NON-GENETIC FACTORS AFFECTING SEMEN QUALITY OF BULLS

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CERTIFICATE

*This is to certify that the thesis entitled **"NON-GENETIC FACTORS AFFECTING SEMEN QUALITY OF BULLS"** submitted to the Department of Animal Nutrition, Genetics and Breeding, Sher-e-Bangla Agricultural University, Dhaka in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE (MS) in ANIMAL BREEDING AND GENETICS,** embodies the results of a piece of bona fide research work carried out by **MD. HAFIZUR RAHMAN,** Registration No.* **13-05440** *under my supervision and guidance. No part of this thesis has been submitted for any other degree or diploma in any other institution.*

I further certify that any help or sources of information received during the course of this investigation has duly been acknowledged.

Dated:

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____

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DEDICATED TO

My Beloved Parents

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The Author

NON-GENETIC FACTORS AFFECTING SEMEN QUALITY OF BULLS

ABSTRACT

Cattle are the main livestock species in Bangladesh having over 25 billion population. Artificial insemination is mainly used for breeding. In the present study, a total of 235 ejaculates from 26 bulls were analyzed to evaluate the non-genetic factors of semen quality of bulls in the Central Cattle Breeding and Dairy Farm (CCBDF). Among three age groups, adult bulls (72-96 months) produced the highest volume (11.65±0.37ml) and concentration (1372.4±53.94) whereas mass and forward movement was higher in mature (3.43±0.03) and adult (69.44±0.55) respectively. Bull having medium weight (500-750 kg) produced more volume of semen (10.39±0.26ml) but concentration and mass movement was higher in light (up to 500 kg) weight group (1382.1±49.37 and 3.52±0.05) while forward movement recorded higher in heavy weight group (69.72±0.55). Seasonal variation was not significant in the study. Early matured (18-24 months) bulls produced more volume (10.15±0.23ml) but late matured (24-36 months) bulls have more concentration (1402.7±43.14), mass movement (3.46±0.04) and forward movement (68.54±0.45). Thus the study showed that, adult bulls having the body weight up to 500 kg (light) from late matured bulls will produce quality semen in any season in terms of volume, concentration, mass and forward movement.

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LIST OFABBREVIATIONS AND SYMBOLS

>	=	Greater than
<	=	Less than
±	=	Plus minus
°C	=	Degree Celsius
%	=	Percentage
AI	=	Artificial insemination
ANOVA	=	Analysis of Variance
AV	=	Artificial vagina
cm	=	Centimeter
CV %	=	Percent Coefficient of Variation
DF	=	Degree of freedom
DLS	=	Department of Livestock Services
DM	=	Dry matter
et al.,	=	And others
e.g.	=	exempli gratia (L), for example
etc.	=	Etcetera
g	=	Gram (s)
GDP	=	Gross Domestic Product
GM	=	Geometric mean
i.e.	=	id est (L), that is
Kg	=	Kilogram (s)
L	=	Litre
Lbs	=	Pound
m^2	=	Meter squares
mg	=	Milligram
ml	=	Milliliter
MS	=	Mean Square
No.	=	Number
SAU	=	Sher-e-Bangla Agricultural University
SAS	=	Statistical Analysis
SE	=	Standard Error
SS	=	Sum of Squares
Viz.	=	Namely

CHAPTER 1
INTRODUCTION

Livestock is one of the major sub-sectors of agriculture in Bangladesh that plays the vital role to meet the requirement of animal protein i.e. milk and meat as well as to the economy of the country. Cattle are the major livestock species that provide meat and milk. According to the Department of Livestock Services (DLS), a total of 412.2 million of livestock population including 24.3 million of cattle are producing animal protein (DLS, 2020). As a result, Bangladesh is sufficient in meat having availability of 126 g meat per day per head instead of 120g of demand and deficit in milk having availability of 175ml milk per day per head instead of 250ml of demand (DLS, 2020). On the other hand, contribution of livestock sub-sector to country's Gross Domestic Product (GDP) is about 1.43 percent which is 13.44 percent of agricultural GDP with a growth rate of 3.04 percent (DLS, 2020). Twenty percent of our population took this sub-sector as permanent occupation whereas fifty percent of the national employment indirectly related to this sub-sector (Ahmed *et al.,* 2008).

Producing meat and milk is the key objective of cattle rearing. However, manure, hides, bones and some other by-products are also found from cattle. But we are facing the problem of low productivity due to different genetic and non-genetic or environmental causes (Forcada and Abecia, 2006). Different diseases of animals are also responsible for the hindrance of livestock development in our country (Islam *et al*., 2001). Breeding is the major technological improvement process in the dairy industry. A cross-breeding program for cattle was introduced in the country in 1976 with a view to improving the milk production efficiency of indigenous cows (Shamsuddoha and Edwards, 2000). The genetic improvement of livestock can be achieved by proper utilization of proven sires and dams by following the artificial insemination (AI) with frozen semen and Embryo Transfer Technology (ETT).

Besides the lack of developed breed, quality semen production in Bangladesh is facing some problems due to age, body weight, seasonal variation and age at sexual maturity of the bull. The findings of Asad *et al*. (2004) suggested that Friesian produced better quality semen than other genotypes while 4 to 6 years age and >600 to 700 kg body weight of bulls performed better than other age and body weight groups. The summer season gave better quality semen than other seasons.

Semen, also called seminal fluid, fluid that is emitted from the male reproductive tract and that contains sperm cells, which are capable of fertilizing the female's eggs. Semen also contains liquids that combine to form seminal plasma, which helps keep the sperm cells viable. The term semen quality refers to the volume, concentration, mass motility and forward motility etc. It also includes the ratio of sperm per ml, ratio of live and dead sperm, ratio of healthy and defected sperms etc.

Artificial insemination (AI) is the process of collecting sperm cells from a male animal and manually depositing them into the reproductive tract of a female. One can cite a number of potential benefits from the use of artificial insemination. It is the first generation reproductive biotechnology that has made a profound contribution to the genetic improvement as well as recognized breeding tool of the cattle (Mazumder *et al.,* 2020). In Bangladesh the artificial insemination (AI) technique has been used near about fifty years and every year this program is extended. Using A.I. the productive and reproductive performances of our cattle population has improved day by day (Islam *et al.,* 2015). About 90 lakh doses of semen were used in 2020 and the current coverage under artificial insemination (AI) is about 65-70% in our country (DLS, 2020). To hasten the development of livestock, we have to maximize our artificial insemination facility from door to door. The current major AI (Artificial Insemination) services providers are government (DLS), Milk Vita, and 5 private sector stakeholders (BRAC, ACI, ADL, Lal Teer and Ejab).

The production as well as the productivity of indigenous cattle are unsatisfactory and can be improved by crossbreeding of indigenous breeds with exotic ones. The semen production, being a quantitative trait, is affected by genetic as well as non-genetic factors. The semen production is affected by season (Singh *et al.,* 2000; Mathur *et al.,* 2002; Bhoite *et al.,* 2005) as well as period (Bhoite *et al.,* 2005). Age also affects semen production as reported by various workers (Sudheer, 2000; Mandal *et al.,* 2010). Due to lack of scientific selection of bull based on their performance, there may be possibility of serious deterioration in the quality of indigenous germplasm. Reproduction parameters are more influenced by environment (Mukhopadhyay *et al.,* 2010). Optimizing the age of breeding bulls in relation to their fertility will help to assess the breeding soundness of bovine bulls. Kuhn and Hutchison (2008) found that the age of the bull at the time of mating will be the major factor and the variation of conception rate and fertility will reach maximum at five years of age then decreased somewhat approximately up to age of 9 or 10 years. A deficiency in the breeding ability of one bull has larger impact on herd productivity. Using a sub-fertile bull may lead to longer calving intervals and lower number of calves produced in the herd (Basu, 1985). A study on four types of crossbred bulls in Bangladesh reported that Holstein–Friesian × Zebu crossbred bull produced better quality semen than Sahiwal × Zebu, Sindhi × Zebu and Jersey × Zebu bulls (Shaha *et al.,* 2008). So, there is little information on the effects of non-genetic factors on production of breeding bulls and semen quality parameters. So, the present study tries to look into these aspects of crossbred bull production.

