STIFFENING AND DARKENING OF *AEDES AEGYPTI* EGGSHELLS DURING EMBRYOGENESIS

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Mosquito's eggshells are white, permeable and soft at oviposition, and turn black, impermeable and stiff with time. Until now no report has shown precisely how much and when eggs darken and elasticity changes. This work have used Atomic Force Microscope tip as micro-indenter to measure *Aedes aegypti* eggs elasticity changes during initial embryogenesis. Darkening of *Ae. aegypti* eggs was observed by stereomicroscopy and digital images densitometry was analyzed. Two controls were used: 1) non-fertilized eggs, which have no embryo but darken normally and 2) eggs treated with darkening inhibitor, which swell normally. A software was developed to analyze the approximation force curves. We have analyzed the first 50 nm (indentation) to fit the Hertz model and between 100 and 150 nm to fit the elastic constant. We verified that eggshell darkening takes 2.5 hours after oviposition (HAO) while stiffening is completed after 3 HAO. Both processes don't require a viable embryo to happen since non-fertilized eggs stiffen normally and they are a consequence of sclerotization, since eggs treated with darkening inhibitor doesn't stiffen. The values founded to Young's modulus are about 1 GPa, consistent with the literature predictions for protein aggregates.