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THE IMPACT OF URBANIZATION ON THE PROLIFERATION OF *AEDES AEGYPTI* (DIPTERA: CULICIDAE) MOSQUITO POPULATION IN DHAKA MEGA CITY, BANGLADESH

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ABSTRACT: The impact of urbanization on various parameters related to *Aedes aegypti*, the vector responsible for dengue transmission, was assessed in the present study. Extensive larval population surveys were conducted from May to December 2022 across different urbanization levels in Dhaka city. Our survey was conducted in three areas: Uttara (23°52’ N, 90°22" E), Jatrabari (23°42’ N, 90°25" E), and Keranigonj (23°69’ N, 90°37" E) that characterized urban, suburban, and rural settings, respectively. The urban areas represented highly populated regions with a high concentration of anthropogenic structures, while the suburban areas encompassed a mix of residential and natural landscapes. The rural areas consisted of predominantly natural habitats with minimal human intervention. The findings revealed significant variations in larval abundance, larval development, egg production, and longevity. Notably, the urban areas exhibited the highest larval abundance compared to the suburban and the rural areas. Furthermore, in urban settings, the larval instars demonstrated accelerated growth compared to their counterparts in the suburban and the rural environment. The urban locales also witnessed a greater number of eggs and female adults displayed an extended lifespan compared to the suburban and the rural areas. The cumulative effects of urbanization resulted in heightened larval abundance, faster developmental rates, increased egg production, and prolonged survival period. These factors collectively contribute to an elevated vector capacity of adult mosquitoes. Considering the potential impact of *Ae. aegypti* on public health, it is imperative to establish entomological surveillance and control strategies tailored to diverse urbanization gradients. Developing mosquito control policies become crucial, with a focus on preventative measures to manage *Ae. aegypti* mosquitoes effectively within urban structures.

Key words: *Aedes aegypti*, urbanization, larval abundance, cumulative effect, longevity

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INTRODUCTION

Mosquitoes are crucial vectors of many diseases, including malaria, dengue fever, Zika virus, and yellow fever, causing significant morbidity and mortality worldwide (WHO 2020). Among these disease-transmitting mosquitoes, *Aedes aegypti* has emerged as a major public health concern due to its widespread distribution and ability to transmit several viral diseases (Bhatt *et al.* 2013). Globalization, human travel, and global trade have accelerated the extent of invasive mosquito species (Medlock *et al.* 2012). In addition, anthropogenic changes within the environment including climate change, urbanization, and biodiversity loss favor the proliferation of *Ae. aegypti* and *Ae. albopictus* in urban areas (Wilke *et al.* 2021, Gubler 2011, Rochlin *et al.* 2016). The epidemiology of mosquito-borne diseases remains uncertain as to the extent to which mosquito species are dispersing and how urbanization processes are affecting their abundance.

To control vector different modern procedures have been developed, including sterile insect technique (SIT), *Wolbachia*-based incompatible insect technique (IIT), entomopathogenic fungi and sterility-inducing gene drives (Hoffmann *et al.* 2011, Wei *et al.* 2017, Zheng *et al.* 2019, Shaw and Catteruccia 2019). In spite of control strategies, habitat management of the vector, as a long-established, traditional approach involving the participation of communities, still plays a critical role to fight against vector-borne diseases. Nevertheless, concomitant with climate changes, fast globalization and urbanization, dengue outbreaks mainly transmitted by *Ae. aegypti* have occurred consecutively in Bangladesh specially in Dhaka city. Furthermore, from biological aspect *Ae. aegypti* behaviors are not quite clear in the urbanized ecosystem. This concern poses challenges for the management of *Ae. aegypti* habitats in the urbanized areas.

