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LARVAL AND ADULT SURVEILLANCE OF MOSQUITOES (DIPTERA: CULICIDAE) IN PABNA, BANGLADESH

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ABSTRACT

An entomological study was conducted at Ishwardi Upazila of Pabna district to identify the species and breeding sites of mosquitoes. Adult mosquitoes were collected using collecting net, human bait, light trap and larvae were collected by ladle spoon, mug, dipper, and pipette from rail junction, bazar, house, cattle shed, rice field, sugarcane field, rice mill, paper mill, drains etc. A total of 2647 mosquito samples including 1426 larvae and 1221 adult mosquitoes were collected from these places. Seven species of larva and eight species of adult mosquitoes under two genera were identified. The recorded genera for both larvae and adults were *Culex* (*Cx*.) and *Anopheles* (*An.*). The collected species for larvae were *Cx. quinquefasciatus*, *Cx. epidesmus*, *Cx.tritaeniorhynchus*, *Cx. mimulus, Cx. sinensis*, *An. vagus* and *An. minimus.* The collected species for adults were *Cx. epidesmus*, *Cx. quinquefasciatus*, *Cx.tritaeniorhynchus*, *Cx. mimulus*, *Cx. sinensis*, *Cx. pallidothorax*, *An. vagus* and *An. minimus*. Among the collected larva, *Culex* was found most prevalent (85.7%) than A*nopheles* (14.3%). Same as larva, adult *Culex* mosquitoes was found most prevalent (84.4%) than A*nopheles* (15.6%). *Cx. quinquefasciatus* (33.5%) found highest among the larval prevalence, in contrast, *Cx. epidesmus* (36.2%) was dominant among adults. On the other hand, *An. vagus* was the most dominant species in both larval and adult surveillance (8.1% and 9.9%, respectively). Both the larva and adult *Culex* mosquitoes were found more prevalent in households (20.3% and 41.2%, respectively), whereas density of *Anopheles* larva was found highest in rice fields (20.6%) and those of adults were concentrated in households (39.5%). The result of this study will help us to develop a sustainable control measures for the mosquitoes and mosquito-borne diseases in the study areas as well as other areas of the country.

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INTRODUCTION

Mosquitoes are unquestionably the most important vectors of diseases (Tandon, 1998). They are important because of the effects on human and animal health and most of all due to the role they play in the transmission of protozoan and viral pathogens. Mosquitoes are regarded as well-known vector of diseases like dengue, Japanese encephalitis, yellow fever, malaria, and filariasis (Bang 1985). They are also important nuisance pests. Mosquito-borne diseases are the major public health problem in Bangladesh (Hossain *et al.* 2000, Ahmed 1987). Over 3,500 species of mosquitoes have already been described from various parts of the world which are grouped in 39 genera and 135 subgenera. More than100 species bite humans. They act as vectors for a number of infectious diseases affecting millions of people per year (Molavi and Afsin, 2003). Others that do not routinely bite humans, but are the vectors for animal diseases, may become disastrous agents for zoonosis of new diseases when their habitats are disturbed, for instance by sudden deforestation (Clements *et al.* 1992) as well as climate change. Furthermore, neither all species of mosquitoes, nor all strains of a given species transmit the same kind of diseases. Their habitats differ and they all transmit the diseases under the same circumstances. For example, some species attack people in houses, and others prefer to attack people walking in forests. Accordingly, in managing public health, knowing which species, even which strains of mosquitoes dealing with which animal is important. Mosquito-borne diseases are becoming important public health problems in Bangladesh. Aslamkhan and Wolfe (1971) gave evidence that filariasis had been present since many years in Bangladesh. Their surveys indicated that bancroftian filariasis is present in all districts with the highest prevalence (14%) in Dinajpur district, especially in the north-west part of the district. *Culex pipiens fatigans* as the primary vector of rural bancroftian filariasis is present in large numbers throughout Bangladesh and it is likely to be the main vector of filariasis all over the country (Wolfe and Aslamkhan, 1971). The study area Pabna district and other areas such as Chittagong Hill Tracts, Rangpur, Barisal districts and Dhaka city has 0.9% infection (Wolfe and Aslamkhan, 1971). Eleven species of larva and adult mosquitoes under three genera (*Culex, Anopheles* and *Aedes*) were identified in Bangladesh Agricultural University and adjacent areas by Akter et al, (2014), Alam et al. (2015) and Farjana et al. (2015). Ten species of larva and adult *Culex* and *Anopheles* were identified by Anik and Farjana (2019) from Bagha Upazilla, Rajshahi.

