BEHAVIORAL RESPONSE OF ADULT ANOPHELES QUADRIMACULATUS AND Aedes albopictus TO DIFFERENT CARBOHYDRATES IN AN OLFACTOMETER

RUI-DE XUE¹, DANIEL L. KLINE³, GUNTER C. MULLER³, AND DONALD R. BARNARD²

¹Anastasia Mosquito Control District, 120 EOC Drive, St. Augustine, Florida 32092, USA.
²USDA, Center for Medical, Agricultural, and Veterinary Entomology, Gainesville, Florida 32608, USA
³Malaria Research and Training Centre, Faculty of Medicine, Pharmacy and Odonto-Stomatology, University of Sciences, Techniques and Technology of Bamako, BP 1805 Bamako, Mali

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ABSTRACT

Eleven different carbohydrates were evaluated to determine the behavioral response of adult Anopheles quadrimaculatus Say and Aedes albopictus Skuse using an olfactometer. The carbohydrates used in the study are arabinose, fructose, glucose, maltose, melezitose, melibiose, raffinose, rhamnose, sucrose, trehalose, and turanose. The results showed that both species of mosquitoes regardless of the sex had significantly higher attraction to arabinose, maltose, melibiose, and trehalose than other 7 carbohydrates tested. Both sexes and both species responded to maltose and trehalose in considerable numbers, and the least responses were to sucrose except by male Ae. albopictus. These findings may provide insights to the development of more effective sugar-based toxic baits for the operational application in mosquito control programs.

Key Words: Carbohydrates, sugar feeding, Anopheles quadrimaculatus, Aedes albopictus, attractive toxic sugar bait

Anopheles quadrimaculatus Say is one of the major vectors of malaria pathogens, while Aedes albopictus Skuse is an important vector of dengue virus and a domestic / peridomestic pest species. Due to many reasons, such as increase of the development of resistance to insecticides, a novel control technique is urged and demanded for control of these vector mosquitoes. Attractive toxic or target sugar baits (ATSB) and bait stations are one of the new control methods. Different toxins have been used as active ingredient to make ATSB to control several species of adult mosquitoes (Xue and Barnard 2003, Muller and Schlein 2006, Muller et al. 2010). ATSB control technique is based on the sugar feeding behavior of adult mosquitoes. Sugar feeding is important for survival, reproduction, and energetics (Foster 1995). Nutrient acquisitions by adult mosquitoes are from nectar resources (Muller et al. 2011, Barredo and DeGennaro 2020). Flowers, fruits, honeydew, and seed pods of certain plants are favored and their carbohydrates could serve as potential attractants for adult mosquitoes (Muller et al. 2010a, 2011). ATSBs contain an attractive odorant and a lethal active ingredient suspended in a sugar source that mosquitoes utilize as a carbohydrate source. The attractiveness of ATSBs to compete with natural sugars available in the environments is still a big challenge. Therefore, research and development of effective attractants for ATSBs are highly demanded. Selecting the carbohydrate source based on increased mosquito response to different sugars would increase the attractiveness of ATSBs and enhance the effectiveness in operational programs. The present study was conducted to determine whether adult An. quadrimaculatus and Ae. albopictus preferentially respond to different carbohydrates and if so, could those carbohydrates be utilized to develop more effective ATSBs in the future.

Ae. albopictus and An. quadrimaculatus mosquitoes used in this study were received from the laboratory colonies maintained in an insectary at 27° C and 80% relative humidity in a 14:10 photoperiod (light:dark) of the US Department of Agriculture-Agricultural Research Service, Center for Medical, Agricultural, and Veterinary Entomology, Gainesville, FL. Adult mosquitoes were maintained in screened cages and provided a 10% sucrose water solution. Male and female adult mosquitoes were 5-7 days old without starvation when used in each test.

Eleven different carbohydrates, arabinose, fructose, glucose, maltose, melezitose, melibiose, rhamnose, raffinose, sucrose, trehalose, and turanose were purchased from Sigma Aldrich online. Each carbohydrate solution was prepared in methanol (99%) to have the same concentration in all. An aliquot of 100 µL of a selected carbohydrate was pipetted into a plastic vial cup (15 mm inner diameter x 9.5 mm height). Prior to use, the solution was allowed to dry for 3 minutes to remove the methanol solvent. A plastic vial cup was treated only with methanol as control.
A homemade olfactometer with 3 cages and dual-ports per cage described by Posey et al. (1998) was used to determine the response to different carbohydrates in the laboratory. Males (100) and females (100) of each species of mosquitoes were transferred to each cage (a total 400 mosquitoes/cage). The plastic cups containing each carbohydrate were loaded immediately onto an aluminum tray to hold the vials and inserted into the olfactometer ports. Six ports of the olfactometer held 6 different carbohydrates in the first run and the other 5 carbohydrates with the control were run in the next time. The test was repeated in 12 days using the Latin-square design so that each carbohydrate was tested against all the other carbohydrates and the control. Each test combination had 2 replicates. After 1 h exposure, all mosquitoes trapped in each port were separated by sex and the species and counted. The cages and ports were cleaned-up after each test and new mosquitoes from the stock cages were introduced to the olfactometer cages for subsequent test runs.