Urbanization stands out as a significant global trend in the twenty-first century, exerting a crucial influence on the proliferation of mosquito-borne diseases. Recent years have witnessed a surge in dengue outbreaks in Bangladesh, attributed to unplanned urbanization. Specifically, urbanized regions, characterized by elevated temperatures conducive to mosquito larval growth, play a pivotal role in shaping the dynamics of life cycles and disease transmission. The urbanization ecosystem postulates a number of habitats for the dengue vectors, which show potent adaptability in Dhaka city. In 2019, the country experienced the most intense dengue fever outbreak (Ahsan *et al.* 2021) and dengue incidence was higher in Dhaka, compared to rural villages (Salje *et al.* 2019). Notably, every year dengue outbreaks in Bangladesh show a tendency to be confined mostly in Dhaka city (Salje *et al.* 2019). In urban areas, resource availability and the absence of resources that were available earlier to their urbanization are accountable for developing or halting the proliferation of vector mosquitoes (Wilke *et al.* 2021). In natural habitat, many vector species are available but due to their ecology and behavior they are inept at invading and colonizing in an urban area (Wilke *et al.* 2019). To understand which vector species are adapting to urban areas is important not only for executing control strategy but also for improving policy to prevent outbreaks (Wilke *et al.* 2019, Trewin *et al.* 2017). Increased temperature and humidity result in mosquito abundance, activity, and the geographic range of vector-borne diseases (Grimmond *et al.* 2015). Such changes in mosquito ecology can influence their breeding patterns, survival rates, and vector competence, ultimately affecting the spread of mosquito-borne diseases (Paaijmans *et al.* 2013).

Aedes aegypti is an invasive mosquito in Bangladesh (Rahman *et al.* 2021) and exploits tree holes, leaf axils and bamboo stumps as a natural breeding sites along with artificial habitats including water bottle, tire and discarded container for the proliferation of larval growth (Sultana *et al.* 2017). Bangladesh, where dengue is endemic, needs information on when and where to increase surveillance and implement control measures for dengue now and in the future in the context of a changing climate. We hypothesized that larval population abundance, and development time will differ among urban, semi-urban and rural areas. By elucidating the ecological factors influencing mosquito populations and their interactions, will contribute to the development of targeted and efficient strategies for mitigating mosquito-borne diseases. In particular, the identification of urban features that will lead to the decrease of breeding habitats will allow the development of mosquito control strategies optimized to preventatively control mosquitoes in urban areas.

MATERIALS AND METHODS

Survey areas: The larval habitat surveys were carried out in urban, suburban and surrounding rural areas of Dhaka city, Bangladesh. The area was chosen to Identify variations in mosquito ecology and breeding preferences across different landscapes. The average temperature in Dhaka is 25.3 °C, and rainfall is approximately 2055 mm. The dengue epidemic in Dhaka city coincided mostly with temperature and rainfall which acts as a leading factor for the development and breeding of *Ae. aegypti*. Hence, Dhaka is an ideal place for the study of impacts of urbanization on *Ae. aegypti*. Our survey was conducted in three areas that characterized urban, suburban, and rural settings. The urban areas represented highly populated regions with a high concentration of anthropogenic structures, while the suburban areas encompassed a mix of residential and natural landscapes. The rural areas consisted of predominantly natural habitats with minimal human intervention. These diverse study areas allowed us to examine the influence of habitat type on the population dynamics of *Ae. aegypti* mosquitoes. Uttara (23°52’ N, 90°22" E) is an urban area of Dhaka city with a population density of 4,874 people/km2. The land is residential with comfortable modern blocks, commercial buildings, schools, colleges, kitchen markets, hospitals, and filled with trees and grasses. Jatrabari (23°42’ N, 90°25" E) could be a suburban area with a population density of around 6,473 people/km2, and land use includes a mix of residential, small industry, and covered with various trees. Keranigonj (23°69’ N, 90°37" E) is a rural area and has a population density of 4,643 people/km2, where land is mainly cultivated by rice and different vegetable plantings.

Larval sampling: The larval habitat studies were conducted in three areas (Uttara, Jatrabari and, Keranigonj) from May to December 2022 (Fig.1). During this time, breeding habitats of *Ae. aegypti* (e.g. discarded plastic bucket, flower pot, disposable food container, tire, building tool, clay pot, constructed building reservoir, garage, footpath hole, oil container and coconut shell) were observed in the study areas monthly with an entomological survey team. Sampling areas were also classified according to the degree of urbanization: urban, suburban, and rural areas. According to Loibl *et al.* (2011), the sampling areas were categorized: urban areas that were in the town heart; suburban parts that were mostly residential, not densely compacted, and situated near an inside of urban area; and rural areas mainly comprised vegetation area and forest, and their population are not densely compacted. In the study, we inspected the outdoor and surrounding areas for breeding habitats of *Ae. aegypti.* During sampling, the geographic location was detected with the use of a hand-held GPS unit (GARMIN Corporation, Taibei, Taiwan).

