

# Studies on different physical characteristics of mongrel dog semen

P. Kumar<sup>1</sup>, A. Kumar<sup>2\*</sup>, C.S. Azad<sup>1</sup>, S.K. Sheetal<sup>1</sup>

<sup>1</sup>Department of Veterinary Gynaecology and Obstetrics, <sup>2</sup>Department of Veterinary Clinical Complex, Bihar Veterinary College, Bihar Animal Sciences University, Patna

\*Corresponding author email ID: dr43ankesh@gmail.com

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## Abstract

Very little experimental work has been done on semen preservation and artificial insemination (AI) in canine in India. Dogs are neither used for production of meat or neither milk nor they are apparently able to boost up the rural economy and for that reason scientist and researcher always ignored works on dogs. Nowadays there is growing interest in dog breeders in breeding with pure and registered breed dogs. Dog breeding is a multimillion-dollar industry worldwide and lack of successful breeding is generally multifactorial. Thus, the dogs are gradually becoming one of the biggest income generating animals for dog lovers and dog breeders. Keeping in views of the above mention facts the present study was done with aim to focus first to study the physical characteristic of mongrel dog semen. In the present study, 3 mongrel dogs were taken for semen collection. Total of 18 ejaculates were collected, 6 from each dog by digital manipulation. The color of semen samples collected from mongrel dogs was observed as milky white to thin milky. The Mean  $\pm$  SEM volume, pH, mass motility and sperm concentration were  $3.55 \pm 0.31$  ml,  $6.56 \pm 0.03$ ,  $3.75 \pm 0.12$ ,  $307 \pm 19.38 \times 10^6$  per ml respectively.

**Key words:** Mongrel dog; Semen; temperature; digital manipulation

## Introduction

Since Stone Age, dogs (*Canis familiaris*) are the companion animals and surprisingly the first animal species domesticated because of its faithfulness and excellent hunting qualities. With progress of industrialization from agriculture, change in social life from joint family to nuclear family there is increased trend to keeping pets as companion animals and also dogs are well known for their honesty, because of these qualities of the dogs attracted the human for their domestication. It is known that dogs are of much importance not only to growing children but also they render physiological benefits to adults. Dog is reliable, affectionate having a sense of routine life. In recent times dog is used for racing, sports, watch dog, blind persons guide and number of other purposes that suit the characteristics of a particular breed. Apart from psychological benefits, people are also kept dogs to protect their property or to guard as this animal can protect human being at the cost of their life.

Very little experimental work has been done on semen preservation and artificial insemination (AI) in canine in India. Dogs are neither used for production of meat or neither milk nor they are apparently able to boost up the rural economy and for that reason scientist and researcher always ignored works on dogs. Nowadays there is growing interest in dog breeders in breeding with pure and registered breed dogs. Dog breeding is a multimillion-dollar industry worldwide and lack of successful breeding is generally multifactorial. Thus, the dogs are gradually becoming one of the biggest income generating animals for dog lovers and dog breeders. In dog breeder, natural mating is used most commonly for conception but many a time there is problem in transportation of animal to long distance due to shyness of male animal, differences in size of male and female animal, moreover, inability of male animal to mount because of muscular skeletal diseases or other congenital conditions, non-availability of stud male at proper mating time result in loss of valuable breeding season. To overcome the above obstacles artificial insemination (AI) can be used as an alternative. So for artificial insemination (AI) we need to preserve the semen at refrigerating temperature (4 0 C) or freezing temperature (-196 0 C). To get optimum quality dog breed, pure breed with physical characteristics and its offspring, AI is most important technique. For successful application of AI technique, the most important step is preservation of semen. In case of dogs, the semen can be preserved at chilling (4<sup>0</sup>C) and freezing temperature (-196<sup>0</sup>C). Due to the scanty reports of evaluation and preservation of mongrel dog semen, the present study was focussed first to study the physical characteristic of mongrel dog semen. Microscopic evaluation of sperm motility, morphology, acrosomal integrity and hypo-osmotic swelling test results of the present study have already previously reported (Kumar *et al.*, 2020).

