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Research Article

Seasonal Prevalence and Host Preference of Some Medically Important *Aedes* Species of Doon Valley, India

Ritwik Mondal[1], N Pemola Devi[2], Sajal Bhattacharya[3]

[1]Department of Zoology, University of North Bengal, West Bengal, India.
[2]Department of Zoology, D.B.S. (P.G.) College, Dehradun, Uttarakhand, India.
[3]Department of Zoology, Asutosh College (University of Calcutta), Kolkata, West Bengal, India.
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Corresponding Author:
Ritwik Mondal, Department of Zoology, University of North Bengal, West Bengal, India.
E-mail Id:
ritwikm.zoology@nbu.ac.in
Orcid Id:
https://orcid.org/0000-0003-3655-6767
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A B S T R A C T

In the recent past, several outbreaks of mosquito-borne diseases in Dehradun city make it necessary for a comprehensive understanding of man-vector contacts, disease transmission and determination of appropriate resting sites for adult mosquitoes. The study was conducted within the periphery of Dehradun city in Doon valley, Uttarakhand state, India consisting of eight (08) locations namely Sahaspur, Premnagar, Raipur, Karanpur, Sahastradhara, Dudhli, Doiwala and Rani Pokhri with the objective to analyze and assess the seasonal prevalence and host choice of the mosquitoes with special reference to *Aedes* species. Among the eight locations of the current study, Raipur registered the highest mosquito abundance (17%) trailed by Sahaspur (16%) and Karanpur (15%). A low abundance of mosquitoes was registered conjointly from Doiwala and Rani Pokhri areas (9% from each area). In these areas, the profile plots of the 2-way ANOVA displayed the highest variation for the data collected from human habitations followed by bovine sheds, whereas the lowest recorded variation was from the poultry. With regards to seasonal prevalence, the highest mosquito abundance was registered during the monsoon followed by the post-monsoon. In provender analysis, a total of 1912 blood meals were collected for antibody test among which 1851 were found positive, with the highest reaction with human antisera (57.75%), followed by bovine antisera (21.34%) and pigs antisera (4.97%). *Ae. aegypti* had the highest Anthropophilic Index (A.I.) with 68.78%, followed by *Ae. albopictus* (64.89%) and *Ae. vittatus* (55.55%). However, Human Blood Predominance (H.B.P.) and Pure Human Blood Pervasiveness (P.H.B.P.) were found highest for *Ae. albopictus* with values 0.43 and 0.44 respectively. The results also show the seasonal prevalence and feeding habits of these vectors. This study would be helpful in formulating season-wise strategies for the control of *Aedes* vector mosquitoes in different eco-epidemiological situations.

Keywords: *Aedes* Species, Double Immunodiffusion, Blood Meal Analysis, Host Preference, Seasonal Prevalence

Introduction

Adult mosquitoes persist within the fringes of human dwellings. However, it is the female mosquitoes that mostly enter the human habitations to get blood meals.[1] The vector capacities of mosquitoes rely upon numerous parameters like biting frequency, dissemination ability, and native profusion; however, the foremost important facet behind the vector capacity lies in host preferences and supported blood meal supply.[2] Mosquitoes belonging to genus *Aedes* perpetually depend on over one host for one gonotrophic cycle, primarily showing their man-landing activity throughout the hours of daylight.[3] Several researchers reported on the impalpable resting habits of *Aedes* species.[4-7] The double immunodiffusion technique has been widely favoured by researchers to determine dipteron blood meal.[8] It is advantageous for the opacity, the murkiness of reactants and purging of the floating debris as these parts get sieved through the gel. Besides that, the technique offers prompt leads to determine the host preferences of mosquitoes in a given neighbourhood.[9,10] Mosquito provender identification has been further in practice to analyse the feeding behaviours of mosquitoes.[11-17]

Given the reportable state of affairs of the several outbreaks of mosquito-borne diseases in Dehradun in the recent past; there is a standing purpose for the evaluation of each man-vector contact, disease transmission and determination of appropriate resting sites for adult mosquitoes. To achieve that goal analysis of multiple blood-feeding behaviours of mosquitoes on different hosts have been recognized as an important step in determining their potential host range and seasonal preferences.[4, 10] This study preponderantly focuses on the analysis and assessment of the seasonal prevalence and host choice of mosquitoes, with special reference to the provender analysis of *Aedes* species, in order to formulate comprehensive management strategies to prevent any potential outbreaks of mosquito-borne diseases.