The mean percent mosquitoes entered into the ports with different carbohydrates were analyzed using multiway ANOVA procedures (SAS 2003). Each count datum for mosquitoes trapped in different ports baited with different carbohydrates were transformed to log (n+1) before the analysis. Differences in the response of each sex of each species to different carbohydrates were compared in separate analyses using Tukey's Honestly Significant Difference (HSD) test. The level of significance in all statistical tests was P = 0.05.

As determined by the olfactometer bioassay, male and female mosquitoes of both species responded to all eleven carbohydrates, compared to the control. The numbers of mosquitoes responded to different carbohydrates varied by the species and the sex of mosquitoes. The most responded carbohydrates by either sex of either species were arabinose, maltose, meliniose, and trehalose (Table 1). The highest numbers of both male and female An. quadrimaculatus responded to maltose (16% and 18.3% respectively). Ae. albopictus males responded the most to trehalose (23.7%) while the females responded mostly to arabinose (35.7%). However, maltose and trehalose had high numbers of both sexes of both species although not statistically significant from the attraction by other carbohydrates in some cases (Table 1). It was surprising that the carbohydrate with the lowest response from both males and females of An. quadrimaculatus (1.7% and 2.7, respectively) and female Ae. albopictus (3.7%) was sucrose. Although not the lowest, the response of male Ae. albopictus (3%) to sucrose was considerably low as well.

The study findings demonstrated that 4 carbohydrates, maltose, trehalose, meliniose, and arabinose were more attractive to both An. quadrimaculatus and Ae. albopictus than the other 7 carbohydrates tested and particularly than sucrose which is the common carbohydrate used in ATSBs. Isolation of the same carbohydrates from the crops of wild-caught adult An. quadrimaculatus (Burkett et al. 1999) and Ae. albopictus (Burkett et al. 1998) indicates their feeding on the same carbohydrates in the natural environment as well. Further, a recent study demonstrated that arabinose enhanced the toxic sugar bait toxicity to Ae. aegypti (Linn.) adult females, but there was no impact on attractiveness of toxic sugar baits when other sugars were present (Airs et al. 2019). Supported with those evidence, the 4 carbohydrates which showed increased attractiveness in our study could be considered as an additional sugar component for the development of ATSBs, and further field evaluation is warranted.

A variety of fruit juices and chemical attractants have been incorporated into ATSBs or TSBs (toxic sugar bait) and evaluated for control of adult mosquitoes (Muller et al. 2010, Xue et al. 2008, Fiorenzano et al. 2017, 2017a). However, the attraction from natural fruits, fruit juices and their extracts did not show a strong attraction. The common chemical attractants (CO2 and Octenol) incorporated with TSBs have increased the attraction of adult female mosquitoes and improved the control efficacy (Fiorenzano et al. 2017a). Toxic sugar baits use sugar as a phagostimulant to induce ingestion of an oral toxin, but sugar alone is not an effective attractant (Fiorenzano et al. 2017). Most ATSB products use brown sucrose which does not have significant attraction. The development and application of more attractive and effective toxic sugar baits and bait stations would provide another useful tool to mosquito management programs and public health officials to continue to combat mosquitoes and mosquito-borne diseases.

REFERENCES CITED


Adult mosquitoes and different carbohydrates


Muller GC, Schlein Y. 2006. Sugar questing mosquitoes in arid areas gather on scarce blossoms that can be used for control. *Intern J Parasitol*. 36:1077-1080.


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Table 1. Mean percent of adult *Anopheles quadrimaculatus* and *Aedes albopictus* attracted to each type of carbohydrates in 1 hour evaluated by olfactometer bioassay

<table>
<thead>
<tr>
<th>Carbohydrates</th>
<th><em>Anopheles quadrimaculatus</em></th>
<th><em>Aedes albopictus</em></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Male</td>
<td>Female</td>
</tr>
<tr>
<td>arabinose</td>
<td>12.7 ab</td>
<td>8.0 bc</td>
</tr>
<tr>
<td>fructose</td>
<td>3.3 b</td>
<td>6.7 bc</td>
</tr>
<tr>
<td>glucose</td>
<td>2.3b</td>
<td>7.3bc</td>
</tr>
<tr>
<td>maltose</td>
<td>16.0a</td>
<td>18.3ab</td>
</tr>
<tr>
<td>melezitose</td>
<td>7.7ab</td>
<td>5.7bc</td>
</tr>
<tr>
<td>meliniose</td>
<td>12.7ab</td>
<td>8.0bc</td>
</tr>
<tr>
<td>raffinose</td>
<td>2.7b</td>
<td>7.7bc</td>
</tr>
<tr>
<td>rhamnose</td>
<td>4.0b</td>
<td>7.0bc</td>
</tr>
<tr>
<td>sucrose</td>
<td>1.7b</td>
<td>2.7c</td>
</tr>
<tr>
<td>trehalose</td>
<td>11.7ab</td>
<td>13.0abc</td>
</tr>
<tr>
<td>turanose</td>
<td>2.7b</td>
<td>6.3c</td>
</tr>
<tr>
<td>Control</td>
<td>0.3ab</td>
<td>0.7ab</td>
</tr>
</tbody>
</table>

Mean percent in each column and row followed by the same letter are not significantly different (Tukey’s HSD, P > 0.05).