In our sub-continent, several research works has been carried out to evaluate the effect of non-genetic factors on semen quality. But in Bangladesh, there is no recent study on this topic except the study of Asad *et al*. (2004). In the mean time, environment specifically seasonal variation took place in our country due to global warming and other natural calamities. Thus, there are some necessities to study on this topic further to update the research findings.

From that stand point the present research work has been undertaken with following objectives:

- To assess the effect of non-genetic factors affecting semen quality
- To find out the effect of age, body weight, season and age at sexual maturity on different qualitative traits of semen i.e. volume, concentration, mass movement and forward movement
- To help in the selection of bulls in producing more and more quality semen for artificial insemination

CHAPTER 2
REVIEW OF LITERATURE

Quality semen is essential for breeding. Many research works have been done to judge the quality of semen of different types of bulls in the world. Some of the research findings are summarized in this chapter according to our research parameters.

2.1 Age

Bhakat *et al.* (2011) reported that all semen traits were significantly ($p<0.01$) affected by age, except sperm concentration in their study. The ejaculate volume (ml) significantly ($p<0.01$) enhanced with the increasing age (year) of bulls (4 to 6 years) but decreased again for older age groups (>6 years). The mass activity of semen significantly ($p<0.01$) increased as the bull matured but decreased again for the older age group. Mass activity was highest in 4-to-5-year age group bulls and lowest in <4 and 5-to-6-year age group bulls. The highest and lowest sperm concentrations were recorded for the bulls in 4-to-5-year age group and <4-year age group, respectively. Sperm concentration per ejaculate was higher in 5-to-6-year age group bulls.

Brito *et al*. (2002a) experimented on sperm production and semen quality in 107 *Bos indicus*, *B. taurus* and cross-bred bulls of 3 artificial insemination (AI) centers in Brazil and found that, ejaculate volume, total number of spermatozoa and number of viable spermatozoa increased ($p<0.05$) with age. However, there was no significant effect of age on sperm concentration, motility, major and total defects. The proportion of spermatozoa with minor defects was highest ($p<0.05$) in bulls 37–60 months of age.

Brito *et al*. (2002b) evaluated on 7 *Bos indicus* and 11 *Bos taurus* bulls from one artificial insemination (AI) center and found that, increased bull age was associated with decreased sperm motility ($p<0.10$) and increased minor sperm defects ($p<0.001$).

Fuerst-Waltl *et al.* (2006) collected semen data from two Austrian AI centers in the years 2000 and 2001. In total, 3625 and 3654 ejaculates from 147 and 127 AI bulls, respectively, were analyzed regarding ejaculate volume, sperm concentration, percentage of viable spermatozoa in the ejaculate, total spermatozoa per ejaculate and motility. Age of bull significantly affected all traits ($p<0.01$ to $p<0.001$) except motility score in center 2. Ejaculate volume and total number of spermatozoa increased with age of bull while sperm concentration was lower in higher age classes (center 1).

Sarakul *et al*. (2017) collected the data of 11,121 ejaculates from 130 dairy bulls having 62.5-100% Friesian pedigree from 2001 to 2015 and presented that, semen quantity and quality varied by year-month, ejaculation number, age, ambient temperature at collection time. Ambient temperature had no effect on volume and appearance whereas bulls with age from >24 to >72 months produced favorable quality and quantity semen.

Asad *et al*. (2004) collected data of 1422 semen samples from 22 different bulls of seven genotypes and reported that, the bulls of 4 to 6 years old provided the highest semen volume (13.60±0.18 mL), sperm concentration (1219.89±10.72 million sperm mL^{-1}), mass movement (4.66±0.06 grade) and forward movement (61.34 ± 0.28%) of sperm.

Rodionovskii (1977) studied semen characters in 2540 ejaculate of 19 bulls, aged 2 years and in ejaculates of surviving bulls up to 10 years of age. Ejaculate volume increased from 3.48 at 2 years to 5.70 ml at 9 years Sperm concentration decreased gradually with age.

Jamrishka and Novy (1980) studied 782 first inseminations with semen of 119 Slovakian Pied bulls from 21 lines and the repeatability of the semen volume was 0.25 for data on individual bulls and 0.06 for data on lines.

Sarder (2008) studied impact of age, body weight, body conditions, and scrotal circumference on sperm abnormalities of bulls used for artificial insemination (AI) in Bangladesh on 1390 ejaculates of semen from 71 bulls. Total head abnormalities, free loose head, mid-piece, tail abnormalities, proximal and distal cytoplamic droplets, total tail abnormalities and total sperm abnormalities were recorded. Age of the bulls had significant effect on sperm abnormalities ($p<0.01$) except for proximal cytoplasmic droplet. He concluded that the middle (8 to <10 yrs) and older (>10 yrs) bulls having 36 to <38 cm scrotal circumference had the lowest sperm abnormalities.

Ziegler (1984) observed that in 990 ejaculates from 35 Simmental bulls, the main difference in semen characters was for ejaculate volume (703 Vs 4.22 ml). Almost all semen characters were significantly affected by bull and age. The 90 days non-return rate was significantly correlated with ejaculate volume (0.11) and with sperm concentration in the older bulls (0.32)

Barbosa *et al.* (1991) evaluated semen traits at 27 and 39 months of age in 7 Canchim and Nelore bulls. Volumes were significantly greater in Canchim than in Nelore bulls at 27 months, but not at 39 months of age. There were no significant breed differences or breed × age interactions for ejaculate volume or sperm motility, concentration or abnormalities, but semen quality improved with increasing age of bull.

Siratskii (1992) studied over a 20 year period from 642 bulls belonging to the pedigree breeding associations in Sumy, Chernigov and Kharkov Oblasts was studied. The relationships between age of the bull and the volume of ejaculate, concentration of spermatozoa, mass activity were studied. Age effects were found in all these traits. A significant dependence of ejaculate volume and the total number of spermatozoa per ejaculate on live weight was also found.

Correlation between the various measures of semen quality and fertilizing ability were calculated.

Zhang *et al.* (1993) evaluated 33 Holstein bulls at the Beijing Bulls Station were grouped by age (11 animals per group): 1 to 2 years (group A), 2 to 4 years (group B) and > 4 years (group C). The semen quality of each ejaculate (volume, sperm concentration, total number of spermatozoa and sperm motility) was recorded for 1 month. The results showed that scrotum circumference, ejaculate volume and blood testosterone level in groups B and C were greater (p<0.01) than those in group A.

Adamou *et al.* (1996) studied over a 3 month period, the ejaculates collected once a week from 10 and twice a week from 6 Borgere bulls. For once and twice a week collection, ejaculate volume averaged, 343 and 3.16 ml respectively, sperm motility rating 3.85 and 4.30, motility percentage 75.7 and 70.5, sperm concentration 1.19×10^9 and 0.76×10^9 and sperm number per ejaculate 4.09×10^9 and 2.45×10^9.

Diarra *et al.* (1997) studied 294 young Holstein bulls between 10 and 18 months old. The sire effect was significant for the volume (p<0.001), concentration (p<0.05) and the number of motile spermatozoa per ejaculate (p<0.001). Intra class regression studies indicated that each increase of 1 day of age in young bulls increased the volume of the ejaculate by 0.004%, the concentration and the motility of the ejaculate by 0.024×10^8 spermatozoa/ml and 0.03% and the concentration of the ejaculate by 0.14×10^8 spermatozoa/ml.

Javed *et al.* (1997) studied 16 bulls, aged 3 to 4, 6 to 7, 8 to 9 or 12 to 15 years, for a period of 1 year. In the summer there was a higher (p<0.05) percentage of dead spermatozoa in the semen samples. There was no difference in sperm size among age groups but bulls aged 8 to 9 years had a higher percentage of dead spermatozoa.

Boge (1998) observed from two experiments carried out with semen of progeny tested bulls of average age 7.2 year, 67 hulls awaiting their progeny test results (4.9 years), 45 young bulls undergoing progeny testing (1.2 years) and 67 young bulls whose semen had been used in test inseminations (1.80 years). In each experiment, 2 ejaculate of each bulls were examined. There were no significant differences between the 4 groups of bull in respect of sperm volume (measured by a computerized cell analyzer). Spermatozoa in a hypotonic medium had a volume 1.7 to 1.9 times greater than under isotonic conditions (p<0.05). The NR (non return) rates to insemination, which were available for some of the bulls, were not significantly correlated with semen quality traits or sperm volume.

