

## Progress on the rearing, release and establishment of the horehound plume moth, *Wheeleria spilodactylus* (Curtis), for the biological control of horehound in Tasmania

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### Summary

In Tasmania, horehound is regarded as a significant problem in the drier sheep grazed pastures in the midlands, east and south-east of the state. A biological control agent, a French biotype of the horehound plume moth, *Wheeleria spilodactylus* (Curtis), was introduced into Tasmania in October 1997. Mass rearing of the agent was carried out on caged horehound plants contained in pots in a controlled temperature glasshouse at about 23°C. Releases were made at five sites during the spring and summer of 1997/98 and at an additional 20 sites during the same period in 1998/99. By February 1999, 2370 pupae, 1380 adults, and 81 300 larvae had been released. Most of the releases consisted of middle to late instar larvae by direct transfer of infested foliage from the culture to the release sites. The foliage was deposited at the field sites to enable the larvae to transfer to the established plants. The majority of releases involved approximately 1500 larvae per site. Assessments at the five earliest release sites during March 1999 showed that *W. spilodactylus* was surviving well at all sites and had spread up to a maximum distance of 800 m. These preliminary results indicate that the agent will establish well in Tasmania. Further releases are planned, together with a study to determine the efficacy of the agent and its use in integrated control programmes.

### Introduction

Horehound, *Marrubium vulgare* L., is a Declared Secondary Weed in Tasmania. Infestations are commonly found in the agricultural areas of the State on roadsides, waste areas, stockyards and dry banks near farm buildings. It is a significant problem in pastures grazed by sheep in the midlands, east and south-east where the mean annual rainfall is 500–600 mm. Infestations on individual properties in these areas range in size from scattered patches of 1–5 ha to much larger areas of ca. 15 ha or more. No detailed study on losses attributable to horehound has been carried out in Tasmania. However, many Tasmanian wool growers regard it as a significant problem due to the costs

involved in applying management strategies to avoid fleece contamination by horehound burrs. In addition, as horehound is of low palatability, the productivity of pastures carrying heavy infestations is considerably reduced.

Interest in a biological control program for horehound in Tasmania was initiated after the successful establishment of a French biotype of the horehound plume moth, *Wheeleria spilodactylus* (Curtis), in southern mainland states following releases during the summer of 1993/94. The larvae of *W. spilodactylus* feed on the shoot tips and leaves and reduce plant vigour. A mass rearing and release programme commenced at New Town Research Laboratories near Hobart in September 1997, with the importation of 400 pupae of *W. spilodactylus* from Keith Turnbull Research Institute, Victoria. The stock used was a biotype collected from Cape d'Agde in southern France in 1996 (Clarke *et al.* 1999). This paper outlines the mass rearing and release strategies used to permanently establish *W. spilodactylus* in Tasmania and presents the results of establishment and dispersal assessments at field release sites to the end of February 1999. The use of *W. spilodactylus* in the integrated management of horehound infestations in Tasmania is discussed.

### Materials and methods

#### Propagation of horehound

Horehound seedlings were collected from field sites and placed singly in 15 cm diameter pots containing a peat, pine-bark and sand mix together with a slow release fertilizer. About 500 plants were maintained in a temperature range of 20–25°C using a controlled temperature glasshouse. The plants were watered daily (or as required) and fumigated fortnightly with Dichlorvos (Insectigas-D DDVP Insecticide®) to reduce attack from pests, particularly greenhouse white fly, *Trialeurodes vaporariorum* (Westwood).

