Phoresy of *Trichouropoda shcherbakae* Hirschmann, 1972 (Acari: Mesostigmata) on beetles of the genus *Tetropium* Kirby, 1837 (Coleoptera: Cerambycidae) in Białowieża Forest, Poland

1. Introduction

Mesostigmatid mites are small arachnids, whose body length is usually less than 1 mm. Such a small body size impedes migration over long distances and colonization of specific isolated micro-habitats such as rotten wood, the nests of birds and mammals, anthills, animal excrement, carcasses etc. [1-9]. Therefore they have developed the ability to disperse using other, more mobile animals, such as insects [10-13]. Phoretic relations between insects and mites have been the subject of numerous research papers, which usually have faunistic character or analyze ecological links [2,10,13]. The phenomenon of mite phoresy on insects that inhabit tree bark (e.g. Curculionidae: Scolytinae) has been studied by different authors [14-17]. However, the relationship between insects and mites during different stages of the life cycle of the host - for example, immediately after the insect leaves its feeding site under the bark, during insect swarming, and shortly before the insect’s death has not been examined.

Longhorn beetles (Cerambycidae) are among the best known beetle families in Poland. Polish fauna includes 195 species, grouped in six subfamilies [18]. In Europe, there are six known species of the genus *Tetropium* (subfamily Spondylidinae): *T. aquilonium* Plavilstshikov, 1940, *T. castaneum* (Linnaeus, 1758), *T. fuscum* (Fabricius, 1787), *T. gabrieli* Weise, 1905, *T. lucorum* Eichhorn, 1846, *T. longipennis* (Olivier, 1789). The study was conducted in the Białowieża Forest, which is recognized as one of Europe’s last natural forest areas. Insects were caught over a period of 8 weeks in 72 attractant traps (type Intercept IPM). In total 1250 specimens of genus *Tetropium* were collected. We analyzed 524 beetles, including 295 specimens of *T. castaneum* and 229 specimens of *T. fuscum*. On 49 beetles (9.4%) there were 785 individuals of *Trichouropoda shcherbakae* (Trematuridae). Mites were more common on *T. fuscum*, which carried 82% of all collected deutonymphs. Most of the mites found on beetles were attached to their legs. This study reports on changes in the intensity of phoresy in time and location of mite deutonymphs on their host species.

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**Abstract:** The aim of this study was to assess the phoretic relationship between two beetle species of the genus *Tetropium* and mites from order Mesostigmata. The study was conducted in the Białowieża Forest, which is recognized as one of Europe’s last natural forest areas. Insects were caught over a period of 8 weeks in 72 attractant traps (type Intercept IPM). In total 1250 specimens of genus *Tetropium* were collected. We analyzed 524 beetles, including 295 specimens of *T. castaneum* and 229 specimens of *T. fuscum*. On 49 beetles (9.4%) there were 785 individuals of *Trichouropoda shcherbakae* (Trematuridae). Mites were more common on *T. fuscum*, which carried 82% of all collected deutonymphs. Most of the mites found on beetles were attached to their legs. This study reports on changes in the intensity of phoresy in time and location of mite deutonymphs on their host species.

**Keywords:** Mites • Brown Spruce Longhorn Beetle • Longhorn beetles • Uropodina
T. gracilicorne Reitter, 1889 and T. tauricum Shapovalov, 2007 [19]. Their development proceeds under the bark and wood of coniferous trees. The first three species prefer spruce (Picea spp.), T. gabrieli is monophagous and feeds on larch (Larix spp.) [20,21], and T. gracilicorne inhabits trees of the genus Abies, Larix, Picea and Pinus [22]. Tetropium tauricum is a recently described species and little is known about its biology [23].

Beetles of the genus Tetropium inhabit weakened, dying and dead trees, but they are not classified as dangerous forest pests (three species are classified as quarantine pests) [22,24,25]. Adults of T. fuscum appear in the Białowieża Forest around mid-May, and the last specimens can be found even in July. Maximum abundance falls on the second and third decade of May and the first decade of June. The appearance period of T. castaneum takes place around the same time, but its maximum falls in the first decade of June. Generations of both species are annual [26,27]. Details of the biology of both species can be found in Juutinen [21]. One of these species – T. fuscum – was introduced to Canada (near the port of Halifax, Nova Scotia), and was first observed there in 1999 [28]. There it began to spread rapidly, and presented a much greater threat to local forests than to those in Europe. It attacks trees of the following species: Picea rubens, P. glauca, P. mariana and P. abies, and even infests healthy trees [24,29,30]. The Canadian Forest Service began extensive research in order to develop tools for survey, detection and control of this species [24,31-33]. Some of the experiments were carried out in the Białowieża Forest, in cooperation with one of the authors of the present paper.