2.2 Body Weight

Gopinathan *et al.* (2018) investigated the influence of body weight and scrotal circumference and its phenotypic correlation with semen production traits in crossbred Holstein Friesian bulls. Overall least-squares mean for semen volume, sperm concentration, mass activity, initial sperm motility, post-thaw motility and number of doses/ejaculate were 4.10±0.06 ml, 842.7±21.7 million/ml, 2.11±0.03, 55.6±0.0%, 49.8±0.0% and 196.92±5.4, respectively. The body weight and scrotal circumference of crossbred bulls influenced (p<0.01) all semen production traits. The body weight was positively correlated (p<0.01) with scrotal circumference (0.85), semen volume (0.56), sperm concentration (0.12), mass activity (0.41) and number of doses/ejaculate (0.49). The phenotypic correlations of scrotal circumference with semen volume (0.47), sperm concentration (0.06), mass activity (0.39) and number of doses/ejaculate (0.36) were positive (p<0.05). In brief, the bulls having body weight between 450 to >650 kg and scrotal circumference >36 cm exhibited better performance with regard to majority of the semen production traits.

Asad *et al*. (2004) collected data of 1422 semen samples from 22 different bulls of seven genotypes and reported that, the maximum volume of semen

(12.16±0.22 mL) and forward movement (60.25±0.45%) of sperm was obtained in the bulls of 400 to 600 kg body weight.

Sarder (2008) studied impact of age, body weight, body conditions, and scrotal circumference on sperm abnormalities of bulls used for artificial insemination (AI) in Bangladesh. During 1997-2002, 1390 ejaculates of semen from 71 bulls from Central Cattle Breeding Station and Dairy Farm, Savar, Dhaka, Rajshahi Dairy and Cattle Improvement Farm, Rajabarihat, and District Artificial Insemination Centre, Rajshahi, were collected and examined. Total head abnormalities, free loose head, mid-piece, tail abnormalities, proximal and distal cytoplamic droplets, total tail abnormalities and total sperm abnormalities were recorded. Body weight had significant effect (p<0.05) except for the total head abnormalities, while body conditions had no significant effect on mid-piece, total tail abnormalities and total sperm abnormalities except for the total head abnormalities. He concluded that the bulls having body weight of <450 kg were very good body condition had the lowest sperm abnormalities.

2.3 Season

Fiaz *et al*. (2010) collected semen production data of Holstein-Friesian and Jersey bulls (n = 18 for each breed) over a period of three years to examine seasonal effects on quantity and quality of semen. The number of ejaculates per bull recorded during three seasons were not significantly different (p>0.05) in both breeds. Friesian bulls produced lower ejaculatory volume during dry summer season, whereas Jersey bulls produced higher (p<0.05) volume during wet summer compared to other seasons. Seasonal pattern of mass motility and individual motility of semen was different between two breeds. The mass motility of semen in both breeds was significantly lower during wet summer. However, individual motility in the semen of Friesian bulls did not differ among seasons (p>0.05) but in Jersey bulls it was lower during wet summer than other seasons. Wet summer resulted in reduced number of semen doses frozen per bull in both breeds. The study indicated that wet summer season deteriorated semen quality in terms of mass motility, individual motility and number of doses in both breeds, except individual motility in semen of Friesian bulls.

Solanki *et al*. (2019) carried out a study on six Gir bulls (4-6 years) for a period of 8 months during summer and winter to study the seasonal variations and correlations amongst the sexual behavior traits. After studying on a total of 96 observations at fortnightly interval, they concluded that, the Gir bulls had good sexual behavior scores throughout the year. However, bulls showed better sexual behavior in winter as compared to summer.

Brito *et al*. (2002b) evaluated on 7 *Bos indicus* and 11 *Bos taurus* bulls from one artificial insemination (AI) center and found that, ambient temperature and humidity did not significantly affect sperm production and semen quality, probably because there was little variation in these variables. Month accounted for less than 2% of the variation in sperm production and semen quality. Increased bull age was associated with decreased sperm motility ($p<0.10$) and increased minor sperm defects ($p<0.001$).

Fuerst-Waltl *et al.* (2006) collected semen data from two Austrian AI centers in the years 2000 and 2001. They reported that, effects for temperature before semen collection were stronger than for temperature on day of collection. However, results were partly inconsistent for AI centers. In center 1, ejaculate volume was significantly affected by average temperature during epididymal maturation ($p<0.01$ for quadratic regression coefficient) and spermatogenesis ($p<0.001$ for linear regression coefficient). In center 2, however, solely ambient temperature during spermatogenesis was found to have a significant effect on ejaculate volume ($p<0.05$ and $p<0.01$ for linear and quadratic regression coefficient).

Sarakul *et al*. (2017) collected the data of 11,121 ejaculates from 130 dairy bulls having 62.5-100% Friesian pedigree from 2001 to 2015 and presented that, semen quantity and quality varied by year-month, ejaculation number, age, ambient temperature at collection time. Ambient temperature had no effect on volume and appearance whereas bulls with age from >24 to >72 months produced favorable quality and quantity semen.

Asad *et al*. (2004) collected data of 1422 semen samples from 22 different bulls of seven genotypes and reported that, the volume was maximize (12.15±0.23 mL) in summer season.

Djimide and Weniger (1984) observed 9000 ejaculates of 296 bulls and sexual behavior of the bulls was recorded. The bulls were of the breeds or crosses like: Sahiwal, Red Sindhi (RS), Friesian, Jersey, Local×Friesian, Local×Jersey, Sahiwal×Friesian, RS×Friesian, Sahiwal and Jersey and RS×Jersey. Semen volume (ml), sperm concentration (million/mm^3) pH and sperm mass motility were determined and also forward motility on the 1^{st}, 2^{nd}, 3^{rd} and 4^{th} day after semen collection. Differences in semen quality were found between breeding groups and seasons. The Zebu breeds had significantly better semen quality and poorer libido than other groups. Crossbreeds, especially of local breeds with Friesian or Jersey, had better semen quality and libido than bulls of imported breeds.

Amir *et al.* (1982) worked with 10 Friesian bulls of proven fertility in every winter (January-March) and summer (July-September) for a period of 3 years. They reported that the bulls and years significantly affected semen volume, sperm concentration and sperm motility before and after freezing. There were no significant differences between samples collected in summer and winter for semen characteristics.

Saxena and Tripathi (1984) studied 83 ejaculates collected from 4 bulls over a 12 months period. There were no significant differences between seasons in ejaculate volume, sperm motility, sperm concentration, numbers of spermatozoa per ejaculate, number of live spermatozoa per ejaculate, sperm resistance and methylene blue reduction time. The proportion of live spermatozoa averaged 86.64%, 85.00%, 83.75%, 90.50% and 85.05% in spring, summer, rainy, autumn and winter respectively and the proportion of abnormal spermatozoa was 13.27%, 13.13%, 13.00%, 8.56% and 7.15% (both p<0.01).

Tomar *et al.* (1985) studied 9 bulls aged <24, 25-30, and >37 months. Ejaculate volume, mass motility score and the percentage of live spermatozoa increased significantly with advancing age, while the percentage of abnormal spermatozoa decreased significantly. Season had a significant effect on the mass motility score (lowest during November-February) and on sperm concentration (lowest during March-June).

Sexena and Tripathi (1986) observed 59 ejaculates collected over 1 year period from 204 Danish Red bulls. In the rainy, autumn, winter, spring and summer seasons, semen volume ranged from 2.80 to 4.31 ml, progressive sperm motility (scale 0 to 5) from 3.63 to 4.25, sperm motility from 70.31% to 77.50%, sperm concentration ($\times 10^6$/ml) from 650 to 1310, number of spermatozoa per ejaculate ($\times 10^6$/ml) from 2585 to 3352. The effects of season were significant for progressive motility and for the percentage of sperm head abnormalities. The effects of storage at 20×-28×C for 0 to 72 hours were studied. There were no significant differences in sperm motility or survival during storage between seasons. Satisfactory sperm motility was maintained up to 72 hours in autumn Vs 48 hours in the other seasons.