Methodology

Study Area and Time Period

The study was conducted within the boundary of Dehradun town in Doon valley (latitude 30°19'N, 78°04'E, longitude 77°35´E to 78°20´E) of district Dehradun (Uttarakhand) consisting of eight locations *viz.* Sahaspur, Premnagar, Raipur, Karanpur, Sahastradhara, Dudhli, Doiwala and Rani Pokhri. The study time was alienated into four distinct seasons from Sep-14 to Aug-15 *viz.* Post-Monsoon (Sep-Nov), Winter (Dec-Feb), Summer (Mar-May) and Monsoon (Jun-Aug) (Figure 1).

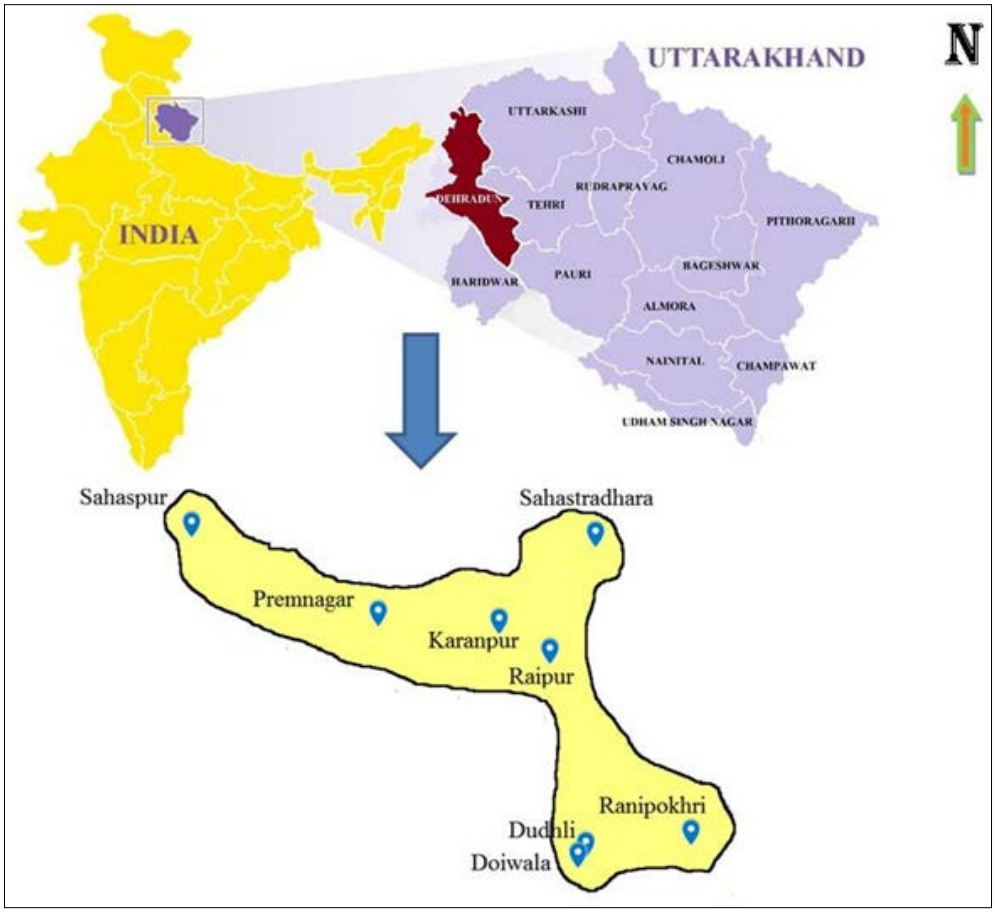

Figure 1.Study Sites on the Periphery of Dehradun District

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Mosquito Surveillance

A door to door survey was performed for the detection of indoor-resting mosquitoes between 06:00 and 08:00 hrs within the morning and between 16:00 and 18:00 hrs in the evening for six min from every habitation weekly, following hand collection methodology and mouth aspirators.[18] Permissions were sought-after from inhabitants to perform indoor-resting mosquito collections for the study. The habitations of the locations were chiefly categorized into four distinct habitations *viz.* human habitations, cattle sheds, pig farm and poultry.

Blood Meal Analysis

Blood meal of mosquito was detected by antibody checking by the agar-gel diffusion technique involving the diffusion of antigen (extract proteins contained in blood from the abdomen of mosquito) and antibody (specific to human, cattle, pig and fowl collected from the Institute of Serology, Kolkata, India) through agar gel so that precipitin reaction leaves a detectable precipitate band according to the outline of Ouchterlony O, Nilson LA.[19]

Blood Meal Estimation

The estimation of blood meal parameters was performed with slight alteration following the protocols developed by Collins RT *et al*.[20]

