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Water mite parasitism is associated with body condition and sex of the whirligig beetle *Dineutus nigrior* (Coleoptera: Gyrinidae)[1]

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Abstract: Larval water mites parasitize a variety of aquatic insects and may have a negative impact on host fitness. Here we tested whether mite parasitism is associated with sex and body condition in the whirligig beetle *Dineutus nigrior* (Coleoptera: Gyrinidae), a host where mite parasitism has not previously been reported. There was a pattern of higher mite intensity in males indicating that males were more heavily exposed or more susceptible to mite parasitism. However, there was not a consistent sex bias in mite prevalence. Host body condition was negatively correlated with mite abundance in both sexes. This could suggest that mites reduce host body condition and thus negatively impact host fitness. Alternatively, individuals in poor body condition may be more susceptible to mite parasitism, suggesting that there may be a condition-dependent mite resistance mechanism in this host.

Keywords: body condition, Gyrinidae, sex-biased parasitism, water mites, whirligig beetle.

Résumé : Les larves d'acariens aquatiques parasitent une variété d'insectes aquatiques et peuvent avoir un impact négatif sur la valeur adaptative de l'hôte. Nous avons ici évalué si le parasitisme par les acariens est relié au sexe et à la condition physique chez le gyrin *Dineutus nigrior* (Coleoptera : Gyrinidae), un hôte chez qui le parasitisme par les acariens n'a pas été précédemment rapporté. Il y avait une plus forte intensité d'acariens chez les mâles indiquant que ceux-ci ont été plus fortement exposés ou sont plus susceptibles au parasitisme par les acariens. Cependant, il n'y avait pas de biais sexuel systématique dans la fréquence d'acariens. La condition physique de l'hôte était négativement corrélée avec l'abondance d'acariens chez les 2 sexes. Cela peut suggérer que les acariens réduisent la condition physique de l'hôte et ont ainsi un impact négatif sur la valeur adaptative de celui-ci. Alternativement, les individus en mauvaise condition physique peuvent être plus susceptibles au parasitisme par les acariens suggérant qu'il puisse y avoir un mécanisme de résistance aux acariens dépendant de la condition chez cet hôte.

Mots-clés : acariens aquatiques, condition physique, gyrin, Gyrinidae, parasitisme biaisé en fonction du sexe.

Nomenclature: Cook, 1974; Davies & Tobin, 1984; Bousquet, 1991; Markow & O'Grady, 2005.

Introduction

Parasitism may be associated with several host characteristics, including body condition and sex. Body condition refers to the amount of energy reserves an individual has available for allocation to various functions, an important factor in determining fitness (Rowe & Houle, 1996). A negative correlation between degree of parasitism and host body condition is present in several parasite–host systems (Polak, 1998; Rolff, Antvogel & Schrimpf, 2000). This could suggest that parasites reduce host body condition. Parasites remove host resources for their own use (Price, 1980) and can cause increased energy expenditure by the host (Booth, Clayton & Block, 1993; Ots *et al.,* 2001) and thus may reduce body condition. Experimental evidence supports the idea that parasites can reduce host body condition (Polak, 1998; Newey, Thirgood & Hudson, 2004). Alternatively, a negative correlation between degree of parasitism and host body condition could be due to individuals in poor nutritional state being less able to invest in costly parasite resistance mechanisms (Leung, Forbes & Baker, 2001; Koskimäki *et al.,* 2004). A negative correlation between host body condition and degree of parasitism is not universal, as a positive correlation (Blanco, Tella & Potti, 1997) and no correlation (Perez-Orella & Schulte-Hostedde, 2005) can also exist.

Sex-biased parasitism exists in a wide variety of host taxa (Poulin, 1996; Sheridan *et al.,* 2000; Moore & Wilson, 2002) and is expected when one sex is more susceptible (Wedekind & Jacobsen, 1998) or more heavily exposed (Lajeunesse, Forbes & Smith, 2004) to parasites. Male-biased and female-biased parasitism is present in invertebrates, but no clear pattern exists (for arthropods see Sheridan *et al.,* 2000), and causes of sex-biased parasitism in invertebrates are generally not well understood (Zuk & McKean, 1996).