Mosquito surveillance is a prerequisite to an effective, efficient, and environmentally sound mosquito control program. Surveillance is used to define the nature and extent of the mosquito problem and to gauge daily mosquito control operations. It provides a basis for evaluating the effectiveness of control operations, the data needed to comply with state rules and regulations regarding the justification for treatments, and a basis for evaluating the potential for transmission of mosquito borne diseases. Mosquito surveillance is most effective when combined with an ongoing program for monitoring meteorological, astronomical, and environmental factors that may influence mosquito population change. Therefore, emphasis should be given on the effective and integrated vector control studies through detail knowledge of prevalence and species distribution of mosquito vectors in certain localities. This study was therefore, designed to identify the different mosquito species and their breeding sites at Ishwardi upazila in Pabna district of Bangladesh.

MATERIALS AND METHODS

The present study was conducted to record the status of mosquito species as well as their breeding sites and larval habitat in Ishwardi Upazila and its surrounding areas (Saragopalpur, Pakshey, Dashuria) in Pabna Districts of Bangladesh. Ishwardi has a tropical type climate, with a hot and rainy summer and a pronounced dry season in the cooler months. January is the coolest month of the year, with temperatures averaging near 28◦ C (82.4◦ F), and April the warmest month, with temperatures ranging between 33◦ and 36◦ C (91◦ F and 96◦ F). Most places receive more than 1,237 mm (in) of rain a year. Most rain falls during the monsoon (June- September) and little during the dry season (November-February). Ishwardi Upazila located in between 24°03' and 24°15' north latitudes and in between 89°00' and 89°11' east longitudes.

Identification of breeding sites for mosquitoes

Sugarcane field, Sugarcrop research Institute (SRI) drains and small bushy area near households, cattle sheds, paper mills, rice mills, rice fields, Export Processing Zone (EPZ), Bazars, discarded broken rail compartments are considered as favorable habitats of adult mosquito in Ishwardi Upazila. So these places were given priority for larval collection. We searched for the breeding sites of mosquitoes in drains, broken pots, flower tubs, containers, bamboo stump, tree holes, fruit shells, swampy areas, agricultural pits etc in the study areas.

Collection of mosquito larvae

Larvae were collected by ladle spoon, pipette, dipper, mug and dropper from different spot and kept in a labeled plastic bottle and records were taken like collection date and time, and were brought to the laboratory for counting and identification. The collected larvae were brought to the laboratory in small plastic jars or bottle with water. During shipment care was taken so that the mosquito larva does not get shrinked or squished. Sample collection from breeding places was made at different periods of the day and from different situations. The most common and easiest technique to investigate the presence or absence mosquito larvae in a habitat is dipping. While dipping, care was taken so that shadow was cast away from the habitat as larvae are very sensitive and will dive to the bottom once shadow is cast on the water. The dipper was gently lowered in an angle of 45° just below the surface so that water flows in together with any larvae that might be present.

Collection of adult mosquitoes

Adult mosquitoes were collected by insect collecting net, mosquito spray, light trap and human bait etc. Collections were made different times in a day in each spot. Collection of mosquitoes was performed from outside the home by using insect collecting net and also using light traps (LTs) to collect the adults. Mosquitoes were collected by mosquito spray and bed nets from inside the house. Collected adult mosquitoes were kept in labeled plastic pot with 70% alcohol.

Identification of mosquito species

Mosquitoes were identified by simple method and by making permanent slides. In simple method, the sample was taken in a clean glass slide and then one or two drops paraffin was added and examined under Microscope using10X. Mosquitoes (larvae and adult) were identified in the laboratory of Entomology, Dept. of Parasitology, BAU using the keys of Barraud (1934) and Bram (1967). The mosquitoes were identified on the basis of their morphological characteristics.

Statistical analysis

Students “t” test was performed and the significance level of various mosquito species was calculated using statistical package SPSS-11.5.