- The *Anthropophilic Index (A.I.)* is the proportion of antibody from one *Aedes* species that is positive for human blood. The A.I. typically combines information from many different biotopes and thus is also powerfully biased towards one host or another.
- The *Human Blood Predominance (H.B.P.)* refers to the proportion of the full precipitin containing human blood (both mixed and pure) from all species combined, which are comprised of selected species.
- The *Pure Human Blood Pervasiveness (P.H.B.P.)* refers to the precipitin that contains only pure (unmixed) human blood.

Statistical Analysis

A two-way ANOVA check was performed to determine the impact of seasons and habitations over the variation of mosquitoes with a p-value ≤ 0.05 was considered statistically important performed in SPSS (version 17.0).[21] To determine the fluctuation among feeding behaviour of *Aedes* species a two-tailed Z test was performed in XLSTAT (Version 7.5.2) wherever a p-value ≤ 0.05 was considered statistically significant.[21]

Results

Among the 08 locations in the current study, the Raipur area had the highest mosquito abundance (17%) trailed by Sahaspur (16%) and Karanpur (15%) whereas the lowest abundance was registered collectively from Doiwala and Rani Pokhri (with 9% each) (Figure 2). *Aedes* (13.22%) and *Armigeres* (0.17%) mosquitoes were found most prevalent in human habitations throughout monsoon (Jun-Aug) among all the seasons. However, the abundance of *Anopheles* and *Culex* mosquitoes were found highest during the Post-Monsoon (Sep-Nov) among all seasons from human habitats. The abundance of *Anopheles* and *Culex* mosquitoes were 7.65% and 8.04% respectively. The abundance of all the 04 genera of mosquitoes studied recorded lowest during the Winter (Dec-Feb) from poultry (Figure 3). The two-way ANOVA showed that the abundance of mosquitoes considerably varied in keeping with completely different habitats and also as different seasons (p ≤ 0.05).

