Comparison of Culicidae Present in Phytotelmata versus Stream Pools at Springfield Station in Dominica, West Indies

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Abstract: Mosquito borne diseases are proliferating all around the world. Mosquito habitats can be categorized into temporary and standing pools, both of which influence human health. To understand the extent of their presence in Dominica, a survey of mosquitoes was conducted by observing phytotelmata (plant-held water) and pools in streams present at the Archbold Tropical Research and Education Center (ATREC) in Dominica, West Indies from May 23rd, 2017 to June 14th, 2017. To test phytotelmata and pool stream habitats, samples were collected from different water holding plants and standing pools in various areas surrounding the station. The larvae obtained from these samples were then reared and identified. Fifteen mosquitoes were reared from pool streams, and 27 mosquitoes were reared from phytotelmata. The species *Aedes busckii* (Diptera: Culicidae) (Coquillet) were found to be the most common in phytotelmata, and *Culex quinquefasciatus* (Diptera: Culicidae) (Say) were found to be the most common in pool streams. Past research suggested that *Aedes busckii* were the only species found in phytotelmata. However, *Culex inflictus* (Diptera: Culicidae) (Theobald) was also found to inhabit water-holding plants.

Keywords: Dominica, *Culex quinquefasciatus*, *Aedes busckii*, phytotelmata, pool streams

Mosquito borne diseases are becoming more prevalent in the world (Tolle 2009). In Dominica, Zika and Chikungunya have recently made an appearance (Fischer 2014; Gulland 2016). Several research projects have been conducted in Dominica to explore the diversity of mosquitoes that are present. It has been shown that 24 species of mosquitoes are present on the island (Brown 2007). To understand the diversity of species present, a survey was conducted around ATREC. The survey was conducted to observe the mosquito species present in phytotelmata versus pool streams.

The most common genera of mosquitoes that are responsible for vector borne pathogens are *Culex*, *Aedes*, and *Anopheles*, with *Culex* and *Aedes* being the common genera present in Dominica (Brown 2007). In 2007, a comprehensive survey (Brown et al. 2007) was completed in Dominica to determine every species of mosquito present. From the ornamental Bromeliads and *Heliconia caribaea* which were tested, four species of mosquitoes were identified; *Culex bisulcatus* (Diptera: Culicidae) (Micraedes), *Isostomyia perturbans*

(Diptera: Culicidae) (Williston), *Toxorhynchites portoricensis* (Diptera: Culicidae) (Roeder), and *Wyeomyia grayii* (Diptera: Culicidae) (Theobald) (Brown et al. 2007). However, research previously performed on phytotelmata in 2014 observed that *A. busckii* were the only species of mosquito that prefer phytotelmata at ATREC(Bowman 2014).

Previous reports have explained that *C. quinquefasciatus* are common in Dominica (Brown 2007), however the larval habitats were unknown. Due to fluctuation in species diversity in phytotelmata, this survey was conducted again to observe if additional species, such as *C. quinquefasciatus*, might have presented themselves.

