MORPHOMETRIC EVALUATION OF AEDES AEGYPTI (LINNAEUS, 1762) AND CULEX QUIQUEFASCIATUS (SAY, 1823) (DIPTERA: NEMATOCERA: CULICIDAE) OF DIFFERENT METAMORPHOSIS STAGES

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ABSTRACT: Morphometric comparison of *Aedes. aegypti and Culex. quiquefasciatus* larval instars and pupae were done through the measurements of total body length, length of head, thorax and abdomen as well as length and width of different external body parameters. From linear regression of the data from different larval instars showed that the important morphological features were the size of the head, thorax and abdomen of the *Ae. Aegypti* and *Cx. Quinquefasciatus*. They increased significantly with the advanced metamorphosis stages (p≤0.05). Size of the external larval organs viz. antennae, comb spine, siphon, anal papilla were also significantly (p≤0.05) increased during stages of metamorphosis.

Key words: Morphometric comparison, Aedes aegypti and Culex quiquefasciatus.

INTRODUCTION

Morphometric study was a useful alternative technique to identify insect species using anatomical landmarks (Bookstein, 1982). This technique was popular and cost-effective, which requires very little entomological experience compared to standard morphological identification (Lorenz, *et al.*, 2017). More than 3100 species of mosquitoes regarding to 34 genera have been recorded and arranged under three sub families named Anophelinae, Culicinae and Toxorhynchitinae (Knight *et al.* 1977). Mosquitoes were found in almost all different climatic conditions and they can also occupy appropriate places in the environment so they are very successful species in environment (Manimegalai *et al.* 2014) and also were responsible for the transmission of important infectious diseases, like dengue, malaria and zika causing millions of deaths every year and endangering approximately 3 billion people around the world (Wilke *et al.* 2016). *Ae. aegypti* was one of the most widely distributed among other mosquito species in the world and the main vector of many arboviruses. In recent years, viral diseases like Dengue fever and Chikungunya that are transmitted by

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infected female *Ae. aegypti* cause a great threat to public health. (Begum *et al.* 2015). Bangladesh has experienced sporadic dengue fever outbreaks since 1964 until the first large outbreak in 2000 and in 2023 most severe dengue outbreak were observed where nearly 170000 cases were reported and approximately 1000 dengue related deaths were recorded (ACAPS 2023 and Hossain *et al.* 2000). Another mosquito-borne illness of humans that is also considered as a public health threat is Chikungunya fever (CF). Chikungunya virus (CHIKV) causes CF. The main vectors of chikungunya in Asia and the Indian Ocean islands was *Ae. aegypti* (Pialoux *et al.* 2007). The Chikungunya virus was first recorded in Bangladesh at Rajshahi and Chapainawabganj in 2008. A massive outbreak of CF with clinically confirmed cases of > 13000 were documented in Dhaka, Bangladesh during the period of April–September 2017. (Hassan *et al.* 2014 and Amin *et al.* 2022).

Another important vector of the insect family culicidae was Cx. quiquefasciatus which was one of the main vectors of West Nile virus, Japanese encephalitis, avian malaria virus of birds, humans and other animals. It played a major role in the transmission of the nocturnal periodic form of Bancroftian filariasis as an obligatory ectoparasitic vector, all over the world (WHO, 1972). Vector capacity varies between different mosquito species, e.g. species-specific host preferences (Börstler, et al., 2016) or vector competence (Vogels 2017). Accurate species identification was essential to understand patterns of pathogen transmission. Mosquitoes are commonly identified by morphology, (Becker et al. 2010). Thereby, an amount of considerable knowledge is required for the proper identification of the different taxonomic characters of different variation of species. To identify vector mosquitos precisely, morphometric tools can be very promising. The identification mostly depended upon the adult female and 4th instar larvae stages (Choudhury et al. 1997). The present research describes the morphometric comparison between different larval and pupal stages of Ae. aegypti that reared in laboratory and Cx. guiguefasciatus, collected from Dhaka University campus area.