#### Mass rearing

Rearing was carried out in cages in a separate controlled temperature glasshouse at about 23°C with a 16 hour photoperiod. These conditions enabled *W. spilodactylus*

to pass through a complete generation in about eight weeks. Cages used were 1 m high, 750 cm wide and 500 cm in depth consisting of insect proof nylon mesh set in aluminium framing with an aluminium base. A gauze sleeve, framed into the front of the cage, enabled access without the need for the cages to be fully opened. Sticky traps were hung in each cage to reduce pest activity, mainly *T. vaporariorum*, and were removed when the emergence of *W. spilodactylus* was imminent. Eight leafy horehound plants were placed in each cage in plastic saucers (14 cm diameter) and hand watered daily. A maximum of 20 cages were utilized over one generation. To infest these cages, 60 newly emerged adults were collected daily usually over 1–3 days as they emerged from pupae produced from the previous culture. Rearing for field release was carried out in spring and summer. Flowering plants were provided, if available, to nourish the adults and increase their fecundity and longevity. Larval counts showed that about 1500 larvae could be produced from each cage. During autumn and winter, a maintenance culture of 2–4 cages was held in the glasshouse at ambient conditions and the larvae allowed to enter diapause until spring.

#### Field release

The majority of releases were carried out in spring and summer, with some later autumn releases if material was still available from the cultures. The size of the horehound infestations selected for release was typical of most Tasmanian sites and ranged from small infestations of 1 ha to the larger infestations of 15 ha, although most infestations were in the order of 3–5 ha.

Most field releases were carried out by direct transfer to field sites of foliage infested mainly with mid to late instar larvae, although in some cases it was noted that some larvae had already reached the pupal stage. Foliage was removed from the caged pots by cropping the stems at the base. For transport, it was placed in large insulated plastic containers with close fitting lids and kept cool with freezer blocks. At the release site, the infested foliage was scattered over several square metres amongst well established horehound plants, to enable the larvae to transfer onto these. The mean number of larvae in each release was estimated by removing 2–3 pots at random from selected rearing cages, and counting the number of larvae on each individual plant. At most sites, larval numbers released ranged from 1000 to 3000 per site, although the minimum number of larvae released was 500 (2 sites) and ranged to a maximum of 30 000 (1 site) (Table 1). Pupae and adults were occasionally collected from the cages with the larvae and these were also released

(Table 1) if not required to produce the next glasshouse generation.

Because of the possibility of sheep grazing reducing agent establishment, all releases of *W. spilodactylus* were made within fenced areas of 20 × 20 m.

#### Assessments for establishment and dispersal

Establishment assessments for *W. spilodactylus* were carried out during February and March 1999, at all sites where releases had been carried out 12 months previously. Horehound plants were examined around the release point for the presence of eggs, larvae, pupae or adults. If *W. spilodactylus* was present at the release site, dispersal was assessed by examining horehound plants every 50–100 m from the release point. Straight line transects were followed along the four main compass points if the infestation was continuous. However, if the density of plants became sporadic as the distance from the release point increased, examinations were carried out at the nearest patch of horehound. A site was classified as established if *W. spilodactylus* had survived at the site for at least a year and was reproducing and spreading.

#### Results

##### Rearing and release

By February 1999, the original stock of 400 pupae imported from Victoria in October 1997 had been bred through seven generations and produced 2370 pupae, 1380 adults, and 81 300 larvae for field release. During this time, releases were carried out at 25 sites (Table 1, Figure 1). Of these, 24 were on sheep grazed properties in the drier midland and south-eastern areas where most of the major horehound infestations are located (Figure 1). One release was carried out in a localized horehound infestation of 1–2 ha, in a dairy pasture at Montana in northern Tasmania, where the mean annual rainfall is about 900 mm (Figure 1).

##### Establishment and dispersal

*Wheeleria spilodactylus* established at all of the five sites assessed and, during the 12 months since its release, had dispersed to a maximum distance of 800 metres at Sorell and Avoca (Table 1). It was recorded throughout smaller infestations of about 1 ha at Richmond 1 and Maria Point (Table 1). At Richmond 1, establishment was achieved with the smallest larval release (1500 per site). At Sorell, it was evident that sheep grazing on horehound could be an important factor in preventing the establishment of *W. spilodactylus*. The agent was found only within the fenced release site, and had moved along fence lines and into nearby infestations of horehound not grazed by sheep.