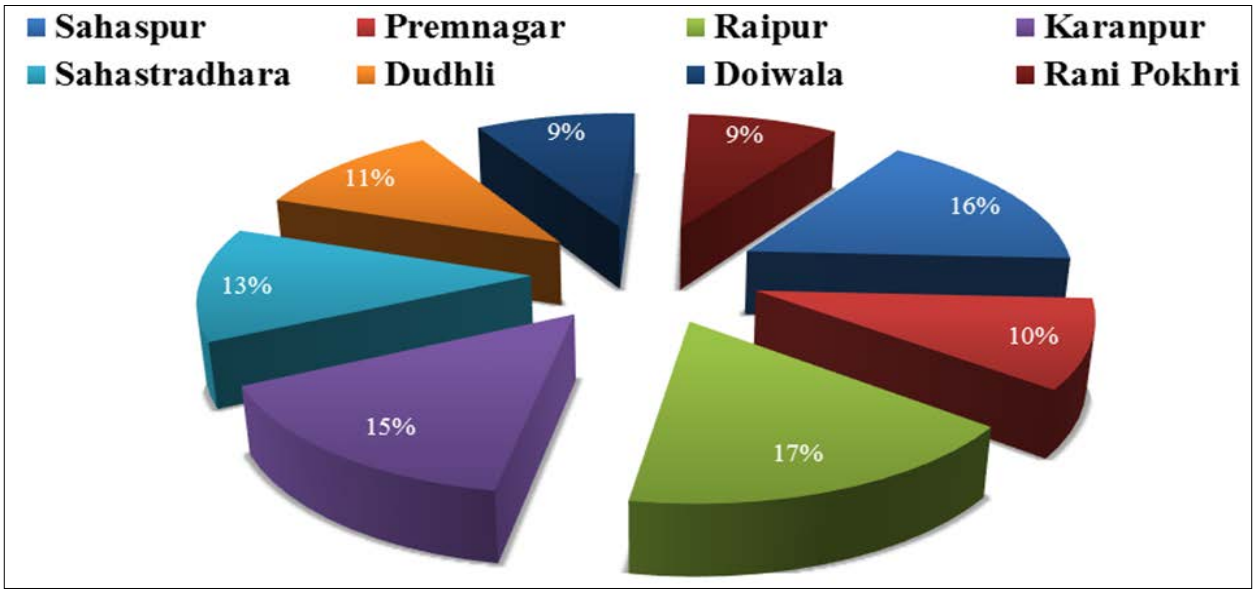

Figure 2.Abundance of the Mosquito Genera Location Wise

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Figure 4, depicts the profile plots of the 2-way analysis of variance estimating the means that of the *Aedes* species, showing the variation is highest for human habitations followed by bovine sheds, whereas recorded lowest within the poultry. Among *Aedes* mosquitoes, season-wise parameters monsoon had the highest variation followed by post-monsoon, whereas the lowest variation among the four habitats was recorded in summer.

Statistical analysis of provender by Z-test disclosed that vital variation ($p \leq 0.05$) was found among the preference of *Aedes* species for the total antibody of human blood is Z = 16.07, among that vital variation for the pure human blood, is Z = 26.919 and mixed-blood is Z = 2.848. In infeed analysis, a total of 1912 blood meals were checked for antibody test, and among them, 1851 samples were found positive. The best reaction was found with human antisera 1069 (57.75%), followed by bovine antisera 395 (21.34%) and the lowest from poultry antisera 92 (4.97%). Mixed blood accounted for 127 positive blood meals, consisting of 6.86% of the total positive samples (Table 1).

