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Phoretic mites in uni- and bivoltine populations of *Ips typographus*: a 1-year case study

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Abstract: Members of the order Mesostigmata frequently use other animals to colonize suitable habitats, and thus their phoretic exploitation of bark beetles is not exceptional. We compared the abundance and species spectrum of phoretic mites in univoltine and bivoltine populations of the European spruce bark beetle *Ips typographus*. Two localities with high population densities of *Ips typographus* in the eastern part of the Czech Republic (400 and 1100 m a.s.l.) were studied. Beetles were sampled with pheromone traps baited with IT Ecolure (5 per locality). In total, 1268 *I. typographus* individuals were captured, from which 1662 mite individuals were collected and 8 species of phoretic mites were identified. The most numerous species were *Dendrolaelaps quadrisetus* and *Trichouropoda polytricha*. *Trichouropoda polytricha* was dominant at the higher elevation (1100 m a.s.l.). Neither the number of phoretic mites per beetle nor the percentage of beetles carrying the mites differed between the localities. At the locality with bivoltine beetle populations, however, the number of phoretic mites per beetle was greater in the spring generation than in the summer generation.

Key words: Mites, spruce bark beetle, generation, altitude, Central Europe

1. Introduction

The European spruce bark beetle, *Ips typographus* (Linnaeus, 1758), is an important component of every Eurasian spruce forest ecosystem. As a pioneer species, it colonizes dying and newly dead trees and thus initiates the decomposition of bark and wood (Wermelinger, 2004). It nevertheless is also one of the most significant pests of Norway spruce trees in Eurasia (Schroeder, 2001). Substantial research has established that *I. typographus* lives in connection with different groups of organisms, such as fungi (Christiansen et al., 1983), nematodes (Forsse, 1987), and mites (Moser and Bogenschütz, 1984). Some of them, like mites, use the beetle for their dissemination (Moser and Bogenschütz, 1984).

Order Mesostigmata is one of the most numerous and largest orders of all mites. They inhabit many different habitats. Most species are wild predators, parasites, or symbionts of mammals, birds, reptiles, and arthropods (Kranz and Walter, 2009). Mesostigmata live very frequently in relationships with other invertebrates (Hunter and Rosario, 1988), and many mites from that order are found in association with various species of bark beetles (Kiełczewski et al., 1983). The mites accompanying bark beetles also form ecologically interesting relationships with fungi (Klepzig et al., 2001). Phoretic mites are common in temporary and diverse habitats and need other animals for transport (Hunter and Rosario, 1988). These phoretic mites can transfer hyperphoretic ascospores of the ophiostomatoid fungi as with *I. typographus* (Moser et al., 1997), *Scolytus scolytus* (Fabricius, 1775) (Moser et al., 2010), *Dendroctonus frontalis* Zimmerman, 1868 (Hofstetter et al., 2006), and their phoretic mites.

Mites that are dependent on bark beetles can be divided into 2 groups. The first group feeds on the substrate, most frequently fungi, and some of these species can be beneficial for bark beetles (Kiełczewski et al., 1983). The second group is parasitic or predacious on beetles (Kenis et al., 2004). Both groups can use bark beetles for transport. Mortality of eggs in *Ips typographus* can be 10% (Mills, 1985) and it is probable that mites are involved in consumption of eggs (Kiełczewski and Bałazy, 1966; Kinn, 1967; Hofstetter et al., 2009). The possibility that the mites of the second group could be used as biological control agents of bark beetles has been previously suggested (Lindquist, 1969). To date, however, it is not clear whether this approach constitutes an effective and practical option for controlling the numbers of bark beetles. In addition, we do not yet know the entire spectrum of phoretic mites accompanying the beetles, nor do we know their direct influence on beetle populations or the details of their ecology (Kenis et al., 2004).

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Among central European countries, the topic of phoretic mites of *I. typographus* has been addressed in Germany (Moser and Bogenschütz, 1984), Slovakia (Feketová, 2011; Vrabec et al., 2012), and Poland; in Poland, many studies have been conducted on mites that live in the galleries of many species of bark beetles and these studies were summarized by Gwiazdowicz (2008). There also are studies from Poland focusing on the spectrum of phoretic mites on *I. typographus* (Gwiazdowicz et al., 2011, 2012). The same species have been found to be dominant in most studies. To date, however, researchers have not investigated the ecological relationships between phoretic mites and bark beetles in general and between phoretic mites and *I. typographus* in particular. Because *I. typographus* occurs in many different habitats at many different elevations (Dutilleul et al., 2000), the species of mites associated with *I. typographus* can be expected to differ with habitat and elevation. Similarly, differences in the abundance of mites can be expected among seasonal generations in bivoltine *I. typographus* populations.

