CHAPTER 6: Biological Control

Biological control is the use of natural enemies (biological control agents) to control a target weed. The objective is to establish self-sustaining populations of the biological control agents that will proliferate and attack the target weed throughout its range. Most noxious weeds in North America are exotic and without specialized natural enemies that occur in their area of origin. As a result, these plants have a competitive advantage over our native species, which have their own specialized herbivores and diseases.

Use of biological control to manage a noxious weed differs from other methods in that management measures are not directed at particular patches or infestations. Biological control agents are living organisms and land managers cannot accurately direct their activity. Instead, the goal of these programs is to release control agents at strategic locations throughout the infested area with the intention that the control agent will establish, build up high populations, and spread throughout the infestation. Eventually, all areas infested by the target weed will be colonized. The establishment, build-up, and spread of a control agent usually requires years, so this method is directed at long-term control of the weed. Biological control methods do not eradicate; rather they provide sustained suppression of the target weed populations. Insect agents can achieve this by defoliation, seed predation, boring into roots, shoots and stems, or extracting plant fluids. All these effects can reduce the competitive ability of the plant relative to the surrounding vegetation (Wilson and MacCaffery 1999).

Many years are necessary to research, test, and release biological controls for use on a target weed. As a result, biocontrol is usually developed for the most damaging and widespread weeds. In the development of weed biological control, scientists examine the target weed in its area of origin and identify the most promising natural enemies for use as potential agents. These natural enemies are subjected to a series of host-specificity tests to examine their safety for introduction into the United States. A high degree of host-specificity is critical for successful biological control of a weed, and natural enemies that attack agricultural crops or related native species are rejected. For yellow starthistle, research on biological control began in the mid 1960s and continues today.

Bud weevil. Bangasternus orientalis *is one of many biocontrol agents released in California to control yellow starthistle. (Photo: B. Villegas)*

Natural Enemies Associated with Yellow Starthistle Control

INSECTS

The United States Department of Agriculture (USDA), Agricultural Research Service Exotic and Invasive Weed Research Unit in Albany and the California Department of Food and Agriculture (CDFA) Biological Control Program are actively pursuing several biological control agents for use against yellow starthistle in California and the

Table 2. Distribution, impact and publications on yellow starthistle seed head insects

Species	Common Name	Distribution	Impact	References
Bangasternus orientalis	bud weevil	Wide	Low	Campobasso *et al*. 1998 Maddox *et al*. 1986, 1991 Maddox and Sobhian 1987 Pitcairn *et al*. 2004 Sobhian 1993a Sobhian *et al*. 1992
Chaetorellia australis	peacock fly	Limited	Low	Balciunas and Villegas 1999 Maddox *et al*. 1990 Turner *et al*. 1996 Pitcairn *et al*. 2004 Villegas *et al*. 1997, 2000b White *et al*. 1990
Chaetorellia succinea (accidental introduction)	false peacock fly	Wide	Moderate	Balciunas and Villegas 1999 Pitcairn *et al*. 1998a, 2003, 2004 Pitcairn 2002 Villegas 1998 Villegas *et al*. 1997, 1999, 2000b
Eustenopus villosus (=E. hirtus)	hairy weevil	Wide	Moderate	Clement *et al*. 1988 Connett and McCaffrey 1995 Fornasari *et al*. 1991 Fornasari and Sobhian 1993 Pitcairn *et al*. 2004 Villegas *et al*. 2000a
Larinus curtus	flower weevil	Limited	Low	Fornasari and Turner 1992 Pitcairn *et al*. 2004 Sobhian and Fornasari 1994 Villegas *et al*. 1999, 2000c
Urophora sirunaseva	gall fly	Wide	Low	Maddox *et al*. 1986 Pitcairn *et al*. 2004 Sobhian 1993b Turner 1994 Turner *et al*. 1994 White and Clement 1987 White *et al*. 1990

General articles on insect biological control of yellow starthistle

Topic	References
Discovery	Clement 1990, 1994 Clement and Sobhian 1991
Effects of natural insect populations on starthistle	Johnson *et al*. 1992 Pitcairn *et al*. 1999b
Reviews	Jette *et al*. 1999 McCaffrey and Wilson 1994 Pitcairn *et al*. 2000c, 2004 Rosenthal *et al*. 1991 Turner 1992 Turner and Fornasari 1992 Wood 1993

