



INDOOR AND OUTDOOR DENSITY OF ADULT *PHLEBOTOMUS ARGENTIPES* THE VECTOR OF *VISCERAL LEISHMANIASIS* IN AN ENDEMIC FOCI OF BIHAR INDIA

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ABSTRACT Since, 1976 to 2014, DDT, and from 2015 till date, alpha cypermethrin insecticide is being sprayed for two rounds per year as indoor residual spray up to 1.8 meter i.e 6 feet height in human dwellings as well as in cattle sheds in Bihar, India, as a part of Kala -azar elimination programme. This is being practiced out of the belief that *Phlebotomus argentipes* the vector of kala azar is endophilic in nature and cannot hop beyond 6feet (1.8 meters) height. A sixteen months study was therefore conducted in three villages in a kala azar endemic block Warisnagar, district Samastipur, Bihar from September 2016 to November 2017 to note the vertical distribution of *P. argentipes* in indoors and their distribution in outdoors. In all the three study villages, density of adult *P. argentipes* was found more in Cattle sheds than in human dwellings. Irrespective of IRS and non IRS villages, significant numbers (p value 0.47) of *P. argentipes* were found above 1.8 meter (above 6 feet) in CS and HD of which 33.37 percent blood fed and gravid females. Sizable number of *P. argentipes* was also found in outdoor peridomestic situations throughout the study period. Therefore it may be the time to evaluate vector control strategies in kala azar endemic areas of Bihar for successful elimination of the disease from India.

KEYWORDS : Distribution of *Phlebotomus argentipes* in indoor and outdoor. Kala azar endemic villages. Bihar

Introduction:

Phlebotomus (Euphlebotomus) argentipes, the vector of visceral leishmaniasis or Kala azar in the endemic states like Bihar, West Bengal are predominantly endophilic in nature. They are light shy species, rest in cracks and crevices on walls, inside rodent burrows in dark corners of rooms, especially in mud huts and cattle sheds. They are found more in cattle sheds than human dwellings. It has also been noted that *P. argentipes* do not fly / rest above 1.8meter (6 feet) from ground level^{1, 2, 3}. Since 1976, as a part of kala azar vector control programme, indoor residual spray of DDT at the rate of 1gm /sq. meter is being done in human dwellings and cattle sheds up to 1.8meter i.e. 6 feet height for control of *Phlebotomus argentipes* in India. Noting the wide spread DDT resistance problem in *P. argentipes*^{4,5,6,7}, Government of India along with Government of Bihar as a part of Kala azar Elimination Programme, in the year 2015 have started vector control programme with two rounds of IRS with synthetic pyrethroid insecticide like Alpha-cypermethrin at the dose of 25mgm per sq. meter twice a year up to 1.8meter (6 feet) level of height^{7,8,9}. The state of Bihar is yet to achieve successful Elimination of Kala azar (VL), may be one of the reason that the flies have changed their resting behaviours. A sixteen months study was therefore conducted in a highly kala azar endemic Block Warisnagar, District Samastipur, Bihar starting from August 2016 till November 2017 to note the population of *P. argentipes* in different heights at indoor of cattle sheds and human dwellings as well as to note availability of the flies in outdoor per domestic conditions in the same study villages.

Materials and Methods

In comparison to India, Bihar state alone contributed more than 72 percent of total kala azar cases per year between 2012 to 2016 (Dpt. of NVBDCP, Government of India). District Samastipur, (25°5' N, 85°5' E) alone has contributed more than 8.5 percent Kala Azar cases of Bihar in 2014 to 2016 (Personal communication Dpt. NVBDCP, Government of Bihar). Two villages i.e. Mannipur and Satmalpur which are reporting perennial transmission of kala azar and are under insecticidal pressure for more than ten years and village Kishanpur which is without any kala azar case and is not under indoor residual spray (IRS) for vector control since last six years in Block Warisnagar, were selected for the study.

