RANGE

Operational Field Guide

to the propagation and establishment of the bioagent
Larinus minutus
(Knapweed seedhead weevil)

March 1999

Province of
British Columbia
Ministry
of Forests

Information contained in this Field Guide is comprised of fact and field observations as of March 1999. Site specific experiences may vary.

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Forest Practices Branch
Range Section
Noxious Weed Control Program
British Columbia Ministry of Forests

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1. PURPOSE

This document summarizes information for the biocontrol agent *Larinus minutus* while it was classified as 'primary' and the responsibility of the Forest Practices Branch. The information is a combination of hard facts, observations and best guesses. Intended as a 'field guide' for those unfamiliar with *Larinus minutus,* the summary contains pertinent information for field propagation and establishment of the bioagent as well as a historical background of its introduction into British Columbia.

2. INTRODUCTION

The goal of the Ministry of Forests weed control program is to reduce target weed populations to ecologically and economically acceptable levels and to prevent their encroachment into new areas. The biological control portion of the program includes biocontrol agent screening, propagation, release, collection and monitoring.

Implicit in the use of biocontrol methods is the acknowledgment that weed eradication is not a goal. Rather, bioagent species and host weed species exist in predator-prey relationships with the weeds held at acceptable population levels and the insect populations self-sustaining.

The biocontrol program is cooperative among the Ministry of Forests, Ministry of Agriculture and Food, Agriculture and Agri-Food Canada, Centre for Agriculture and Bioscience International Institute for Biological Control (CAB IIBC) in Switzerland, the BC Cattlemen's Association, Regional Districts, Montana State University, the US Department of Agriculture (USDA) and the Animal and Plant Health Inspection Service (APHIS).

Screening of seedhead-feeding insects was necessary to address the need to decrease knapweed's large seed supply. The combined attack of several seedhead feeders is advantageous. *U. affinis* and *U. quadrifasciata* together has resulted in a decrease in knapweed's seed production from approximately 24 000 to 1500 seeds per square meter (Harris 1991) while in 1991 Cranston reported the decrease to reach as high as 95%. The knapweed at White Lake, B.C. is considered to be at or just beneath its replacement level as a result of the attack of *U. affinis*, *U. quadrifasciata* and the root-feeder *Sphenoptera jugoslavica* (Powell and Meyers 1988, cited by Harris pers. comm.,

July 1990). As of 1990, the decrease to 1500 seeds per square meter had not caused a decrease in plant density and since these insects have limited habitat ranges, the screening and importation of *L. minutus* took place (Groppe 1990c). *L. minutus* and *T. virens* supplement the impact of *U. affinis* but in different habitat niches.

Harris, 1988, in his paper Feeding strategy, coexistence and impact of insects in spotted knapweed seed-heads discussed the compatibility of *L. minutus* (soft achene feeder in achene growth stage) with other seedhead insects released in B.C., namely, *Urophora affinis* (woody gall former in immature buds), *U. quadrifasciata* (ovary gall former in floret growth stage), *Chaetorellia* spp. (ovary feeder in floret growth stage), *Terellia virens* (soft achene feeder in achene growth stage) and *Metzneria paucipunctella* (ripe achene feeder/predator in ripe achene growth stage). Firstly, competition will not occur between species preferring separate habitats. For example, while *T. virens* and *L. minutus* have the same feeding strategy and *T. virens* is reported to seem dominant in competition with *L. minutus* (CAB IIBC 1988), *T. virens* is found in western Europe and *L. minutus* is found in a more eastern distribution in Europe which is drier. It is suspected, therefore, that *L. minutus* will inhabit the more continental parts of the North American knapweed range (Harris 1991). C*haetorellia acrolophi* does well on very dry slopes (Harris pers. comm., Aug. 1990). Also, both *C. acrolophi* and *T. virens* are still to date present in very low numbers and in few sites in B.C. *U. affinis*, on the other hand, has low attack rates in hot dry places and instead prefers moist sites (Harris pers. comm., Aug. 1990). Secondly, attack of the seedheads occurs at different stages of growth so direct competition is often avoided. For example, the woody galls of *U. affinis* have hardened off by the time *L. minutus* attacks the seedhead and so are not damaged (Groppe 1990c). Thirdly, these insects have developed a variety of feeding strategies for survival. Some agents, like *U. affinis* and *T. virens*, tend to colonize dense infestations of knapweed while other agents, like *Chaetorellia* spp. seem specialized in attacking isolated plants. *U. quadrifasciata* disperses well and avoids seedheads with many *U. affinis* but can coexist in lightly attacked *U. affinis* seedheads. *U. affinis* produces a woody gall that serves to protect the larvae from other insects and tends to be the most damaging of the different strategies since the woody galls draw on the resources of the entire plant. *M. paucipunctella* larvae will feed on larvae of its own or other species. Yet, due to the proximity of the different species in the seedhead and their varying developmental stages, the effects are not significant. Again, both *L. minutus* and *T. virens* use the same feeding strategy and oviposit into open flower heads thus avoiding heads which do not open, ie. heads that have been attacked by receptacle feeders and woody gall formers. The extent of these interactions has yet to be investigated in B.C.