The larval abundance of mosquitoes was estimated from a standardized volume of water collected from each habitat using a dipper or pipette. The larval density data provided insights into the abundance of *Ae. aegypti* mosquitoes within the study area. The collected samples were transferred to the entomological laboratory for rearing and species identification. The mosquitoes that emerged from the collected samples were pooled by sample sites according to urban, suburban, and rural areas. The mosquito larvae had been placed at room temperature (26°C) until becoming adult mosquitoes. The developmental stage of each larval instar was observed, and their numbers were counted at each sampling area. The identification of *Ae. aegypti* was made using taxonomic keys suggested by Becker *et al.* (2020).

Fecundity: To estimate the fecundity, five-day-old females were kept together with male for a day for mating and females were provided with blood meal. The blood-fed engorged females were isolated into vials (3 cm diameter × 6 cm height) with a piece of wet filter paper on the bottom and a cotton ball soaked with sugar solution on mesh net covering the vial mouth. All females were tested for oviposition five days later. Number of eggs were counted, and which females laid no eggs were omitted from the analyses.

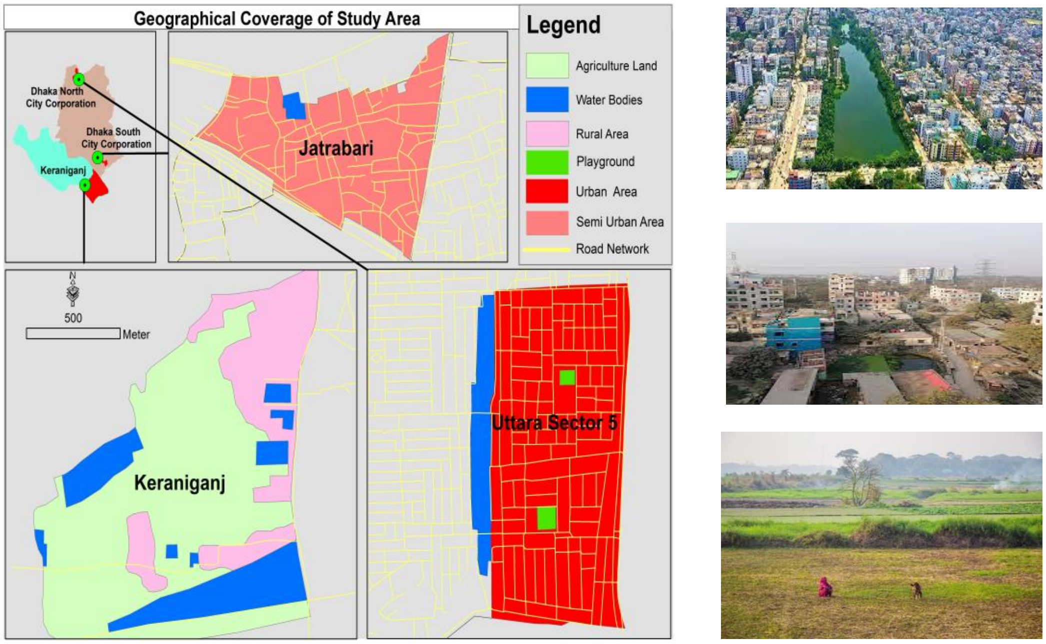

Fig. 1. Map of the study area in Dhaka city, Bangladesh. The study was conducted in three areas from May to December 2022 Urban: Uttara, Suburban: Jatrabari and Rural: Keranigonj.

Longevity: The mosquitoes used for longevity tests were all considered for individuals which were collected originally from the above mentioned three different areas. The emerging females (24 hours old) from each group (i.e. urban, suburban, and rural) were isolated in vials (3 cm diameter × 6 cm height) with a wad of cotton soaked with water to estimate their longevity. The survival rates of the female mosquitoes were monitored daily; and the data collected would provide insights into the longevity and survival dynamics of *Ae. aegypti* in different months.

Data analysis: The data were analyzed to explore the effects of urbanization on the larval abundance, larval development, fecundity and longevity. To determine the significance of differences in larval abundance in each sampling area ANOVA post hoc Tukey's Honestly Significant Difference (HSD) tests were used. Tukey's HSD tests of ANOVA post hoc were also used to further explore the significance differences of different larval instars during development times. Variances in fecundity amongst different zones were tested using one-way analysis of variance (ANOVA). Kaplan-Meier survival analysis was employed to determine the result of different urban settings on adult mosquito survivorship. These statistical analyses were executed using SPSS version 23 (SPSS Inc. IBM) and Graph Pad PRISM (version 8) for support.