All human dwellings and cattle sheds were searched thoroughly by standard entomological methods¹⁰. Cattle sheds and human dwellings had typical conditions for sand fly breeding and therefore good density of *P. argentipes* were noted and marked. Out of marked houses, three human dwellings and three cattle sheds in each village were selected by random method. For outdoor collection, ten dark and damp

outdoors with five to eight meters diameter surrounding each of the noted house and cattle shed were selected. These places had bushes of banana, litchi, palm trees etc. Therefore, in each village three human dwellings, three cattle sheds and ten peridomestic outdoor situations were selected for longitudinal study between August 2016 to November 2017. In each human dwelling and cattle shed, three CDC miniature light traps model 512, specially designed for sand fly collection (made by John W. Hock) were installed overnight (dusk to dawn) at the heights of 0.6 meter (2feet), 1.2 meter (4feet) and above 1.8 meters (above 6 feet) by standard WHO method¹⁰. In outdoor situations ten CDC light traps per village were installed for overnight at a height of 0.6 meter (2feet).

In each village a total of twenty eight light traps (nine in human dwellings, in nine cattle sheds and ten at peridomestic outdoor situations) were installed per night per month. The installation of light traps were undertaken every third week of each month from August 2016 to November 2017 (sixteen months). The sand flies, collected in individual trap were brought to the laboratory separately and height wise species identification has been done by the method of Lewis¹¹.

Results and Discussion:

Results on distribution of indoor and outdoor (peridomestic) population of *P. argentipes* in three study villages of block Warisnagar, district Samastipur from September 2016 to November 2017 are shown in Table 1 and Table 2.

Month wise adult *P. argentipes* were collected from different heights in indoors of selected human dwellings (HD) and cattle sheds (CS) using CDC Light traps in three study villages which are shown in Table 1 and Graphs 1,2 and 3.

In Kishanpur, which, is non endemic and non IRS village, sixteen months observation from September 2016 to November 2017 was done, where 46.7% flies were found above 1.8 meter (Above 6feet) height in human dwellings. As per Table1 and Graph 1, density of the flies in Cattle sheds in the same village was noted 40.4 % above 1.8 meters. 26.5 and 27.8 percent flies were found up to 0.6 meter height in HD and CS respectively. 24.8 and 31.8 percent *P. argentipes* were also found up to 1.4 meter (4feet) height in HD and CS respectively. Therefore statistically significant (p value 0.051) number of *P. argentipes* were found above 1.8 meters in unsprayed non endemic village Kishanpur. Very low density of *P. argentipes* in both human dwellings and cattle sheds were noted in the winter months between December 16 to February 2017. Density started increasing from the month of March on the onset of warm weather with a peak in the

months of June-July and September – October. Number of *P. argentipes* found much higher in CS (557) than HD (274). Villages Mannipur and Satmalpur are endemic for Kala azar transmission and under Indoor Residual Spray with Alpha-cypermethrin. During study period from September 2016 to November 2017 the villages were covered with three rounds of IRS. The results are shown in Table 1 and Graphs 1 and 2.

During study period, village Mannipur were sprayed in every third week of months September 2016, April 2017 and September 2017. The results were shown in Graph 2. Like Kishanpur, lowest density was noted here in the winter months of December 2016 to February 2017. Throughout the study period 32.0 and 29.2 percent *P. argentipes* were found above 1.8 meter height in HD and CS respectively. It was noted that population of *P. argentipes* in CS (411) is more than that of in HD (366). Peak density was noted in September- October, 2016 and in the month of May 2017. Throughout the study period maximum number of flies in this village was found as 33.8 percent in HD and 43.2 percent in CS up to 1.4 meter height. (*p* value 0.390). Although *P. argentipes* were found highly susceptible to Alpha cypermethrin¹⁵, but our present study shows that present IRS do not have action on as per desired level.

Village Satmalpur, as shown in Graph 2 and Table 1, had a very high density of *P. argentipes* in CS as 962 in comparison to HD as 281.26.0 and 37.9 percent *P. argentipes* found above 1.8 meters in HD and CS respectively which is statistically significant with *p* value 0.565). 50.9 and 36.3 percent of *P. argentipes* were found within 1.4 meters in HD and CS respectively. Like other two study villages, minimum density was also noted in the winter months of December 2016 to February 2017 and maximum in the months of September, October 2016 and May 2017 in Satmalpur. During study period the village was sprayed with Alpha cypermethrin in the months of October 2016, May 2017 and October 2017.

