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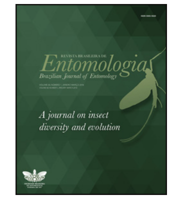

Review

Host–parasite interaction and impact of mite infection on mosquito population

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ABSTRACT

During the present study, the host–parasite relationship between mosquitoes and parasitic mites was investigated. The 8954 individuals of male and female mosquitoes belonging to 26 genera: seven each of *Aedes* and *Culex*, six of *Anopheles* and one each of *Toxorhynchites*, *Coquillettidia* and *Uranotaenia* were collected from 200 sites. The male and female mosquitoes were collected from the State of Uttar Pradesh, located at 26.8500° N, 80.9100° E in North India by deploying Carbon dioxide-baited and gravid traps. The intensity of mite's infection, type and number of mites attached to mosquitoes, mite's preference for body parts and host sexes were the parameters used to determine host–parasite relationship. Eight species of mites: *Arrenurus acuminatus*, *Ar. gibberifrons*, *Ar. danbyensis*, *Ar. madaraszi*, *Ar. kenki*, *Parathyas barbigera*, *Leptus* sp., and *Anystis* sp., parasitized mosquitoes. Parasitic mites preferred host's thorax for attachment as compared to the head, pre-abdomen or appendages. The present study suggests phoretic relationship between parasitic mites and mosquitoes. Wide occurrence, intensity of infection, parasitic load, and attachment preferences of the mites suggested their positive role in biological control of adult mosquitoes. The present study will set the path of future studies on host–parasite relationships of mites and mosquitoes and define the role of parasitic mites in the biological control of mosquitoes.

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Introduction

Aedes, *Anopheles* and *Culex* species of mosquitoes transmit diseases to humans and animals. They are most prevalent in developing and under developed countries, and spread diseases like malaria, dengue, chikungunya, yellow fever, filaria (Esteva et al., 2007). Despite decreasing incidence of human mortality, mosquito borne diseases are still the cause of serious health issues to over 214 million people (WHO, 2015) in developing and under developed countries.

Parasitic mites are ubiquitous and prevalent in the fresh-water habitats, their population density reaches up to 500 individuals with more than 50 species within 1 m^2 (Di Sabatinol et al., 2010). They parasitize insects, including mosquitoes and predate upon them. Larval mites of Arrenuridae, Thyasidae, Anystidae, Hydryphantidae (Mullen, 1975; Smith, 1983) are obligate parasites, which ingest hemolymph by piercing exoskeleton of the host (Smith et al., 2009; Gerson et al., 2003). Attached to mosquito pupae as parasite, the larval stages of mites transform to adults upon ecdysis (Smith and McLever, 1984). In contrast, *Parathyas* larvae attach to their hosts, when host returns to oviposit at the surface of the water (Mullen, 1997). Studies made by Lanciani and Boyt (1977), Lanciani and McLaughlin (1989), Rajendran and Prasad (1992), Nelson (1998), Sarkar et al. (1990), Mathieu et al. (2006), Esteva et al. (2007), Kirkhoff et al. (2013), and Worthen and Turner (2015) have generated significant interest in parasitic mites and their possible role in biological control of insects.

The biphasic (parasitic and predation) life cycle of parasitic mites consists of egg, pre-larva, larva, three nymphal stages and adult stage (Smith, 1988; Esteva et al., 2006). Parasitic mites hatch in the water, and attach to the host during emergence as a phoretic partner (Worthen and Turner, 2015). After completing parasitic phase, larval mites transform into deutonymph and adults, becoming predatory in nature and feeding upon insects and mosquitoes alike. Mites grasp and puncture prey-using chelicerae, secrete stylostome to feed on digested tissues (Smith, 1988) much like plant-parasitic nematodes, which make feeding-plugs to suck host contents (Bilgrami and Gaugler, 2004). Mites can also attach to previously uninfected adults through transfers during mating (Hussell et al., 2010). The larval development completes upon dropping of mites by insects, which return to water bodies, leaving scars as indicators of parasitism (Rolff et al., 2000). Mites grow in size

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(80–90 times) during feeding and up to 47 mite's infected one mosquito individual at a time (Mitchell, 1967; Kirkhoff et al., 2013), significantly enough to affect and reduce host diversity and mosquito population in the area.

The contacts between the mosquito and mite are co-incidental, except in some cases where chemical or other cues play a role (Mullen, 1997). The larval stages of terrestrial mites (e.g. Erthraeidae and Trombellidae) affect mosquito populations (Welbourn and Young, 1988; Southcolt, 1992), whereas others *e.g. Charletonia* and *Leptus* parasitize adult mosquitoes during inactive and resting phases (Wohltmann and Wendt, 1966).

The use of chemical pesticides impacts mosquito populations but alongside, it leaves toxic and adverse effects on human and animal populations. Parasitism (Mullen, 1975; Williams and Proctor, 1991; Gerson et al., 2003) and predation (Bilgrami and Tahseen, 1992; Bilgrami, 1994; Bilgrami, 1997a,b) are ecological interactions that may act alone or concomitantly during biological control process of the pests and vectors. Such is the relationship between mosquitoes and aquatic mites (Acari: Hydrachnidia) (Esteva et al., 2006).

A few options such as *Bacillus thuringiensis*, *B. sphericus*, and *Gambusia affinis* are available to biologically control mosquito larvae but none is available to use against adult mosquitoes. The parasitic mites possessing biological control potentials, few studies made on their biology and behavior, and the need of an effective biological control agent to control adult mosquitoes have led us to carry out this study.

