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Comparative Morphometric Analysis for Differentiation of Three Demodex Mite Species causing Canine Demodicosis

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A B S T R A C T

In the present study, Demodex mite species of dog were differentiated based on their morphometric analysis. For mite's micrometry, skin scarping materials were collected from 40 demodicosis clinically infected dogs, presented with different clinical manifestations. Out of 150 dog mite's species *D. canis* (n=50), *D. cornei* (n=50) and *D. injai* (n=50) were randomly selected for ocular micrometry of mites, body size and its segments. Correlation between body size and its body segments of all three Demodex species were estimated by descriptive statistical data analysis. There was a significant correlation between total body length and length of the podosoma and opisthosoma ($p<0.05$) of *D. injai* as compared to D. *canis* and *D. cornei* species of dog mites.

Introduction

Demodex canis specie was first discovered demodex mite specie of dog by Simon in 1844. Morphological description and name was given by Leydig in 1859. *Demodex injai* was first reported by Desch and Hillier in 2003 and *Demodex cornei*, was firstly discovered by Mason in the 20th century. Based on mitochondrial marker 16S rDNA and Cytochrome Oxidase- I, study of Rojas *et al.,* (2012) concluded that, remaining two forms of reported dog mite *Demodex injai* and *Demodex cornei* are polymorphs of the *Demodex canis* species. Remarkable study of Sastre *et al.,* (2012) who analysed phylogenetic trait of these three species based on partial sequence of mitochondrial 16rDNA and proposed that, *Demodex injai* is the separate species of dog mite and latter on Milosevic *et al.,* (2013) confirmed that, *Demodex injai* are the valid separate species of dogs mites. Family Demodicidae, have a small, thin, usually elongated body, with four pairs of legs. Their bodies are divided into three distinct part called as gnathosoma (mouth and head part), podosoma consists of four pairs of legs and opisthosoma part of

elongated tail. All the three demodex mites species have been reported in India as well as abroad Veena *et al.,* 2017 and Fathima *et al.,* 2017. The present investigation was aimed to study the morphometry of three *Demodex* mites, i.e. *D. canis, D. injai* and *D. cornei* in dogs with demodicosis. It might be helpful in the identification of mite species based on their morphology and its measurements.

Materials and Methods

Identification and morphometric measurements were performed in clinical laboratory of Division of Medicine, ICAR-Indian Veterinary Research Institute, Izatnagar, UP, India. The exact size of a microorganism was determined by using calibrated ocular lens and stage micrometer (Erma objective micrometer, Japan) under compound light microscope (Olympus microscope Model CH-20i.) as per method described by Gonzale and Bendall, (1995) and mites were identified on the basis of morphological characteristics as described by Soulsby, (1996). All the ratios were calculated by using simple arithmetic formulas.

Results and Discussion

Micrometry of 150 mites revealed that, mean length of total body of *Demodex injai* species was 263.610 ± 2.83$^{c.}$ It was significantly larger as compared to *D. canis* and *D. cornei* species of mites ($p<0.05$). It was more or less similar to study of Swathi *et al.,* 2016 who reported the mean body length for *Demodex injai* was 264 ± 6.89µ. Maximum mean length for Demodex *injai* has been measured by Desch *et al.,* 2003 it was found that, 361.3 ± 43.9 µm of mean length of total body. Among three demodex species, *Demodex cornei* was shorter in mean length of total body (156.887 ± 1.35 ^{b} µm). Recently, some researchers was also reported shorter mean length of same mites species, Sakulpoy and Sangvaranond 2010 found to be very closely with finding of the present study (156.92µm) and other were also agreed with similar findings Fathima *et al.,*2017 and Swathi, *et al.,* 2016. The mean length of total body of *Demodex canis* was 223.822 ± 1.69^{a} µm. It was larger as compared to *Demodex cornei* body length but shorter to *Demodex injai.* It was matched with the findings of [11], who had reported mean body lengths of *Demodex canis* as, 211.81 ± 14.86 and 214.32 ± 13.81µm, respectively.

