Heliozela resplendella (Stainton, 1851) and *H. hammoniella* Sorhagen, 1885: two valid species distinguishable in the genitalia of both sexes and life histories (Heliozelidae)

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Abstract. The taxonomy of a sibling species pair of *Heliozela*, i.e. the alder-feeding *H. resplendella* (Stainton, 1851) and the birch-feeding *H. hammoniella* Sorhagen, 1885 has been somewhat unstable for a long time. In this study, we use reared material to show that the two taxa differ not only in their life histories, but also in the genital morphology of both sexes. We also show, using geometric morphometric tools, that the wing shape and pattern differences presented earlier are not reliable, but that there are some other average differences in wing pattern as well. A lectotype is designated for *H. resplendella*.

Introduction

Heliozelidae is a relatively small, non-ditrysian Lepidopteran family with thirteen genera worldwide (Scoble 1995). Four genera, altogether comprising eight species, have been reported from Europe (Nieukerken 2004). Adult moths are small and usually dark with silvery markings on their forewings. The larvae of the northern European species are leaf miners of Betulaceae and Fagaceae (Emmet 1976). The genus *Heliozela* Herrich-Schäffer, 1853 comprises three northern European species, of which the oak-feeding *H. sericiella* (Haworth, 1828) is easily distinguishable by characters of external morphology, genitalia, and life history (Emmet 1976). The other two species, *H. resplendella* (Stainton, 1851) and *H. hammoniella* Sorhagen, 1885 are difficult to distinguish by their external or internal morphologies (Emmet 1976). Practically all that has been known is that the former species feeds on *Alnus* spp. and the latter on *Betula* spp. (Emmet 1976). Subtle differences in forewing shape and patterns were presented by Meyrick (1928) and Benander (1953). According to their observations, the forewings of *H. hammoniella* are greyer (lighter) and less bronze, while the hindwings are darker, the termen of the forewing less oblique and the distance between the two dorsal spots smaller than in *H. resplendella*. The validity of these characteristics has apparently never been confirmed, and Emmet (1976) considered Meyrick's observations doubtful. No diagnostic genital characters have been presented, probably due to the very complex male genital structure and possibly also to the scarcity of reared material. Reared moths are difficult to obtain, due to their peculiar habits of mining and larval pupation (see below). Since both alder and birch commonly occur together, the adult specimens collected from nature can rarely been identified reliably using the food plant as a cue. In the absence of accurate diagnostic characteristics, the species were synonymized by Kuchlein & de Vos (1999). Similarly, in Denmark, both species were placed under the label *H. resplendella* in a local distributional checklist (Karsholt et al. 1985), and the

validity of *H. hammoniella* was questioned, based on the above-mentioned differential problems. In Finland, this principle was not followed (Kullberg et al. 2001) because the Finnish distributional data is predominantly based on reports on early stages rather than adults, but particularly because of the different distributions of the species: the northern border of *H. hammoniella* is about two hundred kilometres south of that of *H. resplendella* (personal observations). Moreover, it seems that the fluctuations in population sizes are not similarly synchronized between species (personal observations).

On the basis of vacated mines, both taxa are known to occur widely in Finland. In the early 1980's, the second author (JI) noticed that a careful search helps to locate larval cases on the ground below vacated mines, particularly at sandy sites. During the next two decades, the authors, accompanied by Tomi Mutanen and Panu Välimäki, succeeded to rear dozens of specimens of both species.

In this paper, we revise the life history and morphological differences of the species. The mines, adults, and genitalia of both species are illustrated. We also discuss the possible differences in wing shape and patterns using geometric morphometric tools (Bookstein 1989, 1991; Rohlf and Marcus 1993). Despite many attempts we found that male genitalia are not appropriate for shape analysis due to their cylindrical form and small size, which did not permit even separated parts of genitalia to be mounted in a sufficiently uniform way. This method uses sets of two- or three-dimensional coordinates of landmark points, which are superimposed (that is, variation in size, rotation and location eliminated) and subjected to standard multivariate statistical analyses. The crucial distinction between the two morphometric approaches is that traditional metric morphometric methods capture mostly size information, but only limited information of shape (Bookstein et al., 1985, Rohlf and Marcus, 1993), while the geometric morphometric method eliminates all non-shape variation from the data through superimposition, and captures *all* shape variability of the raw landmark data. Moreover, the method enables illustrative visualizations of shape differences by thin-plate-spline deformation grids or vector plots. In the past ten years, geometric morphometrics have increasingly been used in taxonomic and systematic studies (Rohlf & Archie 1984; Rohlf et al. 1996; Fulford & Rutherford 2000; Drotz et al. 2001; Monti et al. 2001; Pretorius & Scholtz 2001; Querino et al. 2002; Baylac et al. 2003; Gumiel et al. 2003; Becerra & Valdecasas 2004; Pretorius 2005; Mutanen 2005).