The aim of the present studies was to investigate the phenomena of phoresy occurring on beetles of the genus Tetropium with special reference to the time between the imagines fly out from under the bark and become less active after swarming. The project constitutes a part of comprehensive studies on the impact of beetles of the genus Tetropium on forest ecosystems and their role in the dispersal of other invertebrates, including mites [21,26,27]. The need for these studies is shown by the fact that phoresy on beetles of the family Cerambycidae is only very poorly known [34,35].

2. Experimental Procedures

The research area was Białowieża Forest, which is considered to be the best preserved forest area in the European Lowland [36]. It is widely recognized as a model forest, that various observations and research in natural deciduous and mixed forests of this climatic zone can be referred to [37]. The research site was located in the old growth forest, of almost natural character, in mixed forest and fresh forest habitat, compartment 496C (N52°40’, E23°47’), about 4 km SW of the village of Białowieża. The forest here includes species such as Norway spruce Picea abies, English oak Quercus robur, European hornbeam Carpinus betulus, Scots pine Pinus sylvestris, aspen Populus tremula, and European silver birch Betula pendula.

Mites were obtained during research coordinated by Dr. Jon Sweeney (Canadian Forest Service) and devoted to the search for the most effective means to detect and collect Tetropium fuscum in Canada. The material was collected in 72 traps, from April to June 2007. Traps were established on April 30th and monitored for a period of 8 weeks. During the experiment, traps were emptied four times (every 2 weeks). The traps were baited with the following attractants: racemic fuscumol, R-fuscumol, S-fuscumol, spruce blend, ethanol [33].

Distribution of the attractant substances was random. Traps within blocks were spaced 25-30 m apart, and blocks within 50-100 m of one another. Traps were suspended from a rope tied between two trees so that the trap was at least 1 m away from each tree, and the collecting bucket was ~10 cm above the ground (IPM-Intercept traps). The IPM-Intercept trap has two intersecting panels (30.5 by 80.5 cm) with a top and bottom funnel (40 cm square at the top opening by ~20 cm deep), a collecting cup (13.5 by 13.5 by 14.5 cm deep), and a 0.18 m² cross-sectional area between the top and bottom funnels [32].

The killing agent in the collecting cups was a 50:50 mixture of ethylene glycol and water. All Cerambycidae specimens were preserved in 70% ethanol. All specimens of Tetropium fuscum and T. castaneum were then individually examined in the laboratory under a stereomicroscope to determine the sex, and to determine whether they were infested with mites. Beetles with attached Uropodina mites were selected for further research. These beetles were then analyzed under a stereomicroscope Olympus SX16 in order to identify the number of present deutonymphs and their location on host. From selected mite specimens permanent preparations in Faure’s medium were made. The remaining specimens were identified from temporary preparation after clearing in lactic acid using an open slide technique. We did not find any detached deutonymphs of T. scherbakae that fell off the beetles, as some authors noticed [17]. Since the number of detached deutonymphs was not determined directly in traps, we should state that number of transported deutonymphs might be underestimated.

The material was deposited in Natural History Collections, at the Faculty of Biology, Adam Mickiewicz University in Poznań.
3. Results

3.1 Changes in intensity of phoresy in time

Phoretic mites were found on *Tetropium castaneum* and *T. fuscum*. A total 1250 specimens of genus *Tetropium* was collected. The study material consisted of 524 specimens of *Tetropium* beetles (42% of all), including 295 specimens of *T. castaneum* and 229 specimens of *T. fuscum*. Mites were found on 49 specimens of beetles (9.4%), and included 785 specimens, all of which were deutonymphs (Table 1).

We observed variation in the intensity of phoresy in consecutive collection periods. In the first period no phoresy was observed (beetles emerge from pupal cells). The highest number of beetles (38) carrying mites species was collected in the second half of May (period when beetles swarm and lay eggs). In the case of *T. fuscum* over 2/3 of specimens carried *T. shcherbakae* Hirschmann, 1972, and in the case of *T. castaneum* about 1/5. From 29th of May to 25th of June we recorded a clear decrease in the number of infested beetles, although the number of transported mites was still relatively high (general decrease in the activity of beetles). A significant decrease in both the intensity and the number of carried mites was observed in the second half of June.