**Table 1 Release, establishment and dispersal details for horehound plume moth, *Wheeleria spilodactylus* (Curtis), in Tasmania (October 1997–February 1999).**

Site no.	Site location	Release period	Number agents released			Establishment category <sup>A</sup>	Max dist/area spread
			Larvae	Pupae	Adults		
1	Sorell	Oct/Nov-97	2926	1267	243	A	800 m
2	Ouse	Dec-97	11 000	0	0	A	150 m
3	Richmond 1	Dec-97	1570	0	0	A	1 ha
4	Avoca	Jan/Feb-98	30 000	0	290	A	800 m
5	Maria Point	Feb/Mar-98	4142	0	58	A	1 ha
6	Colebrook 1	May-98	2400	0	0	B	
		Sep-98	3178	1272	791		
		Nov-98	1500	0	0		
		Feb-99	1500	0	0		
7	Colebrook 2	Sep-98	1563	0	0	B	
8	Colebrook 3	Sep-98	2415	107	0	B	
9	Colebrook 4	Sep-98	1280	0	0	B	
10	Bothwell 1	Sep-98	1480	0	0	B	
11	Campania 1	Oct-98	500	0	0	B	
12	Campania 2	Oct-98	1000	0	0	B	
13	Tea Tree	Oct-98	1500	0	0	B	
14	Richmond 2	Oct-98	530	0	0	B	
15	Buckland	Oct-98	1000	0	0	B	
16	Jericho	Oct-98	1000	0	0	B	
17	Campania 3	Nov-98	1000	0	0	B	
18	Gretna	Nov-98	1500	0	0	B	
19	Dunalley	Nov-98	1500	0	0	B	
20	Kempton	Jan-99	1500	0	0	B	
21	Bothwell 2	Jan-99	1500	0	0	B	
22	Bothwell 3	Jan-99	1500	0	0	B	
23	Montana	Jan-99	3000	0	0	B	
24	Colebrook 5	Jan-99	1500	0	0	B	
25	Woodbury	Jan-99	2500	0	0	B	

<sup>A</sup> Establishment category ratings.

A = Definitely established - population has survived at site for 1 year and is reproducing and spreading.

B = No check made as yet.