RESULTS AND DISCUSSION

A total of 2647 mosquito samples including 1426 larvae and 1221 adults were collected from different places of Ishwardi Upazilla during the study. Seven species of larvae and eight species of adult mosquitoes under two genera were identified. The collected mosquitoes were of the family Culicidae. The identified two genera were *Culex* (*Cx*.) and *Anopheles* (An.). The prevalence of *Culex* species were 85.7% and 84.4%; and *Anopheles* were 14.3% and 15.6% in case of larvae and adults, respectively (Table 1). In both cases *Culex* were more abundant than *Anopheles* during the study period and this prevalence of *Culex* species recorded in this study is similar to the findings of Akter et al. (2014), Farjana et al. (2015) and Alam et al. (2015). Presence of *Culex* species in this survey indicates that there is a high risk of filariasis and Japanese encephalitis and the presence of *Anopheles* species indicates the risk of malaria infection in this area. A total of seven larval species were identified in this study, among which 5 species were *Culex* e.g. *Cx. quinquefasciatus*, *Cx. epidesmus*, *Cx. mimulus*, *Cx. tritaeniorynchus*, *Cx. sinensis*, and two species were *Anopheles* e.g. *An. vagus* and *An. minimus* (Table 1). Among eight identified species of adult mosquitoes, six species were *Culex*, namely, *Cx. quinquefasciatus*, *Cx. epidesmus*, *Cx. mimulus*, *Cx. tritaeniorynchus*, *Cx. pallidothorax*, *Cx. sinensis*, and two species were *An. vagus* and *An. minimus* (Table 2). Among the larvae *Culex* species, the highest prevalence was found in case of *Cx. quinquefasciatus* (33.5%) followed by *Cx. epidesmus* (27.7%), *Cx. tritaeniorynchus* (10.7%), *Cx. mimulus* (8.3%), and *Cx. sinensis* (5.4%). Several previous studies reported that *Cx. quinquefasciatus* is the predominant species in Dhaka (Ameen and Moizuddin, 1973), Mymensingh (Akter et al, 2014, Farjana et al. 2015, Alam et al. 2015) and Rajshahi (Anik and Farjana, 2019). This may because of this species can breed in wide range of habitats including polluted organic-rich water like drains (Akter et al, 2014, Farjana et al. 2015, Alam et al. 2015) and in clear stagnant water like artificial containers (Farjana et al, 2020).

Table 1. Prevalence of mosquito larvae at Ishwardi Upazila, Pabna

Sub family	Species	No. of larva	%
Culicinae	***Cx. quinquefasciatus***	**478**	**33.5**
	Cx. mimulus	119	8.3
	Cx. tritaeniorhynchus	153	10.7
	Cx. epidesmus	395	27.7
	Cx. sinensis	77	5.4
	Sub total	1222	85.7
Anophelinae	*An. vagus*	**116**	**8.1**
	An. minimus	88	6.2
	Sub total	204	14.3
	Total	1426	

The scenario is different in case of adult surveillance where the highest prevalence was found in case of *Cx. epidesmus* (36.2%) followed by *Cx. quinquefasciatus* (28.0%), *Cx. tritaeniorynchus* (8.9%), *Cx. mimulus* (3.9%), *Cx. sinensis* (3.8%) and *Cx. pallidothorax* (3.5%) (Table 2). This difference might be due to period of time and method of collection of adult mosquitoes. In case of *Anopheles* species, both of the larvae and adult survey demonstrated that *An. vagus* (8.1% and 9.9%, respectively) was dominant over *An. minimus* (6.2% and 5.6%, respectively) (Table 1 and 2). *An. vagus* was the common *Anopheles* spp. found in several previous studies in Bangladesh (Akter et al, 2014, Farjana et al. 2015, Alam et al. 2015).

Table 2. Prevalence of adult mosquitoes at Ishwardi Upazila, Pabna

Sub family	Species	No. of adult	%
Culicinae	***Cx. epidesmus***	**442**	**36.2**
	Cx. mimulus	48	3.9
	Cx. tritaeniorhynchus	109	8.9
	Cx. quinquefasciatus	342	28.0
	Cx. pallidothorax	43	3.5
	Cx. sinensis	47	3.8
	Sub total	1031	84.4
Anophelinae	*An. vagus*	**121**	**9.9**
	An. minimus	69	5.7
	Sub total	190	15.6
	Total	1221	

Table 3. Distribution of mosquito larvae in different breeding sites of Iswardi, Pabna

