Four leaf mining flies on *Ranunculus* sp. new for Belgium (Diptera : Agromyzidae)

Jonas Mortelmans¹, Daan Dekeukeleire² & Jean-Yves Baugnée³

Abstract

Specific search for leaf-mining flies on *Ranunculus* sp. resulted in four species new to Belgium. Leaf mines, larvae and puparia of *Phytomyza ranunculivora* Hering, 1932, *Phytomyza fallaciosa* Brischke, 1880, *Phytomyza stolonigena* Hering, 1949 and *Phytomyza buhriana* Hering, 1949 were found. Phenology, distribution and ecology of these flies are discussed. For three species, rearing succeeded and observations were confirmed based on adult characteristics.

Keywords: Diptera, Leaf-mining flies, Agromyzidae, new species for Belgium, Ranunculus.

Samenvatting

Nauwgezet zoeken naar bladmineerders op *Ranunculus* sp. resulteerde in de vondst van vier nieuwe soorten voor Belgie. Mijnen, larven en puparia van de soorten *Phytomyza ranunculivora* Hering, 1932, *Phytomyza fallaciosa* Brischke, 1880, *Phytomyza stolonigena* Hering, 1949 en *Phytomyza buhriana* Hering, 1949 werden gevonden. Fenologie, verspreiding en ecologie van deze vliegen worden besproken. Voor drie van deze soorten lukte het uitkweken, en werden identificaties bevestigd aan de hand van adulte vliegen.

Résumé

Des recherches spécifiques sur les diptères mineurs de *Ranunculus* sp. ont mené à la découverte de quatre espèces nouvelles pour la faune belge. Des mines, larves et pupes de *Phytomyza ranunculivora* Hering, 1932, *Phytomyza fallaciosa* Brischke, 1880, *Phytomyza stolonigena* Hering, 1949 et *Phytomyza buhriana* Hering, 1949 ont été trouvées dans diverses localités du pays. La phénologie, la répartition et l'écologie de ces espèces sont discutées. L'identité de trois de ces espèces a été confirmée sur base des adultes obtenus en élevage.

Introduction

Agromyzidae is a large family consisting of over 2800 described species (WINKLER *et al.*, 2009). They are commonly known as leaf-mining flies due to the larvae that create typical mines in leaves. Nearly all species are dull coloured, without wing markings and are amongst the smallest flies ranging from 1mm to 6mm (OOSTERBROEK, 2006). Moreover, identification of adults is hard due to dispersed literature and genitalia that have to be dissected in many cases. Due to these challenges, they are often neglected in faunistic surveys.

Leaf-mining flies are known to have a high degree of host specificity (SPENCER, 1989; WINKLER *et al.*, 2009) and most species occur on only one or two plant genera. By identifying the well-characterized mines and larva/puparia on specific host plants, knowledge of Agromyzidae can be increased at an strong rate in a short time span. For mining Lepidoptera, this has been going on since

¹ Sint-martensblindeken 37, 9000 Gent, Belgium (e-mail: jonas.mortelmans@vliz.be)

² Polderdreef 37, De Pinte, Belgium (e-mail : daan.dekeukeleire@gmail.com)

³ Service public de Wallonie, Département de l'Etude du Milieu naturel et agricole, Avenue de la Faculté 22, B-5030 Gembloux (e-mail : jybaugnee@gmail.com)

2009 (WULLAERT, 2009), but unfortunately, Diptera were not in the scope of this project. This publication illustrates only a tip of this blank area of dipteran faunistics. The checklist by GROOTAERT *et al.* (1991) lists 84 species of Agromyzidae correcting for the synonyms established since. The online checklist by MARTINEZ (2012) lists 135 species, although several dubious records are present. This checklist should be handled with care. Recent unpublished data reveal the occurrence of at least 250 species in Belgium: 207 species have been recorded already and 43 species are expected, since they occur in all countries neighbouring to Belgium (MORTELMANS, in prep).

Recently PAKALNIŠKIS (2004) found 47 species of Agromyzidae feeding on 11 genera of Ranunculaceae, an exceptional number of which many species are not known to the Belgian fauna. On several field trips by the first author in autumn 2012, specific search effort on *Ranunculus* has been conducted. Six species of Agromyzidae were found, of which four appeared to be new to the Belgian fauna.

Results

Phytomyza stolonigena Hering, 1949

MATERIAL EXAMINED: Lavendelven, Herentals (prov. Antwerp), 17.XI.2012, empty mine on *Ranunculus* sp., leg. det. & coll. J. Mortelmans (Fig. 1); Oostende, Maria Hendrikapark (prov. Western Flanders), 6.XII.2012, larvae reared and pupated, leg. det. & coll. J. Mortelmans; Landskouter (prov. Eastern Flanders), 20.XII.2012, empty mine, leg. det. & coll. J. Mortelmans; Bourgoyen, Gent (prov. Eastern Flanders), 24.XII.2012, empty mine, leg. det. & coll. J. Mortelmans; Steenbergse bossen, Zottegem (prov. Eastern Flanders), 27.XII.2012, larvae reared and pupated, leg. det. & coll. J. Mortelmans.