The most common method for catching bark beetles are pheromone traps (Grégoire and Evans, 2007) and this method has been previously used for the collection of phoretic mites from their bodies (Moser et al. 1989).

The goals of the present study were to compare the spectrum and abundance of phoretic mites of the order Mesostigmata on univoltine and bivoltine populations of *I. typographus*, and to compare the spectrum and abundance of phoretic mites in the spring and summer generations of bivoltine populations of *I. typographus*.

2. Material and methods

Beetles were collected from 2 localities in the Czech Republic, i.e. at Pustá Polom village (49°52′9.817″N, 18°0′7.290″E; 400 m a.s.l.) and at Keprník Mt. (50°10′52.998″N, 17°7′1.732″E; 1100 m a.s.l.). Both localities support Norway spruce (*Picea abies* (L.) H.Karst.) forests. The Pustá Polom locality is a spruce plantation where *I. typographus* produces 2 generations per year (Lubojacký and Holuša, 2011). The Keprník locality, however, contains virgin Norway spruce forests and supports only 1 generation of *I. typographus* per year (Plašil and Cudlín, 2005). In recent years, *I. typographus* numbers have increased at both localities (Plašil and Cudlín, 2005; Lubojacký and Holuša, 2011).

At each locality, beetles were collected from 5 pheromone Theysohn traps, which were baited using IT Ecolure pheromone vaporizers (http://www.fytofarm.cz/). Traps were monitored from April to September and beetles were collected every 7 days. We analyzed only 1 and 2 samples collected at the peak of bark beetle flight activity, respectively. Beetles stayed in the traps for up to 1 week. Beetles were sampled twice at the Pustá Polom locality, on 10 May and 3 August 2012 (constituting mature- and offspring-generation beetles), and once at the Keprník locality on 23 May 2012. The collected beetles were stored in bottles containing 70% ethanol.

Mite specimens of the order Mesostigmata were collected from these beetles in the laboratory. The position of the mites on the beetle body (on the coxae, on the elytra, on the elytral declivity only, and under the elytra) and the number of mites per beetle body were recorded. The elytral declivity is characteristic for some genera of bark beetles including the genus *Ips*, and therefore this location was defined separately. Mites were also collected from the sediment in the bottles into which the beetles had been placed after collection. The collected mites were placed in 70% ethanol and then temporarily mounted in 80% lactic acid. Permanent mounts of certain specimens were prepared for improved lighting and subsequent determination, using Hoyer's solution as the mounting medium. All mite specimens of the order Mesostigmata were subsequently identified to the species level. Taxonomical determination keys were used for identification (Hirschmann and Wiśniewski, 1982; Karg, 1989, 1993; Mašán, 2001). Mites were also compared with material that was determined by Prof Gwiazdowicz (University of Life Sciences, Poznań, Poland) and with specimens of *Schizosthetus simulatrix* Athias-Henriot, 1982 revised by Assoc Prof Feňda (Comenius University in Bratislava, Bratislava, Slovakia).

A chi-square test was used to determine whether the percentage of beetles with phoretic mites differed between the overwintering generations from the Pustá Polom locality and the Keprník locality. The Wilcoxon signed-rank test was performed using the R programming language to compare whether the number of phoretic mites per beetle differed between the localities and between the 2 generations at the Pustá Polom locality. Mites found in the sediment of collection bottles were not included in these analyses.

3. Results

A total of 1268 *I. typographus* beetles were captured, and a total of 1662 mite specimens of the order Mesostigmata were collected from their bodies and from sediment in the collection bottles. From the overwintering generation at Pustá Polom, 500 *I. typographus* beetles were collected, and 454 mite specimens were obtained from their bodies and collection bottles; 37.2% of the beetles had at least 1 mite on their bodies. From the offspring generation at Pustá Polom, 233 *I. typographus* beetles were captured, and 216 mites were obtained from their bodies and collection bottles; 25.3% of the beetles had at least 1 mite on their bodies (Figure). During the single generation at Keprník, 535 *I. typographus* beetles were collected, and 992 mites were obtained from their bodies and collection bottles; 44.3% of the beetles had at least 1 mite on their bodies.