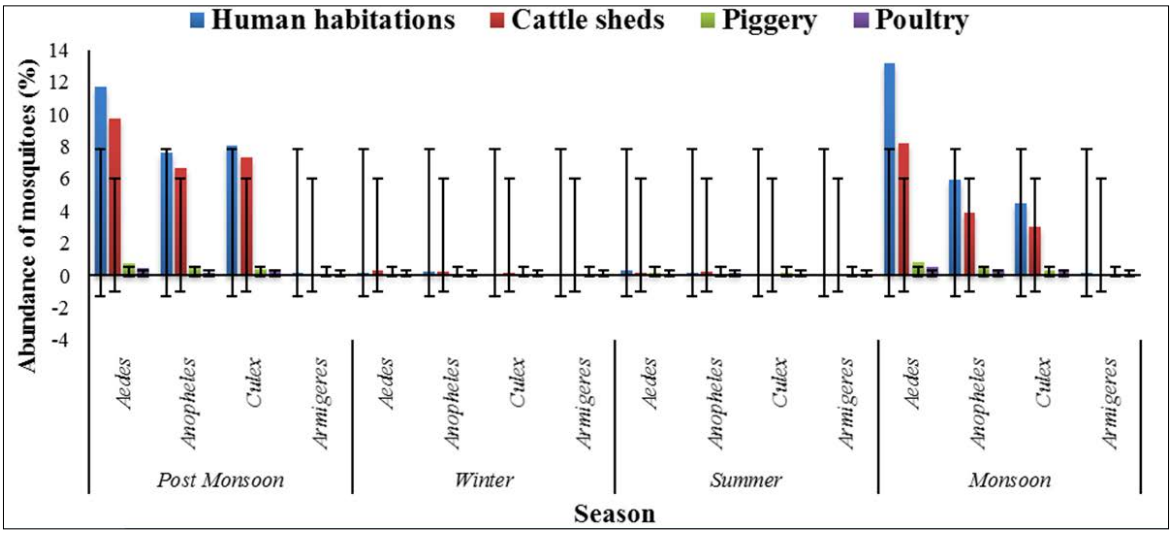

Figure 3.Mosquito Abundance (%) in Different Seasons at 04 Categorized Distinct Habitations

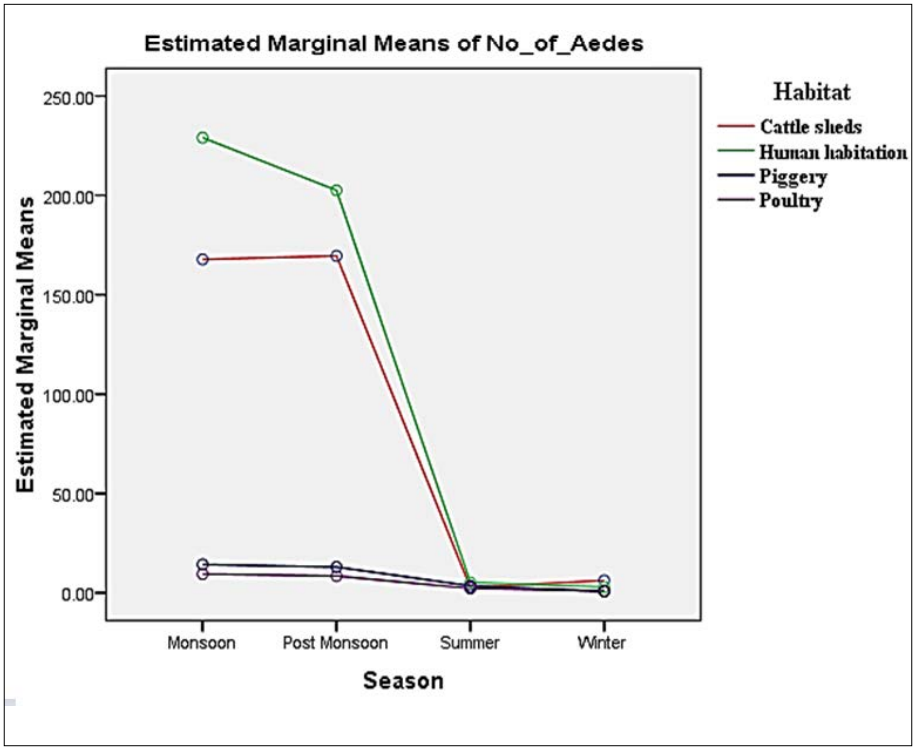

Figure 4.Profile Plot of No. of *Aedes* Species in 04 Distinct Habitats during Different Seasons (from Sep-14 to Aug-15) around the Periphery of Dehradun City

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Table 1.Blood Meal Analysis of *Aedes* Species from Sep-14 to Aug-15