Adults of *Phytomyza stolonigena* are very small, ranging only from 2 to 3 mm in wing span. Phenological data for this species are not known (pers. comm. M. Cerny). Eggs are laid on stems of Ranunculaceae, after which larvae will mine the petiole. From here the larvae makes corridors fanning out in the leaf. Typical for this species are the parallel sided mines with nearly no side branches, all leaving from the stem of the leaf (Fig. 1). Pupation takes place outside the mine (ELLIS, 2012; HERING, 1957). Larvae can be found from August-October (HERING, 1957), although our observation indicates they can actually be found until the end of November.

The species occurs in most of western Europe, ranging from Great Britain to Poland. It has recently been found in Lithuania (OSTRAUSKAS *et al.*, 2003; MARTINEZ, 2012). From the Netherlands, only one observation is known based on larval characteristics only (ELLIS, 2012). All known Belgian observations are listed above. In only one mine, the larva was present. Rearing the larvae was not successful.

Phytomyza fallaciosa Brischke, 1880

MATERIAL EXAMINED: Heiberg, Herentals (prov. Antwerp), 17.XI.2012, several mines with pupa on *Ranunculus* sp., one male reared, leg. det. & coll. J. Mortelmans; Lavendelven, Herentals (prov. Antwerp), 17.XI.2012, several mines with pupa on *Ranunculus* sp., two males reared, leg. det. & coll. J. Mortelmans; Zonnegem (prov. East Flanders), 25.XI.2012, mines on *Ranunculus acris*, leg. & det. D. Dekeukeleire; Erquelinnes, Grand-Reng (prov. Hainaut), 12.VI.2012, one mine with larvae on *Ranunculus acris*, leg. det. & coll. J.-Y. Baugnée (Fig. 2); Blaarmeersen, Gent (prov. East Flanders), 8.XII.2012, mine with pupae on *Ranunculus* sp. rearing failed, det. leg. & coll. J. Mortelmans.

In Belgium, the first mines were found respectively on 17.XI.2012, 25.XI.2012 and 8.XII.2012. Subsequently, pictures of a mine with larvae from 12.VI.2012 were identified. The wide spread of our observations indicates the species is probably common all over Belgium.

Adults of *P. fallaciosa* have wing lengths ranging between 2.1 to 2.5 mm and are active from April till November (HERING, 1957), although highest numbers can be found in spring. It presumably has two or more generations. Eggs are laid on most species of *Ranunculus*, after which the larvae will mine the leaf. Larva form linear mines, often at the apex of leaves, forming a secondary blotch which turns brown in colour soon (Fig. 2). Primary and secondary feeding lines are present with frass in long strings. Finally no islands of unmined green tissue are left over (ELLIS, 2012). Larva are active from

May-June until August-September (HERING, 1957). Pupation is internal, often removed from the mine itself (SPENCER, 1976a, b) and pupa are black in colour.

Phytomyza fallaciosa occurs nearly everywhere in Europe (MARTINEZ, 2012), and it seems to be a common species within its range. In the Netherlands and Great Britain, plenty of mines are found every year (ROBBINS, 1991; SPENCER, 1972). Its discovery in Belgium was pending a long time.

Phytomyza ranunculivora Hering, 1932

MATERIAL EXAMINED: Olmen, Asbeek (prov. Antwerp), 15.VI.2011, empty mine on *Ranunculus* sp., det. & leg. R. Barendse; Oudenaarde, Bos 't Ename (prov. East flanders), 24.IX.2011, empty mine on *Ranunculus repens*, det. & leg. P. Blondé; Opbrakel, Brakelbos (prov. East Flanders), 13.XI.2011, det. & leg. D. Volckaert & J. Mortelmans; Mouscron (prov. Hainaut), 24.V.2012, mine on *Ranunculus* sp., det. & leg. C. Gruwier; Laplaigne, Coupure de Bléharies (prov. Hainaut), 30.VI.2012, mines on *Ranunculus* sp., det. C. Gruwier (Journée des 1000 espèces); Betserbroek, Geetbets (prov. Vlaams Brabant), 22.VI.2012, mine on *Ranunculus acris*, det. & leg. P. van Sanden; Grootenhout, Giels Bos (prov. Antwerp), 18.XI.2012, mine with larvae on *Ranunculus* sp., two males reared, det., leg. & coll. J. Mortelmans (Fig. 3); Laplaigne, Coupure de Bléharies (prov. Hainaut), 1.VII.2012, mines on *Ranunculus* sp., det. C. Gruwier (Journée des 1000 espèces); Blaarmeersen (prov. East Flanders), 8.XII.2012, empty mine, det. leg. & coll. J. Mortelmans.

