

**FOREIGN EXPLORATION & HOST SPECIFICITY
TESTING OF
BIOLOGICAL CONTROL AGENTS OF
FRENCH BROOM IN CALIFORNIA
*PHASE 2***



***INTERNATIONAL BROOM INITIATIVE
PROJECT
2002-2003***

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Foreign Exploration & Host Specificity Testing of
Biological Control Agents of
French Broom in California

Phase II

Final Report

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Executive summary

This is the final report on phase 2 work carried out for the biological control of French broom in California by Commonwealth Scientific and Industrial Research Organisation at its European Laboratory in France in collaboration with the USDA European Biological Control Laboratory. Survey trips associated with the first objective have been completed to southern Spain and western Portugal and southern France to finish pod collections as well as faunal surveys. The planned trip to Morocco has had to be delayed due to unavailability of collecting permits. Analysis of the results of all trips to-date is presented. Eighty five species of phytophagous arthropod have now been found on French broom. Two potential agents, the stem-mining beetle (*Agrilus antiquus*) and fly (*Chyliza leptogaster*) appear to be capable of killing the plant prior to maturity, while the highly specific apionid weevil (*Lepidapion ?argentatum*) can destroy up to 63% of the seed production in the native range. The specialist psyllid, *Arytinnis hakani*, also appears to be highly damaging. The natural enemy fauna was most diverse and the levels of predispersal seed losses to specialist seed feeders were highest in the Western Mediterranean in particular southern Spain, but there is high variation between sites, probably due to habitat, weed infestation size and age. A regional analysis is presented of the diversity of the natural enemy community and levels of seed loss.

In completing the second objective 47 plant species relevant to the USA have now been tested against the first potential agent, the psyllid (*A. hakani*) including 17 native lupines. This is more than double the agreed milestones in phases 1 & 2 of this project. The psyllid laid eggs on 10 test plants species from which it has not been recorded in the field including 6 native lupines. Survival to adult was also accomplished on one of the American native lupines, *Lupinus longifolius*, and one common European species, Spanish broom (*Spartium junceum*) not recorded as a host in the native range. The resulting adults failed to survive to lay eggs. This has led to the recommendation to broaden the range of testing of the psyllid against native lupine species to include nymphal survival tests to better evaluate the risks to these native species in the USA. Post hoc host range studies of the bruchid beetle *Bruchidius villosus* currently being released across the western USA have found that this beetle can lay eggs and develop in the seeds of the native tree lupine (*Lupinus arboreus*). The results of this work were presented at the IXth International Symposium on Biological Control of Weeds. The resulting poster is included in this report. Progress has been made in developing host specificity testing protocols for the seed feeding apionid (*L. argentatum*) and the stem mining fly (*C. leptogaster*). Phase 2 of the project required accommodation of a 17% budget shortfall due to poor exchange rate performance of the dollar against the euro during the life of this project.

Background

Research started at CSIRO's European Laboratory on the biological control of French broom (*Genista monspessulana* (L.) L. Johnson) for California in 2000 through funding available from the California Department of Forestry and Fire Protection (CDF). These resources were added to an existing research effort underway for Australia funded by the Australian Cooperative Research Centre for Weed Management Systems. The results of this preparatory phase of the project were published under an Oregon Department of Agriculture project 801 GR (http://groups.ucanr.org/ceppc/Selection_and_Testing/). Research on this project was then funded by California to continue Phase 1 of the research through CDF, California Department of Transportation (CALTRANS) and California Department of Food and Agriculture (CDFA) via a cooperative agreement with United States Department of Agriculture-Agricultural Research Service for a funding period from February 1, 2001 until May 30, 2002 (http://groups.ucanr.org/ceppc/Foreign_Exploration/).

A budget was drafted for Phase 2 of this project in an informal agreement between CDF, CSIRO and USDA-ARS in March 2002 for a project due to operate from July 1, 2002, until June 30, 2003 under a similar cooperative agreement with USDA-ARS. The reporting schedule was for a progress report December 31, 2002 and a final report on June 30, 2003. Significant delays in the availability of funds prevented the signing of the formal cooperative agreement with USDA-ARS for the disbursement of funds until March 12, 2003, by which time the US\$ had lost 20% of its value against the € compared to the time the budget had been drafted. The slight recovery since then means that the budget was effectively c.17% less than originally expected over the whole project period. This budget deficit had to be accommodated within the project. The late signing of the contract also prevented a progress report being submitted as planned on December 31, 2002.

This report covers all research carried out for the whole funding period until June 30, 2003 against agreed objectives. This final report is an extension of the final report for Phase 1.

This project is part of the International Broom Initiative that is seeking long-term funding for broom biological control research in the US

http://www.caleppc.org/Gorse_and_Broom_Information/

CSIRO staff acknowledges assistance provided in the administration of this project by Mike Pitcairn at CDF and Chuck Quimby (EBCL USDA-ARS). CDF staff have also provided direct assistance through the development of a formal host specificity test list of French broom potential biological control agents and by supplying seeds of test plant species.

Objective 1: Carryout quantitative field surveys for potential biological control agents for French broom in Portugal, southern Spain, and Morocco. Produce an updated and justified priority list of potential biological control agents for French broom..

SURVEY TRIPS.

Introduction

Quantitative survey trips have now been carried out throughout most of the native distribution of *G. monspessulana*, around the Mediterranean on the north coast from Greece to Portugal and to Tunisia on the south coast and the Canary Islands. Areas intensively searched were typical native habitat, i.e. rainfall 600+ mm per annum, less than 1000 m altitude on acid soils that support oak or pine overtopped *maqui* vegetation. The surveyed areas support many co-occurring species in the Genisteae, so sites were selected to include several species in the tribe, where possible, and where not, samples were taken in large mono-specific stands of the common species present. Particular effort was made to find sites where *G. monspessulana* co-occurred with *C. scoparius* or *U. europaeus* for comparison, as focused survey trips have been made for these species in the past and the natural enemy community found on them is relatively well understood (Zwölfer 1963; Syrett *et al.* 1999).

Two trips were made to each site. On the first ‘mid-flowering’ visit (between March and May) sampling consisted of three sharp taps (with a shortened broom handle) to 10 plants per Genisteae species per site (where possible) with a 1.5 m x 1.5 m beating sheet held under each plant. All arthropods were collected with an aspirator except for very numerous species where a sub-sample was collected from a random section of the beating sheet and the numbers of individuals calibrated up for the whole sheet. Immature stages of herbivorous species where adults clearly were not present (e.g., Lepidoptera larvae) were placed in separate rearing boxes with the food plant. Attempts were made to rear out adults for identification. Plants were also searched visually to collect any obvious endophagous species not sampled by beating, including leaf miners, gall formers, stem and root borers and obvious pathogenic fungi. Such species were recorded as present or absent. Samples from individual plants by host species by site were kept separately. Herbarium samples were taken to confirm plant identifications.

All arthropods were sorted, counted and identified as far as possible (to family or genus) in the laboratory in Montpellier on return and voucher specimens were sent for identification from all species clearly on *G. monspessulana* alone and all species in the following orders/families; Lepidoptera, Diptera, Curculionidae, Apionidae, Chrysomelidae, Bruchidae, Aphididae, Cicadellidae, Psyllidae and Miridae.

Another visit was made to each site just before seed pod maturation (in June to July) either in the previous, same or subsequent year and all the pods from 10 randomly selected plants per species were collected and dry stored separately per plant in ventilated plastic boxes. If arthropod species exited the green pods as larvae to pupate in the soil prior to collection then they were noticed from their emergence holes in the pods. Those larvae that did emerge from the pods soon after collection were placed in rearing dishes of moist vermiculite until adult emergence. After a minimum of 3 months storage the samples were sorted for emerged adult phytophagous arthropod species from the whole sample and then 30 pods per plant were dissected to quantitatively assess

the attack rate and impact of the different arthropod species to total plant seed production by relating damage characteristics to phytophagous species.

The quantitative natural enemy species data from the beating trays were used in a regional analysis to determine a) the average number of individuals of each recorded species per *G. monspessulana* plant sampled and b) the percent frequency with which *G. monspessulana* plants were found with particular herbivore species on them. The pod dissection data were used to calculate the percentage seed loss per plant for each given insect seed predator identified a) for the seven most common Genisteae species sampled across all sites and b) between regions where *G. monspessulana* was sampled.