MATERIAL AND METHODS

Larval *Ae. aegypti* and *Cx. quiquefasciatus* and, were collected from various regions of Dhaka University campus area and reared in laboratory condition (25±6°C temperature and 70-80% relative humidity). The rest of the work were also done in the Entomology laboratory, Department of Zoology, University of Dhaka. The total research work was done from April, 2017 to March 2018. Eggs of *Ae. aegypti* were collected four days after blood feeding, by placing an ovipositor cup containing tap water and a particularly submerged fluted coffee filter. To hatch the eggs, the egg strap was placed in a 2-L side-arm flask

containing 1L of distilled water and placed under a laboratory house vacuum for 1h to deoxygenate the water. After the deoxygenation step, the sample were poured the flask contents into an empty larval flat and a pinch (approximately 0.2g) of ground larval food.

The food was prepared at the ratio of 6:4 of dried yeast and water. Larvae shed their exoskeleton four times. The entire larval stage lasts from 6-8 days. With a favorable temperature (26-30°C), the 4th instar larvae began to pupate. After the pupal stage the adults were emerged. Upon emergence, adult mosquitos are collected by an aspirator. The egg rafts of *Cx. quinquefasciatus* were transferred to a larval flat tray filled with 1L of tap water and a pinch (approximately 0.2g) of ground larval food. Then the intact egg rafts were transferred gently and picked up with a wooden applicator stick and placed them of the larval flat in the same orientation as in the ovipositor cup.

For morphometric comparisons, specimens were dissected under a binocular stereo microscope at a suitable magnification (20X). Various body parts were soaked in 70% alcoholic eosin overnight for staining. Larval, pupal and adult mosquitoes were dissected in the same way. The dissected parts were subsequently mounted on slides in Canadian balsam and measured by an ocular and stage micrometer, using two magnifications (10X). The adult mosquito-heads were separated from the body and placed on a slide. The head capsules were also removed carefully under a binocular stereo microscope at a suitable magnification (10X) and measured same manner as stated above. Photographs were taken using a Cannon Powershot 200 digital camera. Morphometric comparisons were done on two species of Nematocera through observation of the variable markers. Morphometric analyses of the adult Ae. aegypti, and Cx. quinquefasciatus included comparative study of the length of proboscis and antenna, maxillary pulp, thoracic appendages (fore leg, mid leg, hind leg), wing length and width and head, thorax, abdomen. The morphometric analyses of four larval instars included the comparative study of the length of antenna, length and width of anal papilla, siphon, number of comb spine and head, thorax, abdomen. The morphometric comparison of different body parts of the pupal stages (the length of antenna, respiratory trumpets, length and width of anal papillae, cephalothorax and abdomen) were also performed.

Mean values of the length and width of the head, thorax and abdomen of larva, pupa and adult of same mosquito species were compared by Tukey's Honest Significance Difference (HSD) test. Mean values of the length and width of various larval and pupal organs of both species were also compared by paired t-test. The mean value of length and width of the pupal and adult cephalothorax, abdomen and other organs were compared by same manner. Simple linear regression lines were also produced using the size the different larval organs of the four larval instars.

RESULTS AND DISCUSSION

Results of the morphometric study of different bodyparts of two insect species, *Ae. aegypti, Cx. quinquefasciatus* were stated as following figures and tables.