3. *LARINUS MINUTUS*

Coleoptera: Curculionidae

Common name: Knapweed seedhead-feeding weevil

BIOLOGY

GENERATIONS PER YEAR: one

ADULT STAGE: Adults are mottled weevils that range in colour from brown to rust or even green depending on age and habitat. Most often they are a mottled rusty brown. They are 5 to 10 mm long (Powell et al. 1994). Adults emerge from overwintering in soil and leaf litter prior to (two to four weeks) the formation of flower buds. In the Kamloops and Nelson areas adults have been seen at the end of May but generally emergence occurs in June with a large increase in numbers at the end of June and beginning of July. The peak in numbers may occur one to two weeks earlier in the Nelson area depending on the weather. Adults feed throughout their five to fourteen week life span. They feed on stems, rosette leaves and florets during the vegetative period of the plant. In particular, the females feed on florets, promoting the development of their ovaries that are immature when they emerge in spring (Groppe 1990a). Adults preferentially feed on flowerheads when they become available and since all plants do not come into flower at the same time, the weevils tend to congregate on small numbers of flowering plants. As more flowers become available the weevils leave other parts of the plants and focus on the flowerheads (Stinson and Marquardt 1987a). The weevils have been observed to copulate when the plants enter the bud stage (about two weeks following emergence (Stinson and Marquardt 1987a)) and continue throughout their life span. The females, which have been feeding in the center of the flowerheads, preferring freshly opened capitula, make holes in the capitulum. Here, they oviposit their eggs, placing three to five eggs in each capitulum. In the field in Greece and Romania oviposition lasts for approximately eight weeks, whereas in the lab, it lasts for up to eleven weeks. Also in laboratory conditions, average fecundity rates were observed at 28 to 130 eggs/female, with an average of 66 eggs (Groppe 1990a) or about 50 eggs/female according to Stinson and Marquardt (1987a).

LARVAL STAGE: Larvae have been observed to hatch three days after eggs are laid when the temperature is 25^{o}C or higher. They feed on the pappus hairs and then work their way down to the achenes. Whole contents of individual seeds are eaten, leaving the seed coat behind, and sometimes part of the receptacle. In the small capitulum of diffuse knapweed, only one *L. minutus* can complete its development. In contrast, in the larger spotted knapweed capitulum several larvae may complete development (Groppe 1990a). Indeed, a single larva consumes approximately 25% of a spotted knapweed seedhead. One hundred percent of the seedhead may be eaten if several larvae are present (Stinson and Marquardt 1987a). There are three instars that take place over a span of four weeks to reach pupation.

PUPAL STAGE: Pupation occurs in late summer. The pupae are white initially and turn brown prior to emergence. They are found within a cocoon attached to the receptacle. The cocoon is made partly with seedcoats (Groppe 1990a).

F1 ADULTS: New adults leave exit holes in the flowerheads upon emergence in late August and September. They are pale when they first emerge and darken shortly after. *L. minutus* are not inactive during the summer, they do not have an aestivation period (Stinson and Marquardt 1987a). After briefly feeding, the weevils move into the leaf litter and soil to overwinter.

DISPERSAL METHOD: The adult weevil walks and flies to reach its mate and host plants.

Photo 1: *Larinus minutus* (adult)

RANGE

Native (European) Distribution

Larinus minutus has been reported throughout the central and western range of spotted knapweed (*Centaurea maculosa*) and diffuse knapweed (*C. diffusa*). It is found in Greece, Romania, Israel, Bulgaria, the Caucasus, the Kazakhstan SSR and the southern part of the European USSR (Groppe 1990b).

L. minutus exists in very dry sites, typically the driest and hottest of the European knapweed range. These areas are generally associated with diffuse knapweed (unknown pers. comm.). It is found at sites in the sand dunes of Hanu Conachi in Romania and in rocky areas in Greece (CAB IIBC, pers. comm., Jan. 1989). *L. minutus* is located in eastern Mediterranean and summer warm continental climates (Stinson and Marquardt 1987b).

See Appendix A for detailed information on Spotted Knapweed (*Centaurea maculosa*) and Diffuse Knapweed (*Centaurea diffusa*).

Predicted North American Distribution

Although the genus is not found in the tropical parts of Asia or in Australia or America, reports indicated that *L. minutus* was expected to establish throughout the majority of the target weeds' North American habitat (Groppe 1990b). In B.C., the weevil is predicted to do best on the dry western prairie and in the dry interior valleys (unknown pers. comm.).

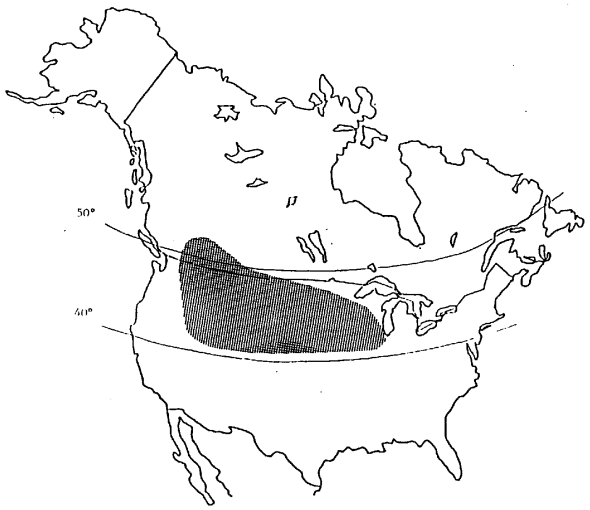

Figure 1: Predicted distribution of *Larinus minutus* in North America (Groppe 1990b)

Subsequent propagation and distribution of *L. minutus* in B.C. has proven this weevil's range to be greater in this province than Groppe's 1990 prediction based on European distributions.

HABITAT

L. minutus attacks both spotted and diffuse knapweeds. The weevils are mostly found in the drier range of the latter weed's habitat. On a 1991 release site in the Kamloops area, the weevils moved off spotted knapweed growing in a slightly moist site with thick smooth brome grass onto diffuse knapweed growing about 23 m away. The diffuse knapweed grows here on a southeast slope that is hotter and drier than the original site and has little competing vegetation covering the coarse soil. The three collection sites in the Kamloops area are infested with diffuse knapweed although the Aberdeen site has both spotted and diffuse knapweed. The weevils have not been noted to show a preference for either species at this site except they are more easily seen on the spotted seedheads. These sites are all Bunchgrass zones ranging from very dry hot to very dry warm subzones with southeast to southwest facing slopes of approximately 10% and an elevation range of 520 to 770 m.

