MICROMETRY FOR DIFFERENTIATION OF DEMODEX MITE SPECIES CAUSING CANINE DEMODICOSIS IN INDIA

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Introduction:
Canine demodicosis can be diagnosed by identifying mites in skin samples. There are three species of Demodex mites with different location and consequently, with different courses and varied symptoms in dogs (Sakulpoy and Sangvanand, 2010). Demodex canis which lives in hair follicles causes demodectic folliculitis and/or furunculosis in dogs; D. injai inhabits in pilosebaceous glands induces oily skin and hair coat on trunk of dogs; however, D. cornei can cause a pruritic canine skin disease by inhabiting in stratum corneum.

Follicular mite, Demodex canis is the most common species and there have been two other morphologically different types of Demodex mites named as Demodex cornei and Demodex injai being reported in different countries in the recent past (Sivajothi et al., 2013a). Morphometric studies were useful in differentiation of Demodex species were less in India. Hence, the present study of micrometry was undertaken to record the morphometry of Demodex mites, i.e. D.canis, D. cornei and D. injai in dogs with demodicosis.

Materials and methods:-
Skin samples were taken from 20 dogs suffering from demodicosis, which were presented to Teaching Veterinary Clinical Complex (TVCC), College of Veterinary Science, Rajendranagar, Hyderabad. The skin samples thus collected using deep skin scrapings, tape impression smears and hair plucks were processed using 10% KOH (Soulsby, 2005) and were preserved in 70% ethanol till micrometry was done. Micrometry was performed by using ocular and stage micrometers under compound microscope (Sriram, 2006) for measuring mites, after calibration under 40X magnification (Figure 1).
Smears of processed skin samples were used for morphological studies and measurements of the *Demodex* mites were made. Total of 179 mites were measured using calibrated micrometers (µm) and mean body length (µm), mean lengths (µm) of gnathosoma, podosoma and opisthosoma, mean width (µm) of gnathosoma, podosoma and opisthosoma, ratio of prosoma to opisthosoma, ratio of total body length and opisthosoma length, ratio of length opisthosoma length to body length (%) and the mean length and mean width of *Demodex* mite eggs were measured.

**Results and Discussion:**
Using micrometry analysis, among the skin samples collected from 20 *Demodex* positive dogs, 19 dogs were positive for *Demodex canis*, among them synhospitalic mite infestation with *Demodex canis* and *Demodex cornei* were noted in 40% (8) dogs. *Demodex injai* was identified in the skin samples taken from one dog with generalized demodicosis.

The morphometrics of three *Demodex* species were presented in table 1 and the micrometry of *Demodex* mites and their eggs shown in figure 2.
Figure 2: Micrometry of *Demodex* mites and their eggs.

- *Demodex canis* mite measuring 224μm
- *Demodex canis* egg measuring 84μm
- *Demodex cornei* mite measuring 120μm
- *Demodex cornei* egg measuring 68μm
- *Demodex injai* mite measuring about 264μm
- *Demodex injai* egg measuring 108μm
Table 1:- The morphometrics of three Demodex species.

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Parameters</th>
<th>D.canis (n=98)</th>
<th>D.cornei (n=52)</th>
<th>D.injai (n=29)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Gnathosoma (µm)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Length</td>
<td>23.1 ± 0.33</td>
<td>18.2 ± 0.6</td>
<td>23.65 ± 0.69</td>
</tr>
<tr>
<td></td>
<td>Width</td>
<td>25.6 ± 0.7</td>
<td>17.6 ± 0.65</td>
<td>24.03 ± 1.05</td>
</tr>
<tr>
<td>2</td>
<td>Podosoma (µm)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Length</td>
<td>64.16 ± 0.73</td>
<td>58.5 ± 1.86</td>
<td>74.31 ± 1.36</td>
</tr>
<tr>
<td></td>
<td>Width</td>
<td>40.71 ± 0.41</td>
<td>36 ± 3.3</td>
<td>42.72 ± 0.86</td>
</tr>
<tr>
<td>3</td>
<td>Opisthosoma* (µm)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Length</td>
<td>135.59 ± 3.17</td>
<td>80.84 ± 7.33</td>
<td>168.6 ± 7.78</td>
</tr>
<tr>
<td></td>
<td>Width</td>
<td>36.2 ± 0.98</td>
<td>25.71 ± 2.6</td>
<td>32.35 ± 0.49</td>
</tr>
<tr>
<td>4</td>
<td>Total body length* (µm)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>217 ± 2.39</td>
<td>138.99 ± 3.21</td>
<td>264 ± 6.89</td>
</tr>
<tr>
<td>5</td>
<td>Egg (µm)*</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Length</td>
<td>82.3 ± 3.96</td>
<td>69 ± 4.12</td>
<td>105.5 ± 3.36</td>
</tr>
<tr>
<td></td>
<td>Width</td>
<td>23.5 ± 0.99</td>
<td>21.6 ± 1.26</td>
<td>28.4 ± 1.03</td>
</tr>
<tr>
<td>6</td>
<td>Ratio of prosoma to opisthosoma*</td>
<td>0.65 ± 0.02</td>
<td>1.26 ± 0.02</td>
<td>0.56 ± 0.03</td>
</tr>
<tr>
<td>7</td>
<td>Ratio of mean total bodylength and opisthosoma length*</td>
<td>1.64</td>
<td>2.05</td>
<td>1.57</td>
</tr>
<tr>
<td>8</td>
<td>Ratio of opisthosoma length to bodylength (%)*</td>
<td>57.96</td>
<td>48.54</td>
<td>63.69</td>
</tr>
</tbody>
</table>