The present study was made on the collected individuals of 23 species of mosquitoes in order to determine prevalence, parasitic load, host preference, attachment site preference, host–parasite relationships, and biological control potential of mites against adult mosquitoes.

Materials and methods

Collection of mosquitoes

The Carbon dioxide-baited and Gravid Traps were used to collect male and female mosquitoes from more than 200 sites in the State of Uttar Pradesh, located at 26.8500° N, 80.9100° E in North India. Each trap was set from dusk to dawn, once a week between May 1st and October 30th 2014. The following morning, mosquitoes were collected and mite infested mosquitoes were sorted out based on mosquito species and parasitic mites. Mosquito individuals infected by the mites were stored at −80 °C for further analysis. No animal specimens were exported out of the country for any purpose. During present study, *Toxorhynchites splendens*, *Uranotaenia compestris* and *Coquillettidia* sp., are referred to as "others", since they were not available in sufficient numbers. They are included in this study for comparison purposes.

Collection of parasitic mites

The parasitic mites carefully separated from mosquitoes, and preserved in the Alcohol-Glycerin-Acetic Acid solution (AGA) (Gibb and Oseto, 2006) for identification. Five to seven mites were mounted in AGA solution on a glass slide, under 12 mm circular cover slip, in such a way that the legs of the mite stayed separated (Smith et al., 2009). Mites were identified by using taxonomic keys provided by Prasad and Cook (1972), Mullen (1974, 1975) and Pesic et al. (2010).

Analysis of host–parasite relationship

The infested mosquito individuals were examined for the intensity of mite infection, type and species of mites, number of mites attached, and preference for host species, sex and body parts. Mosquito–mite relationship was determined in terms of infection intensity (defined as the number of aquatic mites on a host individual) and the mean infection intensity (defined as the total number of parasitic mites divided by total number of parasitized hosts) (Margolis et al., 1982). Preference of mites for male or female mosquitoes was determined on the basis of the number of individuals parasitized. The attachment sites were grouped into five categories: head, thorax, pre-abdomen (between metathoracic and first abdominal segment), abdomen and appendages (legs and wings) (Kirkhoff et al., 2013).

Statistical analysis

Statistical analysis of the data was performed by using Ky-Plot version 2 (Yoshioga, 2002). Student's '*t*-test' and Tukey's multiple range test were applied to determine significant differences at $p \le 0.05$.

Results

A total number of 8954 individuals belonging to six mosquito genera and 23 species i.e., seven species each of *Aedes* and *Culex*, six of *Anopheles* and one each of *Toxorhynchites*, *Coquillettidia* and *Uranotaenia* were collected (Tables 1–4). From the collection, 43.73% mosquito individuals were parasitized by eight species of parasitic mites i.e., *Arrenurus acuminatus*, *Ar. gibberifrons*, *Ar. kenki, Ar. danbyensis*, *Ar. madaraszi*, *Parathyas barbigera*, *Leptus* sp., and *Anystis* sp. Fig. 1 shows *Aedes* sp., infected with *Ar. danbyensis, Cx. pipiens* infected with *Ar. danbyensis*, and *Coquillettidia* sp. with *Leptus* sp.

Aedes *parasitized by parasitic mites*

Parathyas barbigera parasitized all species of *Aedes. Arrenurus acuminatus* and *Ar. kenki* parasitized *Ae. pallidostriatus* and *Ae. pipersalatus* whereas, *Ar. gibberifrons* infected *Ae. novalbopictus* (Table 1). *Arrrenurus danbyensis*, *Ar. madaraszi*, *Leptus* sp., and *Anystis* sp., did not parasitize any individual of *Aedes*.

Parathyas barbigera parasitized maximum number of *Ae. aegypti* (63.13%) with mean infection intensity of 5.59 ($p \le 0.05$) and parasitic load of 1–21 (Table 1). Mites parasitized fewer individuals of *Ae. albopictus* (11.49%) but the mean infection intensity (4.29) and parasitic load (1–9) was significantly higher than other *Aedes* species. The other *Aedes* species were parasitized between 2.35 and 8.21% of the collected population, with mean infection rate and parasitic load ranging between 1.48–5.7 and 1–10 respectively (Table 1).

Anopheles *parasitized by parasitic mites*

Arrenurus acuminatus and *Pr. barbigera* parasitized all species of *Anopheles* mosquitoes except *An. thompsoni* (Table 2). The 67.30% of *An. stephensi* were parasitized with mean infection intensity of 7.30 and parasitic load of 1–12 (Table 2). In terms of parasitized individuals, *An. thomsoni* was the second most preferred mosquito species (20.00%) ($p \le 0.05$), which carried less parasitic load (1–6) and mean infection intensity (3.0) as compared to other species of *Anopheles*. *Arrenurus kenki* was parasitic on *An. thomsoni* and *An. quinquefasciatus*. *Anopheles barbarostris* was least preferred with only 1.25% of it's population parasitized at the mean infection intensity of 3.35 and parasitic load of 1–4 (Table 2).

Culex *parasitized by parasitic mites*

Mites preferred *Culex* species more than others. Seven species of mites parasitized 64.24% of collected individuals of mosquitoes. *Arrenurus kenki* and *Pr. barbigera*, each was parasitic on four species

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