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RESEARCH ARTICLE

ESTROUS CYCLE IN FEMALE MUDHOL HOUND DOGS-AN INDIGENOUS BREED

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INTRODUCTION

The Mudhol Hound breed of dogs is the indigenous breed of dogs found in and around Mudhol taluka of Bagalkot district, Karnataka. These dogs have not been selected for any particular purpose and therefore their reproductive potential have not yet been harnessed. The reproductive potential of a particular breed of animal is measured by considering its reproductive parameters and efficiency.

In the recent, though the efforts are going on assessment of desi canine breeds both at phenotypic as well as molecular level, again those studies are limiting majorly to Rajapalayam, Kanni and Chippiparai dog breeds in Tamilnadu. But as such no such systematic efforts have been carried out to establish reproductive parameters, reproductive efficiency parameters in Mudhol Hound breed of dogs. As a part of standardizing, improve and optimize breeding, increased knowledge regarding factors which might influence litter size is essential. The aim of this cross-sectional study was to record the stages and duration of proestrus, estrus and inter estrus interval in purebred female Mudhol Hound dogs

MATERIALS AND METHODS

The present study was conducted in Mudhol Hound female dogs belonging to Canine Research and Information Centre (CRIC), Timmapur, Mudhol and to the farmers in and around Bagalkot district. CRIC (MH), Timmapur, Mudhol was established by Karnataka Veterinary Animal and Fisheries Sciences University (KVAFSU), Bidar with the primary objective of popularizing Mudhol breed of dog and to provide technical services to Mudhol dog breeders. In addition CRIC (MH), Timmapur, also breeds the pure bred Mudhol Hound dogs and distributed the puppies to SC-ST farmers free of cost under SCP-TSP project entitled "Livelihood security through Mudhol Hound dog rearing".

Mudhol is a taluka place located in Bagalkot district of Karnataka which is the northern part of state. It is positioned between 15° 46' and 16° 46' North latitude and 79°59' and 76°20' East longitude.

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The most elevated position of the district lies between 450 to 800 meters above sea level and extends over an area of 6593 Sq. Km. The climate of Bagalkot district is warm and dry throughout the year and the rain fall is scarce.. Annual rainfall of the district varies from 562 to 978 to and 41.30°C

This present study was conducted in two phases. The first phase of the study was aimed to record the reproductive efficiency parameters of Mudhol dogs maintained on nutritionally balanced diet. The second phase was designed to study the reproductive parameters in Mudhol dogs are influenced by feeding of a homemade diet.

Duration of proestrus and estrus

During the first estrus after puberty, the duration of pro-estrus as well as estrus during first heat after attainment of puberty was determined to assess whether there were any differences between duration of pro-estrus and estrus at puberty and those during subsequent estrus.

Following onset of proestrual bleeding, the animals were examined by vaginal exfoliative cytology technique to record the duration of pro-estrous and estrous in individual animals. For this purpose, a small amount of sterile normal saline was drawn into bovine AI sheath attached to 2 ml syringe. The AI sheath containing sterile normal saline was gently advanced to the anterior vaginal canal and expelled. The succetion force was applied to draw vaginal sample into the AI sheath. The AI sheath was then withdrawn and contents were slowly expelled on to a clean greece free slide. The access fluid was drained off by tilting slide and slide was allowed to dry at room temperature. The air dried smear was then fixed with methanol for 2 minutes followed by staining with Giemsa stain for 30 minutes. Subsequently, the slide was washed under running water; air dried and examined first under 40x and then under 100x to determine the stage of cycle. The determination of stage of cycle was based on identification of the type of cells and proportion of each type of cell. The identification of different type of cells by vaginal smear was made on the basis of following criteria.

1. **Parabasal cells:** Round or oval cells with a large vesicular nucleus and somewhat dark stained cytoplasm were considered as parabasal cells.

2. **Intermediate Cells:** Intermediate cells varied in size and shape, but typically had a diameter two to three times more than that of parabasal cells.
3. **Superficial cells:** Superficial cells were the largest cells seen on a vaginal smear. They were polygonal in shape and distinctly flat, sometimes having the appearance of being rolled up. Their nucleus were either absent or pyknotic (very small and dark). Superficial cells without nucleus are often referred to as being "fully cornified".