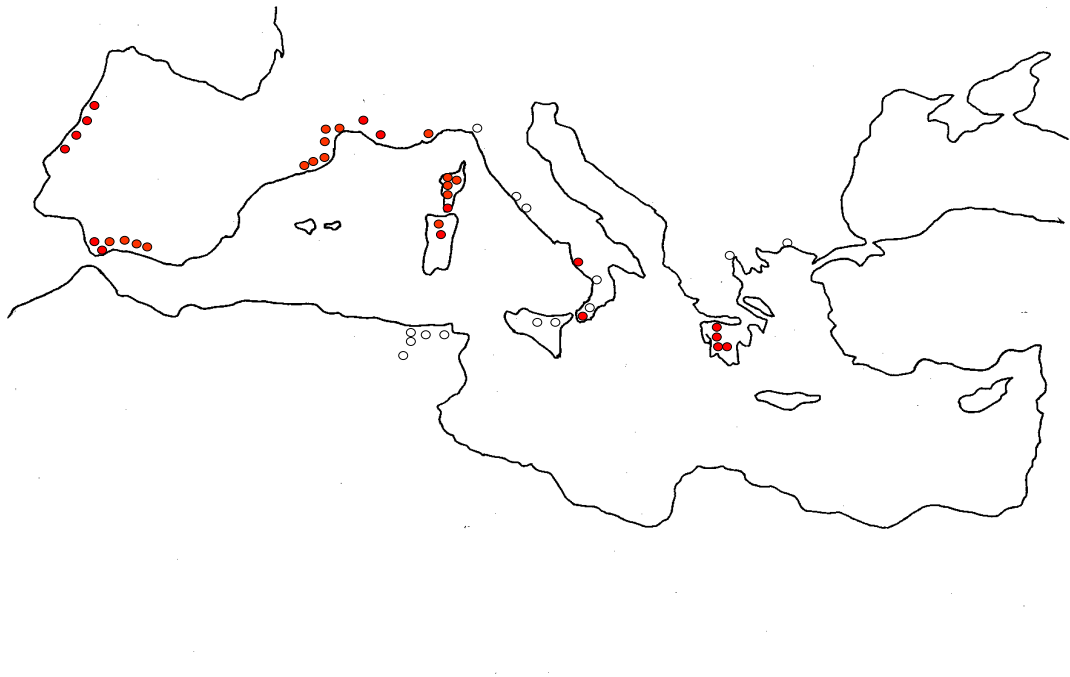


Figure 1. Map of the sample sites (o) of agent prospection surveys for the biological control of *Genista monspessulana* around the Mediterranean. Sites marked in red are populations of *G. monspessulana* of sufficient size for quantitative sampling.

Current status

The coastal surveys have so far included ten sites in Spain (NE and SW), four sites in Portugal, four sites in southern France, ten sites in Corsica, three sites in Sardinia, five sites in western Italy and Sicily and four sites in Greece. The density of sampling reflected the frequency and abundance of *G. monspessulana*. Sites in mainland Italy and Greece were localized (e.g. only one patch in a 10 x 10 km area in Greece) and had few specialist species. As a result the second trip to sample pods at these sites was considered a low priority and has yet to be carried out. Sampling was also carried out at 16 sites in the Canaries and six sites in Tunisia on other species in the Genisteae. The remaining regions still to be surveyed within the native range of *G. monspessulana* include the eastern coast of Italy and the Balkan coast, Turkey and Morocco. Phase 2 of the project completed the ‘mid-flowering’ samples of all eleven sites in southern Spain

and Portugal and pod collections from 3 of the sites in southern France. The planned survey trip to Morocco this year could not be made due to a failure to obtain an official collecting permit from the Moroccan authorities. Negotiations are continuing. Data from ‘mid-flowering’ visits to sites containing *G. monspessulana* in Greece, Italy, and both visits in two sites in Sardinia, Mediterranean France (including Corsica), NW Spain, southern Spain and Portugal have been analyzed quantitatively.

Species in the Genisteae sampled throughout these surveys are included in Table 1. Sites surveyed to date and analyzed in this report are given in Figure 1.

Table 1. Species of Genisteae sampled since January 1999 and whether or not arthropods were found. Surveys included Greece, France, Italy, Spain Portugal and Tunisia.

Species	Number of sites sampled alone	Number of sites sampled together with other Genisteae	Total number of sites sampled	Number of sites where arthropods were found on the plant
<i>Genista monspessulana</i> ^a	9	34	43	42
<i>Genista stenopetala</i> ^b	0	3	3	2
<i>Genista canariensis</i> ^b	0	1	1	1
<i>Genista corsica</i>	0	2	2	1
<i>Genista ferox</i>	2	0	2	2
<i>Genista linifolia</i> ^a	0	1	1	0
<i>Genista microcephala</i>	3	0	3	3
<i>Genista tricuspidate</i>	2	0	2	2
<i>Cytisus villosus</i>	5	16	21	21
<i>Cytisus scoparius</i> ^{a,b}	1	8	9	7
<i>Cytisus arboreus</i>	0	7	7	7
<i>Chamaecytisus proliferus</i> ^a	4	7	11	10
<i>Spartium junceum</i> ^{a,b}	2	6	8	6
<i>Calicotome spinosa</i> ^a	0	3	3	3
<i>Calicotome villosa</i>	4	10	14	14
<i>Adenocarpus foliolosus</i> ^b	0	4	4	2
<i>Adenocarpus telonensis</i>	0	4	4	4
<i>Spartocytisus filipes</i> ^b	0	1	1	1
<i>Stauracanthus boivinii</i>	0	1	1	1
<i>Retama raetam</i> ^{a,b}	1	1	2	0
<i>Ulex europaeus</i> ^{a,b}	1	7	8	8

^a Species that are also exotics in the USA.

^b Species only (or also) sampled in the Canary Islands.

Mid-flowering survey results

The quantitative beating-tray and pod sample surveys in the northern Mediterranean region have so far found 85 species of phytophagous arthropod on *G. monspessulana*. Of these 34 are considered to be specific to the level of the tribe Genisteae and 8 are specific to the genus *Genista* (Table 2). The percentage of beat samples from *G. monspessulana* that included an identified natural enemy species and the number of individuals of that species per sampled plant are also given in Table 2. The rust, *Uromyces genistae* Fuckel, was also observed attacking old leaves in late spring and summer..

Table 2. The abundance and frequency of the 34 phytophagous arthropod species that the literature suggest are at least specific to the tribe Genisteae, and that were sampled during the beating-tray survey of 30 *Genista monspessulana* sites in Greece, Italy, France, Spain and Portugal. Information is includes a) their likely specificity, b) their phytophagous feeding guild and c) other genera of the Genisteae from which these species were also collected during these surveys.

Species	Specificity ^a	Guild ^b	Insects plant ⁻¹	% frequency ^c	Other Genisteae Genera
Hemiptera					
<i>Arytaina genistae</i> (Latreille)	2	1	4.00	3	<i>Cytisus</i> ,
<i>Arytinis hakani</i> (Loginova)	1	1	11.98	70	<i>Cytisus</i> , <i>Calicotome</i>
<i>Acyrtosiphon pisum</i> ssp. <i>spartii</i> (Koch)	2	1	0.15	7	Genisteae
<i>Gargaria genistae</i> (F.)	1	1	0.43	37	<i>Cytisus</i> , <i>Spartium</i>
<i>Heterocordylus</i> ? <i>leptocerus</i> (Kb)	2	1	8.05	7	<i>Cytisus</i>
<i>Globiceps/Orthotylus/Heterocapillus</i> sp. ^e	2	1	11.82	40	<i>Cytisus</i>
Diptera					
<i>Chyliza leptogaster</i> (Panzer)	1?	7	1.5	16	
<i>Asphondylia</i> sp. (galls) ^d	2	3	0.22	53	
Lepidoptera					
<i>Agonopterix nervosa</i> (Haworth)	2	2	0.71	17	<i>Calicotome</i>
<i>Agonopterix scopariella</i> (Heinemann)	1	2	0.09	25	<i>Cytisus</i>
<i>Callophrys rubi</i> (L.)	2	2	0.04	3	
<i>Pseudoterpna pruinata</i> (Hufnagel)	2	2	0.10	3	<i>Cytisus</i> , <i>Calicotome</i>
Oecophoridae sp. ^d	2	2	0.08	13	
Pyralid sp. ^d	2	2	0.12	13	<i>Cytisus</i>
Tortricid sp. ^d	2	2	0.19	13	<i>Genista</i> , <i>Cytisus</i> , <i>Calicotome</i>
Coleoptera					
Chrysomelidae					
<i>Gonioctena Spartoxena</i> sp. ^d	2	2	0.20	7	
Bruchidae					
<i>Bruchidius villosus</i> (F)	2	5	0.67	33	<i>Cytisus</i> , <i>Calicotome</i> , <i>Spartium</i>
<i>Bruchidius lividimanus</i> (Gyll.)	2	5	3.61	57	<i>Genista</i> , <i>Cytisus</i> , <i>Calicotome</i>
Buprestidae					
<i>Anthaxia</i> sp., <i>Agrilus antiquus</i> & <i>Agrilus cinctus</i>	2	7	0.09	7	
Apionidae					
<i>Exapion fuscirostre</i> (F)	1	5	0.20	3	<i>Cytisus</i> , <i>Calicotome</i>
<i>Exapion</i> nr. <i>putoni</i> (Ch. Brisout)	1	5	0.60	7	<i>Genista</i> , <i>Calicotome</i>
<i>Lepiapion</i> ? <i>argentatum</i> (Gerstäcker)	1	5	1.39	37	
<i>Oryxolaemus</i> ? <i>scabiosus</i> (Weise)	1	3	0.20	3	<i>Cytisus</i> , <i>Calicotome</i>
<i>Pirapion</i> ? <i>immune</i> Kirby	2	3	0.13	3	<i>Cytisus</i>
<i>Protopirapion atratulum</i> (Gemar)	2	4	0.08	7	
Curculionidae					
<i>Pachytychius sparsutus</i> (Ol)	2	5	0.38	7	<i>Cytisus</i> , <i>Calicotome</i>
<i>Peritelus senex</i> (Boheman)	2	6	0.93	3	
<i>Pleurodrusus carinula</i> (Olivier)	2	6	0.09	7	<i>Cytisus</i> , <i>Spartium</i>
<i>Sitona gressorius</i> (F.)	2	6	1.00	3	
<i>Sitona regensteiniensis</i> (Herbst)	2	6	0.84	20	<i>Cytisus</i> , <i>Calicotome</i> , <i>Spartium</i>

^a Specificity; 1 - specific to genus, 2 – specific to tribe

^b Guild; 1- sap sucker, 2 – defoliator, 3 – leaf miner/galler, 4 – flower feeder, 5 – seed feeder, 6 – root feeder, 7 – stem feeder

^c % of *G. monspessulana* sites where species sampled.