Body segments of all three mite species were also showed significant difference. The mean length/width of first segments of body (gnathosoma) of *D. canis, D. injai* and *D. cornei* were 21.76 ± 0.29^{a} / 20.49 ± 0.32^{c}, 18.85 ± 0.64^{b} µm /16.83±0.88^{b} µm and 22.42 ± 0.60^{a} µm / 24.04 ± 0.70^{c} µm respectively. Mean length and width of gnathosoma in *Demodex cornei* was significantly shorter and wider as compared to *D. canis* and of *Demodex injai* gnathosoma. It agrees with findings of Fathima *et al.,* 2017 and Swathi, *et al.,* 2016.

Mean length/width of second segments of body (podosoma) of *D. canis, D. injai* and *D. cornei* were 70.38±1.05 ^{a}/39.02±0.42 ^{a}, 74.94±0.77 ^{c}/43.03±0.63 ^{c} and 56.89±0.90 ^{b}/23.91±0.90^{b} respectively. Among these larger length of podosoma was measured in *Demodex injai.* In contrast to, shorter length and comparatively broader width of podosoma were measured in *D. cornei.* It was agreed with Desch and Nutting 1998.

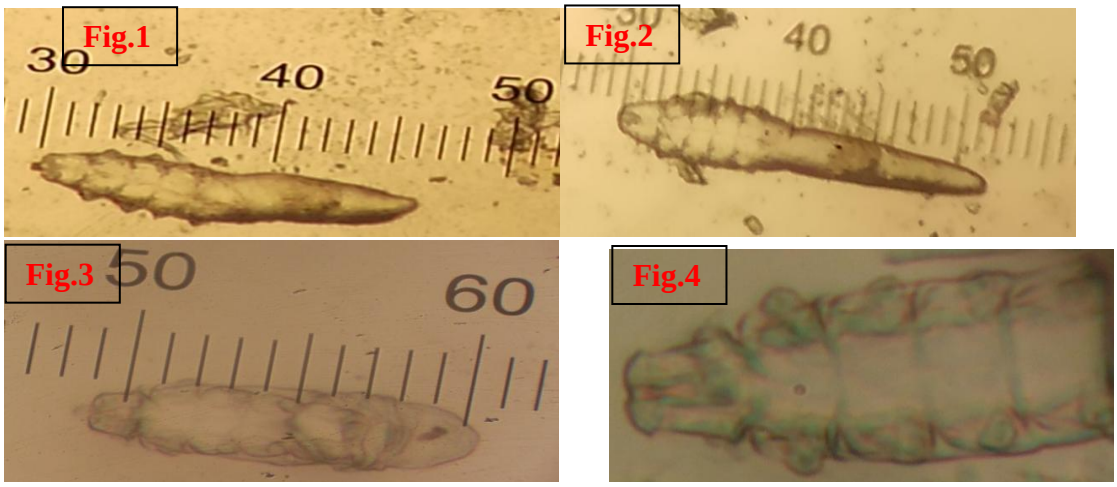
Significant variation was noticed in length and width of opisthosoma (third body segment) in all three reported species of demodex. Larger opisthosoma was observed in *D. injai* with mean length 166.246±2.55^{c} µm and width 37.552 ± 4.39^{a} µm. Blunt and shorter opisthosoma was noticed in *D. cornei* species as 81.137 ± 1.03^{b}µm and width was

24.24 ± 0.9 [a] µm and in *D. canis* species, it was medium in length 131.674 ± 1.20 [a] µm and width 34.8 ± 0.52 a µm. It was similar to those findings of Desch and Nutting 1998 (Fig. 1–4).

