MACROSCOPIC, MICROSCOPIC AND BIO-CHEMICAL CHARACTERISTICS OF FRESH DOG SEMEN*

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ABSTRACT

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Forty semen ejaculates obtained from 10 healthy adult mongrel dogs by digital manipulation technique were examined to assess the macroscopic, microscopic and biochemical characteristics. In the sperm rich fraction, the volume (0.9 to 1.5 ml), colour (white to milky white), consistency of semen (thin to medium), overall mean sperm concentration (319.00 + 11.79 million per ml), pH (5.80 to 6.70), initial sperm motility (80 to 90 per cent), live spermatozoa (74 to 91 per cent), abnormal spermatozoa (7 to 10 per cent), intact acrosome (88 to 96 per cent) and hypo-osmotic swelling response (73 to 96 per cent) were recorded. The aspartate aminotransferase enzyme leakage into the seminal plasma ranged from 36 to 48 units per ml. The mean level of the aspartate aminotransferase recorded was 40.00 +1.11 units per ml of seminal plasma in dog semen.

Key words : Sperm motility, Sperm concentration, Acrosome integrity, HOST, Aspartate Aminotransferase, Dog semen.

INTRODUCTION

A pre-breeding examination of the semen sample should be performed to ensure the optimal health status of male dogs which are intended for breeding purpose. Semen samples should be evaluated based on the appearance, volume, concentration, motility, sperm morphology, membrane integrity, acrosomal integrity and enzyme level. These parameters are necessary not only to provide an accurate estimate of the reproductive performance of the stud dogs but are also essential for further processing of semen for cryopreservation. Hence, the present study was undertaken to evaluate the macroscopic, microscopic and biochemical characteristics of fresh dog semen.

MATERIALS AND METHODS

Semen samples were obtained twice a week from 10 mongrel dogs by digital manipulation technique as described by Allen (1991) with minor modifications. The pre-sperm, sperm-rich and post-sperm fractions were collected separately in clear, graduated semen collection cups. After discarding the pre-sperm and post-sperm fractions the sperm-rich fraction was transferred to a water bath kept at 37°C for further processing. Sufficient care was taken to ensure that the semen was not exposed to unfavourable conditions during and after collection. A total of 30 ejaculates were used for studying fresh semen characteristics and 10 ejaculates were utilized for aspartate aminotransferase enzyme leakage. Immediately after collection, the samples were evaluated macroscopically for volume, colour, consistency and pH and microscopically for motility, spermatozoal concentration, live and dead spermatozoa, sperm morphology, acrosomal integrity, HOST and aspartate aminotransferase enzyme leakage

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The sperm viability and acrosomal integrity were assessed by Supra vital triple staining technique as per Vazquez *et al.* (1992). The abnormal spermatozoal morphology percentage was assessed using three per cent Rose Bengal stain for a staining period of 15 minutes (Yubi *et al.*,1987). Hypo-osmotic swelling test (HOST) was done based on the technique described by England and Plummer, (1993). 0.1ml each from 10 fresh semen ejaculates was taken and centrifuged at 1500 rpm for 20 minutes. The supernatant fluid was collected into sterile tubes. The aspartate aminotransferase enzyme leakage was estimated by colorimetric method as per Reitman and Frankel (1957).

RESULTS AND DISCUSSION

The technique of digital manipulation was adopted to collect semen from experimental dogs. No difficulties were observed in this technique during semen collection. The quality and quantity of semen collected were found to be superior, as it prevented the mixing up of first and third fractions. Similar observations were reported by Allen (1991) and Kadirvel (1998).

The mean volume of second fraction of semen recorded in this study was 1.30 + 0.02 ml with a range from 0.9 to 1.5 ml. Similarly, England and Allen (1989) recorded a mean of volume of 1.2 + 0.7 ml. Allen (1991) reported that the volume of second fraction was within the range of 0.5 - 3.5 ml. Dobrinski *et al.*(1993) recorded a higher volume of 2.2 + 0.2 ml for the sperm-rich second fraction of different breeds of dogs. They stated that the higher volume might be due to the variation in the breeds.

The colour of the second fraction varied from white to milky white. The colour observed in this was in agreement with Allen (1991), Daiwadnya *et al.* (1995) and Kadirvel (1998).

The consistency of second fraction of semen in this study was thin to medium. This observation was in agreement with Nair *et al.* (1999) who recorded a thin to medium consistency for mongrel dog semen. In the present study 83.33 per cent of samples were medium in consistency while 16.67 per cent were thin. Daiwadnya *et al.* (1995) in their studies reported only thin consistency. More of medium consistency obtained in this study might be due to the collection of three fractions of semen separately as explained by Christiansen (1984).

The overall mean concentration of spermatozoa in the study was 319.00 + 11.79 millions per ml. Similar finding was made by Nair *et al.* (1999) who recorded a concentration of 313 + 30.74 millions per ml. The higher concentration in this study might be due to strict measures taken to prevent the dilution of the second fraction with other fractions.

The pH of semen in the study ranged from 5.80 to 6.70 with a mean of 6.37 + 0.03. This pH was within the range reported by Choudhary and Dubay (1974), who recorded an average pH of 6.47 + 0.35 for the second sperm-rich fraction.

Initial sperm motility in this study ranged from 80 to 90 per cent with a mean of 84.00 + 0.91 per cent. It was in accordance with the report of England and Allen (1989). However a lower motility of 79 per cent was recorded by Olson (1992) for various breeds of dogs.

The mean live sperm percentage recorded was 85.67 + 0.81 with a range of 74 to 91 per cent. In the present study, the mean value was slightly higher when compared to 82.81 + 0.93 per cent reported by Daiwadnya *et al.* (1995) for mongrel dogs. The live sperm percentage range was in accordance with the value of 62 to 90 per cent reported by England and Allen (1991) for different breeds of dogs.

In this study the mean value of abnormal sperm morphology was 8.33 + 3.16 per cent (range : 7 to 10 per cent) which was lower when compared to 10.8 + 0.95 per cent recorded for mongrel dogs by Daiwadnya *et al.* (1995). In the present study the predominant type of abnormal spermatozoa noticed included detached heads and tails, presence of proximal and distal protoplasmic-droplets, bent mid-piece, coiled tails, double heads, double tails and bent tails. Similar types of abnormalities were reported by Yubi *et al.*(1987).

The mean value for intact acrosome in this study was 92.00 + 0.28 per cent (88 to 96 per cent) which was comparable to the findings of Nair *et al.(*1999).

The average value for the hypo-osmotic swelling response in fresh semen was 91.30 + 0.92 per cent with a range of 73-96 per cent and was in agreement with the finding by Nair *et al. (*1999) in fresh semen of mongrel dogs.

The enzyme leakage in fresh semen recorded in this study was 40 + 1.11 units per ml which was lower than the value for Beagles dogs recorded by James and Heywood (1979). The aspartate aminotrasferase enzyme leakage into the seminal plasma ranged from 36 to 48 units per ml. The lower value obtained in the present study might be attributed to breed differences.

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