^d Detailed rearing and identification required

^e Hemiptera for which species level study may show greater level of specificity within this genus

Most foliar damage observed was caused by the psyllid *Arytinnis hakani* (Loginova). The psilid fly *Chyliza leptogaster* (Panzer) and the buprestid *Agrilus antiquus* Mulsat et Rey were the only species observed killing mature plants, although only in a restricted part of the native range in southeast France. Amongst the seed feeders, the bruchid beetle, *Bruchidius lividimanus* (Gyllenhal) was the commonest species, occurring throughout the western Mediterranean followed by the apionid, *Lepidapion ?argentatum* (Gerstäcker) and weevil *Pachytychius sparsutus* (Olivier). A population of *Bruchidius villosus* (F.) in NE Spain was found restricted to *G. monspessulana* despite the presence of other known host plants in the genus *Cytisus*, but was not found at sites in southern Spain and Portugal.

The beating tray insect samples were combined for each site sampled and the number of each species found per plant was calculated for each site. These data along with the total number of species sampled per region are presented in Figure 2a for species sampled that were a) specific to the *Genista monspessulana*, b) specific to the tribe Genisteae, c) specific to the family Fabaceae and d) all other insects found including generalist flower visitors. These data were then used to calculate Shannon diversity indices, H , per plant for each site for a) species found at least specific to tribe Genisteae, b) species found at least specific to the family Fabaceae and c) all insects found including generalist flower visitors. Mean values of H were calculated for each region surveyed and are presented in Figure 2b. H is considered to be a better measure of species diversity than a simple count of the number of species (Figure 2a).

Figure 2a shows the the number of more specific natural enemies is higher in the Western Mediterranean and France and Spain show the highest numbers sampled overall and on average per site. Figure 2b shows that the natural enemy community diversity is also higher in the Western Mediterranean being very low in Greece and somewhat lower on small islands like Corsica. Portugal also had lower diversity for specialist natural enemies by region. This diversity is generally considered to become higher the closer you are to the centre of origin of the weed, a zone where prospecting should be concentrated. Natural enemy species diversity is, however, likely to be influenced by several factors other than closeness to the center of origin of the target species including a) size of the target species population sampled b) regional abundance of the target species, c) number of closely related plant species and d) the season and weather at the time of collection..

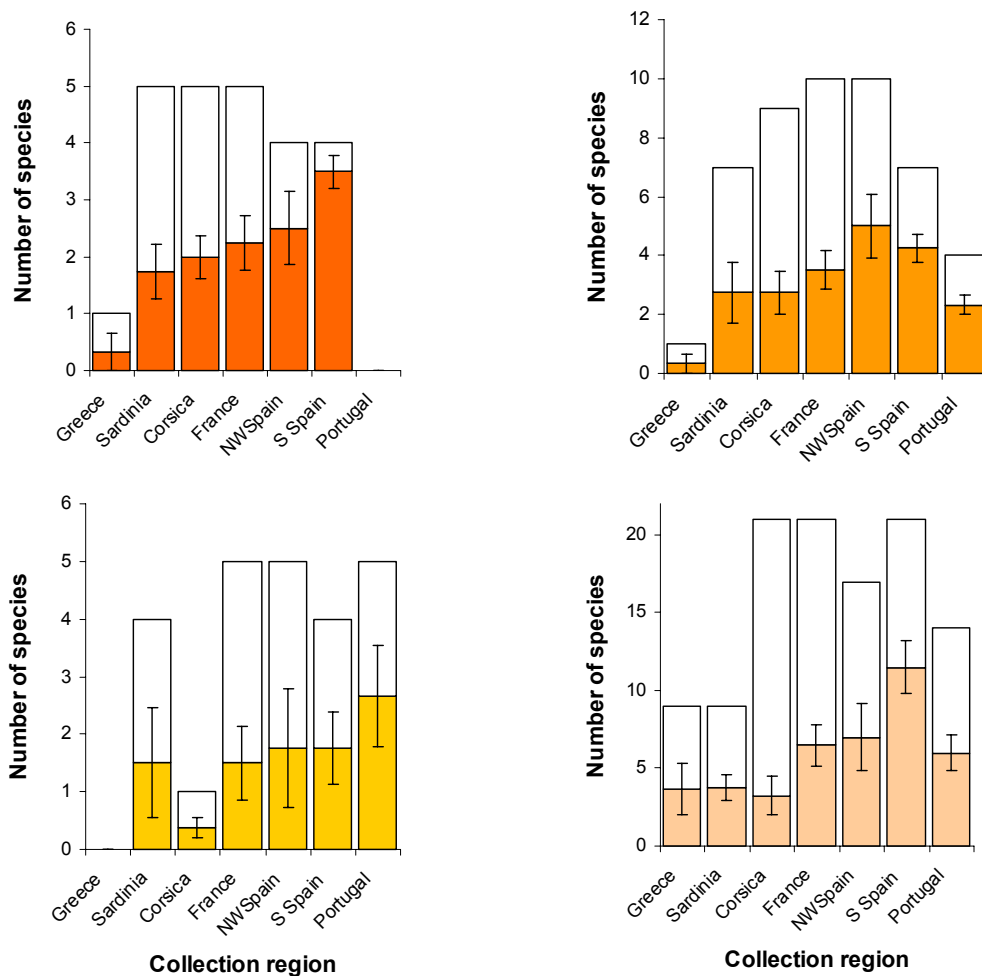


Figure 2a. Mean number of species sampled on French broom per site (filled section \pm SE) and total number (bar ht) for each region sampled for species a) specific to *Genista*, b) specific to the Genisteeae, c) species specific to the Fabaceae and d) generalist flower visitors etc.

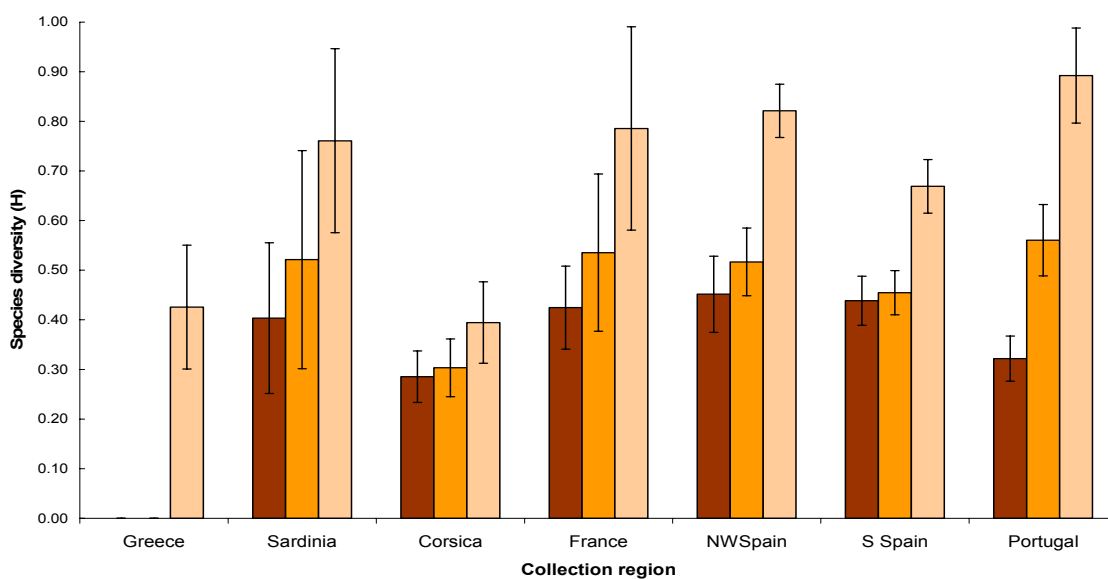


Figure 2b Mean Shannon species diversity indices per French broom plant for each region sampled at flowering for natural enemies a) at least restricted to the tribe Genisteeae, b) at least restricted to the family Fabaceae and c) all natural enemies found (\pm SE).