Adults of *Phytomyza ranunculivora* are small flies. Adults can be found from June until August (pers. comm. M. Cerny). Eggs are laid on several species of *Ranunculus*, after which a larva will make long mines in the upper-surface of the leaf. There is little frass present, and it are large, widely dispersed fragments (Fig. 3). Larvae are active from July until August. The larvae makes an exit slit at the lower side of the leaf, in which the pupation occurs. In most cases pupation occurs outside the mine, but it often remains stuck in the newly made exit slit of the mine (ELLIS, 2012; HERING, 1957; DE MEIJERE, 1938; PAKALNIŠKIS, 2004). It has a yellow/orange puparium (SPENCER, 1972).

The species has a European distribution, occurring in nearly all countries (MARTINEZ, 2012). From the Netherlands and the UK, plenty of observations are known (SPENCER, 1972; BLAND, 1994). Its discovery in Belgium was expected.

Phytomyza buhriana Hering, 1949

MATERIAL EXAMINED: Oostende, Maria Hendrikapark (prov. Western Flanders), 6.XII.2012, one mine on *Ranunculus* sp., larvae reared and pupated, one female emerged, leg. det. & coll. J. Mortelmans (Fig. 4).

Adults of *Phytomyza buhriana* are again very small flies, with wing lengths up to 1 mm. They can be found from late spring till autumn. After eggs are laid on *Ranunculus*, larvae become active from September till December, and they will create a broad mine, closing itself often. The loops are close together and create a secondary blotch with islands of green. Primary and secondary feeding lines can be seen. In initial mining, this blotch is only badly visible, and species identification should be done with care. The walls are irregular with frass in little grains, scattered through the blotch. This frass will never be in strings like *P. ranunculi* (Fig. 4) (HERING, 1957). Larvae have typical characteristics, they lack the frontal appendage and the lower rear arm of the cephalic skeleton is weakly sclerotized (DE MEIJERE, 1938). Pupation occurs outside the mine.

Phytomyza buhriana is probably the rarest of all Diptera discussed in this publication. For now, it is only known from Poland and Germany (MARTINEZ, 2012) and recently, as larvae, from the Netherlands (CUPPEN & DROST, 2007) and Luxemburg (ELLIS, 2012). Identifying mines is difficult: multiple leaves mines, identified as *P. buhriana* have shown to be *P. ranunculi* based on the reared imagos. Only one Belgian observation, based on adult characteristics is known (Fig. 4).

Discussion and prospects

For all species, leaves with larvae were reared in plastic freezer bags, and pupation occurred about 5 days after collecting the leaves. Pupa were transferred in a glass bottle and stored, until adults emerged about 1 month after pupation. These imago's have only confirmed previous identifications.



Fig 1. Typical mine of *Phytomyza stolonigena* (empty mine) on *Ranunculus repens* (photo J. Mortelmans, Lavendelven, 17.XI.2012). Fig 2. Typical mine of *Phytomyza fallaciosa* (with larvae inside) on *Ranunculus acris* (photo J.-Y. Baugnée, Erquelinnes, 12.VI.2012). Fig 3. Typical mine of *Phytomyza ranunculivora* (with larvae inside) on *Ranunculus spec*. (photo J. Mortelmans, Grootenhouten bos, 18.XI.2012). Fig 4. Typical mine of *Phytomyza buhriana* (with larvae inside) on *Ranunculus repens* (photo J. Mortelmans, Oostende, 6.XII.2012).

Three other species of *Phytomyza* are expected on *Ranunculus* in Belgium: *Phytomyza clematidis* Kaltenbach, 1859, *Phytomyza rydeni* Hering, 1934, and *Phytomyza notata* Meigen, 1830. They all share a wide European distribution, from Great Britain to eastern European countries and often, from north to southern Europe (MARTINEZ, 2012). One other rare species, *Phytomyza linguae* Lundquist, 1947 is less expected, but can possibly be found at specific locations where the host plant *Ranunculus lingua* is present in big numbers. For these four species, searching in the right season in the presence of various types of *Ranunculus*, will definitely result in new additions to the Belgian fauna.

Acknowledgements

Thanks to Willem Ellis for kindly confirming identifications of the mines and his useful site, www.bladmineerders.nl. Diederik Volckaert, Pieter Blondé, Rutger Barendse, Christophe Gruwier and Paul van Sanden for the use of their observations. Milos Cerny for his appreciated comments on the observations. Wouter Dekoninck and Patrick Grootaert for their useful comments on the manuscript.

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