L. minutus has established in knapweed infestations in the Bunchgrass, Ponderosa pine, Interior Douglas-fir and Interior Cedar-Hemlock biogeoclimatic zones, primarily on sites characterized as xeric-hot and xeric-warm to xeric-dry but also including moist-warm sites in the Salmon Arm and Arrow Forest Districts. Evidence of the weevil has also been found in the transition of the Coastal Douglas-fir/Coastal Western Hemlock biogeoclimatic zones. *L. minutus* is to date well adapted to southern B.C. Of the 21 sites monitored in the Kamloops Region in 1998, 86% had positive establishment. Of the 28 sites monitored in the Nelson Region in 1998, 24 sites or 86% had positive establishment.

The weevils seem to prefer well drained, coarse textured soils that lack dense vegetation other than knapweed. Stinson and Marquardt (1987c) report that the weevils appear to like sandy soil around the base of the plants for overwintering. Finer textured soils do become established but observation has shown that at a single site where knapweed is intermittently mixed with other vegetation, the weevils concentrate in the areas where there is less competing vegetation. Photos 2, 3 and 4 show two preferable and one non-preferable examples of ground cover, respectively. Note in photo 2 that even though there is ground cover other than live knapweed plants the debris is not matted and is elevated off the soil. Sites that accumulate heat are preferred, such as south facing slopes or flat, open areas. *L. minutus* prefers sites without shade.

Photo 2: Preferable ground cover at Verigin's Tomb.

Photo 3: Preferable ground cover at Lac Du Bois.

Photo 4: Non-desirable ground cover at Lac Du Bois.

4. HISTORY OF INTRODUCTION

Screening of *L. minutus* occurred between 1985 to 1989. It was approved for release in North America in late 1990 and the first shipments to B.C. occurred in 1991. The Ministry of Forests Propagation Facility (MOFPF) at Kamloops received a total of 4466 adults in 1991. Two thousand and three hundred adult weevils were placed into a propagation tent populated with spotted knapweed. One thousand, nine hundred and fifty-six weevils were released in the Kamloops and Penticton Districts in the Kamloops Forest Region. The F1 generation successfully emerged in the B.C. sites by late summer. Two hundred and ten F1 adults were released in the Arrow District in the Nelson Forest Region later that same year.

In 1992, adults were discovered in the propagation tent and at the Aberdeen and Lac Du Bois field sites in the Kamloops District as well as at Verigin's Tomb in Arrow District. Of the 106 weevils found inside the propagation tent, 33 were put back into the tent while 73 were added to the Aberdeen site.

Since *L. minutus* established it was decided to not import any further weevils from Europe and risk contaminating the population in B.C. with a parasite from the site of origin. *L. minutus'* effectiveness would be decreased if heavily parasitized. Lacking parasites, the weevil would be limited by inter and intra-specific competition (refer to the Introduction) and the quantity of its host plant in favourable habitat.

Adult weevils, with a wide range in size, were found in the propagation tent in May 1995. The variation in size might be explained by intraspecific competition among the weevils or interspecific competition with the *Urophora* spp. Forty weevils from this plot plus an additional 170 from Lac Du Bois were used to establish two more tents of *L. minutus* at the propagation facility. These three tents then, were maintained until 1998 at which time the agent was designated as secondary and the tented plots were dismantled.

Three field collection sites have been discovered in the Kamloops area: Lac Du Bois in 1994, Aberdeen in 1995 and Barnhartvale in 1998. In the Nelson Region, a site near Castlegar named Verigin's tomb has yielded insects for collection since 1995. The low numbers harvested from this last site are due to manpower restrictions, not a shortage in insects. The 1996 collection of *L. minutus* varied more with demand than with availability of weevils. The sites still had abundant weevils present when collection ended so efforts were made to spread the weevils further afield at the Kamloops Region collection sites. In the Nelson Forest Region, collection ended when it was felt sufficient weevils had been removed from the site, all primary release sites had been saturated and priorities were placed elsewhere. Collection has continued to date from these sites. The monitoring of field releases in the province has located additional field sites for the collection of *L. minutus*.

Photo 5: Barnhartvale collection site in Kamloops Forest Region.

Photo 6: Verigin's tomb collection site in Nelson Forest Region.

SUMMARY OF COLLECTIONS AND RELEASES

The following tables summarize collection and redistribution data.

Collection and release summary of *Larinus minutus* in B.C.

	1991	1992	1993	1994	1995	1996	1997	1998
Reared & Collected		106	104	773	4140	14 830[a]	37 451	31 571
Released	4466	106	104	773	4140	14 630	37 451	31 529[b]

[a]Two hundred *L. minutus* perished in captivity.

[b]Forty-two *L. minutus* perished in captivity.

Propagation Facility

Insects Reared in MOF Propagation Facility Tents

	1991	1992	1993	1994	1995	1996	1997	1998
Insects Received for Propagation Tents (# tents)	2300 (1)	33 (1)	(1)	(1)	168 (3)	(3)	(3)	(3)
Reared at MOFPF		106	104					204

Kamloops Forest Region

Field collections

#Insects

SITE	1994	1995	1996	1997	1998
Aberdeen		200	4600	29 565	27 842
Barnhartvale					400
Lac Du Bois	773	3770	7030	3586	1000
TOTAL	773	3970	11 630	33 151	29 242

Field releases by district

#Insects(#Releases)

DISTRICT	1991	1992	1993	1994	1995
Clearwater					200 (2)
Kamloops	1486 (3)	73 (1)	104 (1)		400 (4)
Lillooet					200 (2)
Merritt				300 (3)	200 (2)
Penticton	470 (1)				200 (2)
Salmon Arm					200 (2)
Vernon					400 (4)
TOTAL	1956 (4)	73 (1)	104 (1)	300 (3)	1800 (18)