## Material and Method

### Selection of experimental animals

Experimental animals include 3 (Three) mongrel dogs of more than one year of age with normal libido having average body weight of 20- 30 kg. The dog had housed in Kennel of Bihar Veterinary College, Bihar Animal Sciences University Patna and were kept on uniform diet (Two times in day with ad lib access to drinking water) and management during the entire study. All animals were dewormed orally with Tab Skyworm<sup>R</sup> (1 tab per 10 kg b.wt) and also vaccinated for Rabies (Rabigen Mono<sup>R</sup> 1 ml s/c) 15 days before the start of the experiment.

Semen from each dog was collected by digital manipulation method without harming or giving stress to the animal in a calibrated plastic vial connected with funnel after proper cleaning of prepuce and perineal region with antiseptic solution following the method described by Christiansen (1984) with slight modifications with using an estrous bitch as a teaser.

### Method of Semen Collection

Dog semen is generally collected by digital manipulation and stimulation of the bulbus glandis. Semen collection process can be started by massage of the dog prepuce at the level of the bulbus glandis until partial erection developed, followed by the quick retraction of the prepuce and penile exposure. A circular, fixed pressure was maintained with the left hand to simulate the copulatory lock or tie. When a crystal clear prostatic fluid begins to flow into the collection tube, a circumferential pressure was applied posterior to the bulbus glandis till ejaculation complete. The pre-sperm, sperm rich and post-sperm fractions of semen were collected into a single 15 ml graduated transparent centrifuge tube attached to the funnel.

### Samples collection

All three dogs were trained for semen collection by mechanical stimulation. Semen was collected twice a week from all the 3 mongrel dogs by digital manipulation in the morning hours. A total of 18 ejaculates were collected out of which 6 (Six) ejaculate from each dog were collected.

### Evaluation of semen

After semen collection, each individual semen sample was immediately assessed for volume, colour, pH and then transfer immediately to the laboratory and stored at 37°C in a water bath for further evaluation of seminal attributes.

#### Colour

Colour of semen ejaculates was observed/ recorded by visual observation immediately after collection in graduated transparent centrifuge tube in natural day light and recorded as watery, milky white and watery clear. Deviation from normal if any was also recorded.

#### Volume (ml)

Immediately after semen collection the total ejaculate volume (ml) obtained from each collection was recorded directly from the transparent graduated centrifuge tube.

#### pH

pH of each semen sample was recorded after collection using pH paper.

#### Mass motility

Mass motility of each semen ejaculate was assessed immediately after the collection on a scale of 0 to 5 by Herman and Madden (1953). A drop of neat semen was spread uniformly on a clean dry glass slide and observed without cover slip under low power magnification of microscope (10 x).

#### Sperm concentration

Sperm concentration was determined with the help of a Neubauer hemocytometer after a dilution with a diluting fluid and expressed in million per milliliter of semen. Composition of diluting fluid: Eosin yellow- 0.05 gram, Sodium chloride (NaCl)- 1.00 gram, Formalin - 1ml, distilled water add up to 100 ml. Diluting fluid should be capable of killing and dispersing the sperm cells evenly.



**Fig 1.** Stimulation of bulbus glandis



**Fig 2** Collection of semen by massaging of bulbus glandis

## Results and Discussion

The different physical characteristics of semen observed during this study are presented in table - 1.

#### Colour

The colour of majority of semen samples (N=18) collected from mongrel dogs in present study was observed as milky white (N=13) and remaining samples (N= 05) was thin milky.

Silva *et al.* (2006) and Kadirvel (1998) reported milky white color of dog fresh semen in his study which supports our findings. The colour of the semen was clear and watery in the first fraction and milky white and opaque in the sperm-rich fraction observed by Zorinkimi *et al.* (2017b) in 3 mongrel dogs during different

ejaculates. The variation in the colour of different ejaculate of semen is due to different spermatozoa concentrations in the semen.

**Table 1:** The mean  $\pm$  S. E.M of Physical characteristics of semen in dogs (N=18)

S. No	Parameters	Observations
1	Volume (ml)	3.55 $\pm$ 0.31
2	pH	6.56 $\pm$ 0.03
3	Mass motility (Scale 0-5)	3.75 $\pm$ 0.12
4	Sperm concentration (10 <sup>6</sup> /ml)	307 $\pm$ 19.38

### Volume

The mean volume of the semen samples (N=18) collected from mongrel dogs in this study was presented in the table 1. The mean volume of semen collected from mongrel dogs in this study was 3.55 $\pm$ 0.31 ml and ranges from 2.0 ml to 7.0 ml in a single ejaculation.