Stalhammar *et al.* (1988) studied 215 Swedish Red and White and Swedish Friesian bulls in 1983-85 in respect of libido, ejaculate volume, number of spermatozoa per ejaculate, sperm motility before and after freezing and the number of semen doses per collection was generally significantly affected by season of collection and age of bull.

Stalhmmar *et al.* (1989) studied the effects of bulls, season and year of collection and age at collection on ejaculate volume (3 to 8 ml), sperm motility, total number of sperm cells per ejaculate and number of semen doses per collection. Phenotypic correlations among them were derived. Motility and ejaculate volume were affected by size ($p<0.01$). Bull, season of collection and age at collection each affected ($p<0.001$) the 4 traits mentioned above.

Muhuyi (1991) investigated that factors affecting the quality of ejaculates collected from 39 bulls in 1983-86. Year had a significant effect on ejaculate volume and there were significant year season effects on sperm concentration and post-dilution sperm motility ($p<0.05$). Post thawing sperm motility was affected by season (higher in the rainy than in the dry season, $p<0.05$). There were no significant age of bulls, year×age or season×age effects on any of the traits investigated.

Verma *et al.* (1991) observed 96 ejaculates from 6 Jersey bulls (8 ejaculates per bull per season) in winter and summer. Season had no effect on live sperm percentage in undiluted or diluted semen. Progressive sperm motility in undiluted semen was not affected by season, but was higher in undiluted semen and in thawed semen collected in summer. In undiluted semen total sperm abnormalities were not influenced by season. It was concluded that seasonal effects on sperm characters are not important.

Luthra and Marinoy (1995) studied semen from 10 Holstein Friesian bulls. There was a 9% increase in the incidence of total sperm abnormalities in frozen semen compared with fresh semen.

Alvarez *et al.* (1996) observed in 6 hours after thawing of semen of Holstein bulls with significantly higher sperm motility and percentage of live spermatozoa than Siboney bulls. Semen quality was significantly better in the rainy than in the dry season. Acrosomal status differed significantly between the breeds.

Younis *et al*. (1998) studied in 18 Nili-Ravi buffalo bulls of different ages ranging from 3 to 15 years. The ejaculatory volume was higher ($p<0.05$) in adult and old than young bull during the low (May-July) than the peak (September-November) breeding season. Mass activity was significantly higher

($p<0.05$) in the peak than the low breeding season. Sperm concentration did not differ among bulls during two seasons. Total sperms per ejaculate were higher ($p<0.05$) in semen collected from adult and old than in young bulls and in the low than the peak breeding season.

Usmani *et al*. (1993) and Li-junjie *et al*. (2001) reported that motility was reduced significantly in hot and humid season. Similarly, Fawzy and Rabie (1996) and Mostari *et al*. (2005) reported that sperm motility was deteriorated during summer in Friesian bulls. However, Brito *et al*. (2002b) and Fonseca (1995) could not find any significant effect of season on mass motility and individual motility during summer.

Bhakat *et al*. (2011) demonstrated that season had significant effect ($p<0.05$) on volume (VOL), total volume per day (VOLD) and sperm concentration per ejaculate (SPCE) of Sahiwal bulls (VOL at 5% level, VOLD and SPCE at 1% level). Semen volume and total volume per day was highest during rainy (3.87±0.04 and 6.09±0.09 respectively) and the lowest on winter (3.72±0.04 and 5.63±0.08, respectively) season. Significant differences ($p<0.05$) in volume between rainy and winter were observed. Total volume per day in summer (5.72±0.10) and winter (5.63± 0.08) seasons significantly differ ($p<0.01$) with rainy season (6.09±0.09). Mass activity (MA), initial motility (IM) and sperm concentration per ml (SPC) were highest during rainy (2.35±0.02, 55.97±0.003, and 779.77±8.41) and lowest during winter (2.30±0.02, 54.88±0.002, and 754.58±7.97) seasons but non-significant ($p<0.05$). During the rainy season average SPCE was significantly higher (3,141.01±46.05) as compared to winter (2,919.64±43.68) season, respectively.

Mathevon *et al.* (1998) reported that semen characteristics generally improved significantly with age of young bulls. Season significantly affected all semen traits in young bulls but did not significantly affect volume and sperm motility of mature bulls. Performance was better in winter than in summer. The highest numbers of motile spermatozoa per ejaculate were obtained with intervals of at least 4 to 5 d between collections. Although the bull handler and semen collector caused less than 10% of the variance, the collection team significantly affected semen volume, number of sperm, and number of motile sperm per ejaculate for both growing and mature bulls.

2.4 Age at Sexual Maturity

Brito *et al*. (2002a) experimented on sperm production and semen quality in 107 *Bos indicus*, *B. taurus* and cross-bred bulls of 3 artificial insemination (AI) centers in Brazil and found that, semen concentration, total number of spermatozoa and number of viable spermatozoa was significantly higher in *B. indicus* than *Bos taurus* ($p<0.05$). Here, indigenous breed became mature lately and improved breed early.

Asad *et al*. (2004) collected data of 1422 semen samples from 22 different bulls of seven genotypes and reported that, the highest volume (12.01±0.22 mL) of semen, sperm concentration (1182.29±14.30 million sperm mL^{-1}), mass movement (4.40±0.08 grade) and forward movement (58.74 ± 0.54%) of sperm was found in 3 years age of sexual maturity.

Lunstra & Echternkamp (1982) evaluated semen characteristics every 2 weeks from 7 through 13 months of age in 31 beef bulls representing six breed groups (Hereford, Angus, Hereford × Angus crossbreds, Angus × Hereford crossbreds, Red Poll and Brown Swiss). Breeds differed in age at puberty, defined as the age at which an ejaculate was first obtained that contained a minimum of 50×10^6 total spermatozoa with at least 10% progressive motility (Hereford, 326±9 days; Angus, 295×4 days; Hereford×Angus, 300±8 days; Angus×Hereford, 296×9 days; Red Poll, 283±9 days and Brown Swiss, 264±9 days). Significant breed differences also were observed in concentration of spermatozoa, progressive motility, seminal protein concentration, abnormal spermatozoa and acrosomal morphology. Considerable variation was observed for the majority of pubertal traits among the 31 bulls, indicating that differences in stage of pubertal development existed among and within breeds of beef bulls between 7 and 13 months of age. However, adjustment of data to age at puberty negated breed differences ($p>0.10$), indicating that the pubertal patterns of change occurring in each semen characteristic were similar for the breeds evaluated. Concentration of spermatozoa, progressive motility, seminal protein concentration, percentage spermatozoa with normal head and tail morphology and percentage spermatozoa with normal acrosomal morphology increased ($p<0.01$) from puberty through 16 weeks after puberty in all bulls and all breeds. During the first 6 weeks after puberty, rapid increases ($p<0.01$) were observed in percentage spermatozoa exhibiting normal head morphology (excluding acrosomes) and progressive motility, and a rapid decrease ($p<0.01$) was observed in percentage spermatozoa with proximal cytoplasmic droplets, with values at +6 weeks approaching those reported for mature bulls. Percentage spermatozoa with normal acrosomal morphology and concentration of spermatozoa improved more slowly and had not reached mature levels by 16 weeks after puberty. Because age at puberty varied by 62 days among breeds and 88 days among bulls and important characteristics of semen quality improved slowly after puberty, careful evaluation of the stage of pubertal development in individual bulls is recommended before selecting young bulls for natural breeding or artificial insemination.

CHAPTER III

MATERIALS AND METHODS

3.1 Study Site

The study was conducted at the Central Artificial Insemination Laboratory, in the Central Cattle Breeding and Dairy Farm (CCBDF), Savar, Dhaka Bangladesh. Data of semen quality of bulls were collected from the record book.

Plate 3.1: Central Cattle Breeding and Dairy Farm (CCBDF) and Central Artificial Insemination Laboratory

3.2 Duration of Study

The study was conducted from January to December, 2020.