Table 1. Lengths and width of	various body regions of four	larval instars of Ae. aegypti
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Regions	Parameter	Larval instars of Ae. Aegypti					
	(mm)	1 st	2 nd	3rd	4 th		
Head	Length	0.28±0.03a	0.35±0.02b	0.55±0.02c	0.88±0.01d		
	Width	0.24±0.004	0.42±0.01	0.008±0.65	1±0.003		
Neck	Length	0.07±0.002	0.08±0.004	0.002±0.09	0.11±0.002		
	Width	0.11±0.011	0.18±0.025	0.009±0.27	0.46±0.002		
Thorax	Length	0.28±0.04a	0.43±0.02b	0.67±0.03c	1.11±0.003d		
	Width	0.25±0.001	0.49±0.002	0.002±0.75	1.456±0.04		
Abdomen	Length	1.56±0.07a	2.54±0.14b	0.15±3.95c	4.95±0.23d		
	Width	0.27±0.01	0.55±0.003	0.007±0.82	0.73±0.43		

*Mean value indicated as different letters (a,b,c,d) are significantly different from each other (p< 0.05)

 Table 2. Lengths and width of various body regions of four larval instars of Cx.

 Quinquefasciatus

Regions	Parameter	Larval instars of Cx. quinquefasciatus					
	(mm)	1 st	2 nd	3rd	4 th		
Head	Length	0.14±0.01a	0.32±0.02b	0.43±0.01c	0.67±0.06d		
	Width	0.24±0.02	0.36±0.03	0.61±0.02	0.86±0.02		
Neck	Length	0.06±0.003	0.07±0.002	0.08±0.002	0.1±0.004		
	Width	0.27±0.38	0.13±0.004	0.19±0.01	0.4±0.04		
Thorax	Length	0.16±0.01a	0.386±0.04b	0.53±0.01c	0.77±0.02d		
	Width	0.23±0.01	0.4±0.02	0.65±0.02	1.13±0.08		
Abdomen	Length	0.91±0.10a	1.809±0.08b	2.11±0.13c	3.91±0.79d		
	Width	0.23±0.02	0.38±0.02	0.58±0.05	1±0.01		

*Mean value indicated as different letters (a,b,c,d) were significantly different from each other ($p\leq 0.05$).

Table 3. Morphometric data of various larval organs of different larval instars of Ae. aegypti

Larval organs	Parameter	Larval instars of Ae. aegypti				
		1 st	2 nd	3rd	4 th	
Antennae	Length (mm)	0.11±0.004a	0.17±0.002b	0.28±0.001c	0.36±0.003d	
Comb spine	Number	4.4±0.52a	7.3±0.82b	8.9±0.74c	9.7±0.95d	
Shipon	Length(mm)	0.29±0.003a	0.46±0.003b	0.68±0.01c	0.80±0.01d	
	Width(mm)	0.15±0.003a	0.27±0.08b	0.4±0.003c	0.5±0.01d	
Anal papilla	Length(mm)	0.22±0.004a	0.36±0.01b	0.61±0.004c	0.89±0.02d	
	Width(mm)	0.10±0.002a	$0.10 \pm 0.002 b$	0.16±0.003c	0.2±0.01d	

*Mean value indicated as different letters (a,b,c,d) are significantly different from each other (p<0.05).

Larval organs	Parameter	Larv	atus		
		1 st	2 nd	3rd	4 th
Antennae	Length (mm)	0.1±0.003a	0.16±0.01b	0.22±0.01c	0.32±0.01d
Comb spine	Number	4.2±0.42a	7±0.82b	8.5±0.53c	9.4±1.07d
Shipon	Length(mm)	0.3±0.01a	0.5±0.01b	0.71±0.01c	0.81±0.02d
	Width(mm)	0.12±0.003a	0.21±0.07b	0.32±0.02c	0.36±0.01d
Anal papilla	Length(mm)	0.2±0.003a	0.31±0.01b	0.60±0.02c	0.79±0.02d
	Width(mm)	0.09±0.01a	0.10±0.001b	0.13±0.01c	0.19±0.01d

Table 4. Morphometric data of various larval organs of different larval instars of Cx. quinquefasciatus

*Mean value indicated as different letters (a,b,c,d) are significantly different from each other (p<0.05)

Table 5. Lengths and width of various body regions of pupa of Ae. aegypti and Cx. quinquefasciatus