As per present observations significant numbers (*p* value 0.051, 0.390, & 0.565) of *P. argentipes* adults prefers to rest more in Cattle sheds than Human dwellings in villages Kishanpur, Mannipur and Satmalpur respectively, which corroborates with the early findings of Smith¹, Hati et al^{2,3}. In earlier literatures it was mentioned that *P. argentipes* in India cannot hop or rest beyond 6feet i.e 1.8 meters height^{1,2,3,4,7, & 9}. In our present study is clearly showing presence of significant number (*p* value 0.47) of *P. argentipes* beyond the height of 1.8 meter in three study villages of which 33.37 percent females were full blood fed and gravid females.

The present observations corroborate with our previous four months observations from July to November 2016 in the same area¹². Thus our present findings clearly show that *P. argentipes* adults have changed

their behaviour over the period of time and significant numbers are available above 1.8 meter. This invites for a change in policy of IRS (on the basis of findings) by national programme.

Month wise and village wise number of *P. argentipes* collected by CDC light traps in outdoor and indoor are shown in Table 2. During the study period ten Light traps were installed per village per month in outdoor peridomestic situations. In indoor 18 traps were installed per month per village in different height in HD and CS. In outdoor no *P. argentipes* were trapped in the month of January 17 and Maximum numbers were recorded 122 (average 4.06/ trap/night) and 138 (average 4.6/trap/night) in the months of May and June 2017 respectively. In indoor collection only 2 *P. argentipes* were trapped (average 0.03/ trap/night) in the month of January 2017. Maximum numbers were trapped 591 (Average 10.94/trap/night) in September'17, 494 (Average 9.14/trap/night) in October 2016 and 341 (average 6.31/night/trap) in the month of May 2017. Significant numbers of *P. argentipes* were found in outdoor (*p* value 0.001) in comparison to indoor (Table 2).

In the present study levels irrespective of IRS and non IRS villages *P. argentipes* found in greater numbers in cattle sheds than human dwellings. In general significant numbers (*p* value 0.47) were found above 1.8 meter height. They are also found in significant numbers (*p* value 0.001) in outdoor peridomestic situations. Therefore it is the time to evaluate vector control strategies in kala azar endemic areas of Bihar for successful elimination of the disease from India.

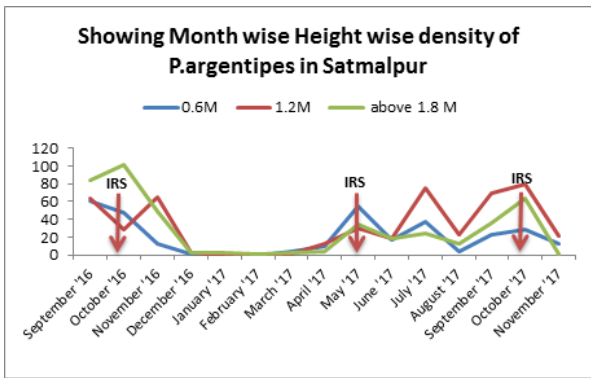
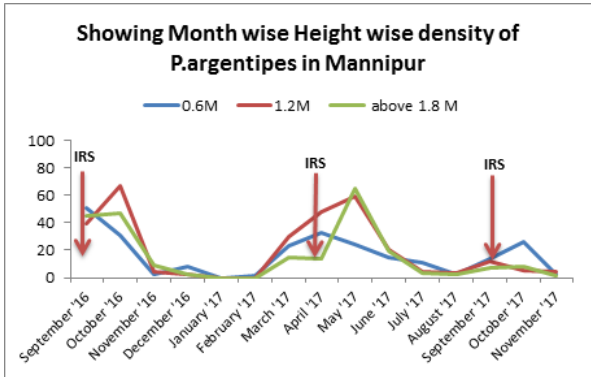
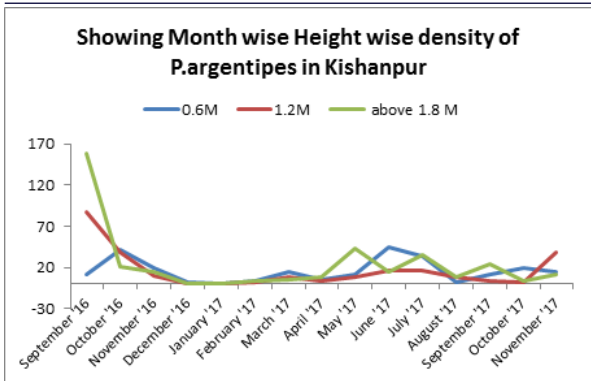
Table 2. Month wise traps wise collection of *P. argentipes* in Kishanpur, Mannipur, Satmalpur, Bihar.