In all geometric morphometric analyses, a combination of true landmarks and sliding semi-landmarks was applied (for landmarks, see Bookstein 1997 or Adams et al. 2004). This approach was used to simultaneously test differences in forewing outline shape and wing markings. The points on the outline which could be defined precisely across all specimens and species within a species group were applied as true landmarks, while all the others were allowed to slide along the outline trajectory in order to reduce uninformative variation in that direction. The landmark digitations were carried out using the TPSDIG 1.40 program (Rohlf 2004a) and the definitions of sliders with the TPSUTIL 1.26 program (Rohlf 2004b). To eliminate all non-shape variation (variation by location, scale, and orientation), the digitized landmark data was subjected to Procrustes superimposition (Rohlf and Slice 1990). The superimposition and calculation of partial warp scores were done using the TPSRELW 1.35 program (Rohlf 2003). The partial warp scores of the superimposed landmarks were applied
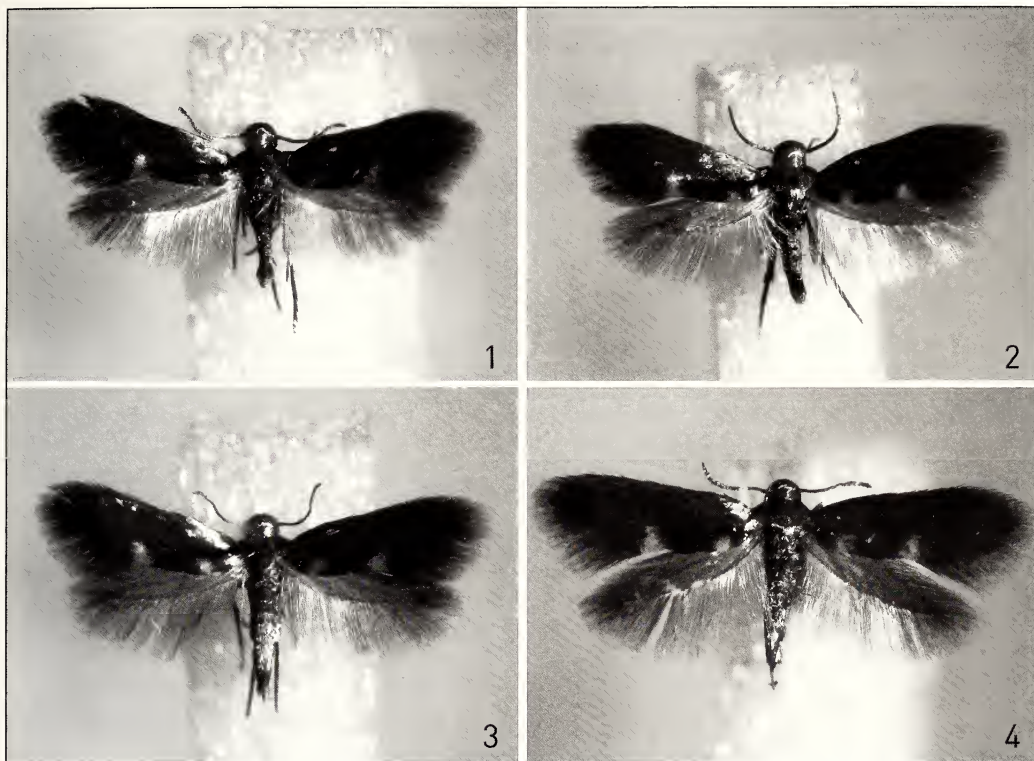
Figs 1–4. Adults of *Heliozela*. **1.** *H. resplendella*, male. **2.** *H. resplendella*, female. **3.** *H. hammoniella*, male. **4.** *H. hammoniella*, female.

for exploratory relative warp analysis (=principal component analysis of partial warp scores) and confirmatory statistical analyses (Multivariate ANOVA).

Redescription of species

Heliozela resplendella (Stainton, 1851)   (Figs 1–2, 5–6, 8, 10–12, 16, 18, 20–24, 28–29)

Aechmia resplendella Stainton, 1851: 6.

