Original Research Article

Epidemiological Studies on Canine Microfilariosis due to *Dirofilaria repens* in and around Mangalore- a Coastal Region of Karnataka

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**ABSTRACT**

A study was conducted to ascertain the epidemiology of canine microfilariosis in and around Mangalore a coastal region of Karnataka for a period of one year from March-2018 to February-2019. A total of 214 blood samples were collected from dogs suspected for microfilariosis and were screened for microfilaria by modified knott's method. Among 214 samples screened, 95 samples were found positive for microfilaria with an overall prevalence of 44.39 per cent. The species of microfilaria was identified as *D. repens* based on the morphology and micrometry. Age wise prevalence was found highest in adult dogs and least in puppies. During the study, highest prevalence was observed during North-east monsoon season from October to December months (71.42%). The breed wise prevalence showed highest in Labrador, followed by Doberman and Golden retriever dogs. The gender wise prevalence was found higher in males (71.05%) than females. The infection was found more in dogs kept outdoor as well as near drainage area.

**Keywords**
Epidemiology, Canine microfilariosis, *D. repens*, Mangalore

**Article Info**

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**Introduction**

Canine filariasis is caused by several species of filarial nematodes which are widely prevalent throughout the world, more specifically in the coastal region. They belong to the super family Filarioidea and family Onchocercidae. About nine filarial nematode species known to infect dogs worldwide are *Acanthocheilonema reconditum*, *Acanthocheilonema dracunculoides*, *Brugia malayi*, *Brugia pahangi*, *Brugia ceylonensis*, *Brugia patei*, *Cercopithifilaria grassii*, *Dirofilaria immitis* and *Dirofilaria repens* (Nelson, 2011). *D. immitis* is also known as heartworm, distributed worldwide and endemic in North, Central and South America, Africa, southern Europe and Asia. Subcutaneous dirofilariasis due to *D. repens* is widely distributed throughout Europe, Asia and Africa, with higher prevalence reported...
from Sri Lanka (30-60%), Iran (60.8%) and Italy (20.5-25%) (Tarello, 2002).

Most common filarial species reported in India are *D. immitis*, *D. repens*, *Acanthocheilonema* spp. and *Brugia* spp. They are distributed in various parts of India, mainly Kerala, Tamil Nadu, Karnataka, Orissa, West Bengal, Bihar, Uttar Pradesh and Maharashtra (Ravindran et al., 2014). The filarial species detected in dogs from different states of India includes *D. immitis* from Kerala (Valsala and Bhaskaran, 1974), *C. grasi* from Tamil Nadu (Balasubramaniam et al., 1975), *D. immitis* from Himalayas (Sarkar et al., 1976), *D. immitis* and *A. reconditum* from West Bengal (Chakrabart and Choudhury, 1983), *D. immitis* and *D. repens* from Orissa (Patnaik, 1989), *D. repens* from Kerala ( Radhika, 2001), *D. repens* and *A. reconditum* from Karnataka (Ananda and D'Souza 2007), *A. reconditum*, *D. immitis* and *D. repens* from Maharashatra and New Delhi and *Microfilaria auquieri* and a novel species of *Acanthocheilonema* from Ladakh, India (Rani et al., 2010). In general, it is believed that, *D. immitis* is mostly prevalent in north eastern India (Bortharkur et al., 2006) while *D. repens* is confined to southern parts of the country (Ananda et al., 2006 and Sabu et al., 2005).

Domestic dogs, foxes, wolves, coyotes and wild canids act as definitive hosts for *D. immitis* and the parasite also found in more than 30 species of animals, including domestic cats and wild felids, ferrets, seals, sea lions, bears and humans (Nelson, 2011). Human is the dead end host of *D. immitis* (Diessanaik et al., 2000), since worms cannot reach maturity. Dogs, cats and wild carnivores are final hosts of *D. repens* and it accidentally infects humans.

*D. immitis* and *D. repens* uses mosquitoes of several genera including *Culex, Aedes* and *Anopheles* as vectors. The distribution of *Dirofilaria* species is not determined by the availability of the vectors but rather by the ability of microfilariae to mature into infectious larvae in the mosquito vector, as maturation is temperature dependent. India has a wide range of climatic zones, from montane (cold, wet, pine) and semi-arid regions to the wet tropics, which make it suitable for a diverse range of vectors and pathogens of medical and veterinary importance, whose transmission and geographical distribution are closely linked to regional temperature, rainfall and humidity (Patz et al., 2005).