Regions	Parameter	Ae.	Cx.
	(mm)	aegypti	quinquefasciatus
Cephalothorax	Length	2.13±0.09a	1.93±0.03
Abdomen	Width	2.57±0.08	2.19±0.04
	Length	2.54±0.14b	2.66±0.36
	Width	1.16±0.08	1.06±0.16

*Mean value indicated as different letters (a,b) are significantly different from each other (p< 0.05)

Table 6. Lengths and widths various organs of pupae Ae. aegypti and Cx. quinquefasciatus

Pupal organs	Parameter(mm)	Ae. Aegypti	Cx. quinquefasciatus
Antennae	Length	0.58±0.06	0.49±0.02
Respiratory trumpets	Length	0.32±0.01	0.3±0.03
Paddle	Length	1.02±0.04	0.94±0.03
	Width	0.4±0.01	0.33±0.01

Table	7.	Lengths an	d widths	of var	ious regi	ions of a	dult A	le. aegypt	i and	Cx.	quin	quej	fasci	atus
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Regions	Parameter	Ae.	Cx.
	(mm)	aegypti	quinquefasciatus
Head	Length	0.66±0.03	0.48±0.04
	Width	0.79±0.01	0.66±0.05
Thorax	Length	1.49±0.19	0.95±0.07
	Width	0.8±0.04	0.76±0.04
Abdomen	Length	3.51±0.06	2.84±0.16
	Width	0.74±0.03	0.65±0.03

Average length and width of different body parts of the larvae *Ae. aegypti* were presented in Table 1 and *Cx. quinquefasciatus* in Table 2. Total body length of each larval instar were compared by a simple linear regression line (Fig. 1 and Fig. 2). Results indicated that changes of larval instars also changed the size. The 1st instar larvae of *Ae. aegypti* were smallest (Head 0.23±0.03 mm, thorax 0.28±0.04 mm and abdomen 1.56±0.08 mm long) (Table 1), where, the 4th instar larvae were largest(Head 0.88±0.01 mm, thorax 1.11±0.003 mm and abdomen 4.95±0.23 mm long) (Table 1). Similarly the larval instars of *Cx.*

quinquefasciatus also changed the size. The 1^{st} instar larvae of *Cx.* quinquefasciatus were smallest (Head 0.14±0.01 mm, thorax 0.16±0.01 mm and abdomen 0.91±0.10 mm long) where the 4th instar larvae were biggest (Head 0.67±0.06 mm, thorax 0.77±0.02 mm and abdomen 3.91±0.79 mm long) (Table 2). The results clearly showed that the size of larvae was significantly increased

Table 8. Lenghts and widths of various external body parts of adult Ae. aegypti and Cx. quinquefasciatus

Regions	Adult body parts	Parameter	Ae. Aegypti	Cx. quinquefasciatus
Head	Antennae	Length	1.88±0.11	1.43±0.13
	Pulpi	Length	0.68±0.05	0.57±0.04
	Proboscis	Length	2.21±0.06	2.08±0.06
	Maxillary pulps	Length	0.68±0.05	0.57±0.04
Thorax	Wing	Length	3.67±0.18	2.91±0.13
	-	Width	0.91±0.04	0.88±0.04
	Fore leg	Length	10.40±0.23	9.34±0.61
	Mid leg	Length	11.26±0.19	10.01±0.28
	Hind leg	Length	13.05 ± 0.05	12.05 ± 0.05



Fig. 1. A Simple linear regression line comparing the total body length of the different larval instars of *Ae. aegypti* showed that body length were increased significantly (p≤0.05) with the developmental stages.



Fig. 2. A Simple linear regression line comparing the total body length of the different larval instars of *Cx. quinquefasciatus* showed that body length were increased significantly ($p \le 0.05$) with the developmental stages.



Fig. 3. Percentage of the length of head, thorax and abdomen in proportion to the total body length of *Ae. agypti* larval instars



Larval instars



Fig. 4. Percentage of the length of head, thorax and abdomen in proportion to the total body length of *Cx. quinquefasciatus* larval instars



Fig. 5. Simple linear regression showed the increasing of antennal length with the increased larval development of *Ae. agypti*. Results indicated that length of the antennae of larvae of *Ae. agypti* were increased significantly during larval development ($p\leq0.05$).

during the larval development, $p \le 0.05$ (Fig. 1, Fig. 2). The average sizes of each instar larvae were larger in *Cx. quinquefasciatus* than *Ae. aegypti*. As the larval

instars of *Ae. aegypti* reared in laboratory and sufficient food was given so they emerged larger adults. The change of larval head, neck, thorax and abdomen were clearly observed during larval development. The results clearly showed that the length of larval head, thorax and abdomen were significantly increased in proportion to their body length as per instar stages, $p \le 0.05$ (Fig 3, Fig 4).



Fig. 6. Simple linear regression showing the increasing of antennal length with the increased larval development of *Cx. quiquefasciatus*. Results indicated that length of the antennae of larvae of *Cx. quiquefasciatus* were significantly increased during larval development (p<0.05).



Fig. 7. Simple linear regression showing the increasing of comb spine number with the increased larval development of *Ae. agypti.* Results indicated that number of comb spine of larvae of *Ae. agypti* were significantly increased during larval development (p<0.05).</p>

The significant increases of the length of antennae, siphon and anal papillae and the number of comb spines were noticed with larval development. The length antennae of 1^{st} instar larvae of *Ae. aegypti* were smallest (0.11±0.004 mm, Table 3) and largest (0.36±0.003 mm, Table 3) in 4th instar larvae. Also, in case of *Cx. quinquefasciatus* the length antennae of 1^{st} instar larvae were smallest (0.10±0.003 mm, Table 4) and biggest (0.32±0.001 mm, Table 4) in 4th instar



Larval instars

Fig. 8. Simple linear regression showing the increasing of comb spine number with the increased larval development of *Ae. agypti.* Results indicated that number of comb spine of larvae of *Cx. quinquefasciatus* were significantly increased during larval development ($p \le 0.05$).



Fig. 9. Simple linear regression showing the increasing of length and width of the respiratory siphon with the increased larval development of *Ae. agypti*. Results indicated that length and width of the respiratory siphon of larvae of *Ae. Agypti* were significantly increased during larval development (p≤0.05).



Fig. 10. Simple linear regression showing the increasing of length and width of the respiratory siphon with the increased larval development of *Cx. quiquefasciatus*. Results indicated that length and width of the respiratory siphon of larvae of *Cx. quiquefasciatus* were significantly increased during larval development ($p \le 0.05$).



Fig. 11. Simple linear regression showing the increasing of length and width of the anal papilla with the increased larval development of *Ae. aegypti*. Results indicated that length and width of the respiratory siphon of larvae of *Ae. aegypti* were significantly increased during larval development (p≤0.05).



Fig. 12. Simple linear regression showing the increasing of length and width of the anal papilla with the increased larval development of *Cx. quiquefasciatus*. Results indicated that length and width of the respiratory siphon of larvae of *Cx. quiquefasciatus* were significantly increased during larval development ($p \le 0.05$).



Fig. 13. Percentage of the cephalothorax and abdomen in proportion to the total body length of *Ae. aegypti* and *Cx. quinquefasciatus* pupae.



% of the body parts in proportion to the total body length

Fig. 14. Percentage of the Head, thorax and abdomen in proportion to the total body length of adult *Ae. aegypti* and *Cx. quinquefasciatus.*

larvae. The number of comb spine of 1^{st} instar larvae of *Ae. aegypti* were smallest (4.40±0.52, Table 3) and largest (9.7±0.95, Table 3) in 4th instar larvae.

