

Overwintering Ecology of *Philonthus rotundicollis* (Coleoptera, Staphylinidae) in the Nest of the Carpenter Ant *Camponotus herculeanus* on the Coast of the Sea of Okhotsk

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Abstract—A widespread rove beetle species, *Philonthus rotundicollis*, whose distribution range stretches across different climatic zones, including the coldest regions of the Asian northeast, was discovered as an inquiline within the nests of the carpenter ant *Camponotus herculeanus* on the coast of the Sea of Okhotsk in winter. It remained unclear if the beetles had significant cold-hardiness and whether they overwintered deep in the soil or were confined to particularly warm habitats. To clarify these aspects, the following metrics of cold-hardiness were measured: supercooling point (SCP), freezing point (FP), supercooling capacity (SCP–FP), and temperature minima at the beetles' overwintering sites. In *Ph. rotundicollis*, mean SCP was $-11.1 \pm 0.7^\circ\text{C}$ (ranging from -7.9 to -18.8°C , $n = 15$), which was insufficient for successful overwintering even on the coast, since temperature minima in leaf litter during a snow-deficient winter fell to -14°C at the depth of 5 cm and -12°C at 20 cm. The beetles could not burrow deep into stiff soil and made use of crevices in dry peat-like soil layers as well as tunnels of soil- and root-dwelling animals, including carpenter ants. The presence of this rove beetle species in the ant nest was probably due to feeding on ant larvae because, at near-zero temperatures, the activity threshold of the beetles was lower than that of the ants that guarded the larvae.

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Ant nests harbor numerous symbiotic invertebrates, including neutral cohabitants. The greatest number of such inquilines is found in the nests of Palearctic ants of the genus *Formica*, where only beetles amount up to 100 species, about 60 of which are rove beetles (Dlussky, 1976). One of the latter, *Philonthus rotundicollis* (Menetriés, 1832) (Coleoptera, Staphylinidae) was discovered by us during winter excavations of the nests of the carpenter ant *Camponotus herculeanus* Linnaeus, 1758 (Hymenoptera, Formicidae) in the environs of Magadan. The nature of their relationship was unknown. This is a Palearctic species distributed across different climatic zones, except for the Arctic (Tikhomirova, 1973). Previous finds of this beetle in the upper reaches of the Kolyma River (Ryabukhin, 2011), which is a region with very cold winters, suggest that this species is highly cold-tolerant or, alternatively, overwinters deep in the soil or is confined to particularly warm habitats.

The aim of this work was to study the cold-hardiness of *Ph. rotundicollis* and compare it with thermal conditions at the overwintering site.

MATERIALS AND METHODS

The upper reaches of the Kolyma River would be a more suitable collection site, given the aims of this study, since this is the coldest part of the species' distribution range. However, the beetle is not abundant there, and so it is not possible to collect a sufficient number of experimental animals. We were able to measure their cold-hardiness owing to a fortuitous discovery of about a hundred overwintering beetles in abandoned chambers of a nest of the carpenter ant *Camponotus herculeanus* in the environs of Magadan (the excavations were carried out in March to study the ecology of the ant). The beetles were taken out of the ant nest, transported to the laboratory at a temperature not higher than -5°C and not lower than -10°C , and kept in a refrigerator at -5°C during the experimental work. Cold-hardiness of the beetles removed from the nest was measured in accordance with previously described methods (Berman et al., 2007; Leirikh and Meshcheryakova, 2015) as the supercooling point (SCP) and the fraction of beetles that have recovered from dormancy. The SCP was determined in a custom-designed freezing chamber that allowed cooling down

the experimental animals (weighing up to 10 g) at a rate of 1°C/min. Manganin-constantan thermocouples for SCP measurements were embedded in a lockable thick-walled copper box (5×8×8 cm) that ensured uniform cooling of the animals. The beetles were attached to thermocouple junctions with a thin layer of petroleum jelly. The box with beetles was put in the freezing chamber.

