# The insect fauna associated with horehound (Marrubium vulgare L.) in western Mediterranean **Europe and Morocco: potential for biological control** in Australia

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### **Summary**

Marrubium vulgare L. (Lamiaceae) was surveyed in western Mediterranean Europe and Morocco to identify the phytophagous insect fauna associated with this weed and to select species having some potential as biological control agents. Twenty-seven insect species were found to feed on the plant. Two Lepidopteran species have been introduced into Australia to control the weed. Five additional species (three Lepidoptera and two Coleoptera) are considered as potential candidates for introduction and host-specificity tests.

#### Introduction

Marrubium vulgare L. (Lamiaceae) is an invasive weed of pastures and natural habitats in southern Australian states. The genus Marrubium comprises of 48 species, all originating from Eurasia (Seybold 1978). In Europe the genus Marrubium includes twelve species (Tutin et al. 1972). M. vulgare L., an erect perennial herb, is the most common species being present in all regions but the most northern countries. M. vulgare is also common in north Africa (Parsons and Cuthbertson 1992). It is found mostly on well-drained calcareous soils and is more common on lands where grazing occurs. In this paper, I present the results of surveys for phytophagous insects that could be used as biological control agents against M. vulgare in Australia.

### Methods

Horehound populations were regularly surveyed around Montpellier, southern France. Plants were randomly collected and taken to the laboratory for measurements and dissection. Outside of France, searches were conducted in regions with suitable alkaline soils and climate, with searches mostly done along roadside populations and on lands close to roads. Surveyed countries were Spain, Portugal, Morocco in 1991-97 with minor searches made in Italy and the former Yugoslavia in 1991 (Figure 1). Plants were randomly collected and dissected in the field. Adult insects found on plants or after sweeping

were preserved. Immature stages were kept with fresh plant material until the adult stage for identification. Insects were observed either in the field or the laboratory to confirm that they fed on the plant. Insects were sent to museum specialists for identification and literature searches were conducted to assess their plant host-range. Selection of candidate biological control agents was based on their supposed degree of specificity, their known host-plants from the literature and feeding behaviour. Priority was given to insects having the strongest observed impact on plants, either through plant biomass reduction (in the case of larvae feeding on leaves) or through reduction in the plant's reproductive capacity.

### Results

### Insect abundance

Significant populations of M. vulgare were found on calcareous soils with little or no plant competition and with occasional or regular grazing by sheep. In six countries 55 M. vulgare populations were examined (Table 1), and 27 insect species

belonging to eight orders were found feeding on the plant (Figure 2, Table 2). The insects included 12 polyphagous species (44%), 8 oligophagous species (30%) and 7 monophagous species (26%). At the larval stage, there were five root-feeding species (22%), one stem-boring species (4%), nine leaf-feeding species (39%), eight flower, ovary or seed feeding species (34%). Based on adult feeding behaviour there was one root-boring species (74%), six leaf-feeding species (40%) and eight species feeding on flowers or ovaries or seeds (53%).

# Wheeleria spilodactylus (Curtis) (Lepidoptera: Pterophoridae)

Wheeleria spilodactylus was abundant at most sites in France and Spain, and had been recorded feeding on M. vulgare (Gielis 1996) and Ballota nigra (Bigot and Picard 1983). The insect's distribution includes Western and Continental Europe, British Islands, the Mediterranean region. Asia Minor and Northern Africa (Savela 1999a). Females lay eggs underneath the leaves and the green larvae feed on leaves, stems and tips of shoots, often causing abortion of buds, and pupae can be found on leaves. It was the first candidate insect to be considered for the biological control of horehound. A first consignment of larvae originating from Cap d'Agde, Herault (43°32'N, 003°51'E) was sent to Keith Turnbull Research Institute, Frankston, in 1991 for host-specificity tests, and subsequently M. vulgare was shown to be the preferred host-plant of the insect (J. Weiss unpublished data). W. spilodactylus was released at many sites in Victoria and at Wyperfeld National Park in 1994 (Weiss 1996) and later in South Australia, ACT and Tasmania (Wills 1998). To tackle

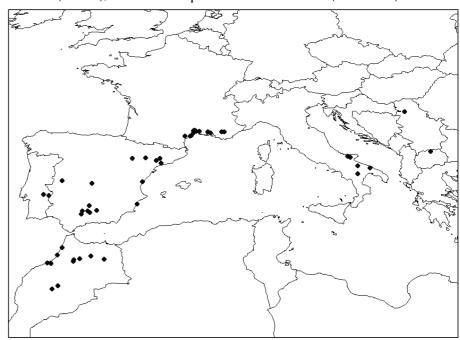


Figure 1. Marrubium vulgare L. populations surveyed in western Europe and Morocco.

climatic differences occurring within the horehound distribution in southern Australia, a second biotype of W. spilodactylus originating from regions of Spain with long dry summers, was imported into Australia in 1996.