Also, in the case of *Cx. quinquefasciatus* the number of comb spine of 1st instar larvae were smallest (4.20±0.42, Table 4) and largest (9.4±1.07, Table 4) in 4th instar larvae. The length shipon of 1st instar larvae of *Ae. aegypti* were smallest (0.29±0.003 mm, Table 3) and biggest (0.80±0.01 mm, Table 3) in 4th instar larvae. Also, in the case of *Cx. quinquefasciatus* the length shipon of 1st instar larvae were smallest (0.30±0.01 mm, Table 4) and biggest (0.81±0.2 mm, Table 4) in 4th instar larvae. The length anal papila of 1st instar larvae of *Ae. aegypti* were smallest (0.22±0.004 mm, Table 3) and biggest (0.89±0.02 mm, Table 3) in 4th instar larvae. Also, in the case of *Cx. quinquefasciatus* the length anal papila of 1st instar larvae were smallest (0.20±0.003 mm, Table 4) and largest (0.79±0.02 mm, Table 4) in 4th instar larvae.

The results clearly showed that the length of antennae, number of comb spine, length and width of respiratory siphon and anal papillae were significantly increased as per instar stages as their body size increased per molt, $p \le 0.05$ (Fig. 5, Fig 6, Fig 7, Fig. 8, Fig. 9, Fig. 10, Fig. 11, Fig 12). It was observed that the length of the siphon of *Ae. aegypti* was smaller than that of *Cx. quinquefasciatus* in relation to their body length as per instar stages. There was a correlation between the larval body length and the larval organ size. As a result, the larval organ gradually increased in size with the size of larval body during the larval development. The correlation between larval organs with their body length is more or less similar in both *Ae. aegypti* and *Cx. quinquefasciatus*.

No significant difference was found between the larvae of *Ae. aegypti* and *Cx. quinquefasciatus* in relation to their total body length.

Average length and width of different body parts of the pupae *Ae. aegypti* and *Cx. quinquefasciatus* were presented in Table 5. The average length and width the cephalothorax of *Ae. aegypti* pupae were measured 2.13 ± 0.09 mm and 2.57 ± 0.08 mm, respectively (Table 5). In the case of *Cx. quinquefasciatus* the average length and width the cephalothorax were measured 1.93 ± 0.03 mm and 2.19 ± 0.04 mm, respectively (Table 5). Here, the width of the cephalothorax were larger than the length in both species. The average length of the abdomen of *Ae. aegypti* and *Cx. quinquefasciatus*, pupae were measured 2.54 ± 0.14 mm and 2.66 ± 0.36 mm, respectively (Table 5). The results showed that there was a correlation between the size of body region, cephalothorax, abdomen and their body size of pupae in both species (Table. 5). But there is no significant difference in the length and width of cephalothorax and abdomen in proportion to their body length between the pupae of *Ae. aegypti* and *Cx. quinquefasciatus* (Fig. 13).

Various pupal organs, antennae, respiratory trumpets and paddle of *Ae. aegypti* and *Cx. quinquefasciatus*, were also measured (Table 6). The average length of antennae of *Ae. aegypti* and *Cx. quinquefasciatus* pupae measured 0.58 ± 0.06 mm and 0.49 ± 0.02 mm, respectively (Table 6). The average length of respiratory trumpets of *Ae. aegypti* and *Cx. quinquefasciatus* pupae were measured 0.32 ± 0.01 mm and 0.3 ± 0.03 mm, respectively (Table 6). The average length and width of the paddle of *Ae. aegypti* pupae were measured 1.02 ± 0.04 mm and 0.4 ± 0.01 mm, respectively (Table 6). The average length and width of the paddle of *Cx. quinquefasciatus* pupae were measured 0.33 ± 0.01 mm, respectively (Table 6). The average length and width of the paddle of *Cx. quinquefasciatus* pupae were measured 0.33 ± 0.01 mm, respectively (Table 6). The results showed that there was a correlation between of the size of antennae, respiratory siphon, paddle and their body size of pupae in both species. But there is no significant difference in the length of antennae, respiratory siphon and length and width of paddle in proportion to their body length between the pupae of *Ae. aegypti* and *Cx. quinquefasciatus*.