Month & Year	Human Dwellings			Cattle Sheds			Human Dwellings			Cattle Sheds		
	I	II	Total	I	II	Total	I	II	Total	I	II	Total
September '16	11	13	24	33	31	64	11	13	24	33	31	64
October '16	8	17	25	33	31	64	11	13	24	33	31	64
November '16	6	12	18	15	15	30	6	12	18	15	15	30
December '16	0	0	0	0	0	0	0	0	0	0	0	0
January '17	0	0	0	0	0	0	0	0	0	0	0	0
February '17	3	0	3	1	0	1	3	0	3	1	0	1
March '17	22	3	25	7	0	7	29	3	32	14	0	14
April '17	14	24	38	31	38	69	18	24	42	31	38	69
May '17	19	77	96	26	122	148	63	148	211	130	341	
June '17	62	41	103	35	138	173	76	50	126	90	216	
July '17	25	24	49	46	95	141	84	18	102	138	240	
August '17	1	7	8	19	7	26	19	7	26	40	66	
September '17	4	43	47	17	64	81	38	30	68	164	232	
October '17	17	47	64	19	83	102	25	5	30	173	203	
November '17	3	7	10	22	32	42	58	7	65	72	100	
Total	111	133	244	333	311	644	111	133	244	333	311	644

Abbreviations: I = Height up to 0.6 meter (2 feet). II= Height up to 1.4 meters (4 feet). III= Height above 1.8 meters (6 feet).

Table 2 : Month wise density of *P. argentipes* trapped from Indoor and Out door

Month & Year	OUTDOOR				Avg den /N/T/Vill	INDOOR				Avg den /N/T/Vill	OD:ID
	Kishanpur	Mannipur	Satmalpur	Total		Kishanpur	Mannipur	Satmalpur	Total		
September '16	14	32	31	77	2.56	258	135	198	591	10.94	01:05
October '16	8	11	19	38	1.26	102	145	247	494	9.14	01:09
November '16	6	12	24	42	1.4	43	15	171	229	4.24	01:04
December '16	0	0	2	2	0.06	4	16	5	25	0.46	03:23
January '17	0	0	0	0	0	0	0	2	2	0.03	00:03
February '17	3	0	1	4	0.13	8	3	0	11	0.2	13:20
March '17	22	3	7	32	1.06	29	68	129	226	4.18	01:04
April '17	14	38	31	83	2.76	18	95	24	137	2.53	01:01
May '17	19	77	26	122	4.06	63	148	130	341	6.31	02:03
June '17	62	41	35	138	4.6	76	50	90	216	4	01:01
July '17	25	24	46	95	3.16	84	18	138	240	4.44	03:04
August '17	1	7	19	27	0.09	19	7	40	66	1.22	00:01
September '17	4	43	17	64	2.13	38	30	164	232	4.29	01:02
October '17	17	47	19	83	2.76	25	5	173	203	3.75	02:03
November '17	3	7	22	32	1.06	58	7	35	100	1.85	01:01



A.K. Vertical distribution of indoor resting adult *Phlebotomus* (*Euphlebotomus*) *argentipes* the vector of kala azar in Kala azar endemic foci of Bihar, India. *Int. J. Curr. Res. Med. Sci.* (2017). 3(1): 1-6.

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