The proportion of each type of cell was studied by counting a minimum of 200 cells and expressed in per centage. The study of vaginal exfoliative cytology procedure was first carried out on first day of proestrous bleeding and was repeated every second day until all the cells examined were superficial cells. The interval between first day of proestrous bleeding and first day on which 100 per cent superficial cells were recorded as duration of pro-estrus.

The first day on which 100% superficial cells were seen was determined as first day of estrus and subsequent vaginal exfoliative cytological examination were carried out every second day from day of onset of estrus and the proportion of cornified cells were determined during each examination. The first day on which the proportion of superficial cells decreases from 100 per cent to atleast 90 per cent was recorded as first day of onset of diestrus and the duration of estrus was determined as the interval between the first day of estrus to last of before the reduction in the number of superficial cells from 100 to 90 per cent. Similarly, the duration of proestrus and estrus was period subsequent to pubertal proestrus and estrus period.

Interval between estrus periods

The time taken for onset of second estrus after attaining puberty was recorded and the interval between first day of pubertal estrus and first day of subsequent estrus was calculated to determine interval between estrus periods

RESULTS

The present study was conducted with the primary objective of determining, if the reproductive efficiency parameters in Mudhol Hound dogs are influenced by the type of diet on which they are maintained. The two methods of feeding systems compared were, feeding of nutritionally balanced dog food and feeding of homemade dog food. The influence of the type of diet fed to dog on the reproductive efficiency was assessed in terms of exhibition pubertal estrus, the duration of pubertal estrus and estrus, inter estrus interval, duration of pro-estrus and estrus during subsequent estrus periods

Duration of pro-estrus and estrus in Mudhol Hound female dogs maintained on nutritionally balanced diet and on homemade diet

The mean duration of proestrus in 30 dogs maintained on nutritionally balanced food was recorded as 9.53 ± 0.31 days and ranged between 7 to 13 days (Table 1).

Table 1 Duration of pubertal pro-estrus and second pro-estrus (days) in Mudhol Hound female dogs maintained on nutritionally balanced diet and on homemade diet

| Sl. No | System of feeding and management | Pubertal Proestrus (Days) | | Second proestrus (Days) | |
|--------|----------------------------------|---------------------------|--------------|-------------------------|--------------|
| | | Mean \pm SE (Days) | Range (Days) | Mean \pm SE (Days) | Range (Days) |
| 1 | Nutritionally Balanced food | 9.53 ± 0.31 a | 07 to 13 | 9.23 ± 0.21 a | 07 to 12 |
| 2 | Homemade diet (n=30) | 8.93 ± 0.32 a | 07 to 12 | 9.73 ± 0.34 a | 07 to 13 |
| | Overall (N=60) | 9.22 ± 0.32 | 07 to 13 | 9.48 ± 0.28 | 07 to 13 |

*Mean values bearing similar superscripts along the column are not significantly different (p<0.05).

Its duration in dogs maintained on homemade diet was 8.93 ± 0.23 days and was not significantly different from mean duration of proestrus recorded in animals maintained on nutritionally balanced food.

Table 1 also presents the mean duration of proestrus in animals maintained on two different systems of management. In dogs maintained on nutritionally balanced food, the mean duration of pubertal estrus was recorded as 7.40 ± 0.57 days and ranged between 5 to 9 days. Further, the mean duration of pubertal estrus in dogs maintained on homemade diet did not differ significantly from the mean duration recorded in animals maintained on nutritionally balanced Diet, the mean duration and range of pubertal estrus being 8.27 ± 0.46 and 6 to 10 respectively (Table 2).

Table 2 Duration of pubertal estrus and second estrus (days) in Mudhol Hound female dogs maintained on nutritionally balanced diet and on homemade diet

| Sl. No | System of feeding and management | Pubertal Estrus (Days) | | Second Estrus (Days) | |
|--------|------------------------------------|------------------------|--------------|----------------------|--------------|
| | | Mean \pm SE (Days) | Range (Days) | Mean \pm SE (Days) | Range (Days) |
| 1 | Nutritionally Balanced food (n=30) | 7.83 ± 0.31 a | 6 to 10 | 7.40 ± 0.57 a | 5 to 9 |
| 2 | Homemade diet (n=30) | 8.23 ± 0.32 a | 07 to 11 | 8.27 ± 0.46 a | 6 to 10 |
| | Overall (N=60) | 8.03 ± 0.31 | 06 to 11 | 7.84 ± 0.51 | 5 to 10 |

*Mean values bearing similar superscripts along the column are not significantly different (p>0.05)

Interval between pubertal estrus and subsequent estrus period of Mudhol Hound female dogs maintained on nutritionally balanced diet and on homemade diet.