Table 3. A comparison of the specialist arthropod community on *Genista monspessulana*, *Cytisus scoparius* (Syrett *et al.* 1999) and *Ulex europaeus* (Zwölfer 1963) generated from the literature search and field collections. Species in bold type are the extreme specialists restricted to one host or the other. (* released or studied as a biocontrol agent).

Family	Genus	Species on <i>G. monspessulana</i>	Species on <i>C. scoparius</i>	Species on <i>U. europaeus</i>
Eriophyidae	<i>Aceria</i> <i>Tetranychus</i>		<i>genistae*</i>	<i>genistae</i> <i>linterarius*</i>
Psyllidae	<i>Arytaina</i> <i>Arytinnis</i> <i>/Arytainilla</i>	<i>Arytinnis hakani*</i>	<i>genistae</i> <i>Arytainilla</i> <i>spartiophila*</i>	
Aphididae	<i>Acyrtosiphon</i> <i>Aphis</i>	? <i>spartii</i> <i>genistae</i>	<i>spartii</i> <i>sarothamni</i>	<i>ulicis</i>
Membracidae	<i>Gargaria</i>	<i>Genistae</i>	<i>genistae</i>	<i>genistae</i>
Pentatomidae	<i>Piezodorus</i>	<i>Lituratus</i>	<i>lituratus</i>	<i>lituratus</i>
Miridae	<i>Heterocordylis</i> <i>Globiceps</i> <i>Orthotylus</i>	<i>genistae, leptocerus</i> <i>fulvicollis, genistae</i> <i>adenocarpi, beieri, virescens</i>	<i>tibialis,</i> <i>leptocerus</i> <i>fulvicollis</i> <i>adenocarpi</i> <i>beieri, virescens,</i> <i>concolor</i> <i>bicolor</i>	<i>parvulus</i> <i>bicolor</i>
Geometridae	<i>Platycranis</i> <i>Chesias</i> <i>Isturgia.</i> <i>Pseudoterpna</i>	<i>boreae</i>	<i>legatella</i> <i>limbaria</i> <i>pruinata</i>	<i>bicolor</i> <i>pruinata</i>
Oecophoridae	<i>Agonopteryx</i>	<i>scopariella</i> <i>nervosa</i>	<i>assimilella</i> <i>scopariella</i> <i>nervosa</i> <i>spartifoliella*</i>	<i>ulicetella</i> <i>nervosa</i>
Lyonetiidae	<i>Leucoptera</i>	<i>laburnella</i>	<i>immundella</i>	
Nepticuliidae	<i>Trifurcula</i>	<i>serotinella</i>	<i>mulinella</i>	<i>ulicinella</i>
Gelechiidae	<i>Mirificarma</i>	<i>cytisella</i>	<i>scopariella</i>	<i>ulicicolella</i>
Gracillariidae	<i>Phyllonorycter</i>	<i>stainoniella</i>	<i>succedana</i> <i>scopariana</i>	<i>succedana*</i> <i>ulicetana</i> <i>internana</i>
Tortricidae	<i>Cydia</i>	<i>succedana</i>		
Psilidae	<i>Chyliza</i>	<i>leptogaster</i>		
Cecidomyiidae	<i>Asphondylia</i>		<i>sarothamni</i> <i>pilosa</i> <i>genistae</i>	<i>ulicis</i>
Tenthredinidae	<i>Rhogogaster</i>		<i>fugax</i>	<i>fugax</i>
Cerambycidae	<i>Deilus</i>	<i>fugax</i>	<i>antiquus</i>	
Buprestidae	<i>Agrilus</i>	<i>antiquus</i> <i>cinctus</i>	<i>antiquus</i> <i>cinctus</i>	
Bruchidae	<i>Anthaxia</i> <i>Bruchidius</i>	<i>funerula</i> <i>lividimanus</i> <i>villosus</i>	<i>funerula</i> <i>lividimanus</i> <i>villosus*</i>	<i>funerula</i> <i>lividimanus</i>
Chrysomelidae	<i>Gonioctena</i>	<i>sexnolatus,</i> <i>gobanzi, variabilis</i>	<i>olivacea,</i> <i>variabilis</i>	
Apionidae	<i>Lepidapion</i> <i>Exapion</i> <i>Pirapion</i> <i>Protopirapion</i>	? <i>argentatum*</i> , <i>squamigerum</i> <i>?plutoni</i> <i>immune</i> <i>attratulum</i>	<i>squamigerum</i> <i>fuscirostre*,</i> <i>plutoni</i> <i>immune</i> <i>attratulum</i>	<i>pseudogallaecianum</i> <i>squamigerum</i> <i>ulicis*</i> <i>immune</i> <i>attratulum</i>
Curculionidae (roots)	<i>Sitona</i> <i>Polydrusus</i> <i>Peritelus</i>	<i>regensteinensis</i> <i>gressorius</i> <i>?cervinus</i> <i>prasinus</i> <i>senex</i>	<i>regensteinensis</i> <i>puberulus</i> <i>confluens</i> <i>prasinus</i> <i>?</i>	<i>regensteinensis</i> <i>striatellus</i> <i>?</i>
Curculionidae (seeds)	<i>Tychius</i> <i>Pachytichius</i>	<i>sparsutus*</i>	<i>parallellus</i> <i>sparsutus</i>	<i>sparsutus</i>

A comparison of Table 2 with a similar list for *C. scoparius* (Syrett *et al.* 1999), suggests it contains very few species in all orders except the Coleoptera suggesting that there may remain many species not yet detected in our surveys from *G. monspessulana*. Several species were also found during a previously presented literature search, which have not yet been seen in the field. For comparative information, Table 3 summarizes the currently known specialist arthropod community on *G. monspessulana*, *C. scoparius* and *U. europaeus* developed from both the literature search (Zwölfer 1963, Syrett *et al.* 1999) and field collections from *G. monspessulana*. This table focuses on arthropod genera where the literature suggests there are different species using these three closely related hosts.

Pod sample survey results

Data from the pod dissections from each of 25 of the *G. monspessulana* sites estimated pre-dispersal seed losses to insects for the different Genisteae species sampled across sites (Table 4) and variation in seed loss by the different pod feeding insects to *G. monspessulana* between regions (Figure 3). The overall average seed predation level in the pods of *G. monspessulana* was 22% (Table 4). This was higher than for any other co-occurring species in the Genisteae except *Calicotome spinosa* (L.) Link, although only two populations of the latter were sampled. *Genista monspessulana* seed losses to *Lepidapion ?argentatum* were highest, but the bruchids also caused comparable losses (Table 4). There was large variation in seed losses to the different seed predators across plants and sites and in overall seed losses per seed predator species between regions in the native range where *G. monspessulana* occurs (Figure 3), ranging from 6% to 39% across regions and 1% to 63% across sites.

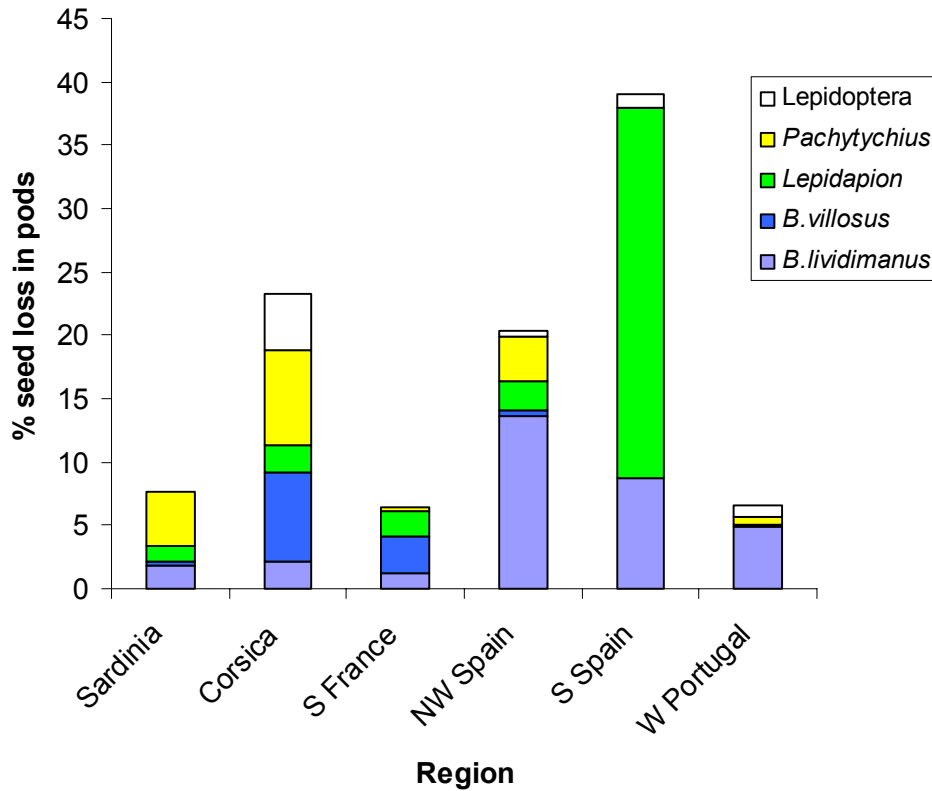
Table 4. Percent seed loss per plant overall and for the five seed predators across seven species of Genisteae at 25 sites containing natural populations of *Genista monspessulana* in the native range in Spain, France and Italy.