Ethics statement: All entomological studies and collections conducted on private residential areas were done with the resident's permission, consent and presence. These studies did not involve endangered or protected species.

RESULTS AND DISCUSSION

Larval abundance: A total of 11 habitat types were inspected at the investigation sites and 2767, 1559, and 1549 *Ae. aegypti* larvae were found in urban, suburban, and rural areas, respectively. In urban areas, the monthly abundance of *Ae. aegypti* larvae was significantly ($P<0.0001$) higher from September to November (Fig. 2). Likewise, the comparison of larval abundance was significantly higher in the suburban area than that of the rural area (Tukey HSD test, $P<0.05$). (Fig. 2). In the present study, the larvae of *Ae. aegypti* were found in all three areas, but the number of *Ae. aegypti* larval presence varied depending on the degree of urbanization. This mosquito species appearing in the urban, suburban and rural areas might be due to artificial breeding sites and the presence of a blood-feeding host range. Bartlett-Healy *et al.* (2012) also reported higher pupal and larval density of *Ae. aegypti* in an urban area. It may be assumed that urban

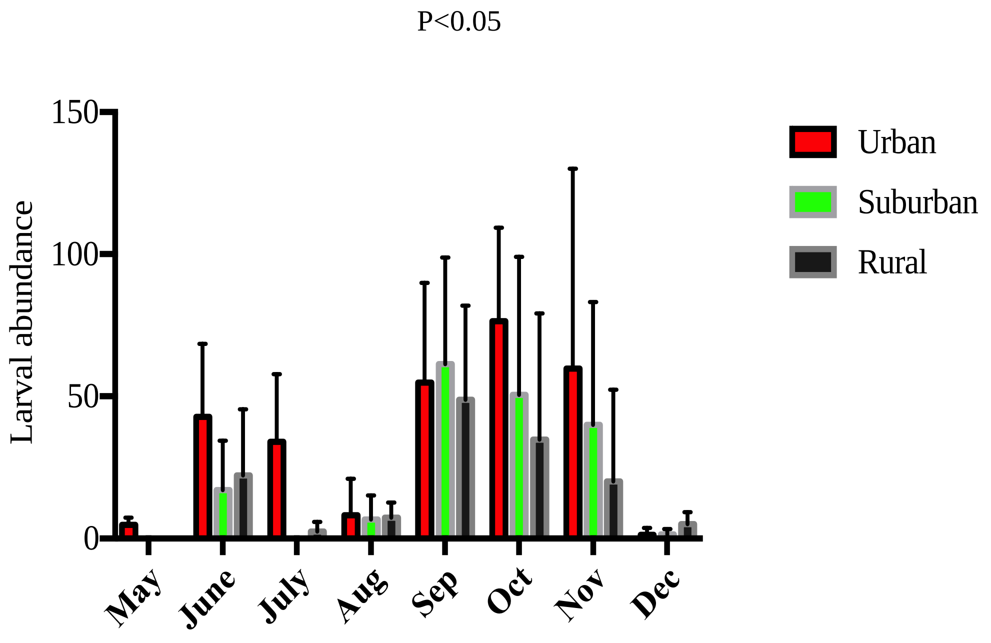

Fig. 2. Larval abundance of *Aedes aegypti* in urban, suburban and rural areas from May to December 2022.

environments provide ample breeding sites for *Aedes* mosquitoes. The larval density in urban areas can be attributed to increased availability of artificial breeding sites, stagnant water in man-made containers, and warmer microclimates (Muhammad *et al.* 2019, Akhtar *et al.* 2016). Moreover, zero or lower densities of immature *Ae. aegypti* mosquitoes were recorded in July and August (Fig. 2) due to the late start monsoon period.

