**Physical performances and semen characteristics of bulls at Central Cattle Breeding & Dairy farm, Savar, Dhaka, Bangladesh.**



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**Physical performances and semen characteristics of bulls at Central Cattle Breeding & Dairy Farm,Savar,Dhaka, Bangladesh.**



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 **List of contents**

|  |  |  |
| --- | --- | --- |
| **Chapters** | **Topics** | **Page No.** |
|  | Abstract | I |
| 1 | Introduction | **1-2** |
| 2 | Materials and Methods | **3-5** |
| 3 | Results | **6-7** |
| 4 | Discussion | **8-9** |
|  5 | Conclusion  | **10** |
|  | References | **11-14** |
|  | Acknowledgement | **15** |
|  | Biography | **16** |

**LIST OF THE TABLES**

|  |  |  |
| --- | --- | --- |
| Sl.No. | Table Title | Page No. |
| 1. | **Grading of semen according to motility.** | **05** |
| 2. | **Results of the clinical examination and libido test of bulls.** | **06** |
| 3. | **Results of the evaluation of fresh semen.** |  **07** |

**ABSTRACT**

The present investigation was aimed at evaluating the bulls and their semen from production to insemination in a routine artificial insemination (AI) programme in Bangladesh. Five crossbred bulls were examined for breeding soundness and their semen was preserved and evaluated with respect to sperm motility. Semen was sampled immediately after collection, pre-diluted, cooling down to +40°C, and storage at -196°C in Central Cattle Breeding Station (CCBS).The percentage of motile spermatozoa varied from 60±10 to 68±3 depending on the occasions when evaluation was made.However pulse rate, body condition scores and scrotal circumference of different bulls varied from 54.3 to 64.3/min, 3.1 to 3.8 (1-5 Scale) and 36.7 to 40.0 cm respectively.This variation may be occurred due to feeding,season and other factors.On the other hand the ejaculate volume of the bulls varies from 4.6 to 9.2 and highest volume found in one bull whose body condition score is 3.8 and body weight is 712kg . Highest sperm motility found in 68 and 60 is the lowest sperm motility. The sperm motility dropped due to dilution, chilling , freezing and storing. It could therefore be inferred that freezing of semen should be undertaken with special care to maintain optimum semen quality.

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| **Key words:** Artificial insemination, breeding soundness, cattle, frozen semen evaluation |

**CHAPTER-1**

**Introduction**

 Artificial insemination (AI) is the single most important technique used for the genetic improvement of cattle. This is possible because a few highly selected bulls produce enough spermatozoa to inseminate thousands of cows per year. The goal of the AI field services is to maximize the number of viable offspring per breeding animal per unit time. This can be achieved by inseminating cows with sufficient progressively motile spermatozoa from a given ejaculate without reducing their fertilizing capacity. Thus, the small number of frozen-thawed spermatozoa in each insemination dose must be of very high quality to ensure acceptable pregnancy rates (Brinsko and Varner 1993).

A thorough examination of bulls for breeding soundness is a prerequisite for investigating their fertility (Spitzer and Hopkins 1997). Although the outcome of a breeding soundness examination for a bull depends to some extends on the clinical reproductive knowledge and experience of the veterinarian concerned, the test was found good enough to identify diseases and/ or problems which often result in poor quality semen in respect to both freezability and fertilizing capacity (Spitzer et al 1988; Johnson et al1995; Lewis et al1996; Bhuiyan et al1997).

It is important to collect as many ejaculates as possible within a certain period without compromising the quality of semen for a routine AI practice. In case of bulls, sperm motility is a fairly reliable indication of the viability of fresh and frozen semen (Saacke and White 1972; Grahman et al1980). The fertilizing ability of spermatozoa, depends not only on the initial quality of semen, but also on the subsequent laboratory processes that end up with deposition of semen in the genital tract of a cow. The processes of semen dilutions, chilling, freezing, storage, transportation and thawing for insemination invariably result in some reduction of the viability and fertilizing capacity of spermatozoa.

Thus the fertilizing capability of spermatozoa in the AI dose is determined not only by the inherent quality of the material produced by the bull but also by man's interaction with the product (Saacke 1983; Serres et al 1997). Moreover, the process of transportation from one tank to another can warm - up the semen straws if is not accomplished quickly and accurately (Hafez 1993). Maintaining optimum nitrogen level in the container is important during prolonged storage and transportation of semen (Sherman 1990; Quintin et al 1997). The aim of the present investigations was to characterize the physical, sexual and semen characteristics of crossbred bulls (Friesian ´ Sahiwal and Friesian ´ local) and to determine the occasions when the quality of frozen semen deteriorate in a routinely running AI field service.

**Objectives:**

 **1.**To ensure successful calving.

 **2**.To obtain knowledge about the motility of bull semen.

**CHAPTER-2**

**Materials and Methods**

**Sample collection**

**2.1. Study site:**

The trial was carried out on 5 breeding bull at the Central Cattle Breeding Station and Dairy Farm, Savar, Dhaka. The farm was situated adjacent to the Dhaka Aricha highway and 30 km northwest side from Dhaka city. The farm was surrounded by plain land with plenty of green vegetation. The highest and lowest ambient temperature of the experimental area was recorded as 37-380C and 15-210C, respectively. Average humidity was 76.6%.

 **2.2. Collection of semen :**

The semen was collected by artificial vagina . Prior to collection of semen, all parts of artificial vagina (AV) set were cleaned, sterilized and assembled. The inner liner was put into the cylinder and both ends of inner liner were reflected over the cylinder forming water like space between them.The cone along with vial was slipped over one of the ends of the cylinder and then tightened with rubber band. Two third of the outer jacket of vagina was filled with warm water.

 The temperature inside the artificial vagina was 1100-1150F. An air screw was used for blowing air between two layers to create desired pressure. Required amount of lubricant was applied inside the artificial vagina with a glass rod. When the bull was sufficiently excited to jump over the dummy, the penis of the bull was directed into the artificial vagina by holding the sheath to collect the semen in a vial.

After collection, the vial containing semen was put into hot water at 1100F for preventing cold shock. It was closed with cotton and labeled.

**2.3** **Semen processing, preservation :**

Semen was collected at homosexual mount (using male as a teaser) using artificial vagina. After collection, the ejaculates in receptacles and prepared diluents in conical flasks, were placed in a water bath at 37°C. After estimation of sperm motility and concentration, semen was diluted with TRIS-fructose-egg yolk (TFEY) extender. Egg yolk was added with the buffer (20%; v/v). The complete extender was divided into two equal parts and 12.8% glycerol was added to one part of the extender. The other part of the diluent was used to make the initial dilution of semen.The equal parts of initially diluted semen and double concentration glycerol-containing extender were mixed together at four steps during a 3 to 4 hrs cooling process.

**2.4** **Semen evaluation :**

The volume of semen was measured directly from the graduated collecting tube. The density was scored into 1-5 scales. To evaluate mass activity, a drop of undiluted semen was placed on a slide without coverslip and examined under phase contrast microscope(100X) and scored into 1-5 scales. To evaluate sperm motility, a small drop (10 ml) of semen was placed on a prewarmed (37°C) slide, covered by a coverslip and examined under phase contrast microscope (400X).

The concentration of the spermatozoa was determined by using haemocytometer (Bane1 952). Semen was diluted with distilled water at the ratio of 1:200 fresh samples. Eosin nigrosin stain was used to determine the percentage of live spermatozoa. The stain was prepared according to Evans and Maxwell (1990). One drop of semen and one drop of eosin-nigrosin stain were placed closely on a clean slide, and the semen and the stain were mixed with a clean stick, a thin smear was made, dried in air and examined under microscope (400X). At least 200 spermatozoa were examined from each smear.

**Table: Grading of semen according to motility:**

|  |  |  |
| --- | --- | --- |
|  Scale |  Grade | Characteristics |
|  **5** |   (+++++) Excellent | More than 80% of the spermatozoa are in vigorous motion. Swirls and eddies formed due to movements of the sperms are extremely rapid and changing constantly.Movements are so vigorous that it is impossible to observe individual spermatozoon in undiluted semen. |
|  **4** |  (++++) Very good | About 70-80% of the spermatozoa are in vigorous motion. Waves and eddies. |
|  **3** |  (+++) Good | About 50-75% of the spermatozoa are in motion. Motion is vigorous but waves. |
|  **2** |  (++)  Fair | About 30-50% of the spermatozoa are in motion. Movements are vigorous. No waves and eddies. |
|  **1** |  (+)  Poor | Less than 30% of the spermatozoa are in motion. The motion is mostly weak. |
|  **0** |  (O)  Zero | No motility found. |

 **CHAPTER-3**

 **Results**

The data on the general and sexual health of crossbred bulls and the results of their libido tests are presented in the Table 1. The bulls were 5.3 to 9.5 years old. Their pulse rate, breathing rate and rectal temperature varied from 54.3 to 64.3/min, 26.0 to 35.0/min, and 38.5 to 38.9ºC, respectively. The bull’s body condition scores varied from 3.1 to 3.8 (1-5 Scale), they weighed between 547 and 725 Kg and their scrotal circumference varied from 36.7 to 40.0 cm.

**Table 1. Results of the clinical examination and libido test of bulls :**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Parameter** | **Bull-1** | **Bull-2** | **Bull-3** | **Bull-4** | **Bull-5** |
| Pulse rate/min. | 64.3±3.2  | 57.3±4.7 | 54.3±1.5 | 63.3±4.7 | 59.7 ±4.9 |
| Respiration rate/min | 31.0±6.2  | 31.3±2.5 | 35.0±4.6 | 27.7±3.2 | 26.0±1.7 |
| Rectal temperature, °C | 38.5±0.1  | 38.9±0.2 | 38.5±0.2 | 38.8±0.2 | 38.7±0.4 |
| Body condition score,(1-5 scale) | 3.5±0.5  | 3.1±0.2 | 3.8±0.8 | 3.8±0.6 | 3.3±0.6 |
| Body weight, kg | 612±10  | 725±15 | 712±10 | 705±18 | 547±9 |
| Scrotal circumference, cm | 36.8±0.7  | 39.7±1.5 | 38.3±2.3 | 40.0±1.7 | 36.7±2.1 |
| Consistency of testes(1-5 scale)\* | Left 4.0±1.0 Right 4.0±1.0  | 3.7±1.23.7±1.2 | 4.3±1.24.3±1.2 | 4.0±1.74.0±1.7 | 4.0±1.04.0±1.0 |

*\* Scale 5 was the best and 1 was the worst. The values are mean ± SD*

The score of the testicular consistency on the basis of firmness and resilience varied from 3.7 to 4.3 (1-5 scale)

In table 2, the ejaculate volume of the bulls varies from 4.6 to 9.2 and the mass activity of bull-4 is highest which is 3.7 and lowest is 2.7 which found in bull-2.

**Table 2. Results of the evaluation of fresh semen :**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Parameters** | **Bull-1** | **Bull-2** | **Bull-3** | **Bull-4** | **Bull-5** |
| Ejaculate volume, ml | 6.4±0.6a  | 6.4±2.4a | 9.2±2.3a | 5.9±0.6a | 4.6±1.1b |
| Density of semen (1-5 scale)  | 3.3±0.6  | 3.0±0.0 | 3.0±0.0 | 3.3±0.6 | 3.7±0.6 |
| Mass activity (1-5 scale) | 3.3±0.6  | 2.7±0.6 | 3.3±0.6 | 3.7±0.6 | 3.3±0.6 |
| Sperm motility, % | 67±6  | 60±10 | 65±5 | 67±6 | 68±3 |

*\* Scale 5 was the best and 1 was the worst. The values are mean ± SD*

Highest sperm motility found in bull-5 which is 68 and 60 is the lowest sperm motility found in case of bull-2.

**CHAPTER-4**

**Discussion**

The bulls used for routine AI in the present investigation can be regarded as sound exceptone that delivered leukocytes in semen. The presence of leucocytes in semen is anindication of infection in the genital tract of the bull (Bhuiyan et al1997). Individual bullsdiffered with regard to the semen volume and semen motility. The differences insemen parameters among bulls may be attributed to the variation in the secretoryactivities of the accessory sexual glands, scrotal circumference, age and body weight(Graves 1978; Leon et al 1991; Sharma et al 1991; Jainudeen and Hafez 1993).

Average motility of sperm before freezing was varied from 63.7 to 68.8 % and Average motility of sperm after freezing varied from 62.2-63.6% (Hossain et al. 2012 Bang. J. Anim. Sci. 41 (1): 1- 5 ***)*** . But in my study average motility of sperm varied from 60-68%. Motility is one of the most important requirements of fertile semen. Donham et al. (1926) found that semen below normal motility (≥ 90 %) was less than half as effective in producing optimum conception rate. Davis (1939) reported motility of spermatozoa as one of the best single evidence of viability. Duration of motility in stored semen was reported by Comstock (1939) as another reliable index of fertility. Lasley (1943) found no significant difference in fertility of semen containing 55 to 95 per cent live sperm, however, semen containing 20 per cent.

Visual estimation of the percentage of motile spermatozoa is the most commonly used technique for semen evaluation. Accordingly, in the present study, sperm motility dropped significantly due to dilution of fresh semen, chilling, freezing, storing in the bull station and in the District AI centre and transportation from the AI centre to sub-centre.

It could therefore be summarized that if semen diluted,chilled,freezed and stored properly then motility of the semen could be upgraded which helps to successful AI.

**CHAPTER-5**

 **Conclusion**

In the present study, the physical and reproductive parameters of crossbred bulls

(Friesian ´ Sahiwal and Friesian ´ local) have been characterized to some extent

in a selected population.It was also observed that different seasonal factors

can affect the semen volume,semen motility and density of semen.

The freezing protocol and handling of frozen semen during transportation and

management during storage need to be improved to ensure at least 50% sperm

motility at the time of AI, given 30 million total spermatozoa per cow dose.

In my study the sperm motility and mass activity of semen was good enough because of well management and absence of disease.

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**The Author**

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**Biography**

I’m Shibu Das, an intern student at Chittagong Veterinary And Animal Sciences University (CVASU), originate from Raojan, Chittagong. After completing one year intern period,I’ll receive my Doctor of Veterinary Medicine (DVM) degree with lots of real life experience. I finished my primary, secondary and higher secondary education from Chittagong board and engaged with seveal extracurricular activities. I have more interest on microbiology,medicine,surgery,nutrition, and epidemiological field area.