Genisteae	n	Mean total % seed loss per plant	<i>Bruchidius lividimanus</i>	<i>Bruchidius villosus</i>	Apionid spp. ^a	<i>Pachytychius sparsutus</i>	Lepidoptera
<i>Genista monspessulana</i>	25	22.03 ± 3.40	5.33 ± 1.18	2.92 ± 1.12	8.38 ± 3.49	2.00±0.55	3.40 ± 0.96
<i>Cytisus villosus</i>	8	9.73 ± 4.54	4.76 ± 1.24	0.06 ± 0.05	4.74 ± 4.49	0.02±0.02	0.15 ± 0.12
<i>Cytisus scoparius</i> ^b	2	1.19 ± 0.26	1.01 ± 0.43	0.00	0.00	0.00	0.17 ± 0.17
<i>Cytisus arboreus</i>	2	12.78 ± 4.56	8.60 ± 1.90	0.00	2.05 ± 1.81	0.88 ± 0.77	1.25 ± 0.08
<i>Calicotome villosa</i>	6	16.35 ± 5.14	10.94 ± 5.65	0.00	4.78 ± 3.58	0.36 ± 0.40	0.03 ± 0.03
<i>Calicotome spinosa</i>	2	34.27 ± 2.02	2.94 ± 1.87	0.00	0.00	31.02 ± 0.16	0.31 ± 0.31
<i>Adenocarpus telonensis</i>	2	4.36 ± 3.52	0.00	0.00	0.00	0.00	4.36 ± 3.52

^a*Lepidapion ?argentatum* on *G. monspessulana*, *Exapion ?subparallelum* (Dbr.) on *C. villosa*, *Exapion fuscirostre* on *C. villosus*, and *Exapion fuscirostre* on *C. arboreus*.

^bData only for sites where *C. scoparius* co-occurs with *G. monspessulana* (Corsica), other studies in the native range have estimated seed loss of *C. scoparius* to be 15-23% (Hosking 1995), 0.4-24% (Mazay 1993), 26% (Hinz 1992).

Figure 3. Percent seed loss per plant in *Genista monspessulana* to pre-dispersal seed predators (Lepidoptera, the weevil, *P. sparsutus*, the apionid *L. ?argentatum* and the two bruchids, *B. villosus* and *B. lividimanus* at 25 sites in 6 distinct geographic regions throughout south west Europe.



Conclusions

The western Mediterranean and particularly southern Spain had the largest diversity of specialist insect species. This suggests that the centre of origin of *G. monspessulana* is in the western Mediterranean. Southern Spain also had the highest level of specialist pre-dispersal seed losses per plant. High seed loss may reflect high densities of the host plant allowing the predators to build up in high numbers or low levels of parasitism or predation or high climatic favorability of this region for the performance of the specialized seed predators. The cause has not yet been determined and may therefore either be coincidental with high overall natural enemy species diversity or reflect the importance of this region in selecting potential biocontrol agents.

PRIORITIZING POTENTIAL BIOLOGICAL CONTROL AGENTS

Survey information has been used to revise a list of potential biological control agents (Table 5) and other species that given further work may attain that list. The psyllid, *A. hakani*, and the apionid, *L. ?argentatum*, are illustrated and discussed in CSIRO (2000, 2002).

Table 5. Current short list of potential biological control agents for French broom, based on surveys across the native distribution.

Current short list of potential biological control agents

- Agent 1. *Arytinis hakani* (Loginova); – Sap-sucking plant louse
(Only ever recorded from French broom)
- Agent 2. *Lepidapion ?argentatum* Gerstaecker; – Small weevil feeding on seeds
(also recorded from *G. anglica*, *G. umbellata*, *?Adenocarpus* sp.
– taxonomy under revision)
- Agent 3 *Agrilus antiquus* Mulsat et Rey – small stem and root boring beetle found in
stems that died prematurely before flowering (also recorded from
Cytisus, but not found on co-occurring *Cytisus* plants at study site where
abundant)

Agent 4

Other species which upon further investigation could be added to the short list.

Moths

- Trifurcula serotinella* H.S., – leaf miner
(not seen, specific to tribe - literature search)
- Coleophora trifarella* Zeller; – leaf miner
(not seen, specific to tribe -literature search)

Beetles

- Agrilus cinctus* Olivier – stem boring buprestid found in the stems
- Bruchidius lividimanus* Gyll. – seed feeding bruchid causing widespread
predispersal seed predation. Quite a broad host range, but not yet
recorded from *Lupinus*.
- Peritelus senex* Boheman – root feeder
(collected only from target, but host range supposedly includes
Astragalus and *Ulex*)
- Pachytychius sparsutus* (Ol) –seed feeder
(collected from most Gensiteae, but may be the most damaging seed
weevil on *G. monspessulana* and *C. scoparius* (Mazay 1993) – Further
investigations necessary

Flies

- Chyliza leptogaster* (Panzer) – stem miner found killing only *G. monspessulana*
plants when growing next to *C. villosa* and *C. villosus*
(recorded also from *Physocarpus*, *Forsythia*, *Neottia*, existence of host
races needs investigation)

Data supporting the seed feeding apionid, *Lepidapion ?argentatum*, as a potential biological control agent has increased as very high levels of pre-dispersal seed predation (>60%) have been recorded at 2 sites in southern Spain. The species name of this apionid, while it fits with current published literature is probably incorrect (M. Alonzo-Zarazaga pers. comm.) and an alternative name of *Lepidapion acuminatum* (Schilsky) applied to an apionid species recorded from *G. monspessulana* in southern Spain may eventually take precedence. This species is unusual in that the males and females are morphologically distinct in that the male rostrum (snout) is relatively short and yellow, while the female rostrum is black (Plate 1).



Plate 1. Female and male *Lepidapion ?argentatum* showing sexual dimorphism in rostrum color and length

The inclusion of two other beetle species as potential biological control agents for broom results from field observations and data analysis during phase 2 of the project. At a site in southern France (Cote d'Azur) prematurely dead *G. monspessulana* plants were observed in mixed populations with *Cytisus villosus* and *Calicotome villosa*. These other two species exhibited no such premature mortality at this site. Dead plants were dug up and found to contain tunnels of a coleopteran running up the lower part of the stem and occasionally into the roots with mature larvae inside. From these roots adults of *Agrilus antiquus* Mulsat et Rey emerged together with an adult of the cerambycid, *Deilus fugax* (Ol). The latter is only known from dead stems of shrubby legume species and so *A. antiquus* was assumed to be the causal agent of premature plant death. This species is known to be specific within the Genisteae. The potential importance of an agent that can kill plants prior to reproduction is evident and therefore we considered this species was worth placing in the list of potential biological control agents for French broom. A sibling species, *Agrilus cinctus* Olivier, has also been recorded from *Genista* and may also deserve further attention (Schaefer 1949). The status of *Pachytychius sparsutus* (Ol) may also require reappraisal

as a potential biological control agent as this species can regularly cause 8% damage to seed production of *G. monspessulana* across several sites and has not shown any capacity to date to attack *Lupinus* species on site at the laboratory in southern France, despite having quite a broad host range in the Genisteae.

It is premature to suggest that the natural enemy species with the greatest potential as biological control agents have already been discovered. This is largely due to the relatively few species of Lepidoptera found feeding on *G. monspessulana* in the field so far compared to the number known from e.g. *Cytisus scoparius*. Surveys particularly targeting known species of potentially specific Lepidoptera will need to be made as these may attack the plant during seasons, such as autumn, so far poorly surveyed. A similar emphasis on members of the chrysomelid genus *Gonioctena* subgenus *Spartoxena* would also be merited as these are hard to find in the field and poorly studied, but are potentially specific. More effort also needs to be placed on successfully rearing and understanding larvae in general observed on *G. monspessulana* in the field that have not yet been successfully reared to adult sufficiently well for identification purposes.

Objective 2: Complete as far as possible laboratory testing of the available test plants (min 10 species) on the approved plant test-list for *A. hakani* that is sufficient to ensure these agents to be ready for importation into a USA quarantine should this project continue.

DRAFT TEST PLANT LIST FOR BROOM BIOCONTROL AGENTS.

Introduction

Significant progress has been made in the preparation of a draft test list for testing biological control agents for French broom through collaboration between CSIRO staff and Mike Pitcairn and Patrick Akers at CDFA and a draft proposal was prepared in July 2002 (Pitcairn, Ackers & Sheppard in prep.). For reference, the draft list of test plants developed in this proposal is given in Table 6, however, a full justification of the species included in this list will be made available in the proposal once it has been finalized and circulated.

