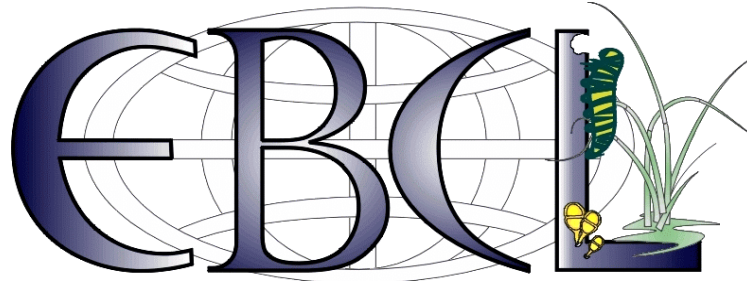


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European Biological Control Laboratory

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Photo: Javid Kashefi

**Survey of Natural Enemies of Rush Skeletonweed in
Eurasia, 2001**

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In collaboration with
George Markin, United States Forest Service, Bozeman, Montana

Introduction

In 1996 a cooperative effort between the United States Forest Service (USFS) and the European Biological Laboratory (EBCL) undertook to implement a new program for biological control of Rush skeletonweed (*Chondrilla juncea*), a new invasive weed of the Interior Pacific Northwest. The program initially concentrated on studying and host testing of the moth *Bradyrrhoa gilveolella*, a biological control agent released in Australia but not in North America, in an earlier biocontrol effort. Testing of this moth was completed in 2000, and a petition requesting permission to release it has been submitted to USDA-APHIS.

The emphasis of the present program has now shifted to the selection of the next agent, or series of agents, to be studied. In the previous program, primarily conducted by Australia between 1970 and 1985, the natural enemies studied all came from the border of the Mediterranean Sea, an area climatically similar to the areas of Australia where Rush skeletonweed was a problem. For the present program the decision was made to shift the search for new agents northward to areas climatically more similar to the Interior Pacific Northwest. The area to be targeted included the mountains of the Balkan Peninsula and other countries around the Black Sea, an area not investigated during the earlier program since the area was inaccessible to Westerners during the Cold War. Preliminary surveys in this area by EBCL personnel, incidental to their work on other programs, had indicated that Rush skeletonweed was widely distributed through that area and had a rich complex of natural enemies attacking it. The attached reports summarize the findings of these surveys.

Budget

A grant awarded to EBCL by the Idaho Department of Agriculture in the fall of 2000 allowed a much larger and more intense survey to be conducted in 2001.

Trust Fund—Idaho Department of Agriculture: \$25,000 (Signed by Idaho Dept. of Ag. 14 November 2000)

Item	Amount Spent / obligated
Indirect Program Support Costs (HQ)	2 500
Republic of Georgia Res. Mat. Exch. Agree. (signed)	5 000
Rep. of Georgia - Travel, D. Coutinot	3 000
Bulgaria Res. Mat. Exch. Agree. (signed)	5 000
Bulgaria - car rental/hotels for cooperators	590
Travel to Bulgaria, Rep. Of Georgia, France, G Markin	5 000
Ukraine Res. Mat. Exch. Agree. (to be signed Mar. 2)	3 910
	25 000

Budget continued

Funds provided to skeletonweed project by USDA -ARS, EBCL and USDA -USFS to support Idaho Trust Fund Grant

Item	Amount spent / obligated
Salaries – EBCL	
D. Coutinot (10%)	4 600
J. Kashefi (15%)	3 900
R. Sforza (5%)	2 500
T. Widmer (2%)	1 800
P. Quimby (1%)	1 350
Admin. Salaries (1%)	1 200
Space charges (labs - offices)	1 800
Space charges (quarantine)	2 000
Travel and vehicle use	
J. Kashefi - Greece/Turkey	2 000
Obligation to complete Ukraine Agree.	1 090
	22 240
Salaries – USFS	
G. Markin (10%)	10 000
Travel	
G. Markin (Nov. 2001 to Bulgaria, Greece, France and Switzerland)	6 000
	16 000*

*Does not include other aspects of work dedicated to Chondrilla juncea project estimated at 1/3 of budget or approximately \$93,000.

