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ABSTRACT

Blood feeding and mating are critical behaviors that regulate both mosquito population maintenance and disease transmission. However, our understanding of mosquito mating systems remains incomplete. One of the most critical issues is a lack of understanding regarding how and where males and females encounter one another. This study was performed to investigate changes in key mating behaviors of *Ae. aegypti* relative to female blood feeding opportunities, taking into account male density. We compared courtship latency and copulation activity between single and pooled males in a range of assays performed in the presence or absence of a blood source and after blood feeding. The time taken by grouped males to initiate courtship in the presence of a host was much shorter than that in single males. There was no significant difference in courtship latency between pooled and single males in the absence of a blood source or after blood feeding. At low male density, the presence of the host and blood meal ingestion provided better conditions for copulation. At high male density, however, copulation activity was decreased after blood feeding, but remained high regardless of the presence or absence of the host. In addition to providing insight into the mating ecology of *Aedes aegypti*, this study indicated that the presence of a blood source influences how males encounter and copulate with females. The observation that copulation activity decreases after blood feeding when males are numerous provides new avenues for improving mass release programs of sterile mosquitoes.

Introduction

Aedes aegypti is a known vector of several viruses that affect public health, including those that cause dengue, chikungunya (ECDC, 2016), and Zika virus disease (Hennessey et al., 2016; CDC, 2016). Together, these diseases pose significant and continuous public health threats (Fares et al., 2015) with great economic impacts worldwide (WHO, 2014). Alternative vector control strategies are needed due to the narrowing spectrum of effective insecticides (Dusfour et al., 2011) caused by the development of resistance (Dorta et al., 1993), along with the lack of effective vaccines and specific therapeutic agents to combat these diseases (Laughlin et al., 2012). Transmission of these viruses generally requires a female mosquito to bite an infected host (Cox et al., 2012) or lay viable infected eggs (Buckner et al., 2013). As blood feeding is the primary mechanism by which disease transmission occurs (Lehane, 2005), and mating, which can trigger vertical transmission (Martins et al., 2012), is closely related to blood feeding (Soghigian et al., 2014), focusing on these behaviors may facilitate the discovery of new strategies for vector control.

Due to the close interplay between blood feeding and mating in *Aedes* mosquitoes, and their roles in disease transmission and vector population maintenance, there have been a number of studies regarding their order of importance. Overall, these observational studies yielded controversial results or relied on unclear assumptions. Flight is an

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