Seasons	*Aedes* species	Blood meal tested	Blood meal +ve	Number and % positive for respective antisera				
				Human	Bovine	Pig	Fowl	Mixed
Post monsoon	*Ae. aegypti*	312	303	192 (63.37%)	55 (18.15%)	14 (4.62%)	7 (2.31%)	35 (11.55%)
	Ae. albopictus	352	343	206 (60.06%)	73 (21.28%)	29 (8.45%)	18 (5.25%)	17 (4.96%)
	Ae. vittatus	187	179	91 (50.84%)	47 (26.26%)	22 (12.29%)	16 (8.94%)	3 (1.68%)
Winter	*Ae. aegypti*	12	8	4 (50.0%)	2 (25.0%)	1 (12.5%)	0	1 (12.5%)
	Ae. albopictus	8	6	2 (33.33%)	3 (50.0%)	0	1 (16.67%)	0
	Ae. vittatus	7	5	2 (40.0%)	1 (20.0%)	1 (20.0%)	0	1 (20.0%)
Summer	*Ae. aegypti*	17	10	3 (30.0%)	2 (20.0%)	2 (20.0%)	1 (10.0%)	2 (20.0%)
	Ae. albopictus	21	19	10 (52.63%)	5 (26.32%)	0	2 (10.53%)	2 (10.53%)
	Ae. vittatus	11	8	3 (37.50%)	2 (25.0%)	1 (12.5%)	1 (12.5%)	1 (12.5%)
Monsoon	*Ae. aegypti*	394	390	211 (54.10%)	81 (20.77%)	33 (8.46%)	24 (6.15%)	41 (10.51%)
	Ae. albopictus	429	421	256 (60.81%)	83 (19.71%)	46 (10.93%)	17 (4.04%)	19 (4.51%)
	Ae. vittatus	162	159	89 (55.97%)	41 (25.79%)	19 (11.95%)	5 (3.14%)	5 (3.14%)
Total		1912	1851	1069 (57.75%)	395 (21.34%)	168 (9.08%)	92 (4.97%)	127 (6.86%)