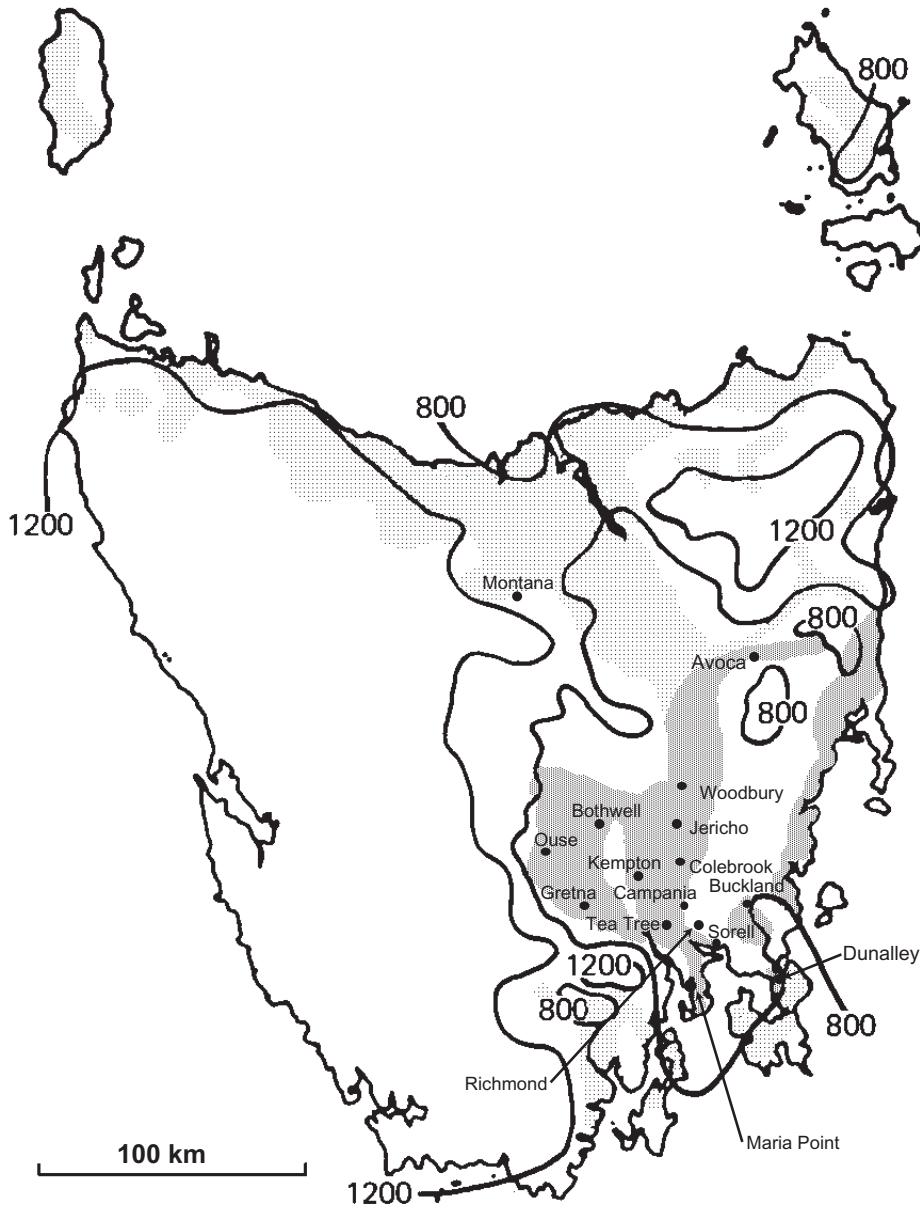
#### Discussion

The successful establishment of *W. spilodactylus* at all five of the earliest releases sites indicates that the agent will be easy to establish at other Tasmanian sites, and that this can be achieved using releases of 1500 larvae per site or less. The dispersal rate of 800 m in 12 months is also indicative that the agent has the potential to spread rapidly. However, it is possible that the dispersal of *W. spilodactylus* could be restricted by the lack of continuity of many Tasmanian horehound infestations. Agent dispersal can be accelerated by further releases from glasshouse cultures and eventually, as population densities start to increase at release sites, through field redistribution programs involving community groups.

If *W. spilodactylus* becomes widely established in Tasmania, and population densities increase to levels high enough to have an impact on horehound, further investigations will be required to determine the best methods of integrating this biological control agent with other control strategies, such as sheep grazing and herbicides.

In Tasmania, sheep often graze on the young, lush growing tips of horehound which are the refuge of young, overwintering larvae of *W. spilodactylus*. The extent of sheep grazing, particularly during winter and in early spring when the young larvae become active, could be an important factor affecting site establishment and population density of *W. spilodactylus*. Information will also be required by landholders on the impact of herbicides currently registered for horehound control. Studies by Ainsworth (1999) have already demonstrated the potential of using a low herbicide rate (2,4-D) in combination with feeding by *W. spilodactylus* to produce acceptably low horehound densities. The integrated control studies would ultimately provide landholders with a comprehensive, cost effective integrated control package for horehound.

*Wheeleria spilodactylus* will be the only biological control agent used against horehound in Tasmania, at least in the short term. A second biological control agent, *Chamaesphacia mysiniiformis* (Boisduval), is not being considered for release in Tasmania as the species requires summer



**Figure 1.** Location of release sites for the horehound plume moth, *Wheeleria spilodactylus* (Curtis), in Tasmania. Sites are shown in relation to the 800 mm and 1200 mm annual isohyets. The shading (light and dark) represents the approximate area (900 000 ha) of sown pastures. The darker shading is the area of sown pasture where the major horehound infestations are located.

temperatures regularly exceeding 30°C in order to complete its development (Weiss personal communication). No further introductions of additional agents are planned at present due to financial constraints (Clarke *et al.* 2000).

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#### References

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