Table.1 Micrometric analysis of mite's species of dog causing demodicosis

Part of body	**Statistics**		***D. canis***	***D.cornai***	***D. injai***	**P value**
			N=110	N=30	N= 50	<0.05
Gnathosoma (µm)	L	Mean	21.76±0.29[a]	18.85±0.64[b]	22.42±0.60 [a]	
		Range	27.27-10.10	20.20-10.10	30.30-20.20	
		G.mean	21.51	18.42	22.08	
	W	Mean	20.49±0.32 [a]	16.83±0.88[b]	24.04±0.70[c]	<0.05
		Range	28.00-12.00	20.20-10.10	30.30-20.20	
		G.mean	20.19	16.03	23.56	
Podosoma (µm)	L	Mean	70.38±1.05 [a]	56.89±0.90 [b]	74.94±0.77 [c]	<0.05
		Range	90.90-50.50	60.60-50.50	90.90-70.70	
		G.mean	69.51	56.68	74.75	
	W	Mean	39.02±0.42 [a]	23.91±0.90 [b]	43.03±0.63 [c]	<0.05
		Range	40.40-20.20	30.3-020.20	50.50-40.40	
		G.mean	38.68	23.44	42.81	
Opisthosoma (µm)	L	Mean	131.674±1.20 [a]	81.137±1.03 [b]	166.246±2.55 [c]	<0.05
		Range	161.60-101.00	90.90-70.70	212.10-141.40	
		G.mean	131.08	80.95	165.32	
	W	Mean	34.8±0.52 [a]	24.24±0.92 [a]	37.55± 4.39 [a]	>0.05
		Range	50.50-30.30	30.30-20.20	43.43-19.19	
		G.mean	34.49	23.76	26.61	
Total body length (µm)	L	Mean	223.822±1.69 [a]	156.887±1.35 [b]	263.610±2.83 [c]	<0.05
		Range	266.64-175.74	171.70-141.40	313.10-232.30	
		G.mean	223.12	156.72	262.88	
Differences in divisions, fraction and ratios of mites species body segments						
Ratio of G: TB		Division	0.097	0.120	0.085	
		Fraction	7/72	3/25	4/47	
		Ratio	07: 72	03:25	04:47	
Ratio of P:TB		Division	0.314	0.363	0.284	
		Fraction	11/35	33/91	27/95	
		Ratio	11:35	33:91	27:95	
Ratio of O: TB		Division	0.588	0.517	0.631	
		Fraction	10/17	15/29	41/65	
		Ratio	10:17	15:29	41:65	
Ratio of G:P		Division	0.309	0.331	0.299	
		Fraction	17/55	1/3	3/10	
		Ratio	17:55	01:03	03:10	
Ratio of P: O		Division	0.535	0.701	0.451	
		Fraction	31/58	61/87	32/71	
		Ratio	31:58	61:87	32:71	

Fig.1 Adult *Demodex* canis, **Fig.2** Adult *Demodex injai*, **Fig.3** Adult *Demodex cornei* and **Fig.4** Gnathosoma (first body segment) and podosoma of adult *Demodex canis*



Calculated ratios of Gnathosoma length mean and total body length mean of *Demodex canis it* was 07: 72, in *Demodex cornei* was 03:25 and in *Demodex injai* was 04:47. Ratios of Podosoma length mean and total body length mean (P: TB) of *D. canis* was 11:35 in *D. cornei* was 33:91 and in Demodex *injai* was 27:95.

Ratios of opisthosoma length mean and total body length mean (O: TB) of *D. canis* was 10:17 in D. *cornei* was 15:29and in *Demodex injai* was 41:65.The ratio between gnathosoma and opisthosoma (G: O) length in of *D. canis* was 17:55 in *D. cornei* was 1:3and in *Demodex injai* was 3:10. The ratio between podosoma and opisthosoma (P: O) length in *D. canis* was 31:58, in *D. cornei* was 61:87 and in *Demodex injai* was 32:7. Calculated ratios were similar to findings of Swathi, *et al.*, 2016.

In conclusion three species of demodex mite were observed in the study viz., *Demodex injai*, *D. canis* and *D. cornei*. The morphometry of mites revealed that mean total body length of *Demodex cornei* was much less than that of *Demodex canis* and *Demodex injai*. *D. cornei* had short opisthosoma and blunted posterior end as compared to other remaining species of dog's demodex mite. Demodex *injai* had tall and thinner opisthosoma with pointed end. Calculated ratios indicate that approximated relationship within or between species of body segments of Demodex mites of dog.

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