Larval development time: The development time of different larval instars were shown in Figure 3. The development time was significant in different areas (e.g. urban, suburban, and rural) (One-way ANOVA: F= 115, *P*<0.0001) (Fig. 3). Early larval instars (L1 to L2) and late larval instars (L2-L3) developed significantly faster in urban areas compared to suburban and rural areas (Tukey HSD test, all *P*<0.001). However, 3rd and 4th instar took longer development time to develop than first and second in different areas. (Fig. 3). This result explains urbanization might enhance the modification of the environment. Urbanization leads to biodiversity loss triggered by anthropogenic changes that may impact on the mosquito community (Ferraguti *et al.* 2016). By and large, larva development time was remarkably longer in rural areas excluding L3-L4 larval stage. It was speculated that urban areas had less predators, more nutrition from nature, or even less drift from farming insecticides resulting in higher population density of *Ae. aegypti*.

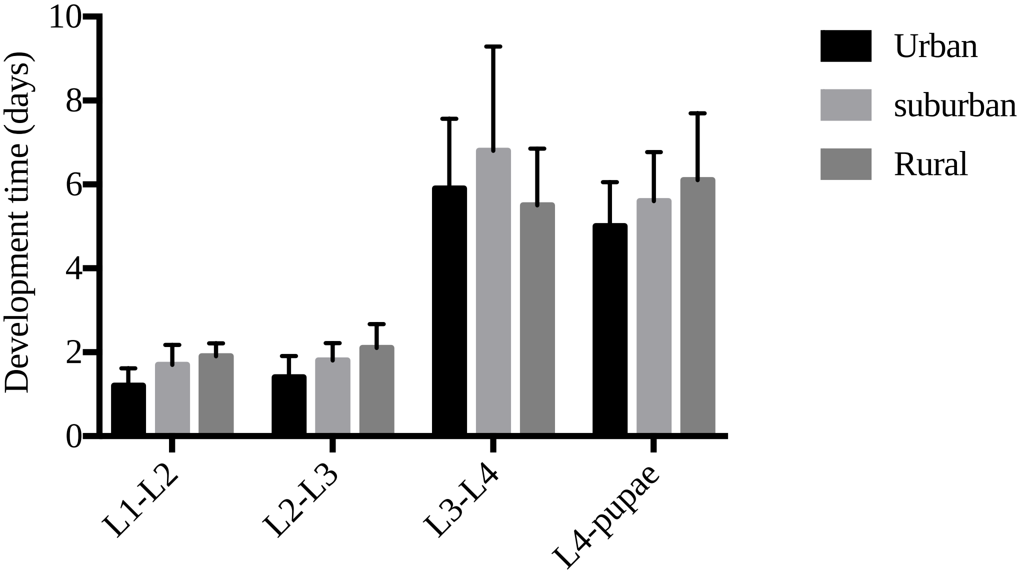

Fig. 3. Development time of *Aedes aegypti* larvae in urban, suburban and rural areas.

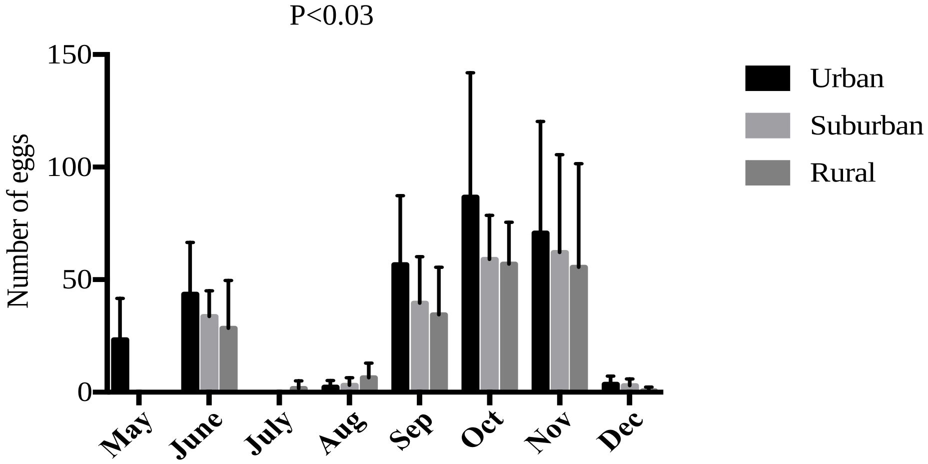

Fig. 4. Number of eggs of adult female *Aedes aegypti* in urban, suburban and rural areas from May to December 2022.