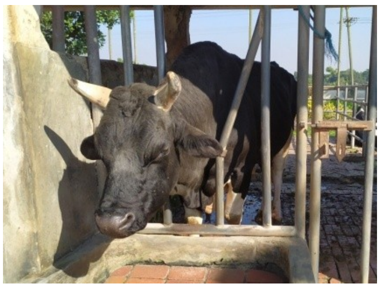
	
Friesian (F)	Sahiwal×Friesian (SL×F)
	
Shahiwal (SL)	Friesian (F) 62.5%
	
Local (L)	Friesian (F) 75%
	
Local ×Friesian (L×F)	Red Chittagong Cattle (RCC)

Plate 3.2: Different breeds of bull in AI Center

3.3 Study Animal

- Local (L)
- Friesian (F)
- Sahiwal (SL)
- Red Chittagong Cattle (RCC)
- Local ×Friesian (L×F)
- Sahiwal×Friesian (SL×F)
- Friesian (F) 62.5%
- Friesian (F) 75%

3.4 Data Collection

The records of genotype, age, body weight and age at sexual maturity of bulls as well as data of volume of ejaculate, density or semen concentration, motility (mass movement and forward movement) in three different seasons were kept for the experimental animals.

In the present study the experimental animals were divided into three age groups: 48-72 months, 72-108 months and 108-144 months for the analysis of breed and age effects.

The number of animals in each genotypes and the number of total ejaculates are shown in Table 3.1.

Table 3.1. Number of animals and total number of ejaculates of different genotypes of bulls

Genotype	**Female**	**Male**	**No. of animals**	**Number of ejaculates**
Friesian purebred (F)	F	F	3	27
Shahiwal purebred (SL)	SL	SL	3	27
RCC purebred (RCC)	RCC	RCC	3	26
Local purebred (L)	L	L	3	27
Shahiwal x Friesian (SI x F)	SL	F	3	27
Local x Friesian (L x F)	L	F	3	27
Local x Friesian x Friesian (L x F x F)	LxF	F	5	44
Local Friesian (L x F2)	L	Lx FxF	3	27
Total			26	232

3.5 Feeding and Management of Bulls

Bulls were fed in a regular feeding practice with emphasis of concentrates. Concentrate feeds were divided into two and fed two times a day in the morning and in the evening.

Concentrate feeds include chickpea, wheat bran, till oil cake, rice bran, urea molasses and common salt. The green grasses like napier, napier pukchung, para, maize and oats and in the form of silage were also supplied on the basis of year round availability. The bulls were fed a balanced ration consisting of 50 percent mixed concentrates and 50 percent hay with supplementation of vitamins and minerals. Feeds were given on the basis of TDN (total digestible nutrient) and CP (crude protein). The animals fed at the rate of

- ✓ Dry mater (DM) = 1.5 kg/100 kg body weight
- ✓ TDN = 60% of the total DM
- ✓ CP = 16%- 18 % of total feed or 11% of the DM

On the basis of DM, concentrate consists of 1/3 of the ration and rest 2/3 of roughages in which 50% straw and 50% green grasses.

Bulls were tested for fertility before putting them in the breeding herd. Semen was collected using a standard artificial vagina from the bulls thrice a week (Sunday, Tuesday and Thursday). Immediately after collection semen sample was kept in warm water at about 100 to 105°F.

3.6 Semen Characteristics Analyzed

The following records on semen quality were analyzed:

- ➢ Volume of ejaculate: Volume per ejaculate was recorded from the graduated collection vials.
- ➢ Concentration: The total number of spermatozoa per 3 ml of raw semen was enumerated by haemocytometer method according to Herman and Madden (1963). The following formula was used for calculating total number spermatozoa per ml of fresh semen.

  $$N = \frac{C \times 4000}{S} \times D$$

  Where,

  N = Number spermatozoa per ml of semen

  C = Number of spermatozoa counted in given number of small squares

  S = Number of small squares counted

  D = Dilution ratio
- ➢ Motility (mass movement and forward movement %): Percentage of motility (mass movement and forward movement of spermatozoa) was estimated by microscopic examination and was recorded in intervals of ten percent.

Volume, sperm concentration and motility (undiluted semen under phase contrast microscope) were recorded for every ejaculate.

3.7 Factors Included in the Analysis

The data on the above parameters were analyzed to see the effect of the following factors:

- Genotypes of bulls: There were 8 (eight) genotypes of bulls as mentioned in the Table 3.1.
- Age groups of bulls: Bulls were divided into three age groups as up to 72 months (≤ 6 years), 72 to 96 months (6 to 9 years) and above 96 months (≥ 9 to 12 years) and termed as mature, adult and aged chronologically.
- Body weight groups of bulls: There were three body weight groups of bulls as up to 500 kg at light, 500-750 kg as medium and above 750 kg as heavy weight.
- The seasons were divided as follows:
    - i. Summer season (March to June)
    - ii. Rainy season (July to October)
    - iii. Winter season (November to February)
- Age at sexual maturity: Bulls were included having two years age of sexual maturity and three years age of sexual maturity.

3.8 Design of Experiment

There was sufficient hierarchy in the data structure. The number of observation varied from class to class. So, the statistical design of this experiment was non orthogonal factorial in nature (Snedecor and Cochran, 1980)

3.9 Statistical analysis

All values were expressed as (Mean±SE). Statistical significance of differences between different parameters was evaluated by using student's t-test. The statistical analysis was done by SPSS program (Version 16.0; SPSS Inc., Chicago, IL, USA).

CHAPTER IV

RESULTS AND DISCUSSION

Data of semen quality of bulls were collected from the record book of the Centrall Artificial Insemination Laboratory, in the Central Cattle Breeding and Dairy Farm (CCBDF), Savar, Dhaka Bangladesh. A total of 235 ejaculates from 26 bulls were collected in different aspects to evaluate the effect of non-genetic factors on semen quality.

4.1 Effect of Age on Semen Quality

All the bulls were divided into three groups according to their age. Bulls of up to 72 months were termed as mature, 72-96 months of age as adult and more than 96 months were termed as aged. According to this grouping, 127 ejaculates from 17 bulls were from mature bulls whereas 54 ejaculates from 6 bulls were from both adult and aged group of bulls. Table 4.1. Semen quality of bulls from different age groups

Table 4.1. Semen quality of bulls from different age groups

Age Group	**Observations**	**Volume (ml)**	**Concentration (million/ml)**	**Mass movement (grade)**	**Forward movement (%)**
		Mean±SE	**Mean±SE**	**Mean±SE**	**Mean±SE**
Mature	127	$8.54^{c}\pm0.24$	$1292.4^{a}\pm35.17$	$3.43^{a}\pm0.03$	$67.52^{b}\pm0.36$
Adult	54	$11.65^{a}\pm0.37$	$1372.4^{a}\pm53.94$	$3.39^{a}\pm0.05$	$69.44^{a}\pm0.55$
Aged	54	$9.82^{b}\pm0.37$	$1281.1^{a}\pm53.94$	$3.19^{b}\pm0.05$	$68.52^{ab}\pm0.55$

Mean values in the same column with different superscripts (a, b) differ significantly at $p<0.05$.

4.1.1 Volume of semen

Significant difference found in the volume of semen in different age groups. The highest volume of semen was found in adult bulls (11.65±0.37) followed by aged (9.82±0.37) and mature (8.54±0.24) age groups (Table 4.1 and Figure 4.1). All three groups differed significantly ($p<0.05$). Lowest age in mature age group of bulls was 30 months. Physiologically it was very near to their maturation. So, low semen volume was a normal phenomenon. On the other hand, aged bulls of more than 96 months were in the decreasing phase of their production life. Similar result found in the study of Bhakat *et al.* (2011). Asad *et al,* (2004) found highest volume in bulls of 4-6 years. Brito *et al.* (2002a) and Fuerst-Waltl *et al.* (2006) showed that, semen volume increased with the age of the bull. Bulls with age from >24 to >72 months produced favorable quality and quantity semen in the study of Sarakul *et al.* (2017). All the reports supported the result of this study.

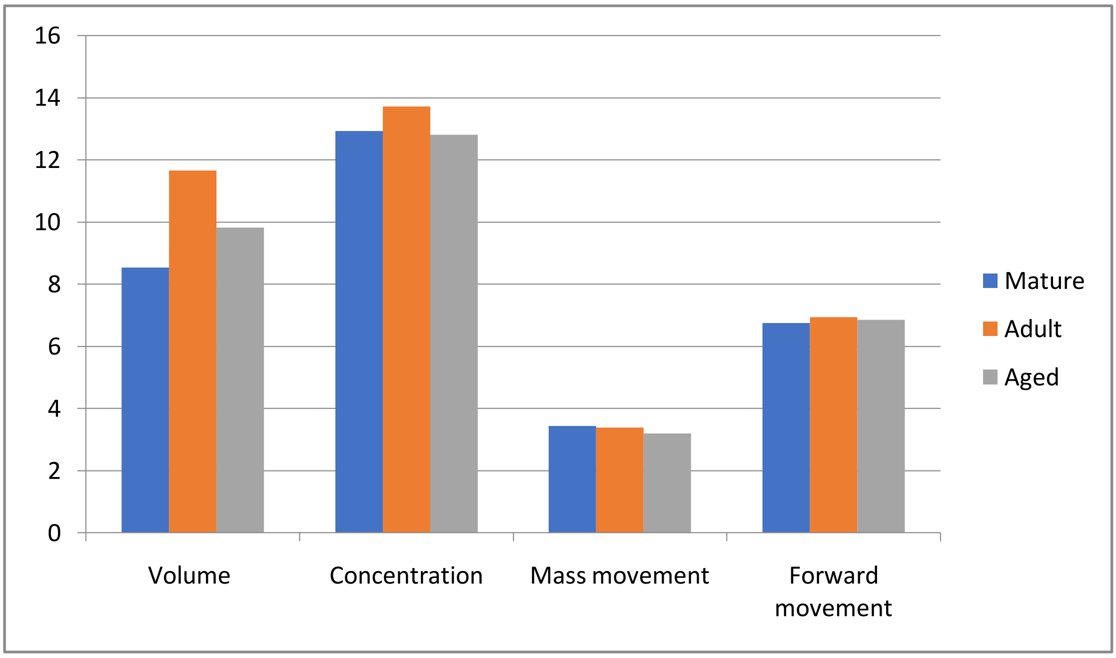


Figure 4.1. Semen quality of bulls from different age groups

4.1.2 Concentration of semen

Concentration of semen was found higher in the adult (1372.4±53.94) group followed by mature (1292.4±35.17) and aged group (1281.1±53.94) in the study (Table 4.1 and Figure 4.1). But the differences were not statistically significant ($p>0.05$).

The result is similar with the result of Asad *et al* (2004) and Bhakat *et al.* (2011) while the result is opposed by Brito *et al.* (2002a) who found no significant difference in different age groups in terms of concentration.

4.1.3 Mass movement of sperm

Mass movement of the semen showed a declining manner with increase of age. Most movement was recorded in mature group (3.43±0.03) followed by adult group (3.39±0.05). However the least movement was recorded in aged group (3.19±0.05) of bulls under the study (Table 4.1 and Figure 4.1). Mature and adult group showed statistically similar whereas aged group differed significantly from other two groups ($p<0.05$).

Asad *et al* (2004) and Bhakat *et al.* (2011) found similar result in their study.

4.1.4 Forward movement of sperm

The study revealed that, semen from bulls of adult age group had the highest forward movement (69.44±0.55%) followed by aged (68.52±0.55%) and mature (67.52±0.36%) age group (Table 4.1 and Figure 4.1). In terms of statistics, there was a significant difference between adult and mature age group whereas no difference with aged group ($p>0.05$).

Asad *et al* (2004) found that mean forward movement decreased with the increase of age. However, the highest forward movement found in the bulls of 4-6 years of age that supported the result of this study.

4.2 Effect of Body Weight on Semen Quality

All the bulls under the study were divided into three groups according to their body weight. Bulls of up to 500 kg were termed as light weight, 500-750 kg as medium and more than 750 kg were termed as heavy weight. According to this grouping, 64 ejaculates from 10 bulls were from light weight bulls whereas 117 ejaculates from 13 bulls were from medium and 54 ejaculates from 6 bulls were from heavy weight group of bulls.

Table 4.2. Semen quality of bulls from different body weight groups

Body weight group (kg)	**Observations**	**Volume (ml)**	**Concentration (million/ml)**	**Mass movement (grade)**	**Forward movement (%)**
		Mean±SE	**Mean±SE**	**Mean±SE**	**Mean±SE**
Light	64	$7.77^{b}\pm0.35$	$1382.1^{a}\pm49.37$	$3.52^{a}\pm0.05$	$67.97^{b}\pm0.51$
Medium	117	$10.39^{a}\pm0.26$	$1293.9^{a}\pm36.51$	$3.37^{b}\pm0.03$	$67.61^{b}\pm0.37$
Heavy	54	$9.83^{a}\pm0.38$	$1251.5^{a}\pm53.75$	$3.17^{c}\pm0.05$	$69.72^{a}\pm0.55$

Mean values in the same column with different superscripts (a, b) differ significantly at $p<0.05$.

4.2.1 Volume of semen

Among three body weight groups, semen from medium weight group showed the highest (10.39±0.26) volume of semen followed by heavy (9.83±0.38) and light (7.77±0.35) weight group (Table 4.2 and Figure 4.2). Volume of semen from medium and heavy weight bulls were found similar whereas light weight groups differed significantly from rest two groups ($p<0.05$).

The result is similar to the findings of Asad *et al,* (2004) who found the highest volume in bulls of 400-600 kg body weight. Gopinathan *et al.* (2018) found a positive correlation of body weight with semen volume.

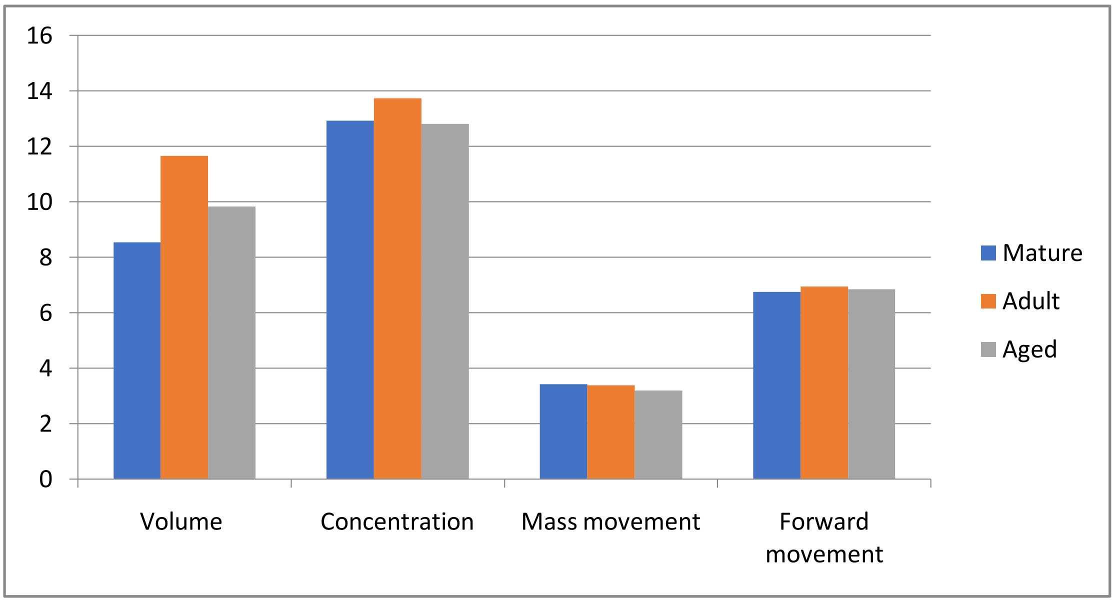


Figure 4.2. Semen quality of bulls from different body weight groups

4.2.2 Concentration of semen

Concentration of semen in light, medium and heavy weight groups of bulls were recorded as 1382.1±49.37, 1293.9±36.51 and 1251.5±53.75 chronologically (Table 4.2 and Figure 4.2). Numerically there were some difference but statistically they are similar (p>0.05).

The body weight was positively correlated (p<0.01) with sperm concentration in the study of Gopinathan *et al.* (2018) while Asad *et al,* (2004) found significant differences in different body weight groups in terms of sperm concentration.

4.2.3 Mass movement of sperm

There were significant differences in mass motility of semen in three weight groups. Motility decreased with weight. The highest motility found in light weight group (3.52±0.05) followed by medium (3.37±0.03) and heavy (3.17±0.05) weight bulls (Table 4.2 and Figure 4.2). The differences among three groups were statistically significant (p<0.05) and all three means were significantly different from one another.