The present results corroborated with the finding of Daiwadnya *et al.* (1995) who reported that the average semen volume to be 2.57  $\pm$ 0.14 ml collected from 4 mongrel dogs at 3 days interval in 48 ejaculates. However, Zorinkimi *et al.* (2017b) collected 42 ejaculates of semen by digital manipulation from 3 mongrel dogs and observed the over-all mean volume of first and second fractions of the ejaculate in the study was 2.95 $\pm$ 0.41 which is slight lower than our observed value. Kadirvel (1998) observed 36 ejaculates from 6 mongrel dogs the mean total volume of ejaculate was recorded 6.15 $\pm$ 0.75 with a range of 2.2 to 10 ml which is higher than our observed value.

These variations in the semen volume have been due to variation in size of dog, age, body weight, and breeds of dog, size of prostate gland, frequency of semen collection, volume of 3<sup>rd</sup> fraction collection (Dubiel 1975).

### pH

The mean value of pH of the semen samples (N=18) collected from mongrel dogs in this study was presented in the table 1. The mean pH value of mongrel dog semen was observed 6.56 $\pm$ 0.03 with a range from 6.4 to 6.8.

pH value of semen in the present study was similar to the values reported by Daiwadnya *et al.* (1995) and Zorinkimi *et al.* (2017b) who reported pH values of 6.57 $\pm$  0.04 and 6.59  $\pm$  0.76 in mongrel dog. Variation in the pH of dog semen (6.23- 7.01) has been reported due to the different method of semen collection (with an artificial vagina without a teaser, by hand manipulation with a teaser and by hand manipulation without a teaser in dogs (Boucher *et al.*, 1958).

### Mass motility

The mean value of mass motility of the semen samples (N=18) collected from mongrel dogs in this study was presented in the table 1. The mass motility in the fresh semen immediately after collection from dog showed tremendous activity and a peculiar wave movement. The mean mass motility value of mongrel dog semen was observed to be 3.75 $\pm$ 0.12 on a scale of 0 to 5 and it ranges from 3.0 to 4.5.

Average mass activity as observed in the present study was higher with the earlier findings of Daiwadnya and Hukeri (1993) who recorded 2.05 mass activity in mongrel dog semen. Daiwadnya *et al.* (1995) collected 48 ejaculates from 4 mongrel dogs at 3 days interval. They reported average mass activity of semen sample to be 2.05  $\pm$ 0.10 and initial motility to be 3.60 $\pm$  0.09 on a grade of 0 to 5.

### Sperm concentration

The mean value of sperm concentration of the semen samples (N=18) collected from mongrel dogs in this study was presented in the table 1. The mean concentration of spermatozoa calculated in this study was 307 $\pm$ 19.38 x 10<sup>6</sup>/ml with the range of 210 to 574x 10<sup>6</sup> spermatozoa /ml.

The present findings are in agreement with the findings of Zorinkimi *et al.* (2017b) who reported the sperm concentration of 42 ejaculates of semen by digital manipulation from 3 mongrel dogs and observed the over-all mean concentration of spermatozoa was 298.73  $\pm$  35.46 million per ml. However, Daiwadnya *et al.* (1995) reported average sperm concentration in mongrel dogs to be 264 $\pm$  15 million/ml, which is lower than present findings. Biswas (2010) observed from 3 dogs of Alsatian breed the mean of sperm concentration of ejaculate was 274.33 $\pm$ 2.66 million per ml. The number of spermatozoa per ejaculate also varies according to age, testicular weight, sexual activity and the size of the dog (Amann 1986).

### Summary and Conclusion

The colour of semen samples collected from mongrel dogs was observed as milky white to thin milky. The Mean  $\pm$ S.E.M volume, pH, mass motility and sperm concentration were  $3.55\pm 0.31$  ml,  $6.56\pm 0.03$ ,  $3.75\pm 0.12$ ,  $307\pm 19.38\times 10^6$  per ml respectively.

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