Peacock fly. *Although* Chaetorellia australis *has established populations on yellow starthistle, it has not established in densities sufficient to produce a significant reduction in starthistle. (Photo: B. Villegas)*

Hairy weevil. Eustenopus villosus *is the most damaging biological control insect established against yellow starthistle. It feeds by chewing a small hole in young flower buds and feeding on the soft internal tissues. This feeding damage kills young buds and stops their development. (Photo: B. Villegas)*

Release in the field. *Biocontrol agents are released only after years of host-specificity testing. (Photo: M. Pitcairn)*

western United States. Six insect species and a rust disease have been introduced against yellow starthistle in the United States. Of the six insects, five have established (see Table 2). Three of these are widespread: the bud weevil (*Bangasternus orientalis*), hairy weevil (*Eustenopus villosus*), and gall fly (*Urophora sirunaseva*). Two of the other insects, the peacock fly (*Chaetorellia australis*) and the flower weevil (*Larinus curtus*), occur only in a few isolated locations and have failed to build up numbers high enough to substantially reduce seed production. The sixth insect, *Urophora jaculata*, failed to establish and does not occur here. In addition to the five insects established as control agents, another insect, the false peacock fly (*Chaetorellia succinea*) was accidentally released in southern Oregon in 1991 and is now widespread throughout California (Balciunas and Villegas 1999). The false peacock fly is not an approved biological control agent and did not undergo host specificity testing prior to its accidental introduction. Fortunately, follow-up surveys of commercial safflower crops and native *Cirsium* thistles showed the fly to be fairly host specific to yellow starthistle (Villegas *et al*. 1999, 2000b; Balciunas and Villegas 1999).

All of the insects released for control of yellow starthistle attack the flower heads. All deposit their eggs either inside or on the immature flower buds and their larvae feed directly on the developing seeds or destroy the disk area on which the seeds develop. A statewide survey of the seed head insects found the false peacock fly to be the most common insect—it was recovered at 99% of the sample locations. The second most common insect was the hairy weevil, which was found at 80% of the sample locations (Pitcairn *et al*. 2003).

Several plant pathogens are known to attack yellow starthistle seedlings and rosettes in California: *Sclerotinia minor*, *Colletotrichum gloeosporioides* and a new species of *Ascophyta* (Woods 1996, Woods and Fogle 1998, Pitcairn *et al*. 2000b). All three species are naturally present in California. Seedlings of yellow starthistle were observed to be infested with *Ascophyta* n. sp. during two winters at one location in central California (Woods 1996). Unfortunately, infestations by *Ascophyta* n. sp. have not been observed since then. More commonly, *S. minor* and *C. gloeosporioides* have been observed to cause high mortality rates in starthistle seedlings at several locations, particularly in areas where skeletons of previous years starthistle plants provide shading. Both of these pathogens are not host specific and are able to infect important crops including lettuce (Pitcairn *et al*. 2000b). In contrast to *Ascophyta* n. sp., these pathogens are more aggressive at warmer temperatures, causing symptoms characterized by wilting and yellowing (Woods and Fogle 1998). It is important to note that none of these pathogens has been approved for use as a biological control agent and land managers need to rely on naturally occurring infection if their benefits are to be realized. It may be possible to isolate a host-specific form of *S. minor* or *C. gloeosporioides* that could be used as a mycoherbicide for use in infested grasslands, but this is many years away from development.

Under laboratory conditions, Klisiewicz (1986) looked at the effect of several pathogenic fungi on yellow starthistle rosettes. The species evaluated included *Fusarium oxysporum* f. sp. *carthami*, *Verticillium dahliae*, *Phytophthora* spp., *Botrytis cinerea*, and *S. sclerotiorum*. Starthistle plants developed symptoms following innoculation and,

False peacock fly. *Accidentally introduced, the false peacock fly is one of the most common seed head insects found on yellow starthistle. (Photo: B. Villegas)*

Biological control damage. *Buds attacked by the hairy weevil* (E. villosus) *early in development fail to flower (upper center). (Photo: B. Villegas)*

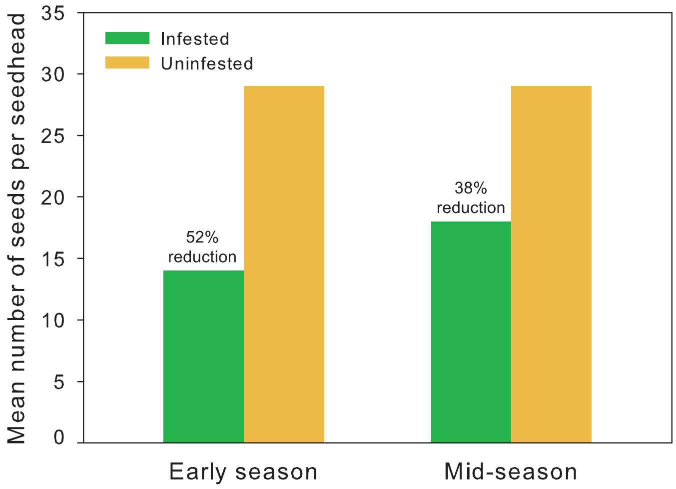

Fig. 18. Effect of insect control agent on seed production. *In a 1999 study near Folsom, CA, insect control agents reduced yellow starthistle seed production (mean number of seeds per head) by 45% on average (M.J. Pitcairn and J.M. DiTomaso, unpubl. data).*

with the exception of *B. cinerea*, the diseases were frequently lethal. However, with the exception of *S. sclerotiorum*, none of these pathogens has been observed to attack starthistle under field conditions. As with the endemic seedling pathogens, none of these diseases is host specific and, thus, all have the potential to attack other economically or ecologically important plant species.

Recently, the Mediterranean rust fungus (*Puccinia jaceae* var. *solstitialis*) has been approved for release in the western United States. Research on this pathogen was initiated in 1978 from isolates collected in Turkey. Since then, the pathogen has undergone a long series of host specificity tests in the USDA-ARS quarantine lab in Fort Detrick, Maryland. The test results showed that this pathogen is highly host-specific, it can infect only a couple of exotic *Centaurea* species, and that its preferred host is yellow starthistle (Shishkoff and Bruckart 1993). The first release of this rust occurred on a private land trust in Napa County in 2003 (Woods *et al*. 2004). In 2004, releases occurred at 25 locations in 22 counties, and, in 2005, releases occured at 99 locations in 38 counties. The rust attacks the leaves and stem of the rosettes and early bolts of starthistle, causing enough stress to reduce the number of flower heads and seed production. Thus, it complements the damage caused by the seed head insects. Preliminary laboratory data suggest that it is well suited to the environmental conditions found in California (Bennett *et al*. 1991), but it is too early to know for sure. It may be limited to areas with sufficient dew period to allow sustained infection during spring; however, this is yet to be determined.

Current Status of Yellow Starthistle Biological Control

The combined impact of five of the insects (except the peacock fly) has been evaluated at three long-term study sites in central California (Pitcairn *et al*. 2002). The hairy weevil and the false peacock fly are the most abundant insects and appear to cause the largest amount of seed destruction. The other three insects failed to build up high numbers and have had little impact on seed production. Since 1995, seed production at the three study sites has steadily declined due to the steady increase in attack by the hairy weevil and the false peacock fly. Recently, the density of mature plants has declined at two sites (Pitcairn *et al*. 2002). Although it is too early to know the stable level of control provided by the seed head insects, as of 2004 mature plant density had declined over 50% at both sites. It is important to note that these sites experienced no

Seed head damage. *This seed head has been damaged by the false peacock fly* (Chaetorellia succinea). *(Photo: B. Villegas)* disturbance from grazing, mowing, or other control methods and it is likely that the endemic plant community also contributed to the suppression of yellow starthistle through interspecific competition. By comparison, control of yellow starthistle at disturbed sites, such as along roadsides, may not occur due to the lower level of plant competition.