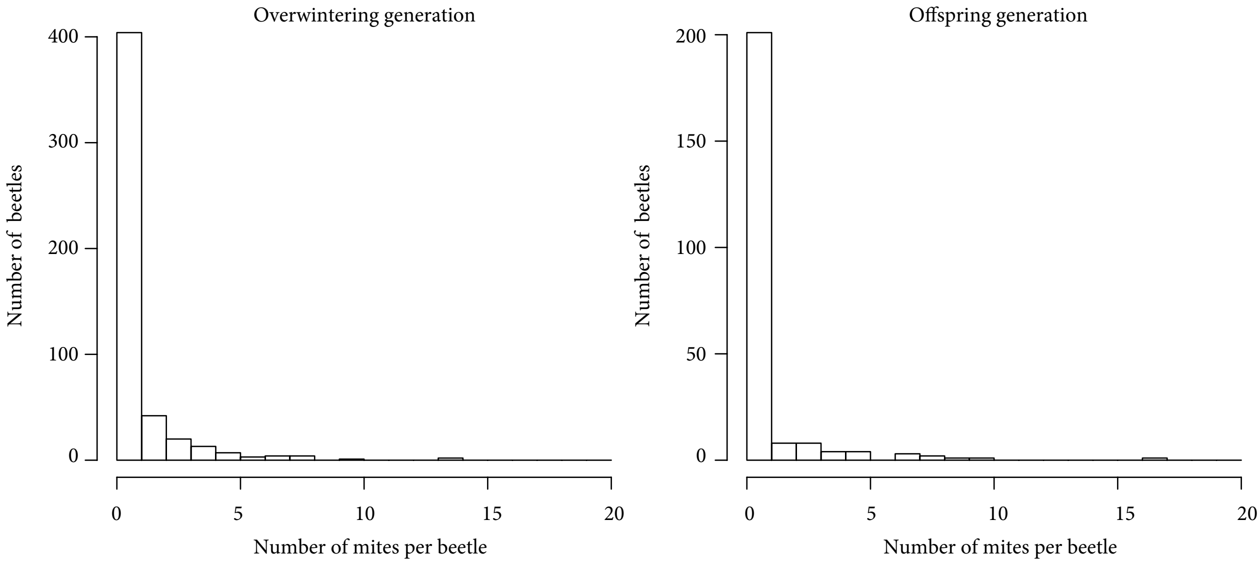

Figure. Number of mites per *Ips typographus* beetle at the Pustá Polom locality in overwintering (left) and offspring (right) generations.

When no distinction was made for generations, the overall percentage of beetles carrying phoretic mites (33.4% at Pustá Polom and 44.3% Keprník) did not significantly differ between the 2 localities ($\chi^2 = 4.1409$, df = 1, P > 0.01). Similarly, the number of phoretic mites per beetle was generally small (<3) and did not significantly differ between the 2 localities (Wilcoxon test, W = 176,420.5, P > 0.01). The percentage of beetles with mites did not significantly differ between the overwintering generation at Pustá Polom and the single generation at Keprník ($\chi^2 = 0.4049$, df = 1, P > 0.01); the same was true for the number of mites per beetle (Wilcoxon test, W = 125,041.5, P > 0.01). At Pustá Polom, however, the number of phoretic mites per beetle was greater in the overwintering than in the offspring generation (Wilcoxon test, W = 64,663.5, P < 0.01) (Fig. 1).

In total, 8 species of phoretic mites were identified from the 2 localities. There was a difference in species composition (6 species at Pustá Polom and 8 species at Keprník), because the species of *Schizosthetus simulatrix* and *Trichouropoda polonica* Wiśniewski and Hirschmann, 1988 were missing in Pustá Polom. The mite *Dendrolaelaps quadrisetus* (Berlese, 1920) was the most abundant and most represented (53.0%) of all mites collected. The second most numerous species was *Trichouropoda polytricha* (Vitzthum, 1923) (31.5% of the mites), followed by *Uroobovella ipidis* (Vitzthum, 1923) (12.0% of the mites). Each of the other species represented <5% of the mites, and these included *Pleuronectocelaeno austriaca* Vitzthum, 1926; *Proctolaelaps fiseri* (Samšiňák, 1960); *Schizosthetus simulatrix*; *Trichouropoda polonica*; and *Uroobovella vinicolora* (Vitzthum, 1926) (Tables 1 and 2).

Table 1. Total number of mites by species on *Ips typographus* beetles (and in sediment of collection bottles) from Pustá Polom. The total number of studied beetles was 733.

Mite species	Numbers of mites at Pustá Polom as a function of position on body (or in the collection vial)					
	On coxa	On elytra	Elytral declivity	Under elytra	Sediment	Total
Dendrolaelaps quadrisetus		4		577	46	627
Pleuronectocelaeno austriaca				3		3
Proctolaelaps fiseri					1	1
Trichouropoda polytricha	1		6	6	9	22
Uroobovella ipidis	3		6		1	10
Uroobovella vinicolora		5	2			7
Total	4	9	14	586	57	670

Table 2. Total number of mites by species on *Ips typographus* beetles (and in sediment of collection bottles) from Keprník. The total number of studied beetles was 535.

