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Immature mosquitoes (Diptera: Culicidae) on the bromeliad *Nidularium innocentii* in ombrophilous dense forest of Santa Catarina Island, Florianópolis, Santa Catarina State, southern Brazil

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Resumo

Mosquitos imaturos em *Nidularium innocentii* em floresta ombrófila densa na Ilha de Santa Catarina, Florianópolis, Santa Catarina Brasil. Formas imaturas de mosquitos (Diptera: Culicidae) foram coletadas por um ano em bromélias *Nidularium innocentii* var. *paxianum* em floresta ombrófila densa, na Ilha de Santa Catarina, Estado de Santa Catarina. Duzentos e onze mosquitos foram obtidos, sendo 37,44% *Culex (Microculex) albipes,* 19,91% *Cx. (Mic.) davisi*, 0,95% *Anopheles (Kerteszia) cruzii* e 0,47% *Wyeomyia (Phoniomyia) tripartita.* A correlação entre a quantidade de mosquitos e a temperatura média foi significativamente positiva, mas a correlação com a pluviosidade não o foi. A quantidade de anofelíneos coletados foi bem menor que a relatada em estudos anteriores na região. Mesmo considerando a pequena quantidade de anofelíneos coletados, a grande densidade de bromélias da espécie torna a planta digna de atenção como criadouro.

Unitermos: bromélia, Anopheles, Wyeomyia, Culicidae, Microculex

Abstract

Immature forms of mosquitoes (Diptera: Culicidae) were collected from bromeliads *Nidularium innocentii* var. *paxianum* in dense ombrophilous forest of Santa Catarina Island, state of Santa Catarina. Two hundred and eleven mosquitoes were collected; 37.44% *Culex (Microculex) albipes*, 19.91% *Cx. (Mic.) davisi*, 0.95% *Anopheles (Kerteszia) cruzii* and 0.47% *Wyeomyia (Phoniomyia) tripartita*. Correlation between the quantities of collected mosquitoes and mean temperatures was significantly positive, but that between the quantities and the rainfall was not. The quantity of anophelines was much smaller than that reported by other workers in the same region. Even considering the small quantity of anophelines collected, the great density of this bromeliad in the area indicates the need for of attention to it as a breeding ground for these mosquitoes.

Key words: bromeliad, Culicidae, Anopheles, Wyeomyia, Microculex

Introduction

Bromeliads are endemic to the Neotropical Region, and many species occur in southern Brazil (Reitz, 1983; Mestre et al., 2001). Plants of this family grow in the soil, on stones and on tree trunks, and are very plentiful in open and preserved forests in tropical and subtropical regions of the American continent (Padilla, 1973).

Bromeliads can retain water and decomposing vegetal debris in the imbrications of their leaves as a natural resource for their subsistence, absorbing food by special structures at the base of the leaves. The tanks formed by their leaves can be utilized by several animals, including mosquitoes.

Due to the great diversity of bromeliads and mosquitoes, demonstrated by the finding of more than 200 species of these insects on the plants (Frank and Curtis, 1981), their relationship is not well known. A close association between certain species of mosquitoes and bromeliads is supposed, due to the liberation of substances by bromeliads, which accumulate in the tank and probably influence the composition of their water (Rachou et al., 1952; Laessle, 1961; Torales et al., 1972).

In Santa Catarina State, studies on bromeliadassociated mosquitoes have been mostly on *Anopheles (Kerteszia)* spp., due to their importance in the transmission of malaria (e.g., Rachou and Ferraz, 1951; Veloso et al., 1956; Aragão, 1968).

The objective of the present study was the evaluation of the mosquito fauna in tanks of *Nidularium innocentii* Lem. var. *paxianum* (Mez) L. B. Sm. in a secondary ombrophilous forest on Santa Catarina Island, southern Brazil.

Material and Methods

The study was developed at the Desterro Environmental Conservation Unit (UCAD – Unidade de Conservação Ambiental Desterro), situated in the north of the Santa Catarina Island (headquarters -27°31'50.8"S; 48°30'44.3"W, 100m a.s.l., area – 495ha). It is composed of ombrophilous dense forest in several stages of regeneration, and it is very rich in bromeliads of several species. *Nidularium innocentii* var. *paxianum* was studied due to its importance to the maintenance of vectors of malaria in southern Brazil (Reitz, 1983) and also because it is a rupicolous bromeliad which is very frequently found in the soil level at UCAD.