Greece: Javid Kashefi
(EBCL-ARS-USDA, Substation—Thessaloniki, Greece)

Since 1997 Greece has been the source of the root attacking caterpillar, *Bradyrrhoa gilveolella* (Figure 1). The caterpillars were collected and shipped to quarantine facilities in Bozeman, Montana, for host testing to determine their suitability as a new biological control agent for *Chondrilla juncea*. During the collection of this caterpillar, numerous other natural enemies were encountered. As time permitted, larvae of these insects were collected and returned to the EBCL substation laboratory in Thessaloniki and reared to provide scientific specimens. The new support provided by Idaho in 2001 not only allowed me to expand this survey but also to complete the sorting, mounting, and labeling of this *Chondrilla juncea* collection. The specimens were also shipped to the USDA Systematics Entomology Laboratory at Beltsville, Maryland, USA, for identification. The attached table (Table 1) summarizes the information available to date on these natural enemies of *Chondrilla juncea* in Greece. Unfortunately, this systematics laboratory often kept the specimen if it was new or unusual, sending me only the scientific name. Without the specimen and the collection information on its label, I was not able to identify the exact location at which many of the specimens had been collected.



Photo: Rouhollah Sobhian
Figure 1. Feeding tubes and damage caused
By the larvae of the moth, *Bradyrrhoa gilveolella*
on the tap root of *Chondrilla juncea*



Photo: Javid Kashefi
Figure 2. Cerambycid beetle (*Agapanthia violecca*)
reared from the stem of Rush skeletonweed.

Table 1. Insects reared and identified from *Chondrilla juncea* in Greece by Javid Kashfi

Order/Family	Identification	Location	Elevation	Habitat	Type of Attack	Abundance
Lepidoptera, Tortricidae	<u>Lobesia botrana</u>	Road Hraklion to Agios Nikolaou – Island of Crete	Mid elevation	Road side	Seed head	Common
Lepidoptera, Noctuidae	<u>Schinia cognata</u>	University Farm, Thessaloniki, N. Greece	Sea level	Orchard	Seed head	Common
Lepidoptera Cosmopterigidae	<u>Pyroderces argyrogrammos</u>	Notes on specimen kept by SEL	?	?	Reared from <u>C. juncea</u>	?
Diptera, Sciaridae	Pending	Bridge on Lake Aliakmonas road Kozani-Larisa, NW Greece and University Farm, Thessaloniki, N. Greece	700 m	Road side	Root	common
Diptera, Tephritidae	<u>Tephritis sp. near separata</u> Rondani	Notes on specimen kept by SEL	?	?	Reared from <u>C. juncea</u>	?
Diptera, Tephritidae	<u>Ensina sonchi</u>	Notes on specimen kept by SEL	?	?	Reared from <u>C. juncea</u>	?
Diptera, Cecidomiidae	<u>Cystiphora schmidtii</u>	University Farm, Thessaloniki, N. Greece	Sea level	Orchard	Leaf	Common
Coleoptera, Curculionidae	<u>Cicloderes canescens</u>	Omalos, Island of Crete	1,000 m	Dry in summer, cold and snowy in winter	Rosette root crown	common
Coleoptera, Cerambycidae	Pending	Klidi, road Thessaloniki to Veria, N. Greece	10 m	Road side	Stem miner	Not common
Coleoptera, Cerambycidae, Lamiinae	<u>Agapanthia cardui</u> (?)	Klidi, road Thessaloniki to Veria, N. Greece	10 m	Road side	Stem miner	Not common
Coleoptera, Cerambycidae, Lamiinae	<u>Calamobius filum</u>	Klidi, road Thessaloniki to Veria, N. Greece	10 m	Road side	Stem miner	Not common

Order/Family	Identification	Location	Elevation	Habitat	Type of Attack	Abundance
Coleoptera, Cerambycidae, Lamiinae	<u>Agapanthia violacea</u>	Klidi, road Thessaloniki to Veria, N. Greece	10 m	Road side	Stem miner	Not common
Coleoptera, Chrysomelidae, Oedemeridae	<u>Oedemera sp.</u>	Klidi, road Thessaloniki to Veria, N. Greece	10 m	Road side	Stem miner	Not common
Hymenoptera, Pteromalidae	<u>Pteromalus sp.</u>	Notes on specimen kept by SEL	?	?	Reared from <u>C. juncea</u>	?
Hymenoptera, Eurytomidae	<u>Eurytoma strigifrons</u>	Neapoli road Kastoria to Ioannina, NW Greece	Mountain about 800 m	Road side	Gall	Rare

Table 1. (continued)

Ukraine: Rene Sforza (EBCL-ARS-USDA, Montferrier-sur-Lez, France)

Ukraine is one of the largest countries of eastern Europe bordering the Black Sea (Figure 3). Climate matching showed that central Ukraine is very similar to the Northwestern states of the United States. Three field trips in 2001 were dedicated in part to surveys of native *Chondrilla juncea*. A contact has been established at Bila Tserkva State Agrarian University, located 120 km south of Kiev, with Dr. Mickael Baranovski, vice rector of the university. A cooperative agreement, which will allow us to continue working in the Ukraine, is to be signed in March 2002.