3.2 Preference of *T. shcherbakae* for host species and place of attachment on its body

Of the two analyzed beetle species, mites were more common on *T. fuscum*, which carried 82% of all collected deutonymphs. The number of deutonymphs of *T. shcherbakae* found on this species was also higher in particular research periods than on *T. castaneum*.

Most of mites found on beetles were attached to their legs (Figure 1, Table 2). They constituted 92% of all mites found on *T. fuscum* and 82% found on *T. castaneum*. The pronotum of both species carried 6% and 7% of deutonymphs respectively, and the abdomen 0.5% and 5%. Only in one case did we find *T. shcherbakae* on the head of *T. castaneum*. This confirms earlier observations that mites do not attach themselves to the head of insects, which could hinder its orientation in space [4]. In the case of *T. fuscum* most mites attached to the legs were found on first legs (Table 2).

![Acarina on Brown Spruce Longhorn Beetle. A - deutonymphs of Trichouropoda shcherbakae attached to the legs of Tetropium fuscum, B - close-up of deutonymphs.](image-url)

Table 1. Number of collected beetles, percentage of infested specimens and number of mites per host. N - number of collected beetles, I (infestation) - % of beetles transporting mites, D (density) - number of mites on host beetle.

<table>
<thead>
<tr>
<th>Host species</th>
<th>Collection period</th>
<th>N</th>
<th>I</th>
<th>D</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>T. fuscum</em></td>
<td>30.04-14.05.2007</td>
<td>20</td>
<td>-</td>
<td>0</td>
</tr>
<tr>
<td><em>T. castaneum</em></td>
<td>30.04-14.05.2007</td>
<td>7</td>
<td>-</td>
<td>0</td>
</tr>
<tr>
<td><em>T. fuscum</em></td>
<td>15.05-28.05.2007</td>
<td>42</td>
<td>78.6%</td>
<td>480</td>
</tr>
<tr>
<td><em>T. castaneum</em></td>
<td>15.05-28.05.2007</td>
<td>27</td>
<td>18.5%</td>
<td>120</td>
</tr>
<tr>
<td><em>T. fuscum</em></td>
<td>29.05-11.06.2007</td>
<td>134</td>
<td>3.7%</td>
<td>166</td>
</tr>
<tr>
<td><em>T. castaneum</em></td>
<td>29.05-11.06.2007</td>
<td>234</td>
<td>1.7%</td>
<td>17</td>
</tr>
<tr>
<td><em>T. fuscum</em></td>
<td>12.06-25.06.2007</td>
<td>33</td>
<td>3.0%</td>
<td>1</td>
</tr>
<tr>
<td><em>T. castaneum</em></td>
<td>12.06-25.06.2007</td>
<td>27</td>
<td>3.7%</td>
<td>1</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>524</td>
<td>9.4%</td>
<td>785</td>
</tr>
</tbody>
</table>

Table 2. The percentage of deutonymphs (D) attached to various parts of host body.

<table>
<thead>
<tr>
<th>Part of beetles' body</th>
<th><em>T. fuscum</em></th>
<th><em>T. castaneum</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>head</td>
<td>0.0%</td>
<td>0.9%</td>
</tr>
<tr>
<td>pronotum</td>
<td>6.1%</td>
<td>7.4%</td>
</tr>
<tr>
<td>elytra</td>
<td>0.7%</td>
<td>3.5%</td>
</tr>
<tr>
<td>legs I</td>
<td>55.6%</td>
<td>31.4%</td>
</tr>
<tr>
<td>legs II</td>
<td>24.7%</td>
<td>15.3%</td>
</tr>
<tr>
<td>legs III</td>
<td>10.8%</td>
<td>34.9%</td>
</tr>
<tr>
<td>ventral surface</td>
<td>1.4%</td>
<td>1.3%</td>
</tr>
<tr>
<td>abdominal sternites</td>
<td>0.3%</td>
<td>5.2%</td>
</tr>
<tr>
<td>Total</td>
<td>295 D=100 %</td>
<td>229 D=100 %</td>
</tr>
</tbody>
</table>
pair of femora (60%) (Figure 1). Significantly fewer mites occurred of the second pair (27%) and third pair (12%) of femora. A different situation was observed in *T. castaneum*. In this case, the highest number of mites was observed on the femora of third pair of legs (42%). Mites were also quite numerous on the femora of second pair of legs (37%). The lowest number of mites was found on the femora of the first pair of legs (18%), and we did not find mites attached to the other parts of the legs.