DISTRICT	1996	1997	1998
12		1000 (10)	
Kamloops	1700 (17)	8400 (84)	3400 (16)
Lillooet	1200 (12)	2000 (20)	1400 (7)
Merritt	1730 (17)	2500 (25)	2000 (10)
Penticton	1600 (16)	1000 (10)	2000 (10)
Salmon Arm		200 (2)	600 (3)
Vernon	1500 (15)	2000(20)	1400 (7)
TOTAL	7730 (77)	17 051 (171)	10 800 (53)

Nelson Forest Region

Field collections

#Insects

SITE	1995	1996	1997	1998
Verigin's tomb	170	3200	4300	2125
TOTAL				

Field releases by district

#Insects(#Releases)

DISTRICT	1991	1992	1993	1994	1995	1996	1997	1998
Arrow	210 (1)				772 (5)	2500 (11)	4700 (22)	4125 (16)
Boundary				473 (4)	400 (2)	1800 (8)	9600 (27)	6400 (13)
Cranbrook							1500 (6)	5600 (12)
Golden								
Invermere								
Kootenay Lake						1000 (5)	3700 (14)	1600 (3)
Revelstoke								
TOTAL				473 (4)	1172 (7)	5300 (24)	20 400 (69)	17 725

Other Forest Regions

Field releases by region

#Insects(#Releases)

REGION	1995	1996	1997	1998
Cariboo	200 (2)			2400 (12)
Quesnel				
Williams Lake				
100MileHouse				
Prince Rupert	200 (2)			
Vancouver	200 (2)		100 (1)	
Campbell River				
Chilliwack		500 (5)		
Duncan				
Port Alberni				
Sunshine Coast				
TOTAL	600 (6)	500 (5)	100 (1)	2400 (12)

Other Release Destinations

Distribution numbers recorded in this table are restricted to the releases given directly to municipalities, native Bands and a contractor by this office. Total field distribution numbers for these groups will be higher since MOF regions and districts maintain relations with these and other groups and share a portion of their releases with them.

#Insects (#Releases)

	1995	1996	1997	1998
Native Bands				204 (1)
City of Kamloops		600 (6)		
Highland Valley Copper Mine reclamation (JS Jones)				400 (2)
MOE Ecological Reserves	400 (1)			
Ministry of Transportation & Highways		300 (3)		
Thompson Nicola Regional District		200 (2)		
TOTAL	400 (1)	1100 (11)		604 (3)

5. REDISTRIBUTION

Redistribution of agents is a critical part of the biocontrol program. To ensure distribution throughout *L. minutus*' potential provincial range, personnel must be able to recollect from field releases and make releases into new sites.

FIELD COLLECTION

What and where to collect

Field collection of *L. minutus* involves sweeping or visually locating adult weevils in a knapweed infestation and hand picking them from plants. Weather conditions have some effect on locating the weevils. The weevils are more plentiful when it is warmer. Yet, much of the monitoring has been done with sweep nets so the general location of the weevils on the plants when comparing warm and cool weather is not known. A seemingly more important factor in collection than daily temperature variations is the position in the weevils' cycle. Prior to flower formation, the weevils are located on the stems, rosette leaves and florets. Following flower formation, and particularly when ovipositing begins, the weevils are found on the flowerheads.

The following are suggested Collection Site Criteria*:

- Sites should be between 0.5 and 1.5 ha in size.
- The average estimated knapweed density should be greater than 5 plants/metre^2.
- Topographies of successful sites have varied. All have been able to accumulate heat units. A hot climate is needed.
- Course to uniformly fine-textured soils with little ground cover and mainly infested with knapweed are preferred.
- Sites receiving cold air drainage may be poor choices, especially if they are relatively flat, allowing cold air to pond.
- Preferably on Crown land with easy access within 100 km of Regional or District Offices.
- Sites should be easily traversed for collection.

*These criteria also need to be kept in mind when establishing future collection sites (current release sites) to ensure a future population of *L. minutus* in the field. See discussion under Field Release.

How to collect

L. minutus weevils are swept from knapweed plants and separated from other debris by either aspirating the weevils out of the sweep net or putting the net contents in a refrigerator to cool the insects down and then separating out the weevils, or a combination of both methods. Aspirating the weevils from the net while in the field

works best as seeds, plant parts, other insects, etc. can be left at the site and the weevils are not jeopardized by storing them with spiders and other predators. It is easier for an experienced person to obtain a 'clean' container when it is hot since the weevils are more active and climb out of the debris found in the bottom of the net more readily. Depending on care taken, experience and luck, the collection may be free of any contaminants listed above, but, it is good practice to still clean the collection after placing the weevils in a refrigerator. It has been observed that approximately two to three weeks after the numbers of weevils greatly increase (~ mid-July), the weevils begin to tenaciously cling to the plants and are difficult to collect with sweep nets. This action may coincide with ovipositing. At this time, it is more efficient, and less harmful to the insects and their host plants, to hand pick with bare hands or aspirate the weevils as they cling to the flowerheads. Aspirating directly off the plants is not as effective when the weevils are really clinging. Plants need to be approached slowly when using these methods. If the weevils notice movement or the plant is shaken, they drop to the ground and lie motionless on the soil where they are well camouflaged. The weevils are placed into containers with a mesh opening in the lid. Mesh screen on the storage lids is critical to allow ventilation and to prevent a build up of condensation that can drown the insects. Depending on experience the task can be accomplished several ways: tipping the plant over the collection containers (this works best with large numbers of weevils per plant); tipping the plant over your hand; or plucking the weevil off the plant from below to avoid the weevil dropping to the ground. When it is hot and the weevils active, all methods that require constantly opening the lids of the containers are not effective as the weevils will fly back out. It is best in this case to combine hand picking and aspirating. Avoid pulling a clinging weevil off a plant. They need to be gently persuaded to avoid injury. What ever the method used, beginner collectors may find it easier to crouch down at the plant level as it takes experience to develop 'an eye' for the weevils on the plants.