# Carcharodus boeticus Rambur (Lepidoptera: Hesperiidae)

Carcharodus boeticus was common in France and Spain where it was found at most of the sites. According to Rungs (1979) this species is absent from Morocco. In France, adults were observed flying in April, June and September and larvae have been recorded on M. vulgare and Ballota foetida Lamk. (Lhomme 1935, Savela 1999b). Larvae are nocturnal and feed on horehound leaves, hiding during the day in a nest made with rolled leaves spun with silk. This species was considered to be a potential agent for horehound and sent to quarantine in Australia in 1994 and 1995. However the introduction was unsuccessful due to difficulties in obtaining matings in quarantine. No further attempt has been made with this species.

## Chamaesphecia mysiniformis (Boisduval) (Lepidoptera: Sesiidae)

Chamaesphecia mysiniformis was found in northern Spain, around Zaragoza (Aragon Province) where the largest horehound populations occur. According to Lastuvka and Lastuvka (1995) and Leraut (1997) the insect is present in France but was not found during this study. In Morocco its status is not clear. Rungs (1979) does not mention it, while Savela (1999c) reports its presence, but it was not found in Morocco during this study. C. mysiniformis has one generation per year, females laying eggs at base of plants in June in Spain. Larvae develop in roots of mature plants generally causing death of the plant, sometimes before the adult emergence, or during the following year. Due to the damage observed in the field, and its probable host-specificity, this species was considered to have a strong potential as a biological control agent. Biology and host-specificity of the insect were studied at CSIRO European Laboratory (Sagliocco and Coupland 1995) and approval was given by the Australian Quarantine and Inspection Service and Environment Australia to import C. mysiniformis as eggs into quarantine in 1995. However the transfer of the insect from the northern to the southern hemisphere posed several problems. A temperature protocol was established to allow re-synchronization of the insect in quarantine during larval development, and reduction of its life cycle from twelve to seven months allowed emergence of adults during the Australian summer. Initial releases were made in 1997 and further releases should occur in southern Australia in 1999.

# Meligethes spp. and Meligethes rotroui Easton (Coleoptera: Nitidulidae)

The Meligethes genus in Europe comprises about 125 species (Schenkling 1913). Easton (1955) clarified the taxonomic position of the north African species within this group. Four species have been described as associated with horehound (Easton 1952), but the small size of insects and the small taxonomic features used for identification make the sorting of species difficult and uncertain. Both adult and larval stages feed on M. vulgare flowers and pollen and destroy significant numbers of seeds.

The distribution of *M. nanus* in France extends from the Perpignan region to Provence Easton (1951), and the species is also found in Spain (Schenkling 1913). M. tropicus occurs in southern France, Spain and along the coastal areas of North Africa (Reitter 1919), while the distribution of M. villosus extends from Germany to Morocco. The species M. rotroui was described by Easton (1952) after close examination of specimens from Morocco, initially identified as M. villosus or M. tropicus. His conclusion was that M. villosus was the commonest species to occur in this country. Easton (1952) concluded from his observations, as well as from the general literature on Meligethes, that these four species were very likely to be specific to horehound. M. rotroui seems to be the best adapted to the dry and warm regions of Australia because of the climate in the regions where it occurs in Morocco. Like the three other Meligethes species mentioned above, M. rotroui overwinter as adults and in Morocco becomes active on horehound plants when flowers are first produced, in March. Adults feed on the flowers especially on pollen and anthers. After sufficient food intake they become sexually mature, mate and lay eggs on the flower clusters. Larvae hatch and develop in the calyxes, feeding on pollen and ovaries, moving from flower to flower to complete their development. Gradually, adults from the previous generation die, and fully developed larvae pupate in the soil litter at the base of plants in April. In May, adults of the new generation can be found on plants were they feed until early June, before they disperse, probably into the woodlands where they aestivate during the dry summer.