Water mites (Acari: Hydrachnidia) are found in virtually all freshwater habitats. The larvae of many species are ectoparasitic, typically on aquatic insect hosts, while adults are typically free-living and predatory. Larval mites attach to

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the host with their mouthparts and then begin to engorge by feeding on haemolymph. The removal of nutrient-rich haemolymph could result in a decrease in host body condition if hosts are not able to compensate for the nutrient drain. A negative correlation between water mite parasitism and host body condition is present in a water mite–damselfly system (Rolff, Antvogel & Schrimpf, 2000), suggesting water mite parasitism may reduce host body condition. Furthermore, water mite parasitism can cause a substantial reduction in investment in egg production (Martin, 1975; Davids & Schoots, 1975; Lanciani, 1982) and a decrease in survival (reviewed by Smith, 1988), suggesting that mite parasitism may reduce the pool of nutrients available for allocation to reproduction and survival. Water mites parasitize a wide range of aquatic insect taxa (Smith, 1988), including the gregarious, surface-inhabiting whirligig beetles (Coleoptera: Gyrinidae) (Zawal, 2003). Reports of water mites parasitizing gyrinids are scarce, and very little is known about parasitic water mite–gyrinid interactions. Here we determine if mite parasitism is associated with sex and body condition of the whirligig beetle *Dineutus nigrior*, a host where mite parasitism has not previously been reported.

Methods

In *D. nigrior*, adults eclose in the summer and fall (Istock, 1966) and typically do not live for more than one winter (Balfour-Browne, 1950). Thus, from early spring until the new cohort begins to eclose, one cohort of adults is present in the population; 2 cohorts are then present for the remainder of the active season. We made frequent collecting trips to our study site throughout the spring and summer of 2006 and observed the first teneral *D. nigrior* specimen on July 7. To avoid potential problems arising from including 2 cohorts of beetles we used specimens collected prior to July 7 (May 8, May 22, June 16, and June 26) in this study. A sample consisted of all of the beetles collected on a particular date or an arbitrarily selected sub-sample of specimens collected.

The specimens were collected from Swan Lake, a small lake (*ca* 6 ha surface area, 8.5 m maximum depth) near Sudbury, Ontario, Canada. The lake was historically acidified by local sulphur deposition and is presently fishless. We used nets to opportunistically sample beetles that were observed swimming on the water surface. Specimens were transported alive to the laboratory and then killed with ethyl acetate fumes (May 22 sample) or 70% ethanol (all other samples). With the aid of a dissecting microscope we searched the external body surface, metathoracic wings, body tergites under the elytra, and the undersides of the elytra for mites. The sex of the host and the number of attached mites was recorded.

We removed all mites from beetles collected on May 22 by gently teasing them apart from the host using an insect pin. We then determined the mass of each beetle (nearest 0.001 g) using an OHAUS TS400D digital scale (OHAUS, Pine Brook, New Jersey, USA). We also measured the length of the elytra of each host to the nearest 0.1 mm using a micrometer-equipped Olympus SZ30 dissecting microscope (Olympus, Tokyo, Japan).

We use intensity to refer to the number of parasites on an infested individual, abundance to refer to number of mites on an individual (including uninfested individuals), and prevalence to refer to the proportion of hosts with at least one parasite (Bush *et al.,* 1997). We tested for sex difference in mean intensity using bootstrap *t*-tests (Rózsa, Reiczigel & Majoros, 2000) and in prevalence using Fisher's exact tests for each individual sample and when the 4 samples were pooled. Confidence intervals were calculated for mean intensity using bootstrap tests (Rózsa, Reiczigel & Majoros, 2000) and for prevalence using Sterne's exact method (Reiczigel, 2003).

We used the May 22 sample to test for a correlation between body condition and $\log_{10}(x + 1)$ mite abundance. We used residuals from a regression of $\log_{10}$-transformed mass on $\log_{10}$-transformed body size (elytral length) as an index of body condition. Size-corrected mass is a common measure of body condition (Schulte-Hostedde *et al.,* 2005).

Fisher's exact tests, bootstrap *t*-tests, and confidence intervals of prevalence and mean intensities were performed using the software Quantitative Parasitology 3.0 (Reiczigel & Rózsa, 2001: Budapest). All other analyses were performed using Statistica 6.0 (StatSoft Inc., Tulsa, Oklahoma, USA).

Results

The mites were identified as *Eylais* spp. by Dr. Heather Proctor (University of Alberta), but they could not be identified to species. *Eylais* mites are large mites that commonly parasitize aquatic Hemiptera and Coleoptera.

Mite intensity ranged from 1 to 8 in females and 1 to 11 in males (Figure 1), and most of the hosts with the highest mite intensities were males despite the fact that more females were collected and found to be infested in the 4 samples (Figure 1). Male mean intensity was significantly higher for the May 8 sample ($t = 3.06$, $P = 0.007$) and when the 4 samples were pooled ($t = 4.45$, $P < 0.001$) (Figure 2). While the bootstrap *t*-test did not indicate that the higher male mean intensity in the June 16 ($t = 2.06$, $P = 0.120$) and June 26 ($t = 1.68$, $P = 0.143$) samples were significant, the 95% confidence intervals did not overlap (Figure 2). There was no significant sex difference in the May 22 sample ($t = 1.11$, $P = 0.291$).