M a t e r i a l . Lectotype male, here designated: 'Type' [rounded label with red margin]; West Wickham, ENGLAND, 2.VI.1850, DGL. Coll. (Mason 1906), (168); Walsingham Collection, 1910-427; Aechmia resplendella Dgl. Mss., Stn. Sppl. Cat. Br. Tin. Pbi. 6 (1851) TYPE Dgl. Mss 168 [handwritten in label bordered with black]; resplendellum, Dougl. [printed]; B. M. Genitalia slide No. 29829. – **Finland:** PPs: Hailuoto 720:39, 12♂, 14♀, larva viii.2000 (Alnus incana), M. & T. Mutanen leg. (slides MM 614, 616, 620, 621, 624, 626, 628, 630, 632, 634, 655); PPs: Hailuoto, Pöllä 720:39, 1♂, 11.vi.1999, T. Mutanen leg.; PPs: Hailuoto, Pöllä 720:39, 1♀, larva 1999 (Alnus), M. Mutanen leg.; PPp: Kemi, Ajos 728:38, 1♀, e.l.1996 (Alnus incana), M. Mutanen leg.; KP: Kalajoki, Saarenkari 712:34, 1♀, 19.–20.vi.2000, M. Mutanen leg.; KP: Kalajoki, Saarenkari 712:34, 1♀, 28.vi.2000, M. Mutanen leg. (slide MM 654); KP: Kalajoki, Rahja 712:33, 1♂, 1♀, 19.–20.vi.2000, M. Mutanen leg. (slide MM 618); KP: Lohtaja 711:32, 1♂, 1♀, 28.vi.2000, M. Mutanen leg. (slide MM 656); PPs: Hailuoto, Pöllä 720:39, 6♂, 7♀, e.p.viii.2000 (Alnus incana), P. Välimäki leg.; PPS: Hailuoto 720:39, 7♂, 3♀, pupa 16.viii.2003 (Alnus incana), J. Itämies leg. (slides MM 644, 648, 649, 650); PP: Tornio, Kalkkimaa 7313:384, 1♂, larva 29.vii.1989, J. Itämies leg.; St: Rauma mlk. 680:20 1♂, 1♀, vii.1988, J. Itämies leg.
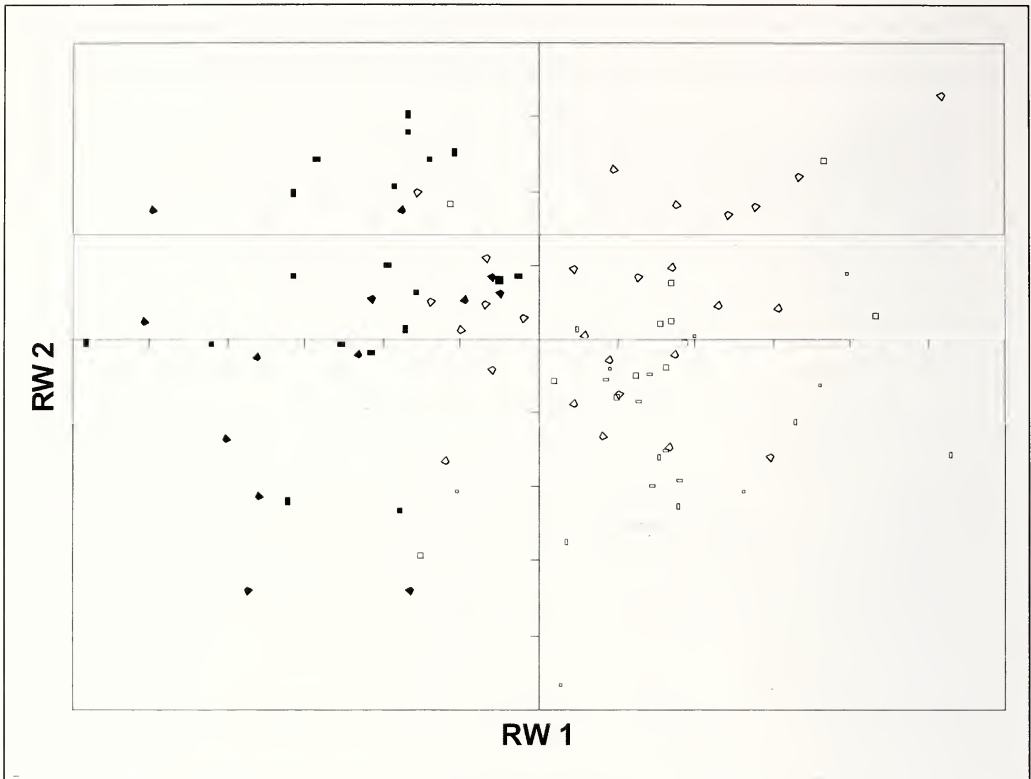
Fig. 5. Results of Relative Warp Analysis (Principal Component Analysis of partial warp scores) of Heliozela forewing outline and pattern shape. Black squares: *H. hammoniella* male, black diamonds: *H. hammoniella* female, open squares: *H. resplendella* male, open diamonds: *H. resplendella* female.

Diagnosis. E x t e r n a l   m o r p h o l o g y. The male average forewing length is 2.61 mm (n=29), that of the female is 2.72 mm (n=29). The female is larger than the male (t=3.196, df=56, p=0.002), and the male is slightly larger than that of *H. hammoniella* (t=-2.135, df=46, p=0.038). Female size is the same in both species (t=0.7414, df=40, p=0.463). The forewing ground colour and the amount of bronze varies, but does not allow reliable differentiation from *H. hammoniella*. Similarly, the hindwing colour appears the same in both species. Meanwhile, statistical treatment of the geometric forewing shape data indicates consistent differences between the species in this respect (F=12.767, df=28, p<0.005). The relative warp analysis showed that the species are distinguishable by the relative warp one, but not by the relative warp two or the other relative warps (Fig. 5). As shown by thin-plate spline deformations grids (Fig. 6), the relative warp axis one reflects mostly the change in the shape of the proximal dorsal spot, which in *H. resplendella* is more hook-shaped and bent towards the wing apex. There is some overlap, but the majority of specimens can be distinguished by this character only. There is a statistically significant difference between the sexes as well (F=2.621, df=56, p<0.005), and it seems that males are more reliably distinguished by this character than females. Wing shape analysis did not reveal differences in the obliqueness of the termen between the species.
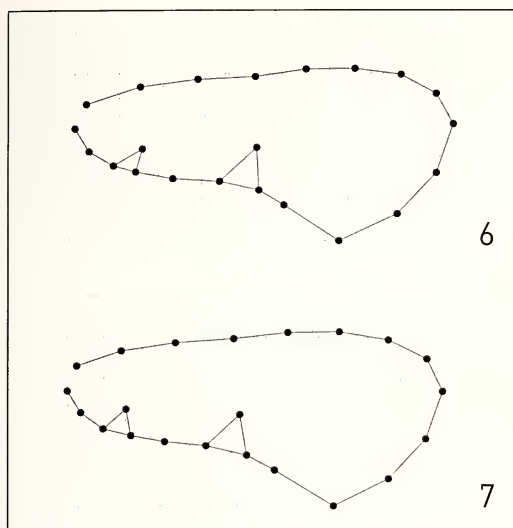
Figs 6–7. Average wing pattern differences between *H. resplendella* (Fig. 6) and *H. hammoniella* (Fig. 7), shown with thin-plate-spline deformation grids and compared to a consensus configuration over both species.