Figure 3. Country of Ukraine:
Dotted circle indicates area surveyed for *Chondrilla*

The area investigated is 600km long and 300km wide. *Chondrilla juncea* was found in all the southern part of Ukraine close to the Crimea. *Chondrilla juncea* was not found in the northern area surveyed. *Chondrilla juncea* seems to occur on sandy soil in the southern part of Ukraine, primarily in the steppe zone. Average winter minimum temperature in central Ukraine is minus 35°C and may have a strong influence on development of these species.

- In the South, I collected a large number of scale insects (Hemiptera: Coccoidea) infesting apices on 2-4 cm length and inducing severe symptoms with absence of flower buds. These insects are stored in 70% alcohol and are waiting for identification. These insects were not observed on other adjacent plant species.
- Some plant pathogens have been observed damaging stems and preventing flowering. Fungi or rusts were collected in four different places in Southern Ukraine. In the region of Odessa, a black rust (*Puccinia* sp.?) was found and collected. That rust covered the entire plant and prevented flowering.
- Hemipterian eggs were found on external parts of stems, but all were parasitized.
- An unidentified insect larva that mines under the cuticle was found at one location (Figure 4).



Photo: René Sforza

Figure 4. Mine of a new and unidentified insect larva under the cuticle of a *Chondrilla juncea* plant.

Bulgaria: Ivanka Lecheva and Anna Stantcheva
(Bulgarian Institution of Higher Agriculture, Plovdiv, Bulgaria)

In the first three months our efforts were focused on organizing the supporting scientific team, getting acquainted with the different aspects of the research on *Chondrilla juncea* worldwide, and conducting preliminary surveys to identify areas with natural populations of *Chondrilla juncea* within Bulgaria. The problem with providing transportation for field work was not resolved; therefore, field surveys were limited in June and July to areas near the town of Plovdiv where fortunately excellent populations of *Chondrilla juncea* were located (Figure 5).



Photo: Anna Stantcheva

Figure 5. Abandoned farmland with scattered *Chondrilla juncea* plants in central Bulgaria.

Later in August with the help of Javid Kashefi a car was obtained and areas along the Black Sea coast, the Danube River, and the mountains of West Bulgaria were searched. During these surveys a possible viral disease, at least three pathogens, ten insects and mites associated with *Chondrilla juncea* were observed. Pictures were taken of most of the sites and samples of the natural enemies were collected whenever possible.

Insects and mites

1. *Cryptocephalinae* sp. (Coleoptera, Chrysomelidae). Leaf-feeding beetle. Relatively abundant. Sample available for confirmation of identification.
2. *Gastroidea polygony*. (Coleoptera, Chrysomelidae). Leaf feeding beetle. Relatively abundant. Sample available for confirmation of identification.
3. *Coptocephala ecopoln*. (Coleoptera, Chrysomelidae). Leaf feeding beetle. Relatively abundant. Sample available for confirmation of identification.
4. Gall midge. (*Cystiphora schmidtii*). Leaf and stem galls. Common in all areas.
5. Unidentified leaf-feeding caterpillar # 1. Collected as larvae. Pupated and emerged in a petri dish. Sample available for identification. Possibly *Schinia cognata*.
6. Unidentified leaf-feeding caterpillar # 2. Collected as larvae. Pupated and emerged in a petri dish. Samples available for identification.

7. Unidentified case-forming, root-feeding caterpillar #3. Collected as larvae, but rearing failed. No samples available for identification, but possibly *Bradyrrhoa gilveolella*.
8. Unidentified flower-and-bud-feeding caterpillar # 4. Rearing failed so no adult insects available for identification.
9. Unidentified beetle larvae (possibly Mordellidae). Mining inside roots and crowns; while abundant and widespread, no adults were obtained for identification (Figure 6).
10. *Aceria chondrillinae*. The gall mite was observed at only two sites and in low abundance.



Photo: René Sforza

Figure 6. Beetle larvae possibly Mordellid mining inside a *Chondrilla juncea* root.

Pathogens

Heavy damage by rusts and powdery mildew was common. Species identification and more in-depth studies are needed. Symptoms matching those of a virus disease reported earlier were also observed on *Chondrilla juncea* in the town of Plovdiv. Heavy attack of a parasitic plant, *Cuscuta* sp., was observed at two sites.