There was no significant sex difference in mite prevalence when the samples were pooled ($P = 0.282$). Prevalence was significantly higher in females on May 22 ($P = 0.035$) and in males on June 16 ($P = 0.044$), and there was no significant difference in prevalence for the other 2 samples ($P > 0.334$) (Figure 3).

Host body condition (residual mass) was negatively correlated with mite abundance for both female ($R^2 = 0.170$; $F_{1,32} = 6.55$, $P = 0.015$) and male ($R^2 = 0.208$; $F_{1,34} = 8.91$, $P = 0.005$) *D. nigrior* (Figure 4).

Discussion

There was an overall trend for higher mite intensity in male *D. nigrior*. There are examples of both male-biased (Lajeunesse, Forbes & Smith, 2004) and female-biased (Robb & Forbes, 2006) parasitism of insect hosts by water mites. Very little is known about gyrinid–water mite interactions or

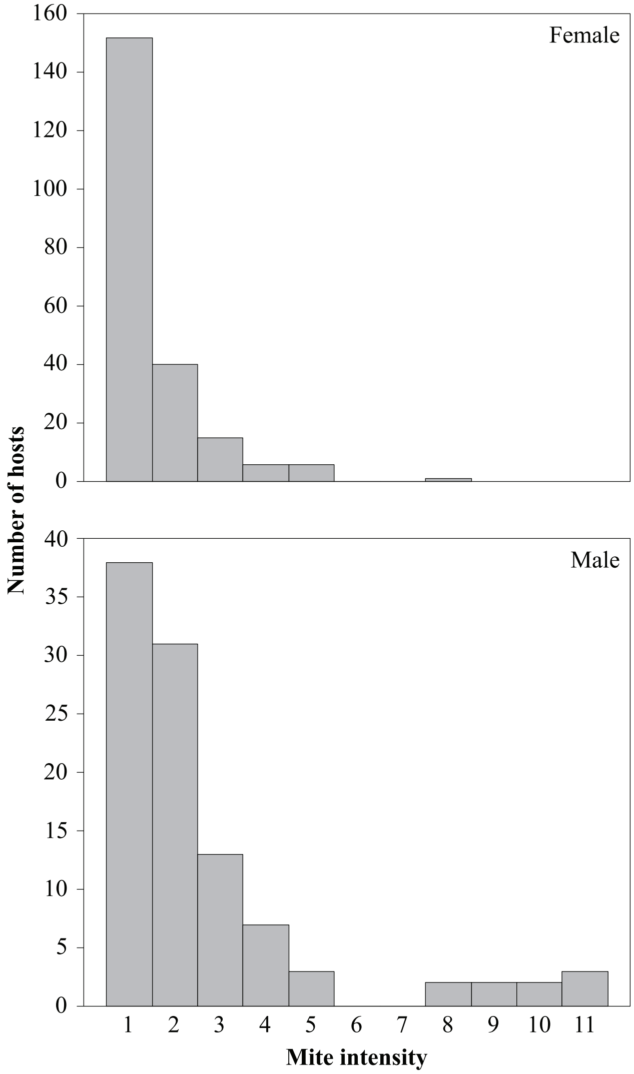

FIGURE 1. Mite intensity frequency for female and male *D. nigrior* hosts when all 4 samples were pooled. Female n = 220, male n = 101.

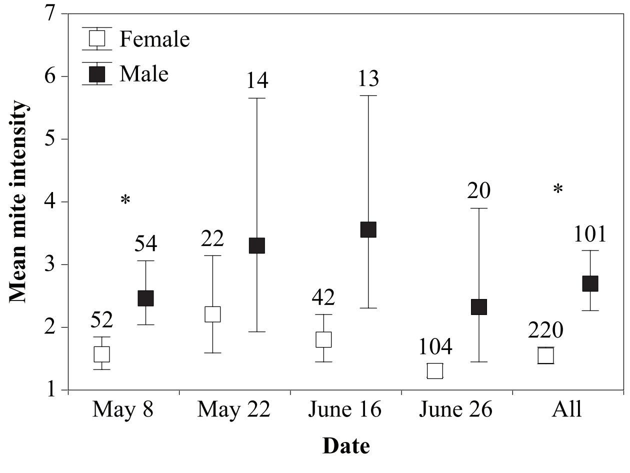

FIGURE 2. Mean mite intensity of each sample and when the samples were pooled for female and male *D. nigrior*. Bars indicate 95% confidence interval calculated by bootstrap tests. Numbers above bars indicate sample size. Significant sex differences ($P < 0.05$) are indicated by asterisks.