Male genitalia. The species can only be safely identified by the male fultura inferior (might be attributable to anellus) that surrounds the phallus. In *H. resplendella*, there are 8–10 teeth that are comparatively small and not bent at the tip and therefore never have a hook-like appearance (Figs 10–12, 16). In *H. hammoniella*, there are ten stout teeth that are larger than those of *H. resplendella*, and many of them are clearly hook-shaped. There is almost no variation in this character, but to see the teeth clearly, we recommend removal of the phallus from the external genitalia.

Female genitalia. Females are easily distinguishable by the absolute lengths of the apophyses anteriores and posteriores. Both are statistically very significantly longer in *H. resplendella* (apophyses anteriores: $t=-9.869$, $df=13$, $p<0.005$; apophyses posteriores: $t=-7.141$, $df=13$, $p<0.005$), and there is no overlap (Figs 20–21). In *H. resplendella*, the mean length of the apophyses anteriores is 0.993 ± 0.030 mm and the mean length of the apophyses posteriores 1.659 ± 0.044 mm, while in *H. hammoniella* the corresponding values are 0.841 ± 0.029 mm and 1.460 ± 0.061 mm. Therefore, it seems that if the apophyses anteriores are longer than 0.91 mm and the apophyses posteriores longer than 1.56 mm, one can safely assume that the species is *H. resplendella*.

Life history. The life history of the species was described in detail by Emmet (1976). The larva mines the leaves of *Alnus incana* and *A. glutinosa*. Young seedlings are preferred, and on larger trees larvae are usually found on branches up to 1.5 meter high, but rarely higher up. Sometimes, especially on sandy sites with young alder, larvae may be abundant. The larva starts mining from the petiole and continues to the midrib and usually mines a 3–5 cm length of it after turning to the lateral rib. After making a short (1–3 cm) mine to the lateral rib, the larva crosses to an adjacent, more basal rib and turns backward to the midrib. The crossing mine in the blade usually turns reddish or yellowish and is clearly visible (Figs 22–24). Finally, the larva makes an oval blotch mine between the lateral ribs to the blade, situated proximally and usually on the same side of the blade as the turning mine, cuts an oval case from the blotch, and drops down to the ground. According to Emmet (1976), the larva mines the blotch for only about 24 hours. Due to this and the fact that larvae mining in the rib are hard to find, inhabited mines are seldom seen. The larva does not continue feeding on the ground. The case is attached to the ground particles, e.g. small stones or dead leaves near the place of landing, deep in the litter. In places with scarce detritus, such as sand pits or sandy sea shores, cases are rather easy to find, provided that the mines are low down and not
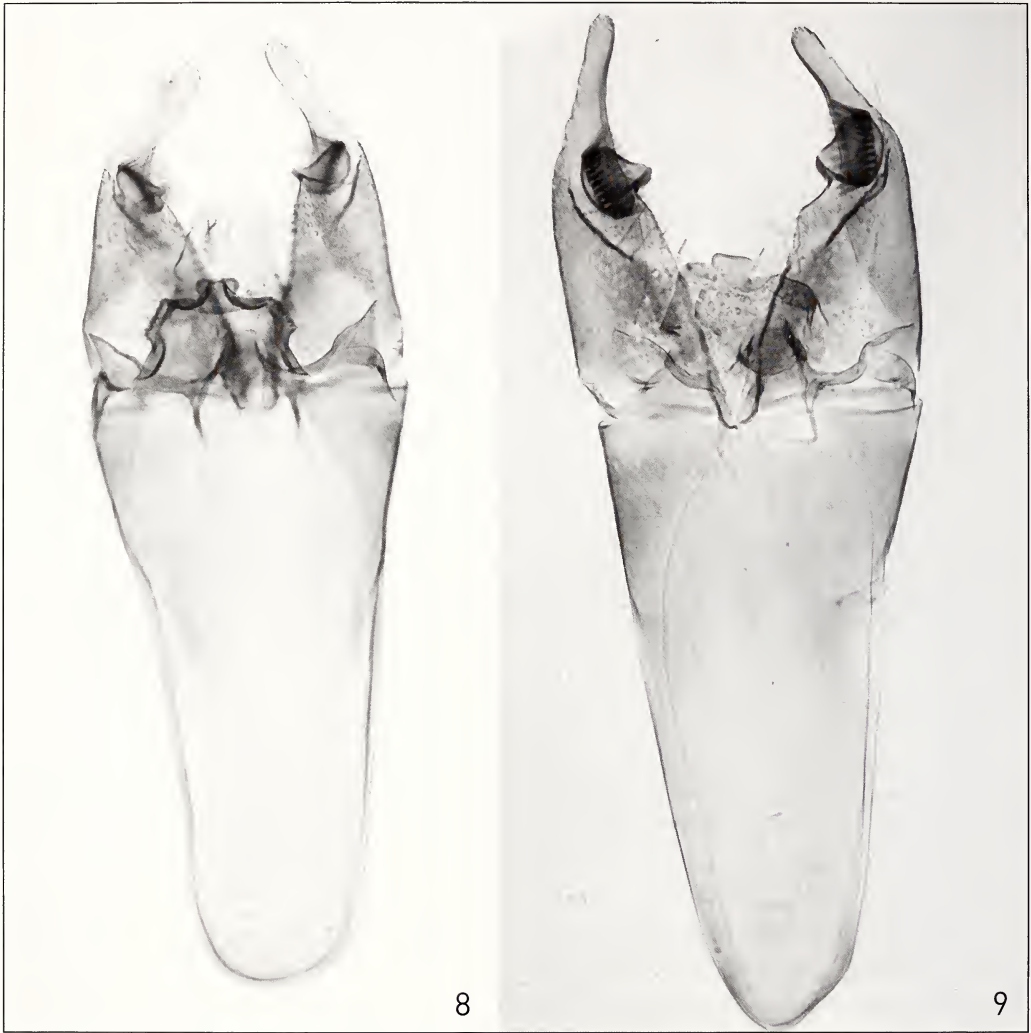
Figs 8–9. Male genitalia (phallus removed) of *Heliozela*. **8**. *H. resplendella*. **9**. *H. hammoniella*.