Table 6. First draft of host test list for French broom for TAG.

		Notes
	Family : Fabaceae	
	Subfamily : Papilionoideae	
	Tribe : Genisteae	
	Subtribe Genistinae	
1	<i>Genista monspessulana</i> (L.) Johnson	target
2	<i>Cytisus scoparius</i> (L.) Link	
3	<i>Cytisus</i> 'Porlock'	
4	<i>Cytisus x spachianus</i>	
5	<i>Spartium junceum</i> L.	
6	<i>Ulex europeus</i> L.	
	Subtribe Lupininae	
7	<i>Lupinus texensis</i> Hook.	E New World Clade
8	<i>Lupinus perennis</i> L.	W New World Clade, eastern distribution
9	<i>Lupinus arboreus</i> Sims	W New World Clade: shrub
10	<i>Lupinus albifrons</i> Benth.	W New World Clade: shrub
11	<i>Lupinus longifolius</i> (S. Wats.) Abrams	W New World Clade: shrub
12	<i>Lupinus littoralis</i> Dougl.	W New World Clade: perennial
13	<i>Lupinus microcarpus</i> var. <i>densiflorus</i> (Benth.) Jepson	W New World Clade: annual
	Tribe : Thermopsidae	
14	<i>Thermopsis macrophylla</i> Hook. & Arn. OR <i>T.</i> <i>Montana</i>	native: Genistoid Alliance
15	<i>Pickeringia montana</i> Nutt. ex Torr. & Gray	native: Genistoid Alliance
16	<i>Baptisia bracteata</i> Muhl. ex Ell.	native: Genistoid Alliance
	Tribe : Crotalariaeae	
17	<i>Crotalaria sagittalis</i> L.	native: Genistoid Alliance
	Tribe : Sophoreae	
18	<i>Sophora tomentosa</i> L. OR <i>S. secundiflora</i> e (Ortega) Lag. ex DC.	native
	Tribe : Galegeae	
19	<i>Astragalus hoodianus</i> T.J. Howell, <i>A. nuttalli</i> (Torr. & Gray) J.T. Howell, <i>A. pomonensis</i>	native

Jones, *A. saurinus* Barneby, **OR** *A.*
lonchocarpus Torr.

- Tribe** : Amorpheae
20 *Amorpha canescens* Pursh, *A. fruticosa* L. **OR**
A. californica Nutt. native
- Tribe** : Vicieae
21 *Lathyrus vestitus* Nutt. native
22 *Pisum sativum* L. crop: garden pea
23 *Vicia faba* L. crop: fava bean
- Tribe** : Psoraleae
24 *Hoita (Psoralea) macrostachya* (DC.) Rydb. native
- Tribe** : Robinieae
25 *Sesbania exaltata* (Raf.) Cory native
- Tribe** : Hedysareae
26 *Hedysarum boreale* Nutt. native
- Tribe** : Phaseoleae
27 *Phaseolus vulgaris* L. crop: table beans
28 *Glycine max* (L.) Merr. crop: soybean
- Tribe** : Trifolieae
29 *Medicago sativa* L. crop: alfalfa
30 *Trifolium repens* L. crop white clover
- Tribe** : Loteae
31 *Anthyllis vulneraria* L. **OR** *Coronilla varia* L. crop: kidney vetch or trailing crownvetch
- Subfamily** :
Caesalpinioideae
- Tribe** : Cercideae
32 *Cercis occidentalis* Torrey native
- Family** : Polygalaceae
33 *Polygala californica* **OR** *P. cornuta* native
- Family** : Sapindaceae
34 *Dodonaea viscosa* (L.) Jacq. **OR** *Sapindus*
saponaria L. native
- Family** : Rosaceae
35 *Fragaria x ananassa* Duchesne crop: strawberry
36 *Rubus idaeus* L., e.g., 'Heritage' crop: raspberry
- Family** : Hydrangeaceae
37 *Hydrangea quercifolia* Bartr. native, ornamental
- Family** : Solanaceae
38 *Lycopersicon esculentum* P. Mill. crop: tomato
- Family** : Brassicaceae
39 *Brassica oleracea* L. crop: broccoli
- Family** : Chenopodiaceae
40 *Beta vulgaris* L. crop: beet
- Family** : Malvaceae
41 *Gossypium hirsutum* L. crop: cotton
- Family** : Apiaceae
42 *Daucus carota* L. crop: carrots
- Family** : Poaceae
43 *Zea mays* L. crop: corn
44 *Triticum aestivum* L. crop: wheat

HOST RANGE TESTING OF *ARYTINNIS HAKANI*

Methods

Progress against this objective is nearing completion. Testing of *A. hakani* has been conducted using test cages containing 5 to 10 test plants run in parallel with control cages containing two plants of *G. monspessulana* only. Testing was carried out as series at 4 separate times three of which (B, C & D) were completed after the end of phase 1 of this project. Five replicate cages were set up with one plant of each of 5 or 6 test plant species. Two (series A & B) or 5 (series C & D) parallel cages were set up containing the two *G. monspessulana* control plants. Five adult *A. hakani* couples (5 male and 5 female) per test plant were introduced into each cage with test plants and into the control cages. After 7-10 days, depending on the level of oviposition observed on the control plants, each series of test cages were dismantled and the adults were removed. All plants were observed for eggs, nymphs and adults once a week for 8 weeks. Adults started to appear on controls after 6 weeks.

Results

In total 47 plant species have so far been tested of which 32 are on the draft test plant list in Table 6. Of the remaining 11 test plant species that need to be tested on the draft test list, 8 could not be tested due to lack of available seeds or plant material and 3 could not be tested as the plants failed to grow big enough to allow for testing. Eggs were observed on all *G. monspessulana* control plants and hybrids (*Cytisus* “Porlock” and *Cytisus x spachianus*), oviposition was also observed on *C. proliferus* (4 reps), *S. junceum* (3 reps) *C. spinosa* (2 reps), *L. anagyroides* (2 reps), *L. arboreus* (4 reps), *L. longifolius* (4 reps), *L. pilosus* (3 reps), *L. perennis* (1 rep), *L. affinis* (1 rep), *L. bicolor* (1 rep). Survival beyond early instar nymphs and greater than 2 weeks occurred on *G. monspessulana* and hybrids, *S. junceum* and *L. longifolius*. Survival to adult did take place, however development time to adult was slower on these species than on the control *G. monspessulana* plants and the adults that completed development failed to survive to lay eggs. Adults on control plants quickly laid eggs and continued to build up populations on the control plants. Furthermore another 24 test plant species have also been tested as part of the risk assessment for Australia including *Lupinus angustifolius* L. and none of these species received eggs

Conclusions

The unexpected survival of *A. hakani* on *L. longifolius* would suggest that there is a risk to native members of the genus *Lupinus* should this agent be released in the USA. It suggests *A. hakani* adult oviposition choice and nymphal survival may be correlated. In order to evaluate this risk more objectively, it would be necessary to carry out further testing of this agent against native *Lupinus* species. This further testing might include a broader range of *Lupinus* species with wider taxonomic, biological and geographic variation than those currently on the test list. This could focus around *Lupinus* species related closely (see Ainouche & Bayer 1999) to those that have already received eggs or supported larval development during the tests reported here. Such tests might also include direct nymphal survival tests in addition to adult oviposition testing in case nymphal survival might occur on *Lupinus* species not currently selected by adults during oviposition tests. Biochemical analysis of several *Lupinus* species including *L. longifolius* and *G. monspessulana* may also help identify factors that are correlated with significant nymphal survival. This would probably require one to two more seasons of host specificity testing.