Overall, larval duration in urban areas resulted in faster emergence than in suburban and rural areas ($P < 0.001$) (Fig. 3). Mosquito species adjusted to urban environments resulting in an increase in larval abundance (Wilke *et al.* 2021). However, the extended development time in the rural areas might prolong the larval exposure to mortality factors, including predation, parasites and desiccation prior to adult development.

Number of eggs: The number of eggs was significantly enhanced in the urban areas than in the suburban and rural areas (One-way ANOVA, F = 3.36, $P<0.03$) (Fig. 4). The egg-laying capacity of *Ae. aegypti* gradually increased from September to November 2022 in the urban areas compared to other two areas (Fig. 4). It might be the fact that *Ae. aegypti* species is anthropophilic, and in the urban areas, where the human population is greater, the mosquito species showed high blood-feeding rate, thus changing the oviposition capacity of *Ae. aegypti* (Muhammad *et al.* 2020). In the study, it was observed that there was a negligible or zero number of eggs laid in the months of July and August during monsoon in all the three areas. The female *Ae. aegypti* might have used a strategy to cease her reproductive rate till the favorable season to come. During the following months temperature conditions are adverse, as suggested in the findings of Sultana *et al.* (2024). Sultana *et al.* (2022) also investigated the egg retention ability of *Ae. albopictus* and indicated that the smaller females retained a higher number of embryos compared to the larger females. Egg laying capacity was peaked in between October and November in all three areas (Fig. 4). In the year 2022, Bangladesh encountered an unusual weather pattern resulting in a different ovipositional pattern in *Ae. aegypti.*

Longevity: We observed a seasonal pattern in longevity of *Ae. aegypti* in three different areas. The survival of females from May to August was significantly different among the three sites (Log-rank test: *P*<0.001). The longevity was higher from August to November in urban, suburban and rural areas (Fig. 5, Fig.6 and Fig. 7). In urban areas, we found that female *Ae. aegypti* mosquitoes had the longest life spans than suburban and rural. These results are in agreement with former findings in which adult mosquitoes had the longest survival in urban areas (Li *et al.* 2014). It might be due to the variations of environmental factors including air temperature and humidity in urban areas.