scarce. Compared to most soil particles, the case appears reddish. The larva pupates inside the case in the autumn (Emmet 1988). Adults are on the wing in June and July (Finland).

Distribution. The species is widespread in North Central Europe, mostly lacking in southernmost Europe, but reported from Corsica (Nieukerken 2004). We are not aware of records outside Europe. In Finland, the species is distributed up to 67°N in the north. Unlike *H. hammoniella*, it is common in southern Lapland (own observations). Distribution is most easily determined based on vacated mines.

Remarks. The original account by Stainton (1851) as well as the label data of the lectotype indicate that the primary material was not reared. The external characters and genitalia of the lectotype are identical to those of our study material of *H. resplendella* (Figs 28–29).
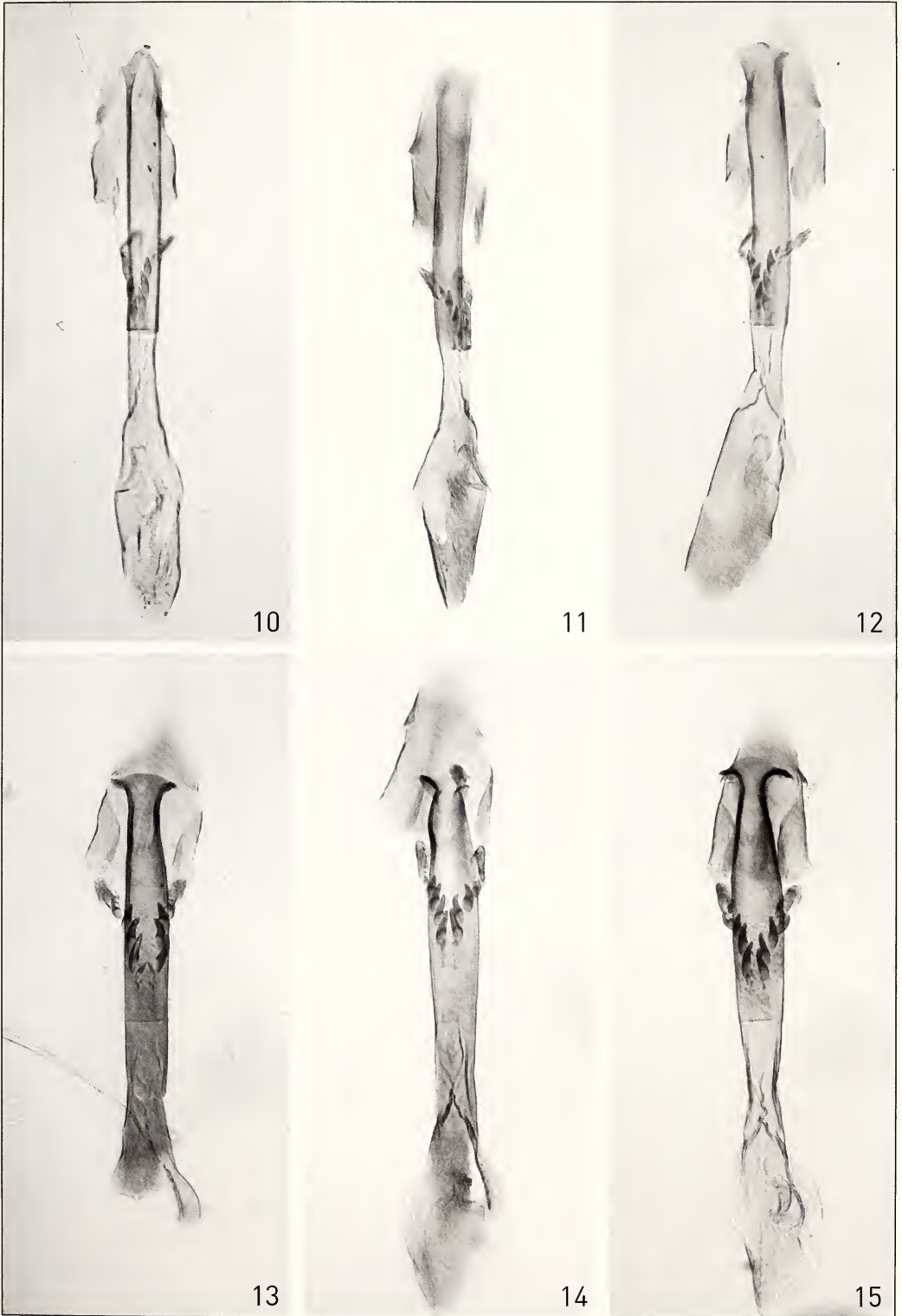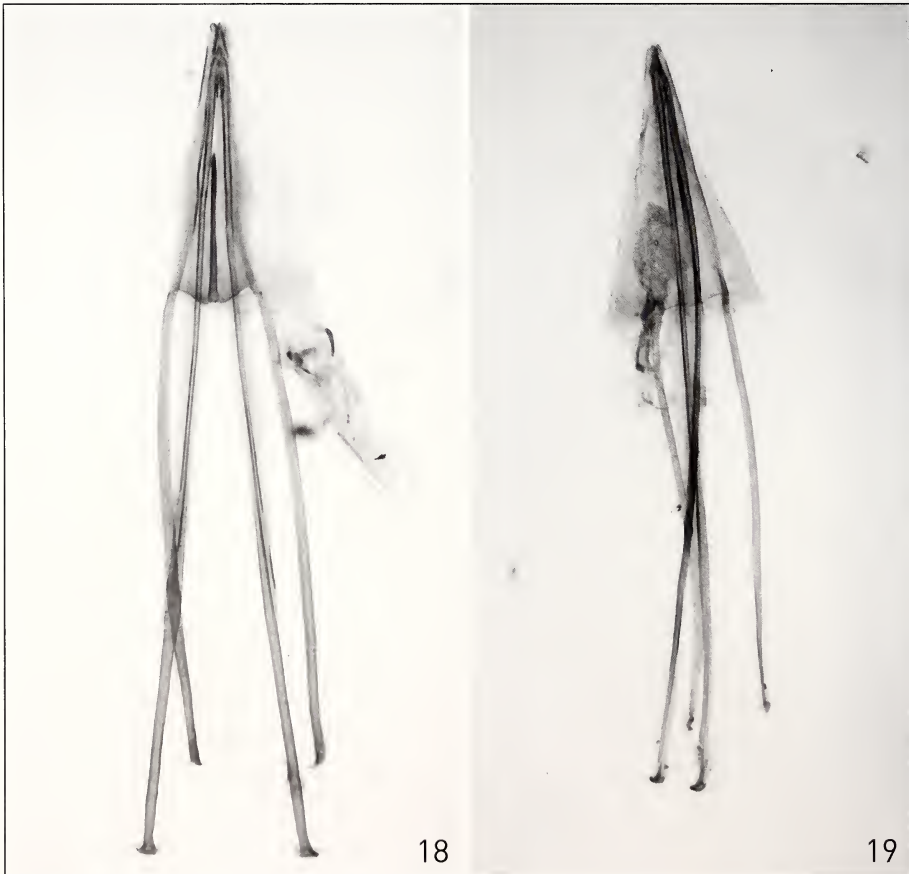
Figs 10–15. Male phallus of *Heliozela*. **10–12.** *H. resplendella*. **13–15.** *H. hammoniella*.
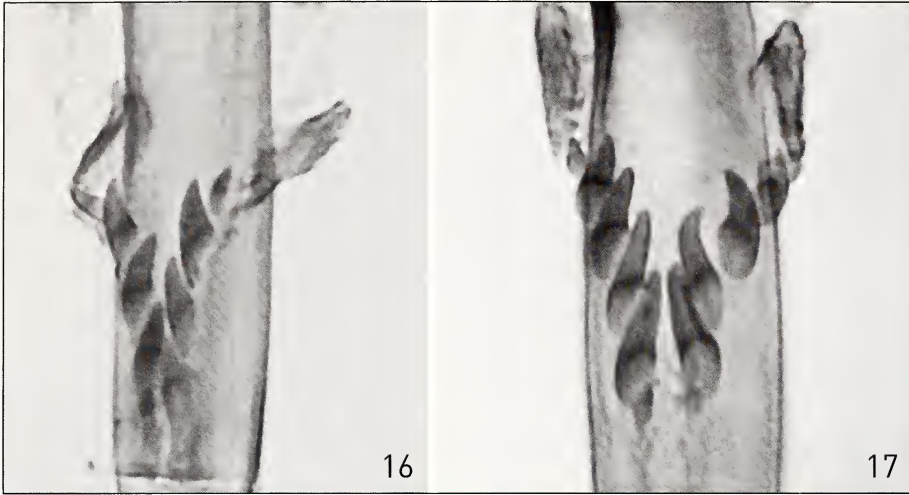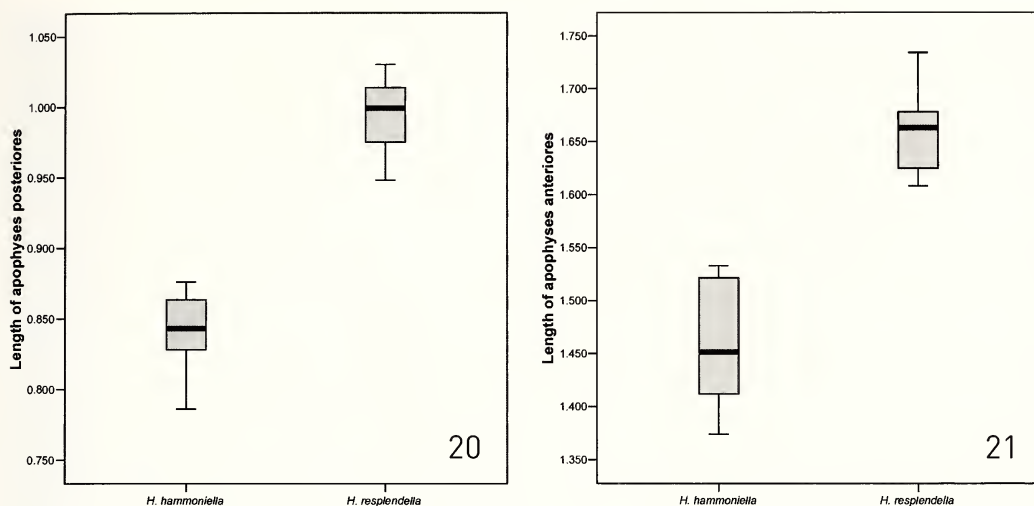
Figs 16–17. Teeth of fultura inferior of *Heliozela*. **16**. *H. resplendella*. **17**. *H. hammoniella*.