* Significant (P<0.05)

The mean total body length of Demodex canis (217 ± 2.39 µm) was almost agreeable with the findings of Tamura et al (2001), Sakulpoy and Sangvaranond (2010), Sivajothi et al (2013a) and Sivajothi et al (2013b), who had reported mean body lengths of Demodex canis as 229 ± 35.1, 217.83 ± 30.06, 211.81 ± 14.86 and 214.32 ± 13.81µm, respectively.

The mean lengths (µm) of gnathosoma, podosoma and opisthosoma of Demodex canis reported in the present study were 23.1 ± 0.33, 64.16 ± 0.73, 135.59 ± 3.17, respectively, were coinciding with the findings of Tamura et al. (2001), who reported 18.9-24.4, 94.11 ± 8.4 and 136.6 ± 18.5µm, Sakulpoy and Sangvaranond (2010) reported (mean ± SD) 24.84 ± 1.70, 60.89 ± 2.10 µm and 147.50 ± 19.56 µm, Sivajothi et al. (2013a) 19.52 ± 0.10, 62.68 ± 0.33µm and 130.52 ± 2.47 µm and Sivajothi et al. (2013b) 18.89 ± 0.18, 60.98 ± 0.21, and 129.68 ± 3.3µm, respectively.

The mean width (µm) of gnathosoma, podosoma and opisthosoma of Demodex canis were 25.6 ± 0.7, 40.71 ± 0.41, and 36.2 ± 0.98 were comparable with the reports of Tamura et al. (2001), who documented measurements of gnathosoma width 18.9-20 µm, and opisthosoma width 26.5 ± 4.2 µm using scanning electron microscopy (SEM).

The ratio of prosoma to opisthosoma was 0.65 ± 0.02; which was similar to the reports of Sivajothi et al. (2013a), who reported the ratio as 0.62 ± 0.02 for Demodex canis.

The ratio of mean total body length and opisthosoma length was 1.64, which was closer to the findings of Sakulpoy and Sangvaranond (2010), who reported the ratio as 1.39, the difference could be due to more variations in lengths of opisthosoma of D. canis mites of the study undertaken.

Ratio of opisthosoma length to total body length in Demodex canis was found to be 57.96%, which coincided with the findings of Izdebska (2010), who found this parameter in D. canis (51 in males, 59 in females) lesser than that of D.injai and greater than that of D.cornei.

The mean length and mean width of Demodex canis eggs found in the present study were 82.3 ± 3.96 and 23.5 ± 0.99 µm, respectively, which coincided with the reports of Nutting and Desch (1978), who documented 81.5 ± 3.5 and 26.6 ± 2.4 egg length and width of Demodex canis, respectively.

The mean body length (µm) of Demodex corni in the present study 138.99 ± 3.21 (95.90-164.4) was matched with the reports of Tamura et al. (2000), who reported 97.5-167.5 (139) µm mean body length of Demodex corni, and Tamura et al. (2001), who reported 139 ± 21.6µm, Sakulpoy and Sangvaranond (2010) found 156.92µm, Lopez et
al. (2011) documented 139.3 ± 10.4, Sivajothi et al. (2013a) as 137.15 ± 22.84μm and Sivajothi et al. (2013b), who reported mean body lengths of *Demodex canis* 132.21 ± 14.6 μm.