The result is dissimilar with Gopinathan *et al.* (2018) and similar with Asad *et al,* (2004).

4.2.4 Forward movement of sperm

The highest forward movement was recorded in heavy weight group (69.72±0.55%) whereas lower in light weight group (67.97±0.51%) and the lowest in medium weight group (67.61±0.37%) as found in the study (Table 4.2 and Figure 4.2). The means of light and medium weight groups were statistically similar but different from heavy weight group (p<0.05). The result is similar with Asad *et al,* (2004).

4.3 Effect of Season on Semen Quality

The time period of the study were divided into three seasons to find out the effect of seasons on semen quality. Months between November and February were termed as winter, between March and June as summer and between July and October as rainy. According to this grouping, 78 ejaculates in each of winter and summer whereas 79 ejaculates in rainy season from all 26 bulls were collected for this study.

Table 4.3. Semen quality of bulls from different season groups

Seasons	**Observations**	**Volume (ml)**	**Concentration (million/ml)**	**Mass movement (grade)**	**Forward movement (%)**
		Mean±SE	**Mean±SE**	**Mean±SE**	**Mean±SE**
Winter	78	9.56^{a}±0.34	1276.0^{a}±44.72	3.38^{a}±0.04	68.33^{a}±0.47
Summer	78	9.95^{a}±0.34	1377.1^{a}±44.72	3.33^{a}±0.04	68.01^{a}±0.47
Rainy	79	9.14^{a}±0.34	1271.9^{a}±44.43	3.37^{a}±0.04	68.23^{a}±0.46

Mean values in the same column with different superscripts (a, b) differ significantly at p<0.05.

4.3.1 Volume of semen

There were no significant changes in volume of semen in different season. Numerically higher volume was found in winter season (9.56±0.34) followed by summer (9.95±0.34) and rainy (9.14±0.34) accordingly (Table 4.3 and Figure 4.3). But the means were statistically similar and there were no significant difference among the means (p>0.05).

The result is in agreement with Brito *et al*. (2002b) and Sarakul *et al*. (2017) whereas Asad *et al*. (2004) found maximum volume in summer and Bhakat *et al*. (2011) during rainy season. Fuerst-Waltl *et al.* (2006) also found a significant effect of temperature on ejaculate volume.

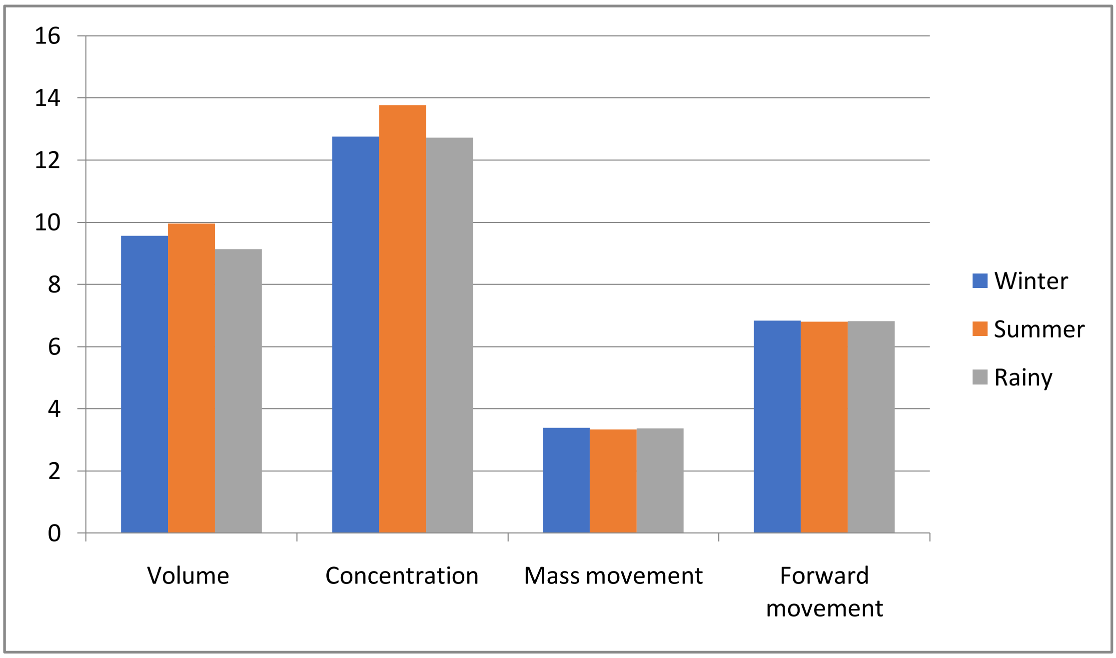


Figure 4.3. Semen quality of bulls from different season groups

4.3.2 Concentration of semen

Semen concentration was slightly higher in summer (1377.1±44.72) than winter (1276.0±44.72) whereas it was lowest in rainy season (1271.9±44.43) according to the result of the study (Table 4.3 and Figure 4.3). Statistically there were no significant difference among the means (p>0.05).

Brito *et al*. (2002b) found similar result and Bhakat *et al*. (2011) found the highest sperm concentration during rainy seasons but it was non-significant ($p>0.05$).

4.3.3 Mass movement of sperm

Numerically slightly higher mass movement of semen was found in winter (3.38±0.04) than rainy (3.37±0.04) and summer (3.33±0.04) season in current study (Table 4.3 and Figure 4.3). But the differences were not statistically significant ($p>0.05$).

Fonseca (1995), Brito *et al*. (2002b) and Bhakat *et al*. (2011) could not find any significant effect of season on mass motility while Fiaz *et al*. (2010) found lower movement during wet summer that supported the current result.

On the other hand, Usmani *et al*. (1993), Li-junjie *et al*. (2001), Fawzy and Rabie (1996) and Mostari *et al*. (2005) reported reduced motility during summer in Friesian.

4.3.4 Forward movement of sperm

Forward movement of semen showed more or less similar means in different seasons both numerically and statistically. Forward movement was recorded higher in winter (68.33±0.47%) followed by rainy (68.23±0.46%) and summer (68.01±0.47%) in the study (Table 4.3 and Figure 4.3). Statistically there were no significant difference among the means ($p>0.05$).

The result is similar to Fonseca (1995), Brito *et al*. (2002b) and Bhakat *et al*. (2011) who found no significant effect of season on forward movement while it is dissimilar to Asad *et al*. (2004) who found significantly lower forward movement in summer.

4.4 Effect of Age at Sexual Maturity on Semen Quality

Breeds of Indian subcontinent become sexually mature later than foreign breeds. Thus the bulls were divided into early (18-24 months) and late (24-36 months) maturity. According to this grouping, 153 ejaculates from 17 early matured bulls whereas 82 ejaculates from 9 late matured bulls were collected to study the effect of the age at sexual maturity on semen quality.

Table 4.4. Semen quality of bulls from different age at sexual maturity groups

Age at sexual maturity	**Observations**	**Volume (ml)**	**Concentration (million/ml)**	**Mass movement (grade)**	**Forward movement (%)**
		Mean±SE	**Mean±SE**	**Mean±SE**	**Mean±SE**
Early	153	10.15^{a}±0.23	1257.5^{b}±31.61	3.31^{b}±0.03	68.01^{a}±0.33
Late	82	8.43^{b}±0.32	1402.7^{a}±43.14	3.46^{a}±0.04	68.54^{a}±0.45

Mean values in the same column with different superscripts (a, b) differ significantly at $p<0.05$.

4.4.1 Volume of semen

Early matured (10.15±0.23) bull produced more semen in terms of volume than late matured (8.43±0.32). Imported and crossbred i.e. early matured bulls produced more semen (Table 4.4 and Figure 4.4). Statistically all two means were significantly different from one another ($p<0.05$).

The highest volume (12.01±0.22 mL) of semen was found in 3 years age of sexual maturity in the study of Asad *et al*. (2004) that is dissimilar to the current study.