4. Discussion

A substantial number of publications on phoresy of *Uropodina* treat it as a kind of curiosity, without deeper analysis. Only a few papers contain information about the preferences of deutonymphs for its host or attachment to specific parts of its body [4,17,34,38-41]. Some studies have confirmed that mite distribution on the host’s body is nonrandom [4,42,38,43]. Al-Deeb et al. [42] showed that mites (*Sancassania* sp.) clearly preferred abdominal segments, apparently to protect themselves from high temperature.

In our research we found that most of mites (92% for *T. fuscum* and 82% for *T. castaneum*) were attached to legs. A similar situation was observed in the study of Bajerlein and Błoszyk [4]. In their research the number of deutonymphs decreased from the third to the first pair of legs. They suggested that deutonymphs climb onto the back of beetle and move forward and that this spatial distribution only slightly affects the locomotion of beetles. A similar pattern of attachment occurs in *T. fuscum*, but a different situation is observed for *T. castaneum*. Further studies are required to explain this issue.

Individual specimens of *Uropodina* have no significant impact on the condition of their host beetle, unless they are located on sensory organs (e.g. mouthparts, antennae, eyes or wings) [44]. Individual phoretic mites may also influence host behavior if their size is relatively large comparing to the beetle (e.g. *Uropoda orbicularis* (Müller, 1776) on hydrophilid beetles) [45]. Moreover Kinn and Witcosky [46] report that despite the fact that mites do not prevent dispersal of their insect host, they may alter the flight behavior, shorten dispersal distance and influence the height of flight. Fronk [47] suggested that for *Dendroctonus frontalis* Zimmerman, 1868, the upper limit of the number of transported deutonymphs over which the beetle could not fly was 40. Moser [14] however found that flying individuals of the same species of bark beetle could carry 60 specimens of *Trichouropoda australis*

Hirschmann, 1972. Nevertheless, a very large number of deutonymphs of *T. scherbakae* found on some beetle individuals (maximum number of deutonymphs on *T. fuscum* - 149, on *T. castaneum* - 97) can be considered as an example of transport parasitism. Consumption of energy for the flight of an insect is in this case much larger, and the distance covered is shorter. It is also possible that such a heavily loaded beetle could be easier prey for predators. This problem is showed on the example of *Sancassania* deutonymphs and *Oryctes agamemnon arabicus* beetles [42]. In that study the authors found 3057 deutonymphs on one female of *O. a. arabicus*. Some of the deutonymphs were attached to host eyes and mouthparts or even spiracles, probably affecting vision and therefore its behavior.

However, this raises the question of why the deutonymphs should attach themselves in such large numbers. After all, in such cases, they would be eaten with their carrier, removing at each time large number of individuals from population. So it appears that such cases occur relatively infrequently and from an evolutionary point of view, it is more normal for each beetle to carry a low number of mites.

We also noted that phoresy was the most intensive in the second half of May, just before the time when the highest number of beetles occurred. In the second half of June, despite the presence of similar number of beetles to the second half of May, the intensity of phoresy was very low. That suggests that deutonymphs of *T. scherbakae* show seasonality or most likely, the synchronization of life cycle of mite and beetle occurs. The primary task of phoretic stages is in fact colonization of as many micro-habitats as possible. With very limited dispersal ability, mites may do so only through transportation on beetles. Other interactions of mites and beetles are poorly known, but they may include fungivores and parasitoids of immature stages of beetles (as were described in bark beetles) [17, 48]. Whether the observed associations between certain species of beetles and mites are only limited to phoresy requires further detailed study.

Phoresy on beetles is a very widespread phenomenon among mites of the suborder *Uropodina*. It was reported on representatives of various families of beetles, but so far we have little information on the presence of specific species of *Uropodina* on representatives of the family Cerambycidae. Mašán [49] in his work dedicated to *Uropodina* of Slovakia lists four species of the genus *Trichouropoda* Berlese, 1916 carried by four different species of longhorn beetles. These are: *Trichouropoda polysetosa* Mašan, 1999 found on *Cerambyx cerdo cerdo* Linnaeus, 1758, *T. scherbakae*, recorded on *Aegosoma scabricorne* (Scopoli, 1763), *T. livorniana*
Phoresy of Trichouropoda shcherbakae Hirschmann, 1972

Wiśniewski and Hirschmann, 1988 associated with *Rhagium inquisitor* (Linnaeus, 1758) and *T. tuberoza* Hirschmann and Zirngiebl-Nicol, 1961 occurring on *Rusticoclytus rusticus* (Linnaeus, 1758). Our observation of *T. shcherbakae* on two species of the genus *Tetropium* extends the range of vectors of this mite.

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