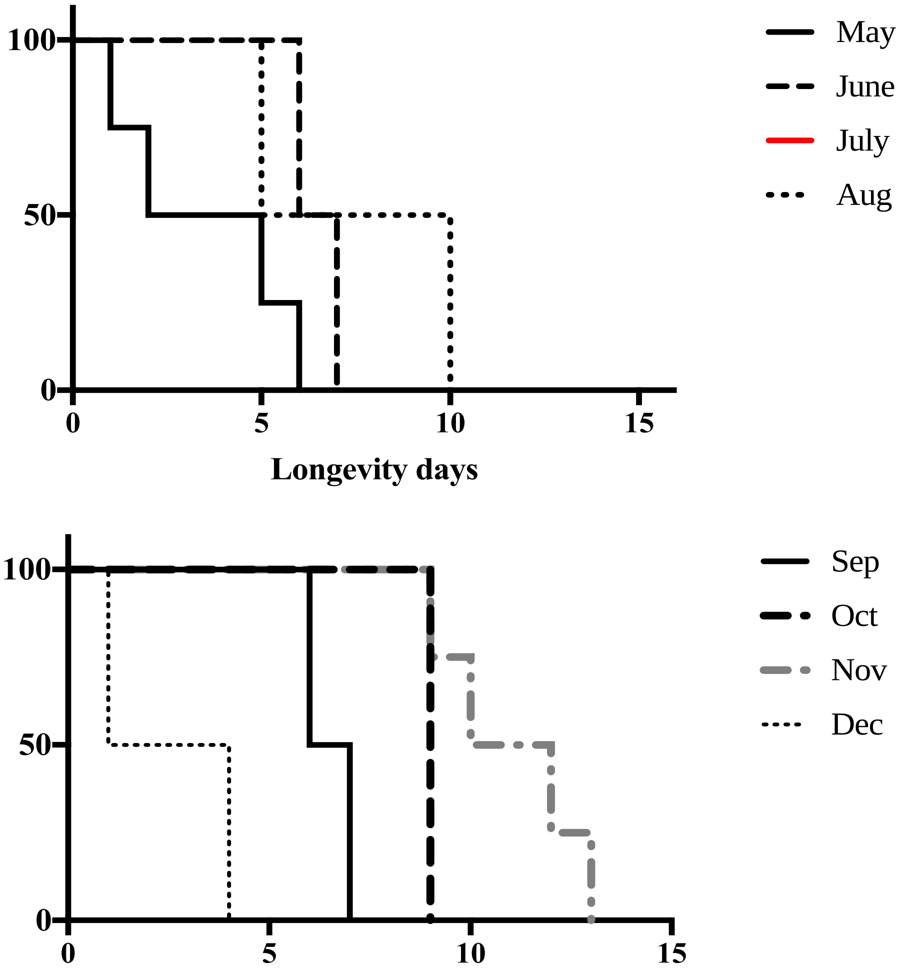

Fig. 5. Kaplan-Meier curves showing the longevity pattern of adult female *Aedes aegypti* in urban areas from May to December 2022.

Additionally, the urban environment provides abundant blood meal sources, such as humans and domestic animals, which could contribute to the increased survival of mosquitoes in urban areas (Shragai *et al.* 2017). From October to November, the female longevity was significantly (Log-rank test: $P<0.001$) long in all three areas (Fig. 5, Fig.6 and Fig. 7). Overall, the longevity was lower in May and December. The weather pattern at the starting and end of the season may shorten mosquito lifespan, as we have observed in our study, which in turn decreased the potential dengue season. One of the warmest years in the past 20 years was observed in 2022 that facilitated the faster transmission of the dengue virus in the community. Survival time was longest in urban areas and smallest in rural areas (Log-rank test: $P<0.04$) (Fig. 8). Prolonged longevity may enhance disease transmission, although the exact relationship between vector and adult life span needs to be explored. Our study helps as a cornerstone for future studies that are required for concluding the exact role of urban features to control vector mosquitoes. Thus, the endeavor will allow the development of targeted mosquito control strategies to be optimized for controlling vector mosquitoes in urban areas.

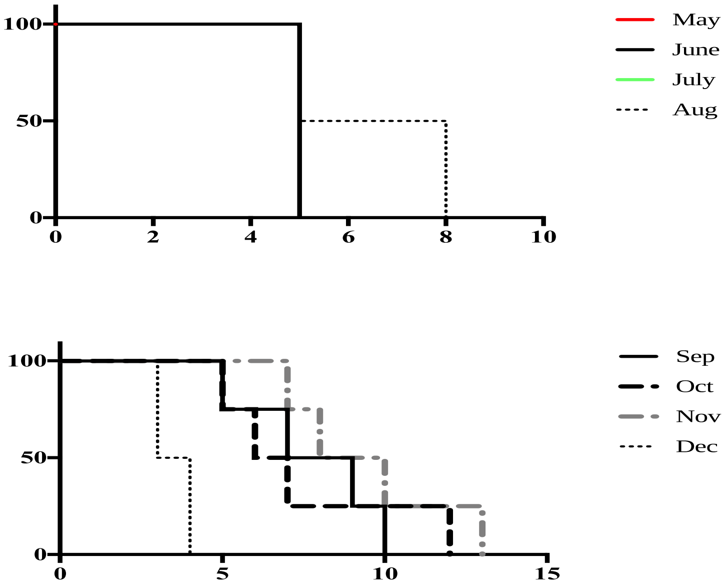

Fig. 6. Kaplan-Meier curves showing the longevity pattern of adult female *Aedes aegypti* in suburban areas from May to December 2022.

Our findings highlight the links between urbanization and the increase of *Ae. aegypti* in Dhaka city. The field sampling reveals that the higher prevalence of *Ae. aegypti* was in densely populated urban areas. In the urbanizing area, the different environments became more suitable for the abundance and development of *Ae. aegypti*. The dense human population produced more kinds of habitation around the living areas that preserve rainwater produces favorable hachure of *Ae. aegypti*. This might be the reason for quick adaptation of *Ae. aegypti* in urban areas. In addition, *Ae. aegypti* is likely to be fully established in rural areas in the upcoming years, initiating nuisance and rising the transmission. Our results also indicate an ongoing plasticity of *Ae. aegypti* in