Species		RLJ	BZR	SRI	HH	CS	RFD	SFD	PM	EPZ	RM	Species / genus total
Cx. quinquefasciatus	No.	26	19	29	105	92	67	37	47	26	30	478
	%	5.4	4.0	6.1	**22.0**	19.2	14.0	7.7	9.8	5.4	6.3	33.5
Cx. epidesmus	No	27	17	34	82	81	63	33	21	17	20	395
	%	6.8	4.3	8.6	20.8	**20.5**	15.9	8.4	5.3	4.3	5.1	27.7
Cx. mimulus	No	06	08	07	16	10	30	08	16	08	10	119
	%	5.0	6.7	5.9	13.4	8.4	**25.2**	6.7	13.4	6.7	8.4	8.3
Cx. tritaeniorhynchus	No.	10	05	12	27	29	27	11	8	06	17	153
	%	6.5	3.3	7.8	17.6	**19.0**	17.6	7.2	5.2	3.9	11.1	10.7
Cx. sinensis	No.	03	05	11	18	07	12	02	05	08	06	77
	%	3.9	6.5	14.3	**23.3**	9.1	15.9	2.6	6.5	10.4	7.8	5.4
Subtotal	No.	162	54	93	248	219	199	91	97	65	83	1222
	%	13.3	4.4	7.6	**20.3**	17.9	16.3	7.4	7.9	5.3	7.0	85.7
An. vagus	No.	05	02	02	23	15	30	10	10	10	08	116
	%	4.3	1.7	1.7	19.8	12.9	**25.7**	8.6	8.6	8.6	6.9	8.2
An. minimus	No.	03	02	08	18	19	12	02	12	06	07	88
	%	3.4	2.3	9.1	20.4	**21.6**	13.6	2.3	13.6	6.8	8.0	6.2
Subtotal	No.	8	4	10	41	34	42	13	22	16	15	204
	%	3.9	2.0	4.9	20.1	16.7	**20.6**	6.4	10.8	7.8	7.4	14.3
Total												**1426**

Here; RL= Rail Junction, BZR= Bazar, SRI= Sugarcrop Research Institute, HH = Household, R FD= Rice Field, S FD= Sugarcane Field, CS= Cattle Shed, PM= Paper Mill, EPZ= Export Processing Zone, R M= Rice Mill

In this study, larvae of *Culex* species were found more prevalent in household (20.3%), followed by cattle shed (17.9%) and rice field (16.3%) (Table 3). The highest density of this species in these areas might be due to the presence a lot of aquatic area, drains, small ponds, water containing cow dung, and surrounding by polluted water breeding places and comparatively dark region (Jannat et al, 2005). Larval density was varied among the areas of larva collection in this study. Highest density of larvae of *Cx. quinquefasciatus* and *Cx. sinensis* was found in households (22.0% and 23.3% respectively), *Cx. epidesmus* and *Cx. tritaeniorynchus* was found in cattle shed (20.5% and 19.0%, respectively), and *Cx. mimulus* in rice fields (25.2%) (Table 3). Globally, many studies have displayed that the breeding habits of the *Cx. quinquefasciatus* are water bodies that are not seriously polluted, such as drains near households with relatively clear water, sewer ditches, clear liquid wastes, stagnant water in low-lying land, and small pools (Shaman et al, 2010). The *Cx. quinquefasciatus* has better capacity of adaptation towards diverse breeding sites. Evidence has shown that *Cx. quinquefasciatus* was well adapted to various breeding site types including ditches, catch basins, flowerpots, and buckets with diverse water quality (Weitzel et al, 2015). Blood-meal analysis of wild-caught females of *Cx. epidesmus* showed that the most of them had fed on bovines and temporary pools of rainwater and paddy fields containing freshwater and filamentous green algae were found to be suitable larval habitats (Kanojia et al, 2003). On the other hand, larvae of *Anopheles* species found more prevalent in rice fields (20.6%), followed by households (20.1%). Highest density of larva of *An. vagus* was more prevalent in the rice field (25.7%), whereas *An. minimus* was predominant in cowshed (21.6%) (Table 3). It is proved that the chief breeding places of *Anopheles* are paddy fields, where larvae are found throughout the year in varying densities (Gunathilaka et al, 2015).