Knapweed is placed in the containers to provide feed and a place for the weevils to cling (it also helps to avoid weevils crawling over each other, fighting and injuring one another). Do not use plant material that is too lush as it produces a lot of condensation inside the containers that may drown the insects. Do not include seed heads in the container so as to avoid seed spread. Also, females may oviposit into these heads, thus, some eggs will be lost. Containers are kept cool and out of direct sunlight in a portable cooler with ice packs wrapped first in plastic bags and then in paper towels to absorb any condensation. Avoid contact between the containers and the wrapped ice packs as condensation still may occur.

In the lab, the numbers of weevils are confirmed. *L. minutus* can be sexed, looking for characteristics similar to those used for sexing *Cyphocleonus achates*, yet, a strong hand lens or a microscope is needed and the weevils must be cooled sufficiently to slow them down, so generally this is not done. If the weevils are to be kept for any length of time before releasing, which usually does not happen, they

should be stored in quantities of 100/container which should be cleaned every two days with warm water and fresh knapweed added. When shipped, quantities of 200/container are used due to their small size.

L. minutus may also be collected by harvesting seedheads in late August, just prior to the emergence of the next generation of adults. Enough stem should be left attached to the seedheads so several may be tied together in small bundles which are then tied to individual plants at the release site. Do not pull up plants when collecting if the intent is to maintain the infestation as a collection site. In order to obtain an estimate of the survival rate of the weevils, open the heads of several plants and calculate an average number of weevils per plant, percentage of seedheads infested or number of weevils per seedhead if harvesting spotted knapweed. This method is more time consuming and there is a high potential to spread seeds from site to site.

When to collect

Time of Year

The collection period is between late June to late July with peak collection generally occurring in the second and third weeks of July in the Kamloops Region. Peak collection may be up to two weeks earlier in the Nelson Region, depending on the weather. *L. minutus* can be found until the end of July, however, it is felt that collection needs to be terminated earlier than this (at the end of the third week in July) to allow for establishment on release sites. Eggs require time to hatch, larvae and pupae must develop and adults must be able to emerge and feed for a short time before entering the leaf litter prior to frost.

The dependence of emergence and hence collection on the accumulation of sufficient heat units has not been observed or investigated. The literature does, however, describe the dependence of larvae hatching on a minimum temperature of 25^{o}C.

It is unknown whether male *L. minutus* emerge before the females from the soil/leaf litter in the spring to create an uneven sex ratio of newly emerged weevils. It is possible to identify male and *female L. minutus* weevils but it requires either a strong hand lens or a microscope and a method of slowing the weevils down, such as putting them in a cooler or refrigerator, to investigate the shape of their abdomens. This is not practical for field work. This fact, combined with the time period needed for the females' ovaries to mature, leads to the practice of leaving the first observed weevils in the field until the population increases and then collecting without determining the sex of the weevils.

Time of Day

Sunny days have been found to be better for collecting than cooler days. Groppe, 1990, reports that in Greece, the weevils are more active in the morning and late afternoon. They can be easily seen on the plants at these times. Yet, even though Groppe reported the weevils to hide from the “extremely hot midday hours” by remaining under flowerheads, leaves and generally in the lower regions of the plants, observations and collection in the Kamloops Region appears to peak around 3:00 pm when the sun is indeed very hot. It could be that in their country of origin, the extreme temperatures are greater than in B.C.

It is recommended not to sweep for the weevils in or following rain. Caution should be taken when hand-picking the weevils following a rain shower. The weevils can either drown in small amounts of water or their elytra (wing covers) can be damaged when they get stuck to the canvas net or the wall of the containers.

Additional considerations

The CAB IIBC September 30, 1994 Quarterly Report on Weeds notes that continual yearly collection of weevils from a site will cause a significant decline in their numbers. Refraining from collecting for a year or more may be necessary to allow the population to recover. Depending on operational goals, it may be worthwhile to rotate collection sites from year to year to sustain their insect populations, but keeping in mind the potential for the knapweed infestation to decline below the level that provides for a good collection site.

SHIPPING

Collected insects are shipped to new release sites in 1 litre bulk food containers. Traditionally, one container (100 insects) was used for each release site unless larger releases were designated. To ensure population establishment and a faster increase in population numbers, releases of a minimum of 200 insects are recommended. When releases of 100 were used it was found that evidence of establishment took two years to find and collection, although small numbers, could not occur until the third year (if the site proved collectable). When releases of 200 have been used, establishment evidence was often found in the first year following release and small numbers of weevils could be collected in the second year. The containers are well ventilated and contain sufficient seedhead-free knapweed to feed the weevils during transport. Containers are packaged into carefully sealed boxes to avoid insect escape during shipment. Cold packs are wrapped first in plastic bags to contain the majority of condensation and then in newspaper or paper toweling to prevent further condensation from building up inside the containers and damaging or drowning the insects. These are used to keep the weevils cool and reduce their activity if they are traveling any distance. Avoid contact between the containers and the wrapped ice packs by using packaging material as condensation

still may occur. The agents are shipped quickly via courier or bus to release locations.

FIELD RELEASE

Potential release sites

A potential release site needs to meet certain criteria to ensure success and longevity. It must meet Ministry needs from a program and logistic standpoint i.e. travel distance, land tenure, and accessibility. It must also be conducive to agent survival and establishment.

Below are suggested release site criteria and considerations. They are based on observations of past sites which have been successful.

Criteria

- Release sites should be large enough to support a viable insect population with potential for natural dispersal.
- Soils may be coarse to fine and uniformly textured, but preferably well-drained.
- Exposed soil with little litter and competing vegetation is particularly desirable, especially when agent numbers are low.
- The average estimated knapweed density should be greater than 5 plants per metre^2. (This is only a guideline. Soil conditions and lack of litter/soil cover seem to be more critical than plant spacings.)
- Sites should not be shaded.
- Topographies of successful sites have varied. All have been able to accumulate heat units. A hot to warm climate is needed.

