Meanwhile, the abnormality of the high group was lower than that of the low group. Individual motility ranged from 79.17% - 82.32%, sperm concentration per ejaculate was between 757.49 $x10^6$ - 1320.21 $x10^6$, and sperm abnormalities were between 4.40% - 10.37%.

The average individual motility in this study was higher than that of Nugraha *et al.*[14] at 62.5% and Nugraha *et al.* [17] at 65 %, while a suitable bull for breeding must have an individual motility value of at least 70% (SNI 4869-1:2017). Bali bulls in this study were included in the category of good semen quality, which was more than 60% - 75% for sperm motility [5]. The sperm concentration in this study was lower than that in PO cattle [18], which was 2250 $x10^6$ / mL and higher than Madura bull that is 797.526 $x10^6$ / mL [19]. The semen of the Bali bull in this study is included in the criteria for good quality semen because the requirements for good semen are sperm concentrations of more than 500 million/ml [5]. At the same time, the abnormality is higher than Suyadi [16], which is 2.99%. According to Ax *et al.* [5], good spermatozoa abnormality is less than 20%.

Table 2. Number of semen successes that were successfully accommodated in two different TSM groups

Productivity Group	Bull Code	Age (year)	Total Shelter (ejaculation) in 1 Year	Total Shelter (ejaculation) Succeed in 1 Year	
			(times)	(times)	(%)
High	Bull 1	10	47	47	100,00
	Bull 2	10	47	47	100,00
	Bull 3	4	47	47	100,00
Low	Bull 29	4	47	18	38,30
	Bull 30	6	47	42	89,36
	Bull 31	10	47	7	14,89

Table 2 shows that all bulls in the high group in one year were successfully accommodated 47 times out of a total of 47 shelters (ejaculation) carried out. While in the low group, no bulls were successfully accommodated 47 times in one year. Bull 29 as many as 18 times in a total of 47 shelters (ejaculation) a year. Bull 30 as many as 42 successful shelters (ejaculation) in one year. Bull 31 only seven successful shelters (ejaculation) in one year. Percentage of total shelter (ejaculation) succeed in one years in bull 29, bull 30 and bull 31 were 38,30; 89,36; 14,89, respectively.

The results of this study, bull 1, bull 2, and bull 3, which are the high productivity group, are included in the category of superior bulls because the percentage of successfully accommodated reaches 100%. This study shows that there is a wide variation in Bali bull at SNAIC-Singosari. This reflects the genetic diversity of the Bali bull, which still has the opportunity to be improved. [20] stated that the application of genetic standards needs to be considered to maximize the potential and maximum and sustainable sperm production.

Table 3. The potential productivity of Bali bulls and the estimated number of straws produced in one year

Productivity Group	Bull Code	Age (year)	N ejaculate	Total TSM in 1 Year (10^6)	∑ Straw generated in 1 Year (straw)
High	Bull 1	10	47	501.803	20.072
	Bull 2	10	47	427.069	17.083
	Bull 3	4	47	408.973	16.359
Low	Bull 29	4	18	67.688	2.708
	Bull 30	6	42	67.251	2.690
	Bull 31	10	7	37.213	1.489

The potential for frozen semen production per ejaculation has a reasonably high range between the high and low groups. The potential for frozen semen production in the high group (Bull 1, Bull 2, and Bull 3) in one year were 20,072 straws, 17,083 straws, and 16,359 straws per ejaculate, respectively (Table 3). Meanwhile, in the low group (Bull 29, Bull 30, and Bull 31), the potential for frozen semen production was 2,708 straws, 2,690 straws, and 1,489 straws.

Bali bull productivity is closely related to the amount of straw produced. Potential productivity is the ability of each bull to produce straw or frozen semen. Productivity in this study is calculated for one year. The difference in straw production between the high and low productivity groups was caused by the number of successful collections in one year. In the low group, the moderate success per individual is only 22.3 times, while in the high group, the average success per individual reaches 100% or 47 times.

The results showed that the straw produced by each bull in the high group for one year was higher than Ditjennak [21]; which is, the minimum local bull each individual produced was 7,500 straws annually. The research results are also higher than those of other Bali cattle studies Indriastuti *et al.* [13], with 11,843.17 straws per year. This shows that Bali cattle at BBIB Singosari have the potential to produce high straw. The potential for frozen semen production is influenced by volume, concentration, and the number of motile sperm [21, 23].

4 Conclusion

Based on the number of successful shelters (ejaculation), the total TSM, and the number of straws produced in one year, it can be concluded that the two groups of high and low productivity in Bali bulls were different. Bull 1, Bull 2, and Bull 3 at SNAIC-Singosari are the best with high productivity potential and potential as a source of semen.

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