The mean lengths (μm) of gnathosoma, podosoma and opisthosoma of *Demodex cornei* in the present study were 18.2 ± 0.596, 58.5 ± 1.86, and 80.84 ± 7.33 respectively, which were similar to the findings of Tamura et al. (2001), who reported the lengths of gnathosoma, leg region of protosoma and opisthosoma of *D. cornei* 14–21.4, 13.0 and 31.5 ± 6.2 μm, Sakulpoy and Sangvaranond (2010) reported in microns ± SD 23.50 ± 1.93, 60.00 ± 2.50 and 59.25 ± 9.68, respectively. Lopez et al. (2011) documented gnathosoma, podosoma and opisthosoma length of *D. cornei* ranged from 18 to 25 μm (21.9 ± 2.2), 52 to 68 μm (61.2 ± 4.6) and 48 to 69 μm (56.2 ± 8.4), respectively.

The mean width (μm) of gnathosoma, podosoma and opisthosoma of *Demodex cornei* were 17.6 ± 0.65, 36 ± 3.3, 25.71 ± 2.597 respectively, which was similar to the findings of Lopez et al. (2011), who reported Gnathosoma, podosoma and opisthosoma width of *Demodex cornei* ranged from 18 to 28 μm (20.6 ± 2.3), 29 to 44 μm (33.5 ± 5.6) and 26 to 35 μm (29.9 ± 2.6), respectively.

The ratio of prosoma to opisthosoma was 1.26 ± 0.02, which was similar to the reports of Sivajothi et al. (2013a), who reported the ratio as 1.37 ± 0.04 for *Demodex cornei*.

The ratio of mean total body length and opisthosoma length was 2.05, which was closer to the reports of Sakulpoy and Sangvaranond (2010), who reported 2.49. The difference could be due to more variations in lengths of opisthosoma of *D. cornei* mites of the study undertaken.

Ratio of opisthosoma length to total body length in *Demodex cornei* was found to be 48.54%, which is coincide with the findings of Izdebska (2010), who found this parameter in *D. cornei* very much less than that of *Demodex canis* and *Demodex injai*.

The mean length (μm) and mean width (μm) of *Demodex cornei* eggs were 69 ± 4.12 and21.6 ± 1.26, respectively, which were matched with the findings of Lopez et al. (2011), who reported egg length and width 71.7 ± 2.5 (68 - 79) and 22.1 ± 1.4 (20 - 24) of *Demodex cornei*.

Mean body length of *Demodex injai* was found to be 264 ± 6.89 μm, which was matched with lower limit reported by Hillier and Desch (2002), who documented *Demodex injai* mite body length 270-390μm, the difference may be due to small sample size with size variations.

The mean lengths(μm) of gnathosoma, podosoma and opisthosoma were 23.65 ± 0.69, 74.31 ± 1.36, and 168.6 ± 7.78 in *Demodex injai* of present study were similar to the findings of Hillier and Desch (2002), who reported 23.4 ± 1.1, 87.0 ± 3.6 and 225.4 ± 29.1, respectively.

The mean width(μm) of gnathosoma, podosoma and opisthosoma of *Demodex injai* were 24.03 ± 1.05, 42.72 ± 0.86 and 32.35 ± 0.49 which matched with the findings of Hillier and Desch (2002), who reported 28.0 ± 1.7, 44.3 ± 2.5 and 35.6 ± 2.9, respectively.

The ratio of prosoma to opisthosoma and ratio of mean of total body length and opisthosoma length of 0.56 ± 0.03 and 1.57, respectively; which were lesser than other two species due to longer opisthosoma. The previous findings of authors were not available for these parameters.

The ratio of opisthosoma length to body length (%) in *Demodex injai* mites was 63.69 which was more compared to other two species, matched with the findings of Izdebska (2010), who reported 68% in *Demodex injai*, which was more than respective parameter in other two species of *Demodex* mites.

The mean length and mean width of *Demodex injai* eggs (μm) were 105.5 ± 3.36 and 28.4 ± 1.03, respectively, which was almost in agreement with the findings of Hillier and Desch (2002), who reported the mean length and width of *Demodex injai* egg were 104.6 ± 5.0 and 29.93 ± 1.9, respectively.
References: