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*Biological
Control*

BIOLOGY AND BIOLOGICAL CONTROL OF KNAPWEEDS



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CHAPTER 1: INTRODUCTION

Overview

Knapweeds belong to the genus *Centaurea* in the family Asteraceae. This genus can be found on all continents of the Northern Hemisphere, but is particularly species-rich in the Middle East. Taxonomic classification within the *Centaurea* is still uncertain; this polyphyletic group includes the starthistles and cornflowers as well as the knapweeds. All three groups are among the 28 *Centaurea* species and three additional hybrids currently present in North America. Twenty-one of these species are knapweeds.

Unlike cornflowers, knapweeds typically have grayish-green and often rough or hairy leaves sometimes deeply divided into irregular lobes. Some knapweed species are similar to starthistles, in that bracts subtending flower heads can end in stiff spines; however, unlike all starthistles, the spines of these knapweeds are never longer than the bracts. Only knapweeds are discussed in this manual.

All knapweeds (and all *Centaurea*) are exotic in North America. They had arrived on this continent by the early 1900s with settlers from Europe and Asia, typically as seed in contaminated hay. Together, these knapweeds form a large complex of invasive species that currently infest more than 5 million acres throughout the United States and Canada (Figure 1). They have adapted to a wide range of habitats and environmental conditions. Though knapweeds are some of the most common rangeland pests in the West, they are frequently found invading pastures and fields in the Midwest and eastern states. They are responsible for millions of dollars of damage annually in reduced agricultural yields and lowered forage value. Knapweeds displace native vegetation, thus negatively impacting wildlife and threatening the delicate ecological balance within many habitats. Allelopathic compounds have been isolated from knapweed plant material in North America, though the role of allelopathy in some knapweed plant interactions is under debate.



Figure 1 Infestations of spotted knapweed (L.L. Berry, www.bugwood.org).

Of the 21 knapweed and three hybrid species in North America, six species are especially problematic. This manual addresses the biological control of these six species (Table 1). Among the most troublesome are spotted, diffuse, and squarrose knapweeds. The lesser-known meadow, brown, and black knapweeds are closely related to the others and are included in this manual because they share similar biology and some of the same biological control agents. Please note that the problematic range species known as Russian knapweed (*Acroptilon repens*) is not included

Table 1 Exotic knapweeds discussed in this manual.

| SCIENTIFIC NAME | COMMON NAME | DURATION |
|-----------------------------|-------------------------|--------------------|
| <i>Centaurea diffusa</i> | Diffuse knapweed | Annual/perennial |
| <i>Centaurea jacea</i> | Brown knapweed | Perennial |
| <i>Centaurea nigra</i> | Black knapweed | Perennial |
| <i>Centaurea nigrescens</i> | Tyrol (Meadow) knapweed | Perennial |
| <i>Centaurea stoebe</i> | Spotted knapweed | Biennial/perennial |
| <i>Centaurea virgata</i> | Squarrose knapweed | Perennial |

in this manual. This species has been assigned to a different genus, and is sufficiently distant in its relation to the *Centaurea* knapweeds as to have its own complex of biological control agents.

Herbicides have traditionally been the most popular method for treating knapweed infestations; however, herbicides are not always the appropriate management tool due to their high costs, potential for environmental contamination, plant resistance, non-target effects, and prohibition in environmentally sensitive areas. Non-chemical weed control methods are becoming increasingly important treatment alternatives. The most effective knapweed management programs to date incorporate multiple control methods, including the introduction of biological control agents. This manual addresses the biological control of six invasive knapweed species within the larger context of integrated knapweed management.

Classical biological control of weeds

Most invasive plants in North America are not native; they arrived with immigrants, through commerce, or by accident from different parts of the world. These non-native plants are generally introduced without their natural enemies, the complex of organisms that feed on the plants in their native range. The lack of natural enemies is one reason non-native plant species become invasive pests when introduced in areas outside of their native range.

Biological control (also called “biocontrol”) of weeds is the deliberate use of living organisms to limit the abundance of a target weed. In this manual, “biological control” refers to “classical biological control,” which reunites host-specific natural enemies from the weed’s native range with the target weed. Biological control agents may feed on a weed’s flowers, seeds, roots, foliage, and/or stems. This damage may kill the weed outright, reduce its vigor and reproductive capability, or facilitate secondary infection from pathogens—all of which reduce the weed’s ability to compete with other plants.

Natural enemies used in classical biological control of weeds include a variety of organisms, such as insects, mites, nematodes, and fungi. In the US, most weed biological control agents are plant-feeding insects, of which beetles, flies, and moths are among the most common. To be considered for release in the US, biological control agents must feed and develop only on the target weed and, in some cases, on a few closely related plant species. A potential biological control agent’s life cycle should be closely matched, or synchronized, with the development of the target weed. If properly synchronized, foliage-feeding insects would be in the feeding stage when the weeds are actively growing, and seed-feeding insects would be in the feeding stage when the plant is actively developing seeds.

The most effective biological control agents tend to be those that damage the most vulnerable or most problematic and persistent parts of the host plant. Root- and stem-feeding biological control agents are usually more effective against perennial plants that primarily spread by

root buds. Alternatively, flower- and seed-feeding biological control agents are typically more useful against annual or biennial species that only spread by seeds. There are advantages and disadvantages to biological control of weeds by any agent (Table 2).

Table 2 Some advantages and disadvantages of classical biological control as a management tool.

| ADVANTAGES | DISADVANTAGES |
|--|---|
| Target specificity | Protracted time until impact is likely |
| Continuous action | Unpredictable level of control |
| Long-term cost-effective | Uncertain “non-target” effects in the ecosystem |
| Gradual in effect | Irreversible |
| Generally environmentally benign | Not all exotic weeds are appropriate targets |
| Self dispersing, even into difficult terrain | Will not work on every weed in every setting |

Host specificity is a crucial point of consideration for a natural enemy to be released as a biological control agent. Host specificity is the extent to which a biological control agent can survive only on the target weed. These tests are necessary in order to ensure that the biological control agents are effective and that they will damage only the target weed. Potential biological control agents often undergo more than five years of rigorous testing to ensure that host specificity requirements are met.

The United States Department of Agriculture’s Animal and Plant Health Inspection Service – Plant Protection and Quarantine (USDA-APHIS-PPQ) is the federal agency responsible for authorizing the importation of biological control agents into the US. The Canadian Food Inspection Agency (CFIA) serves the same role in Canada. Federal laws and regulations are in place to minimize the risks to native plant and animal communities associated with introduction of exotic organisms to manage weeds. The Technical Advisory Group (TAG) for Biological Control Agents of Weeds is an expert committee with representatives from regulatory agencies, federal land management offices, and environmental protection agencies from the US and representatives from Canada and Mexico. TAG reviews all petitions to import new biological control agents into the US and makes recommendations to USDA-APHIS-PPQ about the safety and potential impact of prospective biological control agents. Weed biological control researchers work closely with USDA-APHIS-PPQ and TAG to accurately assess the environmental safety of potential weed biological control agents and programs. The Canadian counterpart to TAG is the Biological Control Review Committee (BCRC) which uses NAPPO RSMP NO.7 as their review/petition requirement.

In addition, each state in the US has its own approval process to permit field release of weed biological control agents.

Code of best practices for classical biological control of weeds

Biological control practitioners have adopted the International Code of Best Practices for Biological Control of Weeds. The Code was developed in 1999 by delegates and participants to the 10th International Symposium for Biological Control of Weeds and is intended to help reduce the potential for negative impacts from biological control of noxious weed activities. In following the Code, practitioners agree to voluntarily restrict biological control activities to those most likely to result in success.

INTERNATIONAL CODE OF BEST PRACTICES FOR CLASSICAL BIOLOGICAL CONTROL OF WEEDS¹

1. Ensure that the target weed's potential impact justifies release of non-endemic agents
2. Obtain multi-agency approval for target
3. Select agents with potential to control target
4. Release safe and approved agents
5. Ensure that only the intended agent is released
6. Use appropriate protocols for release and documentation
7. Monitor impact on the target
8. Stop releases of ineffective agents or when control is achieved
9. Monitor impacts on potential non-targets
10. Encourage assessment of changes in plant and animal communities
11. Monitor interaction among agents
12. Communicate results to public

¹ Ratified July 9, 1999, by the delegates to the 10th International Symposium on Biological Control of Weeds, Bozeman, MT.

Although weed biological control is an effective and important management tool, it does not work in all cases and is not expected to eradicate or completely remove the target weed. Ideally, biological control should be integrated with other chemical, mechanical, and/or cultural methods of weed control to improve overall weed control success.

Biological control of knapweeds

Biological control of knapweeds has long been considered inadequate to reduce infestations of these species throughout North America. However, the most recent evidence indicates that some northern and western knapweed infestations are finally coming under the control of biological control agents. These observations are discussed in greater detail in Chapter 3, along with the biology of all knapweed biological control insects.

Developing a biological control program for invasive knapweeds has a number of unique challenges. Knapweeds are a diverse group of plants exhibiting different life cycles—annual, biennial, and perennial (Table 1, page 2)—and occurring in many habitats. Despite the challenges, the knapweed biological control program is one of the oldest, on-going classical biological control programs in the US and Canada.

One of the key characteristics of a successful biological control agent is host specificity. Although there are no native *Centaurea* in North America, there are closely related native species in different genera. In order for a knapweed biological control agent to be approved for release in the United States or Canada, biological control researchers must first demonstrate that the agent

will not feed and develop on either native species, or any other species of economic importance.

Seed-feeding insects have long been a popular choice for biological control programs because, as a group, they often have relatively high levels of host specificity. Currently, there are eight seed-feeding insects established for the biological control of knapweeds in North America. The first insect introduced against knapweed was the knapweed banded gall fly, *Urophora affinis*, in 1973 (Figure 2).

The knapweed species described in this manual are not often seed-limited, so they are not always vulnerable to seed-feeding biological control agents. To increase the chances of biological control success, researchers identified biological control agents from other feeding guilds. As of 2010, 13 insect species had been introduced to North America for knapweed biological control.

Two fungi and one mite species attack knapweeds; however, because none are approved for distribution in North America, only the insects are described in this manual.



Figure 2 Adult *Urophora affinis* (Linda Parsons & Mark Schwarzländer, University of Idaho).

Integrated weed management

In order to reach the land manager's noxious-weed management objectives, the program usually stretches over many years and incorporates several control methods and activities, including chemical (herbicides), mechanical control, cultural treatments, and biological control. Called Integrated Weed Management (IWM), this approach relies on realistic management objectives, accurate weed identification and mapping, and post-treatment monitoring to answer the question: Are current weed-management activities meeting the weed-management objectives?

Land managers choose weed control methods that will enable them to achieve their goal in the most cost-effective manner. No single control method will enable managers to meet their knapweed management objectives in all environments. The control method(s) employed in a weed-management strategy will depend on the size and location of the infested area and specific management goals (e.g., eradication vs. weed density reduction). Small patches of knapweed may be eliminated with a persistent herbicide program, but large areas will often require the application of additional control methods. A combination of control methods, such as biological control with supplemental cultural practices or chemical controls, consistently applied through time, is usually necessary to attain management objectives for knapweed, especially when it infests large acreages.

Is biological control of knapweeds right for you?

When biological control is successful, biological control agents behave like a pest species of the target weed: they increase in abundance until they suppress the target weed. As local weed populations are reduced, biological control agent populations decline with them due to starvation and/or dispersal.

Some factors to be aware of before starting biological control activities include the following:

- The efficacy of biological control agents cannot be guaranteed.
- Biological control will not work every time in every situation.
- Biological control will not eradicate the weed.
- By itself, biological control may not provide the desired level of control.
- It might take years before you see any results.

We recommend you develop an IWM program in which biological control is just one of several weed control methods. Here are some questions you should ask before you implement a biological control program.

Is my goal to eradicate the weed or reduce its abundance?

Biological control does not eradicate target weeds, so it is not a good fit with an eradication goal. However, depending on the target weed, biological control agent used, and land use, biological control can be effective at reducing the abundance of a target weed.

How soon do I need results: this season, in one to two seasons, or within five to ten years?

Biological control takes time to work, so another weed management method may be a better choice if you need to show immediate results. Generally, it can take one to three years after release to confirm that biological control agents are established at a site, and even longer for agents to cause significant impacts to the target weed. In some weed infestations, more than five years may be needed for biological control to reach its weed-management potential.

What resources can I devote to my weed problem?

If you have a small weed problem (small infested area), weed control methods such as herbicides or hand pulling, followed by annual monitoring for re-growth, may be most effective. These intensive control methods may allow you to achieve rapid control and prevent the weed from infesting more area. However, if an invasive weed is well-established over a large area, and resources are limited, biological control may be your most economical weed control option.

Is the weed the problem or a symptom of the problem?

Invasive plant infestations often occur where desirable plant communities have been disturbed. If the disturbance continues without restoration of a desirable, resilient plant community, biological control may not solve your weed problems.

The ideal biological control program

1. is based on an understanding of weed, habitat, and land use conditions;
2. is part of a broader integrated weed management program;
3. has considered all weed control methods and determined that biological control is the best option based on available resources and weed management objectives;
4. has realistic goals and timetables and includes adequate monitoring.

About this manual

This manual provides information on knapweeds and each of their biological control agents. It also presents guidelines to establish and manage biological control agents as part of a knapweed-management program.

Chapter 1: Introduction Information on knapweeds and their biological control.

Chapter 2: Getting to Know Knapweeds Provides detailed descriptions of taxonomy, growth characteristics and features, habitat, and occurrence of six invasive knapweeds in the US. It also presents a list of related *Centaurea* species present in the US and Canada.

Chapter 3: Biology of Knapweed Biological Control Agents Describes the biological control agents of knapweed, including information on each agent's native range, original source of releases in North America, part of plant attacked, life cycle, description, destructive stages, host specificity, known non-target effects, habitat preferences, and availability. This chapter is particularly useful for identifying biological control agents in the field.

Chapter 4: Elements of a Knapweed Biological Control Program Includes detailed information and guidelines on how to plan, implement, monitor, and evaluate an effective knapweed biological control program. Included are guidelines and methods for

- selecting and preparing release sites;
- collecting, handling, transporting, shipping, and releasing biological control agents;
- monitoring biological control agents and vegetation.

Chapter 5: An Integrated Knapweed Management Program Discusses the role of biological control in the context of integrated knapweed management.

Glossary Technical terms frequently used by those involved in knapweed biological control.

Literature Cited Lists the publications cited directly in this manual.

The **Appendices** are:

- I. Troubleshooting Guide: When Things Go Wrong
- II. PPQ Form 526: Interstate Transport Permit
- III. Sample Biological Control Agent Release Form
- IV. Knapweed Standardized Impact Monitoring Protocol (SIMP) Instructions and Monitoring Forms
- V. General Biological Control Agent Monitoring Form
- VI. Knapweed Qualitative Monitoring Form
- VII. Knapweed Biocontrol-Associated Vegetation Monitoring

CHAPTER 2: GETTING TO KNOW KNAPWEEDS

Introduction

Knapweeds in North America are part of the genus *Centaurea* and the sunflower family (Asteraceae). This family is large and very diverse and includes dandelions, sunflowers, and daisies. The genus is also large and diverse, encompassing nearly 500 species, including knapweeds, starthistles, and cornflowers. The majority of *Centaurea* are native to Eurasia and the Mediterranean; none are native to North America. There are 28 species and three hybrids of *Centaurea* currently established in the U.S. and/or Canada, 21 of which are knapweeds. Six of the most problematic knapweed species are described in detail within this manual. Under favorable conditions, all six are capable of forming large infestations and having serious environmental impacts.

Leaf shape, bract shape, capitula size, and flower color are important traits for distinguishing knapweed species. The taxonomic key (page 12) can be used to differentiate the six knapweed species covered in this manual. Following the key, each of the six knapweed species are described in greater detail. Taxonomic classification and distribution information was taken from the USDA PLANTS database. (PLANTS is a collaborative effort of the USDA NRCS National Plant Data Center, the USDA NRCS Information Technology Center, and many other partners. See www.plants.usda.gov.)

Knapweed development

Knapweeds may be annuals, biennials, or perennials, meaning they take 1, 2, or more years, respectively, to complete their life cycle (Figure 3). The life cycle of all knapweeds begins as a seedling that develops into a rosette with 5 to 12 leaves. The basal leaves of knapweeds are characterized by being grayish-green (usually), having a rough or finely hairy texture, often being either deeply divided or irregularly lobed, and withering at maturity. Biennial and perennial knapweeds remain as rosettes their first year; annuals germinate and flower within the same growing season.

Like all other members of their family, knapweeds produce flower heads (also called seed heads or capitula) that are an aggregation of many individual flowers (Figure 4). These flowers, called florets, are clustered together and attached to a receptacle. Both the receptacle and florets are enclosed by modified leaves called involucre bracts. Flower head size and bract shape are important diagnostic features for distinguishing knapweed species.

Each floret produces one seed (achene) through the summer. Some species produce seeds with a tuft of whitish hairs (pappus) on one end, similar to those on seeds of dandelions. Seeds are 4–5 mm long and dispersed primarily by wind. The seeds of some knapweed species may remain viable for 20 years, though the majority of seeds germinate within one year. Flowering stems are usually branched and have one flower head on each stem. Allelopathic chemicals have been isolated from North American knapweeds, though their role in plant competition is currently under debate.



Figure 3 Generalized knapweed life stages: a. seedling, b. rosette, c. bolting, d. bud, e. flowering, f. senescence. Photos: a & f. Ohio State Weed Lab Archive, Ohio State Univ.; b. Steve Dewey, Utah State Univ.; c. K. George Beck & James Sebastian, Colorado State Univ.; e. Michael Shephard, Forest Service. All www.bugwood.org.

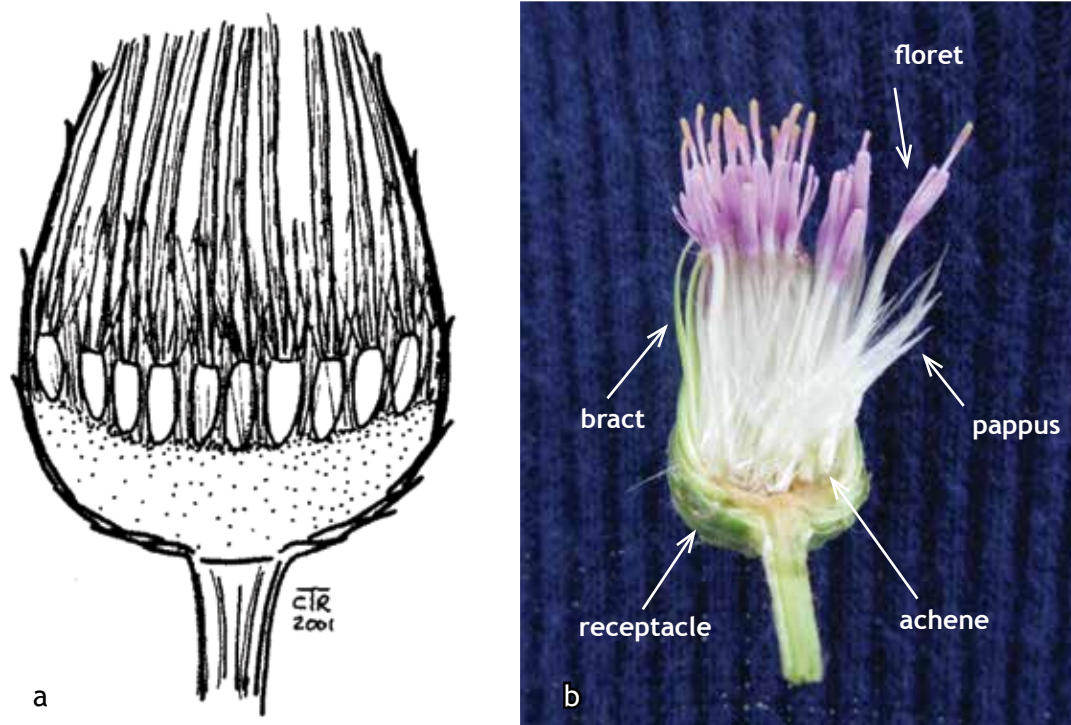


Figure 4 a. Capitulum drawing (Cindy Roche), b. capitulum photo diagram (Rachel Winston, MIA Consulting).

Insects used in knapweed biological control inflict damage to the plant in two places: the seed head and the root. The plant is damaged by the larvae of these insects feeding on and often destroying seed head and root tissue. Only the adult seed head weevils eat foliage. Beyond this, adult knapweed biological control agents generally don't damage the plant.

Seed-feeding biological control agents attack the plant at specific stages of development: some attack the plant early, in the bud stage, and others attack later, when plants are in early to full bloom. Larvae eat and destroy seeds and receptacle tissue, limiting the reproductive output of knapweeds.

Root-boring biological control agents can attack the plant as soon as the root is large enough for the insect to feed. The root is composed of two key tissues: the root cortex and the central vascular tissue (Figure 5). Both tissues are nutritious; the cortex tissue stores nutrients, and the vascular tissue contains the channels in which nutrients and water move up and down the plant. Plants attacked by root-feeding biological control agents are often stunted and produce fewer seeds, if they are not killed outright.

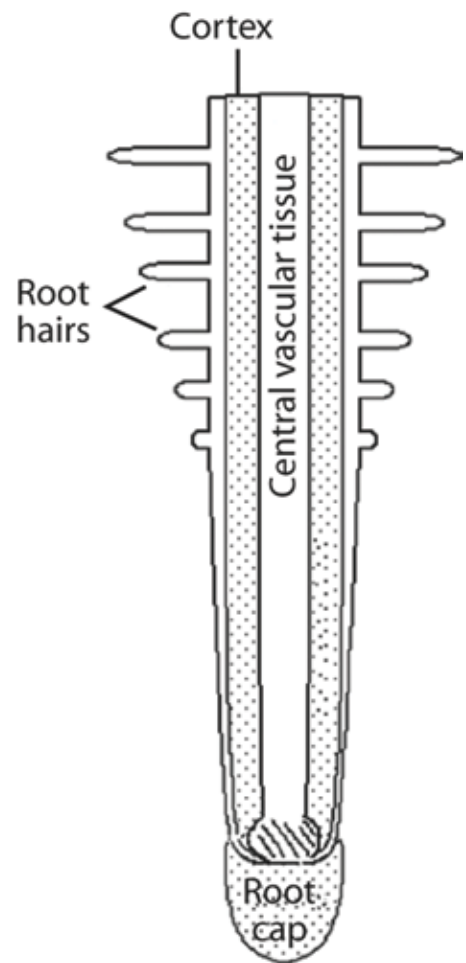


Figure 5 Key tissues in the knapweed root (<http://pc65.frontier.osrhe.edu>).

Simple key to six knapweed species commonly found in Wesern North America

Adapted from Roche and Roche 1993

Illustrations by Cindy Roche

- 1a) Bracts surrounding flower head are spine-tipped
 2a) Central, terminal bract strongly bent backwards

Squarrose knapweed
 (*Centaurea virgata*)



- 2b) Central, terminal bract not strongly curved
 Diffuse knapweed
 (*Centaurea diffusa*)



- 1b) Flower heads without spine-tipped bracts
 3a) Edge of bract is comb-like fringe
 4a) Fringes of bracts short, drawn out and rigid, bract with brown triangular tip

Spotted knapweed
 (*Centaurea stoebe*)



- 4b) Fringes on bracts as long or longer than the width of the bract, not rigid
 5a) Fringe on bract black

Black knapweed
 (*Centaurea nigra*)



- 5b) Fringe on bract tan to brown
 Meadow knapweed
 (*Centaurea pratensis*)



- 3b) Bracts without comb-like fringe, having a brown, papery, translucent tip

Brown knapweed
 (*Centaurea jacea*)



Taxonomic classification of knapweeds

All knapweeds described in this manual fall within the same Subtribe of the Asteraceae (Table 3). Knapweeds differ only in their classification at the level of Species.

Table 3 Taxonomic classification of knapweeds (USDA-PLANTS)

| | | |
|---------------|------------------|------------------|
| KINGDOM | Plantae | Plants |
| SUBKINGDOM | Tracheobionta | Vascular plants |
| SUPERDIVISION | Spermatophyta | Seed plants |
| DIVISION | Magnoliophyta | Flowering plants |
| CLASS | Magnoliopsida | Dicotyledons |
| SUBCLASS | Asteridae | |
| ORDER | Asterales | |
| FAMILY | Asteraceae | Aster family |
| TRIBE | Cynareae | |
| SUBTRIBE | Centaureinae | |
| GENUS | <i>Centaurea</i> | Knapweed |

Species descriptions of knapweeds

The following pages contain descriptions of the six knapweeds covered in this manual. Species are listed in their order of abundance and/or severity in North America: spotted, diffuse, squarrose, meadow, black, and brown knapweed. All six knapweeds are compared to each other in Table 4 on page 26. To aid in proper knapweed identification as you peruse the plant pages, please observe that the first three species are more similar to each other morphologically, and have the typical knapweed leaves of being gray-green and deeply divided or lobed when basal. The remaining three species are also more similar to each other morphologically, and do not have very grayish leaves, nor are leaves divided.

The photos were chosen based on how clearly they illustrate the characteristics of the plants. Consequently, where photos of sparsely branched plants were available, those were used. Please note that, under favorable conditions, knapweeds are also capable of growing with a highly branched and bushy form (Figure 6).



Figure 6 Bushy specimens of knapweed: a. spotted knapweed; b. diffuse knapweed; c. squarrose knapweed. a., c., Steve Dewey, Utah State University; b. Richard Old, www.xidservices.com. Both www.bugwood.org.

Spotted knapweed

Scientific name

Centaurea stoebe L. ssp. *micranthos* (Gugler) Hayek; *Centaurea stoebe* L.

Synonyms

Bushy knapweed; *Acosta maculosa* auct. non (Lam.) Holub; *Centaurea biebersteinii* DC; *Centaurea maculosa* auct. non Lam.

Description

At a glance

A bushy, winter-hardy, biennial or perennial forb (Figure 7a). This upright plant is often found in dense infestations. Plants grow from 1–3½ ft (30 cm–1 m) in height and are supported by a deep taproot. Rosette leaves are gray-green, woolly, and deeply divided. Stem leaves are pinnately divided, becoming smaller and less divided towards the tips of multiple woolly, hairy stems. Mid-plant branches are topped by few to many pink or lavender flower heads producing numerous tiny, bristle-topped seeds. Flowering occurs from June to October. Receptacles are covered by shortly fringed bracts with dark brown tips, thus the common name “spotted.” Dead tops remain in winter with new sprouts appearing in spring. Reproduction is by seed only.

Roots

Spotted knapweed has a deep and stout taproot and does not reproduce vegetatively.

Leaves

Basal leaves are gray-green and hairy to somewhat woolly. They are usually 4–8 inches (10–20 cm) long and deeply divided into elliptic lobes that can appear like leaflets along a wide whitish to purplish midvein (Figure 7c). Stem leaves decrease in size above the mid-stem and are alternate. Lobes become more slender and fewer on upper leaves. Leaves near flower heads are undivided and have no lobes.



Stems

The stems are upright, stiff, usually branched, and often occur in multi-stemmed clumps reaching heights of 3½ ft (>1 m). Stems are roundish in cross-section, covered in dense woolly hairs, and have ridges that extend downward from leaf bases. Upper stems are wiry and slender with many alternate branches that

Figure 7 Spotted knapweed: a. plant (James H. Miller, Forest Service); b. capitulum (Michael Shephard, Forest Service); c. leaf (John Cardina, The Ohio State University). All www.bugwood.org.

end in flower heads. Small plants usually have an unbranched stem and one flower head, whereas older plants can have hundreds of flower-tipped branches.

Flowers

Each branch is topped by a flower bud covered with overlapping rows of shortly fringed (not spiny), green bracts with triangular, dark brown tips (Figure 7b). The black tips give the plant its “spotted” appearance and common name. Bracts turn straw-colored and spread upon maturation and are topped by 20 to 50 pink or purple colored florets. The urn-shaped receptacles are typically 0.25–0.6 inch (6–15 mm) wide and 0.4–0.8 inch (10–20 mm) long. All florets are hermaphroditic, having both male and female reproductive parts. Flowers usually appear from early summer through autumn.

Seeds

Seeds are 0.1 inch (2 ½ mm) long, oval, black or brown with pale, vertical lines. Each seed has a short, bristly pappus about half the length of the seed. Plants can produce 10 to 1,200 seeds, some of which can remain dormant for many years.

Habitat and ecology

Spotted knapweed grows in a wide range of habitats. It rapidly colonizes roadsides and disturbed lands, especially dry sites, then invades adjacent undisturbed grasslands and open forests. It spreads only by seeds, which are equipped for dispersal by wind, water, livestock, wildlife, and human activity and which can remain viable in the soil for many years. Seeds germinate throughout the growing season. Heads persist on the stiff stems through the winter, eventually breaking off when new rosette growth appears the following spring. Both diploid and tetraploid spotted knapweed types are known. Specialized chemicals give this weed a distinctive smell.

Distribution and noxious weed status

Spotted knapweed was introduced in contaminated hay from Europe and Asia as early as 1890. It has the widest distribution of all the knapweed species in North America. It presently occurs in 46 states, six Canadian provinces, and one Canadian territory (Figure 8). It is listed as noxious in 14 states (AZ, CA, CO, ID, MI, MN, MT, NE, NV, NM, ND, OR, UT, WY) and four Canadian provinces (AB, BC, MB, SK), is prohibited in MA and CT, and is a regulated non-native plant species in South Dakota.

Approved biological control agents

All 13 insects described in this manual will attack this species. Some prefer other knapweed species over spotted. The most effective established agents preferring spotted are *Agapeta zoegana*, *Cyphocleonus achates*, *Larinus* spp., *Sphenoptera jugoslavica*, and *Urophora* spp.



Figure 8 States and provinces where spotted knapweed is established (USDA PLANTS).

Diffuse knapweed

Scientific name

Centaurea diffusa Lam.

Synonyms

White knapweed; tumble knapweed; *Acosta diffusa* (Lam.) Soják

Description

At a glance

An annual, biennial, or short-lived perennial winter-hardy forb that usually grows as a biennial (Figure 9a). Stems are 1–3½ ft (30 cm–1 m) tall, with many, spreading branches, giving the plant a ball-shaped appearance and tumble-weed mobility when broken. Rosette leaves are deeply divided, gray-green, and covered in small hairs. Stem leaves are stalkless, and get smaller and less divided higher up the stem. Flower heads with white (sometimes pink or lavender) florets occur at the ends of branches and produce numerous bristle-topped seeds. Flowering occurs from June through October. Receptacle bracts are edged with a fringe of spines. This plant has a taproot, and reproduces only by seed.

Roots

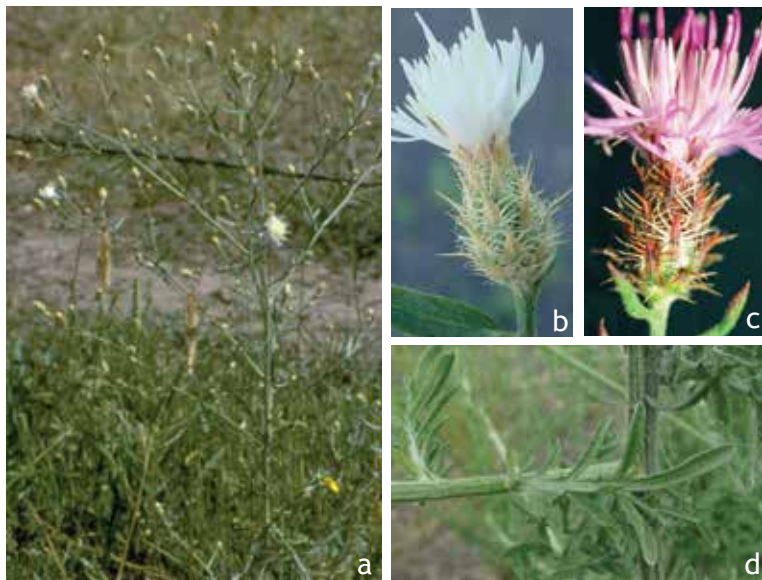
Diffuse knapweed has a deep and fibrous taproot and does not reproduce vegetatively.

Leaves

Basal leaves are gray-green and hairy to somewhat woolly. They are usually 4–8 inches (10–20 cm) long and deeply divided into elliptic or linear lobes (Figure 9c). Stem leaves decrease in size above the mid-stem, are alternate, and aren't stalked. Lobes become more slender and fewer on upper leaves; leaves near flower heads have no lobes.

Stems

At maturity, there is usually one main upright stem. Stems are 1–3½ ft (30 cm–1 m), but most



often 2 ft (60 cm) tall, with numerous, spreading branches that give the plant a ball-shaped appearance and tumble-weed mobility when broken. The stems are roundish in cross-section, are covered in hairs, and have additional short, stiff hairs on the angles. Upper stems are wiry and slender with many alternate branches

Figure 9 Diffuse knapweed: a. plant; b., c., capitula with color variation; d. stem leaves; e. seed. a., d. K. George Beck, Colorado State University; b. Richard Old, www.xidservices.com; c. Steve Dewey, Utah State University. All www.bugwood.org.

that end in flower heads. Small plants usually have an unbranched stem and one flower head while older plants can have hundreds of flower-tipped branches.

Flowers

Each branch is topped by one to a few flower buds covered with overlapping rows of small, narrow, yellow-green bracts fringed with sharp, rigid spines. The terminal spine is distinctly longer than the lateral, spreading spines. Bracts spread upon maturation and are topped by 10 to 20 white florets (may be pink or purple, Figure 9b & c). The urn-shaped receptacle is typically 0.25–0.4 inch (7–10 mm) wide and 0.25–0.6 inch (7–15 mm) long. All florets are hermaphroditic, having both male and female reproductive parts. Flowers appear from early summer through autumn.

Seeds

Seeds are dark brown achenes, 0.1 inch (2 to 3 mm) long, and have a plume of bristle-like hairs that vary from scale-like to $\frac{1}{8}$ the length of the seed. Plants can produce anywhere from 5 to 900 seeds, some of which can remain dormant for many years.

Habitat and ecology

Diffuse knapweed grows in a wide range of habitats. It rapidly colonizes roadsides and disturbed lands, especially dry sites. It prefers habitats in the shrub-steppe zones and dry, open forests. Unlike other knapweeds, the flower heads of diffuse do not open to shed seeds. Instead, seeds are shed primarily after the stiff central stalk breaks and the mature plants tumble in the wind. Secondarily, seeds can be spread by vehicles, animals, and people. Seeds germinate throughout the growing season, and can remain viable for many years. A diploid, fertile hybrid between diffuse knapweed and spotted knapweed has been identified and is known as *C. x psammogena*. Specialized chemicals give this weed a distinctive smell and an extremely bitter taste.

Distribution and noxious weed status

Diffuse knapweed is native to the eastern Mediterranean and western Asia and was first recorded in North America in 1907 in an alfalfa field. It presently occurs in 24 states, five Canadian provinces, and one Canadian territory (Figure 10). It is listed as noxious in 13 states (AZ, CA, CO, ID, MT, NE, NV, NM, ND, OR, UT, WA, WY) and four Canadian provinces (AB, BC, MB, SK) and is a regulated non-native plant species in South Dakota.

Approved biological control agents

All 13 insects described in this manual will attack this species. Some prefer other knapweed species over diffuse. The most effective established agents preferring diffuse are *Bangasternus fausti*, *Larinus* spp., *Sphenoptera jugoslavica*, and *Urophora* spp.



Figure 10 States and provinces where diffuse knapweed is established (USDA PLANTS).

Squarrose knapweed

Scientific name

Centaurea virgata Lam. ssp. *squarrosa* (Willd.) Gugler

Synonyms

Centaurea squarrosa Willd.; *Centaurea triumfettii* auct. non All.; *Centaurea virgata* Lam.

Description

At a glance

A bushy and long-lived perennial forb (Figure 11a). Stems are 1–3½ ft (30 cm–1 m) tall, with numerous, spreading branches. Rosette leaves are deeply divided, gray-green, and covered in small hairs. Stem leaves are stalkless, getting smaller and less divided higher up the stem. Flower heads with pink or purple florets occur singly or in pairs at the ends of branches and produce numerous bristle-topped seeds. Flowering occurs from June through September. Receptacle bracts are edged with a fringe of spines with the terminal spine curving backward. This plant has a taproot, and reproduces only by seed.

Roots

Squarrose knapweed has a very deep taproot and does not reproduce vegetatively.

Leaves

Basal leaves are gray-green and hairy to somewhat woolly. They can be 4–8 inches (10–20 cm) long and are deeply divided into lobes (Figure 11c). Stem leaves decrease in size above the mid-stem, are alternate, and aren't stalked. Lobes become more slender and fewer on upper leaves; leaves near flower heads have no lobes and often appear bract-like.

Stems

Numerous stems arise from the root crown at maturity. Stems are 1–3½ ft (30 cm–1 m) tall, with many spreading branches. The stems are roundish in cross-section, are covered in



hairs, and sometimes appear winged. Upper stems are wiry and slender with many alternate branches that end in flower heads. Small plants usually have one unbranched stem and one flower head while older plants can have hundreds of flower-tipped branches.

Figure 11 Squarrose knapweed: a. plant; b. capitulum; c. leaves. a., b., Steve Dewey, Utah State University; c. Joseph M. DiTomaso, University of California-Davis. All www.bugwood.org.

Flowers

Each branch is topped by one or two flower buds covered with overlapping rows of small, narrow, yellow-green bracts fringed with sharp, rigid spines. The terminal spine is distinctly longer than the lateral, spreading spines and is strongly curved backwards. Bracts spread upon maturation and are topped by 4 to 8 pink or purple florets (Figure 11b & c). The urn-shaped receptacle is typically 0.12–0.25 inch (3–6 mm) wide and 0.33–0.6 inch (8–15 mm) long. All florets are hermaphroditic, having both male and female reproductive parts. Flowers usually appear from early to late summer. The heads are deciduous, falling off the stems after the seeds mature.

Seeds

Seeds are golden to dark brown with pale vertical stripes and a bristle-like, white pappus. The pappus hairs can be up to $\frac{1}{2}$ the length of the seed. Only 3 to 4 seeds are produced per capitulum, each measuring about 0.12 inch (3 mm) in length. Plants can produce anywhere from 3 to a few hundred seeds, some of which can remain viable for many years.

Habitat and ecology

Squarrose knapweed grows in a wide range of habitats in North America, but is specially adapted to withstand harsh, dry climates better than any other knapweed species. It rapidly colonizes roadsides and disturbed lands, and is frequently found in deserts and shrub-steppe. Under favorable conditions, this species may remain as a rosette for multiple years before developing flowering stems. Seeds disperse when whole heads break off from the stem and get lodged in the hair and fur of animals due to the spiny bract fringes, much like the actions of cocklebur and burdock. They can also be spread by vehicles, animals, and people. Seeds germinate throughout the growing season, but can remain viable for many years.

Distribution and noxious weed status

Squarrose knapweed was introduced to North America by the early 1950s from Southwest Asia and the Middle East, presumably in wool, either on sheep or on woolen products. It presently occurs in six states (Figure 12). It is also listed as noxious in six states (AZ, CA, CO, NV, OR, UT).

Approved biological control agents

Seven biological control agents described in this manual will attack squarrose knapweed. These include *Bangasternus fausti*, *Chatorellia acrolophi*, *Larinus minutus*, *Sphenoptera jugoslavica*, and *Urophora* spp. The seventh insect, *Pterolonche inspersa*, is not currently established in the U.S.



Figure 12 States and provinces where squarrose knapweed is established (USDA PLANTS).

Meadow knapweed

Scientific name

Centaurea × *moncktonii* C.E. Britton

Synonyms

Protean knapweed; Bemis grass; *Centaurea pratensis* Thuill.; *Centaurea nigrescens* Willd.; *Centaurea debeauxii* Gren. & Godr. ssp. *thuillieri* Dostál; *Centaurea dubia* Suter ssp. *vochinensis* (Bernh. ex Rchb.) Hayek; *Centaurea vochinensis* Bernh. ex Rchb.

Description

At a glance

A fertile hybrid of black and brown knapweed, meadow knapweed may closely resemble either parent species, or may combine the characteristics randomly. Because chromosome numbers (44) are compatible, the hybrids can backcross with either parent to form highly variable populations. It is typically a perennial growing from a taproot or small cluster of roots beneath its woody crown. Stems grow upright from 1–3½ ft (30 cm–1 m) and branch near their middle (Figure 13a). Rosette leaves are often bright green, undivided, and tapering at both ends. Stem leaves are lance-shaped and stalkless, becoming smaller towards the stem tips. Broadly-oval capitula occur singly on branch tips and contain 20–40 rose-purple (sometimes white) florets. Flowering occurs from July to October. Receptacle bracts are light to dark brown, with deeply fringed margins. This plant reproduces primarily by seed, and the seed sometimes lacks pappus.

Roots

Meadow knapweed seedlings are taprooted while mature plants develop a cluster of fleshy roots beneath the woody crown.

Leaves

Unlike spotted, diffuse, and squarrose knapweed, meadow knapweed leaves are not deeply divided and are a brighter green with only subtle gray tones. Basal leaves grow up



to 6 inches (15 cm) long and ¼ inch (3 cm) wide having entire margins or tiny lobes or teeth. Basal leaves also taper at both ends with the broadest part above the middle of the leaf (Figure 13c). Stem leaves are alternate and become progressively smaller up the stem; uppermost leaves are linear, almost bract-like.

Figure 13 Meadow knapweed: a. plant; b. capitulum; c. stem leaves. All Eric Coombs, Oregon Department of Agriculture.

Stems

The stems are upright, but less so (and less stiff) than other knapweed species. Stems are usually branched midway and reach heights of 1–3½ ft (30 cm–1 m). They are roundish in cross-section and are far less hairy than those of spotted, diffuse, and squarrose knapweed. Upper stems are wiry and slender and sometimes with many alternate branches that end in solitary flower heads.

Flowers

Each branch is topped by a solitary flower bud covered with overlapping rows of light to dark brown bracts bearing papery, deeply fringed margins. At the time of flowering, these bracts reflect a metallic golden sheen (Figure 13b). Bracts spread upon maturation and are topped by 20 to 40 rose-purple colored florets. Receptacles are broadly oval and rounded rather than urn-shaped (the common shape for many knapweeds and starthistles). They are up to 0.5–0.75 inches (12–18 mm) wide and 0.75–1 inch (18–25 mm) long. All florets are hermaphroditic, having both male and female reproductive parts. Flowers usually appear from summer through autumn.

Seeds

Seeds are about 0.12 inches (3 mm) long and ivory white to light brown. They are usually plumeless but sometimes bear a really short row of pappus. Plants can produce 10–500+ seeds, some of which can remain dormant for many years.

Habitat and ecology

Unlike other knapweed species, meadow knapweed grows in moist sites, including irrigated pastures, meadows, river banks, streams, irrigation ditches, and forest openings. It spreads only by seeds, which can be dispersed by water, in hay, or on vehicles. Seeds can remain viable in the soil for many years. Germination occurs primarily in spring and autumn when soil moisture is sufficient.

Distribution and noxious weed status

Meadow knapweed was grown for winter forage in North America prior to 1959. It now occurs in 25 states and four Canadian provinces (Figure 14).

It is listed as noxious in four states (CO, ID, OR, WA) and one Canadian province (BC).

Approved biological control agents

Four biological control agents described in this manual are established on meadow knapweed. These include *Larinus minutus* and *L. obtusus*, *Metzneria paucipunctella*, and *Urophora quadrifasciata* spp. The most promising agent is *Larinus minutus*.



Figure 14 States and provinces where meadow knapweed is established (USDA PLANTS).

Black knapweed

Scientific name

Centaurea nigra L.

Synonyms

Lesser knapweed; common knapweed; *Centaurea jacea* L. ssp. *nigra* (L.) Bonnier & Layens; *Centaurea nemoralis* Jordan

Description

At a glance

Perennial forb growing from a cluster of roots beneath its woody crown. Stems grow upright from 1–3 ft (30 cm–0.9 m) and are branched near the top. Basal leaves are undivided but may be unevenly lobed. Stem leaves are lance-shaped and stalkless, becoming smaller towards the stem tips. Broadly-oval capitula occur singly on branch tips and contain 20–60 pink or purple (rarely white) florets. Flowering is from July to October. Bracts are tipped with comb-like, black teeth. This plant reproduces primarily by seed, but may spread by rooting from leaf nodes of prostrate stems. Seeds have very short, bristled pappus.

Roots

Mature plants develop a cluster of fleshy roots beneath the woody crown; rooting sometimes occurs from the leaf nodes of prostrate stems.

Leaves

Similar to meadow knapweed, basal leaves are not finely divided and are a brighter green than typical knapweeds, with only subtle gray tones and fine hair (Figure 15c). Basal leaves grow up to 6 inches (15 cm) long and 1¼ inch (3 cm) wide having entire margins or tiny lobes or teeth and narrowing close to the stem. Stem leaves are alternate and become progressively smaller up the stem; uppermost leaves are linear, almost bract-like.



Stems

Stems grow upright from 1–3 ft (30 cm–0.9 m), branching near the top. They are round in cross-section and have grooves or ridges; most are covered with fine, cobwebby hairs though some older stems can be hairless. Upper stems are wiry and end in solitary flower heads.

Figure 15 Black knapweed: a. plant; b. capitulum (early bloom); c. leaves. a. Cindy Roche, www.bugwood.org; b. Mikrolit'; c. Brokenearexeter.

Flowers

Each branch is topped by a solitary flower bud covered with overlapping rows of brownish bracts. The middle and outer involucre bracts are tipped with comb-like and black teeth (Figure 15b). Bracts spread upon maturation and are topped by 20 to 60 pink or purple florets. Receptacles are broadly oval and almost globe-shaped rather than urn-shaped (the common shape for many knapweeds and starthistles). They are typically 0.6–1 inch (15–25 mm) wide and 0.6–1.25 inch (16–32 mm) long. All florets are hermaphroditic, having both male and female reproductive parts. Flowers usually appear from summer to early autumn.

Seeds

Seeds are 0.12–0.16 inches (3–4 mm) long, pale brown, sparsely short haired, and topped by a few bristles up to 0.04 in (1 mm) long. Some seeds may lack bristles. Plants can produce from 10 to a few hundred seeds, many of which can remain dormant for many years.

Habitat and ecology

Black knapweed tolerates drier habitats than meadow knapweed, but grows in damper conditions than spotted, diffuse, and squarrose knapweed. Its preferred habitat includes disturbed sites such as roadsides, waste places, fields, and clearings. It spreads by seeds, but occasionally spreads by rooting from leaf nodes of prostrate stems. Seeds are equipped for dispersal by water, hay, livestock, wildlife, and human activity. Seeds can remain viable in the soil for many years. Germination occurs primarily in spring and autumn or when soil moisture is sufficient.

Distribution and noxious weed status

Black knapweed is native to Europe and was introduced to North America via the U.K. It was first recorded in the U.S. in 1895 near Pullman, Washington. This weed occurs in 27 states and six Canadian provinces (Figure 16). It is listed as noxious in one state (WA).

Approved biological control agents

Urophora quadrifasciata is the only biological control agent currently established on black knapweed.



Figure 16 States and provinces where black knapweed is established (USDA PLANTS)

Brown knapweed

Scientific name

Centaurea jacea L.

Synonyms

Brownray knapweed

Description

At a glance

A perennial growing from a taproot or small cluster of roots beneath its woody crown. Stems grow upright from 1–3½ ft (30 cm–1 m) and branch near the top (Figure 17a). Basal leaves are often undivided and taper at both ends. Stem leaves are lance-shaped and stalkless, becoming smaller towards the stem tips. Broadly-oval capitula occur singly on branch tips and contain 20–40 pink florets (rarely white). Flowering occurs from July to October. Receptacle bracts are light to dark brown, with a papery, translucent margin. This plant reproduces primarily by seed, and seed lacks pappus.

Roots

Brown knapweed seedlings are taprooted while mature plants develop a cluster of roots beneath the woody crown.

Leaves

Similar to meadow knapweed, basal leaves are not finely divided and are a brighter green than typical knapweeds, with more subtle gray tones and less hair. Basal leaves grow up to 6 inches (15 cm) long and 1¼ inch (3 cm) wide and have entire margins or shallow lobes. Basal leaves also taper at both ends with the broadest part above the middle of the leaf (Figure 17c). Stem leaves are alternate and become progressively smaller up the stem; uppermost leaves are linear, almost bract-like.



Stems

The stems are upright, usually branched near the top, and reach heights of 1–3½ ft (30 cm–1 m). They are roundish in cross-section, have ridges that are sometimes purple, and are far less hairy than spotted, diffuse, and squarrose knapweed. Upper stems have alternate branches that end in solitary flower heads.

Figure 17 Brown knapweed: a. plant; b. capitulum; c. leaves. a., c., George Slickers; b. Cindy Roche, www.bugwood.org.

Flowers

Each branch is topped by a solitary flower bud covered with overlapping rows of light to dark brown bracts with papery, translucent margins (Figure 17b). Bracts spread upon maturation and are topped by 20 to 40 pink florets (rarely white). Receptacles are broadly oval and almost globe-shaped rather than urn-shaped (the common shape for many knapweeds and starthistles). They are typically 0.5–0.85 inch (12–22 mm) wide and 0.75–1 inch (18–25 mm) long. All florets are hermaphroditic, having both male and female reproductive parts. Flowers usually appear from summer through early autumn.

Seeds

Seeds are light brown, plumeless, and 0.12 inches (3 mm) long. Plants can produce from 5 to a few hundred seeds, some of which can remain dormant for many years.

Habitat and ecology

Similar to meadow knapweed, but brown knapweed prefers moister, cooler conditions than do other knapweed species, including spotted, diffuse, and squarrose. It can be found growing in grasslands, open woods, meadows, pastures, woodland clearings, and in cutover areas of forest. Plants can tolerate partial shade. It spreads only by seeds, which are equipped for dispersal by water, in hay, or on vehicles. Seeds can remain viable in the soil for many years. Germination occurs primarily in spring and autumn when soil moisture is sufficient.

Distribution and noxious weed status

Brown knapweed is native to Europe. It was reportedly grown as a hay or forage crop (known in Quebec in the 1850s as bull clover) and also as a pollen source for honeybees into the late 1800s.

It presently occurs in 26 states and four Canadian provinces (Figure 18). It is listed as noxious in one state (WA).







Approved biological control agents

Urophora quadrifasciata is the only biological control agent currently established on brown knapweed.



Figure 18 States and provinces where brown knapweed is established (USDA PLANTS).

Table 4 Trait comparisons of six exotic knapweed species in North America. Photos: spotted, Michael Shephard, Forest Service; diffuse, Richard Old, www.xidservices.com; squarrose, Steve Dewey, Utah State University; meadow, Eric Coombs, Oregon Department of Agriculture; brown, Cindy Roche; black, Mikrolitt')

| TRAIT | SPOTTED | DIFFUSE | SQUARROSE | MEADOW | BLACK | BROWN |
|-------------------------------|---|--|---|--|--|--|
| Life history | Short-lived perennial | Annual to short-lived perennial (usually biennial) | Long-lived perennial | Perennial | Perennial | Perennial |
| Preferred habitat | Disturbed initially; dry to mesic | Disturbed initially; dry | Disturbed initially; Dry | Moist sites | Mesic to moist | Disturbed initially; Mesic to moist |
| Average height | 2½ ft (0.75 m) | 1½ ft (0.45 m) | 1½ ft (0.45 m) | 2 ft (0.6 m) | 1½ ft (0.45 m) | 2 ft (0.6 m) |
| Basal leaf description | 4-8 in long (10-20 cm); deeply divided into elliptic lobes; gray-green; densely hairy | 4-8 in long (10-20 cm); deeply divided into linear lobes; gray-green; densely hairy | 4-8 in long (10-20 cm); deeply divided into fine lobes; gray-green; densely hairy | 6 in long (15 cm); entire margins (sometimes tiny teeth or lobes); tapered both ends; widest past middle; green; less hair | 6 in long (15 cm); entire margins (sometimes tiny teeth or lobes); wide at base then taper near stem; green; fine hair | 6 in long (15 cm); entire margins (sometimes tiny teeth or lobes); tapered both ends; widest past middle; green; less hair |
| Capitulum diameter | 0.25-0.6 in (6-15 mm) | 0.25-0.4 in (7-10 mm) | 0.12-0.25 in (3-6 mm) | 0.5-0.75 in (12-18 mm) | 0.6-1 in (15-25 mm) | 0.5-0.85 in (12-22 mm) |
| Bract description | Fringe short and rigid; dark brown triangular tip | Narrow; fringed by sharp spines; terminal spine longer than laterals and not curved backward | Narrow; fringed by sharp spines; terminal spine longer than laterals and strongly curved backward | Bearing papery, deeply fringed margins | Tipped with comb-like, black teeth | Bearing papery, translucent margins |
| Capitulum |  |  |  |  |  |  |

North American *Centaurea*

There are 28 species and three hybrids of *Centaurea* established in the U.S. and Canada, including bighead knapweed (*C. macrocephala*), cornflower (*C. cyanus*), and yellow starthistle (*C. solstitialis*) (Figure 19). None of these species are native to North America, and all are covered in greater detail in Table 5.



Figure 19 Other exotic *Centaurea* present in North America: a. bighead knapweed (Richard Old, www.xidservices.com); b. cornflower (Steve Dewey, Utah State University); c. yellow starthistle (Peggy Greb, USDA ARS).

Table 5 Other exotic *Centaurea* in the United States and Canada (USDA-PLANTS)

| SCIENTIFIC NAME | COMMON NAME | DURATION | HABIT | DISTRIBUTION |
|--------------------------|------------------------|---------------------------------|-------|--|
| <i>C. aspera</i> | Rough starthistle | Biennial | Forb | NY |
| <i>C. babylonica</i> | Syrian knapweed | Biennial | Forb | CA |
| <i>C. bovina</i> | Pasture knapweed | Biennial | Forb | MA |
| <i>C. calcitrapa</i> | Red starthistle | Annual Biennial Perennial | Forb | AL, AZ, CA, DC, IA, IL, IN, MD, NJ, NM, NY, OR, PA, UT, VA, WA; ON |
| <i>C. calcitrapoides</i> | Smallhead starthistle | Annual Biennial Perennial | Forb | CA |
| <i>C. cineraria</i> | Dusty miller | Perennial | Forb | CA, MD, NY |
| <i>C. cyanus</i> | Garden cornflower | Annual | Forb | AL, AR, AZ, CA, CO, CT, DC, DE, FL, GA, HI, IA, ID, IL, IN, KS, KY, LA, MA, MD, ME, MI, MN, MO, MS, MT, NC, ND, NE, NH, NJ, NM, NV, NY, OH, OK, OR, PA, RI, SC, SD, TN, TX, UT, VA, VT, WA, WI, WV, WY; AB, BC, MB, NB, NF, NS, ON, PE, QC, YT |
| <i>C. depressa</i> | Iranian knapweed | Annual Biennial | Forb | MD |
| <i>C. diffusa</i> | Diffuse knapweed | Annual Biennial Perennial | Forb | AZ, CA, CO, CT, IA, ID, IL, IN, KY, MA, MI, MO, MT, NE, NJ, NM, NV, OH, OR, TN, UT, WA, WI, WY; AB, BC, ON, QC, SK, YT |
| <i>C. diluta</i> | North African knapweed | Annual | Forb | CA, MO |
| <i>C. eriophora</i> | Wild sandheath | Annual | Forb | CA, CO |
| <i>C. iberica</i> | Iberian knapweed | Perennial | Forb | CA, KS, OR, WA, WY |

Continued next page

Table 5 Other exotic *Centaurea* in the United States and Canada (USDA-PLANTS), *continued*.

| SCIENTIFIC NAME | COMMON NAME | DURATION | HABIT | DISTRIBUTION |
|--|--|-----------------------|-------|--|
| <i>C. jacea</i> | Brown knapweed | Perennial | Forb | CA, CT, DC, DE, IA, ID, IL, IN, KY, MA, MD, ME, MI, MT, NH, NJ, NY, OH, OR, PA, RI, UT, VA, VT, WA, WI, WV; BC, NB, ON, QC |
| <i>C. macrocephala</i> | Bighead knapweed | Perennial | Forb | ID, MI, MT, OR, WA, WI; ON, QC |
| <i>C. melitensis</i> | Maltese starthistle | Annual Biennial | Forb | AL, AZ, CA, GA, HI, IN, MA, MO, NJ, NM, NV, OR, PA, SC, TX, UT, WA, WI; BC |
| <i>C. x moncktonii</i> | Meadow knapweed (hybrid of <i>C. jacea</i> and <i>C. nigra</i>) | Perennial | Forb | BC, NS, ON, QC |
| <i>C. montana</i> | Perennial cornflower | Annual | Forb | AK, ID, ME, MI, MN, MT, NH, NY, OR, PA, UT, WA, WI; BC, NB, NF, ON, QC; SPM |
| <i>C. nigra</i> | Black knapweed | Perennial | Forb | CA, CT, DC, DE, IA, ID, IL, IN, KY, MA, MD, ME, MI, MO, MT, NC, NH, NJ, NY, OH, OR, PA, RI, VA, VT, WA, WI, WV; NB, NF, NS, ON, PE, QC; SPM |
| <i>C. nigrescens</i> | Tyrol knapweed | Perennial | Forb | CA, CT, DC, ID, IL, IN, MA, MD, ME, MI, MO, MT, NE, NH, NJ, NY, OH, OR, PA, RI, VA, VT, WA, WI, WV, WY; BC, NS, ON, QC |
| <i>C. paniculata</i> | Jersey knapweed | Biennial | Forb | NY |
| <i>C. phrygia</i> | Wig knapweed | Perennial | Forb | NY, OH, VT |
| <i>C. x pouzinii</i> | hybrid of <i>C. calcitrapa</i> and <i>C. aspera</i> | Biennial | Forb | CA |
| <i>C. x psammogena</i> | hybrid of <i>C. diffusa</i> and <i>C. stoebe</i> | Perennial | Forb | BC, ON, QC |
| <i>C. scabiosa</i> | Greater knapweed | Perennial | Forb | CT, IA, ID, IN, KY, MD, ME, MT, ND, NH, NJ, NY, OH, UT, WY; BC, NB, ON, QC |
| <i>C. solstitialis</i> | Yellow starthistle | Annual | Forb | AZ, CA, CO, CT, FL, IA, ID, IL, IN, KS, KY, MA, MD, MI, MN, MO, MT, NC, ND, NE, NH, NJ, NM, NV, NY, OH, OK, OR, PA, RI, SC, SD, TN, TX, UT, VA, WA, WI, WV, WY; AB, MB, ON, SK |
| <i>C. stoebe</i> ssp. <i>micranthos</i> | Spotted knapweed | Biennial Perennial | Forb | AL, AR, AZ, CA, CO, CT, DE, FL, GA, HI, IA, ID, IL, IN, KS, KY, LA, MA, MD, ME, MI, MN, MO, MT, NC, ND, NE, NH, NJ, NM, NV, NY, OH, OR, PA, RI, SC, SD, TN, UT, VA, VT, WA, WI, WV, WY; AB, BC, NB, NS, ON, QC, YT |
| <i>C. sulphurea</i> | Sulphur knapweed | Annual | Forb | CA |
| <i>C. transalpina</i> | Alpine knapweed | Perennial | Forb | KY, MA, MN, NJ, OH, OR, VA, VT |
| <i>C. trichocephala</i> | Feather-head knapweed | Perennial | Forb | WA |
| <i>C. uniflora</i> | Singleflower knapweed | Perennial | Forb | NY |
| <i>C. virgata</i> ssp. <i>squarrosa</i> | Squarrose knapweed | Perennial | Forb | CA, MI, MT, NV, OR, UT |

Related species

There are no species of *Centaurea* native to the United States and Canada; however, two species of *Plectocephalus*, formerly classified as *Centaurea*, are native to North America. Both resemble some species of knapweed.

Plectocephalus rothrockii (Figure 20a) is largely restricted to moister canyon sites in the Sierra Madre Occidental of Mexico and associated ranges of Arizona and New Mexico. This range overlaps with only two of the invasive knapweeds of this manual: spotted and diffuse. The receptacle bracts of *P. rothrockii* are very similar to spotted knapweed, however its basal leaves are non-woolly and undivided with entire margins or having tiny teeth.

Plectocephalus americanus (Figure 20b & c) is found throughout the Southwest and Midwest of the U.S., a range overlapping with all knapweeds in this manual except squarrose. Its basal leaves are similar to *P. rothrockii*, and thus distinguish this species from both spotted and diffuse knapweed. Its receptacle bracts are not brown and papery as those of brown knapweed, nor are they fringed in black as in black knapweed. The bracts most closely resemble those of meadow knapweed. *P. americanus* differs from meadow knapweed in that it has 5–7 pairs of fringe lobes per bract, whereas meadow knapweed has more than 12 pairs per bract. Furthermore, *P. americanus* has longer, more delicate florets and prefers dry, sunny, and disturbed sites, whereas meadow knapweed is restricted to more moist conditions.



Figure 20 Species native to North America that closely resemble exotic *Centaurea*: a. *Plectocephalus rothrockii* (G.A. Cooper, courtesy of Smithsonian Institute, USDA PLANTS); b., c., *P. americanus* (pschemp).

CHAPTER 3: BIOLOGY OF KNAPWEED BIOLOGICAL CONTROL AGENTS

History

Biological control of knapweeds is one of the oldest classical biological control programs in the United States and Canada. It began in the 1970s with the importation of the knapweed banded gall fly, *Urophora affinis*. Though this fly established successfully, the knapweed biological control program continued to expand. To date, a total of 13 insect species have been introduced in North America as classical biological control agents of knapweed. These include five beetles, four flies, and four moths.

Basic insect biology

Insects are the largest, most diverse class of animals. An understanding of basic insect biology and anatomy will help land managers recognize and identify the insects used as biological control agents of knapweed. The insects used in this biological control program have complete metamorphosis, which means they exhibit a life cycle with four distinct stages: egg, larva, pupa, and adult (Figure 21). Adult insects have an exoskeleton (a hard external skeleton), a segmented body divided into three regions (head, thorax, and abdomen), three pairs of segmented legs, and may have one or two pairs of wings (Figure 22). The head of an adult insect has one pair each of compound eyes and antennae.

Immature insects have an exoskeleton that must be shed in order for them to grow to the next stage. The process of an insect shedding its “skin” in order to grow is called molting, and larval stages between molts are called “instars.” Larvae generally complete three to five instars before they molt into the pupal stage (Figure 23). During the pupal stage, insects change from larvae to adults. Insects do not feed during the pupal stage.

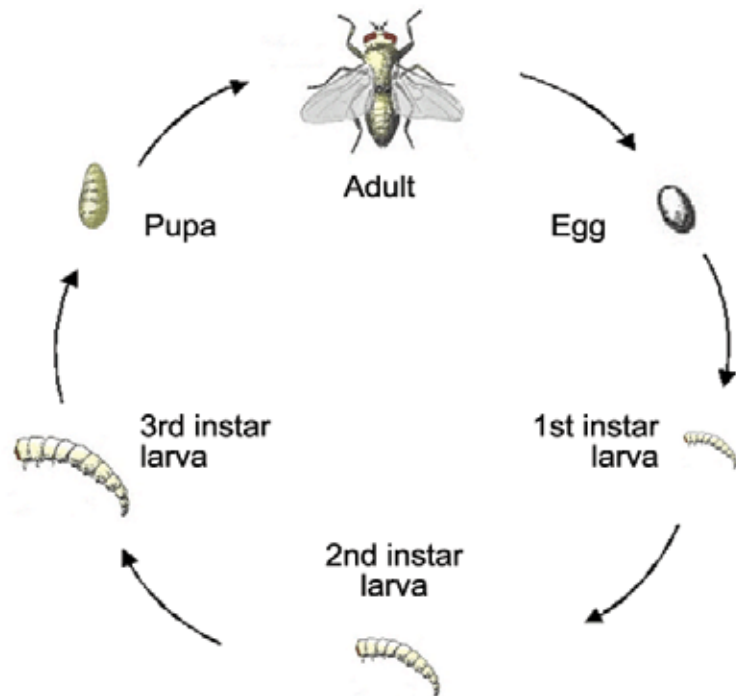


Figure 21 Complete metamorphosis of an insect (www.bugwood.org).

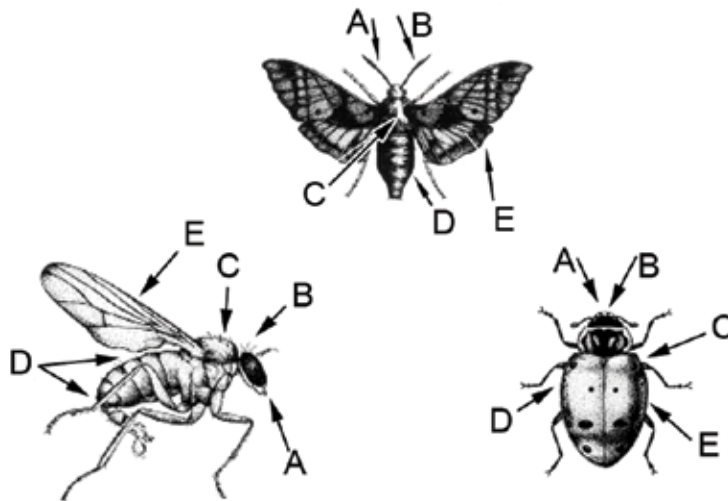


Figure 22 Body parts of adult insects: a. head, b. antenna, c. thorax, d. abdomen, e. wing. All www.bugwood.org.

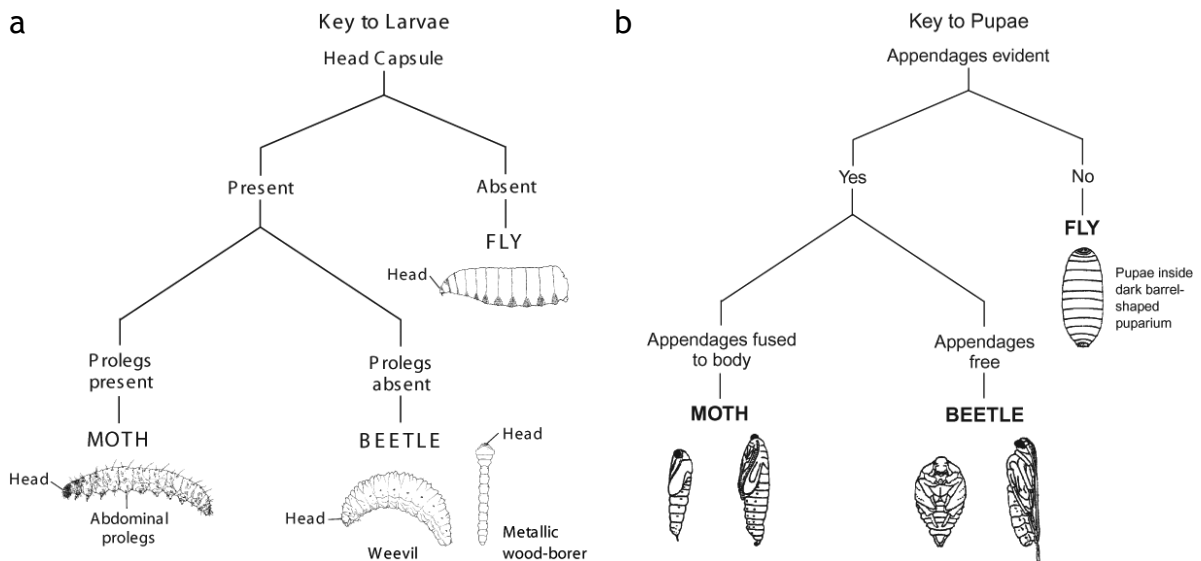


Figure 23 Identification key for: a. insect larvae; b. insect pupae. Both www.bugwood.org.

Beetles (Order Coleoptera)

Most adult beetles are hard-bodied with tough exoskeletons. They have two pairs of wings. The two front wings, elytra, are thick and meet in a straight line down the abdomen of the adult, forming a hard, shell-like, protective covering. The two hind wings are membranous and used for flight. These are larger than the elytra and are folded under the elytra when not in use. Beetle pupae have well-developed appendages that are obviously not fused to the body. Beetle larvae are grub or wormlike with three small pairs of legs. Most are pale white with a brown or black head.

Flies (Order Diptera)

Many insects have the word “fly” in their common name though they may not be true flies. In the common names of true flies, “fly” is written as a separate word (e.g., house fly) to distinguish them from other orders of insects that use “fly” in their common name (e.g., butterfly in the order Lepidoptera and mayfly in the order Ephemeroptera). Adult true flies are easily distinguished from other groups of insects by their single pair of membranous wings and typically soft bodies. Fly pupae are contained inside a barrel-shaped puparium. Larvae of most true flies are legless and wormlike and are called maggots.

Butterflies and moths (Order Lepidoptera)

Adult Lepidoptera have two pairs of membranous wings, covered (usually completely) by minute powder-like scales. Antennae are prominent. Pupae have moderately developed appendages that are fused to the body. The larvae (caterpillars) have a toughened head capsule, chewing mouthparts, and a soft body that may have hair-like or other projections, three pairs of true legs, and up to five pairs of additional prolegs. The pupal stage is known as a chrysalis or cocoon.

Approved knapweed biological control agents

Thirteen knapweed biological control species (five beetles, four flies, and four moths) are permitted for release in the U.S. These insects attack two distinct parts of knapweed plants: eight of the biological control agents are seed feeders and five are root feeders (Figure 24, page 34). Table 6 (page 35) lists the natural enemies of knapweeds in the United States and the species of knapweed they attack.

In general, adults have little impact on the plant, with the exception of two seedhead weevils, *Larinus minutus* and *L. obtusus*. Adults of these species can significantly defoliate knapweed stems, further weakening the plant. Past research demonstrated any one of the 13 knapweed biological control species, alone, could not successfully control knapweed. Consequently, most knapweed biological control programs use a combination of biological control agents which together create multiple stresses on knapweed and have a greater chance of contributing to its suppression. Recent studies, however, indicate that the multiple impacts of *L. minutus*, in particular, have led to knapweed control at various sites in Canada and the northwestern U.S. This species is, therefore, highly recommended for use in any knapweed biological control program.

The remainder of this chapter divides the knapweed biological control agents into two sections: seed feeders and root borers. Each biological control agent listed in Table 6 is described in greater detail within its respective feeding section. Additional information regarding the correct collection and distribution of all insects is described in Chapter 4.

SEED FEEDERS

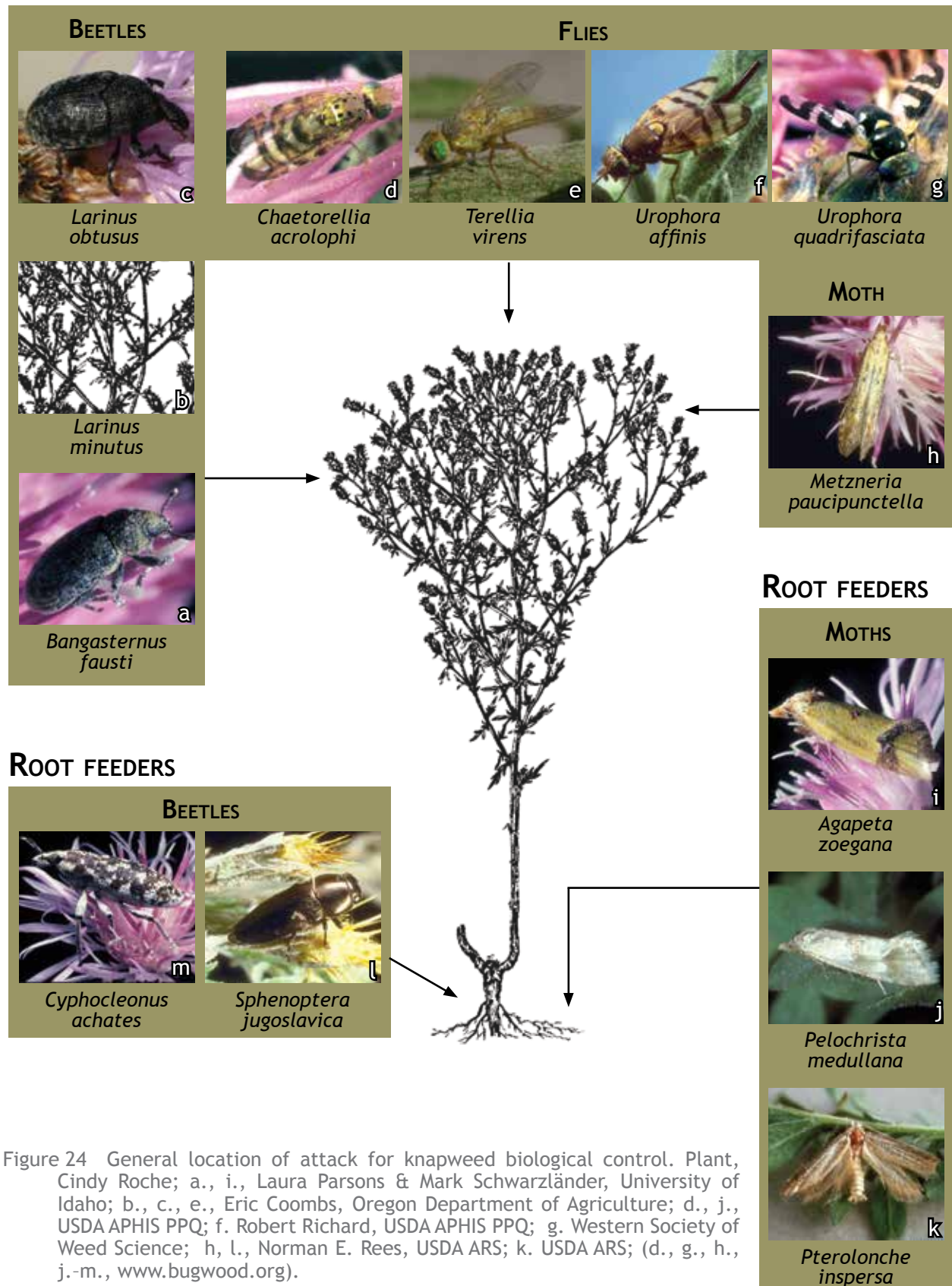


Figure 24 General location of attack for knapweed biological control. Plant, Cindy Roche; a., i., Laura Parsons & Mark Schwarzländer, University of Idaho; b., c., e., Eric Coombs, Oregon Department of Agriculture; d., j., USDA APHIS PPQ; f. Robert Richard, USDA APHIS PPQ; g. Western Society of Weed Science; h, l., Norman E. Rees, USDA ARS; k. USDA ARS; (d., g., h., j.-m., www.bugwood.org).

Table 6 Biological control agents introduced in the United States against exotic knapweeds.

| FEEDING TYPE | INSECT TYPE | SCIENTIFIC NAME | ESTABLISHED | KNAPWEED SPECIES | | | | | |
|-----------------|-------------|---------------------------------|-------------|------------------|---------|-----------|--------|-------|-------|
| | | | | SPOTTED | DIFFUSE | SQUARROSE | MEADOW | BLACK | BROWN |
| Seedhead feeder | Beetle | <i>Bangasternus fausti</i> | X | X | X | X | | | |
| | | <i>Larinus minutus</i> | X | X | X | X | | | |
| | | <i>Larinus obtusus</i> | X | X | X | X | | | |
| | Fly | <i>Chaetorellia acrolophi</i> | X | X | X | X | | | |
| | | <i>Terellia virens</i> | X | X | X | X | | | |
| | | <i>Urophora affinis</i> | X | X | X | X | | | |
| | | <i>Urophora quadrifasciata</i> | X | X | X | X | X | X | X |
| | | <i>Metzneria paucipunctella</i> | X | X | X | X | X | | |
| | | <i>Cyphocleonus achates</i> | X | X | X | X | | | |
| Root borer | Beetle | <i>Sphenoptera jugoslavica</i> | X | X | X | X | | | |
| | | <i>Agapeta zoegana</i> | X | X | X | X | | | |
| | Moth | <i>Pelochrista medullana</i> | X | X | X | X | | | |
| | | <i>Pterolonche inspersa</i> | X | X | X | X | X | | |

Seed feeders

As of 2010, eight seedhead-feeding biological control agents are established in the U.S. and Canada for the control of knapweeds (Table 6). These include three beetles (*Bangasternus fausti*, *Larinus minutus*, and *Larinus obtusus*), four flies (*Chaetorellia acrolophi*, *Terellia virens*, *Urophora affinis*, and *Urophora quadrifasciata*), and one moth (*Metzneria paucipunctella*).

Seed feeder differentiation

The two weevil genera can be differentiated by snout size. *Bangasternus fausti* has a much shorter snout than either of the *Larinus* spp. *Larinus minutus* tends to be smaller, have more grayish hairs and more reddish tibia than the otherwise very similar *L. obtusus*, though the two species may be variants of the same species.

Knapweed seedhead flies can be easily differentiated based upon body and wing coloration (Figure 25a). *Terellia virens* has a straw colored body and clear wings. *Chaetorellia acrolophi* has an orange-yellow and spotted body with light brown bands on the wings. Both *Urophora* spp. have dark bodies. *Urophora affinis* has dark, faint bands on the wings, while *U. quadrifasciata* has thick dark bands making a distinctive “UV” pattern on each wing.

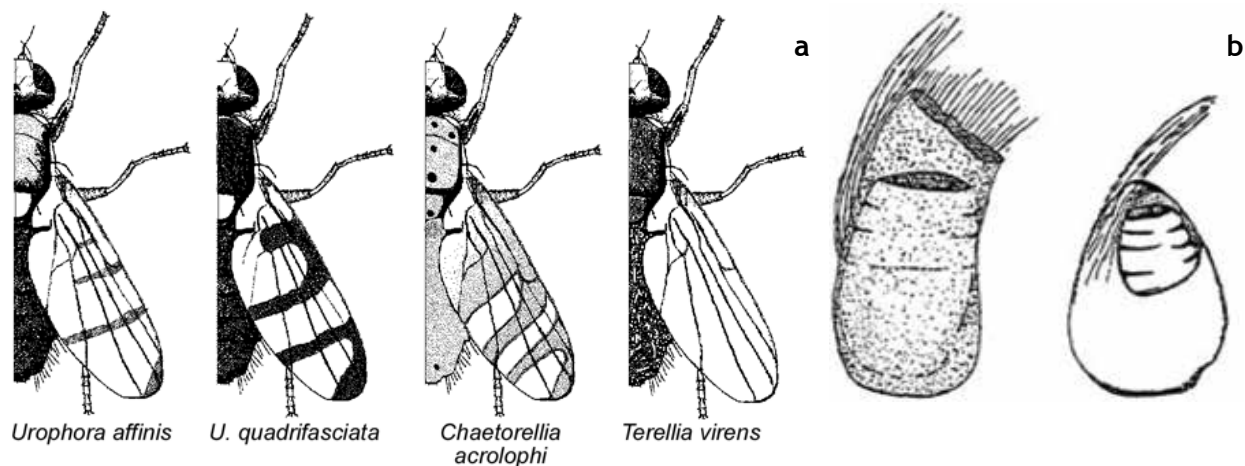


Figure 25 a. Comparison of knapweed seedhead flies (J. Johnson, University of Idaho); b. comparison of *U. quadrifasciata* papery gall (left) and position of larva in the hard, woody gall of *U. affinis* (right) (Cindy Roche).

Seed feeder timeline of attack

Larvae of all the seedhead-feeding insects consume immature seeds and other tissues in the flower head, thus reducing the plant's reproductive output. In addition, two of these species (*Urophora* spp.) are gall formers. Their feeding causes the plant to encase the insect larvae in gall-like structures. The gall formation process acts as a metabolic sink by draining valuable nutrients away from normal plant growth, depleting the plant's resources (Figure 26).

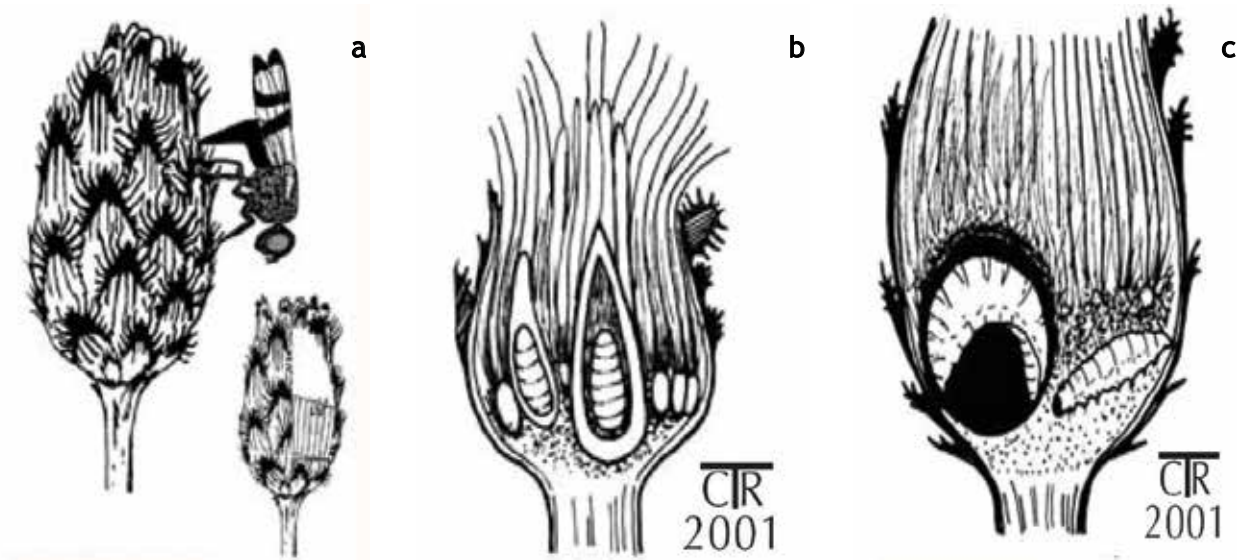


Figure 26 a. *Urophora affinis* ovipositing into a closed knapweed bud (left) and position of eggs amid the young florets in the head (right); b. position of fly larvae inside the knapweed seedhead; c. position of beetle (left) and moth (right) larvae inside the knapweed seedhead. All Cindy Roche.

Seed feeders are separated in time and space by such factors as:

- Type of knapweed patches insects will infest (isolated plants *vs* dense stands)
- Larval feeding habits (e.g., feeding in the receptacle, florets, and seeds)
- Number of generations per year
- Number of larvae in the head
- Overwintering site (in or out of the seedhead)

Knapweeds produce flower heads throughout the spring and summer, creating a constant supply of seedheads of different sizes and stages of development on which the insects can feed. Each seed insect prefers certain seedhead characteristics for oviposition (Table 7).

Table 7 Traits of seed feeding biological control agents introduced in the United States against exotic knapweeds. Underlined knapweed species is preferred host.

| INSECT TYPE | BIOLOGICAL CONTROL AGENT | ATTACK | BUD STAGE PREFERRED FOR ATTACK | GENERATIONS PER YEAR | OVERWINTER |
|-------------|--|---|---|----------------------|--|
| Beetle | <i>Bangasternus fausti</i> Broad-nosed knapweed seedhead weevil | Diffuse, spotted, squarrose | Closed bud 0.14 to 0.2 inch (4 to 5 mm) in diameter | One | Adult in litter |
| | <i>Larinus minutus</i> Lesser knapweed weevil | <u>Diffuse</u> , spotted, squarrose, meadow | Early seed formation | One | Adult in litter |
| | <i>Larinus obtusus</i> Blunt knapweed flower weevil | <u>Spotted</u> , meadow, diffuse | Early seed formation | One | Adult in litter |
| Fly | <i>Chaetorellia acrolophi</i> Knapweed peacock fly | <u>Spotted</u> , diffuse, squarrose | Closed bud 0.14 to 0.3 inch (4 to 7 mm) in diameter | Two, rarely three | Larvae in seedhead |
| | <i>Terellia virens</i> Green clearwing knapweed fly | <u>Spotted</u> , diffuse | Full bloom | One, partial second | Larvae in seedhead (pupae less common) |
| | <i>Urophora affinis</i> Banded knapweed gall fly | Spotted, diffuse, squarrose | Closed bud 0.12 inch (3mm) in diameter | One, partial second | Larvae in seedhead |
| | <i>Urophora quadrifasciata</i> UV knapweed seedhead fly | Black, brown, diffuse, meadow, spotted, squarrose | Closed bud 0.2 to 0.3 inch (5 to 7 mm) in diameter | One or two | Larvae in seedhead |
| Moth | <i>Metzneria paucipunctella</i> Knapweed seedhead moth | <u>Spotted</u> , diffuse, meadow | Late bud to early bloom | One | Larvae in seedhead |

Table 7 (cont.) Photos: 1, Laura Parsons & Mark Schwarzländer, University of Idaho; 2, 3, 5, Eric Coombs, Oregon Department of Agriculture; 4, USDA APHIS 6, PPQ; Robert Richard, USDA APHIS PPQ; 7, Western Society of Weed Science; 8, Norman E. Rees, USDA ARS. 6-8 www.bugwood.org









| ADULT | EGG | LARVA | PUPA |
|---|--|---|--|
|  | Yellow; laid singly on bracts or stem leaves covered with a black egg cap | White legless C-shaped grub with brown head capsule | In a chamber in seedhead, white (brown before emergence) |
|  | Yellow; laid singly or in small clusters in the bud between pappus hairs | White legless C-shaped grub with brown head capsule | Long, white turning brown before emergence |
|  | Yellow; clusters are laid in the bud between pappus hairs | White legless C-shaped grub with brown head capsule | Long, white turning brown before emergence |
|  | White with filament; laid singly or in small clusters under bracts of flower bud | Barrel-shaped; 1st gen. white, 2nd gen. yellow | No gall, white puparium covered in pappus hairs |
|  | Shiny white; multiple eggs laid inside young, opening flower heads, between florets | Barrel-shaped white, turning yellow brown | No gall, yellow-brown puparium |
|  | White; cluster of 1-5 laid inside unopened seedheads, between florets | Creamy white, barrel-shaped, retracted head, circular dark brown anal plate | Inside woody gall, brown |
|  | White; laid in clusters of 1-5 among developing florets of still-closed buds | Creamy white, barrel-shaped, retracted head, elliptical dark brown anal plate | Inside papery gall, brown |
|  | Red-brown then yellow; laid singly on bracts at base of closed bud, or stem just beneath | White with dark brown head capsule, five pair of prolegs | Cocoon brown, appendages fused to body |

Table 8 Comparison of seed feeder life cycles by knapweed growth stage. Underlined knapweed species is preferred host.

| KNAPWEED LIFE STAGE | SPECIES ATTACKED | | | |
|------------------------|--|--|---|--|
| | <i>Bangasternus fausti</i> Diffuse, spotted, squarrose | <i>Larinus minutus</i> <u>Diffuse</u> , spotted, squarrose, meadow | <i>Larinus obtusus</i> <u>Spotted</u> , meadow, diffuse | <i>Chaetorellia acrolophi</i> <u>Spotted</u> , diffuse, squarrose |
| Seedling | Overwinters as adults in plant litter and soil surrounding plant | Overwinters as adults in plant litter and soil surrounding plant | Overwinters as adults in plant litter and soil surrounding plant | Overwinters as larvae in previous year's seedheads |
| Rosette | | | | |
| Bolting | Adults begin to emerge | Adults become active, feeding on leaves of seedlings, rosettes, and bolting plants | Adults become active, feeding on leaves of seedlings, rosettes, and bolting plants | |
| Early bud | Adults feed on foliage, mate, lay eggs on bracts or on end of a stem | | | Adults emerge, mate. Females lay eggs into young buds |
| Late bud | Larvae mine into seedhead and feed on developing florets and ovules | Mating begins | Mating begins | Larvae tunnel to center of seedhead and pupate 10-15 days after they hatch, creating a second gen- eration. A third generation is possible but rare |
| Flowering | Larvae complete development from egg to adult in ~32 days | Eggs laid in between pappus hairs | Eggs laid in between pappus hairs | |
| Seed formation | | Larvae feed on pappus hairs then on seeds and receptacle | Larvae feed on pappus hairs then on seeds and receptacle | |
| Mature | Adults emerge from seedhead through exit hole and overwinter in litter and soil surrounding plant | Pupation occurs. Adults emerge through exit holes, and sometimes feed on foliage and florets, overwintering in litter | Pupation occurs. Adults emerge through exit holes, and some- times feed on foliage and florets, over- wintering in litter | Larvae from second (and possible third) generation feed upon mature seeds then overwinter in seedheads |
| Senescence | | | | |

Continued next page

| SPECIES ATTACKED | | | |
|--|---|---|--|
| <i>Terellia virens</i> <u>Spotted</u> , diffuse | <i>Urophora affinis</i> Spotted, diffuse, squarrose | <i>Urophora</i> <i>quadrifasciata</i> Black, brown, diffuse, meadow, spotted, squarrose | <i>Metzneria</i> <i>paucipunctella</i> <u>Spotted</u> , diffuse, meadow |
| Overwinters as mature larvae (less commonly as pupae) in previous year's seedheads | Overwinters as larvae in previous year's seedheads | Overwinters as larvae in previous year's seedheads | Overwinters as larvae in previous year's seedheads |
| Adults emerge. Mating and egg laying begin with the onset of hot, sunny weather and continue for 4-6 weeks | Late instar larvae begin pupation | Late instar larvae begin pupation | Late instar larvae begin pupation; adults emerge |
| | Adults emerge and mate. Females lay eggs in closed buds, in between immature florets. Slightly older buds are preferred by <i>U. quadrifasciata</i> . | Adults emerge and mate | Adults are active at dusk, laying eggs on bracts or beneath young seedheads |
| Eggs are laid in young, newly flowering seedheads between florets and hatch in 3-5 days. Larvae feed up to 14 days. Second generation may occur, though not typical in North America | Larvae feed on receptacle tissue in developing seedheads. Feeding leads to development of hard, woody galls. | Eggs are laid in closed buds, in between immature florets | Eggs hatch and larvae enter opened seedheads, feeding on florets, seeds, and receptacle tissue |
| | Larvae feed on receptacle tissue in developing seedheads. Feeding leads to formation of thin, papery gall | Larvae feed on receptacle tissue in developing seedheads. Feeding leads to formation of thin, papery gall | |
| 1st generation overwinters as larvae in seedheads (less commonly as pupae). Rare second generation larvae overwinter as pre-pupae; pupation occurs the following spring | 10-33% of larvae pupate and emerge for second generation in late seedheads. The majority overwinter as larvae in seedheads | Second generation adults emerge and lay eggs in late developing seedheads. Second generation overwinters as larvae in seedheads | Larvae overwinter in seedheads |

Continued from previous page

Bangasternus fausti

Broad-nosed knapweed seedhead weevil



| | |
|----------------------------|--------------------------------------|
| ORDER | Coleoptera |
| FAMILY | Curculionidae |
| NATIVE DISTRIBUTION | S Europe, Mediterranean |
| ORIGINAL SOURCE | N Greece |
| FIRST U.S. RELEASE | 1989 Oregon |
| SPECIES ATTACKED | Diffuse, spotted, squarrose knapweed |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | CA, ID, OR, UT, WA |

Figure 27 Adult *B. fausti* (Laura Parsons & Mark Schwarzländer, University of Idaho).

Description

Eggs are yellow ovals covered with dark egg caps. Larvae are white, C-shaped grubs with brown head capsules and can be up to 8 mm (0.3 inch) long. Pupae are white and up to 5 mm (0.2 inch) long. Adults are small and gray to brown/black. They can be 4 mm (0.16 inch) long and have shorter, more blunt snouts compared to the *Larinus* weevils.

Life cycle

Overwintering adults emerge from soil and plant litter in spring and feed on knapweed foliage prior to egg laying. Eggs are laid from late spring through the summer, individually on the underside of leaflets or on stems below the developing flower head. Eggs are covered with masticated plant tissue (which forms a black egg cap) and hatch in 8–12 days. Depending on the egg placement, hatching larvae either mine into the midrib of the leaflet or into the stem prior to tunneling into the flower head. Larvae develop through four instars and feed on developing seed tissue throughout the summer. Pupation occurs in the flower head within a chamber made of frass and fused seeds. Adults emerge in late summer or early fall when knapweeds are senescing. Adults drop to the ground to overwinter. There is one generation per year.

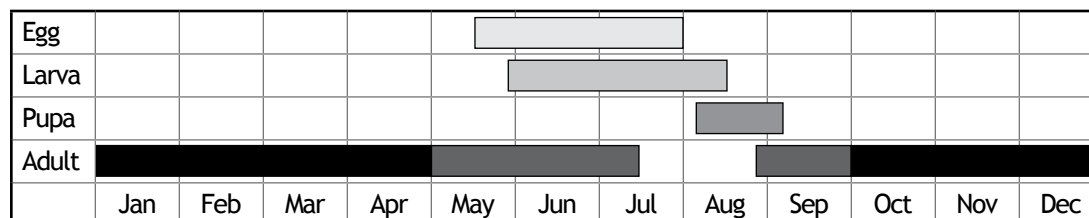


Figure 28 Life cycle of *B. fausti*. Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period.

Habitat preference

This species prefers hot, dry areas and does not do well in areas with prolonged rain or at high elevations.

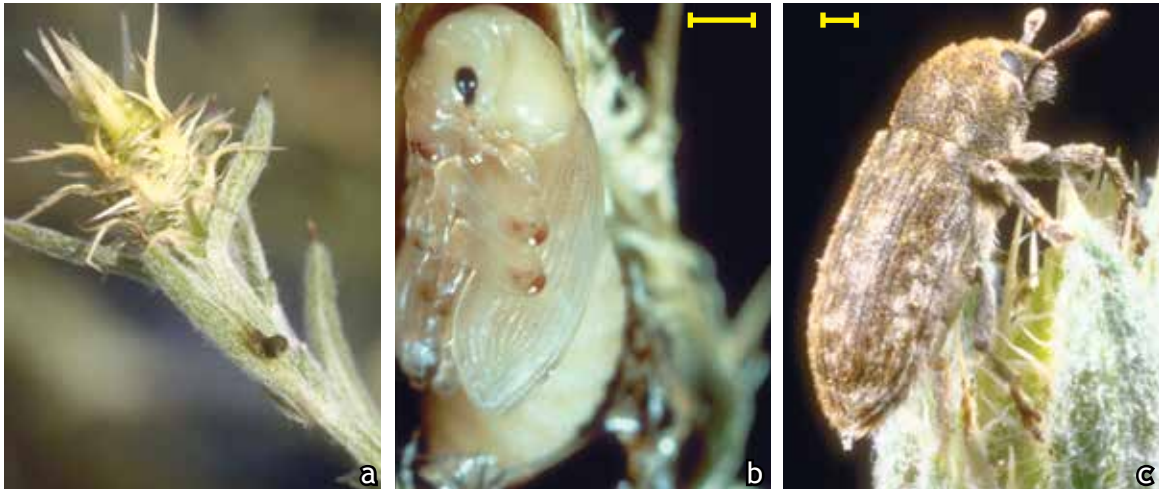


Figure 29 Stages of *Bangasternus fausti*: a. egg (Eric Coombs, Oregon Department of Agriculture); b. pupa (USDA ARS European Biological Control Laboratory); c. adult (USDA APHIS PPQ). All www.bugwood.org.

Impact

Larvae can consume up to 100% of seed in squarrose knapweed, 95% in diffuse knapweed, and less in spotted knapweed (likely an artifact of the size of the capitula for each species). Seed consumption does not damage existing plants, but does help reduce the rate of spread of knapweed populations.

Availability

Populations are widespread and available for mass redistribution in Oregon and Washington.

Comments

Larvae of this species will reportedly attack any other insects occupying the flower head. It is often present with *Larinus* spp., though populations of *B. fausti* are slower to build. The two genera can be distinguished by the shorter, more blunt snout of *Bangasternus*.

Larinus minutus

Lesser knapweed flower weevil



| | |
|---------------------|--|
| ORDER | Coleoptera |
| FAMILY | Curculionidae |
| NATIVE DISTRIBUTION | Mediterranean |
| ORIGINAL SOURCE | Greece |
| FIRST U.S. RELEASE | 1991 MT, WA, WY |
| SPECIES ATTACKED | Diffuse, meadow, spotted, squarrose |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | CA, CO, ID, IN, MN, MT, NV, OR, UT, WA, WY |

Figure 30 Adult *L. minutus* (Laura Parsons & Mark Schwarzländer, University of Idaho).

Description

Eggs are elongate, yellow, and often clustered in the seedhead between pappus hairs. Larvae are white, C-shaped grubs with brown head capsules. They are approximately 8 mm (0.3 inch) long. Pupae are 6 mm (0.24 inch) long and white, turning brown shortly before emergence. Adults are 4–5 mm (0.16–0.2 inch) long, a mottled-brown color, and have a large, bent snout.

Life cycle

Overwintering adults emerge from soil litter throughout the summer. Mating occurs continuously during this long period. Adults feed on the leaves of rosettes and flowering plants, outer stem tissue, and flowers prior to laying eggs. Up to five eggs are deposited in the seedhead between pappus hairs; females lay 28–130 in a lifetime. Larvae hatch in three days and feed on pappus hairs before consuming seeds and receptacle tissue. Larvae feed through the entire flowering period of knapweeds and develop through three instars in four weeks. The number of larvae per seedhead depends on the size of the seedhead and the knapweed species. Pupation occurs in chambers made of chewed seeds and pappus hair within the seedhead. New adults emerge by chewing their way out, leaving behind the now-open pupal chamber. They feed on foliage and flowers before moving to overwintering sites at plant bases. There is one generation per year.

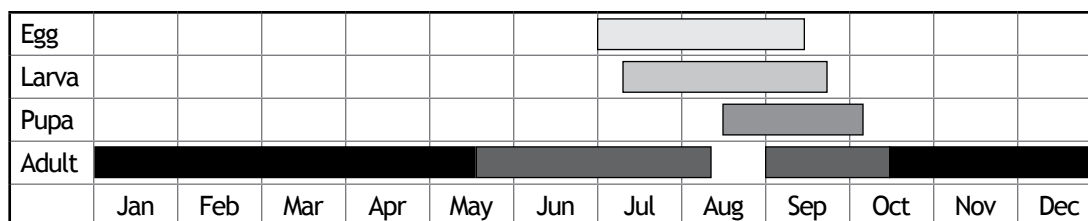


Figure 31 Life cycle of *L. minutus*. Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period.

Habitat preference

Larinus minutus typically prefers sites more dry and hot than those tolerated by *L. obtusus*. It favors dense knapweed stands with little plant competition. It requires well-drained, coarse soils. Compacted sites (especially those grazed with livestock during the bolting stage) or places with prolonged rainfall are not suitable for this insect.



Figure 32 a. Adult *L. minutus*; (Eric Coombs, Oregon Department of Agriculture); b. emergence holes (USDA ARS Archive, www.bugwood.org).

Impact

Knapweed defoliation by adults can be severe. This defoliation can stunt and even kill affected plants. Larval feeding consumes large portions of developing seeds, reducing the rate of knapweed spread even further. Diffuse knapweed plants (the preferred host) under attack by *L. minutus* typically turn a blue-green color, have few leaves, and distorted growth.

Availability

Insects are readily available for redistribution in Oregon and Washington.

Comments

This species has been credited with dramatic reductions of diffuse knapweed in Montana, Oregon, Washington, and British Columbia.

Larinus spp. oviposit later than the other seed-feeder agents. Competing agents can limit their success, though *L. minutus* larvae are reportedly very aggressive. This genus can be distinguished from *Bangasternus* by its longer snout.

Although very similar to *L. obtusus*, *L. minutus* tends to be smaller, have more grayish hairs and more reddish tibia. The two species may be variants of the same species.

At some sites, mice extensively prey on pupae of this species.

Larinus obtusus

Blunt knapweed flower weevil



| | |
|---------------------|---------------------------------------|
| ORDER | Coleoptera |
| FAMILY | Curculionidae |
| NATIVE DISTRIBUTION | Europe, Middle East |
| ORIGINAL SOURCE | Serbia and Romania |
| FIRST U.S. RELEASE | 1991 Colorado |
| SPECIES ATTACKED | Spotted, meadow, occasionally diffuse |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | ID, MT, OR, WA, WY |

Figure 33 Adult *L. obtusus* (Laura Parsons & Mark Schwarzländer, University of Idaho).

Description

This species is very similar to *L. minutus*. Eggs are elongate, yellow, and deposited in the seedhead between pappus hairs. Larvae are white, C-shaped grubs with brown head capsules. They are approximately 8 mm (0.3 inch) long. Pupae are 6 mm (0.24 inch) and white, turning brown shortly before emergence. Adults are 5–7 mm (0.2–0.28 inch) long, a mottled brownish-black, and have a large, bent snout.

Life cycle

Overwintering adults emerge from soil litter throughout the summer. Adults feed on the knapweed foliage and flowers prior to laying eggs. Eggs are deposited in the seedhead between the pappus hairs. Larvae hatch in three days and feed on the pappus hairs and developing seeds. Larvae feed through the entire flowering period of knapweeds, and develop through three instars in 3–4 weeks. Pupation occurs in 9 days in pupal chambers made of chewed seeds and pappus hair within the seedhead. New adults emerge in late summer by chewing their way out, leaving behind the now-open pupal chamber. They feed on foliage and senescing flowers before moving to overwintering sites at the base of plants. There is one generation per year.

| | | | | | | | | | | | | |
|-------|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Egg | | | | | | | | | | | | |
| Larva | | | | | | | | | | | | |
| Pupa | | | | | | | | | | | | |
| Adult | | | | | | | | | | | | |
| | Jan | Feb | Mar | Apr | May | Jun | Jul | Aug | Sep | Oct | Nov | Dec |

Figure 34 Life cycle of *L. obtusus*. Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period.

Habitat preference

Larinus obtusus favors more moist sites with cooler temperatures than those tolerated by *L. minutus*. It establishes on south and west slopes with well-drained coarse soils, often near water. Excess competing vegetation may discourage establishment.

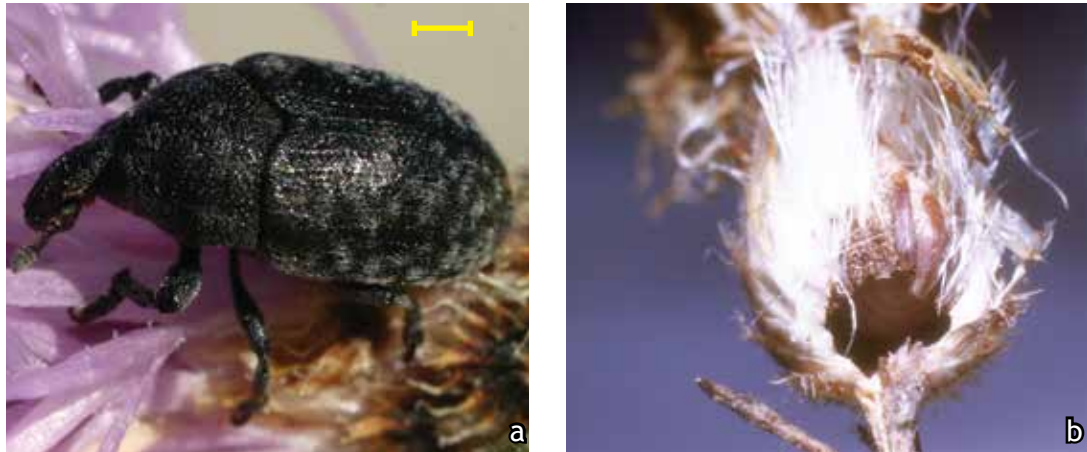


Figure 35 a. Adult *L. obtusus*; (Eric Coombs, Oregon Department of Agriculture); b. damage (Montana State University Archive, www.bugwood.org).

Impact

This weevil prefers spotted knapweed, but may attack meadow and occasionally diffuse. Knapweed defoliation by adults can be extensive when weevil populations are high. This defoliation can stunt and even kill affected plants. Larval feeding consumes large portions of developing seeds, further reducing the rate of knapweed spread.

Availability

Limited supplies of insects are available in Oregon and Washington.

Comments

Populations of this weevil do not build up as quickly on spotted knapweed as *L. minutus* does on diffuse.

This genus can be distinguished from *Bangasternus* by its longer snout. *L. obtusus* is very similar to *L. minutus*, but *Larinus obtusus* tends to be larger, have darker colored tibia, and have somewhat darker colored bodies. The two species may be variants of the same species.

Chaetorellia acrolophi

Knapweed peacock fly



| | |
|----------------------------|---------------------------------------|
| ORDER | Diptera |
| FAMILY | Tephritidae |
| NATIVE DISTRIBUTION | Europe |
| ORIGINAL SOURCE | Austria, France, Hungary |
| FIRST U.S. RELEASE | 1992 Montana |
| SPECIES ATTACKED | Spotted primarily; diffuse, squarrose |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | CO, MT, OR, WA, WY |

Figure 36 Adult *C. acrolophi* (Eric Coombs, Oregon Department of Agriculture).

Description

Eggs are shiny white, elongate, and have a long filament thickened at one end. First generation larvae and pupae are white and barrel-shaped. Second generation larvae and pupae are more yellowish-brown in color. Adults are 4–5 mm (0.16–0.2 inch) long and have bright green eyes, orange-yellow colored abdomens, and overall spotting on the thorax. Wings are clear with light brown bands.

Life cycle

There are usually two generations per year; however, a rare third generation is possible under ideal conditions. Adult flies emerge in early summer as knapweed buds form. Mating occurs immediately and oviposition starts within two days. Females lay eggs individually or in small groups of 2–4 underneath the bracts of unopened buds. A single female may lay 70 eggs in her lifetime. Larvae hatch in 4–5 days and penetrate the buds, and feed on immature florets until they reach the developing seeds, where they feed through three instars. Pupation occurs in the flower head 10–15 days after larvae hatch. Typically, first generation adults emerge throughout July, mate and lay eggs. New larvae of this generation continue to feed on developing seed tissue. Third instar larvae overwinter. Pupation occurs within the flower head the following spring.

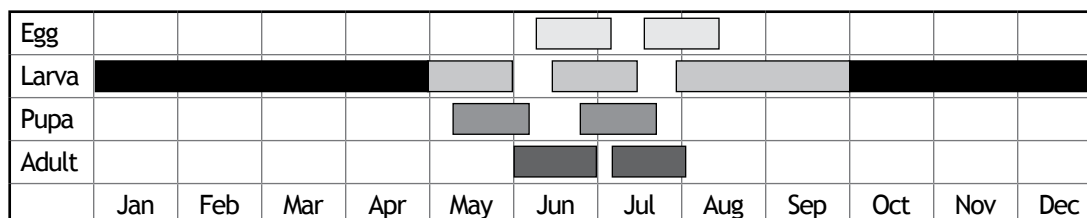


Figure 37 Life cycle of *C. acrolophi*. Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period.

Habitat preference

Chaetorellia acrolophi is most effective in areas with low density knapweed, which is less preferred by other seedhead feeders. The highest rate of attack occurs on sites without *Urophora* species present and only low populations of *Larinus*. It generally does better at higher elevations and in regions with high rainfall.

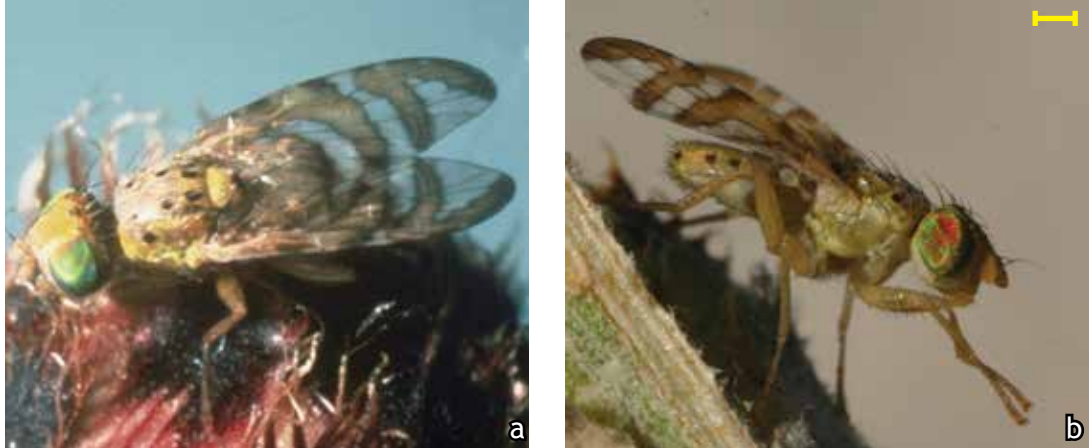


Figure 38 Adult *C. acrolophi*: a. CABI Biosciences Archive (www.bugwood.org); b. Eric Coombs (Oregon Department of Agriculture).

Impact

Spotted knapweed is the preferred host; diffuse and squarrose are occasionally attacked. Larval feeding destroys some developing seeds. This does not damage existing plants, but helps reduce the rate of spread of knapweed populations.

Availability

This species is established in a few states; however, it is available for redistribution in only limited areas.

Comments

C. acrolophi appears to be having difficulty establishing itself in some areas, largely because of competition with other seed-feeding biological control agents.

Terellia virens

Green clearwing knapweed fly



| | |
|---------------------|------------------------------|
| ORDER | Diptera |
| FAMILY | Tephritidae |
| NATIVE DISTRIBUTION | S Europe |
| ORIGINAL SOURCE | Austria |
| FIRST U.S. RELEASE | 1992 Montana |
| SPECIES ATTACKED | Spotted, secondarily diffuse |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | CA, ID |

Figure 39 Adult *T. virens* (Eric Coombs, Oregon Department of Agriculture).

Description

Eggs are elongate, about 1mm (0.04 inch) long, and shiny white. Larvae are a plump barrel shape and white, but turn yellow-brown as they mature. Pupae are yellow-brown. Adults are about 5 mm (0.2 inch long). They have clear wings and large, bright green, and iridescent eyes.

Life cycle

Weather conditions determine the number of generations (one or two) of *T. virens*; however, only one generation has been confirmed at most North American sites.

Adults emerge in spring when knapweed is still in the rosette or bolting stages. Adults feed heavily on nectar when knapweed flower heads bloom. Mating begins with the onset of warm weather and continues throughout summer. In the summer and fall, females lay one to several eggs between florets in young flower heads. The female lays an average of 80 eggs in her lifetime and often marks the bracts of the flower head with a substance to discourage egg laying by other females. Eggs hatch in 3–5 days and larvae feed on ripening seeds and receptacle tissue through three instars. Larvae overwinter within seedheads, then pupate in chambers made of pappus in spring.

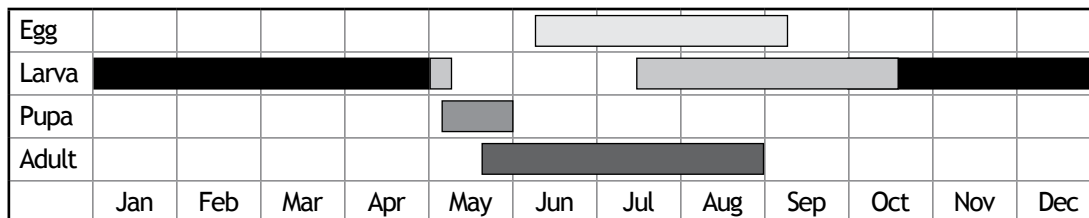


Figure 40 Life cycle of *T. virens* (assuming one generation per year). Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period.

Habitat preference

This species prefers plants on south-facing slopes and dry locations.

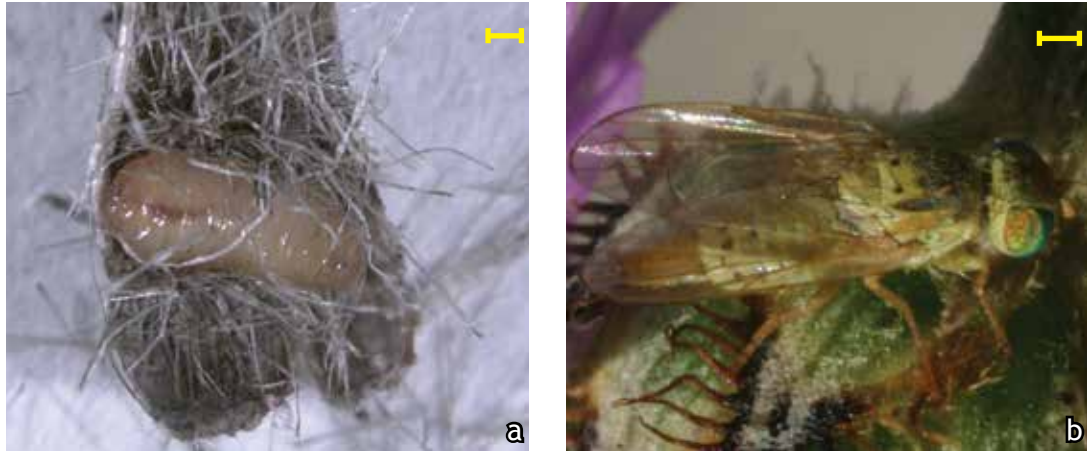


Figure 41 *Terellia virens*: a. larva; b. adult. Both Eric Coombs, Oregon Department of Agriculture.

Impact

Spotted knapweed is the preferred host, but this species will occasionally attack diffuse knapweed. Larvae can consume up to 90% of seed in seedheads. Seed consumption does not damage existing plants, but does reduce knapweed's rate of spread.

Availability

Limited numbers of this fly can be collected in Oregon.

Comments

This fly can coexist in seedheads infested by *Chaetorellia acrolophi* and *Urophora* species, though it thrives when densities of these competing insects are low. It is a very poor competitor in heads infested by *Larinus* species, so only exists where these weevils are absent or at very low densities.

Urophora affinis

Banded knapweed gall fly



| | |
|---------------------|--|
| ORDER | Diptera |
| FAMILY | Tephritidae |
| NATIVE DISTRIBUTION | Europe, W Asia |
| ORIGINAL SOURCE | Austria, France |
| FIRST U.S. RELEASE | 1973 Montana, Oregon |
| SPECIES ATTACKED | Spotted, diffuse, squarrose |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | Throughout knapweed-infested locations in U.S. |

Figure 42 Adult *U. affinis* (Laura Parsons & Mark Schwarzländer, University of Idaho).

Description

Eggs are white, elongate and crescent-shaped. Larvae are creamy white, barrel-shaped, and with heads that retract slightly into the thorax. Larvae of flies do not have head capsules but do develop dark brown anal plates by the end of the feeding period. The pupa is brown, barrel-shaped, and 3 mm (0.12 inch) long. Adults can be up to 4 mm (0.16 inch) long. They have dark bodies and clear wings marked with faint horizontal bars. Females have long, pointed, black ovipositors.

Life cycle

There is usually one generation per year. In warm climates, a second generation may occur. Overwintering as third instar larvae, flies pupate for about 14 days in the spring and emerge as adults at the time knapweed is in the bud stage. Females lay up to 120 eggs in groups of 1–5 among immature florets inside closed seedheads. After 3–4 days, larvae hatch and tunnel into the base of the seedhead where they feed on receptacle tissue. Larval feeding triggers the formation of a hard, woody gall which surrounds the larva. The majority of larvae require a cold period to induce pupation, and thus overwinter in seedheads; 10–25% of larvae may pupate early in suitable climates, with second generation adults emerging in early fall.

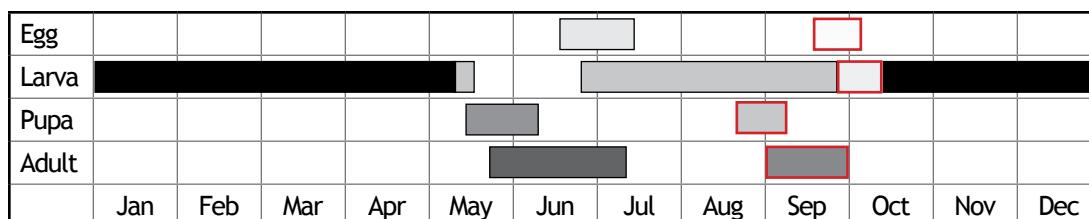


Figure 43 Life cycle of *U. affinis*. Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period. The majority have one generation per year, but a small percentage have two (outlined in red).

Habitat preference

This species is well adapted to a variety of environmental conditions and can be found throughout the majority of spotted and diffuse knapweed infestations. It shows a preference for mesic sites and appears to do better on dense populations.

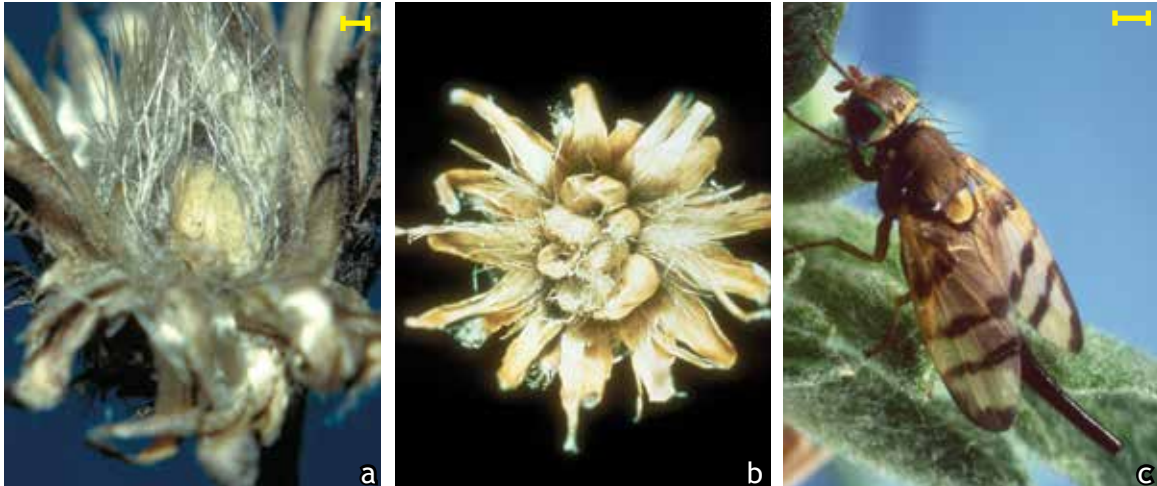


Figure 44 Stages of *Urophora affinis*: a. larva (University of Idaho Archive); b. damage with galls (Jim Story, Montana State University); c. adult (Robert D. Richard, USDA APHIS PPQ). All www.bugwood.org.

Impact

Larvae directly destroy seeds, reducing the rate of knapweed spread. In addition, galls drain nutrients from other parts of the plant, which causes stunting and reduces the number of seedheads produced. Between 2–4 galls in a single seedhead are common, though this depends on seedhead size (often directly related to the species attacked).

Availability

U. affinis is widespread and available for collection throughout the Northwest.

Comments

This was the first biological control agent legally introduced against knapweeds. It does not disperse as rapidly as *U. quadrifasciata*, but is often the dominant species at sites where both flies coexist. Where both flies are present, spotted knapweed seed production is reduced by up to 90%. *Urophora* larvae are often destroyed by *M. paucipunctella* and weevil larvae. Deer mice feed heavily on *Urophora* larvae, and mice populations are known to increase as a result.

Urophora quadrifasciata

UV knapweed seedhead fly



| | |
|----------------------------|---|
| ORDER | Diptera |
| FAMILY | Tephritidae |
| NATIVE DISTRIBUTION | E Europe, W Asia |
| ORIGINAL SOURCE | Russia |
| FIRST U.S. RELEASE | 1981 Immigrated from Canada |
| SPECIES ATTACKED | Black, brown, diffuse, meadow, spotted, squarrose |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | Throughout U.S. |

Figure 45 Adult *U. quadrifasciata* (Western Society of Weed Science, www.bugwood.org).

Description

Eggs are white, elongate, and crescent-shaped. Larvae are creamy white, barrel-shaped, and with heads that retract slightly into the thorax. Larvae do not have head capsules but do develop dark brown anal plates by the end of the feeding period. The pupa is brown, barrel-shaped, and 3 mm (0.12 inch) long. Adults can be up to 4 mm (0.16 inch) long. They have dark bodies and clear wings marked with distinctive dark bands forming a “UV” pattern on each wing. Females have long, pointed, black ovipositors.

Life cycle

There are usually two generations per year. Overwintering as third instar larvae, flies pupate for about 14 days in the spring and emerge as adults at the time knapweed is in the bud stage. Females lay up to 120 eggs in groups of 1–5 among immature florets inside closed seedheads. Also, unlike *U. affinis*, *U. quadrifasciata* females prefer well-developed seedheads. After 3–4 days, larvae hatch and tunnel into the base of the seedhead where, through three instars, they feed on receptacle tissue. Larval feeding induces the formation of a papery gall which surrounds the larva. Pupation occurs in galls in late summer. Second generation adults emerge in early fall and repeat the life cycle, attacking late-developing seedheads. Larvae overwinter in seedheads.

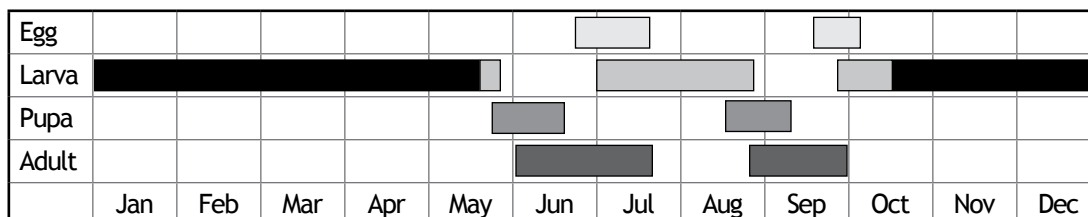


Figure 46 Life cycle of *U. quadrifasciata* (two generations). Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period.

Habitat preference

This species is well adapted to a variety of conditions and can be found throughout the majority of spotted and diffuse knapweed infestations. It is less tolerant of severe winter conditions and requires considerably more protective snow cover than *U. affinis*. Diffuse knapweed is a better host for this species, because it continues to produce suitable floral buds over a long period, which supports the production of second-generation flies.

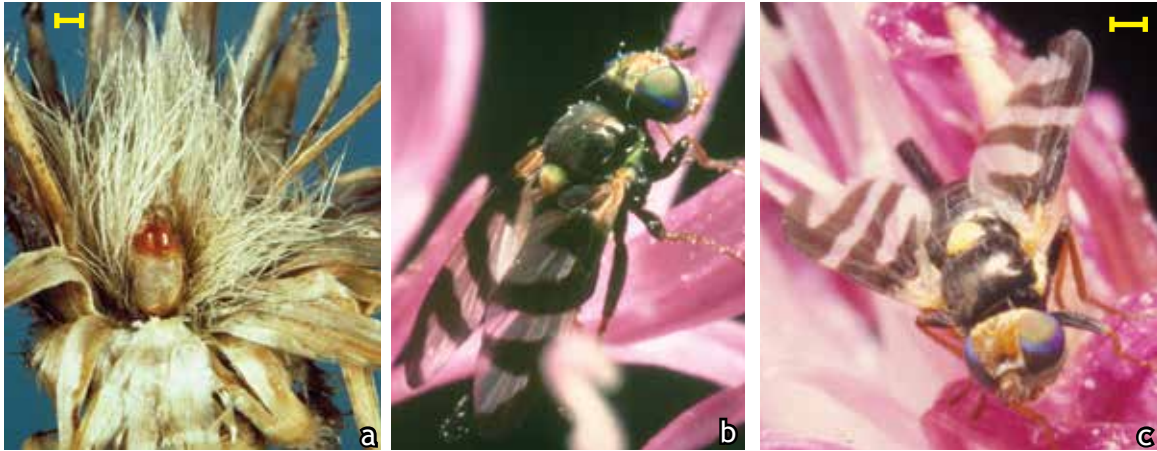


Figure 47 Stages of *Urophora quadrifasciata*: a. larva; b. & c. adult. a. University of Idaho; b. Robert D. Richard, USDAAPHIS PPQ both www.bugwood.org; c. Laura Parsons, University of Idaho.

Impact

Larvae directly destroy seeds, thereby reducing the rate of knapweed spread. In addition, galls drain nutrients from other parts of the plant, causing the plant to produce fewer flowering stems. Between 2–4 galls in a single seedhead are common; the actual number depends on seedhead size (often directly related to the species attacked).

Availability

U. Quadrifasciata is widespread and available for collection throughout the Northwest.

Comments

Initially, this species was not approved for release in the U.S. due to fears it would feed on non-target species; however, in 1980 it migrated into the U.S. from Canada. It has since been approved for redistribution in the U.S., and is now the most widely distributed knapweed biological control agent in the country. Where both *U. affinis* and *U. quadrifasciata* flies are present, spotted knapweed seed production is reduced by up to 90%. *Urophora* larvae are often destroyed by *M. paucipunctella* and weevil larvae. Deer mice populations increase due to heavy feeding on *Urophora* larvae.

Metzneria paucipunctella

Spotted knapweed seedhead moth



| | |
|---------------------|---|
| ORDER | Lepidoptera |
| FAMILY | Gelechiidae |
| NATIVE DISTRIBUTION | Europe |
| ORIGINAL SOURCE | Switzerland |
| FIRST U.S. RELEASE | 1980 Montana |
| SPECIES ATTACKED | Spotted preferred; also diffuse, meadow |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | CO, ID, MT, OR, UT, VA, WA |

Figure 48 Adult *M. paucipunctella* (University of Idaho Archive, www.bugwood.org).

Description

Eggs are elongate, oval, and reddish-brown when first deposited, but turn yellowish as they mature. Larvae are 4–5 mm (0.16–0.20 inch) long, white with dark brown head capsules, distinct body segments, and several pairs of prolegs. Pupae, enclosed in a cocoon, are brown with appendages fused to the body. Adult moths are small (8 mm or 0.32 inch long). Their front wings are slightly fringed and light gray with peppery spotting and dark tips. When at rest, the wings are folded over their backs, giving them a slender appearance.

Life cycle

Adults begin emerging and mating in late spring and early summer when knapweeds are in the rosette and bolting stages. They fly at dusk and are rarely seen. Female moths may lay 60–100 eggs, beginning in early summer. Eggs are placed singly on the bracts at the base of the young flower heads, or on the stems just below the flower head. Larvae hatch in 10–12 days as flower heads are opening. Larvae enter the opened flower heads and feed on the florets, seeds, and receptacle tissue (which reduces the viability of uneaten seeds). There are five instars total; several young larvae can occupy a seedhead early in the season, but only one larva survives beyond the third instar. Larvae overwinter in the seedheads. Pupation occurs in the seedhead in spring and lasts 3–4 weeks. There is one generation per year.

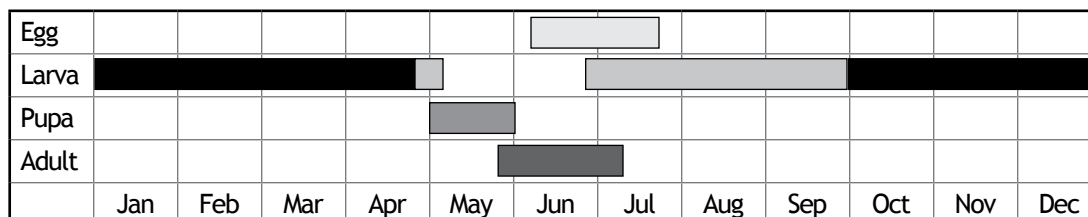


Figure 49 Life cycle of *M. paucipunctella*. Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period.

Habitat preference

This species does not tolerate severe winter temperatures. Favored sites are south slopes in dry, mild-winter climates. Snow cover during winter enhances larvae survival. It appears to do best in areas where spotted knapweed flowers early in the season.



Figure 50 Stages of *Metzneria paucipunctella*: a. Larva and pupa; b. adult (all Norman Rees, USDA ARS www.bugwood.org).

Impact

This species prefers spotted knapweed, but will also attack diffuse and meadow. Feeding larvae can destroy eight seeds per larva (on average) and reduce the viability of others. Older larvae bind seeds together, preventing seeds from dispersing over long distances.

Availability

Populations are widespread and available for collection in Idaho, Montana, Oregon and Washington.

Comments

Larvae of this species will destroy any other insects occupying the flower head. Despite the strong competition between this species and the two *Urophora* species, seed reduction is greater when all three species are present.

Moth larvae are heavily preyed upon by deer mice and are also frequently attacked by parasitoids. Both natural enemies cause heavy mortality of *M. paucipunctella*.

Root borers

Five root-boring insect species have been introduced in the U.S. and Canada for the control of diffuse, spotted and squarrose knapweeds. Two species are beetles (*Cyphocleonus achates* and *Sphenoptera jugoslavica*), and three are moths (*Agapeta zoegana*, *Pelochrista medullana*, and *Pterolonche inspersa*). Only three of these root-feeding biological control agents are established. Though initial releases of *P. medullana* and *P. inspersa* were believed successful, these insects are no longer present in North America.

Root borer differentiation

The adult root-boring beetles are easily differentiated (Figure 24, pag32, or Table 9). *Cyphocleonus achates* is large, rounded, and a dull, mottled gray-black; *S. jugoslavica* is flattened, tapered toward the abdomen, and metallic bronze. Forewing color can be used to differentiate adult knapweed root-boring moths: *Agapeta zoegana* is bright yellow with brown bands; *P. medullana* is tan to gray with mottled tips; and *P. inspersa* is light brown with a silvery sheen.

Root borer timeline of attack

Larvae of all the root-boring insects burrow into and mine the roots of knapweeds. Depending on the species, they feed on the central vascular tissue or the cortex of the root just below the epidermis (Figure 51). In either case, feeding depletes the carbohydrate reserves of the attacked plants, thus hindering both the plant's growth and ability to overwinter. Root-boring may kill plants outright. In addition to mining the roots, the larval feeding by *S. jugoslavica*, *C. achates* and *P. inspersa* cause root galls to form. The gall formation process acts as a metabolic sink by draining valuable nutrients away from normal plant growth, further depleting the plant's resources.

Multiple larvae of *C. achates* and *A. zoegana* may attack the same knapweed root as well as those infested by other species. The remaining knapweed root borers usually do not occur with other species, and there is typically only one larvae per root.

Most larvae of the knapweed root-boring biological control agents complete their development in a single root; however, larvae of *A. zoegana*, have been observed migrating a short distance between roots during the growing season.

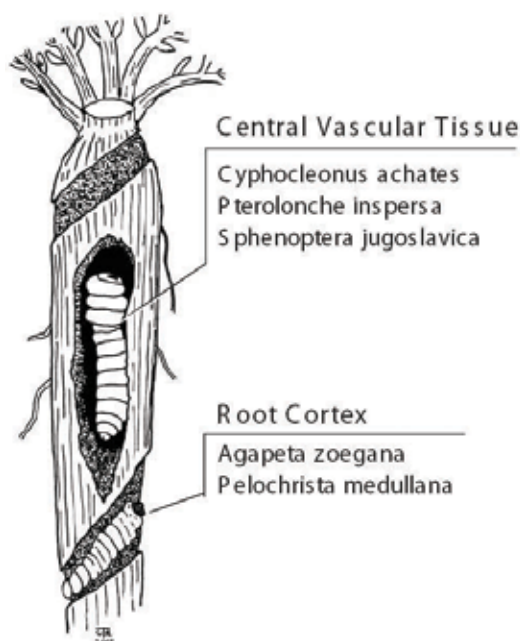


Figure 51 Distribution of knapweed root borers (Cindy Roche).

Table 9 Traits of seed feeding biological control agents introduced in the United States against exotic knapweeds. Underlined knapweed species is preferred host. Photos: Laura Parsons & Mark Schwarzländer, University of Idaho (1,3); Norman Rees, USDA ARS (2); USDA APHIS PPQ (4), USDA ARS (5); 2,4,5 www.bugwood.org.






| INSECT TYPE | BIOLOGICAL CONTROL AGENT | ATTACK | GENERATIONS PER YEAR | OVER-WINTER | ADULT | EGG | LARVA | PUPA |
|-------------|---|--|----------------------|---------------------------|--|---|---|--|
| Beetle | <i>Cyphocleonus achates</i> Knapweed root weevil | <u>Spotted</u> , diffuse | One | Mature larvae in the root |  | Dark; laid in clusters on the root crown, just below the soil surface | Large, C-shaped grubs with light brown head capsules | Large, white, with free appendages |
| | <i>Sphenoptera jugoslavica</i> Bronze knapweed root borer | Diffuse, <u>squarrose</u> , spotted | One | Mature larvae in the root |  | Purple-blue; laid on the bases of rosette leaf petioles | Cylindrical, tapering at the tail end; have dark brown head capsules | Large, white (brown before emergence) with free appendages |
| Moth | <i>Agapeta zoegana</i> Sulfur knapweed moth | <u>Spotted</u> , diffuse | One | Mature larvae in the root |  | Yellow-red; laid singly or 2-3 in stem and leaf crevices | White with brown head capsule and mouthparts | Large, white; appendages fused to body |
| | <i>Pelochrista medullana</i> Brown-winged knapweed root moth | <u>Diffuse</u> , spotted | One | Mature larvae in the root |  | Ribbed, yellow; laid in small clusters under rosette leaves | Elongate, whitish with brown head capsules. Found in mined tunnel in silken web | Large, white; appendages fused to body |
| | <i>Pterolonche inspersa</i> Grey-winged knapweed root moth | <u>Diffuse</u> , spotted, <u>squarrose</u> | One | Mature larvae in the root |  | Black; laid singly or in clusters of 2-3 on underside of leaf | Elongate, pearly white, and with light brown head capsule | Large, white; appendages fused to body. Found in silken cocoon |

Table 10 Comparison of root borer life cycles by knapweed growth stage. Underlined knapweed species is preferred host.

| KNAPWEED LIFE STAGE | SPECIES ATTACKED | |
|---------------------|---|---|
| | <i>Cyphocleonus achates</i> <u>Spotted</u> , diffuse | <i>Sphenoptera jugoslavica</i> Diffuse, squarrose, spotted |
| Seedling | Overwinters as larvae in previous year's roots | Overwinters as larvae in previous year's roots |
| Rosette | | |
| Bolting | Larvae pupate in roots | Larvae pupate and new adults emerge |
| Early bud | | |
| Late bud | | |
| Flowering | Adults mate and females lay eggs on root crown | Adults mate and females lay eggs |
| Seed formation | Larvae burrow into root central vascular tissue, forming a gall in the root | Larvae burrow into root central vascular tissue, forming a gall in the root |
| Mature | | |
| Senescence | Larvae overwinter in roots | Larvae overwinter in roots |

Continued next page

| SPECIES ATTACKED | | |
|--|--|---|
| <i>Agapeta zoegana</i> Spotted, diffuse | <i>Pelochrista medullana</i> Diffuse, spotted | <i>Pterolonche inspersa</i> Diffuse, spotted, squarrose |
| Overwinters as larvae in previous year's roots | Overwinters as larvae in previous year's roots | Overwinters as larvae in previous year's roots |
| Larvae actively feed on roots again | | |
| Larvae pupate and new adults emerge | Larvae pupate and new adults emerge | Larvae pupate and new adults emerge |
| Adults mate and females lay eggs | | |
| Larvae hatch and chew into root cortex May migrate to other nearby roots and continue development | Adults mate and females lay eggs | Adults mate and females lay eggs |
| | Larvae migrate to root cortex to feed | Larvae migrate to root vascular tissue to feed, causing galls |
| | Larvae overwinter in roots | Larvae overwinter in roots |
| Larvae overwinter in roots | | |

Continued from previous page

Cyphocleonus achates



| | |
|---------------------|------------------------------------|
| ORDER | Coleoptera |
| FAMILY | Curculionidae |
| NATIVE DISTRIBUTION | Europe, Mediterranean |
| ORIGINAL SOURCE | Austria, Greece, Hungary |
| FIRST U.S. RELEASE | 1988 Montana, Washington |
| SPECIES ATTACKED | Spotted primarily; diffuse |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | CA, CO, ID, MT, NM, OR, UT, WA, WY |

Figure 52 Adult *C. achates* (Laura Parsons & Mark Schwarzländer, University of Idaho).

Knapweed root weevil

Description

Eggs are <2 mm (0.08 inch) in diameter and white or pale yellow initially, but darken during incubation. Larvae are plump, creamy white or yellowish, with large, light brown head capsules. They can be up to 13 mm (0.5 inch) long. Similar to most weevils, they retain a “C” shape. Adults are large, 13–15 mm (0.5 to 0.6 inch) long, brown–gray mottled, and have short, thick snouts. Females have rounded abdomens, while the males’ are flattened. They are rapid walkers, compensating for their poor ability to fly.

Life cycle

Adults emerge in late summer through early fall and spend most of their life (about 10 weeks) on the root crown, just below the surface. On sunny days they climb to the tops of plants in search of mates, and may mate several times in their lives. Females lay their eggs in notches they excavate on the root crowns, just below the soil surface. A typical female may lay over 100 eggs. Larvae hatch in 10–12 days and mine towards the center of the roots. They develop through four instars, with third- and fourth-instar larvae often causing a gall-like enlargement of the root. Larvae overwinter in the roots, and pupation occurs in the root over a two-week period in early summer. New adult weevils chew through the root and crawl to the surface. They live for 8–15 weeks but do not overwinter. There is one generation per year.

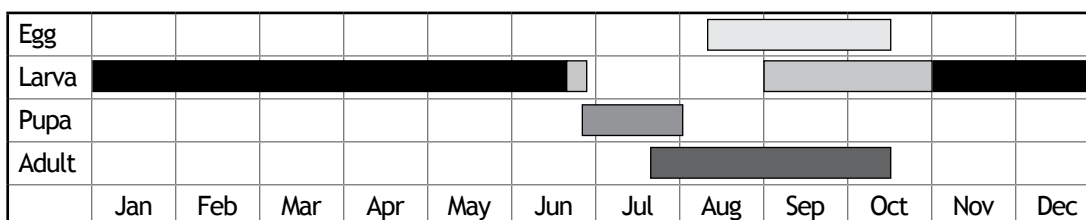


Figure 53 Life cycle of *C. achates*. Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period.

Habitat preference

This weevil prefers hot and dry sites, with loose well-drained coarse soils in temperate areas. It establishes in undisturbed bunchgrass habitat, but favors bare soil surfaces where grasses do not crowd the target plants. Sites need to be somewhat large with a corridor of plants, into which it disperses by walking.

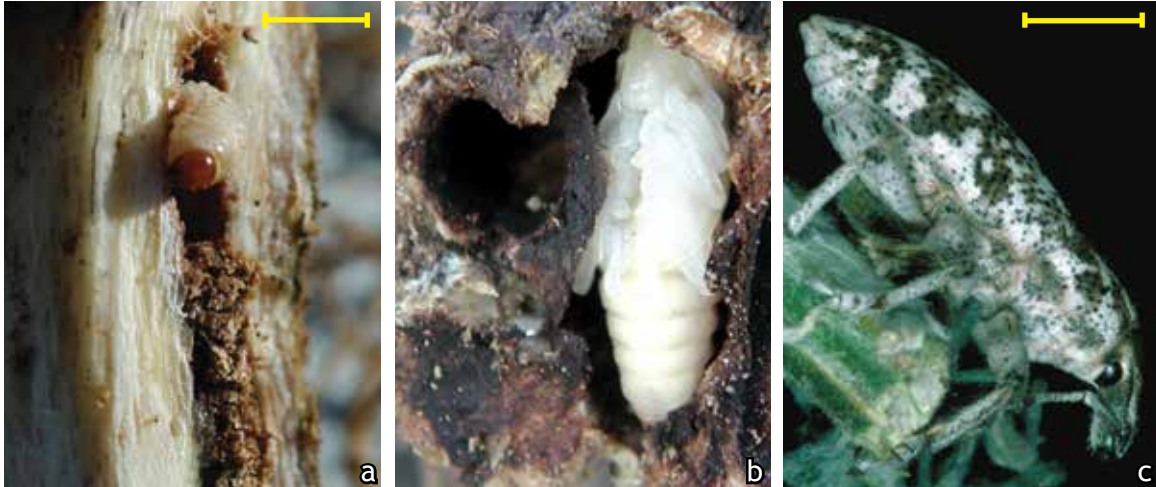


Figure 54 Stages of *Cyphocleonus achates*: a. larva (Eric Coombs, Oregon Department of Agriculture); b. pupa (Biological Control: A Guide of Natural Enemies of North America Web Site, Cornell University); c. adult (J. Johnson, U of Idaho). All www.bugwood.org.

Impact

Spotted knapweed is preferred over diffuse. Small plants can be killed by larval feeding. Most damage is done when multiple larvae occupy a root or when the attacked roots are small, which leads to a reduction in the biomass and density of knapweed populations. Tunneling in the root also exposes the plant to bacterial and fungal infection that can cause secondary injury.



Figure 55 Four larval instars of *C. achates* (USDA ARS Archive).

Availability

This insect is established in numerous states and populations though it has been slow to spread naturally due to its poor flying ability. Mass-rearing efforts have increased populations of this weevil throughout northwestern North America.

Comments

Multiple larvae are often found attacking the same root, along with other species.

Sphenoptera jugoslavica

Bronze knapweed root borer



| | |
|----------------------------|------------------------------------|
| ORDER | Coleoptera |
| FAMILY | Buprestidae |
| NATIVE DISTRIBUTION | S Europe |
| ORIGINAL SOURCE | Greece |
| FIRST U.S. RELEASE | 1979-1980 CA, ID, OR, WA |
| SPECIES ATTACKED | Diffuse; also spotted, squarrose |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | CA, CO, ID, MT, NV, OR, UT, WA, WY |

Figure 56 Adult *S. jugoslavica* (Norman Rees, USDA ARS, www.bugwood.org).

Description

Eggs are flat and white when first laid, but change to dark bluish–purple after five days. Larvae have an enlarged head and a long, thin, cylindrical body that tapers to the end. They are whitish with dark brown head capsules. Pupae are initially white, but later darken. Adults can be up to 10 mm (0.4 inch) long. They are a metallic bronze color and somewhat flattened, with their bodies tapering towards the narrowed abdomen tip.

Life cycle

Adults emerge in summer with the onset of diffuse knapweed flowering. They feed on knapweed leaves for 2–3 days before mating. During late summer, females lay multiple eggs between the bases of rosette leaves. Leaf stems with diameters of 3–6 mm (0.12–0.24 inch) are preferred over smaller leaves. Females lay an average of 50 eggs during their lifetimes. Larvae hatch after two weeks and feed between leaf stalks. As knapweed sets seed, second-instar larvae mine into the upper root; their feeding creates swollen galls and tunnels often filled with frass. Larvae overwinter in roots. Pupation (nine days) occurs within the feeding chamber during early summer the following year. There is one generation per year.

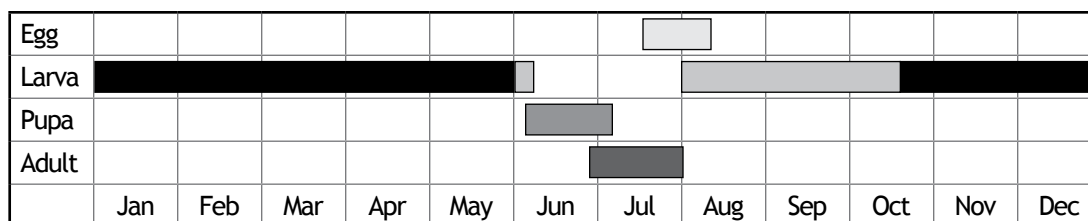


Figure 57 Life cycle of *S. jugoslavica*. Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period.

Habitat preference

This species prefers arid environments with a period of drought in summer. It thrives in well-drained coarse soils on south aspects. Exposed soil between plants will increase the soil temperature, making sites even more suitable.



Figure 58 Stages of *Sphenoptera jugoslavica*: a. larva (University of Idaho Archive www.bugwood.org); b. adult (Laura Parsons & Mark Schwarzländer, University of Idaho).

Impact

Diffuse knapweed is the preferred host, but this species will also attack squarrose and spotted knapweed. Larvae root mining can cause significant damage; in comparison, adult feeding on the leaves is much less damaging. Larval feeding causes gall-like swelling near the knapweed root crown, depleting carbohydrates and interrupting nutrient transfer, which can kill the plant or retard rosette growth. Attacked plants are often stunted and produce fewer seeds the following season.

Availability

Populations are widespread and available for mass redistribution in Oregon and Washington.

Comments

Plants rarely support more than one larva; if two develop on a single root, the larva feeding lowest in the root is usually smaller. Root aphids at some sites attract ants, which kill *S. jugoslavica* larvae and pupae.

Agapeta zoegana

Sulfur knapweed moth



| | |
|----------------------------|------------------------------------|
| ORDER | Lepidoptera |
| FAMILY | Cochylidae |
| NATIVE DISTRIBUTION | Europe, W Asia |
| ORIGINAL SOURCE | Hungary, Austria |
| FIRST U.S. RELEASE | 1984 Montana |
| SPECIES ATTACKED | Spotted, sometimes diffuse |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | CA, CO, ID, MN, MT, OR, UT, WA, WY |

Figure 59 Adult *A. zoegana* (Laura Parsons & Mark Schwarzländer, University of Idaho).

Description

Larvae are white with brown mouthparts and can be up to 7 mm (0.3 inch) long. Adults are usually 11 mm (0.4 inch) long with a wingspan measuring 15–23 mm (0.6–0.9 inch). Forewings are bright yellow with brownish band markings; hind wings are dark gray. Females have a larger, more rounded abdomen than males and lay white, flattened eggs that turn yellow-red in a few days.

Life cycle

Larvae overwinter in roots, and feed in those roots the following spring. Pupation occurs in the roots; adults emerge from summer to early fall when knapweeds are in bud and flowering. Adults mate within 24 hours of emergence and are short-lived. They are most active in early morning or evenings, and rest low on the plants or on the soil surface during the day. Females deposit eggs on knapweed stem crevices and leaves as early as the following day. A single adult female lays 21–78 eggs in her lifetime. Larvae hatch in 7–10 days and migrate to the crown area and mine roots, and develop through six instars. As they mine the outer root layers, larvae produce a whitish web tunnel that encloses them. They create a spiral trail downward before they turn back towards the top of the root. There is usually only one generation per year.

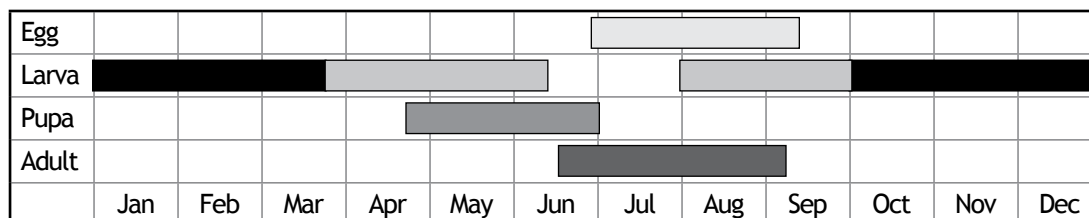


Figure 60 Life cycle of *A. zoegana*. Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period.

Habitat preference

This species is found mostly in dry, well-drained, open sites with scattered (not dense) vegetation. It survives in areas characterized by a moderately humid climate and in areas with arid, subcontinental climates. It can tolerate cold winter temperatures, but requires a long growing season. Suitable host plants have roots with at least 2.4 mm (0.1 inch) in diameter.

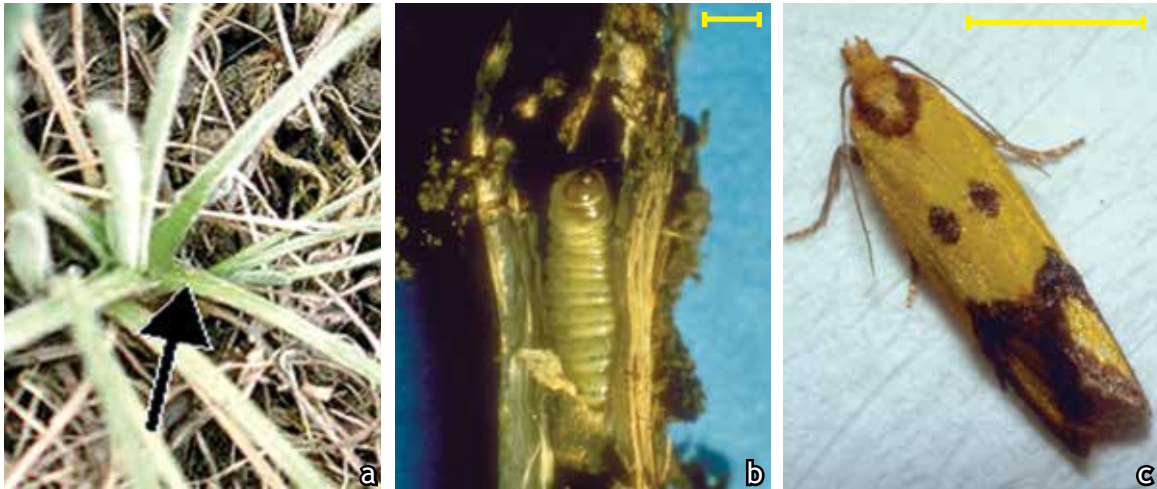


Figure 61 Stages of *Agapeta zoegana*: a. egg (Nez Perce Biological control Center Archive); larva (USDA ARS); c. adult (Jim Story, Montana State University). All www.bugwood.org.

Impact

Spotted knapweed is preferred over diffuse knapweed as a host. When larvae feed within roots, root tissue can be completely consumed. This reduces knapweed biomass and density and may kill small plants.

Availability

This moth is becoming widespread throughout the U.S., but densities decrease as insects disperse.

Comments

Multiple larvae may attack the same root. In one observation, more than 50 *A. zoegana* larvae and 20 *C. achates* larvae were found attacking one very long (8 inches, 20 cm) segment of knapweed root. This insect was released experimentally on squarrose knapweed, but was not recovered.

Pelochrista medullana

Brown-winged knapweed root moth



| | |
|----------------------------|-------------------------------|
| ORDER | Lepidoptera |
| FAMILY | Tortricidae |
| NATIVE DISTRIBUTION | E Europe |
| ORIGINAL SOURCE | E Romania, E Austria |
| FIRST U.S. RELEASE | 1984 Montana |
| SPECIES ATTACKED | Diffuse; occasionally spotted |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | |

Figure 62 Adult *P. medullana* (USDA APHIS PPQ Archive, www.bugwood.org).

Description

Eggs are oval, somewhat flattened, and have a strong outer shell with distinct ribs. Initially they are white, but gradually turn dark yellow during incubation. The segmented larvae are whitish-yellow with brown head capsules. They are usually less than 10 mm (0.4 inch) long. Adult moths are tan to gray with mottled wings fringed at their tips. They can be up to 10 mm (0.4 inch) long.

Life cycle

Adults emerge throughout summer when knapweed is bolting and flowering. They mate within 24 hours of emergence and lay eggs primarily on the lower surface of rosette leaves. Females can lay up to 120 eggs in warm dry weather, but this can be greatly reduced by cold, rainy conditions. Larvae hatch 7–9 days after oviposition, move to the center of the rosette and mine into the root crown. Larvae feed on the outer layers of root tissue, similar to *Agapeta zoegana*. Webbed tubes are produced along feeding tracks, which can be irregular, downward or spiralling, and the tunnels are lined with a silken web. There are six larval instars. This species seems to prefer rosette plants; larvae that feed on the roots of flowering plants develop poorly. Larvae overwinter in the roots and complete development in the spring or early summer. Pupation occurs within the webbing inside the root. There is one generation per year.

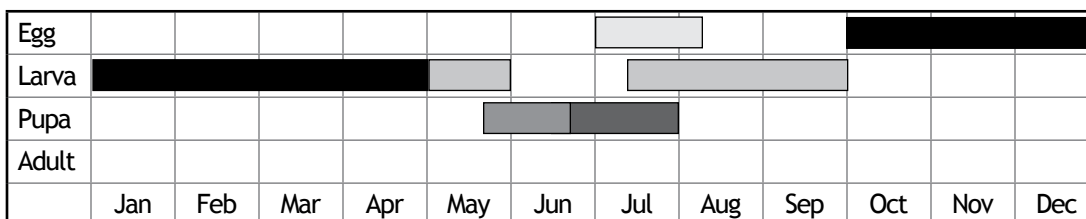


Figure 63 Life cycle of *P. medullana*. Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period.

Habitat preference

This species prefers hot, dry areas. It prefers high density diffuse knapweed. Plants growing in poor, coarse, or gravel soils are ideal.

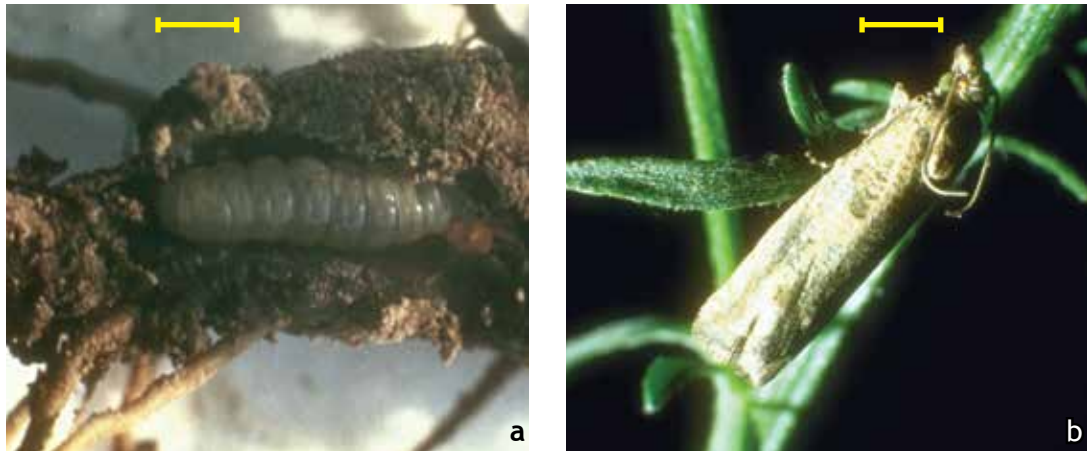


Figure 64 Stages of *Pelochrista medullana*: a. larva (Montana State University Archive); b. adult (Bob Nowierski, Montana State University). Both www.bugwood.org.

Impact

This species prefers diffuse knapweed, but will also attack spotted knapweed at some sites. Damage to the roots is similar to that caused by *Agapeta zoegana*. The larvae reduce root storage capacity and expose the plant to pathogens, but only the third to sixth instars cause measurable damage. Small plants, < 10 mm (0.4 inch) root diameter, can be completely destroyed. Plants that survive insect attack are usually smaller and produce fewer flower heads than unfested plants.

Availability

This moth was released in limited numbers in Idaho, Montana, Oregon and British Columbia. Initially, it was believed to have established; however, to date, there is no evidence of this agent in either the U.S. or Canada.

Comments

Usually only one larva develops per root, likely due to intraspecific competition. Very large roots have been observed to contain up to four.

Pterolonche inspersa

Grey-winged knapweed root moth



| | |
|---------------------|---------------------------------------|
| ORDER | Lepidoptera |
| FAMILY | Pterolonchidae |
| NATIVE DISTRIBUTION | S Europe, Mediterranean |
| ORIGINAL SOURCE | Europe |
| FIRST U.S. RELEASE | 1986 Idaho, Oregon, Utah |
| SPECIES ATTACKED | Diffuse; sometimes spotted, squarrose |
| NONTARGET EFFECTS | None reported |
| U.S. ESTABLISHMENT | |

Figure 65 Adult *P. inspersa* (USDA APHIS PPQ Archive, www.bugwood.org).

Description

Eggs are black and oval-shaped with a slight depressed center. Larvae are pearly white with inflated segments and have small, brown head capsules. Adult moths can be up to 8 mm (0.32 inch) long. Their wings are light brown, exhibiting a silvery sheen. Wingspans are up to 20 mm (0.8 inch). When at rest, the wings are held close to their sides.

Life cycle

Adults emerge from late summer through early fall, mate and lay eggs during their short, 15–20-day life span. Eggs are laid singly or in small groups on the under-surfaces of rosette leaves. A single female may lay 140+ eggs in her lifetime. Larvae hatch within 12 days and mine down the root, feeding on the woody central portion of the root or the soft tissue near the outer edges, causing galls to form. There are five larval instars; third instars typically overwinter within the root and resume feeding the following spring. Larvae construct silken “chimney” tubes that extend from the galls upward to 20 mm (0.8 inch) above the soil surface, where they pupate. The chimneys provide easy exits for the emerging adults. Pupation is 15 days and occurs in early summer. There is one generation per year.

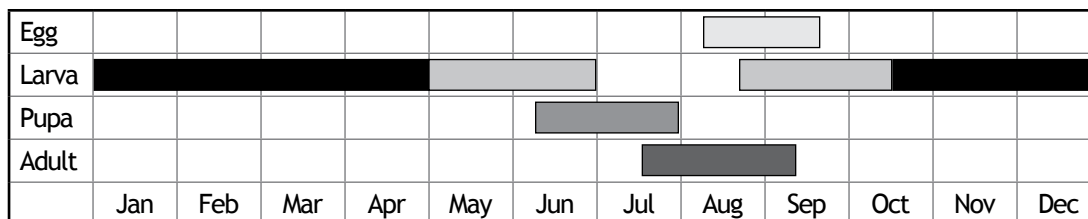


Figure 66 Life cycle of *P. inspersa*. Bars indicate the approximate length of activity for each of the life stages. Black bars represent the overwintering period.

Habitat preference

This species prefers hot, dry sites with low to moderate plant density. It requires a period of drought during summer, so it is only suited to more arid environments. Preferred soils consist of loosely compacted sand or gravel.

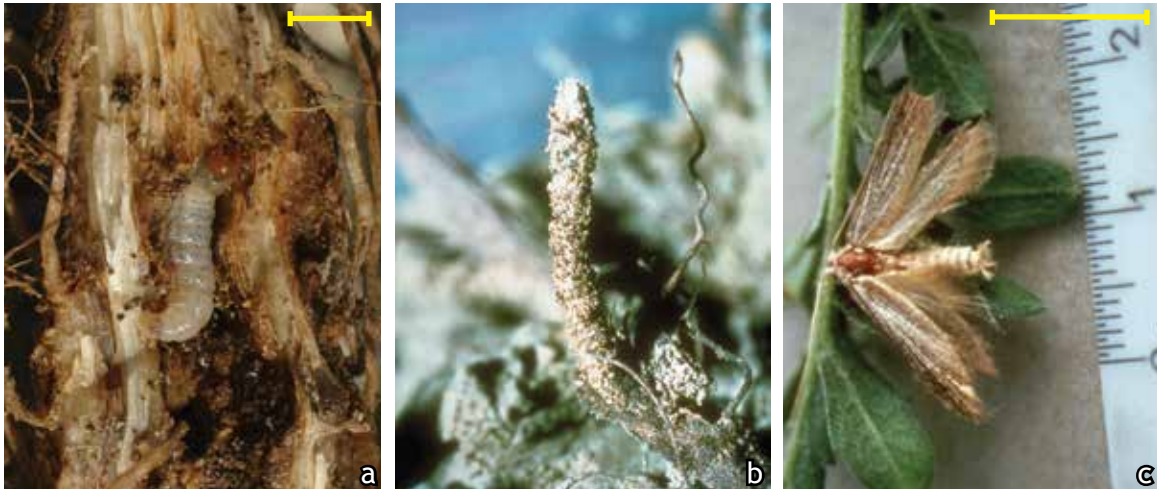


Figure 67 Stages of *Pterolonche inspersa*: a. larva (Eric Coombs, Oregon Department of Agriculture); b. silken chimney tube (USDA ARS European Biological Control Laboratory); c. adult (USDA ARS Archive). b., c., www.bugwood.org.

Impact

Diffuse knapweed is preferred. Larvae feed on roots, which interrupts the vascular flow of nutrients to the plants, thereby decreasing the plant's biomass and flowering ability. Damaged roots become spongy and fragile and easily break apart. Damage attracts other predators, which move into the roots and provide secondary attack.

Availability

This moth was first released in three states in 1986. It established, initially; however, since 2000 it has not been recovered at any of these early sites. There has been evidence of establishment and wide dispersal in British Columbia, but this insect is not currently (2010) available for general distribution in Canada.

Comments

Usually only one larva of this species develops per root, due to aggressive intraspecific competition. However, very large roots have been observed to contain up to four. The moth can co-occur with *Sphenoptera jugoslavica* by feeding below *S. jugoslavica* galls.

At one time, 20% of diffuse knapweed plants at one Oregon site were infested by this species. Populations have since crashed due to the dramatic control of diffuse knapweed by the seedhead weevils (*Larinus* spp.).

CHAPTER 4: ELEMENTS OF A KNAPWEED BIOLOGICAL CONTROL PROGRAM

The results of biological control to treat knapweed may vary greatly from site to site, depending on conditions. Land managers need to develop biological control programs that address management conditions and objectives unique to their area.

Defining your goals and objectives

Defining your weed management goals and objectives is the first and most important step in developing a biological control program. By defining what you want to achieve, you will be able to determine if, when, and where you should use biological control.

As precisely as possible, you must first define what will constitute a successful knapweed management program. For example, the goal of “. . . a noticeable reduction in knapweed density over the next ten years. . .” might be achievable, but is subjective and open to observer bias. Alternatively, the goal of “. . . a 50 percent reduction in knapweed stems over the next three years . . .” is more precise and measurable.

If your goal is to reduce the abundance of knapweed, then biological control might be an appropriate weed-management tool; however, by itself biological control usually will not completely remove knapweed from the landscape. If your goal is to eradicate this weed, then you should plan to employ other weed control techniques instead of or in addition to biological control (see Chapter 5 for more details).

Taking stock: Your infestation and your options

Before embarking on knapweed management activities, you must first understand the scope of your problem, identify areas of special concern, and review and understand all weed management tools available to you in your situation.

Your first step should be to develop a distribution map of your knapweed infestation(s) at a scale that will allow you to address your weed problem in a manner consistent with your land-management objectives and your weed-management resources. For example, in large management areas with significant knapweed infestations and limited resources, aerial mapping of large patches of knapweed may be sufficient to identify priority areas for additional survey and weed-management activities (Figure 68a). In other management areas with small, discrete knapweed infestations, or where an infestation affects your ability to meet management objectives, your weed-management strategy might have to include extensive mapping and analysis of the scope of the infestations (e.g. location, size, density, and cover) (Figure 68b).

Once you determine the scope of your knapweed infestations, review the management tools available (herbicides, mechanical treatments, cultural practices, and biological control) and determine the conditions (when, where, if, etc.) under which it might be appropriate to use each tool or combination of tools (see Chapter 5). Consult your agency or university biological

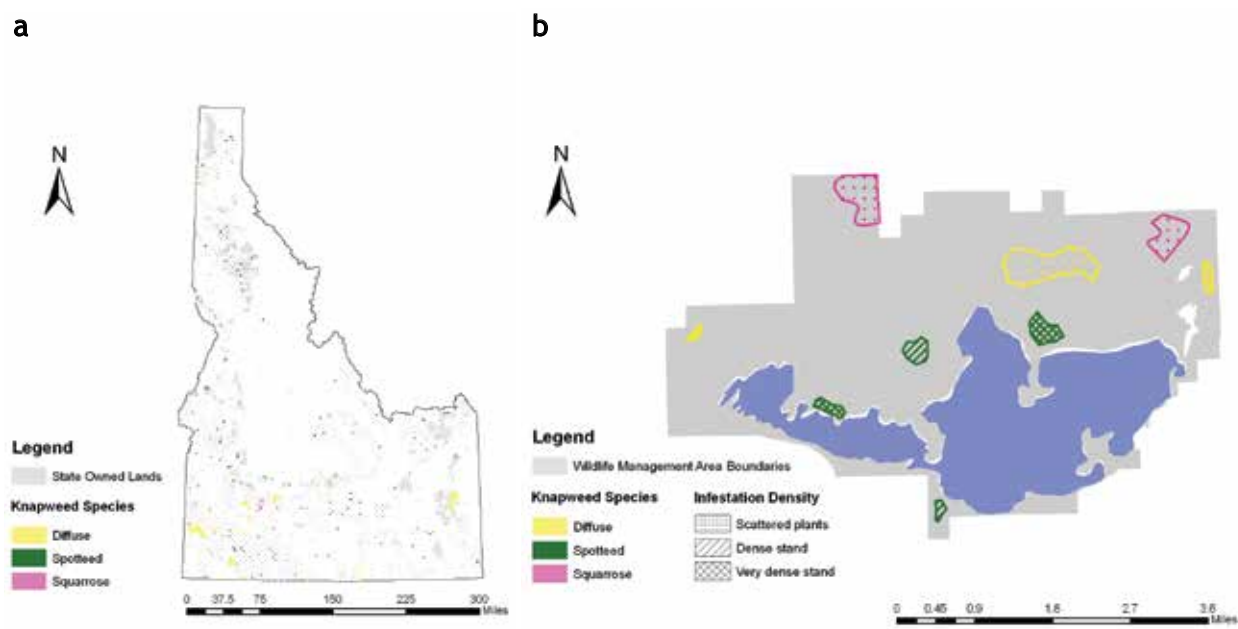


Figure 68 Hypothetical knapweed infestations: a. Idaho land managed by the State of Idaho; b. Mud Lake Wildlife Management Area managed by the Idaho Fish and Game.

control expert, cooperative weed management area, or county weed coordinator/supervisor to learn about other knapweed management activities underway or planned for your area, and the level of control that might be achieved by each.

Identify the resources that will be available for weed management activities, and determine if they will be consistently available until you meet your weed management program objectives. If resources are not available, or will not be available consistently, identify what will happen at the treatment site if the activities are not implemented.

With a map of knapweed infestations in your management area, an understanding of your land management objectives, and a list of the weed management tools available with the level of control you can realistically expect from each, you can identify the sites where biological control would be a good fit.

Developing, implementing, and managing a biological control program

After you've determined that biological control is suitable for treating your knapweed infestations, there are several important factors to consider while planning your approach, including selecting appropriate release sites, obtaining and releasing insects, and monitoring the success of the program (discussed below). If problems are encountered following the initiation of a biological control program, refer to the troubleshooting guide in Appendix I.

Before you begin

There is a fair amount of preliminary work to do before you can implement a biological control release program. Here are a few things you will need to consider.

- Make sure that you can make a long-term commitment to the program. Many biological control programs do not result in visible weed reduction for a number of years (typically 3–5 or more).
- Decide how long you can let biological control activities continue if weed control goals are not being met.

- Discuss your biological control program plans with landowners and land managers. Ask local weed managers about their experiences with biological control. Determine which agents they have used, alone or in combination with other weed control tools, and what level of control they achieved. Would their level of control be acceptable for your management area? Talk to neighboring managers about any activities, such as herbicide use, grazing, or mowing programs they have planned on their land. These measures could have a direct impact on your proposed biological control activities.
- Set short- and long-term goals. For example, a short-term goal might be to release and determine establishment of biological control agents; a long-term goal might be to reduce knapweed density by 50% within 10 years.
- Determine what resources will be consistently available for 5–10 years for implementing, monitoring, and assessing your biological control program. These include
 - committing resources for field equipment and supplies,
 - recruiting and training personnel, and
 - identifying sources of biological control agents.

Selecting biological control agent release sites

Establish goals for your release site

You must consider your overall management goals for a given site when you evaluate its suitability for the release of biological control agents. Suitability factors will differ, depending on whether the release is to be a

- general release, where agents are simply released for knapweed management,
- field insectary (nursery) release, primarily employed for production of biological control agents for distribution to other sites, or
- research release, used to document biological control agent biology and/or the agent's impact on the target weed and nontarget plant community.

A site chosen to serve one of the above roles also might serve additional functions over time (e.g., biological control agents might eventually be collected for redistribution from a research release).

Determine site characteristics

Knapweed biological control agents vary in their habitat and climatic preferences. Consider these preferences when determining the location of your release (see Table 11, page 78). In addition, if your biological control program goals involve establishing permanent monitoring sites, which in turn will require regular inspections, consider the site's ease of accessibility, terrain, and slope.

For practical purposes, a knapweed infestation cannot be too large for biological control releases (Figure 69); however, it might not be large enough. Small, isolated patches might not be adequate to allow biological control agent populations to build up and persist. In such cases, the sites might be better suited for other weed control tactics, such as herbicide applications. An area with at least one acre (0.40 hectares) of knapweed might be considered the minimum size for a release site, but a larger area of infestation is more desirable, especially for field insectaries. Infestations should be contiguous, rather than scattered in patches, so that insects may disperse more easily. Most knapweed biological control agents do best in a moderately dense area of infestation, though some prefer less-dense infestations.

Note land use and disturbance factors

Preferred release sites are those that experience little to no human (or other) disturbance. Fallow sites and natural areas are good choices for biological control agent releases. If a site must be disturbed (e.g., mowed or grazed), the activities should not take place during the spring and summer months when most biological control agents are active above ground.

Sites where insecticide use is routine should not be used for agent releases. Such sites include those near wetlands that are subject to mosquito control efforts, where grasshopper outbreaks routinely require chemical control, or near agricultural fields that are sprayed regularly.

Avoid sites prone to seasonal flooding (Figure 70). Do not use sites where significant conversion will take place, such as road construction, cultivation, building construction, and mineral or petroleum extraction. Do not use sites where burning occurs regularly.

Survey for presence of biological control agents

Examine your prospective release sites to determine if knapweed biological control agents are already present. If an agent you are planning to release is already established at a site, you can still release it to augment the existing population, but it might be better to release it at another site. Also, you should re-evaluate the release of the planned species if a different species of biological control agent is present.



Figure 69 Spotted knapweed infestation suitable for biological control efforts (Norman E. Rees, USDA ARS, www.bugwood.org).



Figure 70 Seasonal floods make this riparian infestation of spotted knapweed less suitable for biological control (Steve Dewey, Utah State University).

Record ownership and access

In general, release sites on public land are preferable to sites on private land. If you must release biological control agents on private land, it is a good idea to select sites on land known for its long-standing, stable ownership and management. Stable ownership will help you establish long-term agreements with the landowner, permitting access to the sites to sample or harvest biological control agents and collect insect and vegetation data for the duration of the project. This is particularly important if you are establishing a field-insectary site, because five years or more of access may be required to complete insect harvesting or data collection. General releases of biological control agents to control knapweed populations require less-frequent and short-term access; you may need to visit such a site only once or twice after initial release. If you are releasing insects on private land, it may be a good idea to obtain:

- written permission from the landowner or land manager allowing use of the area as a release site,
- written agreement by the landowner allowing access to the site for monitoring and collection for a period of at least six years (three years for establishment and buildup and three years for collection), and
- permission to put a permanent marker at the site

You may wish to restrict access to release locations, especially research sites and insectaries, and allow only authorized project partners to visit the sites and collect insects or plants. The simplest solution would be to select locations that are not visible to, or accessible by, the general public. Being practical, most if not all of your sites will be readily accessible, so in order to restrict access you should formalize arrangements with the landowner or public land manager. This will require you to post no-trespassing signs, installing locks on gates, etc. (Figure 71)

Another consideration is the physical access to a release site. You will need to drive to or near the release locations, so determine if travel on access roads might be interrupted by periodic flooding or inclement weather. You might have to accommodate occasional road closures by private landowners and public land managers for other reasons, such as wildlife protection.



Figure 71 Gated access (Chris Schnepf, University of Idaho, www.bugwood.org).

Choosing the appropriate biological control agents for release

You should consider several factors when selecting biological control agents for release at a site, including agent efficacy, availability, and site preferences (Table 11, page 78).

Agent efficacy

Efficacy refers to the ability of the agent to directly or indirectly reduce the target weed's population to below acceptable damage thresholds or cause weed mortality resulting in control. Most of the available data on efficacy is anecdotal, observational, or based on limited experimental data.

It is preferable to release only the most effective, rather than all, biological control agents that might be available against a target weed. Consult with local weed biological control experts, land managers, and landowners to identify the agent(s) that appear(s) more effective given your local site characteristics and management scenarios.

Agent availability

Only 11 of the 13 knapweed biological control insects described in this manual are currently established in the continental U.S., and availability varies greatly between species (see Table 11, page 78). The *Urophora* flies are readily available throughout the U.S. and have several collectable populations in all states with significant knapweed infestations. These insects should be easy to obtain from intrastate and local sources. *Cyphocleonus achates*, *Larinus minutus*, and *Metzneria paucipunctella* are all well-established in the Northwest, with populations continuing to increase. Less widespread but still available for collection in some northwestern states are *Agapeta zoegana*, *Bangasternus fausti*, and *Sphenoptera jugoslavica*. *Chaetorellia acrolophi*, *Larinus obtusus*, and *Terellia virens* are the least common of knapweed biological control agents. Federal agencies and commercial biological control suppliers may be able to assist you in acquiring agents that are not available in your state (see Obtaining and Releasing Knapweed Biological Control Agents, below). County weed managers, extension agents, or federal and university weed or biological control specialists should be able to recommend in-state sources for various knapweed biological control agents.

Release site characteristics

General physical and biological site preferences and characteristics for each agent have been developed from anecdotal observations and experimental data. See Table 11, page 78.

Obtaining and releasing knapweed biological control agents

You can obtain knapweed biological control agents either by collecting them yourself, having someone collect them for you, or by purchasing them from a commercial supplier. Typically, the last two methods will require packaging and shipping from the collection site to your release location (see Collecting Knapweed Biological Control Agents, page 80).

Factors to consider when looking for sources of biological control agents

You do not need to take a “lottery approach” and release all types of biological control agents at a site in the hopes that one of them will work. In fact, some biological control agents will not be available even if you want them, and some have shown to have little or no effectiveness in certain areas. The best strategy is to release the best agent! Ask the county, state, or federal biological control experts in your state for recommendations of agents for your particular region.

- If available, biological control agents from local sources are best. Using local sources increases the likelihood that agents are adapted to the abiotic and biotic environmental conditions present and are available at appropriate times for release at your site. Local sources may include neighboring properties or other locations in your county and adjacent counties. Remember: Interstate transport of biological control agents requires a USDA-APHIS-PPQ permit (see Regulations Pertaining to Knapweed Biological Control Agents, page 85). Get your permits early to avoid delays.
- Some states, counties, and universities have “field days” at productive insectary sites (Figure 72). On these days, land managers and landowners are invited to collect or receive freshly collected knapweed biological control agents for quick release at other sites. These sessions are an easy and often inexpensive way for you to acquire biological control agents. They are good educational opportunities as well, because you can often see first-hand the impacts of various agents on knapweed plant communities.



Figure 72 Collecting insects in diffuse knapweed (Eric Coombs, Oregon Department of Agriculture).

Typically, field days are conducted at several sites in a state and on several dates during the summer. Although designed primarily for intrastate collection and distribution, out-of-state participants may be welcome to participate. (Remember that USDA permits are required for interstate movement and release of biological control agents.) Contact county weed supervisors, university weed or biological control specialists, or federal weed managers for information about field days in your state and/or adjacent states.

Table 11 Summary of general agent characteristics and some components of knapweed biological control agents released in the United States (through 2010). Underlined knapweed species are the preferred hosts.

| AGENT CHARACTERISTICS | | | | | | | |
|-----------------------|------------|--|--|-----------------------------|------------------|---------------------------------------|-----------------------------------|
| TYPE | SPECIES | ATTACK | DAMAGE | EFFICACY | AVAILABILITY | | |
| Seed feeder | Beetle | <i>Bangasternus fausti</i> Broad-nosed knapweed seedhead weevil | Diffuse, spotted, squarrose | Seeds | Low to Moderate | Readily collected from OR and WA | |
| | | <i>Larinus minutus</i> Lesser knapweed weevil | <u>Diffuse</u> , spotted, squarrose, meadow | Seeds, foliage | Very high | Widely established and increasing | |
| | | <i>Larinus obtusus</i> Blunt knapweed flower weevil | <u>Spotted</u> , meadow, diffuse | Seeds, foliage | Moderate | Widely established but not dense | |
| | Fly | <i>Chaetorellia acrolophi</i> Knapweed peacock fly | <u>Spotted</u> , diffuse, squarrose | Seeds | Low | Established but not in high densities | |
| | | <i>Terellia virens</i> Green clearwing knapweed fly | <u>Spotted</u> , diffuse | Seeds | Low | Established but not in high densities | |
| | | <i>Urophora affinis</i> Banded knapweed gall fly | Spotted, diffuse, squarrose | Seeds | Moderate to High | Readily available | |
| | | <i>Urophora quadrifasciata</i> UV knapweed seedhead fly | Black, brown, diffuse, meadow, spotted, squarrose | Seeds | Moderate to High | Readily available | |
| | Moth | <i>Metzneria paucipunctella</i> Knapweed seedhead moth | <u>Spotted</u> , diffuse, meadow | Seeds | Low | Widespread in ID, MT, OR, WA | |
| | Root borer | Beetle | <i>Cyphocleonus achates</i> Knapweed root weevil | <u>Spotted</u> , diffuse | Roots | Moderate to High | Widely established and increasing |
| | | | <i>Sphenoptera Jugoslavica</i> Bronze knapweed root borer | Diffuse, squarrose, spotted | Roots | Moderate to High | Readily collected from OR or WA |
| Moth | | <i>Agapeta zoegana</i> Sulfur knapweed moth | <u>Spotted</u> , diffuse | Roots | Moderate | Becoming widespread; low numbers | |
| | | <i>Pelochrista medullana</i> Brown-winged knapweed root moth | <u>Diffuse</u> , spotted | Roots | Low to Moderate | No longer established | |
| | | <i>Pterolonche dispersa</i> Grey-winged knapweed root moth | <u>Diffuse</u> , spotted, squarrose | Roots | Low to Moderate | No longer established | |

Continued next page

| SITE CONDITIONS | |
|--|--|
| FAVORABLE CONDITIONS | UNFAVORABLE CONDITIONS |
| Hot, dry areas | High elevations; areas with prolonged rain |
| Dry, hot sites (and more so than <i>L. obtusus</i>); dense knapweed; well-drained, coarse soils | Compacted soil; grazed with livestock during knapweed bolting stage; prolonged rainfall |
| Cooler and more moist than <i>L. minutus</i> sites; sometimes near water; well-drained, coarse soils | Overly dry; competing vegetation |
| High elevations with scattered (not dense) knapweed and low populations of other seed feeding biological control agents | Dry sites; dense knapweed patches; large populations of <i>Urophora</i> and <i>Larinus</i> spp. |
| Dry sites; south-facing slopes | Areas with prolonged rainfall; north-facing knapweed populations |
| Variety: well-adapted and widespread; does better on mesic sites; dense knapweed | Sparse knapweed populations |
| Variety- it is well-adapted and widespread; more mild winters with high snow cover; diffuse provides suitable seedheads longer | Severe winters; low snow cover; knapweed populations only flowering early |
| Dry sites; mild winter climates; snow cover during winter enhances larvae survival; early flowering knapweed | Extreme winter temperatures; no snow cover in winter; high elevations with late flowering knapweed |
| Hot and dry; loose, well-drained coarse soils; temperate areas; scattered plants between populations to aid in dispersal | Prolonged rainfall; moist, clayey soil; isolated populations with few plants |
| Hot, dry areas; period of summer drought; loose, sandy or gravel soil; open space between plants, thus increasing soil temperature | Prolonged rainfall in summer; dense vegetation in cold site; clay soils |
| Mesic, cooler areas with long growing seasons; roots 2.4 mm (0.1") diameter | Disturbance from mowing, grazing, cultivation; sites over 1000 m (3300 ft) may not allow sufficient time for development |
| Hot, dry areas; high knapweed density; poor, coarse, or gravel soils | Prolonged rainfall; sparse host plant cover; clay soils |
| Hot, dry areas; period of summer drought; low to moderate knapweed density; loose, sandy or gravel soil | Prolonged rainfall in summer; very dense knapweed; clay soils |

Continued from previous page

Collecting knapweed biological control agents

Planning and timing of collection is critical. The species of biological control agent and weather characteristics at your collection and release sites will determine the best time in the season to collect. Ensure that all necessary collection supplies are on hand. Also, accurate identification of the biological control agents is essential. General guidelines for collecting knapweed biological control agents are listed in Table 12. For all species, collect only on a day with good weather; insects are usually not active in rainy and very windy conditions.

Table 12 Recommended timetable and methods for collecting knapweed biological control agents in the United States.

| TYPE | | SCIENTIFIC NAME | LIFE STAGE | PLANT GROWTH STAGE | TIMING | METHOD | |
|-------------|---------------------------------|--------------------------------|-----------------------------------|---------------------------------------|---------------------------------------|-------------------------|--|
| Seed feeder | Beetle | <i>Bangasternus fausti</i> | Adult | Early to late bud | Early summer (June) | Net/aspirator | |
| | | <i>Larinus minutus</i> | Adult | Flowering | Summer (Jul-Aug) | Net/aspirator | |
| | | <i>Larinus obtusus</i> | Adult | Flowering | Summer (Jul-Aug) | Net/aspirator | |
| | Fly | <i>Chaetorellia acrolophi</i> | Larvae or pupae in heads | Mature, senescing | Fall (Sep-Nov); or early spring (Apr) | Whole plant bouquets | |
| | | <i>Terellia virens</i> | Larvae or pupae in heads | Mature, senescing | Fall (Sep-Nov); or early spring (Apr) | Whole plant bouquets | |
| | | <i>Urophora affinis</i> | Larvae or pupae in heads | Mature, senescing | Fall (Sep-Nov); or early spring (Apr) | Whole plant bouquets | |
| | | <i>Urophora quadrifasciata</i> | Larvae or pupae in heads | Mature, senescing | Fall (Sep-Nov); or early spring (Apr) | Whole plant bouquets | |
| Moth | <i>Metzneria paucipunctella</i> | Larvae or pupae in heads | Mature, senescing | Fall (Sep-Nov); or early spring (Apr) | Whole plant bouquets | | |
| Root borer | Beetle | <i>Cyphocleonus achates</i> | Adult | Flowering | Late summer (Aug) | Hand pick or net | |
| | | <i>Sphenoptera jugoslavica</i> | Adult | Flowering | Summer (Jul; early evening) | Net/aspirator | |
| | Moth | <i>Agapeta zoegana</i> | Larvae or pupae in roots | Senescing; or rosette | Fall (Oct-Nov); or spring (Apr-May) | Transfer infested roots | |
| | | <i>Pelochrista medullana</i> | Not presently established in U.S. | | | | |
| | | <i>Pterolonche dispersa</i> | Not presently established in U.S. | | | | |

Beetles

Beetles are best collected in the adult stage. Adult *Bangasternus fausti*, *Larinus* spp. and *Sphenoptera jugoslavica* can be collected with a sweep net (with or without an aspirator) during summer when plants are in flower (early bud for *B. fausti*). *Cyphocleonus achates* can be netted in late summer, but adults are large enough to be hand-picked, as well.

Flies

Sweeping adult flies is possible, though this is not always the best stage for collection as flies are fragile and can be damaged during collection. Consequently, all knapweed flies are best transferred by placing plants with infested seedheads into uninfested patches during late fall or early spring. Transferring infested seedheads may also transfer unwanted parasitoids or other seedhead insects. To avoid this, seedheads can be collected in fall and stored at 39–46°F (4–8°C). Two to three weeks prior to their normal emergence time, bring them to room temperature in rearing cages or breathable, clear containers. Once they emerge, flies can be transferred to new knapweed infestations.

Moths

Adult knapweed moths are just as delicate as adult flies, so are best transferred as larvae. *Metzneria paucipunctella* is best transferred using methods similar to those used to transfer knapweed flies (above). *Agapeta zoegana* can be redistributed by transferring infested plants, dug up by the roots, to new sites in late fall or early spring. Alternately, they can be reared inside following the methods used for flies (see above).

Sweep net

A sweep net (Figure 73) is made of a cotton or muslin hoop, 10–15 inches (25–38 cm) in diameter, attached to a handle 3 feet (0.9 m) long. They can be purchased from entomological, forestry, and biological supply companies, or you can construct one yourself. As their name implies, they are used to “sweep” insects off knapweed.

A sweep is made by swinging the net through the plant canopy. It is best to alternate between sweeping insects off the weed and aspirating them from the net. Sweep no more than 25 times before aspirating. This reduces the potential harm from knocking biological control agents around with debris or from damage inflicted by other insects inside the net.

Sweep netting is a relatively easy and efficient manner for collecting insects from the above-ground portion of plants, and is the ideal method for collecting adult beetles. The best time for sweeping knapweed insects is during the warmest part of the day (between 1 and 6 p.m.), as this is when the beetles are most active. The adult knapweed moths and midges of *A. plagiata* and *Z. giardi*, respectively, are very delicate, and collecting them with sweep nets can be damaging or fatal. Consequently, it is best to use other methods for collecting those biological control species.



Figure 73 Sweeping (Mark Cole, University of Idaho).

Aspirator

Use an aspirator (Figure 74) to suck the insects directly from knapweed or the sweep net. This provides selective sorting (no unwanted or unknown material is inadvertently collected). Aspirating can be done in the field or indoors. When aspirating indoors, cool the insects to make them less active and easier to collect. A variety of aspirators can be purchased from entomological, forestry, and biological supply companies, or you can construct one yourself. For the latter, make sure that tubing reaching your mouth is covered by fine-mesh screening, so you don't inhale small particles or insects.



Figure 74 Aspirator (Laura Parsons, University of Idaho, www.bugwood.org).

Hand-picking

Simply pick the insects from the knapweed plants by hand or tap them into a net or plastic tray using a tool such as a badminton racquet. Forceps or tweezers may be helpful. Hand-picking works best for stationary or slow-moving insects, such as the larvae of *A. plagiata*. Use clippers if collecting galls.

Containers for knapweed biological control agents

The manner in which biological control agents are handled following collection and during transportation to the release site can affect whether they will survive to multiply at the new site. To reduce mortality or injury, it is best to redistribute the agents the same day they are collected.

Following collection, insects need to be transferred to containers to protect them and prevent them from escaping. Containers should be rigid enough to resist crushing but also ventilated to provide adequate air flow and prevent condensation. Unwaxed paperboard cartons (Figure 75) are ideal for all knapweed biological control agents. They are rigid, permeable to air and water vapor, and are available in many sizes. Unfortunately, they are becoming increasingly difficult to find.

As an alternative, you can use either light-colored, lined or waxed-paper containers (e.g., ice cream cartons are particularly suitable) or plastic containers, providing they are ventilated. Simply cut holes in the container or its lid, and cover the holes with a fine mesh screen.

Untreated paper bags (lunch bags) work well for transporting agents across short distances. However, they are fragile and offer little physical protection for the agents within, you must take care to seal them tightly to prevent the agents from escaping, and some biological control agents are capable of chewing through them. If you do use paper bags, fold over the tops several times and staple them shut. Do



Figure 75 Cardboard containers (Martin Moses, University of Idaho, www.bugwood.org).

not use glass or metal containers; it is difficult to regulate temperature, air flow, and humidity, and the glass containers are breakable.

Fill the containers two-thirds full with paper towels to provide a substrate for insects to rest and hide and to help regulate humidity. Include fresh sprigs of knapweed foliage (as food) before adding the agents. Knapweed sprigs should be free of seeds, flowers, and any other insects. Do not place sprigs in water-filled containers; if the water leaks, it will likely drown your agents. Seal the container lids either with masking tape or label tape. Be sure to label each container with (at least) the biological control agent(s) name, the collection date and site, and the name of the person(s) who did the collecting.

Transporting knapweed biological control agents

Keep the containers cool at all times

If you sort and package the agents while in the field, place the containers in large coolers with frozen ice packs. Do not use ice cubes unless they are contained in a separate, closed, leak-proof container. Wrap the ice packs in crumpled newspaper or bubble wrap to prevent direct contact with containers. Place extra packing material in the coolers to prevent the ice packs from shifting and damaging the containers. Always keep coolers out of direct sunlight, and only open the coolers again when you are ready to remove the containers to release the agents or place them in a refrigerator for overnight storage. If you sort and package your agents indoors, place the packages in a refrigerator (no lower than 40°F or 4.4°C) until you transport or ship them.

Transporting short distances

If you can transport your biological control agents directly to their release sites within 3 hours after collecting them and release them the same day or early the next, you need not take any measures other than those already described.

Shipping long distances

You might need to use a bonded carrier service with overnight delivery (e.g., USPS, FedEx, UPS, or DHL) if your release sites are far from your collection sites or you have to deliver your biological control agents to several sites. In such cases, the containers should be placed in insulated shipping containers with one or more ice packs, depending on the size of the packs. Some specially designed foam shippers have pre-cut slots to hold containers and ice packs (Figure 76). This construction allows cool air to circulate but prevents direct contact between the ice and the containers. Laboratory and medical suppliers sell foam “bioshippers” that are used to transport lab or medical specimens. If neither foam product is available, you can use a heavy-duty plastic cooler.



Figure 76 Commercially made shipping container (University of Idaho, www.bugwood.org).

Common Packaging Mistakes

Excess heat—Do not expose biological control agents to direct sunlight or temperatures above 80° F (27° C).

Excess moisture—Remove spilled or excess water in the container.

Lack of air—Provide adequate ventilation; use only air-permeable containers.

Careful packaging is very important regardless of the shipping container you use. Ice packs need to be wrapped in crumpled newspaper, wrapping paper, or bubble wrap, and should be firmly taped to the inside walls of the shipping container to prevent them from bumping against and possibly crushing the insect containers during shipping.

Empty spaces in the shipper should be loosely filled with crumbled or shredded paper, bubble wrap, packing “peanuts,” or other soft, insulating material. Use enough insulation to prevent agent containers and ice packs from shifting during shipment but not so much that air movement is restricted. Tape the container lids shut. Enclose all paperwork accompanying the agents before sealing the shipping container. For additional security and protection, you may place the sealed shipping containers or coolers inside cardboard boxes.

Other factors to consider

- Make your overnight shipping arrangements well before you collect your biological control agents, and make sure the carrier you select can guarantee overnight delivery.
- Plan collection and packaging schedules so that overnight shipments can be made early in the week. Avoid late-week shipments that may result in delivery on Friday, Saturday, or Sunday, which then could delay release of the agents for several days.
- Clearly label the contents of your containers and specify that they are living insects.
- Check with a prospective courier to make sure that they can accept this type of cargo and will not X-ray or otherwise treat the packages in ways that could harm the biological control agents. If the courier cannot guarantee that such treatments will not occur, choose a different carrier.
- Contact personnel at the receiving end, tell them what you are shipping and when it is due to arrive, verify that someone will be there to accept the shipment, and instruct them not to X-ray the container or open it prior to releasing the agents.

Purchasing knapweed biological control agents

A number of commercial suppliers provide knapweed biological control agents. County weed managers, extension agents, or university weed or biological control specialists may be able to recommend one or more suppliers. Make sure that a prospective supplier can provide the species you want and can deliver it to your area at a time appropriate for field release. (You may want to know where and when the agents were collected.) Interstate shipments of knapweed biological control agents by commercial suppliers also require a USDA permit (see page 86 and Appendix II). Determine in advance whether you or the shipper is responsible for obtaining the permit. Do not purchase or release unapproved, non-permitted biological control organisms.

Releasing knapweed biological control agents

Establish a permanent location marker

Place a steel fence post or plastic or fiberglass pole at least 4 feet (1.2 m) tall as a marker at the release point (Figure 77). Avoid wooden posts; they are vulnerable to weather and decay. Markers should be colorful and conspicuous. White, bright orange, pink, and red are preferred over yellow and green, which may blend into surrounding vegetation. Sometimes, conspicuous posts are not practical or suitable at your release site, due to too much human or large animal traffic or a high risk of vandalism, etc. In such cases, mark your release sites with short, colorful plastic tent or surveyor's stakes or steel plates that can be etched or tagged with release information and located later with a metal detector and GPS.



Figure 77 Permanent marker for biological control agent release site (Rachel Winston, MIA Consulting).

Regulations Pertaining to Knapweed Biological Control Agents

U.S., intrastate Generally, there are few if any restrictions governing collection and shipment of biological control within the same state; however, you should check with your state's department of agriculture or agriculture extension service about regulations governing the release and intrastate transport of your specific biological control agent.

U.S., interstate The interstate transportation of biological control agents is regulated by the U.S. Department of Agriculture (USDA), and an approved permit is required to transport living biological control agents across state lines. You should apply for a Plant Protection Quarantine (PPQ) permit from the Animal and Plant Health Inspection Service (APHIS) as early as possible—ideally, at least six months before actual delivery date of your biological control agent. You can check the current status of regulations governing intrastate shipment of weed biological control agents, PPQ Form 526 (Appendix II), at the USDA-APHIS-PPQ website http://www.aphis.usda.gov/plant_health/permits/organism/index.shtml. A recently initiated ePermit process can be accessed at http://www.aphis.usda.gov/permits/ppq_epermits.shtml; this allows the complete online processing of biological control agent permit requests

Canada Canada requires an import permit for any new or previously released biological control agent. Permits are issued by the Plant Health Division of the Canadian Food Inspection Agency. Redistribution of knapweed biological control agents within a province is generally not an issue; however, you should consult with provincial authorities and specialists prior to moving biological control agents across provincial boundaries.

Record geographical coordinates

The coordinates should be determined using a GPS device, and should be done as a complement to, rather than a replacement for, a physical marker. Accurate coordinates will help locate release points if markers are damaged or removed. Along with the coordinates, be sure to record what coordinate system and datum you are using, e.g., Latitude/Longitude in WGS 84 or UTM in NAD83.

Prepare a map

The map should be detailed and describe access to the release site, including roads, trails, and relevant landmarks. The map should be a complement to, not a replacement for, a physical marker and latitude and longitude or UTM coordinates. It will be especially useful for a long-term project in which more than one person will be involved or participants are likely to change. Maps are often necessary to locate release sites in remote locations that are difficult to access.

Complete relevant paperwork at site before or just after releasing biological control agents

Your agency may have release forms for you to complete. Typically, the information you would provide would include a description of the site's physical location, including GPS-derived latitude, longitude, and elevation coordinates; a summary of its biological and physical characteristics and use; the names of the biological control agent(s) released; date and time of the release; weather conditions during the release; and the names of the person(s) who released the agents (see Biological Control Agent Release Form in Appendix III). The best time to record this information is while you are at the field site; don't wait until you are back in the office to do it. Once back in the office, submit the information to your county extension agent, university, or state department of agriculture. Keep a copy for your own records.

Set up a photo point

A photo point is used to visually document changes in knapweed infestations and the plant community over time following release of biological control agents at a site. Use a permanent feature in the background as a reference point (e.g., a mountain, large rocks, trees, or a permanent structure). Pre- and post-release photographs should be taken from roughly the same place and at the same time of year. Make sure each photograph includes your release point marker (Figure 78).

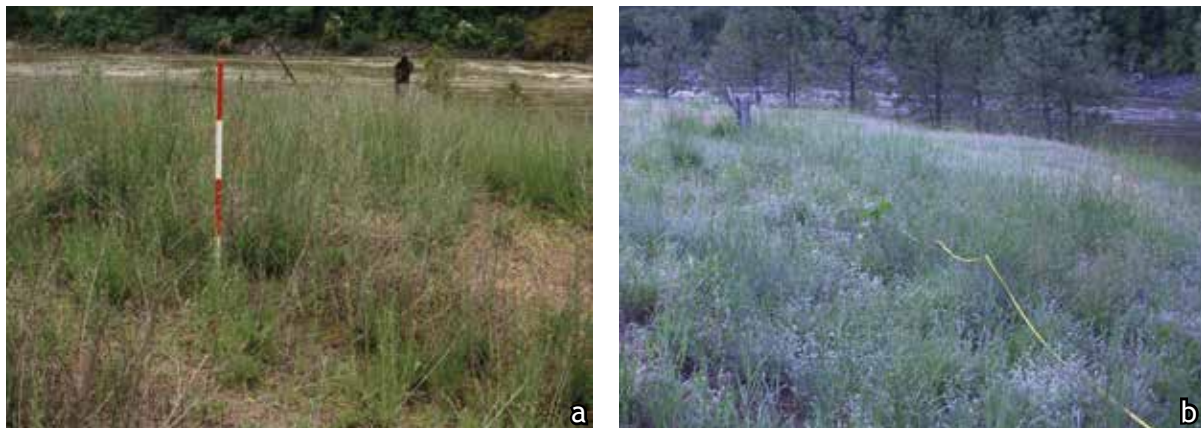


Figure 78 Photo point for spotted knapweed infestation: a. before (Leonard Lake, Forest Service); b. several years after multiple biological control agent releases (Rachel Winston, MIA Consulting).

Release as many agents as possible

For practical purposes, there is probably no maximum number of biological control agents that could be released. In other words, you can never release too many insects. As a general rule of thumb, it is better to release as many individuals of an agent as you can at one site than to spread those individuals thinly over two or more sites. Concentrating the release will help ensure that adequate numbers of males and females are present for reproduction and reduce the risks of inbreeding and other genetic problems. Guidelines for a minimum release size are uncertain for most agents, but releases of 200 individuals or more are encouraged.

Adults of knapweed biological control agents should be released in a group at the marked release point. This is preferred to scattering the agents throughout the knapweed infestation. Releases should be made under moderate weather conditions (mornings or evenings of hot summer days, mid-day for cold season releases). Avoid releasing agents on rainy days; however, if bad weather is expected over several days, it is better to release the insects than wait for conditions to improve, because the agents' vitality may decline with extended storage.

When larvae or pupae are collected in infested knapweed tissue, it is best to broadcast the infested plant material into the litter from previous years' growth. Emerging biological control agents can utilize nearby soil and litter for pupation or shelter. If there are ant mounds or ground dwelling animals in the area, a better option might be to tie a bouquet of infested material to a nearby fencepost.

Documenting, monitoring, and evaluating a biological control program

Weed biological control success is measured by how well the biological control agents reduce the targeted weed densities near or below a pre-determined threshold. Measurement of the weed population in relation to this threshold will determine if your efforts have been successful. The effects of biological control agents usually take much longer to appear than those of herbicide and mechanical control strategies, and at least several years to have full impact on the weed.

The need for documentation

Documenting outcomes--successes and failures--will help generate a more complete picture of biological control impacts, guide future management strategies, and serve education and public relations functions (Figure 79). Documenting initial conditions, coupled with data from periodic evaluations of the biological control agent's establishment and impact, can indicate whether or not the biological control program is working as desired or if additional releases of the same or different biological control agents are needed. Similarly, it can provide critical information for other land managers and help them determine where and when to apply biological controls.

The value of monitoring and evaluation efforts will be greatly enhanced if the information you record is accessible by other land managers and researchers. Institutional memory is short if based



Figure 79 Monitoring *Centaurea* density (Arlie Sommer, University of Idaho).

on personal recollection, and documentation of initial conditions, release locations, successes, and failures will provide critical information to those who will follow you. Documenting successes and failures can help prioritize future research and collection efforts. At the very least, it should help others avoid releasing biological control agents that do not work and concentrate on those that do. Publicly accessible information on release locations, sizes, and outcomes can be extremely useful information for biological control researchers and policy makers. Finally, other land managers need to know the location of your releases so they can avoid engaging in activities, such as summer grazing, mowing, and applying herbicides or insecticides, that would harm your biological control agent populations.

Information databases

Many federal and state agencies have electronic databases for archiving information from biological control releases. We have included a standardized biological control agent release form (Appendix III) that, when completed, should provide sufficient information for inclusion in any number of databases.

At the federal level, the USDA Animal and Plant Health Inspection Service (APHIS) maintains the Cooperative Agricultural Pest Survey (CAPS) database, which is part of the National Agricultural Pest Information System (NAPIS) (<http://ceris.purdue.edu/napis/>). Biological control release information is entered into CAPS by a number of state and federal agency personnel who serve on the state's CAPS survey committee. Contact your local APHIS officials or state department of agriculture for more information on participation.

The USDA Forest Service maintains a database that can store information on biological control agent releases on federal and non-federal lands. As of the writing of this document, biological control releases made on Forest Service lands should be entered into the FACTs database. Other agencies may maintain their own databases for this information. Many of the databases maintained by state and federal agencies have some safeguards in place to prevent undesirable uses of the information they contain.

Information collection

For any weed biological control program, pre- and post-release monitoring is critical to determine if management goals have been achieved. Information on both biological control agent populations and the status of knapweed are collected during monitoring.

Status of knapweed and other plants

- What is the distribution and density of the target knapweed?
- Are the biological control agents causing damage to the target knapweed plants and/or nontarget vegetation? If so, what percentage of the plants are attacked?
- Has there been a change in the knapweed population and distribution since introducing the biological control agents?
- Has there been a change in desirable vegetation at the release site?
- Is there a change in undesirable plants, such as other noxious invasive weeds, at the release site?

Populations of biological control agents

- Are biological control agents established at the release site?
- Are biological control agent populations increasing in size?
- How far beyond the initial release point(s) at a given site have biological control agents spread?
- Are surplus biological control agents collectable at the site?

To address these questions, monitoring activities must be focused on biological control agents, their impacts (damage) on individual knapweed plants, the knapweed population, and the rest of the plant community in the vicinity of the release.

Assessing the status of knapweed and other plants

The ultimate goal of a biological control program is to reduce the abundance of the target weed and enable the recovery of more-desirable vegetation on the site. To determine the efficacy of biological control efforts, there must be monitoring of plant community attributes, such as target weed distribution and density. Ideally, monitoring occurs before biological control efforts are started (pre-release) and at regular intervals after release.

The methods used in pre-release vegetation monitoring should enable land managers to determine later if they are achieving the objectives of the weed biological control program. Often, land managers use reductions in knapweed patch size or density to gauge the success of weed management efforts. Pre-release estimates of knapweed stem density, flowering and vegetative stems, patch size, and patch perimeter at the release sites are frequently measured to enable pre- and post-treatment comparisons.

Land managers may have a goal of changing the structure and composition of the plant community through biological control. Pre-release sampling techniques, which allow managers to describe pre-treatment vegetation, are integral to assessing progress towards this goal.

Pre-release monitoring should include the establishment of control plots where no insects will be released. These plots should be as similar as possible in habitat type (the same soil type, aspect, and exposure) to the release plots. Control sites should be far enough away from release sites so that it is unlikely they will be colonized by biological control agents, at least during the monitoring period of the program. For consistency, the same data collection protocols should be used at control and release sites.

In order to measure biological control agent impact accurately, pre-release and post-release methods for assessing plant densities after biological control agents are released must be the same. Post-release assessments should be planned annually for at least three to five years after the initial agent release.

There are many ways to qualitatively (descriptively) or quantitatively (numerically) assess knapweed populations and other plant community attributes at release sites.

Qualitative (descriptive) vegetation monitoring

Qualitative monitoring uses descriptive elements of knapweed at the management site. Examples include listing plant species occurring at the site, estimates of density, age and distribution classes, visual infestation mapping, and location of the photo points. Qualitative monitoring provides insight into the status or change of knapweed populations. However, its descriptive nature does not generally allow for detailed statistical analyses. Data obtained in qualitative monitoring may trigger more intensive monitoring later on.

Quantitative vegetation monitoring

The purpose of quantitative monitoring is to measure changes in the knapweed population before and after biological control agent release (Figure 80). It may be as simple as counting flowering knapweed plants in an area, or as complex as measuring plant height, flower and seed production, biomass, or species diversity. If designed properly, quantitative data can be statistically analyzed and give precise information on population or community changes.



Figure 80 Quantitative vegetation monitoring (Mark Schwarzländer, University of Idaho).

The Standardized Impact Monitoring Protocol (SIMP) used by the state of Idaho for weed and vegetation monitoring is described in Appendix IV. It is a combination of qualitative and quantitative elements and can be easily modified to meet your personal or agency needs. For additional vegetation monitoring protocols, see Appendices VI and VII.

Assessing impacts on non-target plants

Sampling of vegetation other than knapweed should be included in a vegetation monitoring program in order to assess potential non-target impacts of the biological control program. Specific methods will depend on the species targeted for sampling in particular areas.

Changes in abundance of other desirable or undesirable vegetation

Biological control agent releases, among other land management strategies, can affect the presence and relative abundance of many other plant species, even though the agents do not directly utilize them. If biological control works to remove knapweed at a site, it will create an empty niche to be filled by alternative—hopefully desirable—vegetation. Within the overall management plan for your site, it may be important to document the changes in other vegetation after you release your biological control agents.

Depending on your program goals, you may need to document quantitative and/or qualitative changes for groups of plants, such as native forbs or exotic perennial grasses, or on individual species, such as a rare plant or a food plant for a native butterfly.

Plant species may be considered beneficial (e.g., native and introduced forage plants), or deleterious (e.g., other invasive weeds). One important management goal should be to avoid invasion of a site by another exotic weed after successful biological control of knapweed; in other words, you do not want to replace one invasive weed with another. For this reason, we strongly recommend that you monitor populations of other exotic weeds that are known to be problematic in your area.

You will need to clearly define site management goals and become familiar with the plant communities at your release location and nearby sites. You can easily modify the vegetation sampling procedures described above to monitor changes in density and/or cover for knapweed as well as other plant species, both before and after you release your biological control agents.

Direct impacts of biological control agents on nontarget plants

The host ranges of the currently approved knapweed biological control agents are restricted to *Centaurea* species (see Chapter 3). There are 28 species and three hybrids of *Centaurea* currently established in the U.S. and Canada, though all are introduced and none are native. However, there are two species of closely-related *Plectocephalus* (formerly classified as *Centaurea*) which are native

to North America. Both resemble some species of knapweed.

Plectocephalus rothrockii is largely restricted to moister canyon sites in the Sierra Madre Occidental of Mexico and associated ranges of Arizona and New Mexico. This range overlaps with only two of the invasive knapweeds of this manual: spotted and diffuse. *Plectocephalus americanus* (Figure 81) is found throughout the Southwest and Midwest of the U.S., a range overlapping with all knapweeds in this manual except squarrose. Care should be taken in the management of your knapweed biological control program to ensure that *Plectocephalus* and any other native species are identified and monitored along with knapweed.



Figure 81 American basketflower, *Plectocephalus americanus*, a species closely related to *Centaurea* and native to North America (pschemp).

The first step in addressing possible non-target attacks on native species is to become familiar with the plant communities present at and around your release sites. A visual, pre-release survey will locate native plants that are present. You may have to consult with a local botanist, if available, for help. If your knapweed infestation occurs within the range of the native *Plectocephalus*, a local plant authority or herbarium records from a university or other research institution might be able to identify for you precisely where these natives might be growing.

If you do find one or more native *Plectocephalus* at a potential biological control release site, you should not immediately cancel plans to release biological control agents; knapweed insects have never been recorded feeding on these plant species. The vegetation sampling procedures described above can be modified to help you monitor changes in density and/or cover of the native species, before and after biological control agents are released. Closely monitoring the natives will help you determine if biological control agents utilize the natives in any way.

If you observe approved biological control agents feeding on and/or developing on any native species, collect samples and take them to a biological control specialist in your area. Alternatively, you may send the specialist the site data so he or she can survey the site for non-target impacts. Be sure not to ascribe native plant damage to any specific insect and thus bias its identification.

Assessing knapweed biological control agent populations

All biological control agents go through a population cycle of gradual increase, peak, and decline during the season. It is easier to assess insect establishment when populations are peaking, so we recommend you make multiple visits to a site throughout the season and sample when populations appear highest. Populations of some biological control agents take two to three years to reach detectable levels. Thus, if no agents are detected a year after release, it does not mean that the insects failed to establish. Revisit the site at least once annually for three years. If no evidence of insects is found, either select another site for release or make additional releases at the monitored site. Consult with your county extension educator or local expert on biological control of weeds for their opinion.

General biological control agent surveys

If you wish to determine whether or not a knapweed biological control agent has established after initial release, simply find the biological control agents themselves and/or evidence (that is, distinctive plant damage) of their presence (Figure 82, Table 13). Begin looking for biological control agents where they were first released. If you do not find any, continue to explore the area around the release site. Sometimes biological control agents do not like the area where they were released and move to patches of knapweed nearby.



Figure 82 Diffuse knapweed damaged by *Larinus minutus* (Eric Coombs, Oregon Department of Agriculture).

Additional monitoring methods

A systematic monitoring method is needed to determine the density of insects at the release site or how far the biological control agents have spread from the release point. Numerous approaches can be taken in setting up a monitoring program; these will vary depending on the information you wish to obtain.



Figure 83 Pheromone trap (Sandy Kegley, Forest Service, www.bug.org).

A simple method for monitoring the abundance of knapweed biological control agents is described in Appendix IV. Used by the state of Idaho, this approach is simple, efficient, and sufficiently versatile to allow for the collection of information from the same sites over multiple years. It is based on counting adults. Alternative general biological control agent monitoring forms can be found in Appendix V. These alternative methods rely on dissecting infested knapweed seedheads and roots. The protocols described in both sets of forms can be modified easily to meet your or your agency's needs.

Yet another method for monitoring knapweed biological control agents utilizes pheromone traps. Sex pheromones are chemical attractants (odors) exuded by insects to attract the opposite sex. They are highly specific to one species of insect and are often used in insect pest management. The pheromone is artificially synthesized, packaged, placed in a trap and set out in the field. In the biological control of knapweed, this method is currently only available for the knapweed root moth, *Agapeta zoegana*. The pheromone trap, called a Delta trap (Figure 83), is used to attract male moths and is considered by some to be the best monitoring method for this insect.

Table 13 Life stages/damage to look for to determine establishment of knapweed biological control agents.

| TYPE | SCIENTIFIC NAME | STAGE | WHERE TO LOOK | WHEN TO LOOK | DAMAGE | |
|-------------|---------------------------------|--|-----------------------------------|--|--|--|
| Seed feeder | <i>Bangasternus fausti</i> | Adults | Plant tops during heat of day | Late Spring (May-Jun) | Adults do not do any appreciable damage | |
| | | Larvae | Seedheads | Summer (Jun-Aug) | Dissection reveals feeding damage | |
| | <i>Larinus</i> spp. | Adults | Foliage during heat of the day | Summer (Jun-Jul) | Plants are defoliated and deformed | |
| | | Larvae | Seedheads | Late Summer (Jul-Sep) | Dissection reveals feeding damage | |
| | Fly | <i>Chaetorellia acrolophi</i> , <i>Terellia virens</i> , <i>Urophora</i> spp. | Adults | Foliage during heat of the day | Summer (Jun-Aug) | Adults do not do any appreciable damage |
| | | | Larvae | Seedheads | Sum-Spring (Jul-Apr) | Dissection reveals feeding damage and/or galls |
| Moth | <i>Metzneria paucipunctella</i> | Adults | Foliage at dusk | Summer (Jun) | Adults do not do any appreciable damage | |
| | | Larvae | Seedheads | Sum-Spring (Jul-Apr) | Dissection reveals feeding damage | |
| Root borer | <i>Cyphocleonus achates</i> | Adults | Plant tops during heat of day | Late Summer (Jul-Sep) | Adults do not do any appreciable damage | |
| | | Larvae | Roots | Fall-Spring (Sep-Jun) | Dissection reveals feeding tunnels and frass | |
| | <i>Sphenoptera jugoslavica</i> | Adults | Rosette leaves | Summer (Jul) | Adults do not do any appreciable damage | |
| | | Larvae | Roots | Fall-Spring (Aug-May) | Dissection reveals feeding tunnels and frass | |
| | Moth | <i>Agapeta zoegana</i> | Adults | Low foliage by day; black light at night | Late Summer (Jul-Sep) | Adults do not do any appreciable damage |
| | | | Larvae | Roots | Fall-Spring (Aug-May) | Dissection reveals feeding tunnels and frass |
| | <i>Pelochrista medullana</i> | | Not presently established in U.S. | | | |
| | <i>Pterolonche inspersa</i> | | Not presently established in U.S. | | | |

CHAPTER 5: AN INTEGRATED KNAPWEED MANAGEMENT PROGRAM

Introduction

Integrated weed management (IWM) is a systems approach to management of undesirable plants. IWM is described in the Federal Noxious Weed Act as a system for the planning and implementation of a program using an interdisciplinary approach to incorporate multiple methods for containing or controlling an undesirable plant species or group of species. Components include but are not limited to:

- Education and outreach
- Prevention
- Monitoring
- Early Detection and Rapid Response (EDRR)
- Biological controls (insects, mites, or pathogens)
- Physical or mechanical treatments (tilling, mowing, etc.)
- Cultural practices (grazing, reseeding, etc.)
- Chemical treatment
- General long-term land management practices

An integrated, coordinated approach to weed management has two interdependent goals.

1. To develop a long-term plan to manage all land in a designated area, with landowners and land managers working together towards effective management.
2. To implement the most effective weed control methods for the target weed, regularly assessed and adjusted as needed.

A program that integrates multiple control methods, such as biological agents, chemical controls, and cultural practices, is far more likely to achieve long-term success against knapweed than any single control method used alone.

Integrating biological control

Classical biological control has been applied to many invasive weed species, and there are several examples in which both single- and multiple-agent introductions have successfully controlled the target weeds. The use of biological control agents alone to control weeds can be effective with some invasive plants, including knapweed in some locations, but may take three to five years or more to reduce weed populations to manageable levels. The success rate for classical biological control may increase when multiple species of biological control agent are used, so long as the different species attack different parts (leaves, stems roots) of the target weed at different times during the growing season, or are released over a larger range of infestation.

Biological control agents are not going to work against knapweed every time at every site; integration with other management tools or simply resorting to other tools may be required and is often encouraged in order to attain knapweed management objectives. Some agents, particularly *Larinus minutus* alone and/or in conjunction with root-feeding insects, have helped

reduce knapweed densities in many regions (Figure 84). However, *L. minutus* and other biological control agents have not established in all areas where knapweed occurs. Even when established, biological control agents do not eradicate the target weed. Where ideally suited, biological control can maintain knapweed densities below economically significant levels.



Figure 84 Photo points for diffuse knapweed infestation: a, c., before; b, d., years after *Larinus minutus* introductions (Eric Coombs, Oregon Department of Agriculture).

Land managers appreciate that successful, long-term knapweed control programs must be cost-effective. Because this weed occurs in a wide variety of environments across North America (Figure 85), no single control method can be successful in all infestations. Land managers also know they must operate under social constraints, which will limit the weed management tools they can use in sensitive areas, such as wilderness, near waterways, and on public lands. A wide variety of successful weed management methods, including herbicide mixtures, selective grazing practices, reseeding, and biological control agent releases, have long been used.



Figure 85 Various habitats where spotted knapweed can thrive: a. bays and inlets (John M. Randall, The Nature Conservancy); b. gravel pit (Michael Shephard, Forest Service); c. open pasture (L.L. Berry); d. open forests (Rachel Winston, MIA Consulting); e. dry sagebrush steppe (Steve Dewey, Utah State University); f. along a mountain stream (Steve Dewey, Utah State University) (a-c, e, f. www.bugwood.org).

Weed control methods used to manage knapweed

The most commonly cited activities for the control of invasive plants are listed in the introduction to this chapter. These activities are described in detail below, with emphasis on how to integrate each with biological control methods (where applicable). Knapweed plants within a population may show considerable variation in growth form, response to stress, and flowering frequency. Therefore, management strategies must be site-specific.

Education and outreach

Education and outreach efforts should be a significant component of any IWM strategy and program, regardless of the other weed control methods employed. Efforts should include activities aimed to increase public awareness of noxious weeds, the problems they cause, their distribution, and ways to manage them.

Prevention and exclusion

Prevention and exclusion activities are aimed at areas not currently infested by knapweed, and are intended to keep uninfested areas weed-free. Though spotted knapweed is already present throughout much of North America, there are many areas where it remains at low densities. Other knapweed species are far less widespread. Preventing further introduction and spread to uninfested areas is much more environmentally desirable and cost-effective than is the subsequent treatment of large-scale infestations.

Knapweed is spread by the movement of seed-contaminated hay, wind, wildlife, water, or motorized equipment (Figure 86) into uninfested areas. Where these factors can be controlled, preventing the spread of knapweed requires cooperation among all local landowners. It is important to ensure that possible invasion avenues are identified and management actions are taken to reduce the risk of spread. Actions would include minimizing soil disturbances and regularly monitoring uninfested sites. Prevention and exclusion activities are typically paired with education efforts. Examples of exclusion efforts include weed-free forage programs, state seed laws, weed screens on irrigation water intake, mandatory equipment cleaning before entering uninfested sites, and seeding all disturbed sites with desirable vegetation after soil disturbance.



Figure 86 Diffuse knapweed with mature seed heads being transported by vehicle undercarriage (K. George Beck & James Sebastian, Colorado State University, www.bugwood.org).

An early-detection and rapid-response (EDRR) program is a specific protocol for tracking and responding to infestation spread. It relies heavily on education and outreach activities to be effective. Knapweed EDRR programs take a two-prong approach to controlling knapweed's spread: an education component, which instructs the public on how to detect and identify knapweed, followed by a rapid response to eradicate the weed at all verified locations.

Biological control

Biological control involves the use of living organisms, usually insects, mites, or pathogens, to control a weed infestation and recreate the balance of plant species with their natural predators and pathogens. Classical biological control focuses on the introduction of natural enemies from the origin of the invasive weed. This method of management is most suitable for large scale populations. For new infestations or satellite outbreaks, more rapid control methods, such as chemical treatment with or without mechanical control, should be utilized. Refer to Chapter 3 for detailed descriptions of the biological control agents currently approved for use against knapweed.

Physical or mechanical treatment

Physical treatment utilizes hand pulling, hoeing, tilling, or mowing to disrupt and remove weeds and is the oldest method of weed control. Mulching can be used to control regrowth. Physical methods can be very effective in controlling knapweed, but are labor-intensive.

Hand pulling/hoeing

Hand pulling and hoeing may be successful on small populations of knapweed if they are applied persistently (Figure 87). It is important first to remove as much of the root as possible, while minimizing soil disturbance, and second to remove all pulled/hoed knapweed plant parts from the area to prevent possible seed dispersal.



Figure 87 Hand-pulled spotted knapweed (Steve Dewey, Utah State University, www.bugwood.org).

Tilling

Tilling is not usually compatible with biological control efforts. Tilling and disking frequently disrupt and destroy biological control agents overwintering in knapweed roots and seed heads or in soil litter.

Fortunately, knapweed is not usually a problem in cultivated crops, where tilling is done. Tilling will control knapweed, providing roots are cut at least seven inches below the soil surface, and it is done on a routine basis. A single tilling event may lead to the spread of knapweed in mature stands, because tillage creates an ideal seed bed. Control is enhanced when an herbicide treatment is used, when competitive pasture species or crops are sown and fertilizer is added. Tilling is generally not practical or desirable in wildlands and nature preserves.

Mowing

Cutting back the above-ground portion of a plant will remove top growth and can reduce knapweed seed production and significantly decrease populations, especially if the plants are mowed frequently during the growing season. Mowing should be done as close to the ground surface as possible. If the mowing height is too high, the plant may regrow in a sprawling form and produce flowers below the cutting height. Likewise, mowing conducted after plants have started producing seed may help distribute seed.

A single mowing treatment does not injure the root system, and plants can easily resprout from the root crown. Long-term control with mowing is not possible, especially with perennial species, unless done often enough so that root carbohydrates are depleted and roots die. However, frequent mowing is too costly or infeasible for large and remote infestations on rough terrain. Alternatively, mowing can be used to reduce non-target plant cover and litter prior to fall herbicide applications as this will improve coverage of the chemical on knapweed regrowth.

Prior to mowing, it is important to consider the life cycles of the biological control agents and when and where they will be on the plant. For example, mowing excess plant litter during late winter and very early spring when *Agapeta zoegana*, *Bangasternus fausti*, *Cyphocleonus achates*, *Larinus* spp. and *Sphenoptera jugoslavica* are overwintering in soil litter or knapweed roots is compatible with biological control. However, mowing during this same time period would destroy the larvae or pupae of *Chaetorellia virens*, *Metzneria*

paucipunctella, and *Urophora* spp. Mowing during spring and early summer when most species are active in some stage in the above-ground portion of knapweed could kill large numbers of insects.

Cultural practices

Cultural methods of weed control, including seeding with competitive species, burning, and grazing, can enhance the growth of desired vegetation, which may slow the invasion of noxious weeds. Regardless of which method is used, all cultural control techniques are more successful when combined with other control methods, such as biological controls and chemical treatments.

Seeding competitive species

Knapweeds are all strong competitors, and planting competitive species as the sole control method is unlikely to control knapweed populations. Instead, revegetation is best used in combination with other control tactics. If knapweed is suppressed by one or more alternative control methods, but its ecological niche remains unfilled, reinvasion by knapweed or by other undesirable species will likely occur. Long-term management of this weed requires the establishment and maintenance of desirable competitive species. Studies addressing the effects of different species on *Centaurea* growth have found perennial grasses to provide significant competition to knapweed species. Other studies found that growing taprooted forbs along with grasses increases niche occupation and may be more effective in minimizing invasion of knapweeds than grasses alone.

Seeding can be used to help establish competitive native species, such as grasses and forbs, in a knapweed infestation (Figure 88). The choice of plant species to be seeded should reflect site conditions, management, and future use. In a greenhouse study, bluebunch



Figure 88 Seed sowing equipment (Thomas C. Croker, Forest Service, www.bugwood.org).

wheatgrass (*Pseudoroegneria spicata*) and northern sweetvetch (*Hedysarum boreale*) proved to be good competitors against spotted knapweed when grown together. Both species are native to the American Northwest. However, a different study found bluebunch wheatgrass and Idaho fescue (*Festuca idahoensis*, another native grass) to be ineffective competitors. Numerous introduced grasses help keep knapweeds in check, but these are often invasive weeds themselves. Consult your local county extension agent or Natural Resource Conservation Services representative for the best alternatives for

your region. Controlling knapweed prior to seeding is important. Herbicides may reduce knapweed vigor and allow establishment of the desired species; contact your local county extension agent or weed control authority for current recommendations for chemicals.

Incorporating biological control agents with re-seeding has been difficult, primarily because the methods used to establish a productive stand of competitive species are not always compatible with the establishment and survival of biological control agents. In order to establish a suitable site for re-seeding, either an area must be tilled to provide an acceptable seed bed and/or herbicides such as glyphosate must be applied to reduce competition from knapweed. Tilling can disrupt and destroy biological control agents overwintering in soil

litter and plant roots, and heavy herbicide use will reduce the knapweed shoots on which some biological control agents feed, thus hindering establishment of biological control agent populations.

Seeding of competitive species using a no-till seeder would be less disruptive than conventional seeding techniques to an established knapweed biological control population. Unfortunately, no-till seeding is frequently unsuccessful due to the thick thatch of dead knapweed stems often found in old stands. Some intensive management techniques establish competitive species first, using biological control agents only after the seeded species have become established and knapweed has begun to re-grow.

Prescribed fire

Although this method of cultural control is used against many domestic and exotic plants (Figure 89), the majority of studies addressing this method demonstrate that fire increases the frequency and density of knapweed cover. Fire may kill above-ground growth and has successfully reduced populations of knapweed in Midwestern grass prairies; however, the stout taproot survives most fires and enables plants to re-sprout from the root crown (Figure 90). Burning creates areas of bare soil, increasing the amount of sunlight reaching the soil and encouraging the sprouting of resilient knapweed seeds from the soil seed bank. Likewise, repeated burning may deplete the soil of organic material and thus favor this weed and other undesirable plants. Therefore, burning is not an effective method for controlling knapweed spread. It can, however, be useful for burning off dead plant litter, thus allowing herbicide applications to be more effective.

Biological control agents must be able to survive controlled burns, and this often depends on the timing of the burn. Generally, soil-inhabiting agents are able to survive fast-moving, low- or moderate-intensity fires. If a fire event occurs while the biological control agents are in the adult stage, many can often escape the fire by flying off, readily re-establishing on recovering knapweed not killed during the fire event. Though this is not possible for *Cyphocleonus achates*.



Figure 89 Prescribed fire on diffuse knapweed (Steve Dewey, Utah State University, www.bugwood.org).



Figure 90 Squarrose knapweed re-sprouting after fire (Steve Dewey, Utah State University, www.bugwood.org).

Grazing

Numerous studies exploring grazing on knapweed species have been done, but have yielded contradictory results. In some cases, it has been noted that when knapweed occurs in hay or on rangeland, the feeding value to livestock decreases. Mature knapweed plants, especially diffuse and squarrose knapweed, are coarse and fibrous and the spines on the bracts can be very irritating or even cause injury to the mouths and digestive tracts of grazing animals. Compounds in the leaves of spotted knapweed may make this species unpalatable to livestock by negatively affecting the activity of anaerobic rumen microorganisms.

Despite these findings, other studies show that rosettes of many knapweed species are surprisingly high in protein content and are readily grazed by sheep and goats (cattle have a difficult time eating the low-growing leaves of knapweed rosettes but will feed on bolting plants) (Figure 91). Spring and fall grazing by sheep and goats can be effective at reducing knapweed flower production and density of young plants, respectively. This is especially true when young animals are pre-conditioned to feed on knapweed. Some studies show



Figure 91 Grazing sheep (USDA ARS, www.bugwood.org).

that despite the lower palatability and protein content in late summer, goats will feed readily on knapweed in the bud to flowering stage.

The majority of research indicates that careful grazing practices, including strict observations of both knapweed and desirable forage species before and during grazing events, can help suppress knapweed growth, flower, and seed production in many settings. To minimize weed invasion, grazing systems should alter the season of use, promote litter accumulation, and rotate or combine livestock types and grazing locations to allow grazed plants to recover before being regrazed. Desirable species must fully

recover from the prior grazing before being regrazed. When utilizing grazing as a control tool, keep in mind that timing and execution are very important. Sheep have been observed to pass viable seed of spotted knapweed up to seven days after consumption. Furthermore, improper or over-grazing can result in conditions conducive to knapweed invasion, such as bare ground and the absence of desirable forage species--the very conditions that may have led to the original infestation.

Integration of grazing and biological control agents for knapweed may be complicated. When knapweed is suppressed via grazing, a good proportion of the plants get trampled. Both trampling and consumption of foliage will kill eggs on leaves, juvenile biological control insects developing in buds, and adult insects hiding or overwintering in leaf litter. Grazing can best complement biological control when the life stages of the insects established at a particular site are taken into account before animals are released on a knapweed infestation. Research is currently underway to determine the best means of integrating biological control and grazing management to suppress knapweed.

Chemical control

Herbicides are important tools for controlling noxious weeds and are available for knapweed control in a variety of environments. Herbicide usage is most applicable for small infestations, including new populations and satellite outbreaks, and for use on the leading edge of large advancing populations. Herbicides are often too costly to be of practical value for treating extensive infestations of knapweed. Repeated applications are often required to keep any species of knapweed in check. Small patches may be missed, the soil seed bank must be taken into account for several years, and potential damage to associated vegetation must also be considered, especially in natural areas. Herbicides are best used as part of a larger, integrated system, which might include planting desirable competitive species and utilizing grazing and biological control methods. Some studies suggest grazing prior to herbicide application will help remove nontarget plant growth that would otherwise intercept the spray. Likewise, burning prior to herbicide applications will remove any dead plant litter and thatch, thus increasing the contact area on knapweed leaves following regrowth (Figure 92).



Figure 92 Using herbicides to treat squarrose knapweed regrowth following a burn (Steve Dewey, www.bugwood.org).

Herbicide registrations change frequently, and can result in fewer or more available herbicides or changes in permissible herbicide application practices. In addition, herbicide timing, application rates, and restrictions vary by state and location. Please consult your local weed officer or county agricultural extension agent to learn which herbicides currently work best in your area and when to apply them in your situation. For specific recommendations, refer to the Pacific Northwest Weed Management Handbook, an annually revised Extension publication from the Extension Services of Oregon State University, Washington State University and the University of Idaho. Some of the most widely used products include the following.

- 2,4-D should be applied when the plants are in the rosette to early bolt growth stage. Application of 2,4-D after stem elongation is not as effective. This herbicide has no residual control, so annual spraying is necessary until no seedlings are detected. This may require several years of annual treatment, and should be accompanied by revegetation of desirable species. If used for spot treatments in early spring, it will help control plants germinating from seed and prevent them from flowering. It can be used in tank mixtures for broadcast applications to kill aboveground growth of knapweed and will not kill grasses.
- Glyphosate is best used on actively growing knapweed when most plants are at the bud stage. This is a non-selective herbicide; it kills forbs and grasses and so should only be used where loss of non-target vegetation is acceptable. Glyphosate does not have soil residual effects, so annual spraying is necessary until no seedlings are detected. This may require several years of annual treatment, and its use should be followed by revegetation of desirable species.

- Use picloram on new knapweed growth in fall or in spring on plants in the rosette to bolting stages. Treatment made in bud stage may not prevent seed production in the year of application, but seed germination will be markedly reduced. This is a selective treatment that, at the suggested rate, will not damage perennial grasses, but will kill desirable legume species. It has a long soil residual period, which will reduce regrowth from seedlings for two to seven years.
- Dicamba should be used when the knapweed is in the bud to bloom stage. It is often mixed with others for increased control. This herbicide will provide good control of knapweeds, but with a shorter soil residual period than picloram, so follow up treatments with 2,4-D when plants are in the rosette to early bolt growth stage for a minimum of two years may be needed to prevent reinfestation by seedlings. There is still some residual activity, and this herbicide may kill desirable legume species, so its use should be limited when in conjunction with forb revegetation efforts. It does not kill grasses.
- Clopyralid is best used when applied on fall- or spring-emerged rosettes. It is often mixed with others for increased control. It will provide good control of knapweeds, but with less soil residual than picloram or dicamba, so follow up treatments are often required. There is some residual activity, and this herbicide may kill desirable legume species, so its use should be limited when in conjunction with forb revegetation efforts. This herbicide does not kill grasses.
- Aminopyralid can be applied in the spring or early summer on rosettes or bolting plants or, alternately, in the fall on rosettes. Aminopyralid can be very damaging to desirable forbs, especially legumes. However, this herbicide can often be applied near many tree species where dicamba and picloram cannot be used. This herbicide does not kill grasses. It has a soil residual period, which will reduce regrowth from seedlings in subsequent years.

Application timing is very important to ensure the most effective use of herbicides. It is good to treat knapweed with fall applications when plants are storing reserves in the root system for winter. Spring-emerged seedlings or rosettes are also often vulnerable and easier to control. Location, species, growth stage, stand density, land use, and environmental conditions (e.g., drought or cold temperatures) all determine the best choice of product and application rate. Always refer to the label prior to applying herbicides to knapweed infestations. **Many herbicides have strict grazing restrictions.** If land usage of treated areas includes grazing practices, it is important to refer to the label prior to applications.

Depending on the species and the location, herbicides and biological control agents can be combined or used separately against knapweed. For example, spotted, meadow, black, and brown knapweed can grow in scattered forest understory where most herbicides generally cannot be used. Though most knapweed biological control insects do well in open, sunny locations, *Agapeta zoegana*, *Chaetorellia acrolophi*, *Larinus* spp., and *Urophora* spp. also attack knapweeds in forested locations. In some instances, herbicide application may weaken knapweed plants, making them more vulnerable to attack by root-feeding biological control agents. *Sphenoptera jugoslavica* attack rates were found to increase following herbicide applications on diffuse knapweed.

The actions of herbicides and the seed feeding insects, *Bangasternus fausti*, *Chaetorellia acrolophi*, *Larinus* spp. *Metzneria paucipunctella*, *Terellia virens*, and *Urophora* spp., are less complementary. Successful herbicide applications result in no host plants for feeding and oviposition, thus decreasing biological control agent populations and their ability to control recovering knapweed individuals.

If it is the goal of the land management program to protect any established biological control agents, herbicide applications should take place at a time least disruptive to the agents, even if the timing is not ideal for the chemicals. In order to ensure that the insects maintain viable populations as the knapweed infestation is reduced, 25% of the area should remain untreated to serve as “refuges” for biological control agents.

General long-term land management practices

Knapweed has persistent growth characteristics, and seed can remain viable in the soil for several years. Therefore, you should implement long-term weed management programs. Long-term weed management includes re-treatment with herbicides or continued cultural, mechanical, or biological control practices to maintain low populations of this weed. Range improvements, such as grazing systems, cross-fencing, and water development, will help retard the invasion of many weed species, including knapweed. Sites with no desirable species should be reseeded with a competitive plant species as part of the total management program (Table 14, page 106).

Use Pesticides Safely!

- Read the pesticide label, even if you have used the pesticide before. Follow all the instructions on the label.
- Wear protective clothing and safety devices as recommended on the label.
- Bathe or shower after each pesticide application.
- Be cautious when you apply pesticides. Know your legal responsibility as a pesticide applicator. You may be liable for injury or damage resulting from pesticide use.

Peter M. Rice

Table 14 Comparison of knapweed management options.

| MANAGEMENT TECHNIQUE | ADVANTAGES | DISADVANTAGES | COMMENTS |
|---------------------------------------|---|--|---|
| Herbicides | Fast acting | Expensive for large areas. | Best used on small patches when knapweed foliage first emerges, or on the edges of a large infestation to keep it from spreading while other methods, such as biological control, have time to establish. |
| | High success rate for reducing knapweed densities. | May harm desirable vegetation, especially broadleaf species. | |
| | Rapidly enhances grass production. | Many natural areas are inaccessible to spray equipment. | |
| | | Public resistance to chemical controls. Regulations or policies may prohibit use in some areas. | |
| Biological Control | Can be very selective. | Some risk of undesirable effects on native plants. | Most economical option for large infestations and will control knapweed in a variety of environments in which the weed occurs, especially if multiple agents are introduced. |
| | Agents generally do not have to be reintroduced once established. | Not successful in all situations. | |
| | Public acceptance is generally higher than with other weed control methods. | Permanent: cannot be undone. | |
| | | Measurable changes in weed densities may take many years. | |
| Grazing | Allows use of the land even with heavy knapweed infestations. | Cannot be used in many natural areas such as national parks and wilderness areas. | Will remove top-growth only, and does not reduce the root mass. The same areas must be grazed annually or knapweed will rapidly reestablish; however this is often detrimental to desirable species. |
| | Can be used in combination with biological or chemical control methods. | Improper management can result in livestock poisoning. | |
| | | Non-selective | |
| | | Expensive Can exacerbate the problem. | |
| Mechanical/Cultural Treatments | Very effective | Not appropriate for natural areas and wildlands. | Not always compatible with biological control agents so is best used when an areas is being "reclaimed." |
| | Can be used to reseed native species. | Can be expensive for larger infestations. | |

GLOSSARY

| | |
|--------------------------|---|
| abdomen | The last of the three insect body regions; usually containing the digestive and reproductive organs |
| achene | A small, one-seeded fruit that does not split at maturity |
| allelopathy | One plant harms another by exuding specific molecules |
| alternate | Where leaves appear singly at stem nodes, on alternate sides of the stem |
| annual | A plant that flowers and dies within a period of one year from germination |
| antenna (pl. antennae) | In arthropods, one of a pair of appendages on the head, normally many jointed and of sensory function |
| aspirator | An apparatus used to suck insects into a container. Can be as simple as in a mouth aspirator, or mechanical as in a gasoline- or battery-powered vacuum aspirator |
| basal | Located at the base of a plant or plant part |
| biennial | A plant that flowers and dies between its first and second years and does not flower in its first year |
| biological control | The reduction in the abundance of a pest through intentional use of its natural enemies (predators, parasitoids, and pathogens) |
| bolting | Plant stage at which the flower stalk begins to grow |
| bract | A small, leaf-like structure below a flower |
| capitulum (pl. capitula) | Seed head of a plant in the sunflower family |

| | |
|------------------------|--|
| complete metamorphosis | An insect life cycle with four distinct stages (egg, larva, pupa, adult) |
| compound eyes | Paired eyes consisting of many facets, or ommatidia, in most adult Arthropoda |
| coordinates | A set of numbers used to specify a location |
| crown | Location of where a plant's stems meets its roots |
| density | Number of individuals per unit area |
| dissemination | Dispersal. Can be applied to seeds or insects |
| elytron (pl. elytra) | Hardened front wing of a beetle |
| emergence | Act of adult insect leaving the pupal exoskeleton, or leaving winter or summer dormancy |
| exoskeleton | Hard, external skeleton of the body of an insect |
| exotic | Not native |
| floret | One of the small, closely clustered flowers forming the head of a composite flower in the sunflower family |
| flower head | A special type of inflorescence consisting of numerous florets that actually look like one flower |
| forb | Herbaceous plant (does not have solid woody stems) |
| gall | An abnormal growth on a plant, usually induced by an insect that lives within the gall |
| genera | A taxonomic category ranking below family and above species and consisting of a group of species exhibiting similar characteristics. In taxonomic nomenclature the genus name is used, either alone or followed by a Latin adjective or epithet, to form the name of a species |
| grub | A soft, thick-bodied, C-shaped beetle larva |
| head | Insect segment with the mouthparts, antennae, and eyes |
| head capsule | Hardened covering of the head of an immature insect |
| herbivory | Feeding on plants |

| | |
|----------------------------------|---|
| hermaphroditic | Having both male and female reproductive parts |
| host | The plant or animal on which an organism feeds; the organism utilized by a parasitoid; a plant or animal susceptible to attack by a pathogen |
| host specificity | The highly-evolved, often obligatory association between an insect and its host (i.e., weed). A highly host-specific insect feeds only on its host and on no other species |
| inflorescence | The flowering part of a plant |
| instar | The phase of an insect's development between molts |
| integrated weed management (IWM) | A system for the planning and implementation of a program, using an interdisciplinary approach, to select a method for containing or controlling an undesirable plant species or group of species using all available methods |
| involucre | A circle of bracts under an inflorescence |
| larva (pl. larvae) | Immature insect stage between the egg and pupa (examples include grubs, caterpillars and maggots) |
| lobed | A leaf with shallow or deep, rounded segments, as in a thistle rosette leaf |
| membranous | Thin and transparent |
| metabolic sink | Site of a plant that receives photosynthate (food) produced by the plant, diverting the resource away from the plant's normal use |
| molting | Process of insect development that involves shedding its exoskeleton and producing another for the next instar |
| node | Part of the stem of a plant from which a leaf, branch, or aerial root grows |
| nontarget effect | When control efforts affect a species other than the species they were enacted to control |
| oviposit | To lay or deposit eggs |
| pappus | A tuft of hairs, scales, or bristles at the base of an achene in flowers of the sunflower family |

| | |
|------------------------------|--|
| perennial | A plant that lives more than two years |
| petiole | Leaf stalk that attaches it to a plant stem |
| proleg | A fleshy, unsegmented, abdominal walking appendage of some insect larvae, common among caterpillars |
| pupa (pl. pupae) (v. pupate) | Non-feeding, inactive insect stage between larvae and adult |
| qualitative | Measurement of descriptive elements (e.g., age class, distribution) |
| quantitative | Measurement of quantity; the number or amount (e.g., seeds per capitula) |
| receptacle | Part of the stem to which the flower is attached |
| rosette | A compact, circular, and normally basal cluster of leaves |
| seed head | Synonym for capitulum of a plant in the sunflower family. The seed head consists of a receptacle and florets |
| senescence | Final stage in a plant's life cycle |
| snout | 'Nose' of a weevil. The elongate head of a weevil with mouth parts at the tip (apex) |
| species | A fundamental category of taxonomic classification, ranking below a genus or subgenus and consisting of related organisms capable of interbreeding |
| synchrony | Occurring at the same time (e.g., plant flowering and insect oviposition) |
| taxonomy | The classification of organisms in an ordered system that indicates natural relationships. The science, laws, or principles of classification; systematics |
| thorax | Body region of an insect behind the head and abdomen, bearing the legs and wings |
| transect | A straight line of varying length along which plants are periodically sampled individually or in quadrants |

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APPENDICES

- Appendix I:** Troubleshooting guide: When things go wrong
- Appendix II:** PPQ Form 526 Interstate Transport Permit
- Appendix III:** Sample biological control agent release form
- Appendix IV:** Knapweed Standardized Impact Monitoring Protocol (SIMP) instructions and monitoring form
- Appendix V:** General biological control agent monitoring Form
- Appendix VI:** Knapweed qualitative monitoring form
- Appendix VII:** Knapweed biological control associated vegetation monitoring form

Appendix I: Troubleshooting guide; when things go wrong

This guide is intended to assist those who encounter problems when establishing a biological control program. It identifies the probable cause of typical problems and offers solutions.

| PROBLEM | PROBABLE CAUSE | SOLUTION |
|---|--|--|
| Biological control agents unhealthy when received | Physical damage to agents in transport | Prevent containers from colliding; use crush-proof containers. |
| | Drowning | Do not put water in containers during transport; prevent accumulation of excess moisture; too much plant material causes condensation. |
| | Excess or prolonged heat or cold | Keep containers cool at all times; use coolers and ice packs; avoid exposure to direct sunlight while in transit. |
| | Starvation | Put knapweed foliage (no flowers, seeds, or roots) in containers. |
| | Redistribution time | Transport or ship agents immediately after collection. |
| | | Release agents at new site immediately upon arrival or receipt of agent. |
| Parasitism and/or disease | Check source agents. Ensure the insect population is disease-free when collecting or receiving shipment. | |
| Number of eggs low | Agents past reproductive stage | Collect at peak activity (i.e., insects are mating). |
| | Sex ratio: not enough males or females | Observe mating among biological control agents before collecting; males often emerge earlier than females. |
| | Synchrony | Agents not synchronized with the knapweed growth stage; biological control agents require the weed to be at specific growth stage for optimal oviposition. |
| Few biological control agents collected | Wrong method used | Refer to Table 8 for recommended collection time and technique. |
| | Collection done at wrong time | Refer to Table 8 for recommended collection time and technique. |
| | Collection technique | Biological control agents can be killed/damaged during sweeping or aspirating so sweep lightly. |
| | | Use vacuum aspirator if aspirating by mouth is not working. Practice sweeping to avoid debris. |
| Conditions at time of collection wrong | Refer to Chapter 4 "Collecting Knapweed Biological control Agents" for guidelines on desirable weather conditions. | |
| Agents not found after release | Site is unsuitable | Refer to Chapter 4 "Selecting Biological control Agent Release Sites." |
| | Site too small | Select a larger site with a dense, uniform stand of knapweed. |
| | Pesticide used in area | Select pesticide-free site. |
| | Released on wrong knapweed species | Ensure the correct species of knapweed and biological control agent are used. |
| Cannot locate release site | Permanent location marker not obvious | Use bright-colored wooden, metal, or plastic stake. |
| | Map poorly or incorrectly drawn | Check map; redraw with more detail or add landmarks; GPS. |

Appendix II: PPQ Form 526, interstate transport permit application

Please see http://www.aphis.usda.gov/permits/ppq_epermits.shtml to electronically apply for permits.

According to the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information is 0579-0054. The time required to complete this information collection is estimated to average 0.50 hours per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information. FORM APPROVED
OMB NO. 0579-0054

No permit can be issued to move live plant pests or noxious weeds until an application is received (7 CFR 330 (live plant pests) or 7 CFR 360 (noxious weeds)).

| | | | | | | | |
|--|---|---|---|--|---|------------------------------|--|
| U.S. DEPARTMENT OF AGRICULTURE ANIMAL AND PLANT HEALTH INSPECTION SERVICE PLANT PROTECTION AND QUARANTINE PERMITS AND RISK ASSESSMENT, UNIT 133 RIVERDALE, MARYLAND 20737 APPLICATION FOR PERMIT TO MOVE LIVE PLANT PESTS OR NOXIOUS WEEDS | | SECTION A - TO BE COMPLETED BY THE APPLICANT | | | | | |
| 3. TYPE OF PEST TO BE MOVED * <input type="checkbox"/> Pathogens <input type="checkbox"/> Arthropods <input type="checkbox"/> Noxious Weeds <input type="checkbox"/> Other (Specify) _____ <small>This permit does not authorize the introduction, importation, interstate movement, or release into the environment of any genetically engineered organisms or products.</small> | | 1. NAME, TITLE, AND ADDRESS (include Zip Code) 2. TELEPHONE NO. () | | | | | |
| A. SCIENTIFIC NAMES OF PESTS TO BE MOVED | B. CLASSIFICATION (Orders, Families, Races, or Strains) | C. LIFE STATES, IF APPLICABLE | D. NO. OF SPECIMENS OR UNITS | E. SHIPPED FROM (Country or State) | F. ARE PESTS ESTABLISHED IN U.S.? | G. MAJOR HOST(S) OF THE PEST | |
| 4. | | | | | | | |
| 5. | | | | | | | |
| 6. | | | | | | | |
| 7. WHAT HOST MATERIAL OR SUBSTITUTES WILL ACCOMPANY WHICH PESTS (indicate by the number) | | | | | | | |
| 8. DESTINATION | | 9. PORT OF ARRIVAL | | 10. APPROXIMATE DATE OF ARRIVAL OR INTERSTATE MOVEMENT | | | |
| 11. NO. OF SHIPMENTS | 12. SUPPLIER | | 13. METHOD OF SHIPMENT <input type="checkbox"/> Air Mail <input type="checkbox"/> Air Freight <input type="checkbox"/> Baggage <input type="checkbox"/> Auto | | | | |
| 14. INTENDED USE (Be specific, attach outline of intended research) | | | | | | | |
| 15. METHODS TO BE USED TO PREVENT PLANT PEST ESCAPE | | | | 16. METHOD OF FINAL DISPOSITION | | | |
| 17. <small>Applicant must be a resident of the U.S.A. I/We agree to comply with the safeguards printed on the reverse of this form, and understand that a permit may be subject to other conditions specified in Section B and C.</small> | | | | | SIGNATURE OF APPLICANT (Must be person named in Item 1) | | |
| | | | | | 18. DATE | | |
| WARNING: Any alteration, forgery, or unauthorized use of this document is subject to civil penalties of up to \$250,000 (7 U.S.C. s7734(b)) or punishable by a fine of not more than \$10,000, or imprisonment of not more than 6 years, or both (18 U.S.C. s1001). | | | | | | | |
| SECTION B - TO BE COMPLETED BY STATE OFFICIAL | | | | | | | |
| 19. RECOMMENDATION <input type="checkbox"/> Concur (Approve) <input type="checkbox"/> (Accept USDA Decision) | | 20. CONDITIONS RECOMMENDED | | | | | |
| 21. SIGNATURE | | 22. TITLE | | 23. STATE | 24. DATE | | |
| SECTION C - TO BE COMPLETED BY FEDERAL OFFICIAL | | | | | | | |
| | | | | | | 25. PERMIT NO. | |

PERMIT

(Permit not valid unless signed by an authorized official of the Animal and Plant Health Inspection Service)

Under authority of the Plant Protection Act of 2000, permission is hereby granted to the applicant named above to move the pests described, except as deleted, subject to the conditions stated on, or attached to this application. (See standard conditions on reverse side.)

* For exotic plant pathogens, attach a completed PPQ Form 526-1.

| | | | | |
|---|----------|-------------------|-----------------|-------------------|
| 24. SIGNATURE OF PLANT PROTECTION AND QUARANTINE OFFICIAL | 25. DATE | 26. LABELS ISSUED | 27. VALID UNTIL | 28. PEST CATEGORY |
|---|----------|-------------------|-----------------|-------------------|

Appendix III: Sample biological control agent release form

Exotic weed biological control: *General Release Site Information*
USDA-APHIS-PPQ

| | | | | | |
|---------------|--|----------------|--|-------------------|--|
| | | | | Site code: | |
| State: | | County: | | Site name: | |
| Lat.: | | Long.: | | Elev.: | |

Landowner and/or contact person

| | | | |
|----------------|--|-------------------------|------------|
| Name | | Title (if appl.) | |
| Address | | | |
| City | | State | ZIP |
| Phone | | e-mail | |

Extent of weed infestation: ≤5 ac (2 ha) 5 - 100 ac (2 - 40 ha) 100 - 1000 ac (40 - 400 ha)
 > 1000 ac (400 ha)

Weed distribution: Largely or totally continuous Interrupted ('patches' separated by uninfested areas)

General site topography: Level Slight slope Moderate slope Steep slope Hilly

Aspect: North South East West Northwest Northeast Southwest Southeast

Soil type: Gravel/cobble Sand Sandy loam Loam Silt loam Clay loam Clay

Probability of flooding: Very low (rarely occurs) Low-moderate (occasional years) High (e.g. yearly)

Treatments at site in last year: Herbicide(s) [if so, chemical: _____] Grazing Burning
 Cutting Bulldozing/plowing Other: _____

Native (pre-infestation) plant communities at site, if known:

| | |
|---|--|
| Biocontrol agent(s) released: | |
| Date and time agent(s) released: | |
| Number of agent(s) released: | |
| Weather conditions at time of release: | |
| Released by (name and affiliation): | |

Appendix IV: Knapweed Standardized Impact Monitoring Protocol (SIMP) instructions and monitoring forms

Idaho's statewide monitoring guidelines for *Cyphocleonus achates* and spotted knapweed



Overview

A critical part of successful weed biological control programs is a monitoring process to measure populations of biological control agents and the impact that they are having on the target weed. Monitoring should be conducted on an annual basis for a number of years. The Idaho State Department of Agriculture, in conjunction with the University of Idaho, Nez Perce Biocontrol Center, and federal land management agencies, has developed the monitoring protocol below to enable land managers to take a more active role in monitoring the progress and weed control ability of the knapweed root weevil, *Cyphocleonus achates* (CYAC) in efforts to control spotted knapweed, *Centaurea maculosa*. This monitoring protocol was designed to be implemented by

land managers in a timely manner while providing data which will enable researchers to better quantify the impact of CYAC on spotted knapweed throughout the state.

Spotted Knapweed

Spotted knapweed is an herbaceous, short-lived, perennial reproducing entirely by seed and producing up to 25,000 seeds per plant can remain viable in the soil up to 8 years. Flowers range in color from pink to light-purple and bloom from July to October. The flower head bracts are black-tipped, giving the plant its characteristic "spotted" appearance. Seeds are brown to black in color, smooth, and less than 0.25 inch long. Stems are typically 2 to 4 feet tall with lower leaves that are deeply lobed and upper leaves that are more linear. Spotted knapweed prefers moist rangeland habitats, but is common in waste areas, along roadsides, and in pastures. To date, thirteen biological control agents have been approved for release for the knapweed complex, which includes spotted knapweed.

Knapweed Root Weevil (CYAC)

CYAC is a robust biological control agent that can attack spotted (preferred host) and diffuse knapweeds. CYAC overwinter as larvae in roots. CYAC larvae mine and gall the vascular tissue of knapweed roots. CYAC larval feeding reduces knapweed density and can result in death of small plants. CYAC adults emerge from June to mid-September and feed on knapweed leaves. The adults are 0.5 to 0.6 inches long and generally live 8 to 15 weeks. Females mate several times and deposit more than 100 eggs during their lifetime. CYAC eggs are laid singly in a notch excavated by the female on the root crown, just below the soil surface.



Appendix IV (cont.): Knapweed Standardized Impact Monitoring Protocol (SIMP) instructions and monitoring forms

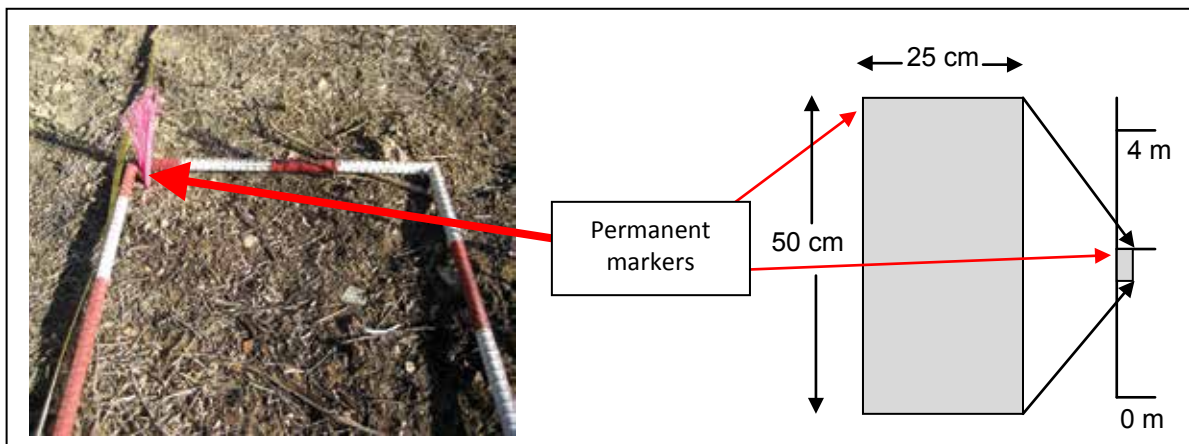
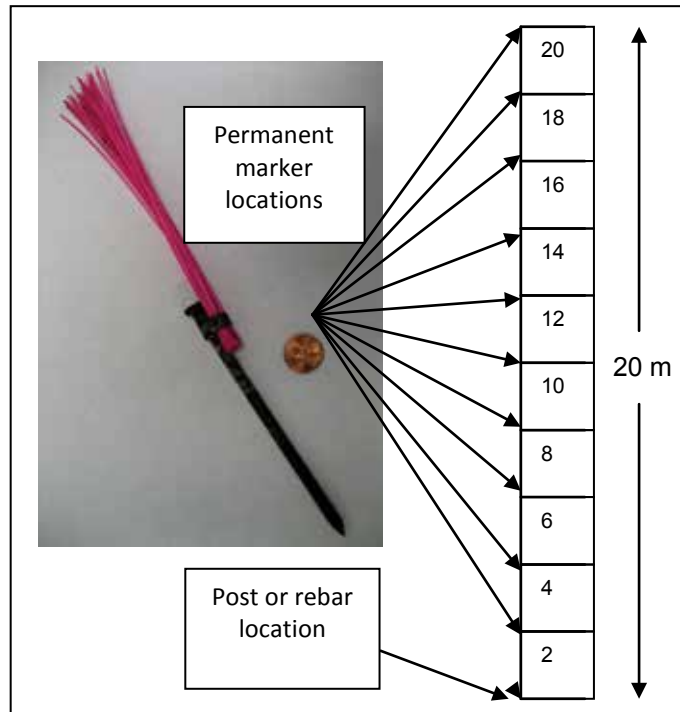
Eggs hatch in 10 to 12 days and larvae begin feeding on roots.

Monitoring

The Statewide Biological Control monitoring protocol is based upon a permanent 20 meter vegetation sampling transect randomly placed in a suitable (at least 1 acre) infestation of spotted knapweed and timed counts of CYAC adults. Annual vegetation sampling will allow researchers to characterize the plant community and the abundance and vigor of leafy spurge. Timed counts of CYAC adults will provide researchers with an estimate of CYAC population levels.

Permanent Site Set-up

To set up the vegetation monitoring transect, you will need: 1) a 25 x 50 cm Daubenmire frame made from PVC (preferred) or rebar, 2) a 20-m tape measure for the transect and plant height, 3) 10 permanent markers (road whiskers and 16 penny nails – see picture below), 4) a post (stake or piece of rebar) to monument the site (see pictures for examples of field equipment), and 5) 30-45 minutes at the site **during the second week of August**. To set up the transect, place the 20-m tape randomly within the infestation. Mark the beginning of the transect with a post. Place permanent markers every 2 m (for a total of 10 markers) beginning at the 2-m mark and ending with the 20-m mark on the tape measure. Place the Daubenmire frame parallel to the tape on the 50-cm side with the permanent marker in the upper left corner starting at 2 m (see pictures). **Refer to the “timed” data sheet** for how to conduct monitoring. Repeat the frame placement at 2-m intervals for a total of 10 measurements (one at each permanent marker).



Appendix IV (cont.): Knapweed Standardized Impact Monitoring Protocol (SIMP) instructions and monitoring forms

Idaho's statewide monitoring guidelines for *Larinus* spp. and spotted knapweed:



Overview

A critical part of successful weed biological control programs is a monitoring process to measure populations of biological control agents and the impact that they are having on the target weed. Monitoring should be conducted on an annual basis for a number of years. The Idaho State Department of Agriculture, in conjunction with the University of Idaho, Nez Perce Biocontrol Center, and federal land management agencies, has developed the monitoring protocol below to enable land managers to take a more active role in monitoring the progress and weed control ability of the knapweed flower weevils, *Larinus* spp. (LA) in efforts to control spotted knapweed, *Centaurea maculosa*. This monitoring protocol was designed to be implemented by land managers in a timely manner while providing data which will enable

researchers to better quantify the impact of LA on spotted knapweed throughout the state.

Spotted Knapweed

Spotted knapweed is an herbaceous, short-lived, perennial reproducing entirely by seed, producing up to 25,000 seeds per plant can remain viable in the soil up to 8 years. Flowers range in color from pink to light-purple and bloom from July to October. The bracts of the flower heads are black-tipped, giving the plant its characteristic “spotted” appearance. Seeds are brown to black in color, smooth, and less than 0.25 inch long. Stems are typically 2 to 4 feet tall with lower leaves that are deeply lobed and upper leaves that are more linear. Spotted knapweed prefers moist rangeland habitats, but is common in waste areas, along roadsides, and in pastures. To date, thirteen biological control agents have been approved for release for the knapweed complex, which includes spotted knapweed.

Knapweed Flower Weevil (LA)

LA is an abundant biological control agent that can utilize spotted, diffuse, and squarrose knapweeds. LA overwinters as an adult. Adults emerge in the spring when they begin to feed on knapweed foliage. Females produce between 28 and 130 eggs which they lay in clusters in open flowers. Eggs hatch and larvae feed on seeds and receptacle tissue for about a month. Larvae construct cocoons within the seed heads using pappus hairs and pupate. Emerging adults chew a characteristic round hole in the top of the cocoon that is visible when viewed from above (see picture).



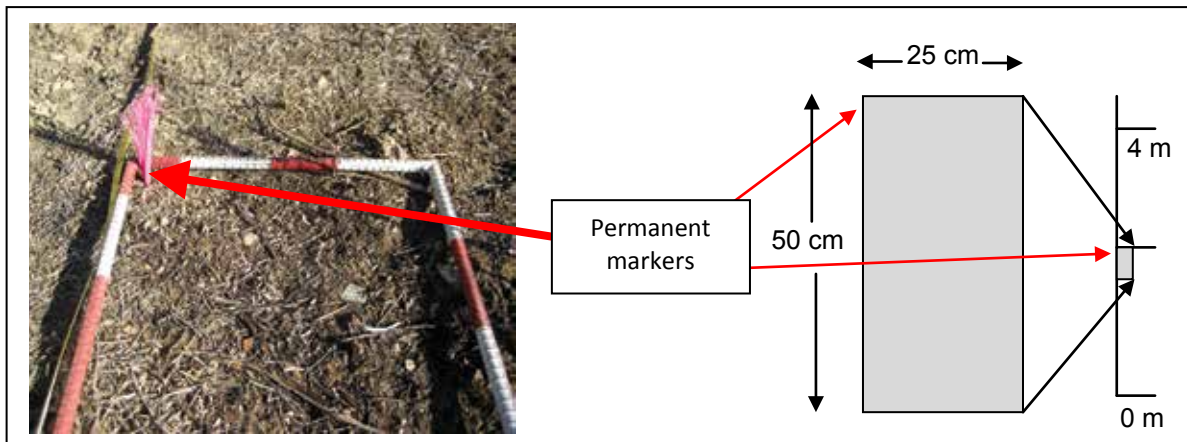
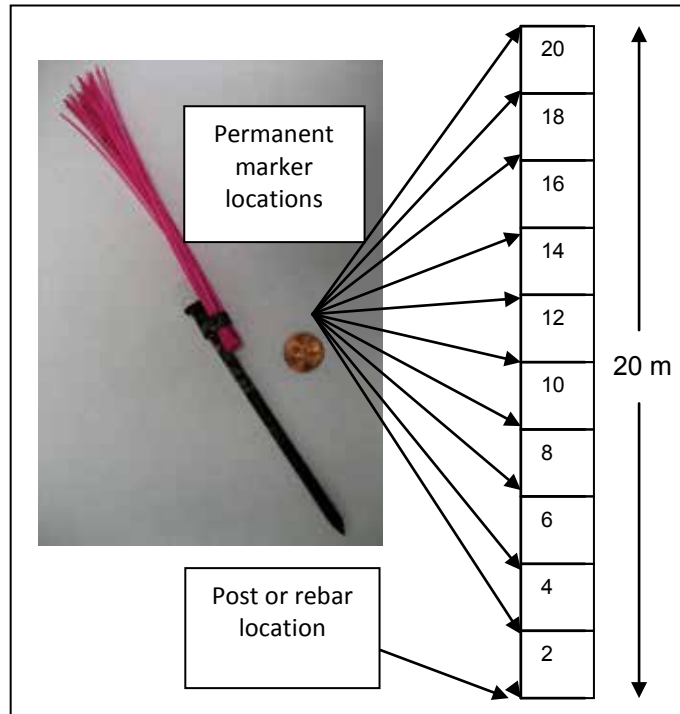
Appendix IV (cont.): Knapweed Standardized Impact Monitoring Protocol (SIMP) instructions and monitoring forms

Monitoring

The Statewide Biological Control monitoring protocol is based upon a permanent 20 meter vegetation sampling transect randomly placed in a suitable (at least 1 acre) infestation of spotted knapweed and sweep net samples of LA. Annual vegetation sampling will allow researchers to characterize the plant community and the abundance and vigor of spotted knapweed. Sweep net samples of LA adults will provide researchers with an estimate of LA population levels.

Permanent Site Set-up

To set up the vegetation monitoring transect, you will need: 1) a 25 x 50 cm Daubenmire frame made from PVC (preferred) or rebar, 2) a 20-m tape measure for the transect and plant height, 3) 10 permanent markers (road whiskers and 16 penny nails – see picture below), 4) a post (stake or piece of rebar) to monument the site (see pictures for examples of field equipment), and 5) 30-45 minutes at the site **during the 3rd week of June**. To set up the transect, place the 20-m tape randomly within the infestation. Mark the beginning of the transect with a post. Place permanent markers every 2 m (for a total of 10 markers) beginning at the 2-m mark and ending with the 20-m mark on the tape measure. Place the Daubenmire frame parallel to the tape on the 50-cm side with the permanent marker in the upper left corner starting at 2 m (see pictures). **Refer to the “sweep” data sheet** for how to conduct monitoring. Repeat the frame placement at 2-m intervals for a total of 10 measurements (one at each permanent marker).



Appendix IV (cont.): Knapweed Standardized Impact Monitoring Protocol (SIMP) instructions and monitoring forms

Idaho's statewide monitoring guidelines for *Larinus* spp. and diffuse knapweed:



Overview

A critical part of successful weed biological control programs is a monitoring process to measure populations of biological control agents and the impact that they are having on the target weed. Monitoring should be conducted on an annual basis for a number of years. The Idaho State Department of Agriculture, in conjunction with the University of Idaho, Nez Perce Biocontrol Center, and federal land management agencies has developed the monitoring protocol below enables land managers to take a more active role in monitoring the progress and weed control ability of the knapweed flower weevils, *Larinus* spp. (LA) in efforts to control diffuse knapweed, *Centaurea diffusa*. This monitoring protocol was designed to be implemented by land managers in a timely manner while providing data which will enable researchers to

better quantify the impact of LA on diffuse knapweed throughout the state.

Diffuse Knapweed

Diffuse knapweed is a biennial or short-lived, perennial reproducing entirely by seed and producing up to 18,000 seeds per plant. Flowers occur singly or in clusters, bloom from July to September and may be white, pink or lavender. The bracts of the flower heads are tipped with a long, slender spine fringed with smaller spines. Seeds are blackish-brown with vertical brown and gray stripes and about 0.1 inch long. Stems are typically 6 inches to 2 feet tall with deeply divided leaves. Diffuse knapweed prefers well-drained, light textured soils and is intolerant of shade. This plant is common along roadsides, at disturbed sites, and in abandoned areas. To date, thirteen biological control agents have been approved for release for the knapweed complex, which includes diffuse knapweed.

Knapweed Flower Weevil (LA)

LA is an abundant biological control agent that can utilize spotted, diffuse, and squarrose knapweeds. LA overwinters as an adult. Adults emerge in the spring when they begin to feed on knapweed foliage. Females produce between 28 and 130 eggs which they lay in clusters in open flowers. Eggs hatch into larvae which feed on seeds and receptacle tissue for about a month. Larvae construct cocoons within the seed heads using pappus hairs and pupate. Emerging adults chew a characteristic round hole in the top of the cocoon that is visible when viewed from above (see below).



Appendix IV (cont.): Knapweed Standardized Impact Monitoring Protocol (SIMP) instructions and monitoring Forms

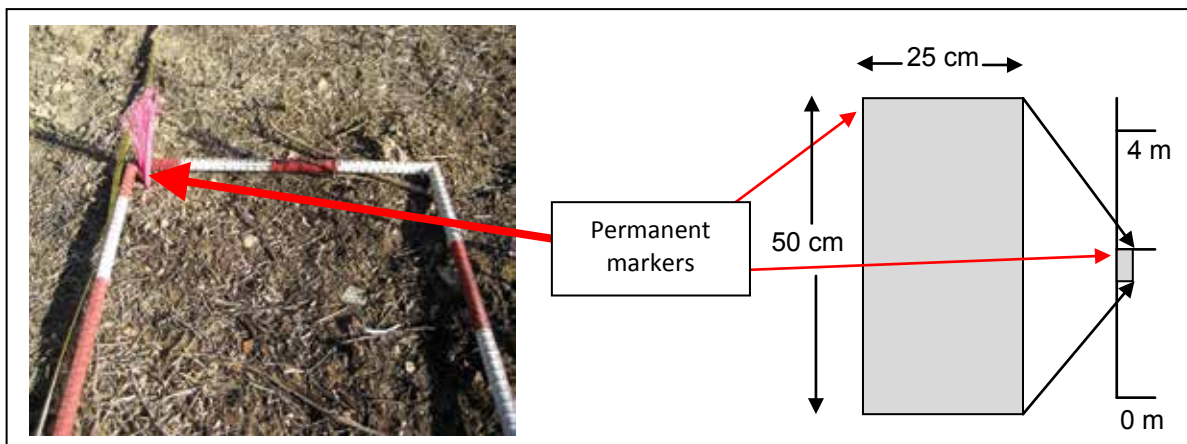
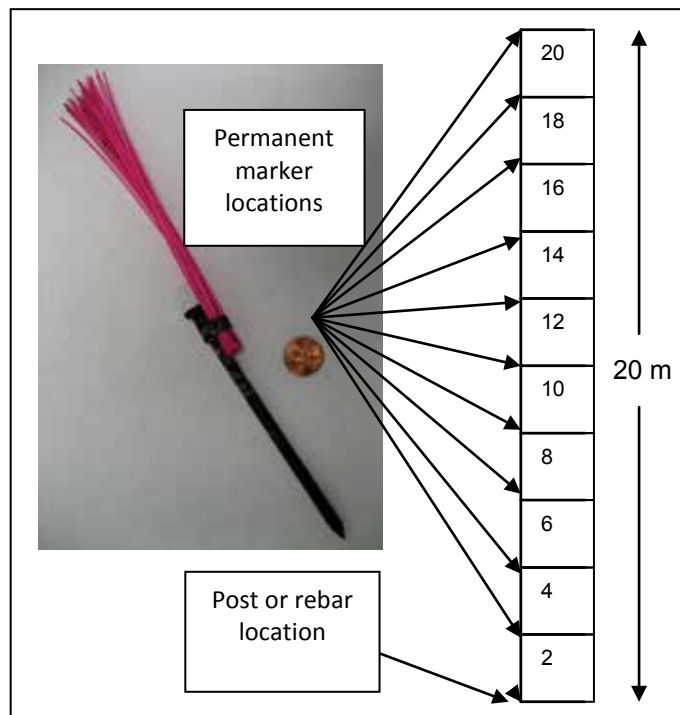
Monitoring

The Statewide Biological Control monitoring protocol is based upon a permanent 20 meter vegetation sampling transect randomly placed in a suitable (at least 1 acre) infestation of diffuse knapweed and sweep net samples of LA. Annual vegetation sampling will allow researchers to characterize the plant community and the abundance and vigor of diffuse knapweed. Sweep net samples of LA adults will provide researchers with an estimate of LA population levels.

Permanent Site Set-up

To set up the vegetation monitoring transect, you will need: 1) a 25 x 50 cm Daubenmire frame made from PVC (preferred) or rebar, 2) a 20-m tape measure for the transect and plant height, 3) 10 permanent markers (road whiskers and 16 penny nails—see picture below), 4)

a post (stake or piece of rebar) to monument the site (see pictures for examples of field equipment), and 5) 30–45 minutes at the site **during the 2nd week of June**. To set up the transect, place the 20-m tape randomly within the infestation. Mark the beginning of the transect with a post. Place permanent markers every 2 m (for a total of 10 markers) beginning at the 2-m mark and ending with the 20-m mark on the tape measure. Place the Daubenmire frame parallel to the tape on the 50-cm side with the permanent marker in the upper left corner starting at 2m (see pictures). Refer to the “**sweep**” data sheet for how to conduct monitoring. Repeat the frame placement at 2-m intervals for a total of 10 measurements (one at each permanent marker).



Appendix IV (cont.): Knapweed Standardized Impact Monitoring Protocol (SIMP) timed data sheet (for use with *Cyphocleonus achates*)

Monitoring biological control agents is an essential component of a successful biocontrol program that can be used to accurately document impact and safety of this weed management practice. This monitoring form has been endorsed by the Nez Perce Biocontrol Center, University of Idaho, Forest Health Protection, Bureau of Land Management, and Idaho State Department of Agriculture. The monitoring information from this form will be used to document vegetation cover, target weed density, and biological control agent abundance and the changes that occur over time.

General Information:

| | | | |
|---------------------|------------|--------------------------------------|------------|
| Observer(s): | | Date: | Landowner: |
| Permanent site? Y N | Site name: | | Weed: |
| Biocontrol agent: | | Insect Stage: Adult Larvae Pupae Egg | |
| Lat/Long: N ° ' " | W ° ' " | UTM Datum: | UTM E: |
| | | UTM Year : | UTM N: |

Weed Infestation:

| | | | |
|----------------|----------------|--------|--------------------------|
| Size in acres: | Picture taken? | Yes No | If Y, picture direction: |
|----------------|----------------|--------|--------------------------|

Vegetation cover (all in %, rows add to 100%):

| Frame | Target weed% | Other weed% | Forb/shrub% | Grass% | Bare ground% | Litter% | Moss% | Total% |
|-------|--------------|-------------|-------------|--------|--------------|---------|-------|--------|
| 1 | | | | | | | | |
| 2 | | | | | | | | |
| 3 | | | | | | | | |
| 4 | | | | | | | | |
| 5 | | | | | | | | |
| 6 | | | | | | | | |
| 7 | | | | | | | | |
| 8 | | | | | | | | |
| 9 | | | | | | | | |
| 10 | | | | | | | | |

Target weed size/density:

| Frame | Number of stems | Height of tallest stem (cm) |
|-------|-----------------|-----------------------------|
| 1 | | |
| 2 | | |
| 3 | | |
| 4 | | |
| 5 | | |
| 6 | | |
| 7 | | |
| 8 | | |
| 9 | | |
| 10 | | |

Biological control agent:

| Count location | # insects per 3 min. count |
|----------------|----------------------------|
| 1 | |
| 2 | |
| 3 | |
| 4 | |
| 5 | |
| 6 | |

Appendix IV (cont.): Knapweed Standardized Impact Monitoring Protocol (SIMP) Sweep Data Sheet (for use with *Larinus* spp.)

Monitoring biological control agents is an essential component of a successful biocontrol program that can be used to accurately document impact and safety of this weed management practice. This monitoring form has been endorsed by the Nez Perce Biocontrol Center, University of Idaho, Forest Health Protection, Bureau of Land Management, and Idaho State Department of Agriculture. The monitoring information from this form will be used to document vegetation cover, target weed density, and biological control agent abundance and the changes that occur over time.

General Information:

| | | | |
|---------------------|------------|--------------------------------------|------------|
| Observer(s): | | Date: | Landowner: |
| Permanent site? Y N | Site name: | | Weed: |
| Biocontrol agent: | | Insect Stage: Adult Larvae Pupae Egg | |
| Lat/Long: N ° ' " | W ° ' " | UTM Datum: | UTM E: |
| | | UTM Year : | UTM N: |

Weed Infestation:

| | | | |
|----------------|----------------|--------|--------------------------|
| Size in acres: | Picture taken? | Yes No | If Y, picture direction: |
|----------------|----------------|--------|--------------------------|

Vegetation cover (all in %, rows add to 100%):

| Frame | Target weed% | Other weed% | Forb/shrub% | Grass% | Bare ground% | Litter% | Moss% | Total% |
|-------|--------------|-------------|-------------|--------|--------------|---------|-------|--------|
| 1 | | | | | | | | |
| 2 | | | | | | | | |
| 3 | | | | | | | | |
| 4 | | | | | | | | |
| 5 | | | | | | | | |
| 6 | | | | | | | | |
| 7 | | | | | | | | |
| 8 | | | | | | | | |
| 9 | | | | | | | | |
| 10 | | | | | | | | |

Target weed size/density:

| Frame | Number of stems | Height of tallest stem (cm) |
|-------|-----------------|-----------------------------|
| 1 | | |
| 2 | | |
| 3 | | |
| 4 | | |
| 5 | | |
| 6 | | |
| 7 | | |
| 8 | | |
| 9 | | |
| 10 | | |

Biological control agent:

| Count location | # insects per 10 sweeps |
|----------------|-------------------------|
| 1 | |
| 2 | |
| 3 | |
| 4 | |
| 5 | |
| 6 | |

Appendix IV (cont) :Knapweed Standardized Impact Monitoring Protocol (SIMP) instructions and monitoring form

General Information:

- Observer(s) – Who are you?
- Date – Today's date.
- Landowner – Who is the landowner/land manager?
- Permanent? – Is this a permanent monitoring site?
- Site name – Which site are you monitoring? This could have a specific name if it is a permanent site.
- Weed – Which target weed are you are monitoring?
- Biocontrol agent – Which biocontrol agent you are monitoring?
- Insect Stage – What is the growth stage of the agent are you monitoring?
- Lat/Long OR UTM – What are the coordinates of the site you are monitoring? If UTM (preferred), what datum and year are your coordinate system?



Annual grass – note stems which are typically solitary or in a few stemmed tufts.

Vegetation Cover (all in %, rows should add up to total 100%) – All percentages are to be estimated to the nearest 5%. Put a "T" on the form for trace amounts less than 5%.

- Frame – Which frame number are you working on (1= 2m, 2= 4m, ...,10 = 20m on transect)?
- Target weed % – What is % cover of the target weed to the nearest 5%?
- Other weeds % – What is the % cover of any other weeds in the frame to the nearest 5%? Count undesirable annual grasses as weeds.
- Forb/Shrub % – What is the % cover of native forbs/shrubs in the frame to the nearest 5%?
- Grass % – What is the % cover of grass to the nearest 5%?
- Bare Ground/Litter % – What is the % cover of bare ground/litter to the nearest 5%?



Perennial grass – note the multiple stem base with multiple year's growth.

Target Weed Size/Density

- Frame – Which frame number are you working on (1=2m,...,10=20m)?
- Number of stems – How many stems of the target weed are in the frame?
- Height of tallest stems (cm) – How tall is the tallest stem in the frame (in cm)?

Biological Control Agent Density Monitoring

Here, you collect data *for the target biocontrol agent* that helps to get an unbiased assessment of the population size of the biological control agents. This is probably the most important part of the data collection.

- Count/Sweep location – Do not count or sweep in the area where the transect is located. Instead, identify 6 similar locations around or close-by but at least 20 paces away from the transect.
- Counting: *In 3 minutes, count the number of target insects. How many insects can you find in the 3 minute period? Carefully approach the plants and be sure to count each insect only once. Repeat the count 5 times (for a total of 6 3-minute counts) in different areas.*
- # of insects per 10 sweeps – *How many insects are in your net after 10 sweeps of the surrounding vegetation? Take one step between each sweep. Repeat 5 more times (for a total of 6 sweep sites, 60 sweeps) moving at least 2 steps away from the last sweep location.*
- *These are replications and provide the unbiased data to calculate the population size of the biological control agents.*

Appendix V: General biological control agent monitoring form

SITE: _____ STATE: _____ COUNTY _____ DATE: _____
year month day

DATA COLLECTOR: _____ TIME: _____
First and last name

LAT/LONG: N _____° _____' W _____° _____' UTM DATUM: _____ UTM YEAR: _____
 UTM E: _____ UTM N: _____

ELEVATION: _____ TEMPERATURE: _____ WEATHER: _____

INSECT COUNTS:

| Species | Method | # insects (use Chart A) |
|--|---|-------------------------|
| <i>A. Zoegana</i> , <i>C. achates</i> , <i>S. jugoslavica</i> | Randomly select 25 plants, dissect root tissue and count larval tunnels | |
| <i>M. paucipunctella</i> , <i>B. fausti</i> <i>C. acrolophi</i> , <i>Larinus</i> spp., <i>T. virens</i> , <i>Urophora</i> spp. | Randomly select 25 plants, dissect galls and count larvae | |

| Chart A: Insect abundance | 1 | 1-10 |
|---------------------------|---|---------|
| | 2 | 11-25 |
| | 3 | 26-100 |
| | 4 | 100-500 |
| | 5 | >500 |

KNAPWEEDS:

| Chart B: Damage Class | 0 | <1% |
|-----------------------|---|-------|
| | 1 | 1-5% |
| | 2 | 6-25% |
| | 3 | > 25% |

| Chart C: Cover Class | 0 | <1% |
|----------------------|---|--------|
| | 1 | 1-5% |
| | 2 | 6-25% |
| | 3 | 26-50% |
| | 4 | 51-75% |
| | 5 | 76-95% |
| | 6 | >95% |

| Quad # | Knapweed | | Stems | | | |
|--------|------------------------|-----------------------|----------------|------------------|-----------------------------|--|
| | % damage (use Chart B) | % cover (use Chart C) | # mature stems | # immature stems | Height 4 tallest stems (cm) | |
| 1 | | | | | | |
| 2 | | | | | | |
| 3 | | | | | | |
| 4 | | | | | | |
| 5 | | | | | | |
| 6 | | | | | | |
| 7 | | | | | | |
| 8 | | | | | | |
| 9 | | | | | | |
| 10 | | | | | | |
| 11 | | | | | | |
| 12 | | | | | | |
| 13 | | | | | | |
| 14 | | | | | | |
| 15 | | | | | | |
| 16 | | | | | | |
| 17 | | | | | | |
| 18 | | | | | | |
| 19 | | | | | | |
| 20 | | | | | | |

Appendix V (cont.): General biological control agent monitoring form

| Quad # | Knapweed | | Stems | | | |
|--------|---------------------------|--------------------------|----------------|------------------|-----------------------------|--|
| | % damage (use Chart B) | % cover (use Chart C) | # mature stems | # immature stems | Height 4 tallest stems (cm) | |
| 21 | | | | | | |
| 22 | | | | | | |
| 23 | | | | | | |
| 24 | | | | | | |
| 25 | | | | | | |
| 26 | | | | | | |
| 27 | | | | | | |
| 28 | | | | | | |
| 29 | | | | | | |
| 30 | | | | | | |
| 31 | | | | | | |
| 32 | | | | | | |
| 33 | | | | | | |
| 34 | | | | | | |
| 35 | | | | | | |
| 36 | | | | | | |
| 37 | | | | | | |
| 38 | | | | | | |
| 39 | | | | | | |
| 40 | | | | | | |

Notes:

Appendix V: Instructions for the general biological control agent monitoring form

Materials needed: 1 meter (1.1 yard) stick, 0.2 x 0.5 m (0.2 x 0.55 yard) quadrat frame, stopwatch, sweep net, monitoring form, pencils, clipboard, camera, and GPS unit to relocate transects.

General: The purpose of this monitoring activity is to estimate the abundance of knapweed and its biological control agents at the site, and to record measurements of a sample of knapweed plants. Conduct the monitoring when the biological control agents are at their peak. Monitoring is easier with two people, one to make the observations and the other to record data.

1) Site information: Fill out the site information at the top of the form.

2) Insect counting: Use the chart for the method to count insects. Carefully approach the site and avoid disturbing the vegetation. Adult insects often drop from the vegetation once you touch stems (or even as you approach the quadrat). Use Chart A to record the category of abundance (1-5).

3) Locate the transect and position the quadrat: After you have completed the insect counts, locate the transect using the GPS coordinates and the permanent marker.

4) Position the quadrat: Position the quadrat along the transect, as close to the ground as possible, carefully positioning the quadrat along that transect line. Be sure not to damage the plants. The quadrat should be in the same location as the previous year's quadrat. Move stems in or out of the frame area so that all stems originating inside the quadrat are included.

5) Estimate feeding damage: Examine the knapweed for any damage to the leaves, shoots, flower heads, etc., such as malformed flower heads due to agents feeding on capitula and seeds. Standing over the frame, estimate the percent of damage over the entire quadrat, using Chart B to determine the category of damage.

6) Estimate percent cover: Standing over the frame, estimate how much of the quadrat is covered by knapweed. Use cover estimates in Chart C to estimate percent cover class.

7) Count stems: Count the number of knapweed stems, beginning at one corner of the quadrat and working systematically across the quadrat. Count the number of mature (floral) and immature (vegetative) stems.

8) Measure stems: Select the four (4) tallest knapweed stems in each quadrat (if there are fewer than 4 stems/quadrat, measure all that are present). Measure the stem height (to the closest cm)

9) Other observations: Record any general observations or useful information; disturbances, grazing, fire, etc., for the sample quadrat or the site in general.

Appendix VI: Knapweed qualitative monitoring form

Name: _____ Date: _____ Time: _____ am/pm

Location: _____ Site #: _____

Biocontrol species: _____ Year of release: _____

| Cover class estimate by plant category | | | | | | |
|--|----|------|-------|--------|--------|---------|
| | 0% | 1-5% | 6-20% | 21-45% | 46-70% | 71-100% |
| Knapweed | | | | | | |
| Annual Grasses | | | | | | |
| Perennial Grasses | | | | | | |
| Forbs | | | | | | |
| Shrubs | | | | | | |
| Trees | | | | | | |

| |
|--|
| Dominant plant species on site: |
| |
| |
| Other noxious weeds: |
| |
| |

| Estimate knapweed density class (✓ check one) | | | |
|---|--|-----------------------|--|
| Flowering plants/meter sq | | Knapweed distribution | |
| 0 | | Isolated | |
| 1-25 | | Scattered | |
| 26-50 | | Scattered-Patchy | |
| 50-75 | | Patchy | |
| >75 | | Continuous | |

| Knapweed phenology class at time of monitoring | |
|--|-------------------|
| Knapweed stage | Estimated percent |
| Seedling | |
| Rosette | |
| Bolting | |
| Flowering | |
| Senescent | |

Comments/Observations _____

Appendix VII (cont): Instructions for knapweed biological control associated vegetation monitoring form

Materials needed: 1 meter stick, 1.0 m² quadrat frame, data sheets, pencils, clipboard, camera, and GPS unit to relocate quadrats.

General: The purpose of this activity is to estimate the abundance of other vegetation in the community, and to record measurements of knapweed plant attributes. Monitoring is easier with two people, one to make the observations and the other to record data.

1) Site information: Fill out the site information at the top of the form.

2) Position the quadrat: Position the quadrat frame as close to the ground as possible, carefully positioning the quadrat along that transect line. Be sure not to damage the vegetation. The quadrat should be in the same location as the previous year's quadrat.

3) Estimate amount of vegetation: Standing over the frame, estimate how much of the quadrat is vegetated, and how much is not vegetated (bare ground, rock, etc). Use cover estimates in Chart A to estimate percent cover.

4) Estimate percent cover of vegetation: Standing over the frame, estimate how much of the quadrat is covered by knapweed, how much is covered by other forbs, grasses, or shrubs. Use cover estimates in Chart A to estimate percent cover. Because vegetation can naturally overlap, it is possible to have a combined total percent cover to exceed 100%.

5) Estimate percent cover of individual species: Standing over the frame, estimate how much of the quadrat is covered by individual species, other than knapweed. Use this section to track specific species, for example perennial grasses, native forbs, etc.

6) Other observations: Record any general observations or useful information, such as disturbances, grazing, fire, etc.