# Studying tachinids at the top of the world



### Notes on the tachinids of Northeast Greenland

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#### Introduction

Over the past seven years, we have spent several weeks every summer exploring the host-parasitoid foodwebs of Northeast Greenland. To sample host larvae for rearing, we have crawled over the ground, turned over rocks and swung our sweep nets. To catch adult parasitoids (and adult hosts) in flight, we have used Malaise traps, pitfall traps, pan traps and extensive hand netting. This field work has primarily been implemented by Tomas Roslin and Gergely Várkonyi, while Jim O'Hara has helped identify the tachinids from the catches. Helena Wirta has coordinated the molecular



Figure 2. Zackenberg Research Station. (Photo: Gergely Várkonyi)

work shedding additional light on the fauna and its host associations. As our previous reports from this area have been focused on disentangling the structure of the larger food web by various means (Roslin *et al.* 2013, Várkonyi & Roslin 2013, Wirta *et al.* 2014), we here wish to summarize our findings from an explicit tachinid perspective. Thus, our primary focus will be on the tachinid fauna and its ecology in Northeast Greenland. From a geographic perspective, our specific emphasis will be on the Zackenberg Valley, with additional observations from Traill Island.

#### Study area

Starting in 2007, the first two summers were spent on Traill Island ( $72^{\circ}33'N 23^{\circ}35'W$ ; Fig. 1). In 2009, we moved our activities some 200 km to the North, to the Zackenberg Valley ( $74^{\circ}30'N 21^{\circ}00'W$ ). Here, the Aarhus University, Department of Bioscience (Denmark), operates the Zackenberg Research Station – a facility providing excellent logistic support in an otherwise inaccessible region (Figs. 2, 3).

Both study sites are located in the high-Arctic zone of Northeast Greenland, and part of the Northeast Greenland National Park. With a size equaling Western Europe, this park forms one of the largest uninhabited areas of the globe. While the interior of the park is covered by the Greenland Ice Sheet, the coastal area is characterized by continuous permafrost soil with a maximum active layer thickness varying from 20–100 cm. The mean monthly air temperature ranges between  $-20^{\circ}$ C and  $+5^{\circ}$ C (Meltofte & Rasch 2008).

On Traill Island, our activities were focused on the Karupelv Valley, at an elevation of less than 200 m.a.s.l. In the southeast, this valley is bordered by the mountain of Svinhufvuds Bjerge (1378 m.a.s.l.) and in the northwest by Kongeberge (1884 m.a.s.l.).

At Zackenberg, we have focused our activities on the Zackenberg river valley. Being located farther north than Traill Island, this area is characterized by a slightly more "lush" vegetation, as shown by a higher component of *Salix* (still only some cm in height). The overall flora of the Zackenberg Valley consists of >150 vascular plant species (Bay 1998). The main vege-



**Figure 3**. Habitats of the Zackenberg Valley from the ground and the top of Dombjerg Mtn. (Photos: Gergely Várkonyi)

tation types include fen, grassland, salt marsh, *Cassiope tetragona* (Linnaeus) Don (Ericaceae) heath, *Vaccinium uliginosum* Linnaeus (Ericaceae) heath, *Dryas* Linnaeus (Rosaceae) heath dominated by the hybrid of *D. octopetala* Linnaeus and *D. integrifolia* Vahl, snowbeds with *Salix arctica* Pallas (Salicaceae), abrasion plateaus, fell-fields, and lake vegetation (Bay 1998).

Within the Zackenberg area, we have distributed our sampling efforts across multiple complementary techniques, with the aim of documenting both the adult parasitoid community associated with Lepidoptera, and the host associations of individual parasitoids. For the specific methods used for both purposes, see Várkonyi & Roslin (2013), Roslin *et al.* (2013) and Wirta *et al.* (2014).

As a specific advantage for anyone attempting to establish trophic interactions within a host-parasitoid web, Northeast Greenland is characterized by a depauperate flora and fauna. With a total terrestrial fauna and (vascular) flora of approximately 500 and 170 species, respectively (Roslin *et al.* 2013), this scores as one of the least diverse regions in the world. At Zackenberg, the local lepidopteran community comprises 20 species representing 11 families (for a list of species involved, see Várkonyi & Roslin 2013; beyond this list, *Psycho*- polata (Duponchel) (Geometridae) and Euxoa drewseni (Staudinger), Polia richardsoni (Curtis), Sympistis nigrita (Boisduval) and Syngrapha parilis (Hübner) (all Noctuidae; Höykinpuro 2010).

### Methods for establishing host associations

Our initial efforts at establishing host associations for the parasitoids of the area were based on the traditional rearing of hosts until the hatching of either the adult host or its parasitoid. For this purpose, we collected and reared more than 400 lepidopteran larvae from Traill Island, and 1450 larvae from Zackenberg. In the next phase of the project, we applied novel techniques based on molecular information for the same purpose. Here, we used order-specific primers for selectively amplifying the DNA of one species in each interspecific interaction. Using this technique, we were able to sequence gut contents of adult flies and parasitoid contents of host larvae (for details, see Wirta *et al.* 2014). Excitingly, the former approach has proven sensitive enough to allow us to amplify DNA remaining in the



**Figure 4.** (Left) A female *Peleteria aenea* reared from a larva of *Apamea zeta*. (Right) Adults of *P. aenea* are often seen on semi-open sandy ground in sunny weather. The two strong setae on the lower portion of the parafacial (arrow) easily distinguish *Peleteria* from other Greenland tachinids. (Photos: Gergely Várkonyi, ex. Zackenberg Valley)

*phora sabini* (Kirby) (Geometridae) was detected as a species new to the area in 2013 by TR). Their hymenopteran parasitoids include 27 species representing three hymenopteran families (Ichneumonidae with 19 species, Braconidae with seven species, and Eulophidae with one species; for species, see Várkonyi & Roslin 2013). Traill Island features a handful of additional lepidopteran species, but here our rearings were focused on eight: *Gynaephora groenlandica* (Wocke) (Lymantriidae), *Boloria* spp. (Nymphalidae; *B. polaris* (Boisduval) and/or *B. chariclea* (Schneider), from neither of which the larva has been described), *Entephria*  gut of the adult parasitoid – even after metamorphosis (discussed by Rougerie *et al.* 2011).

In exploring the best source of host DNA for identifying host remnants from parasitoids, we made an interesting discovery regarding the impact of tachinid morphology and lifestyle: when we extracted DNA separately from the head, thorax, and abdomen of parasitoids, the head of tachinids yielded significantly more identifiable sequences than did other body parts – a pattern contrasting with that observed for wasps (see Wirta *et al.* 2014). This difference among parasitoid orders may be linked to differences in their biology: as tachinids emerge from their puparia by first inflating and then deflating a balloon-like ptilinum from behind the face, host DNA may be caught in the resultant folds of the ptilinum as it is retracted into the head. This shows how basic knowledge of tachinid biology may help the molecular ecologist in selecting appropriate samples.

### The tachinid fauna of Zackenberg

On Traill Island, we detected only one species of tachinid from more than 400 host larvae reared and Malaise traps run over a full summer. This species was *Exorista* sp. (Exoristinae; see below for the identity of this species). At Zackenberg, our even more extensive sampling of the fauna revealed the presence of the same *Exorista* species, along with two other tachinids, *Periscepsia stylata* (Brauer & Bergenstamm) (Dexiinae) and *Peleteria aenea* (Staeger) (Tachininae) (Fig. 4). Quite remarkably, a single sequence of parasitoid DNA detected from within a host larva indicated the presence of a tachinid species never observed as an adult fly in the area (Figs. 5, 6). This sequence differs from *P*.

stylata by 5.4% and was detected within a larva of *Boloria chariclea* (in this case, the host *Boloria* was identified to species on the basis of its DNA barcode) with no higher match for any previously barcoded tachinid species. This pattern either suggests the occurrence of cryptic variation within *P. stylata*, or the existence of another species of *Periscepsia* new to Zackenberg. Hence, our DNA-based methods proved useful in revealing the tachinid species richness in this Arctic area (Wirta *et al.* 2014).

**Figure 5**. A phylogenetic hypothesis for tachinid haplotypes detected in Zackenberg, Northeast Greenland. This phylogeny is based on DNA sequences of the barcoding gene region, and reconstructed by the neighbor-joining technique.

### Which *Exorista* species occurs in Northeast Greenland?

Exorista thula Wood (Exoristinae) was originally described from several localities in northern Canada and reported as a parasitoid of Gynaephora groenlandica (Morewood & Wood 2002). However, in their revision of tachinid parasitoids of Arctic Gynaephora, Morewood & Wood (2002) suggested that the Greenlandic rearing record of the Palaearctic species Exorista fasciata (Fallén) from G. groenlandica reported by Henriksen & Lundbeck (1918) might actually refer to *E. thula*. The situation became further confused when Monty Wood (Canadian National Collection of Insects, Ottawa) examined our Exorista specimens reared from Traill Island and noted that "about half of them resemble the *thula* from Ellesmere Island, Pearyland [northern Greenland], and other far northern Canadian localities, sufficiently closely to be able to call them thula with confidence. However, the other half has wider abdominal bands, although the bands are quite variable in width, and seem more like our specimens of fasciata as identified by Mesnil. This raises the issue



of what really is *fasciata*. These specimens show me clearly why *fasciata* appeared on the Greenland list of Diptera, but whether these variable specimens really represent a different species from the *thula* or whether *thula* is more variable on Traill Island is a matter for gene sequencing to sort out.... Perhaps *thula* is a variant of *fasciata*, but I would not like to say so without seeing more specimens from northern Greenland and Europe." (e-mail from M. Wood to James O'Hara on January 10, 2009).