Figs 18–19. Female genitalia of *Heliozela*. **18**. *H. resplendella*. **19**. *H. hammoniella*. The scale is the same in both figures.

Figs 20–21. Length of female apophyses anteriores (Fig. 20) and apophyses posteriors (Fig. 21) of *Heliozela resplendella* and *H. hammoniella*.

Heliozela hammoniella Sorhagen, 1885   (Figs 3–5, 7, 9, 13–15, 17, 19, 20–21, 25–27)

Heliozela hammoniella Sorhagen, 1885: 338.
*Tinagma betulae* Stainton, 1890: 264.

M a t e r i a l . **Finland:** PPe: Oulu 721:42, 5♂, 1♀, larva viii.2000 (*Betula*), M. & T. Mutanen leg. (slides MM 619, 622, 623, 625, 627); PPs: Hailuoto 720:39, 2♂, 1♀, larva viii.2000 (*Betula*), M. & T. Mutanen leg. (slides MM 617, 629, 633); PPe: Hailuoto 720:39, 2♂, 1♀, larva 2001 (*Betula*), M. & T. Mutanen leg. (slides 615, 631); PPs: Hailuoto, Pöllä 720:39, 1♂, 1♀, e.p. viii.2000 (*Betula* sp.), P. Välimäki leg.; EP: Närpiö 694:21, 1♂, 30.vi.1990, J. Itämies leg.; Kn: Vaala 715:51, 1♂, 5♀, pupa 5.ix.1994 (*Betula pubescens*), J. Itämies leg. (slides MM 636, 651, 653); Kn: Vaala 715:51, 2♂, 3♀, case ix.2002 (*Betula pubescens*), J. Itämies leg. (slides MM 635, 645, 658, 659); Kn: Vaala, Itäranta 715:51, 3♂, 3♀, ex.p. 1993, J. Itämies leg.; Kn:Sotkamo 7089:603, 1♂, larva ix.1993 (*Betula pubescens*), J. Itämies leg. (slide MM 652); Kn:Vaala 715:51, 1♂, larva 3.ix.1993 (*Betula pubescens*), J. Itämies leg. (slide MM 660); Kn:Vaala 715:51, 1♂, larva 5.ix.1990 (*Betula pubescens*), J. Itämies leg.