Materials and Methods

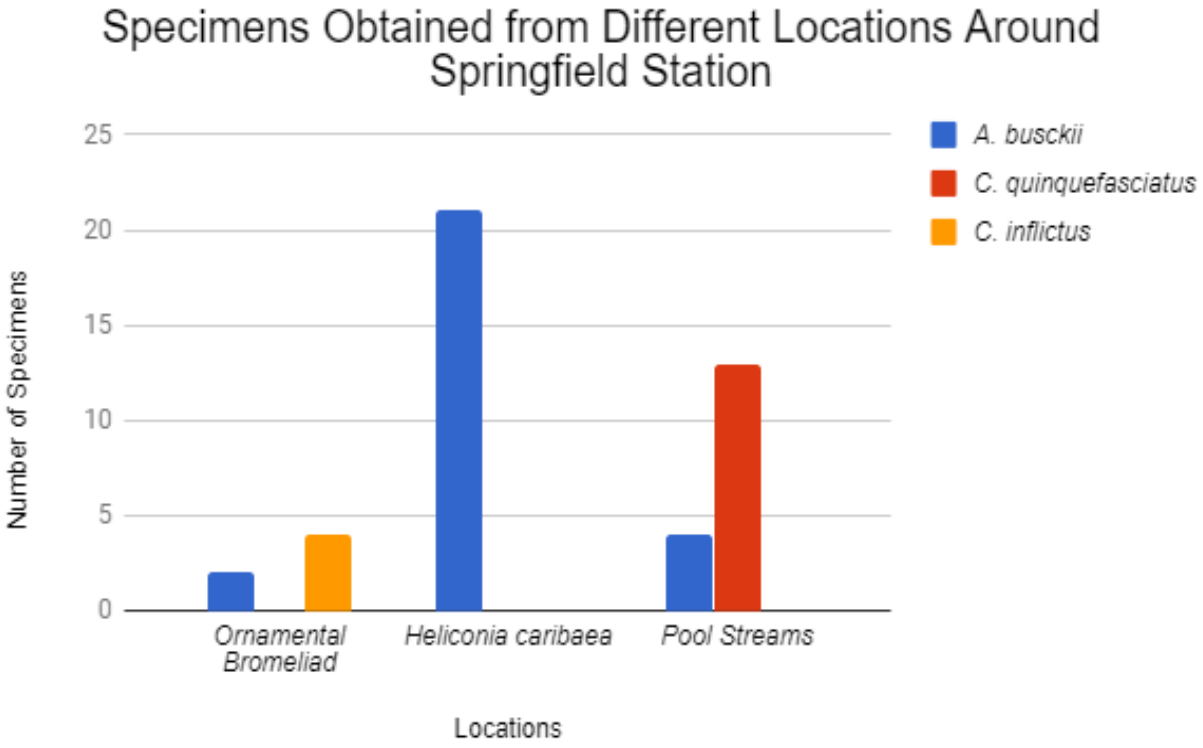
Water was collected from different areas around ATREC for analysis. These areas included rock pools in streams and phytotelmata, which both contained stagnant, discolored water to observe the species present. Samples were collected from *Heliconia caribaea* and ornamental Bromeliads around the station. They were found in the north and west sides of ATREC, Dominica and from areas around the Checkhall River, ATREC, Dominica. These sites were chosen based on the areas surrounding them. Some of the areas were near bodies of water, while others were in more urban environments, such as areas by the station. Samples were collected from varying microhabitats in order to get more diverse results. Four samples were collected from pool streams present at the station. A turkey baster (OXO Good Grips, Chambersburg, PA) was used to collect the water, about 16 oz, and then it was poured into a clear plate (Genpak #9L White Tray, Charlotte, NC) to identify if mosquito larvae were present. If larvae were present, the sample was transferred into a sample collecting bottle (Nalgene 32 oz Widemouth Economy Bottle with 63 mm cap, Lima, OH) and the date and location was recorded. When gathering samples from phytotelmata, the turkey baster was placed at the root of the plant to collect water more likely to obtain larvae. The samples were transferred to rearing containers (Mosquito Breeder, Bioquip, Rancho Dominguez, CA) and the larvae were preliminarily identified to either the genus or species level, depending on the instar using the key in Mosquitoes of Dominica (Brown et al. 2007). The captured larvae were then placed in a rearing container at room temperature. Once reared, the adult mosquitoes were also identified using the key in Mosquitoes of Dominica (Brown 2007). A total of ten rearing containers were used and each container received fish food (Tetramin Tropical Flakes, Tetra, Blacksburg, VA,) (Imam 2014) to feed the larvae as needed. When adults emerged, they were captured with an aspirator (Gempler's, Janesville, WI) and placed in the freezer for five to ten minutes to kill. After they were taken out, they were curated in an ethyl acetate killing jar (Ben Meadows, Janesville, WI). They were then taken out, point mounted, and identified to the species level using the Mosquitoes of Dominica key (Brown 2007.)