Republic of Georgia: Dominique Coutinot (EBCL-ARS-USDA, Montferrier-sur-Lez, France)

In Georgia, native *Chondrilla juncea* is widespread and is not a weed problem; native natural enemies (insects, mites and pathogens) play a major role in the control of the weed. In 1997 three trap plots were set up around Tbilisi with American varieties of Rush skeletonweed; the impact of natural enemies, especially pathogens, was studied. In 1998 and 1999 isolated pathogen cultures were brought to EBCL and delivered to the pathology unit. These cultures are under identification and evaluation. From 1998 to 2000, observations have been made on the impact of pathogens, insects and mites at the 3 Tbilisi sites with American varieties of skeletonweed. In other localities observations have been made with only native skeleton weeds. However, compiling an inventory list and the biology of natural enemies, especially insects and mites, was the goal for 2001.

In 2001, a Research Materials Exchange Agreement: "Investigations on distribution and biology of *Chondrilla juncea* and its natural enemies in Georgia," was signed between the European Biological Control Laboratory (EBCL) at Montferrier, France, and with the Kanchaveli L. Research Institute of Plant Protection, Tbilisi, Georgia. Studies in Georgia were made in 2001 by a student candidate of Sciences Eliso Beradze, selected by and under the supervision of Prof. Guram Aleksidze, Academician Secretary, Academy of Agricultural Sciences, Tbilisi. In 2001, D. Coutinot, Support Scientist and Quarantine Officer (EBCL-USDA-ARS), visited Georgia in April, June and July. He discussed the agreement and research with Georgian cooperators and visited the sites where potential biocontrol agents were surveyed. (Figure 7.)



Photo: D. Coutinot

Figure 7. Collection site in a wheat field in the Kumisi Lake area of Georgia.

In the Republic of Georgia, natural enemies are controlling skeletonweed *C. juncea*, the most common are the mite *Eriophyes chondrilla*, the gall midge *Cystiphora schmidtii*, the rust *Puccinia chondrillina*, and the fungus *Alternaria* sp. (Figure 8).

The first three organisms have already been introduced as biocontrol agents to North America, but these varieties found in Georgia may prove to be more adapted to the colder interior climates of Idaho.



Photos: D. Coutinot
Figure 8(A).



Figure 8(B).

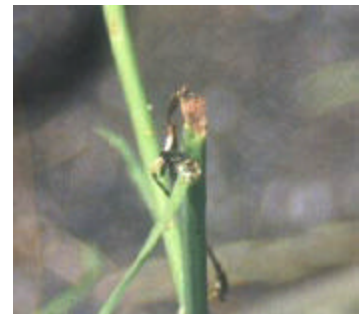


Figure 8(C).

Figure 8(A). A deformed plant heavily attacked by three natural enemies: *Eriophyes chondrillae*, *Cystiphora schmidtii* and *Puccinia chondrillina*.

Figure 8(B). Attack by the mite *Eriophyes chondrillae*

Figure 8(C). The pathogen *Alternaria* sp. has caused a necrosis that killed the growing tips of *Chondrilla juncea* shoots preventing flowering and seeding.

Table 2. Natural enemies of the Rush skeletonweed, *Chondrilla juncea* in the Republic of Georgia

ORDER/Family	Identification or Status	Part attack	Abundance	Location ***	
Homoptera	Aphididae	Uroleucum chondrillae	Stems/tip	Common	Kumisi, Okrokhana, Zahesi,
	Aphididae	ID needed	rosette	Common	Kumisi, Okrokhana, Zahesi
	Coccidae	ID needed	Stems and leaves	Found in July	Funikuliori, Tabakhmela
Diptera	Cecidomyiidae	<i>Cystiphora</i> sp. - in rearing in Georgia	Stems	Common	Funikuliori, Kumisi, Zahesi
	Agromyzidae	<i>Liriomyza</i> "new species"	Rosette	June	Funikuliori
	Agromyzidae	Specimens in rearing in Georgia	Leaves	April-Sept.	Kumisi, Okrokhana
	Agromyzidae	Chromatomya horticola	Roots	April	Kumisi
	Sciaridae	Sciara sp.	Roots	April	Kumisi
Coleoptera	Meloidae	ID needed	Buds	Common	Funikuliori, Kumisi, Zahesi
Lepidoptera	Noctuidae	Specimens in rearing in Georgia	Stems	Common	
Acarina	Eriophyidae	Eriophyes chondrillae	Stems and buds	Common	Funikuliori, Kumisi, Zahesi,
Fungus**	Uredinales	Puccinia chondrillina	Stems and leaves	Common	Funikuliori, Kumisi, Zahesi
	Hyphomycetes	<i>Alternaria</i> sp.	All plant	Common	Funikuliori, Kumisi, Tabakhmela Zahesi
	Sphaeropsidales	<i>ID needed</i>	Stems		Funikuliori
	<i>Unknown</i>	ID needed	Stems		Funikuliori, Kumisi

* More specimens are needed to describe this new species

**All the isolated strains are being cultured at EBCL Pathology Quarantine by T. Widmer EBCL plant pathologist.

***Elevation for those locations is 580 to 800 meters

Some natural enemies are still being reared in the Republic of Georgia. As soon as we get adult specimens, they will be identified and reported.