To resolve the issue of what Exorista species occurs in Northeast Greenland, we have now sequenced a set of tachinids from both Traill Island and Zackenberg for the CO1 barcoding region. To construct a molecular phylogeny for this material, we used the program MEGA5 (Tamura et al. 2011), applying both neighbour joining and maximum likelihood techniques with default values. Here, the specimens from both Traill Island and Zackenberg form a single cluster, and are thus of the same species (Fig. 5). This was a somewhat unexpected outcome given the previously observed variation in morphology. We also sequenced several specimens identified as E. thula by M. Wood from Ellesmere Island and Banks Island and these clustered with our Exorista from Traill Island and Zackenberg. We therefore feel confident that our species is E. thula, if indeed

this is a valid species. We further sequenced other *Exorista* specimens that were identified as *E. thula* (Yukon), *E. fasciata* (Finland), *E. larvarum* (Linnaeus) (Canada and Western Europe), and *E. mella* (Walker) (Canada and United States). For these the results were far from clear, suggesting that the identifications – and the characters upon which they were based – are unreliable. There is clearly a need for further molecular and morphological work to resolve this difficult Holarctic species complex.

## Host associations of individual species

Based on our efforts to understand the biology and host use of tachinids in Traill Island and Zackenberg, we offer the following observations on individual species (see also Várkonyi & Roslin 2013):

*Exorista thula* has been reared thrice from *G*. *groenlandica* at Zackenberg (for details, see Várkonyi & Roslin 2013), and 40 times from the same host species on Traill Island. Our molecular techniques add an interesting twist to these observations, as in Zackenberg this parasitoid was detected from three *Apamea zeta* 



**Figure 6**. The food web of tachinids and their lepidopteran hosts found in Northeast Greenland, as resolved by three different techniques: traditional rearing (applied to 1450 host larvae), MAPL-HL (Molecular Analyses of Parasitoid Linkages; Host Larvae as the source, as applied to 1195 host larvae), and MAPL-AP (Adult Parasitoids as the source, as applied to 76 adult flies; redrawn from Wirta *et al.* 2014).

(Treitschke) larvae and from one of *Sympistis nigrita* ssp. *zetterstedtii* (Staudinger) (Fig. 6). In addition to the material examined as larvae, we also screened a set of host pupae for the presence of parasitoid DNA (Wirta *et al.* 2014): twenty individual pupae were collected in the wild, representing two species (19 pupae of *Gynaephora groenlandica* and 1 pupa of *Polia richardsoni*). Among the pupae of *G. groenlandica*, we detected three cases of parasitism by *E. thula*.

*Peleteria aenea* is a common species at Zackenberg, but was not encountered on Traill Island. Yet, it is known from all around Greenland and the Canadian Arctic (Morewood & Wood 2002). While our observations from traditional rearing have previously led us to concur with Morewood & Wood (2002) that this species parasitizes *A. zeta* and not *G. groenlandica* (Várkonyi & Roslin 2013), our molecular data offers a more diversified view of the host use by this species. While *P. aenea* has been reared twice at Zackenberg from a large larva and a prepupa of *A. zeta*, it is also found in *G. groenlandica* and *S. nigrita* ssp. *zetterstedtii* (Fig. 6). At Zackenberg, adults of this species are most often encountered in July (Várkonyi & Roslin 2013).

### The ecological significance of high-Arctic tachinids

Our quantitative screening of large sets of host larvae tells us about the general prevalence of tachinids in the harsh climate of Northeast Greenland. On Traill Island, the average parasitism rates of Exorista thula on G. groenlandica reached 17 to 31% in two different materials collected in 2007 and 2008 by TR and by Juha Höykinpuro, respectively (summarized in Höykinpuro 2010). Within the Karupelv Valley, these rates differed significantly between smaller sites, reaching levels as high as 67% on sandy soils (Höykinpuro 2010). At Zackenberg, parasitism rates by tachinids were notably lower: of 1195 host larvae screened for the presence of parasitoid DNA, only 2.4% tested positive for tachinids (Wirta et al. 2014). This finding suggests low parasitism rates at Zackenberg as a whole, and is further supported by the rearing records from Zackenberg: out of a total of 1450 rearings of Lepidoptera larvae, only five (0.3%) produced tachinids. In general, these findings suggest substantial variation at both a local (among sampling sites within the Karupelv Valley) and regional scale (between Traill Island and Zackenberg, as separated by >200 km). Nonetheless, they reveal that even in the High Arctic, biotic interactions involving tachinids are diverse.

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