Table 7. Results of host specificity testing completed for *A. hakani* at 4 separate times; A: March 2001, B : May 2002, C : March 2003, D : June 2003. Underlined species still need to be tested

Test Plant	Series & No. reps	No. Reps with eggs	No eggs/ attacked plant ⁻¹	No. L1-L3 nymphs/ attacked plant ⁻¹	No. L3-L5 nymphs/ attacked plant ⁻¹	No. adults attacked plant ⁻¹	survival weeks
Family Fabaceae							
Subfamily Papilionioideae							
Tribe Genisteae							
Subtribe Genistinae							
<i>Genista monspessulana</i> (L.) Johnson	A-6	6	163	77	43	33	7 ^a
<i>Genista monspessulana</i> (L.) Johnson	B-2	2	105	105	28 ^b	28 ^b	8
<i>Genista monspessulana</i> (L.) Johnson	C-5	5	168	212	mv	57	8
<i>Genista monspessulana</i> (L.) Johnson	D-5	5	69.4	33	mv	mv	8
<i>Cytisus x spachianus</i>	B-5	5	22	20	5	4.2	4
<i>Cytisus 'Porlock'</i>	A-5	5	95	100	100	50	8
<i>Cytisus scoparius</i> (L.) Link	A-5	0	0	0	0	0	0
<i>Cytisus X praecox</i>	A-5	0	0.0	0.0	0	0	0
<i>Cytisus striatus</i> (Hill) Rothm.	A-5	0	0.0	0.0	0	0	0
<i>Cytisus proliferus</i> (L.f) Link	A-5	4	9.8	0.3	0	0	0
<i>Spartium junceum</i> L.	A-5	3	20	20.3	13.3	1.6	6
<i>Calicotome spinosa</i> L.	A-5	2	15.0	0.5	0	0	1
<i>Ulex europeus</i> L.	A-5	0	0	0	0	0	0
<i>Laburnum anagyroides</i> Medikus	A-5	2	11.0	1.0	0	0	1
Subtribe Lupininae							
<i>Lupinus texensis</i> Hook.	C-5	0	0	0	0	0	0
<i>Lupinus perennis</i> L.	D-5	1	4	4	0	0	2
<i>Lupinus arboreus</i> Sims	A-5	4	3.8	1	0	0	1
<i>Lupinus albifrons</i> Benth.							
<i>Lupinus longifolius</i> (S. Wats.) Abrams	C-5	4	33.5	34.7	5.5	2.5	6
<i>Lupinus littoralis</i> Dougl.							
<i>Lupinus microcarpus</i> var. densiflorus (Benth) Jepson	A-5	0	0	0	0	0	0
<i>Lupinus cocinnus</i> J. Agardh	A-4	0	0.0	0.0	0	0	0
<i>Lupinus polyphyllus</i> Lindley	A-5	0	0.0	0.0	0	0	0
<i>Lupinus rivularis</i> Lindley	A-5	0	0.0	0.0	0	0	0
<i>Lupinus pusillus</i> Pursh	A-5	0	0.0	0.0	0	0	0
<i>Lupinus luteolus</i> Kellogg	A-5	0	0.0	0.0	0	0	0
<i>Lupinus elegans</i> H.B.K.	A-5	0	0.0	0.0	0	0	0
<i>Lupinus bicolor</i> Lindley	A-5	1	10.0	0.0	0	0	0
<i>Lupinus affinis</i> Agardh	A-5	1	32.0	1.0	0	0	2
<i>Lupinus pilosus</i> L.	A-5	3	24.3	13.0	0	0	2
<i>Lupinus succulentus</i> K. Koch	A-5	0	0.0	0.0	0	0	0
Tribe Thermopsidae							
<i>Thermopsis macrophylla</i> Hook. & Arn.	B-5	0	0	0	0	0	0
<u><i>Pickeringia montana</i> Nutt. ex Torr. & Gray</u>	NO SEEDS						
<u><i>Baptisia bracteata</i> Muhl. ex Ell.</u>	not available at the right time						
Tribe Crotalariaeae							
<u><i>Crotalaria sagittalis</i> L.</u>	not available at the right time						
Tribe Sophoreae							
<u><i>Sophora tomentosa</i> L.</u>	NO SEEDS						
Tribe Galegeae							
<i>Astragalus pomonensis</i> Jones.	D-5	0	0	0	0	0	0

Test Plant	Series & No. reps	No. Reps with eggs	No eggs/ attacked plant ⁻¹	No. L1-L3 nymphs/ attacked plant ⁻¹	No. L3-L5 nymphs/ attacked plant ⁻¹	No. adults attacked plant ⁻¹	survival weeks
Tribe Amorpheae							
<i>Amorpha californica</i> Nutt.	D-5	0	0	0	0	0	0
Tribe Viciaeae							
<i>Lathyrus vestitus</i> Nutt.	C-5	0	0	0	0	0	0
<i>Pisum sativum</i> L.	A-5	0	0	0	0	0	0
<i>Vicia faba</i> L.	A-5	0	0	0	0	0	0
Tribe Psoraleae							
<i>Hoita (Psoralea) macrostachya</i> (DC.) Rydb.	C-5	0	0	0	0	0	0
Tribe Robinieae							
<i>Sesbania exaltata</i> (Raf.) Cory	NO SEEDS						
Tribe Hedysarea							
<i>Hedysarum boreale</i> Nutt.	NO SEEDS						
Tribe Phaseoleae							
<i>Phaseolus vulgaris</i> L.	C-5	0	0	0	0	0	0
<i>Glycine max</i> (L.) Merr.	C-5	0	0	0	0	0	0
Tribe Trifolieae							
<i>Medicago sativa</i> L.	A-5	0	0	0	0	0	0
<i>Trifolium repens</i> L.	A-5	0	0	0	0	0	0
Tribe Loteae							
<i>Anthyllis vulneraria</i> L.	D-5	0	0	0	0	0	0
Subfamily Caesalpinioideae							
Tribe Cercideae							
<i>Cercis occidentalis</i> Torrey	not available at the right time						
Family Polygalaceae							
<i>Polygala californica</i> OR <i>P. cornuta</i>	NO SEEDS						
Family Sapindaceae							
<i>Dodonaea viscosa</i> (L.) Jacq.	NO SEEDS						
OR <i>Sapindus saponaria</i> L.	NO SEEDS						
Family Rosaceae							
<i>Fragaria x ananassa</i> Duchesne	D-5	0	0	0	0	0	0
<i>Rubus idaeus</i> L., e.g., 'Heritage'	D-5	0	0	0	0	0	0
Family Hydrangeaceae							
<i>Hydrangea quercifolia</i> Bartr.	NO SEEDS						
Family Solanaceae							
<i>Lycopersicon esculentum</i> P. Mill.	C-5	0	0	0	0	0	0
Family Brassicaceae							
<i>Brassica oleracea</i> L.	C-5	0	0	0	0	0	0
Family Chenopodiaceae							
<i>Beta vulgaris</i> L.	C-5	0	0	0	0	0	0
Family Malvaceae							
<i>Gossypium hirsutum</i> L.	C-5	0	0	0	0	0	0
Family Apiaceae							
<i>Daucus carota</i> L.	C-5	0	0	0	0	0	0
Family Poaceae							
<i>Zea mays</i> L.	C-5	0	0	0	0	0	0
<i>Triticum aestivum</i> L.	C-5	0	0	0	0	0	0

a two plants died from *A. hakani* attack; b numbers estimated as only data on survival % recorded; mv = missing value (counts in series D not collected as all test plants were insect free after 2 weeks).

HOST RANGE TESTING OF *BRUCHIDIUS VILLOSUS*

The current status of our understanding of the host specificity of *B. villosus* was presented in a poster at the XIth International Symposium on Biological Control of Weeds in Canberra Australia in April 2003. A copy of this poster at the end of this report summarizes this information.

HOST RANGE TESTING OF OTHER AGENTS

Work has started on developing host range testing protocols for two other potential biological control agents. *Lepidapion ?argentatum* has been successfully established in the broom garden at the CSIRO lab for the second year running. Lab-based Perspex cages were used to explore host specificity testing protocols, where branches of different test species were bunched together and passed into the cage through a hole in the base. Adult apionid pairs were placed in the cage containing flowering test plants to assist ovary development. Unfortunately all developing pods of control and test species aborted, probably due to lack of effective pollination. The cages did however allow active observation of adult behavior of these extremely small apionids

Thirty adults of the stem-mining fly, *Chyliza leptogaster* (Panzer), were individually reared in April to May 2003 from field collected material. A rearing cage system was developed that allowed adult survival to last 1 month, compared to only several days in previous years using constant supplies of readily accessible, water, pollen, honeydew and *Genista* branches. To test the specificity of this fly and whether or not oviposition required prior damage scars, single male and female pairs were placed in permeable plastic tubes around stems of *G. monspessulana* and *Forsythia europaea* Degen & Bald, as literature records indicated that adults of this fly had also been reared from this plant genus. Stem sections in the tube cages were cut with a scalpel on one part to provide "prior damage" oviposition sites if necessary. Once the flies had died, experimental plants were placed outside to allow larval development. Whether or not successful oviposition took place and larval development is occurring should be known this autumn.

Progress on previous recommendations

Work in Phase 1 highlighted a number of recommendations progress on these recommendations are documented below

- 1) *A separate taxonomic study of the Apionidae on the Genisteae*: The Cooperative Research Centre for Australian Weed Management funding for this PhD project has undergone protracted negotiations to set up a joint PhD position between an Australian and a French University. These negotiations failed to secure support from the Australian University so the CRC is now considering funding the whole project through the University of Montpellier II near CSIRO's European laboratory and USDA-ARS EBCL laboratory in France. A candidate, Boris Fumanal, has been selected to conduct the research should it get the go ahead.
- 2) *Approval of a test plant list* : A draft test list approval proposal has been drafted by CDFA for submission to the biological control federal Technical Advisory Group. The completion of this proposal can be made with further project funding
- 3) *Rapid completion of agent selection* : four species have now been selected as potential agents with a fifth species, the fly *C. leptogaster*, also undergoing further study to see whether it can attack undamaged plants.
- 4) *Molecular studies of intra-specific variation in French broom*. Progress can not be made on this recommendation without further project support.
- 5) *International Broom Initiative : Expansion of the project to include other weeds in the Genisteae* : Efforts are continuing to secure the funding levels required.