Table 1. Countries and number of Marrubium vulgare populations surveyed.

Country	Years	No. <i>M. vulgare</i> populations	No. insect species found		
France	1991-97	15	24		
Spain	1991-97	20	18		
Portugal	1991-92	2	4		
Italy	1991	5	2		
Former Yugoslavia	1991	2	7		
Morocco	1993-97	11	11		
Total	-	55	-		

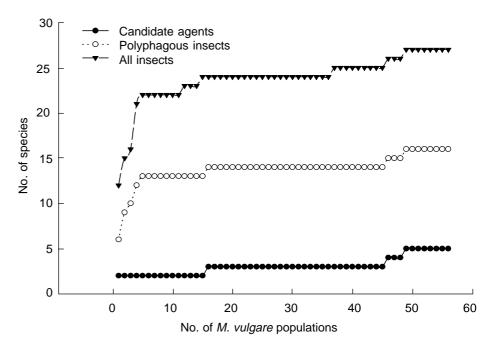


Figure 2. Increase in number of insect species found with number of Marrubium vulgare populations surveyed (France =15, Spain =20, Portugal =2, Italy =5, former Yugoslavia =2, Morocco =11).

Table 2. Insects collected feeding on M. vulgare.

Species	RF <sup>A</sup>	Collected in country <sup>B</sup> (number of sites)	Plant assoc. <sup>c</sup> (larva),[adult]	HSD	References
Coleoptera: Buprestidae Trachys goberti (des Gozis)	R	F (3), S (2)	(I-R)	О	Therond 1975
Coleoptera: Cerambycidae Phytoecia melanocephala Fabricius Phytoecia virgula (Charpentier)	LC R	M (6) F (1)	(I-R) (I-R)	M P	Villiers 1946 Villiers 1978
Coleoptera: Chrysomelidae Chrysomela banksi Fabricius Longitarsus ballotae (Marsh.)	LC LC	F (9), S (1), Y (1) F (7), S (6), I (1), M (2)	(E–L), [E–L] (E–L), [E–L]	0 0	Balachowski 1963, Therond 1976 Therond 1976
Coleoptera: Mordellidae <i>Mordellistena</i> sp.	LC	F (9), S (5), I (3)	(I-S)	P	Therond 1975
Coleoptera: Nitidulidae Meligethes nanus Erichson M. tropicus Reitter M. villosus Brisout M. rotroui Easton	R LC LC A	F (3), S (2) F (2), S (5), Y (2), M (10) F (3), S (2), Y (2), M (10) M (11)	(E-S), [E-S] (E-S), [E-S] (E-S), [E-S] (E-S), [E-S]	M M M	Easton 1952, 1955 Easton 1952, 1955 Easton 1952, 1955 Easton 1952, 1955
Coleoptera: Phalacridae Olibrus bisignatus (Ménétriès)	R	F (1), M (1)	(E-S)	P	
Diptera: Anthomyiidae <i>Delia platura</i> (Meigen)	LC	F (3), S (4), Y (1)	(E-L)	P	Hennig 1976
Diptera: Cecidomy <u>i</u> idae unidentified species	LC	F (4), M (1)	(E-S)	O*	
Diptera: Tephritidae Aciura coryli (Rossi)	R	F (6), S (1), Y (1)	(E-S)	P	White 1992
Embioptera: Embiidae Embia ramburi Rimsky-Korsakow	R	F (4), S (6)	(E-R), [E-R]	P*	
Homoptera: Aphididae  Aphis ballotae (Passerini)	LC	F (5), S (1)	(E-L, St)	M	Tuatay and Remaudière 1964
Homoptera: Cicadellidae Eupteryx melissae (Curtis)	LC	F (7), S (6), M (2)	(E-L), [E-L]	P	Alford 1994, Ribaut 1936
Heteroptera: Tingidae Tingis (Neolasiotropis) marrubii Vallot	LC	F (12), S (4), P (1), Y (2), M (4)	(E-L),[E-L]	M	Péricart 1983
Hymenoptera: Formicidae Messor barbara (L.) Aphaenogaster senilis (Mayr) Camponotus cruentatus Latreille Tapinoma nigerrimum (Nyl.)	LC R R	F (4), S (1) P (1) F (1), P (1) F (1)	[S] [S] [S]	P P P	
Lepidoptera: Hesperiidae Carcharodes boeticus Rambur	С	F (12), S (20), P (2)	(E-L)	0	Savela 1999
Lepidoptera: Pterophoridae Wheeleria spilodactylus (Curtis)	A	F (13), S (20), P (2), I (1),Y (1), M (2)	(E-L, B)	O	Bigot and Picard 1983, Rungs 1979
Lepidoptera: Sesiidae Chamaesphecia mysiniformis (Boisduval)	A	S (8)	(I-R)	O	Sagliocco and Coupland 1995, Lastuvka and Lastuvka 1995, Savela 1999
Lepidoptera: Tortricidae <i>Endothenia pauperculana</i> Staudinger	LC	F (5), S (2)	(E-B, F, S)	O	Lhomme 1935
Thysanoptera: Phlaeothripidae Haplothrips reuteri (Karny)	LC	F (14), S (3)	(E-L), [E-L]	P	Lewis 1973