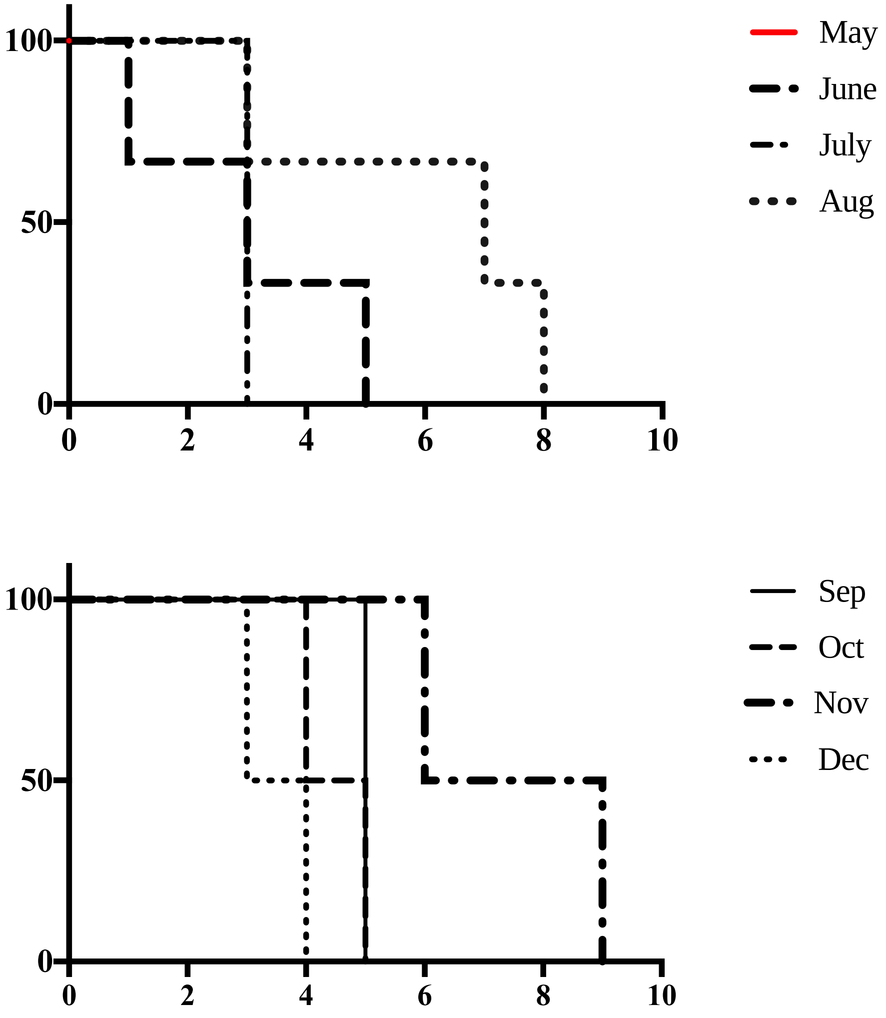

Fig. 7. Kaplan-Meier curves showing the longevity pattern of adult female *Aedes aegypti* in rural areas from May to December.

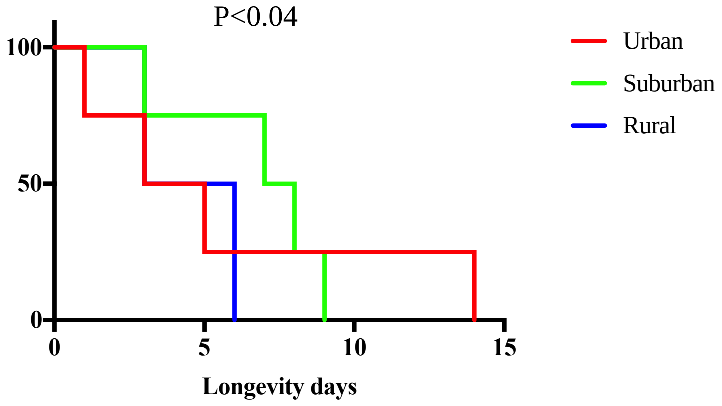

Fig. 8. Overall Longevity days of adult female *Aedes aegypti* in urban, suburban and rural areas.

urbanized areas, threatening us with a high risk of the extent of *Aedes*-borne diseases. The present results may have applied purposes for the design of entomological surveillance in suburban and rural areas. Special attention should be needed to implement new entomological investigation and control approaches and develop disease threat and spread. Future research integrating climate data may provide a more holistic understanding of the linkages between urbanization, local climate changes, and mosquito-borne disease transmission. Hopefully, the management can be combined with other emerging control approaches for an improvement of community-based and sustainable vector management.

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