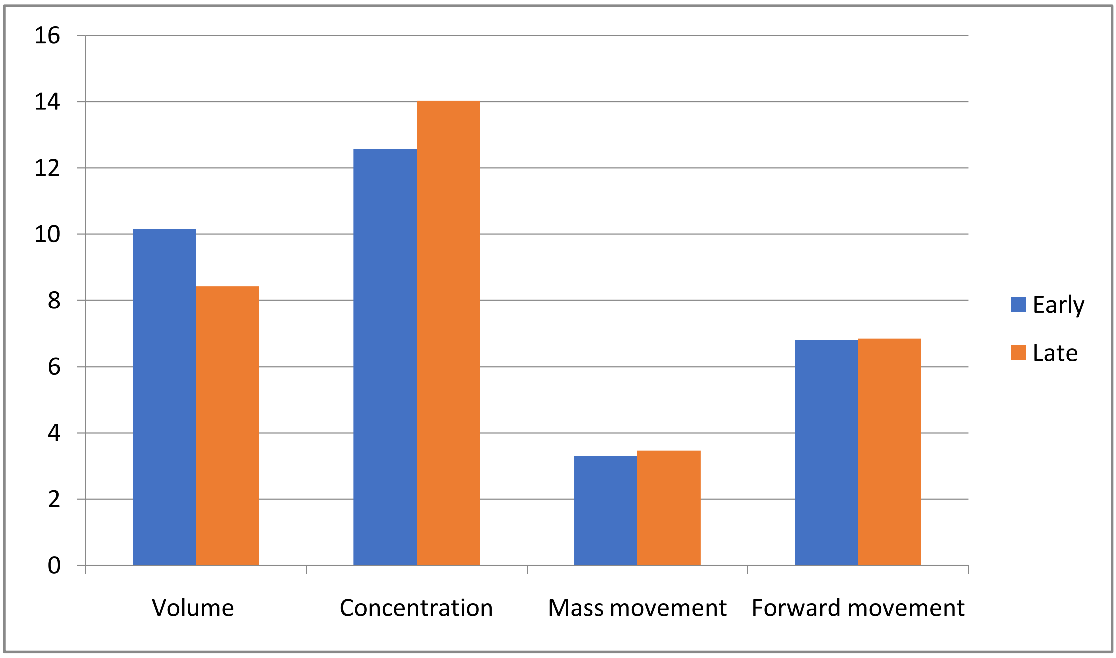


Figure 4.4. Semen quality of bulls from different age at sexual maturity groups

4.4.2 Concentration of semen

Semen concentration was higher in late matured bulls (1402.7±43.14) than that of early matured bulls (1257.5±31.61) according to the study (Table 4.4 and Figure 4.4). The differences between the means were statistically significant (p<0.05).

The result is similar with the result of Brito *et al*. (2002a) and Asad *et al*. (2004).

4.4.3 Mass movement of sperm

The study revealed that, mass movement of semen from native bull was more than imported bulls. Late matured bulls (3.46±0.04) produced semen with more mass movement than that of early matured bulls (3.31±0.03) in current study (Table 4.4 and Figure 4.4). Statistically all two means were significantly different from one another (p<0.05).

The result of the present study is in agreement with Asad *et al*. (2004) who reported the highest mass movement (4.40±0.08 grade) sperm in late matured bulls.

4.4.4 Forward movement of sperm

Numerically slightly higher forward movement was found in the semen of late matured bulls (68.54±0.45%) than that of early matured bulls (68.01±0.33%) under the study (Table 4.4 and Figure 4.4). However, they were not statistically different from one another (p<0.05).

Asad *et al*. (2004) also found the highest forward movement (58.74 ± 0.54%) of sperm in bulls of 3 years age of sexual maturity but it was not statistically significant. Thus the result is completely similar with the result of present study.

CHAPTER V
SUMMARY AND CONCLUSION

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The research work was done on the basis of data of semen quality of bulls collected from the record book of the Central Artificial Insemination Laboratory, in the Central Cattle Breeding and Dairy Farm (CCBDF), Savar, Dhaka Bangladesh. Non-genetic factors of semen quality were analyzed in different aspects from a total of 235 ejaculates from 26 bulls.

According to age, bulls under the study were divided into three groups i.e. mature (up to 72 months), adult (72-96 months) and aged (more than 96 months). According to this grouping, 127 ejaculates from 17 bulls were from mature bulls whereas 54 ejaculates from 6 bulls were from both adult and aged group of bulls.

Volume of semen differed significantly ($p<0.05$) among age groups of bulls. The highest volume of semen was found in adult bulls (11.65±0.37) followed by aged (9.82±0.37) and mature (8.54±0.24) age groups. Higher concentration of semen was found in the adult (1372.4±53.94) group followed by mature (1292.4±35.17) and aged group (1281.1±53.94) in the study. But the differences were not statistically significant ($p<0.05$). Mass movement was higher in mature group (3.43±0.03) followed by adult group (3.39±0.05). However the least movement was found in aged group (3.19±0.05) of bulls. Mature and adult group showed statistically similar whereas aged group differed significantly from other two groups ($p<0.05$). Semen from bulls of adult age group had the highest forward movement (69.44±0.55%) followed by aged (68.52±0.55%) and mature (67.52±0.36%) age group. In terms of statistics, there was a significant difference between adult and mature age group whereas no difference with aged group ($p>0.05$).

All the bulls under the study were divided into light (up to 500 kg), medium (500-750 kg) and heavy (more than 750 kg) according to their live weight.

According to this grouping, 64 ejaculates from 10 bulls were from light weight bulls whereas 117 ejaculates from 13 bulls were from medium and 54 ejaculates from 6 bulls were from heavy weight group of bulls.

Medium weight group showed the highest (10.39±0.26) volume of semen followed by heavy (9.83±0.38) and light (7.77±0.35) weight group. Volume of semen from medium and heavy weight bulls were found similar whereas light weight groups differed significantly from rest two groups ($p<0.05$). Statistically similar ($p<0.05$) concentration of semen were recorded in different weight groups. Numerically it was 1382.1±49.37, 1293.9±36.51 and 1251.5±53.75 in light, medium and heavy weight groups of bulls chronologically. Motility decreased with weight and it was statistically significant ($p<0.05$). The highest motility found in light weight group (3.52±0.05) followed by medium (3.37±0.03) and heavy (3.17±0.05) weight bulls. The highest forward movement was recorded in heavy weight group (69.72±0.55%) whereas lower in light weight group (67.97±0.51%) and the lowest in medium weight group (67.61±0.37%) as found in the study. The means of light and medium weight groups were statistically similar but different from heavy weight group ($p<0.05$).

The time period of the study was divided into three seasons; winter (November-February), summer (March-June) and rainy (July-October). According to this grouping, 78 ejaculates in each of winter and summer whereas 79 ejaculates in rainy season from all 26 bulls were collected for this study.

Statistically similar ($p<0.05$) traits (volume, concentration and movement) of semen were recorded in different season. Numerically higher volume was found in winter season (9.56±0.34) followed by summer (9.95±0.34) and rainy season (9.14±0.34) accordingly. Whereas, semen concentration was slightly higher in summer (1377.1±44.72) than winter (1276.0±44.72) and it was lowest in rainy season (1271.9±44.43). Higher mass movement of semen was found in winter (3.38±0.04) than rainy (3.37±0.04) and summer (3.33±0.04) season in current study. Forward movement was recorded higher in winter (68.33±0.47%) followed by rainy (68.23±0.46%) and summer (68.01±0.47%) season.

The bulls under the study were divided into early (18-24 months) and late (24-36 months) maturity. According to this grouping, 153 ejaculates from 17 early matured bulls whereas 82 ejaculates from 9 late matured bulls were collected to study the effect of the age at sexual maturity on semen quality.

Statistically different ($p<0.05$) traits (volume, concentration and movement) were found between early and late matured bulls except forward movement. Early matured imported and crossbred produced more semen (10.15±0.23) than local breeds of late maturity (8.43±0.32). But semen concentration and mass movement were higher in late matured bulls (1402.7±43.14 and 3.46±0.04) than that of early matured bulls (1257.5±31.61 and 3.31±0.03). On the other hand, numerically slightly higher forward movement was found in the semen of late matured bulls (68.54±0.45%) than that of early matured bulls (68.01±0.33%) under the study. However, they were not statistically different from one another ($p>0.05$).

At the end, we can conclude on above discussion that, adult (72-96 months) bulls having the body weight up to 500 kg (light) from late matured bulls will produce quality semen in terms of volume, concentration, mass and forward movement. Season have no significant difference in the quality of semen.

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