<sup>&</sup>lt;sup>A</sup> Relative frequency: A = abundant (found at most sites, often in large numbers), C = common (found at many sites but not abundant), LC = locally common, R = rare.

 $<sup>^{\</sup>mathtt{B}} \ Collected \ in \ country: F = France, \ I = \ Italy, \ M = \ Morocco, \ P = \ Portugal, \ S = Spain, \ Y = former \ Yugoslavia.$ 

<sup>&</sup>lt;sup>c</sup> Plant association: B = bud, E = external feeding, F = flower, I = internal feeding, L = leaf, S = seed, S = stem, R = root.

<sup>D</sup> Host specificity: M = monophagous, only feeding on M. vulgare, O = oligophagous, feeding restricted to plants within the Lamiaceae, P = polyphagous, \* = status unknown.

Meligethes rotroui is thought to have a strong potential for the biological control of *M. vulgare* through seed number reduction. Another species of Meligethes, *M. planiusculus* has been imported for the biological control of Paterson's curse (*Echium plantagineum*) and in a field experiment caused a 65% reduction of seeds produced (Swirepik *et al.* 1996).

A first consignment of *M. rotroui* was sent to Australia in April 1997. Insects were collected in March in the Meknes region, Morocco and sent to Montpellier then to Melbourne but unfortunately all insects were dead on arrival, possibly because they were collected too early and did not have time to build fat reserves, thus losing vigour during transportation. Further collections should be carefully timed to allow shipments of insects in good condition before their peak in egglaying.

# Phytoecia melanocephala Fabricius (Coleoptera: Cerambycidae)

The genus Phytoecia includes many species throughout the Palaeartic region but only a few species in western Africa. Villiers (1946) records the presence of P. melanocephala in northern Africa (Morocco, Algeria, Tunisia) and Sicily. Adults are diurnal and larvae develop in stems and root. The discovery of larvae of P. melanocephala developing in M. vulgare roots was the first record of this insect's larval host-plant. In Morocco, P. melanocephala was the only root-borer found in horehound roots. Plants collected on roadsides at El Haj-Kaddour in the Meknes region in March 1994, had 41% attack by P. melanocephala (Sagliocco unpublished data). P. melanocephala was also recorded at Bir-Tam-Tam (Taza), Meknes, Mohammedia (Casablanca) and Azilal (Beni-Mellal). The most serious damage was observed at Meknes and at Mohammedia, where a horehound population identified in 1994 had few surviving plants in 1996 and dissected plants showed evidence of P. melanocephala damage.

Adults emerge in May-June and can be found on horehound stems where they feed and mate. Adult life-span seems to be short, not exceeding two or three weeks and adults have also been observed feeding on horehound stems in cages, and on thistles capitula in the wild (Villiers 1946). The larvae develop within the root, eventually causing death of the plant and the life-cycle is possibly completed in one year. Field surveys suggest that P. melanocephala is restricted to horehound. Its distribution shows a likely adaptation to high summer temperatures, which rise to over 40°C in the Meknes region. A second collection of insects was made in June 1996 at Aïn-Toto (18 km east of Meknes), with 290 adults collected by sweeping while resting or mating on horehound plants, and taken

back to the CSIRO quarantine facilities at Baillarguet for rearing and observations. In spite of precautions taken during transportation, all insects died within eight days, suggesting a very short adult lifespan, a feature which seems to be a common within this genus (C. Cocquempot personal communication). However, technical difficulties related to the biology of the insect, such as egg-laying (females lay eggs inside the lower part of stems) and the short adult longevity pose problems for collections in northern Africa and shipment to quarantines laboratories for further studies, either being in Australia or in France.

