

Non-Invasive dsRNA Delivery into Aedes aegypti eggs by Electroporation

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INTRODUCTION: Aedes aegypti is a vector of several diseases such as Dengue, Chikungunya and Zika viruses. The discovery of new drugs and/or novel targets for mosquitoes control strategies are urgent due to the growing development of resistant populations. The combination of RNA interference (RNAi) tools and functional genomics may offer an attractive approach of research to find new targets for vector control and understanding its physiology. Previously, we performed the silencing of AKT and GSK3 by electroporation in eggs of the cattle tick *Rhipicephalus microplus* due to the important roles of both enzymes in tick embryogenesis, performed the same technique in Rhipicephalus microplus eggs, promoting near 50% gene silencing. Moreover, GSK3 silencing by microinjection into adult mosquitoes was also previously reported. **OBJECTIVE:** Investigate the relationship between biological zygotic changes and energy metabolism regulation in Aedes aegypti mosquito embryos. MATERIAL AND METODS: Female mosquitos were blood fed and eggs were obtained 72 hours after bloodmeal in a dark room at 28 °C. Collected eggs were electroporated using 10 pulses at 150V with 1-s intervals (ECM 2001 Electro Cell Manipulation System, BTX). To test dsRNA internalization by electroporation, DAPI (4 ', 6-diamidino-2-phenylindole) entry was tested in mosquito eggs by electroporation under the same conditions. Moreover, hatching rate was determined in eggs electroporated compared with control conditions. **DISCUSSION AND RESULTS**: Here it was demonstrated that DAPI was delivered in *A. aegypti* embryos by electroporation, as observed under fluorescence microscope. CONCLUSION: Conventional RNAi silencing can be applied to study embryogenesis. However, one disadvantage of conventional methods for double-stranded RNA delivery is the potential mosquito egg/embryo structure damage. The advantage of electroporation of embryos is the lower damage when compared to invasive delivery such as microinjection Further studies to test gene silencing via electroporation are on the way.

Keywords: Aedes aegypti, electroporation in eggs, RNAi in embryos.

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