The fraction of beetles that survived overwintering in nature, following their removal from ant winter chambers, was counted after termination of dormancy by keeping the beetles for 1 day at 0°C and then for 1 day at 5°C. Thermal conditions for overwintering were analyzed with the use of previously collected data from soil layers of approximately 100 habitats in the upper Kolyma basin and environs of Magadan (Alfimov et al., 2009, 2012; Berman et al., 2007, and other sources). The temperature dynamics in the *C. herculeanus* nest in the environs of Magadan from which the beetles were sampled was tracked with DS1922L loggers during the winter of 2014–2015. The loggers were installed on the surface of the tree trunk and in the partially decomposed wood beside the winter chambers at the depths of 5, 10, and 20 cm from the soil surface. The temperature was recorded twice a day.

RESULTS AND DISCUSSION

In the northeast of Asia, *Ph. rotundicollis* is known from the upper Kolyma basin and from the north of the Sea of Okhotsk region; on the coast of Tau Bay, it is ubiquitous but never abundant. The beetle can be found primarily in forest leaf litter, in plant debris accumulated at the shores, in the moss, and seldom on carrion; it is encountered infrequently from June to August (Ryabukhin, 2011).

The overwintering ecology has not been studied. Most rove beetle species are known to overwinter in leaf litter; much less commonly, they dig burrows (Tikhomirova, 1973; Babenko, 1997). The *C. herculeanus* nest on the coast of the Sea of Okhotsk, where an aggregation of *Ph. rotundicollis* was discovered, was situated in a sparse larch forest on the lower part of a south-facing slope. The nest was made in a fallen larch tree that had been buried in the ground at the roadside of an abandoned forest track. Winter chambers were located in the partially decomposed butt-log portion and in the trunk of the tree at the depths of

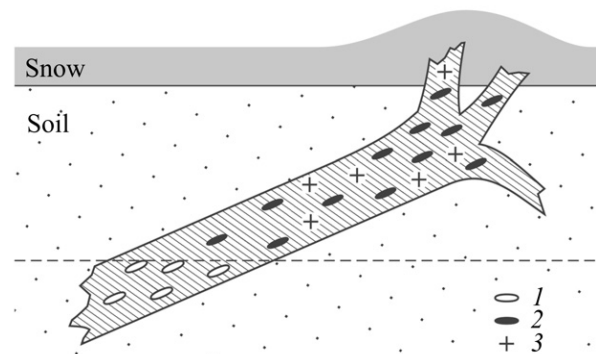


Fig. 1. Schematic representation of the location of winter chambers in the carpenter ant nest within a buried larch trunk: 1, chambers with workers, larvae, and alate males and females; 2, chambers with workers, males, and females, but no larvae; 3, chambers with beetles *Philonthus rotundicollis* only.

5–25 cm from the soil surface. About 20 chambers, connected by tunnels, occupied the lower part of the roots and trunk of the larch and continued inwards (Fig. 1). In the chambers, there were approximately 3000 workers, 500 alate males and females, and about that many early instar larvae, which were very small (up to 1 mm). The larvae were only found in the lowermost chambers, in the layer 20–25 cm deep. In the 9 carpenter ant nests excavated earlier in the upper Kolyma basin and the Sea of Okhotsk region, late instar larvae (up to 2 mm in length) usually constituted up to 30% of the nest inhabitants and were present both in the upper (5–10 cm) and deeper (15–20 cm) chambers (Berman et al., 2007).

About a hundred beetles were dispersed in groups of 3–10 individuals throughout the abandoned ant chambers and tunnels that were filled with bore dust and ant waste (from the surface to the depth of 20 cm). However, ants and beetles never occurred together in any of the chambers. In general, the beetles amounted to 2% of the total ant family and were exclusively represented by adults. No beetle larvae were found in the nest.

Cold-hardiness. Rove beetles *Ph. rotundicollis*, similar to ants, survive negative temperatures in a supercooled state and die at temperatures below SCP, i.e., during freezing. The SCP of the beetles had a mean value of $-11.1 \pm 0.7^\circ\text{C}$ ($n = 15$) and ranged from -7.9 to -18.8°C (Fig. 2). The mean freezing point (FP) was $-5.7 \pm 1.03^\circ\text{C}$, and the mean supercooling capacity (the difference between SCP and FP) was $4.6 \pm 0.4^\circ\text{C}$ (table). The carpenter ants from the same nest turned out to be twice as cold-hardy (table).

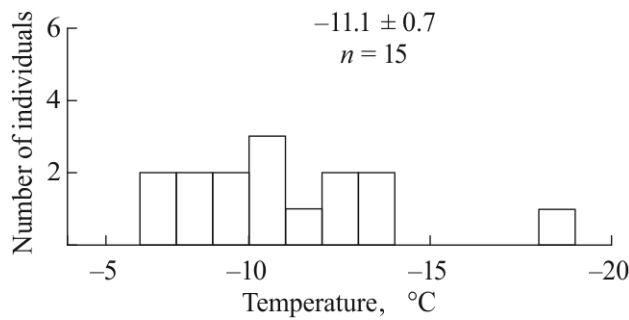


Fig. 2. Distribution of supercooling points of the beetles *Philonthus rotundicollis* from the nest of the ants *Camponotus herculeanus*.

Thermal conditions for overwintering on the coast of the Sea of Okhotsk are relatively mild: the January mean temperature is -17°C and the mean absolute minimum air temperature is -30°C (Handbook on the Climate..., 1990). The temperature minima at a depth of 15–20 cm, given the snow cover of 5–20 cm, barely reach -15°C , while under 60 cm snow these values fall only slightly below -7.0°C (Alfimov et al., 2012; Berman and Zhigulskaya, 2012).

During the cold and snow-deficient winter of 2014–2015, in the *C. herculeanus* nest where the overwintering beetles were discovered, the chambers at depths of 5 and 20 cm had temperatures below -10°C for 10 consecutive days (in the last third of January). The minima of -14°C at the depth of 5 cm and -12°C at 20 cm held for 1 and 2 days, respectively, while the minimum air temperature was -25°C and the snow cover above the tree trunk was 5 cm thick (Fig. 3). During the second half of the winter, even though the air temperature was still that low, the temperature in the nest did not fall below $-6\text{...}-7^{\circ}\text{C}$ due to an increase in snow cover up to 40 cm.

Comparison of cold-hardiness of *Ph. rotundicollis* (SCP values from -7.9 to -18.8°C) and the temperature minima in the soil layers where the beetles overwintered (-14°C at the depth of 5 cm and -12°C at

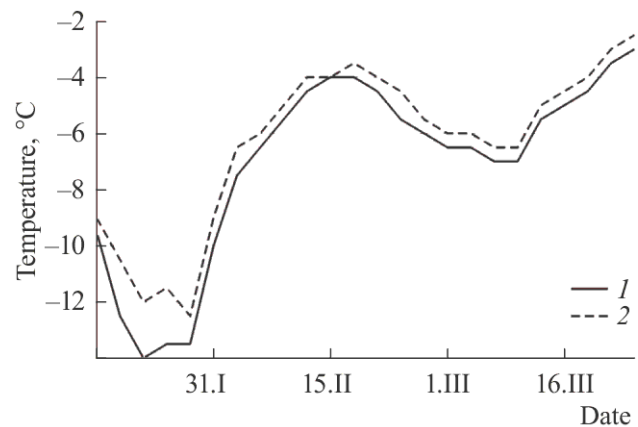


Fig. 3. Thermal conditions for overwintering of the beetles *Philonthus rotundicollis* in the nest of the ants *Camponotus herculeanus* in 2014–2015 in the environs of Magadan. Temperature minima: 1, at the depth of 5 cm; 2, at the depth of 20 cm.

20 cm) shows that the beetles with SCP $> -14^{\circ}\text{C}$, which comprised 70% of the sample, did not even have a minimal “safety margin” to withstand cold during a snow-deficient winter (i.e., a difference between values from -7.9 to -14°C , at which they would not die, and the actual temperature minima in the nest). The only exception was a single individual with SCP = -18.8°C (Fig. 3).

However, it was already clear during the excavation of the nest that *Ph. rotundicollis* had overwintered successfully: disturbed beetles started to move at the air temperature of -6°C . All the 35 individuals that were removed from the nest and kept in Petri dishes with a moistened cotton wool wad for 1 day at 0°C and then for 1 day at 5°C were active.

This comparison suggests that the thermal conditions at the overwintering sites of *Ph. rotundicollis* on the coast of the Sea of Okhotsk in snow-deficient winters or in snowless habitats are marginal even in the carpenter ant nests, the tunnels of which allow the beetle to penetrate the soil to the depth of 20 cm.

Cold-hardiness ($^{\circ}\text{C}$) of the rove beetles *Philonthus rotundicollis* and ants *Camponotus herculeanus* from the environs of Magadan

Species	Date	SCP			FP		SCP–FP	
		mean	minimum	<i>n</i>	mean	<i>n</i>	mean	<i>n</i>
<i>Ph. rotundicollis</i>	27.III.2015	-11.1 ± 0.7	-18.8	15	-5.7 ± 1.03	15	4.6 ± 0.4	15
<i>C. herculeanus</i>	25.III.2015	-27.5 ± 0.5	-33.1	40	-13.0 ± 0.4	40	14.6 ± 0.4	40

The occurrence of the beetles in the ant nest might not be accidental. Having initially colonized partially decomposed wood of the tree stump, they might have fed on the ant waste and also possibly on the ant larvae. The nests could have been particularly attractive to the beetles in autumn, after the first ground frosts. Ants are immobilized even at near-zero temperatures, while the beetles remain active. According to observations made by Babenko (1997), rove beetles are still able to forage under the snow at mild subzero temperatures and attack small enchytraeids. According to our observations, the air temperature during excavation of the nest was $-6...-8^{\circ}\text{C}$, and opening of the chambers induced the beetles to move and hide in bore dust, while the ants remained motionless. The beetles might have fed on the larvae in autumn until the soil became frozen, which is indirectly attested to by the absence of larvae in all of the upper chambers to the depth of 20 cm. There were no beetles further down, exactly where the ant larvae were found in great numbers. In several carpenter ant nests excavated by us in winter in the upper Kolyma basin (Berman et al., 2007), both the shallow and deep chambers harbored larvae.

The thermal conditions in the Kolyma basin, where *Ph. rotundicollis* is also present, are even harsher. The temperature minima during the winter of 1977–1978 in two *C. herculeanus* nests at the depth of 10 cm were $-25...-26^{\circ}\text{C}$; the minimal air temperature on those days reached -38°C . The temperature below 0°C was recorded for 7 months, including 5 months below -10°C and 3 months below -15°C (Berman et al., 2007, and other sources). Thus, overwintering sites for this rove beetle species in continental regions are even more limited.

CONCLUSION

The cold-hardiness of the rove beetle *Ph. rotundicollis* seems to be inadequate, given that its distribution range includes the coldest territories of the Asian northeast. However, there exists an analogy to this phenomenon. Not all of the ant species from the upper Kolyma basin have cold-hardiness that corresponds to background soil temperatures in that harsh ultra-continental climate. Part of the species (*Formica exsecta* Nylander, 1846; *F. sanguinea* Latreille, 1798; *F. candida* F. Smith, 1878; *Myrmica angulinodis* Ruzsky, 1905, and others) are obligatorily confined to

particularly warm yet small biotopes, such as places of snowdrift formation on downwind slopes, talik layers, and loose and moderately wet porous peat. *Formica lemami* Bondroit, 1917 builds nests on loose slope-wash, where tunnels reach the depth of over 120 cm and terminate in winter chambers (Berman et al., 2007).

As shown in this communication, this is exactly how *Ph. rotundicollis* copes with winters on the coast of the Sea of Okhotsk: it makes use of the carpenter ant tunnels to penetrate into warmer (relative to the surface) soil layers. It can be readily appreciated that the distribution of *Ph. rotundicollis* in the northern territories is also facilitated by its ability to penetrate deep into the soil via cavities and crevices. In addition, this rove beetle may be attracted to ant nests due to potential predation on ant larvae late in autumn, when the rove beetles remain active and the worker ants have already entered chill coma and cannot protect their young.

According to the results of the study of cold-hardiness of *Ph. rotundicollis* on the coast of the Sea of Okhotsk, the distribution of this poikilothermic species in colder continental regions of the Asian Northeast does not a priori imply that this species has significant cold-hardiness.

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