Uzbekistan and Turkey: Rouhollah Sobhian (EBCL-ARS-USDA, Montferrier-sur-Lez, France)

Turkey was relatively well surveyed during the previous biocontrol program by the Australians and therefore, was not officially included in the 2001 work. Similarly, Uzbekistan, a country lying immediately to the north of Afghanistan, was not included since it was considered to be climatically different from Idaho (too high in elevation, cold, and dry). However, while no Idaho funds were earmarked for work in these countries, my visit to both areas while working on other weeds allowed me to opportunistically survey *Chondrilla* plants when I encountered them. Several possible new insect associates not reported before and believed to have potential as biocontrol agents are discussed below.

1. Root feeding caterpillars were found in eastern Turkey and Uzbekistan. The adults reared for identification appeared to be different from *Bradyrrhoa gilveolella* Treitschke (Pyralidae) and were sent to CABI in London for identification. The insect was first identified as *Bazariasp.*, but later J. Maxen (Identification Service Manager) informed me that they re-examined the insects and they were probably *Bradyrrhoa gilveolella*. A. Kovalev, from St. Petersburg, suggested to send the material to the Museum in St. Petersburg for confirmation because they know the fauna of the region. But this has not yet been done.
2. Larvae of a small midge were found in seed heads both in Turkey and Uzbekistan. Adults were reared and sent for identification in 1997. Both Ray Gagné (SEL, Washington, D.C.) and M. Skuhrová in Czech Republic checked the material and reported that it is a new species belonging to the Genus *Jaapiella* Rübtsaamen, 1915. Since R. Gagné retired, M. Skuhrová is planning a publication that describes the species. Larvae of the insect pupated in soil on the bottom of my rearing jar. Adults were found ovipositing in open flowers of *Chondrilla juncea*. Near Izmir, Turkey, adults were observed mating in a nuptial swarm, and found ovipositing in open flowers of *Chondrilla juncea*. The eggs were inserted between the flower-petals and the bracts. Up to three females were observed ovipositing in the same flower at the same time.
3. The larvae of an unidentified moth was found feeding on the developing seeds in Turkey. The larvae penetrated through a small hole into the seed-heads, consumed all the developing seeds and left it. Then they penetrated to the next seed-pod and after eating all the seeds were searching for another seed-pod. The larvae were light brown and reached up to two centimeters. No adult could be reared for identification.
4. The larvae of an unidentified fly was found in the seed heads in Uzbekistan. Material collected was collected and kept in Tashkent for rearing adults for identification. Rearing is still in progress and no adults have yet emerged.

Apparently there are several species of *Chondrilla* in Uzbekistan. To identify exactly which species I was collecting from, I prepared herbarium samples, which were hand carried to St. Petersburg by A. Kovalev for identification. However, we are still waiting for their reply.

Summary

Access to the areas we wished to visit turned out to be easier than anticipated despite the problems of local civil wars and occasional civil unrest. We also were fortunate to locate local scientists with whom we could cooperate so we could legally work in their countries and particularly to obtain the permits to export the specimen collected to our laboratory in France.

In general the area was found to have a rich fauna of insects, mites, and plant pathogens attacking Rush skeletonweed. While at least five of the natural enemies collected were found in the earlier survey of the Mediterranean area by the Australians, the great majority of the 25+ organisms are newly reported associations. This number is likely to increase this winter as we continue to sort the collections submitted by our cooperators.

While many of these natural enemies are probably generalists—insects capable of feeding on a variety of different plants, many of them are from groups that are highly specialized in their host range, attacking only a single species of plant or a few very closely related species. These are the types that have given us many highly successful weed biocontrol agents in the past.

Many of the identifications included in this report were provided by local scientists and should be considered tentative. One of our major goals for this winter will be to consolidate the collections made by EBCL personnel and our cooperators and send the specimens to recognized world authorities for definitive identification. Once we are satisfied that our collection of natural enemies of Rush skeletonweed is complete, our next goal will be to identify those that have potential as biocontrol agents, then select and prioritize those on which we would concentrate our studies next year, if the necessary funds to continue the programs can be obtained.