Considerations

- Releases at any elevation should be attempted. The highest recorded elevation where establishment has been successful is 850 m.
- Sites receiving cold air drainage may be poor choices, especially if they are relatively flat, allowing cold air to pond.

The following are suggested steps to take when making a site selection:

1. Plan release site locations prior to requesting agents. Release sites should be pre-selected the fall or spring prior to release of agents. This avoids ‘drop and dash’ releases and promotes overall weed management planning.
2. Determine tenure and stability of land management. Preferably a site will be located on Crown Land with MOF mandated as the steward and have a cooperative tenure holder. Other suitable locations may be land under the

jurisdiction of other agencies with the goals of controlling weeds and establishing/maintaining working relationships. Release sites might be located in or close to relevant municipalities with the goal of future cost effective collection sites. An example is a municipal water reservoir which is long term and most activity, particularly herbicide spraying, is prohibited.

3. Make sure the site will not be disturbed after release. Crown control of the site is preferred with future management known. Discuss future development plans for the site wherever it is located.
4. Check previous release records and maps to ensure no prior release of the agent has been made at a potential site. An unofficial rule is that a distance of 1 km constitutes a separate release.
5. Monitor plants at potential site to ensure the agent is not already present through natural dispersal.
6. Check the immediate vicinity of the proposed release site for ant hills and wasp nests to minimize predation.
7. Mark selected release sites with a stake so that it may be relocated to monitor insect progress and weed population decline.

Insect release

Before any weevils can be released there is preparatory work that needs to be completed at the site. Make sure that all paperwork, photo's, site maps, measurements, etc. are completed before opening any lids, otherwise people will be treading on insects.

In the past, each release consisted of approximately 100 adults transported in a bulk food container, yet 200 adults in one or two containers is recommended.

Agents are released primarily by Ministry of Forests regional and district personnel and occasionally staff from the Ministry of Agriculture and Food. Other people who may receive insects include Native Bands on reserve and private lands, private citizens, Ministry of Transportation and Highways and University staff.

The following are suggested steps to take when making an insect release:

1. Mark the release site with a semi-permanent stake to assist relocation efforts for follow-up agent establishment and weed impact monitoring.
2. Fill out the 'Biological Control Release Record' (see Appendix C) that is shipped with the weevils **accurately** and **completely**. Information on the forms is fundamental to further analysis of the program. One completed copy of the Release Record is kept in the District office and one is returned to the MOF Regional office. The forms are then collated to create a provincial database traditionally administered by the Ministry of Agriculture and Food.
3. Create accurate site maps complete with permanent tie points. This is essential for future monitoring of the release site.

4. Take photographs. They have proven to be a useful tool to both relocate the release site and to provide an ocular comparison of the site over time. A suggested method and form (EM-9) is outlined in the Habitat Monitoring Manual.
5. Gently release the weevils (once the paper work is completed) at one location by the stake. They will disperse themselves from this initial release point. It is more difficult for insects to propagate if they are spread over a large area.

Additional considerations

Initial releases for the season should be made in similar latitudes or further south than collection sites to ensure temperatures are conducive to agent establishment. As northern release sites warm, they can receive insects. Once northerly temperatures begin to drop, releases should be directed south again to provide for the longest possible establishment season.

If more than one collection site is available, it is preferable to redistribute weevils into habitats similar to those they are acclimatized to.

If agent establishment at a release site is uncertain or the knapweed infestation is particularly large, re-release of agents may need to be considered. Before re-releasing at a site the Region/District plan needs to be reviewed, i.e. Can agents be spared for re-release at a site that may or may not be conducive to the agents' survival when they could be placed at a new site?

6. MONITORING

Monitoring of field sites can be carried out to determine:

1. whether the agent has established at the release site
2. the density of agents per plant or area
3. how far the agent has spread from the release point
4. the agent's preferred habitat and current range
5. areas that are unsuitable to the agent
6. any effects the agent has had on the weed population
7. potential collection sites
8. if collecting from the site has had any effects on plant or agent populations
9. agent life cycle information i.e. emergence dates, effects of weather.

Depending on the type of information being sought the monitoring technique will vary. Reconnaissance methods can be used to assess parameters such as site suitability, presence or absence of agents, dates of emergence etc. A suggested monitoring form ('Release Site Monitoring Form') detailing information to collect at each site can be found in the Appendix C. Many details listed on this form can be found on the original release form. Some will not change over time and need not be duplicated if the information is already recorded, while other details, particularly the plant's dispersal description, may change.

A more rigorous method is needed for quantifiable information on insect and plant populations. It is suggested that this type of monitoring be planned at selected sites as dictated by the constraints of program planning, time and budget.

A suggested transect method is as follows:

1. Find the release stake or from the description on the release form, the closest position to the release stake. Mark a starting point.
2. Determine four directions from the starting point for running the transects. If cardinal directions cannot be used, determine 4 non-cardinal right angles. Pace out 25 m in each direction.
3. Pace out three parallel transects at least 2 metres apart: two 33 m long and one 34 m long if the site does not allow for the above transect design.
4. Pace out two 50 m parallel lines lying side by side or staggered (as can be accommodated by the site). The intent is to monitor a uniform number of plants randomly, if the site does not allow for either of the above transect designs
5. Make two sweeps of the plants per metre with a sweep net (a sweep is defined as a single pendulum swing in one direction). This works out to be approximately one sweep per step. Check the sweep net every five metres, count and record the number of weevils.

A form for this method (Biocontrol Agent Monitoring Form) is found in Appendix C. The average number of agents may be calculated per metre and recorded or the count for every five metres may be included on the form. Either way, the percent attack will be equivalent.

AGENTS

Sites can be monitored for the presence of adults from June to the end of July and again at the end of August into September for the F1 generation. Weevils can be observed on the plants in the spring, either on the stems, rosettes or florets prior to flower formation and later on the flowerheads. Characteristic 'shot-holes', the result of adults emerging from the seedheads, can be observed in late August and into the remainder of the fall or even the following spring.