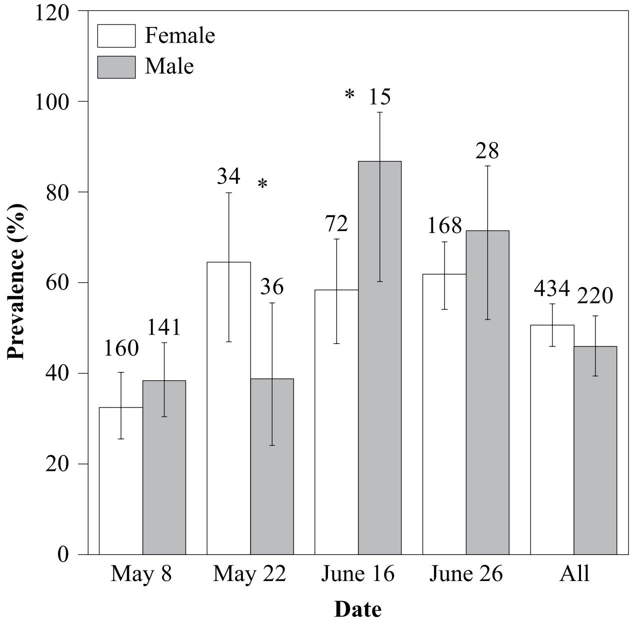

FIGURE 3. Prevalence of mite parasitism for each sample and when the samples were pooled for female and male *D. nigrior*. Bars indicate 95% confidence interval calculated by Sterne's exact method. Numbers above bars indicate sample size. Significant sex differences ($P < 0.05$) are indicated by asterisks.

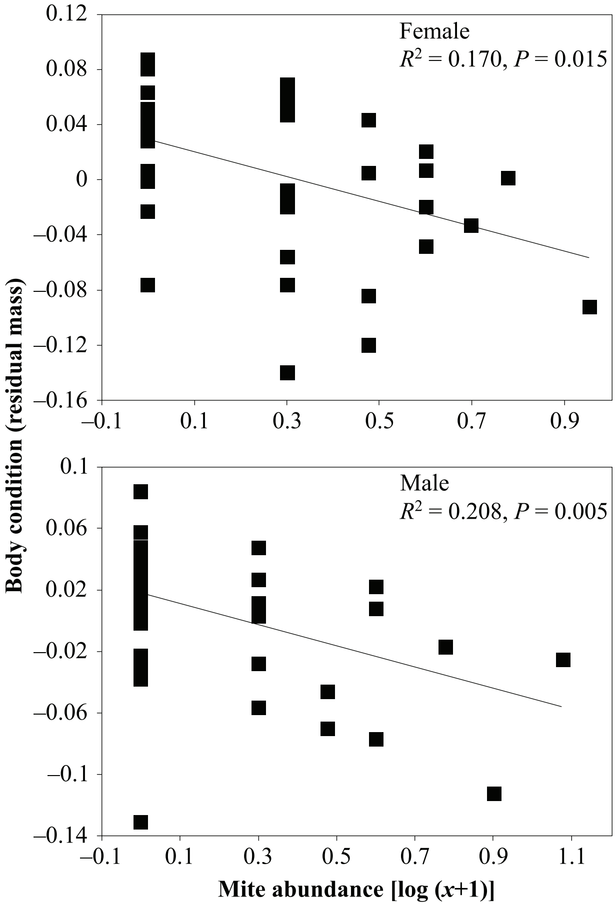

FIGURE 4. Relationship between mite abundance and body condition of female and male *D. nigrior*. A significant negative correlation is present for both sexes. Female n = 34, male n = 36.

about the general behaviour and ecology of gyrinids, and thus speculation into potential causes of the male-biased mite intensity is difficult.

It is possible that male *D. nigrior* are more susceptible to mite parasitism. There may be a trade-off between parasite resistance and other fitness-related functions, such as reproduction (Sheldon & Verhulst, 1996). Mating success is typically more important to the fitness of males than females (Bateman, 1948); thus, males may invest relatively more in functions that increase mating success and relatively less in parasite resistance (Rolff, 2002; Stoehr & Kokko, 2006). To mitigate the costs of mite parasitism some insect hosts employ resistance methods such as melanotic encapsulation of mite feeding tubes (Yourth, Forbes & Smith, 2001) and grooming (Leung, Forbes & Baker, 2001), but it is not known whether gyrinids employ effective resistance against water mites.