Since Lozovei and Silva (1999) reported the absence of significant differences between the results by dismounting and washing the plants' leaves and by sucking their water, the latter method was utilized in the present study. The water was sucked with a siphon bottle, blowing into the tank to mix the material, adding more pure water to the plant, and repeating the suction. All the material was poured out into a translucent white plastic tray, in portions small enough to see the immature forms, illuminating from below by a torch. These forms were transferred to plastic tubes that were subsequently labelled for identification, and the water was poured back onto the plant. Ten plants were examined per month, from November 2002 to October 2003 (total - 120 plants). To prevent the re-examination of the same bromeliads, they were marked with white adhesive tape.

In the laboratory, immature forms were separated into 10ml vials, mixing the original water from the plant with distilled water and small amounts of fish food (Tetramin®). Emerged adults were killed by ethyl acetate and fixed with entomological pins. Exuviae and dead larvae and pupae were preserved in ethanol 80°GL.

Adult mosquitoes were identified based on Kumm (1933), Rozeboom and Komp (1950), Lane and Whitman (1951), Correa and Ramalho (1956), Cotrim and Galati (1977) and Forattini (2002). Genera and subgenera were abbreviated as proposed by Reinert (1975, 1982 and 1991).

All the material was deposited in the collection of the Microbiology and Parasitology Department of the Universidade Federal de Santa Catarina, Florianópolis, and voucher specimens were donated to the collection of the Faculdade de Saúde Pública, Universidade de São Paulo, São Paulo, Brazil (Dr. Iná Kakitani).

Mean temperatures and precipitation during the study period were recorded at Station number 124 of EPAGRI, localised at Itacorubi, Florianópolis – SC (27°34'00"S; 48°30'00"W). The association between the

quantities of collected mosquitoes and the meteorological data were analysed by Statistica 6.0 software.

Results

Two hundred and eleven immature forms of mosquitoes were collected from *Nidularium innocentii* var. *paxianum*. Of these, 101 (47.87%) were collected in the summer, 86 (40.76%) in the autumn, one (0.47%) in the winter, and 23 (10.90%) in the spring (Table 1). The influence of season on the collections was significant at 1% by Qui-square.

Most collected mosquitoes belonged to *Culex* (*Microculex*) albipes (37.44%) and *Culex* (*Microculex*) davisi (19.91%), and only 0.95% of the collected insects were Anopheles (Kerteszia) cruzii and 0.47% Wyeomyia (Phoniomyia) tripartita. Thus, all collected mosquitoes belonged to three groups (subgenera): *Microculex* (98.58%), Kerteszia (0.95%) and Phoniomyia (0.47%).

Multiple regression analysis showed a significant positive correlation ($R^2 = 0.677$; p<0.05) between the quantity of collected mosquitoes and the mean temperatures in each of the four seasons. However, the same analysis did not show any correlation between these quantities and the cumulative rainfall in each of the seasons. No significant correlation for separate species was observed.

Discussion

Bromeliads are breeding places for a diversified invertebrate fauna because they are complex biological structures, varying according to the plant size, the shape of the leaves, and the environment where they live (Frank, 2007).

In the present study, the mosquito fauna found in *Nidularium innocentii* var. *paxianum* (Table 1) was composed of species usually found in bromeliads. Species of *Anopheles (Kerteszia), Wyeomyia (Phoniomyia)* and *Culex (Microculex)* are usually found in bromeliads (Machado-Allison et al., 1986; Forattini, 2002).

Although only two specimens of *An. (Ker.) cruzii* were found in *N. innocentii*, its importance must not be underestimated, because this species has been incriminated as a vector of malaria. The low prevalence of this mosquito is probably related to the low quantity of water in these bromeliads. The quantity of immature forms is proportional to the volume of water (Forattini, 2002), and they can be bioindicators of preserved environments (Dorvillé, 1996). The ratio of infestation of this species of bromeliad by *Anopheles* is much lower than that reported by Reitz (1983), which was 25-30%. This difference is difficult to explain, and probably cannot be attributed to the difference between the methodology; Lozovei and Silva (1999) reported no significant differences between the results obtained by the