### Other insects

Endothenia pauperculana Staudinger (Lepidoptera: Tortricidae) (=Enarmonia nougatana Chrétien)

In France this species is restricted to the southern region and has several generations between spring and autumn. Larvae develop on buds, flowers and seeds of horehound and *Sideritis hirsuta* (Lamiaceae) (Lhomme 1935). This insect was not found in large numbers, however because of its host-plant range this species may deserve further consideration.

# Aphis ballotae (Passerini) (Homoptera:Aphididae)

Aphis ballotae was locally common in France, occurring in small colonies on some individual plants. According to Tuatay and Remaudière (1964) A. ballotae is common in horehound in central Europe and in northern Iran, though Stroyan (1984) mentions uncertainties about status of the species and host-plants. Without field observations in other regions of Europe where the insect may have a stronger impact and a clearer status of this taxon, it is difficult to consider A. ballotae as a potential biological control agent for horehound.

# Longitarsus ballotae (Marsham) (Coleoptera: Chrysomelidae)

Longitarsus ballotae was locally common in France and Spain. Adults feed on leaves making shot-holes and larvae feed on roots of horehound seedlings and young plants. Insect's impact appears to be minimal

# Tingis marrubi Vallot (Heteroptera: Tingidae)

Tingis marrubi is probably specific to *M. vulgare* (Péricart 1983), but this insect was never found to be abundant on horehound. Adults and larvae feed on leaves cells and cause local depigmentation.

# Unidentified Cecidomyiidae

This species was never found to be abundant. Larvae develop in the calyx on the ovary and provoke its deformation and

seed's abortion. No Cecidomyiid has been recorded so far on horehound from France (Dauphin and Aniotsbehere 1993) and the insect could not be identified, suggesting that this species is still undescribed.

### Lepidoptera: Sesiidae

Two other insects are mentioned in the literature, but were not found during this survey. The Sesiid, *Chamaesphecia maurusia* Püngeler is reported on *Marrubium* spp. in Algeria, Morocco and Spain (Savela 1999c). The Sesiid, *C. oxybeliformis* (Herrich-Schäffer), is reported from *M. vulgare* and *M. peregrinum* (Lastuvka and Lastuvka 1995, Savela 1999c) from Macedonia, Greece, the Balkan region and Asia Minor. These insects may have some potential for the biological control of *M. vulgare*, especially if their fecundity is higher than *C. mysiniformis*.

### Discussion

This study on the horehound insect fauna has shown that all parts of the plant are attacked by specialist insects and that some possibilities of biological control exist with two insect orders, Lepidoptera and Coleoptera. Two lepidopteran species have already been introduced into Australia. One species (W. spilodactylus) is well established, has been widely distributed and has an impact on the horehound plants. The second lepidopteran (C. mysiniformis) is not yet established, but recent results are encouraging. Concerning coleopteran insects, some potential exist with the pollen and seed-feeder M. rotroui, which may help to significantly reduce seed set and recruitment, making this insect the next candidate agent to import into Australia. The root-borer, P. melanocephala will have a similar effect as C. mysiniformis but collection and hostspecificity tests pose technical problems. Another lepidopteran species, E. pauperculana, may have some potential and could complement seed control offered by M. rotroui. Two other sesiid insects are known from the literature from regions in Europe climatically comparable to Australia. They may be specific to horehound and could be considered as potential agents to be imported if further searches in northern Africa and the Aegean region are considered necessary.

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Toulouse) Hymenoptera, Formicidae, M. Shaffer (NHM, London) Pterophoridae, J.D. Holloway (IIE) Hesperiidae, I.M. White (IIE, London) Tephritidae, Prof. J. Menier (MNHM, Paris), Drs. M. Martinez and C. Cocquempot (INRA, Montpellier) and R.G. Booth (IIE) Cerambycidae, Embiidae, Mordellidae, Nitidulidae, Tortricidae, G.J. Du Heaume (IIE) Phlaeothripidae, G.M. Stonedahl (IIE) Tingidae. Drs. J.K. Scott and D. Briese are acknowledged for their review and comments on previous version of this paper. I thank C. Espiau for producing the horehound sites map.

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