Mite species	Numbers of mites at Keprník as a function of position on body (or in the collection vial)					
	On coxa	On elytra	Elytral declivity	Under elytra	Sediment	Total
Dendrolaelaps quadrisetus				247	6	253
Pleuronectocelaeno austriaca					1	1
Proctolaelaps fiseri					6	6
Schizosthetus simulatrix					10	10
Trichouropoda polonica					1	1
Trichouropoda polytricha	25		123	2	352	502
Uroobovella ipidis	93	1	17	1	78	190
Uroobovella vinicolora	3	1	1		24	29
Total	121	2	141	250	478	992

For most species, only deutonymphs were found. In the case of *P. austriaca*, all individuals were adults and both sexes were found, while there were only adult females in the case of *P. fiseri*.

With respect to position on the beetle, mites were most abundant under the elytra; 50.3% were found under the elytra, followed by 32.2% in the sediment of the collection bottles, 9.3% in the elytral declivities, 7.5% on the coxae, and 0.7% on the elytra. *Dendrolaelaps quadrisetus* was dominant under the elytra, while *T. polytricha* was dominant in the sediment and in the elytral declivity. *Uroobovella ipidis* appeared more often than other species on the coxae, and *U. vinicolora* appeared more often than other species on the elytra.

4. Discussion

The percentage of *I. typographus* beetles carrying phoretic mites and the number of phoretic mites per beetle was not statistically significant different at lower and higher elevations. The species spectra did differ between the 2 locations and included only a few species, as has been reported from other locations (Moser and Bogenschütz, 1984; Moser et al., 1989; Gwiazdowicz et al., 2011, 2012).

The most abundant mite overall was *D. quadrisetus* which is very often found in association with *I. typographus* (Moser and Bogenschütz, 1984; Moser et al., 1989; Gwiazdowicz et al., 2011). This species is also found with other species of bark beetles (Pernek et al., 2008). It is considered a predator of the eggs of the host bark beetle because mite females have been observed to suck out the contents of one egg after another (Kiełczewski and Bałazy, 1966). The mite *T. polytricha* also occurs very often and is frequently described as a phoretic species on *I. typographus* (Feketová, 2011; Gwiazdowicz et al., 2012). The results presented here indicate that *D. quadrisetus* dominates at lower elevations while *T. polytricha* dominates at higher elevations. The latter species inhabits coniferous stands mainly at medium to high elevations (up to 1200 m a.s.l.) (Mašán, 2001). These 2 are the species most frequently found in association with bark beetles at most locations (Gwiazdowicz et al., 2011; Vrabec et al., 2012).

Other species recorded in this study and in previous studies include *U. ipidis* and *U. vinicolora* (Moser and Bogenschütz, 1984; Feketová, 2011), *P. austriaca* (Gwiazdowicz et al., 2011), *P. fiseri* (Gwiazdowicz et al., 2011; Penttinen et al., 2013), and *T. polonica* (Gwiazdowicz et al., 2011). *Schizosthetus simulatrix*, however, has not been previously mentioned in similar studies with *I. typographus*. It is possible, however, that individuals of this species are mistaken for other representatives of the subfamily Parasitinae (Kalúz et al., 2003). Moser and Bogenschütz (1984) and Moser et al. (1989) reported finding *Vulgarogamasus* sp., but this was probably *S. simulatrix* (Kalúz et al., 2003). This species could also be considered a predator of host eggs because its close relative, *S. lyriformis*, has been observed feeding on the eggs and larvae of the bark beetle *Ips pini* (Say, 1826) (Hofstetter et al., 2009).

The locations on the beetle where the individual mite species were found were consistent with previous reports (Moser and Bogenschütz, 1984; Feketová, 2011; Vrabec et al., 2012). *Dendrolaelaps quadrisetus* is most often found under the beetle's elytra, while *T. polytricha*, *U. ipidis*, and *U. vinicolora* are found in various locations on the beetle body and in the sediment of collection bottles (Moser and Bogenschütz, 1984; Feketová, 2011; Vrabec et al., 2012).

At the Pustá Polom locality, the number of phoretic mites per beetle was higher in the mature beetles of the overwintering generation than in the offspring beetles. We suggest that this could be a result of the fact that bark beetle larvae feed in isolation. Their larval galleries do not intersect and remain filled with frass and wood dust. If bark beetles are not abundant, the galleries developed by the maturation feeding of offspring beetles are unlikely to intersect with those of the maternal beetles. Therefore, transfer of mites from maternal beetles to offspring beetles is restricted. Similar findings have been reported for nematodes that parasitize bark beetles (Tenkáčová and Mituch, 1987).

All of the mite species detected here occur commonly in various parts of the *I. typographus* range (Moser et al., 1997; Penttinen et al., 2013). In this study, as in others (Moser and Bogenschütz, 1984; Moser et al., 1989; Gwiazdowicz et al., 2011, 2012), only a small number of phoretic mite species were found with *I. typographus*, even though approximately 60 species of mites of the order Mesostigmata have been described from *I. typographus* galleries (Gwiazdowicz, 2008). It seems that the number of mites per beetle is similar among beetle populations.

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