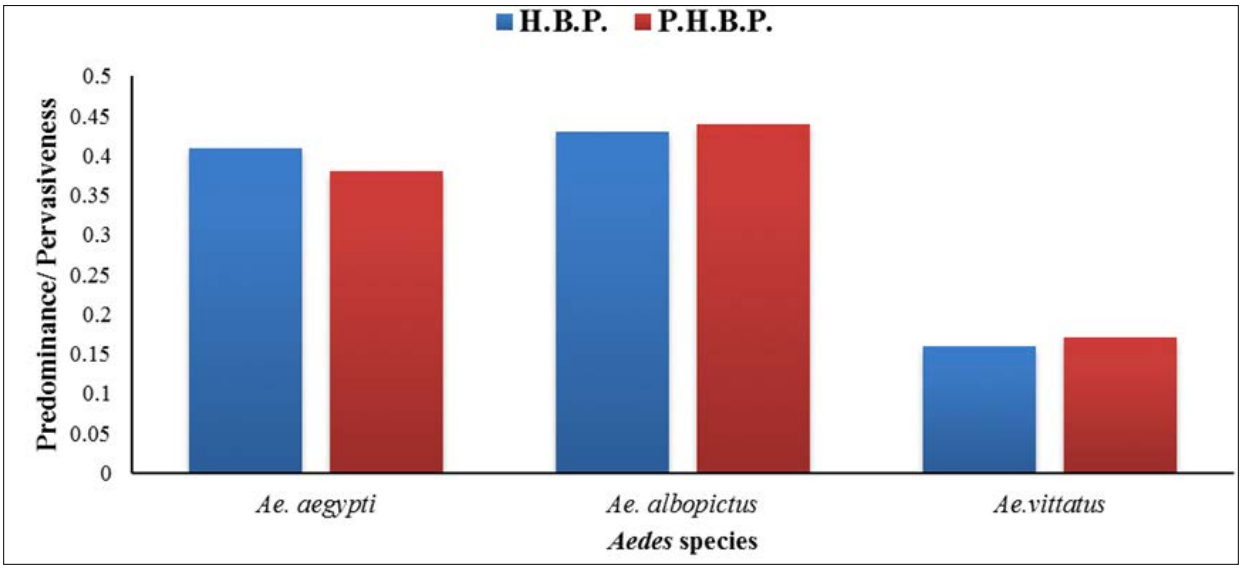

Figure 5.Comparison Showing between Human Blood Predominance (H.B.P.) and Pure Human Blood Pervasiveness (P.H.B.P.) among 03 Species of *Aedes* during the Study Period

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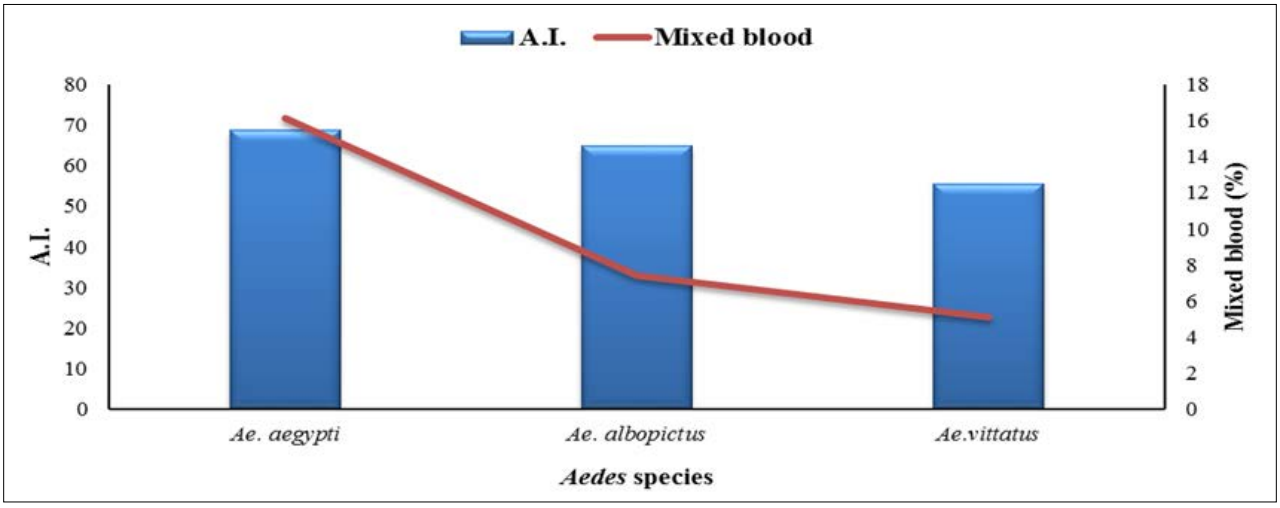

Figure 6.Column-line Diagram Depicts the Anthropophilic Index (A.I.) and Mixed-blood Composition among 03 Species of *Aedes*

Figure 5 depicts the comparison between Human Blood Predominance (H.B.P.) and Pure Human Blood Pervasiveness (P.H.B.P.) among the reported 03 *Aedes* species. Human Blood Predominance (H.B.P.) and Pure Human Blood Pervasiveness (P.H.B.P.) were found highest for *Ae. albopictus* with 0.43 and 0.44 values and lowest for *Ae. vittatus* with the worth of 0.16 and 0.17 respectively (Figure 5). *Ae. aegypti* recorded highest for A.I. with (68.78%), followed by *Ae. albopictus* (64.89%) and *Ae. vittatus* (55.55%). The constant hierarchal pattern was seen in the mixed-blood analysis *viz. Ae. aegypti* (16.16%) > *Ae. albopictus* (7.42%) > *Ae. vittatus* (5.13%). Figure 6 portrays a column-based diagram of Anthropophilic Index (A.I.) and mixed-blood composition among 03 species of *Aedes.*