Prevalence of canine filariasis varies from one geographical area to another mainly because of differences in climatic conditions and distribution of vector. Risk factors for canine filariasis include age, gender, season, topography and living conditions of the dogs. The adult worms of *D. repens* are commonly found in the subcutaneous tissue causes subcutaneous dirofilariasis and are considered as moderately pathogenic. Most infections caused by *D. repens*, *Acanthocheilonema* spp. and *Brugia* spp. have minimal veterinary clinical significance, however all canine filariae can infect humans and remain important from a public health prospective (Irwin and Jefferies, 2004). Therefore, the present study was undertaken to ascertain the epidemiology of microfilariosis in dogs and its species identification, which is important for surveillance programme and therapeutic implications.

**Materials and Methods**

A total of 214 dogs presented to private clinics in Mangalore as well as dogs of non-descript breed captured by a non-governmental organization for sterilization under birth control programme were included in the present study. The blood samples were
collected from the dogs suspected for microfilariosis for a period of one year from March-2018 to February-2019 in ethylene diamine tetra acetic acid (EDTA) from recurrent tarsal vein for screening. The samples were examined on the same day for detection of microfilariae by modified Knott’s method as per Lindsay (1965). The detailed history of dogs regarding age, sex, breed, topography and living conditions was recorded. The Identification of Microfilaria was done as per the description by Soulsby (1992) and Bowman (2014). To study the age wise prevalence, different age groups were categorised as <1 year, 1-3 years, 3-5 years and > 5 years.

The season wise prevalence was recorded in four seasons – Summer (March, April and May), South-west Monsoon (June, July, August and September), North-east monsoon (October, December and November) and Winter (January and February). The breed and gender wise prevalence was also studied during the present study.

**Results and Discussion**

Out of 214 blood samples screened for a period of one year from March-2018 to February-2019, 95 samples were found positive for microfilaria by modified knott’s method with a overall prevalence of 44.39 %. Morphologically, the microfilariae were unsheathed with blunt head and the tail was long, curved with hook like posterior end (Fig. 1 and Fig. 2). The length and width of microfilaria were in the range of 310.9±9.10µm and 6.51±0.14µm respectively. The morphology and morphometry indicates the microfilariae belonged to *D. repens*.

In the present study, the age wise prevalence was observed highest in > 5 years old dogs (63.93%) followed by 3-5 years (60%), 1-3 years (36.47%) and least in <1 year old dogs (3.5%). The difference in respect to age group was significant (P≤0.05). The gender wise prevalence of microfilariosis showed higher in males (71.05%) than in females (29.71 %) and difference was statistically significant (P≤0.05).

The seasonal study revealed, highest prevalence of microfilariosis during north-east monsoon (71.42%) followed by south-west monsoon (40.84%), winter (38.88%) and summer (38.46%).

The difference between seasons was significant (P≤0.05) (Table.1). A total of seven breeds of dogs were screened for microfilariosis. Out of 192 Non-descript dogs samples screened, 83 (43.22%) were found positive.

Among eight Labradors, two Doberman and two Golden retriever blood samples screened, all were found positive for microfilaria whereas, four Boxer, two Great Dane and four Lhasa Apso breeds of dogs were found negative for microfilaria. The breed wise prevalence of microfilariosis was higher in Labrador, Doberman and Golden retriever breeds (Table. 2) and showed a statistically significant difference (P≤0.05).

During the study, it was also observed that, the infection was more in dogs living in both outdoor and indoor conditions (100%), compare to dogs living exclusively in outdoor (44.02%) and indoor conditions (20%). The difference between living conditions was found statistically significant (P≤0.05). The topographical study revealed that, the infection was higher in dogs reared near farm topography (100%), followed by near drainage (44.75%), urban (44.44%) and dogs from marshy areas were found negative for microfilaria. Statistically the difference was non-significant (P≤0.05) (Table. 3).
**Fig. 1** Microfilaria in modified knott’s method (400X)

**Fig. 2** Microfilaria: unsheathed, (a) blunt head (b) tapering tail (400X)
Table 1 Age wise, sex wise and season wise prevalence of microfilariosis in dogs

<table>
<thead>
<tr>
<th>Age (Years)</th>
<th>Sex</th>
<th>Season</th>
<th>South West monsoon</th>
<th>North East monsoon</th>
<th>Winter</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1-3</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3-5</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&gt;5</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>28</td>
<td>85</td>
<td>40</td>
<td>61</td>
<td>76</td>
</tr>
<tr>
<td>Female</td>
<td>138</td>
<td>142</td>
<td>26</td>
<td>142</td>
<td>28</td>
</tr>
</tbody>
</table>