Results

Fifteen mosquitoes were collected and reared from stream pools and 27 mosquitoes were

collected and reared from phytotelmata. *Aedes busckii* were the most common species collected from phytotelmata and *Culex quinquefasciatus* were the most common species collected from stream pools. Previous research suggested that only *A. busckii* were found in phytotelmata, however, this research suggested that *Culex inflictus* also inhabit phytotelmata

Table 1: Sample locations, the total specimens collected from each container, and their final identifications. All of the samples obtained were reared.



	Brown (2007)		**Bowman (2014)**		**Current Study (2017)**	
	Bromeliad	*Heliconia caribaea*	Bromeliad	*Heliconia caribaea*	Bromeliad	*Heliconia caribaea*
A. busckii			x	x	x	x
C. inflictus					x	
Is. perturbans		x				
C. bisulcatus	x					
Toxorhynchites portoricensis		x				
Wyeomyia grayii	x					

Table 2: The various species found in phytotelmata from 2007 to 2017.

Fig 1: Female *Aedes busckii.* The distinctive golden dorsocentral lines are present on the mesoscutum.

Fig 2: Female *Culex quinquefasciatus*; dorsal view.

Fig 3: *Culex quinquefasciatus;* lateral view, note light bands on anterior margins of abdominal tergites.

Fig 4: Male *Culex inflictus;* dorsal view.

Fig 5 & 6: Female *Culex inflictus;* dorsolateral and lateral views, note the dark curved setae on mesoscutum and abdominal tergites without light bands.

Discussion

The larval sampling from phytotelmata and pool streams helped to identify the larval habitats of *Culex quinquefasciatus* present at the station. According to the data presented, *C. quinquefasciatus* was found in stagnant, discolored water. Further research should be conducted on the *C. quinquefasciatus* habitat on the rest of the island to observe if there are other areas where they are prominent in. Previous research only found four species inhabiting water-holding plants, which include *Aedes busckii*, *Isostomyia perturbans*, *Culex bisulcatus*, and *Toxorhynchites portoricensis*. In this experiment, *A. busckii* were found to inhabit phytotelmata, however *Culex inflictus* was also reared from this habitat. From research conducted previously, it is understood that *C. inflictus* is a new finding (Brown 2007).

Culex inflictus is a species that needs more research. This species is known to inhabit areas such as crab holes and hollow logs. Since *C. inflictus* was found in phytotelmata, research should be conducted to understand their correlation with water holding plants (Dunn, 1934.) Understanding the correlation between *C. inflictus* and phytotelmata will help in understanding when the species made the transition from crab holes and hollow logs to phytotelmata. Crab holes and hollow logs are usually drier environments so understanding what caused *C. inflictus* to prefer environments with water over the former would help us understand if any behavioral differences occurred over time.

Although this study helped in understanding habitats of species in phytotelmata and stream pools, difficulties arose when trying to properly identify the adults. One limitation presented itself when identifying *C. inflictus,* due to there not being ample amount of research being performed on the species. It was difficult to differentiate between the different *Culex* species and took a large amount if research time to confirm the species. Although limitations presented themselves during this study, there were also several strengths that helped make this experiment a success, including the method of capturing and rearing the specimens. This method was followed the exact same way each time and only provided positive results. It consisted of collecting samples with a turkey baster, preliminarily identifying specimens before they were reared, and feeding the larvae at the same time each day. Uniformity was very important in this study. Another strength of this study was the killing and identification process. Refrigerating and killing the mosquitoes with the ethyl acetate killing jar proved to be extremely effective throughout the study.

This study helps in providing a better understanding of what habitats are present on the island. The results obtained provide a basis on how to expand on future research on larval habitats.

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