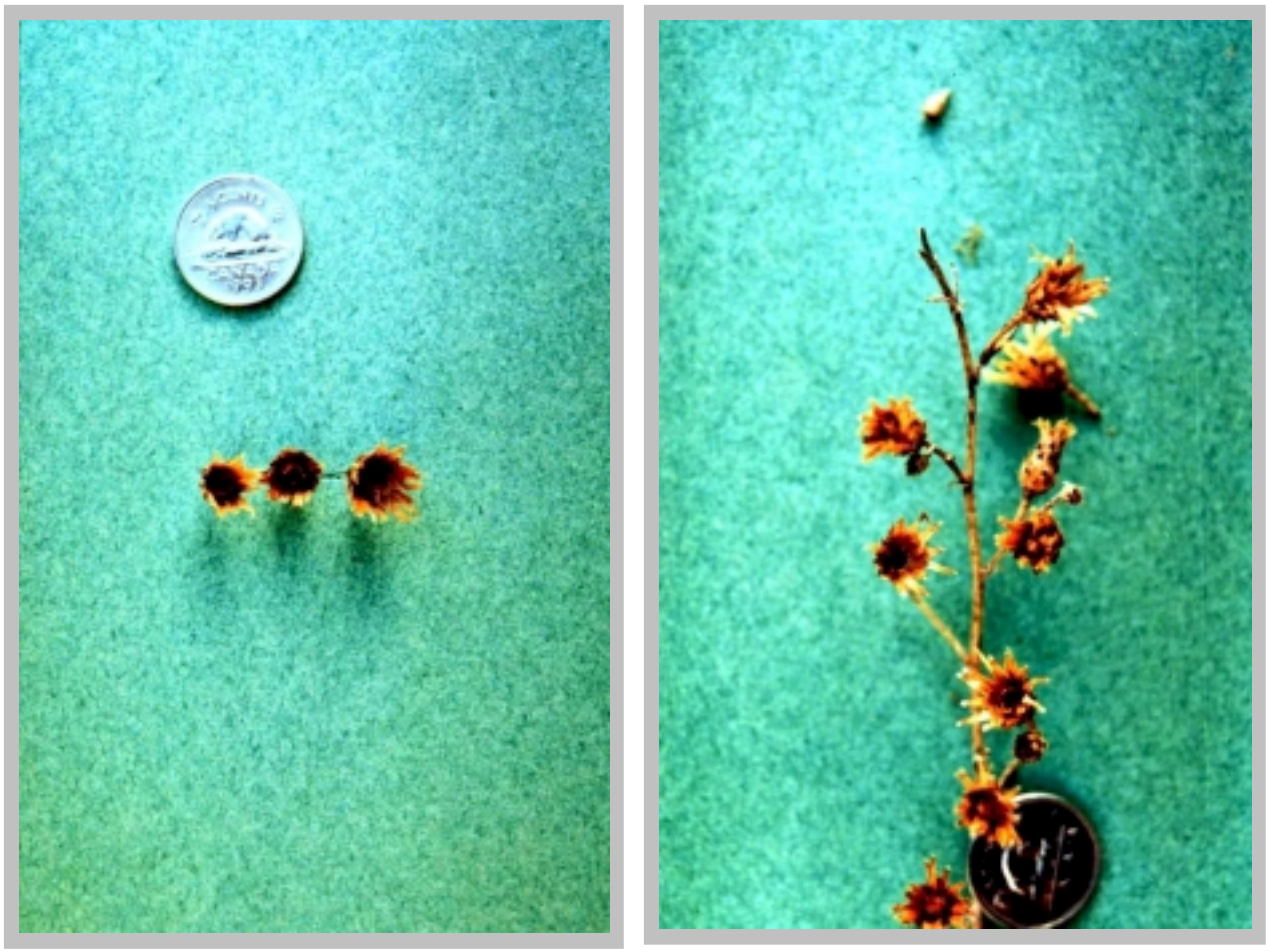

Photo 7: *L. minutus* emergence holes in knapweed seedheads.

PLANTS

A method needs to be developed for measuring responses of the host weed population. Useful parameters to monitor would be: height, density, biomass production, seed production, cover, and frequency.

RESULTS

To date, monitoring has found that *L. minutus* can establish in the Bunchgrass, Ponderosa pine, Interior Douglas-fir, Interior Cedar-Hemlock and Coastal Douglas-fir/Coastal Western Hemlock transition biogeoclimatic zones. Specific site parameters are more difficult to summarize and analyze. However some generalities have been noted previously in the suggested criteria for site collection and release.

With the differences in latitude, future monitoring information may be best analyzed on a Region or District level. Dispersal information is best presented in map form.

APPENDICES

Appendix A - HOST WEEDS

Spotted Knapweed *Centaurea maculosa* Lam.

The plant

- A short-lived perennial, introduced with seed grain (apparently alfalfa) from southeastern Europe. First Canadian collection made at Victoria, B.C. in 1893. Spreads by seed. Mature plants are 0.3-1.5 m tall, with long fibrous tap roots. Stems are somewhat hairy when young and highly branched. The basal and lower stem leaves are pinnately lobed; the upper leaves have smooth margins. Young leaves have a fine coating of hairs. Flowers are 1-1.5 cm long, pink to purple (occasionally white); bracts of the flower head are black tipped, giving the head a spotted appearance. Flowering occurs from July through to September. Prolific seeds are black or brown, 3 mm long, and topped with bristles up to half the length of the seed. The plant contains volatile oils with a distinctive smell and an extremely bitter taste.

Habitat

- Found throughout British Columbia, but primarily at lower to mid-elevations of the southern interior including the entire Kootenays, Okanagan, Thompson-Nicola, Cariboo-Chilcotin, Fraser Valley, Vancouver Island and several Gulf Islands. Occurs in isolated pockets elsewhere in the province (Prince Rupert Forest Region and Peace River area). Estimated to cover about 60,000 ha of semi-arid rangeland in B.C. (Muir 1986) and has potential to occupy 1.1 million ha.

