

MORPHOLOGICAL CHANGES IN EGGS AND LARVAE OF *Aedes aegypti* ADAPTING TO SALINITY

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Aedes aegypti, the principal global vector of arboviral diseases, lays eggs and undergoes larval and pupal development in freshwater (FW) and has recently been observed to develop in coastal brackish water (BW) habitats, evolving various physiological mechanisms that enable these mosquitoes to survive and breed in BW habitats. The present study investigates the morphological changes of eggs and larvae of *Ae. aegypti* reared in FW and BW under laboratory conditions. Five different laboratory colonies were used for the subsequent experiments: two freshwater colonies named JFW (reared in 0 g/L tap water) and NFW (reared in tap water purified by reverse osmosis) and one brackishwater colony (reared in 10 g/L water) named JBW and two reversal colonies where FW larvae transferred and continued to reared in BW designated as JBWR colony, and BW larvae transferred and continued to reared in FW designated as JFWR colony. Cuticle thickness of the 6th to 8th abdominal segments of ten mid-L4 larvae from each experimental setup was measured. The results demonstrated that the JBW colony had significantly thicker ($p < 0.0001$) cuticle (JBW; [70th Generation; G70] = 1530 ± 111 nm) than the two freshwater colonies (JFW [G70] = 996 ± 132 and NFW [G20] = 945 ± 75 nm). Where salinity conditions had been reversed, the cuticle thickness differed significantly from the parent JBW and JFW colonies (JBWR [G3] = 1512 ± 132 , [G11] = 1167 ± 159 , JFWR [G3] = 1196 ± 92 , [G11] = 1402 ± 122 nm). The maximum length and width of four anal papillae of 40 L4 larvae from each of the five colonies were measured. The mean anal papilla length of JBW larvae [G69] was 0.62 ± 0.04 mm, and this was significantly greater ($p < 0.05$) than the mean anal papilla length of two FW colony larvae (JFW [G69] = 0.49 ± 0.06 mm, NFW [G22] = 0.58 ± 0.05 mm). JFWR colony ([G4] = 0.59 ± 0.05 , [G11] = 0.6 ± 0.05 mm) tended to have longer lobes than FW larvae maintained in FW and vice versa in JBWR compared to the JBW colony (JBWR [G4] = 0.54 ± 0.03 , [G11] = 0.55 ± 0.05 mm) and this difference was statistically significant ($p < 0.05$). A total of 100 eggs from ten iso females were analyzed, and the mean egg lengths of all five colonies significantly differed ($p < 0.05$) from each other. It is anticipated that changes to the cuticles of salinity-tolerant *Ae. aegypti* larvae may reduce the effectiveness of insecticides used to control arboviral infections. The morphological changes in anal papillae and eggs may alter ion, water, and gas transport mechanisms to allow *Ae. aegypti* to adapt to increased salinity in natural habitats. The results highlight the necessity for further research on the ultrastructure and physiological mechanism of the cuticle and anal papillae in relation to insecticide resistance and the genomic biology of salt tolerance in *Ae. aegypti*.

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