In the present study, the interval between pubertal estrus and subsequent estrus was significantly shorter in dogs maintained on nutritionally balanced food as compared to the inter estrus interval of animals maintained on homemade diet (275.67 ± 10.65 v/s 306.03 ± 7.14). The inter estrus interval in animals maintained on nutritionally balanced food ranged between 193 to 401 days while, it was 222 to 423 days in animals maintained on homemade diet (Table 3). 193 to 401 days and the mean inter estrus interval was recorded as 275.67 ± 10.65 days.

DISCUSSION

The Mudhol Hound dog is the only breed from Karnataka recognized so far by Kennel Club of India. Even though, it is known as a hunting dog, it is now also used as companion and guard dogs. Its morphological features have been well

established; however, its normal reproductive parameters are not known. This knowledge is essential to achieve a proper reproductive management. The present study is perhaps first comprehensive study on establishing the normal reproductive parameters for any Indian breeds of dogs.

Duration of Pro-estrus and estrus in Mudhol Hound female dogs maintained on nutritionally balanced diet and on homemade diet

In dogs pro-estrus has been defined as that stage of reproductive cycle prior to estrus when bitch attract the male, but not responsive to the mating (Olson and Nett, 1986). Cytological pro-estrus has been defined as period between first day of onset of pro-estrial bleeding to the day on which 100% cornification is seen on study of vaginal exfoliative cytology (Suyash Vardan Bhal, 2002). The second criterion was also used in the present study to define the duration of proestrus. In 60 dogs, irrespective of system of feeding on which Mudhol Hound female dogs were maintained, the average duration pubertal proestrus recorded as 9.22 ± 0.79 days and individual animal it ranged between 07 to 13 days. Further, the duration of proestrus did not differ between groups of dogs maintained on either nutritionally balanced diet or homemade diet.

The average duration of proestrus recorded in the present study is in agreement with duration of proestrus reported (Suyash Vardan Bhal, 2002). The average duration pubertal proestrus is also stated to be 09 days, ranging from 0 to 27 days (Christe and Bell, 1971; Allein and England, 1990). However, Chakrabarthy *et al.* (1980) reported range of proestrus to be 0 to 17 days. Further one study (Chakrabarthy *et al.*, 1980) also suggested that the average length of proestrus shorter for pubertal than mature bitches, but subsequent investigation (Wildt *et al.*, 1981) failed to substantiate this finding.

The present study revealed that duration of proestrus of Mudhol Hound female dogs similar to those observed in other breeds of dogs. Although significant individual variations did occur regarding duration of pro-estrus, it was also interesting to note that the duration of cytological proestrus was not influenced by the system of feeding. Comparable studies on the effect of plane of nutrition on duration of proestrus in dog are not available.

In the present study duration of estrus was defined as interval from first day of which 100% cornification was seen on study of vaginal exfoliative cytology to the first day of diestrus. The mean duration of estrus was recorded as 7.88 ± 0.49 days and the system of feeding did not seem to influence on duration of estrus. The period of estrus is reported to normally lose for about 10 days but, can range from 3 to 21 days (Olson and Nett, 1986). Bell and Christie, (1971) also reported that the average duration of estrus, based on behavioural signs is 9 days, ranging from 4 to 24 days. The mean duration of pubertal estrus was recorded in the present study is within the range of estrus reported in earlier studies.