Average length and width of different body parts of the adult *Ae. aegypti* and *Cx. quinquefasciatus* were presented in Table 7. Result indicated that the sizes of the adult *Ae. aegypti* were larger (Head 0.66 ± 0.03 mm, thorax 1.49 ± 0.19 mm and abdomen 3.51 ± 0.086 mm long) (Table 7) and the size of the adult *Cx. quinquefasciatus* were smaller (Head 0.48 ± 0.04 mm, thorax 0.95 ± 0.07 mm and abdomen 2.84 ± 0.16 mm long) (Table 7). The results also showed that there was a correlation between the size of body region (head, thorax and abdomen) and their body size of adult in both species (Fig. 14, Table. 7). The average length and width of various external body parts (antennae, pulpi, proboscis, maxillary

pupls, wing, fore leg, mid leg and hind leg) of *Ae. aegypti* and *Cx. quinquefasciatus* were presented in Table 8. The average length of antennae of *Ae. aegypti* and *Cx. quinquefasciatus* measured 1.88 ± 0.11 mm and 1.43 ± 0.13 mm, respectively (Table 8). The average length and width of wing of adult *Ae. aegypti were* measured 3.67 ± 0.18 mm and 0.91 ± 0.04 mm, respectively (Table 8). The average length and Cx. quinquefasciatus measured 2.91 ± 0.13 mm and 0.88 ± 0.04 mm, respectively (Table 8). The results showed that there was a correlation between of the size of antennae, proboscis, maxillary pulps, wing, legs and their body size of adult in both species. But there is no significant difference in the length of antennae, proboscis, maxillary pulps, legs and length and width of wing in proportion to their body size between the adult of *Ae. aegypti* and *Cx. quinquefasciatus* also, the sizes of the external organs were larger in adult *Ae. aegypti* than *Cx. quinquefasciatus*.

Presented result offered a sensibly clear picture of the significant dimensions of morphometric features of two species of Nematocera (*Ae. aegypti* and *Cx. quinquefasciatus*). This research showed the variation of size and shape of the above mentioned species. Both species had complete metamorphosis. Four larval instars of *Ae. aegypti* and *Cx. quinquefasciatus* were increased in size during larval development. On hatching, the newly emerged larvae appear transparent. As they grow, they appear dark before molting. In every instar the larvae become transparent immediately after molting and before the next molt the larval cuticle gets darkened. The larval size increases as they grow and molt (Andrew *et al.* 2013).

Comparing with other research works similar to our work we can mention Andrew *et al.* (2013). This study was about the morphology and morphometry of *Ae. aegypti* mosquito at laboratory of St. John's College, Agra. They took 20 male and 20 female adult *Ae. aegypti* for their study. They studied on head, clypeus, vertex, antenna, maxillary palps, thorax, wings, legs and abdomen. Morphometric study was done using Image J software and variance analysed with (ANOVA) and found that the size of head, proboscis, maxillary palp, antenna, thorax, its lyre marking and median longitudinal lines measurements. The size of the wing, legs, abdomen measurements and revealed the morphologic features of *Ae. aegypti* adult in Agra for better understanding of the key characters and also revealed the size of larvae was significantly increased during the larval development (p<0.05) which is also revealed in our research work.

CONCLUSION

The morphometric study revealed that various body parts and overall sizes of different stages of the lifecycle of *Ae. aegypti* and *Cx. quinquefasciatus* were increased significantly ($p \le 0.05$) in size during metamorphosis. Also laboratory

reared larval instars and adults of *Ae. aegypti* are larger than *Cx. quinquefasciatus* larval instars and adults that were collected from various natural sources.

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