Discussion

Earlier, it was explicit that *Ae. aegypti* is strictly domiciliary, preferring less vegetation, biting inside and primarily found indoors, whereas *Ae. albopictus* and *Ae. vittatus* was found outdoors and breed in varieties of natural containers.[15,16,22] Being contradictory, the results of this study shows all the studied species of *Aedes* were found within the human habitations and cattle sheds considerably. However, in a facet of seasonal prevalence, there are similarities with the studies conducted by a group of researchers in Selangor, Malaysia[4], with the highest abundance of *Aedes* recorded throughout monsoon followed by post-monsoon. A decent range of mosquitoes found in human habitations in conjunction with comparatively less number in kine sheds, farm and poultry, indicating that preferring resting sites could be related to their blood-feeding habits (anthropophagic or zoophagic, etc.).[11,17] The abundance of mosquitoes found within the human habitations in the present study is considerably higher than that of the results of Azmi SA *et al*.,[17] which is suggestive that appropriate and varied breeding sites favour the mosquitoes to make up such high-density populations within the human habitations.

Mixed blood as provender ensuing from two or more feeds may not be necessarily in sequences of feeding, and some blood varieties might remain insufficiently digested, which may affect the origin identification of blood samples.[7,11] However, the tendency of the mosquitoes to take multiple blood meals could get genetically explained. It appears more probable that the mixed feeding behaviour is either an expression of most liked feeding or is caused by interrupting feeding because of anti-mosquito actions taken by the intended host, which ultimately results in a cryptic behaviour.[20,23] The results of multiple blood parameters of this study support the studies previously conducted in Chhattisgarh, however, the results vary within the genus.[10] Once the female *Aedes* reaches a threshold level, the blood-induced-distension of the gut wall triggers a refractory period during which females do not take provenders. Also, if the blood meal initiates vitellogenesis in female mosquitoes, a second inhibition accompanies gametocyte development.[8,14] It was reported from Puerto Rico that *Aedes* mosquitoes processed blood by pre-diuresis throughout blood-feeding and by diuresis on completion of blood-feeding, particularly while resting.[13] The studies by Washino RK, Tempelis CH reflected the very fact on blood meal identification that mosquitoes have developed two host choice methods *viz.* (a) 'fixed' or 'active' and (b) 'expedient' or 'passive' by the passage of time.[3] Species belonging to *Aedes* show a 'fixed' host preference pattern and feed primarily on mammals.[9, 24] This study reveals 90.48% of *Aedes* mosquitoes feeding on a single host, whereas 6.64% counted for multiple hosts feeding for a single gonotrophic cycle. The results indicate that, as humans are excitable hosts before a mosquito becomes engorged several interrupted half meals might occur from humans.[24,25]

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One demerit of the provender identification techniques is a scarce sensitivity to reveal mixed feedings. Nevertheless, every sample of mixed blood meal supports the very fact that one or a lot of feeding taken by mosquitoes. It looks plausible that the propensity for a species to change pronto from one host to another may dilute its vector potential.[20] Thus regardless of wherever the mosquitoes got captured, it is essential to analyze their blood meals to establish a rudimentary understanding of their actual feeding behaviour.

Conclusion

Aedes mosquitoes are vectors of several important diseases such as dengue, chikungunya, Zika and yellow fever. Among these diseases first three are endemic to India. Recurrent epidemics of dengue and chikungunya in different parts of India are causing concerns to the public health authorities. *Ae. aegypti* and *Ae. albopictus* are recognized vectors for these diseases. It has been delineated that *Ae. vittatus* also has the potential to be the vectors for several above-mentioned diseases. The present study throws light on the preferred habitats, seasonal prevalence and feeding habits of these vectors. This study would be helpful in formulating season-wise and area wise strategies for the control of *Aedes* vector mosquitoes in different eco-epidemiological situations.

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Conflict of Interest: None

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