It is also possible that males may be more exposed to mites. We observed larval mites attached to teneral hosts in the late summer and early fall (E. Fairn, pers. observ.), and some *Eylais* species can survive the winter on the host and engorge the following spring (Lanciani, 1970). Thus, if male *D. nigrior* eclose earlier in the summer than females (*i.e.,* protandry), they could accumulate more mites before going into the winter period and thus have more mites engorging the following spring. However, it is not known whether protandry is present in this species.

Sex differences in habitat use may also result in males being more exposed to larval mites (Lajeunesse, Forbes & Smith, 2004). One of the few studies of the ecology of *D. nigrior* suggests that males guard larger territories than females for part of the day (Fitzgerald, 1987). Larger territory may increase the probability of encountering larval mites and thus contribute to the higher mite intensity in males. Fitzgerald (1987) also suggests that for part of the day there is a slightly female-biased sex ratio near the pond margins and a slightly male-biased sex ratio near the centre of the pond. If pre-parasitic mites are more heavily concentrated in areas away from the margins, then males may be more heavily exposed.

Female *D. nigrior* are larger than males in terms of several measures of body size, including elytral length, body length, and body height (Fairn, Alarie & Schulte-Hostedde, 2007). Therefore, the hypothesis that the larger sex in sexually size-dimorphic species is more exposed to parasitism (Moore & Wilson, 2002) cannot explain the higher male mite intensity in this system.

An alternative interpretation is that the male-biased mite intensity is not caused by sex differences in exposure or susceptibility to mite parasitism but rather by females suffering higher mite-induced mortality than males at high mite intensity, resulting in females with high intensities being relatively rare. However, it is not known if females are more susceptible to mite-induced mortality, or if mite-induced mortality is present at all.

The negative correlation between host body condition and mite abundance is consistent with water mites of the genus *Arrenurus* on the damselfly *Coenagrion puella* (Rolff, Antvogel & Schrimpf, 2000) and the terrestrial mite *Macrocheles subbadius* parasitizing the fly *Drosophila nigrospiracula* (Polak, 1998). However, no correlation was found between degree of mite parasitism by *Arrenurus* mites and the body condition of the damselfly *Enallagma ebrium*, although heavily parasitized males did not survive as long as lightly parasitized males when food stressed (Forbes & Baker, 1991). The negative correlation is consistent with the hypothesis that water mites reduce host body condition. Mite-induced reduction in body condition of *D. nigrior* could be caused by several mechanisms, including removal of nutrient-rich haemolymph, mobilization of resources to allocate to mounting an immune response, or increased energy expenditure during locomotion due to the burden of carrying the mites. These mites are relatively large (up to approximately 1/5 the length of the host: E. R. Fairn, A. I. Schulte-Hostedde & Y. Alarie, unpubl. data) and appear to engorge a considerable degree on the host (up to approximately 10× in length: Fairn, Schulte-Hostedde & Alarie, unpubl. data). Thus, they may remove considerable resources and may create a substantial burden during locomotion. *Drosophila nigrospiracula* males experimentally infested with parasitic mites had lower body condition than control males that were not exposed to mites, providing experimental support for the ability of mites to reduce host condition (Polak, 1998). There is also evidence that suggests water mites may reduce host body condition. For example, *Hydrachna* mites may reduce survival in a notonectid host (Lanciani, 1982), and *Eylais* and *Hydrachna* mites may substantially reduce investment in egg production in female notonectid and corixid hosts (Davids & Schoots, 1975; Martin, 1975; Lanciani, 1982), suggesting that water mite parasitism may reduce the pool of nutrients available for allocation to reproduction and survival.

The negative correlation between host body condition and mite parasitism is also consistent with the hypothesis that hosts in poor condition are more susceptible to parasitism. If *D. nigrior* employ some form of costly mite defence mechanism, then individuals in poor condition may be less able to invest in the resistance and thus may have heavier mite parasitism. For example, dragonfly larvae in better condition invested more in grooming to prevent mites (Leung, Forbes & Baker, 2001), and body condition was positively associated with encapsulation response in the damselfly *Calopteryx virgo* (Koskimäki *et al.,* 2004). The 2 hypotheses for this relationship are not mutually exclusive and may be synergistic, as a mite-induced decrease in body condition may then make the host less able to invest in an immune response or other defence mechanisms.

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