The duration of proestrus and estrus during subsequent estrus also did not differ significantly when compared with pubertal pro-estrus and estrus (Table 2). However, it has been stated that the duration of proestrus and estrus may differ among pubertal and mature bitches (Johnston *et al.*, 1980), which could not be confirmed in the present study. Further, pubertal bitches are reported to be less likely than mature bitches to demonstrate estrus behavior even when ovulation occurs (Wildt *et al.*, 1989). Additionally, the duration of proestrus and estrus may be shorter, reduced in inconsistent pattern of

circulating progesterone have been reported to occur in some pubertal cycles (Wildt *et al.*,1981; Chakraborty *et al.*, 1980) which again was not observed in the present study. The estrous cycle of pubertal bitch is also reported to differ from those of mature animals in that, pubertal bitches are more likely to manifest a split heat or false heat (Jochle and Andersen, 1977). In the present study, the estrus phase at puberty of all 30 bitches progressed in diestrus as revealed by the vaginal exfoliative cytology. These observations suggest that the duration and characters of pubertal proestrus and estrus of Mudhol Hound bitches is similar to the subsequent proestrus and estrus and that the incidence of split heat either nil or extremely low. The results of the present study also suggest that system of feeding has little influence on duration of pubertal and subsequent pro-estrus and estrus.

Interval between pubertal estrus and subsequent estrus period (days) in Mudhol Hound female dogs maintained on nutritionally balanced diet and on homemade diet

In the present study, the mean inter estrus interval of 60 Mudhol Hound female dogs irrespective of system of feeding was recorded as 292.85 ± 7.67 days and the interval between first day of pubertal proestrus and first day of subsequent cycle was significantly shorter (275.67 ± 10.65 days) in animals maintained on nutritionally balanced food as compared to inter estrus interval recorded in animals maintained on homemade diet (Table 3).

Table 6 Interval between pubertal estrus and subsequent estrus period (days) in Mudhol Hound female dogs maintained on nutritionally balanced diet and on homemade diet

| Sl. No | System of feeding | Inter estrus interval period (Days) | |
|--------|------------------------------------|--------------------------------------|--------------|
| | | Mean± SE (Days) | Range (Days) |
| 1 | Nutritionally Balanced food (n=30) | 275.67 ± 10.65a | 193 to 401 |
| 2 | Homemade diet (n=30) | 306.03 ± 7.14b | 222 to 423 |
| | Overall (N=60) | 290.85± 9.23 | 193 to 423 |

*The mean values with different superscripts are significantly different (p<0.05)

The dog is usually considered as a monoestrus animal and has two estrus cycles a year although some small breeds may have 3 – 4 and large breeds only one (Roberts, 1971). Anderson *et al.*, (1962) reported that in beagles estrus occurred at every 7 months ±2 Months and Burns and Frazer (1966) stated that the grey Hounds and other larger breeds have one estrus period a year. Roberts (1980) also stated that there is evidence that frequency of estrus is genetically conditioned. Burns and Frazer (1966) quoted first in reporting that in nearly 500 bitches, 338 had an inter-estrus interval of 5- 7 Months, over 100 had interval of over 7 months and only 16 had an interval of less than 5 months. The results of the present study suggests that the average inter estrus interval in Mudhol Hound female dogs is around 9 months and is slightly larger than reported range of 5 to 7 months. This is probably because Mudhol Hound dogs have been developed for hunting purpose and Hounds in general have longer inter estrus interval.

The present study also found the inter estrus interval could be shortened by at least 30 days when maintained on nutritionally balanced food. This observation suggests a definite influence of nutrition on shortening of anestrus period through its stimulatory effects on Hypo-thalamo pituitary axis.

Summary

The mean duration of proestrus and estrus of 60 Mudhol Hound female dogs was determined as 9.22 ± 0.79 and 9.48 ± 0.28 and ranged between 7 to 13 days. The duration of proestrus and estrus of Mudhol Hound Female dogs was found to be similar to those reported in other breeds of dogs. It was also observed that the frequency of occurrence of pubertal estrus during hotter months of the year was very low suggestive of influence of estrus.

The duration of pubertal pro estrus and estrus was similar to those observed during estrus subsequent to pubertal estrus and type of nutrition did not seem to influence the duration and character of pro estrus and estrus

The inter estrus interval was recorded as 292.85 ± 7.67 days irrespective of system of feeding. and the interval between first day of pubertal proestrus and first day of subsequent cycle was significantly shorter (275.67 ± 10.65 days) in animals maintained on nutritionally balanced food as compared to inter estrus interval recorded in animals maintained on homemade diet.

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