New recommendations

- 1) *Testing of A. hakani against a broader range of Lupinus species*. The inconclusive results from the host specificity testing to date suggest that more detailed testing of this psyllid is required on Lupinus species that received eggs and those species closely related to them for oviposition and nymphal survival.
- 2) *Evaluation of the impact of potential agents*. If funding were available it would be important to evaluate the potential impacts of selected agents to a) use efficacy as a selection criteria and b) predict the likely impacts following release

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Host plant use variation in the broom bruchid, *Bruchidius villosus*: assessing the risks to North American Lupines



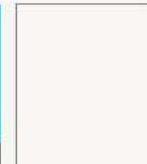
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Andy Sheppard



Melanie Haines

Seed-feeding bruchid beetle, *Bruchidius villosus*, has a checkered history as a biological control agent for Scotch broom *Cytisus scoparius* (L.) Link.

Accidental Introductions

Arriving in the eastern USA sometime prior to 1918, *Bruchidius villosus* feeds exclusively on Scotch broom throughout its range in eastern USA & Canada. An official redistribution program to the western States has started.

Biological Control Introductions

Following no-choice oviposition tests involving *Cytisus* (5 sp & varieties), *Cytisus proliferus*, *Genista* (3 sp), *Laburnum* (2 sp) and *Ulex europaeus* confirmed Scotch broom as the only accepted host, *Bruchidius villosus* from the UK, was released in New Zealand in 1987 and then Australia in 1995 as a biological control agent.

Key non-targets in the Genisteae

Tagasaste/Tree Lucerne *Cytisus proliferus* L.f. – key fodder species in Australia: **Lupines** *Lupinus* species – many commercial and native species in North America

Post release non-target damage

B. villosus released into New Zealand was recorded developing fully on **Tagasaste** in the field in the mid 1990's questioning oviposition test results.



Figure 1 *Bruchidius villosus* emerging from a *Lupinus arboreus* seed.

Laboratory based oviposition no-choice tests failed to predict this. Why?

A. Field surveys, literature sources

1 Host use variation of wild *Bruchidius villosus* populations from Scotch broom, *Genista monspessulana* and *Spartium junceum*

- Populations in the UK and northern France have only ever been recorded infesting Scotch broom despite sympatric *Genista*, *Spartium* and *Laburnum* populations

B. Open field 'choice' trial

A 2-year field choice test using *B. villosus* from *Spartium junceum*

GM GT SJ CR	CG GM CE GM	GM= <i>Genista monspessulana</i>
SJ CT CE GT	SJ CT CR CS	GT= <i>Genista tinctoria</i>
CP CG CP CS	CP CR CP GT	CE= <i>Coronilla emerus</i>
GM CS CR CT	GT CS SJ CT	CG= <i>Coronilla glauca</i>
		CP= <i>Cytisus scoparius</i>

C. Broom garden trial

Cytisus proliferus - test

Plan of one of two blocks

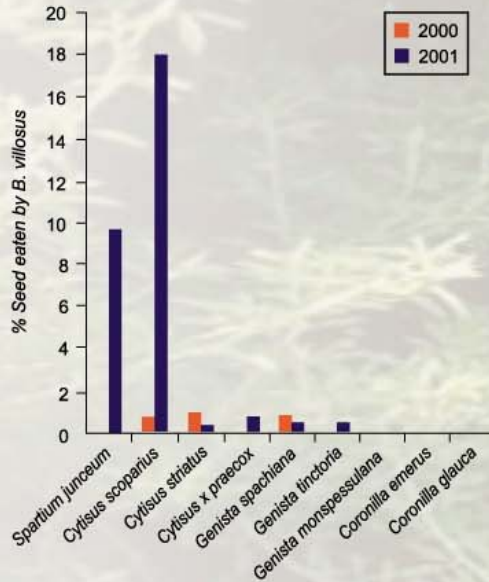
Row 1	<i>Cytisus scoparius</i> 10 plants	<i>Genista monspessulana</i> 10 plants	<i>Cytisus proliferus</i> 10 plants
Row	<i>Cytisus</i>	<i>Cytisus</i>	<i>Genista</i>

- Populations infesting *Genista monspessulana* seeds (1.1 to 19.4%) in southern France, Spain and Italy did not infest intermixed populations of either Scotch broom or *Cytisus villosus*
- Populations infesting *Spartium junceum* seeds (3-10%) in southern France, did not infest intermixed populations of *Genista monspessulana*

GT CP SJ CS	CG CP CE GM	CP= <i>Cytisus praecox</i>
GM CR CS CT	CS SJ CT GT	CS= <i>Cytisus scoparius</i>
CG SJ CP CR	GM CR CP CR	CT= <i>Cytisus striatus</i>
GM GT CT CE	CT GT CS SJ	SJ= <i>Spartium junceum</i>

Figure 2 Open field trial design Montferrier-sur-Lez, France.

In 1st year *B. villosus* attacked 3 sp. and the next year it attacked 7 sp. of the 9 presented. *B. villosus* appeared to adapt to the potential hosts presented



Genista monspessulana, with the most contrasting pod production phenology to *Spartium*, remained poorly attacked.

2 Confirmed *B. villosus* hosts from pod rearings

Fabaceae Hosts	Record Country	References
Tribe Genisteae; Subtribe Genistineae		
<i>Cytisus scoparius</i>	W, EU	Many refs
<i>Cytisus villosus</i>	F	Frick 1962
<i>Cytisus striatus</i>	F E	Syrett Emberson 1997
<i>Cytisus cantabricus</i>	E	Hosking 1995
<i>Cytisus hirsutus</i>	Au, Gr	Frick 1962
<i>Cytisophyllum sessilifolium</i>	F I	Frick 1962
<i>Genista hispanica</i>	Ch	Frick 1962
<i>Genista tinctoria</i>	ICh	Frick 1962
<i>Genista monspessulana</i>	F E	Frick 1962
<i>Genista pilosa</i>	C. Eu	A. Szentesi
<i>Laburnum alpinum</i>	Au Dk F DI	Frick 1962
<i>Laburnum anagyroides</i>	Au F Hu I	Frick 1962
<i>Spartium junceum</i> L.	GB F D I	Frick 1962
<i>Petteria ramentacea</i>	Yu	Bottimer 1968
<i>Cytisus multiflorus</i> x <i>Cytisus scoparius</i>	F (tests)	
<i>Cytisus scoparius</i>	F (tests)	
<i>Cytisus proliferus</i> .	F (tests)	
<i>Genista spachiana</i>	F (tests)	
<i>Genista stenopetala</i> . x <i>Genista canariensis</i>	F (tests)	
Tribe Genisteae; Subtribe Lupininae		
<i>Lupinus arboreus</i>	Fr (tests)	

Summary of evidence for host use

Populations of *B. villosus* exist in the native range that used only one host genus, however every known genus of host-plant within the host range of *B. villosus* remains unattacked in some part of the native range despite being available

Geographic variation of host preference among *B. villosus* populations could not be linked to significant detectable molecular genetic variation (M. Haines & J. F. Martin unpublished)

Apparently specific *B. villosus* populations lost this strict specificity when exposed to long-term host choice conditions even in their natural environment, unless potential hosts showed large differences in phenology to the local host.

B. villosus populations may synchronize their reproductive activity to the locally most abundant host plant within the host genera *Cytisus*, *Genista* or *Spartium* or *Laburnum* while less abundant potential hosts in these genera lose attractiveness.

If true, introduced apparently specific Scotch broom- based populations of *B. villosus* could expand their host range when exposed to abundant suitable alternate hosts of overlapping flowering phenology (e.g. as happened to Tagasaste in NZ)

Native range open field tests allow presentation of plots of test plants brings new insights to host range testing and host use.

2	<i>proliferus</i> 10 plants	<i>scoparius</i> 10 plants	<i>monspessulana</i> 10 plants
Row 3	<i>Genista monspessulana</i> 10 plants	<i>Cytisus proliferus</i> 10 plants	<i>Cytisus scoparius</i> 10 plants

Cytisus proliferus
1st flowering: 15/02/00
2nd flowering: 15/05/00

Genista
Flowering: 20/04/00

Cytisus
Flowering: 15/05/00

Plants of each species reached 2 to 3 metres high and produced hundreds of pods, that were collected in boxes by species.



Lupinus arboreus
Flowering: 20/05/00



All the seeds were dissected

	Total seeds	Seeds with <i>B. Villosus</i> emergence
<i>C. proliferus</i>	1383	61 (44%)
<i>Lupinus arboreus</i>	1564	38 (2,4%)

While all field studies in Europe suggested the host range of *B. villosus* is limited to members of the subtribe Genistineae, we found a population of *B. villosus* from *Spartium junceum* laying eggs on *Lupinus arboreus* and *L. elegans*. Larval development under field conditions was completed only in *L. arboreus*

It appears *B. villosus* may feed on some North American Lupines where their respective North American distributions overlap.

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