Table 4. Distribution of adult mosquitoes in different places in Ishwardi, Pabna

Species		RLJ	BZR	SRI	HH	R FD	S FD	C S	P M	EPZ	R M	Species / genus total
Cx. epidesmus	No.	43	17	21	149	13	21	44	23	57	36	442
	%	9.7	3.8	4.8	**33.7**	2.9	4.8	10.0	5.2	12.9	8.1	36.2
Cx. mimulus	No.	03	02	01	21	04	07	05	04	02	05	48
	%	6.2	4.1	2.1	**43.8**	8.3	14.6	10.4	8.3	4.1	10.4	3.9
Cx. tritaeniorhynchus	No.	06	02	03	62	02	09	13	04	08	09	109
	%	5.5	1.8	2.8	**56.9**	1.8	8.3	11.9	3.7	7.3	8.3	8.9
Cx. quinquefasciatus	No.	17	05	09	166	21	34	47	17	35	23	342
	%	5.0	1.5	2.6	**48.5**	56.5	6.1	13.7	5.0	10.2	6.7	28.0
Cx. pallidothorax	No.	02	-	03	09	02	01	01	10	06	05	43
	%	4.7	-	7.0	20.9	4.7	2.3	2.3	**23.3**	14.0	11.6	3.5
Cx. sinensis	No.	02	01	06	18	01	06	04	06	03	02	47
	%	4.3	2.1	12.8	**38.3**	2.1	12.8	8.5	12.8	6.3	4.3	3.8
Subtotal	*No.*	*226*	*26*	*43*	*425*	*43*	*78*	*114*	*64*	*111*	*80*	*1031*
	%	*21.9*	*2.5*	*4.1*	***41.2***	*4.1*	*7.6*	*11.1*	*6.2*	*10.8*	*7.8*	*84.4*
An. vagus	No.	05	06	09	42	01	01	06	16	20	10	121
	%	4.1	5.0	7.4	**34.7**	0.8	0.8	5.0	13.2	16.5	8.3	9.9
An. minimus	No.	03	03	04	33	01	02	08	04	6	07	69
	%	4.3	4.3	5.8	**47.8**	1.4	2.9	11.6	5.8	8.7	10.1	5.6
Subtotal	No.	8	9	13	**75**	2	3	14	20	26	17	190
	%	4.2	4.7	6.8	**39.5**	1.1	1.6	7.4	10.5	13.7	8.9	15.6
Total												**1221**

Here; RLJ= Rail Junction, BZR= Bazar, SRI= Sugarcrop Research Institute, HH= Household, R FD= Rice Field, S FD= Sugarcane Field, CS= Cattle Shed, PM= Paper Mill, EPZ= Export Processing Zone, R M= Rice mill, Sp= Species

In case of adult surveillance, *Culex* mosquitoes was more prevalent in households (41.2%), followed by railway junction (21.9%) and cowshed (11.1%). Adult *Anopheles* mosquitoes were also found predominant in households (39.5%) like *Culex* mosquitoes. Prevalence of all the species of *Culex* and *Anopheles* were found highest in households except *Cx. pallidothorax* which was predominant in paper mills (23.3%) (Table 4). Rice mill residue inside the drain and comparatively dark region may facilitate the breeding of *Cx. pallidothorax*, though larvae of this species were not found in this study. Poor infrastructure, man-made abandoned container here and there, small bushes, unused plastic and iron particles, abandoned pipes, household dust and residue are the resting habitats of *Culex* larvae and such environment are available in the study sites (Castro *et al* 2010). In this study, though the larvae of *Anopheles* were found more prevalent in rice fields but their adults were dominant in households. Expanding agriculture might produce habitat characteristics favored by *Anopheles* mosquitoes, specifically agricultural uses might increase the availability of the pools of water with little or no surrounding vegetation that are the preferred breeding sites for *Anopheles* mosquitoes (Gunathilaka et al, 2015). Despite the zoophilic and exophilic behavior, *An. vagus* has been shown to oviposit in a range of breeding sites including artificial containers and in close proximity to human habitats (Maheswary et al, 1994), this may be the reason of highest concentration of *An. vagus* in household in this study.

Interestingly, almost all mosquitoes found in this study were present in all areas of collection indicating they share the same habitats. There was a very strong positive association between the habitats of anophelines and culicines in this study. Similar findings have been reported from habitats in rural areas in East and West Africa (Fillinger *et al.* 2004) indicating that there is no clear separation between 'typical' *Anopheles* and *Culex* larval habitats.

CONCLUSION

This survey provides useful information on the prevalence and distribution of mosquito species at Ishwardi Upazila in Pabna district. The mosquito population was found to increase with increase in human or animal population. Environmental pollution also creates favorable condition growth of identified mosquitoes. The results indicate that these places of studied area are very much vulnerable to mosquito-borne diseases. The obtained information will help us for effective mosquito control measures. Although the survey was limited at Ishwardi Upazila and its surrounding in Pabna district, but basic information obtained is also applicable for other regions in Bangladesh.

CONFLICT OF INTEREST

There are no conflicts of interest in the present study.

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