Diagnosis. E x t e r n a l   m o r p h o l o g y . The male average forewing length is 2.52 mm (n=19), that of the female is 2.76 mm (n=13). The female is larger than the male (t=3.851, df=30, p=0.001), but of the same size as that of *H. resplendella* (see above). The male is slightly smaller than that of *H. resplendella* (see above). Otherwise similar to *H. resplendella*, but the proximal dorsal spot is usually more or less evenly rounded or triangular, apparently never strongly curved towards the wing apex (Fig. 7). The spot may be reduced, and in one specimen examined it is nearly absent.
M a l e   g e n i t a l i a . Similar to those of *H. resplendella*, but the teeth of the futura inferior are larger, and many of them are clearly bent at the tip, thus giving a hook-like appearance (Figs 13–15, 17). Otherwise see *H. resplendella* above.
F e m a l e   g e n i t a l i a . Both apophyses anteriores and posteriores are shorter than in *H. resplendella*, see above and Fig. 19.
Life history. The original description of *H. hammoniella* is actually a description of its life history. Wood (1890) also gives a similar, detailed account for the species (as *Tinagma betulae*). See also Emmet (1976). The larva lives on *Betula* spp., preferring
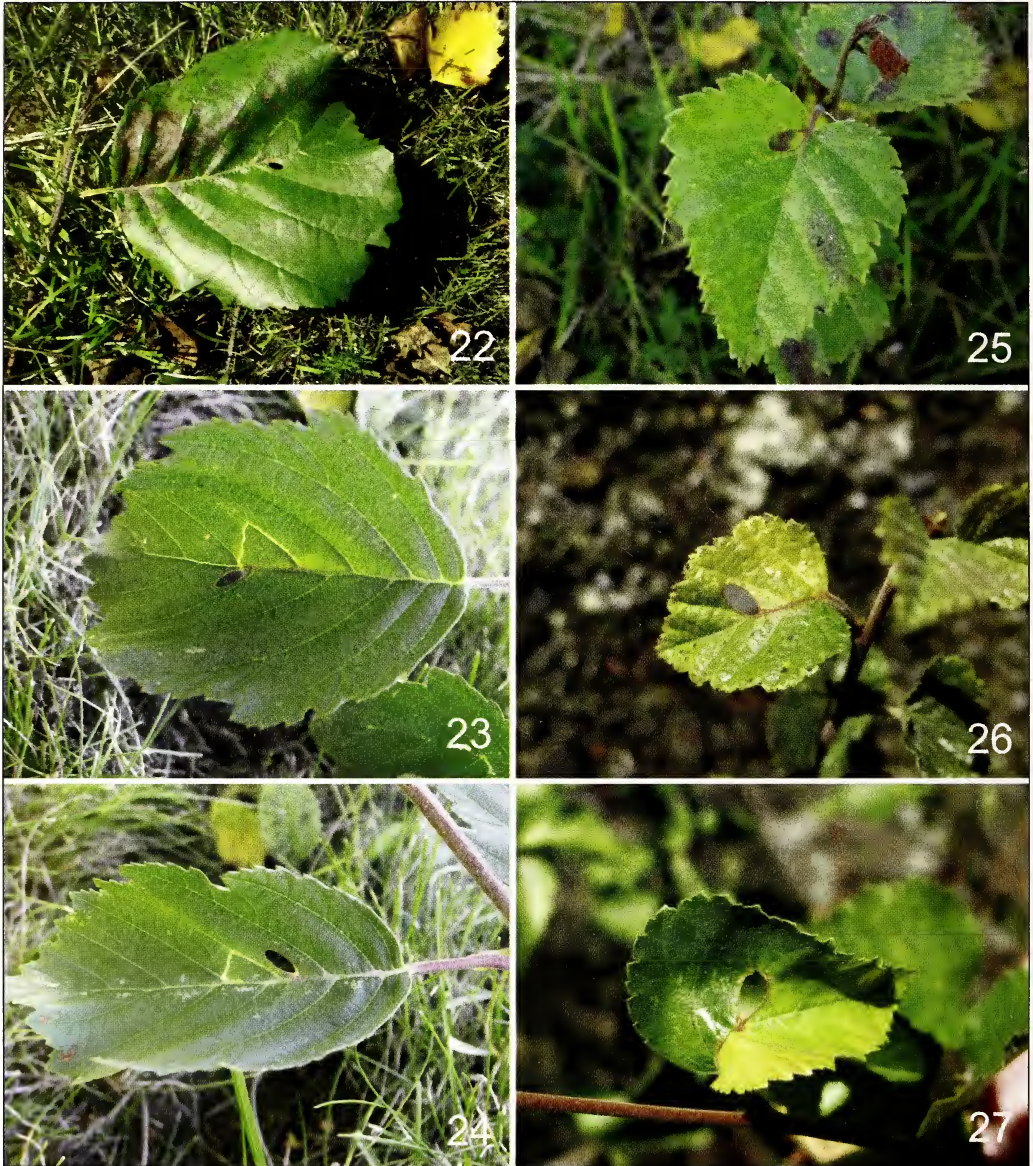
Figs 22–27. Mines and case cuttings of larvae of *Heliozela*. **22–24.** *H. resplendella.* **25–27.** *H. hammoniella.*

saplings. Larvae are usually found below the height of one meter. In Finland, larvae are usually scarcer than those of *H. resplendella.* According to Sorhagen (1886) and Emmet (1976) the larva starts mining from the pith of the twig. We have verified this to be correct. The larva then continues to the petiole and to the midrib of the leaf. Compared to *H. resplendella*, the mine is very short in the blade. Unlike *H. resplendella*, the larva does not make a turn in the blade but makes the blotch mine usually beside the midrib near the leaf base, sometimes closer to the tip (Figs 25–27). Like *H. resplendella*, the larva cuts an oval case from the blotch mine. The case is usually cut beside the midrib,

Figs 28–29. 28. Male genitalia of *H. resplendella*, lectotype. **29.** Teeth of fultura inferior of *H. resplendella*, lectotype.

but sometimes the blotch mine is elongated sidewards, in which case the cutting is made sidewards from the midrib as well. Exceptionally, the cutting may be made on the midrib. Sorhagen (1886) and Hering (1957) reported that mined leaves usually become limp and pale, but this was considered exceptional by Emmet (1976). Our experience is that mined leaves remain stunted and are often paler than adjacent leaves (cf. Figs 26–27).

Distribution. In Europe, the distribution is more or less the same as that of *H. resplendella*, but reported more scarcely, and not around the Mediterranean (Nieukerken 2004). We are not aware of records outside Europe. Due to identification difficulties, current knowledge is possibly partly unreliable. The distributions of both species should be clarified with genital dissections of adult moths or by searching for vacated mines.

In Finland, the species is widespread, but lacking in Lapland, while *H. resplendella* is common even in the southernmost parts of Lapland (own observations).

Remarks. The first description of *H. hammoniella* by Sorhagen (1885) was not mentioned by himself in the subsequent review of the microlepidoptera of the Brandenburg area (Sorhagen 1886). As noted above, the original description was only based on life history, and according to Stange (1891) and Stainton (1891) no adult specimens were reared in connection with the original discovery of the species. Only after Stange managed to rear specimens from larvae *H. hammoniella* proved to be conspecific with *H. betulae* Stainton; both Stange (1891) and Stainton (1891) published this synonymy. Sorhagen's collection was preserved in the Zoological Museum of Hamburg, and if there nevertheless was a type specimen or series, it was destroyed by bombs and fire in 1943 (Horn et al. 1990). Since the type material was reared on birch seedlings, and since the Sorhagen's (1886) description of its biology and our observations perfectly match, it most probably represents the species here referred to as *H. hammoniella*, but this naturally cannot be verified.

Acknowledgements

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