Number of dogs examined 28 85 40 61 76 26 142 28 18

Number positive 1 31 24 39 54 41 10 58 20

Per cent Prevalence 3.5 36.4 7 60 63.9 3 71.05 29.71 38.46 40.84 71.42 38.88

\[X^2 \text{ Value} = 34.45^{\ast\ast}, 33.93^{\ast\ast}, 9.606^*\]

\(**\) - Significant at \(p \leq 0.01\), \(*\) - Significant at \(p \leq 0.05\)

Table 2 Breed wise prevalence of microfilariosis in dogs

<table>
<thead>
<tr>
<th>Sl. No.</th>
<th>Breed</th>
<th>Number of dogs examined</th>
<th>Number positive</th>
<th>Per cent Prevalence</th>
<th>X² Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Non-descript</td>
<td>192</td>
<td>83</td>
<td>43.22</td>
<td>14.63^{\ast\ast}</td>
</tr>
<tr>
<td>2.</td>
<td>Labrador</td>
<td>8</td>
<td>8</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>3.</td>
<td>Doberman</td>
<td>2</td>
<td>2</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>4.</td>
<td>Golden Retriever</td>
<td>2</td>
<td>2</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>5.</td>
<td>Boxer</td>
<td>4</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>6.</td>
<td>Great Dane</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>7.</td>
<td>Lhasa Apso</td>
<td>4</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

\(**\) - Significant at \(p \leq 0.01\)

Table 3 Topography and living conditions wise prevalence of microfilariosis in dogs

<table>
<thead>
<tr>
<th>Topography</th>
<th>Living conditions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Near drainage</td>
<td>Near farm</td>
</tr>
<tr>
<td>Number of dogs examined</td>
<td>181</td>
</tr>
<tr>
<td>Number positive</td>
<td>81</td>
</tr>
<tr>
<td>Per cent Prevalence</td>
<td>44.75</td>
</tr>
</tbody>
</table>
| \[X^2 \text{ Value} = 5.708, NS, 17.36^{\ast\ast}\]

\(**\) - Significant at \(p \leq 0.01\), \(\text{NS: Non-significant}\)
During the present study, 44.39 per cent prevalence of canine microfilariosis was recorded in Mangalore region of Karnataka. Ananda and Placid (2007) reported 38.09 per cent prevalence of canine microfilariosis in Mangalore region whereas, Radhika et al., (2001) reported 7.95 per cent prevalence of canine microfilariosis in Thrissur, Kerala region. The variation in prevalence rate may be due to the fact that, the study conducted in different period of time and in different geographical area because of difference in the distribution of the vector, topography, environment and average age of the study population. Although temperature is the main influencing factor for transmission of dirofilaria as per Brown et al., (2012), many other factors influence the transmission are precipitation, relative humidity, human and animal population density and socio economic status.

The higher prevalence of microfilariosis was recorded in dogs more than 5 years old and males. This is in accordance with the previous studies (Radhika et al., 2001; Christopher and Abel-Danjuma. 2016), who reported higher rate of microfilariosis in male and older dogs. This may be due to the exploratory life style of male dogs and increased exposure risk to mosquitoes in adult dogs. The least prevalence in young puppies may be due to the fact that approximately 10 months is required for L3 larvae to become adults and then to produce microfilariae. In the present study, the higher prevalence was recorded during North-west monsoon season, while Radhika et al., (2001) reported higher prevalence in summer whereas, Deepa and Alex (2011) reported highest prevalence in winter. This variation might be due to the fact that, the mosquito vector plays role in transmission of dirofilariasis and whose development and survival need favourable climatic conditions viz. temperature, rainfall and humidity (Patz et al., 2005).

Prevalence of microfilariosis was found highest in Labrador, Doberman and Golden retriever breeds. This may be due to the representation of these breeds in this study population was highest among those examined, as reported by Bhattacharjee and Sarmah (2014).

The higher prevalence of microfilariosis was in dogs living in both outdoor and indoor conditions as well as near drainage area. This might be due to increased exposure of dogs to mosquito vector and keeping animals indoors may reduce the risk of exposure to the disease transmitting mosquitoes as reported by Walter (1996) and Theis et al., (1999).

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