Growing conditions

- Grows in a wide range of environmental conditions, though mostly in grasslands and open forests of the Bunchgrass, Ponderosa Pine, Interior Douglas-fir and Interior Cedar-Hemlock, as well as the Coastal Western Hemlock and Coastal Douglas-fir biogeoclimatic zones. Local infestations occur in the Montane Spruce and Englemann Spruce-Subalpine Fir zones. A rapid colonizer of disturbed soils, it can also displace native vegetation in undisturbed sites.

Photo 8: Spotted knapweed (*Centaurea maculosa*)

Diffuse Knapweed *Centaurea diffusa* Lam.

The plant

- A biennial to short-lived perennial, spreading primarily by seed. Stems are 60-90 cm in height with many branches. Leaves are 5-20 cm long, hairy, and highly divided. Leaves alternate from the stem, with basal leaves forming a rosette. Flower heads are numerous, urn-shaped, and covered with small, narrow bracts ending in sharp, rigid spines. Flowers are white or occasionally pink or purple. Prolific seeds are black to dark brown, 3 mm long, and lack a developed fringe of hairs (pappus). Diffuse knapweed contains volatile oils with a distinctive smell and extremely bitter taste.

Habitat

- Widely distributed throughout British Columbia, though primarily in the Kootenays, Thompson-Nicola, Okanagan, Kettle River, and Fraser Canyon areas of the southern interior. Occurs in pockets and at lower abundance in the Cariboo-Chilcotin.

Growing conditions

- Occurs over a wide range of ecological types, though it tends to dominate dry valley bottoms in the Bunchgrass zone and transition areas of Ponderosa Pine, and Interior Douglas-fir biogeoclimatic zones. A rapid colonizer of disturbed soils, it can also invade and displace native vegetation in undisturbed areas.

Photo 9: Diffuse knapweed (*Centaurea diffusa*)

Appendix B - LITERATURE CITED

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Appendix C - MONITORING FORMS

BIOLOGICAL CONTROL RELEASE RECORD

SITE NUMBER: ___ ___ ___ ___ /___ ___ /___ ___

BIOAGENT: _____ /_____ **WEED SPECIES:** ___ ___ ___ ___ / ___

SOURCE: _____ **STAGE:** ADULT PUPA LARVA EGG OTHER

COLLECTION:__ __ / __ __ / __ __ **RELEASE:**__ __ / __ __ / __ __ **TIME:**__ __: __ __
Y M D Y M D

 # RELEASED: _____

JURISDICTION: ___ ___ **RELEASED BY:** _____

DISTRICT: _____ **RANGE UNIT NAME:** _____

PRIVATE LAND: **Owner:** _____ **Phone:** ( ) ___ ___ ___ - ___ ___ ___

ADDRESS: _____

LOCATION: _____

BCGS MAP: __ __ __ __ __ __ __ **UTM:** ZONE | EASTING | NORTHING **DATUM** _____

WEED DENSITY: < 1 plant/m^2 ☐ 2-5 plants/m^2 ☐ 6-10 plants/m^2 ☐ < 10 plants/m^2 ☐

SIZE OF INFESTATION: < 100 m^2 ☐ 101-400 m^2 ☐ 401-2500 m^2 ☐ 2501-5000 m^2 ☐

5001-10000 m^2 ☐ > 1ha. ☐

WEED DISTRIBUTION: Continuous Stand ☐ Scattered Patches: ☐

SLOPE %:___ **ASPECT^0:**___ **ELEVATION m:**___ **BIOGEO UNIT:** __ __ __/__ __ __ __ __/
zone/subzone - variant/site series

MONITORING for ESTABLISHMENT

Date Established					Photo:	
Y	M	D	Yes	No	Yes	No
__	__	__	☐	☐	☐	☐
__	__	__	☐	☐	☐	☐
__	__	__	☐	☐	☐	☐

SKETCH MAP (Indicate North)

COMMENTS:

RELEASE SITE MONITORING FORM

DATE: **AGENT:**

SITE NUMBER: **SITE NAME:**

MAP NO.:

WEED DENSITY:

<1 plant/m^2	____	2-5 plants/m^2	____
6-10 plants/m^2	____	>10 plants/m^2	____

SIZE OF INFESTATION:

<100m^2	____	100-400m^2	____
400-2500m^2	____	2500-5000m^2	____
5000-10000m^2	____	>1 ha	____

WEED DISTRIBUTION:

Continuous Stand	____	Scattered Patches	____

ACCESS TO SITE:

Easy	____	Difficult	____

Describe if necessary.

SITE TOPOGRAPHY:

Flat	____	Bowl Shaped	____
Forest Openings	____	Close to River/Lake	____
Terraced	____	Hillside	____
Other (describe)	____		

TRAVERSABILITY OF SITE:

Easy	____	Difficult	____

Describe if necessary.

SOIL DESCRIPTION:

Moss covered	____	Gravel	____
Clay	____	Silt	____
Compact	____	Loose	____
Sandy	____	Other (describe):	____

SLOPE (%): ____ ASPECT (^{o}): ____ ELEVATION (m): ____

BIOGEOCLIMATIC CLASSIFICATION: ____

DISTANCE FROM KAMLOOPS (km): ____

LAND OWNER:

RECOMMENDATION:

COMMENTS:

BIOCONTROL AGENT MONITORING FORM

SITE NUMBER:		DATE: (YR/M/D)	
AGENT:		LOCATION:	
RELEASE DATE:		TARGET PLANT:	

METERS	NORTH	SOUTH	EAST	WEST
1				
2				
3				
4				
5				
6				
7				
8				
9				
10				
11				
12				
13				
14				
15				
16				
17				
18				
19				
20				
21				
22				
23				
24				
25				
MAX DIST.				

PERCENT ATTACK: _____

COMMENTS: _____