Species	Summer	Autumn	Winter	Spring	Total - %
An. (Ker.) cruzii	1	0	0	1	2(0.95%)
Cx. (Mcx.) albipes	51	16	1	11	79(37.44%)
Cx. (Mcx.) davisi	17	24	0	1	42(19.91%)
Cx. (Mcx.) hedys	7	7	0	3	17(8.06%)
Cx. (Mcx.) imitator	7	7	0	0	14(6.64%)
Cx. (Mcx.) neglectus	0	0	0	2	2(0.95%)
Cx. (Mcx.) pleuristriatus	0	0	0	1	1(0.47%)
Cx. (Mcx.) Gr. consolator	5	8	0	2	15(7.11%)
Cx. (Mcx.) Gr. imitator	12	24	0	1	37(17.54%)
Cx. (Mcx.) Gr. pleuristriatus	1	0	0	0	1(0.47%)
Wy. (Pho.) tripartita	0	0	0	1	1(0.47%)
Total (%)	101(47.87%)	86(40.76%)	1(0.47%)	23(10.90%)	211(100%)

TABLE 1: Immature Culicidae collected in the tanks of *Nidularium innocentii* in UCAD, from November 2002 toOctober 2003, examining ten plants per month.

 $X^2=71.93; X^2_{tab 1\% 33 g.l.}=50.89.$

dismounting and careful examination of the bromeliads and the suction of their water, as utilized in the present study.

The high density of *N. innocentii* in several parts of UCAD (up to 170 plants/ $100m^2$, according to L. C. Pinho – personal communication) can indicate some importance of this bromeliad for the production of *Anopheles* mosquitoes. Reitz (1983) reported, in a study of 20,513 plants, a mean density of 67 specimens/ $100m^2$, frequently in groups of 2-5 plants.

Mosquitoes of *Culex (Microculex)* constituted almost the totality of the immature forms collected from *N. innocentii*. These mosquitoes are always associated with bromeliad tanks (Consoli and Oliveira 1994), and have been associated with the feeding in cold-blooded animals (Lourenço-de-Oliveira and Heyden, 1986). They are very rare in collections from humans (Forattini et al., 1989; Paterno and Marcondes, 2003). However, Forattini et al. (1987) reported a small proportion of biting by these mosquitoes in birds and equines.

Wyeomyia (Phoniomyia) was also very rare in the collections from N. innocentii, but these mosquitoes are very common human-biters in the studied area (Paterno and Marcondes, 2003). Possibly, they are more frequent in epiphyte bromeliads, and Culex (Microculex) would be the predominant group in ground plants (Correa and Ramalho, 1956). However, mosquitoes of these groups have not been studied in their breeding places. The productivity of Wyeomyia (not Phoniomyia) from bromeliads was evaluated as 107 adults/plant per year in Florida (Frank et al., 1988), but the ecology of these Sabethini mosquitoes is poorly known.

Other bromeliads were recently studied in UCAD (*Aechmea lindenii* and *Vriesea philippocoburgi*) and in a restinga locality ("RPPN Morro das Aranhas") in the north-east of the island (*A. lindenii* and *V. friburgensis*) (Muller and Marcondes, 2006). *Culex* (*Microculex*) also constituted a great proportion (57.5-98.9%) of the mosquitoes in the localities. *A. lindenii* had no mosquitoes in UCAD, probably because it was open and had no accumulated water, but had a diversified fauna in Morro das Aranhas. A statistical comparison of both studies could not be adequately developed, due to

the non-coincidence of the periods of study. These variations between faunas associated with different bromeliad species and localities emphasise the need for careful studies on the ecology of mosquitoes in their tanks.

The multiple regression analysis indicated a high positive correlation between the quantity of immature forms of mosquitoes per plant and the mean temperature. This is probably caused by the enhancement of the blood-feeding, copulation and egg-laying activities in higher temperatures (Forattini, 2002). However, this analysis did not indicate a significant correlation between rainfall and the quantity of mosquitoes. Although the availability of water in bromeliads is probably important for the mosquitoes, since the amount of water N. innocentii can retain is small (up to 500ml) (Reitz, 1983), the excess of water was probably wasted by the plant in the rainiest months, not reflecting in greater populations of the mosquitoes. The significances of the associations between rainfall and temperature and the quantities of mosquitoes were also similar in the study of Muller and Marcondes (2006).

Bromeliads are very diversified and densely distributed in UCAD, and *Nidularium innocentii* is the most frequent at ground level, forming a "green carpet" in several areas. Therefore, their importance as breedingplaces for mosquitoes cannot be neglected.

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