Two additional biological control agents are now under preparation for use in California. The first is the Mediterranean rust disease, discussed above. It is expected that infection of the rosette and stem leaves by this disease will stress the plant causing reduced growth, fewer number of seed heads, or possibly early death. The second biological control agent (not yet released) is the rosette weevil (*Ceratapion bassicorne*). This weevil deposits its eggs on young rosettes and its larvae burrow into the root and up into the bolting stem. Field observations in Turkey show that attack by the weevil results in shorter plants with fewer seed heads compared to unattacked plants (Uygur *et al.* 2005). Infection by the rust and attack by the rosette weevil occur in late winter through spring. This will be followed by the attack of the seed head insects in summer. It is hoped that these two new biological control agents will complete a guild of herbivores and pathogens sufficient to control yellow starthistle in the western United States.

Choice of Biological Control Agents

Of all the seed head insects, only two, the hairy weevil and the false peacock fly, have proven to consistently build up high numbers and cause a substantial amount of seed destruction (Pitcairn and DiTomaso 2000, Woods *et al*. 2002, Pitcairn *et al*. 2003). The combination of these two insects has been reported to reduce seed production by 43 to 76% (Pitcairn and DiTomaso 2000). Balciunas and Villegas (1999) reported a 78% reduction in seed production when seed heads contained false peacock fly larvae alone. The hairy weevil is an approved agent and is available for use to landowners. The false peacock fly is not a permitted biological control agent and therefore is unavailable. Despite this, the false peacock fly is a very common insect and is found almost everywhere that yellow starthistle is known to occur (Pitcairn *et al*. 2003). It is likely that the false peacock fly is already present at locations identified for yellow starthistle control (to check this, see monitoring methods, below). In developing a biological control program, it is recommended that efforts be directed at establishing the hairy weevil throughout the infested area; it is expected that the false peacock fly will build up on its own.

While the rust has been released in California, its continued release and establishment is regulated

Mediterranean rust fungus. Puccinia jaceae *var.* solstitialis *on yellow starthistle. (Photo: D. Woods)*

Damage by hairy weevils. *Yellow starthistle plants may respond to damage caused by adult hairy weevils by emitting sap around the damaged area. (Photo: B. Villegas)* by the California Environmental Protection Agency (CalEPA) and will likely not be available for general use. Current releases by CDFA are permitted under an experimental use permit and are limited to 10 acres per year. The goal of the CDFA distribution effort is to establish the rust in all regions where yellow starthistle is known to occur. Rusts produce millions of spores that are easily transported by wind. It is expected that, once established, the rust will spread on its own throughout the nearby yellow starthistle populations. In contrast, CDFA and other state agricultural departments will distribute the rosette weevil at no cost to the user. It is unknown when the rosette weevil will be ready for distribution.

Methods and Timing

SELECTION OF RELEASE SITES

The objective of a biological control program is to establish self-sustaining populations of the biological control agents at locations throughout the infested area. First, release sites must be identified for the biological control agents. The release site should contain at least one acre of yellow starthistle that is undisturbed by farm equipment, vehicular traffic, livestock (no grazing), mowing, and pesticide use. These sites are small refugia that allow the control agent to reproduce and build up high numbers that eventually spread outward into adjacent yellow starthistle populations. To build up their population, the control agents require yellow starthistle to reproduce and develop. Insects are killed if the plant is destroyed before flower maturation. The release site should have a moderately dense infestation of yellow starthistle; however, the plant population should not be so dense that plants are stressed and stunted. Ideal release sites are areas where application of herbicides is not permitted, such as near stream corridors, or areas that are inaccessible by equipment such as hillsides or ravines. It is not necessary to release insects everywhere on a landscape. Rather, a few locations strategically spread throughout the property are sufficient. Distance between release locations can be as much as a five miles and still result in effective spread and coverage by the biological control agents.

Post-release monitoring. *After release, biocontrol insects are carefully monitored for effectiveness and spread. (Photo: M. Pitcairn)*

RELEASE OF THE HAIRY WEEVIL

The hairy weevil has one generation per year. It overwinters under plant litter near the base of yellow starthistle plants, along fence rows, or at the base of trees (Pitcairn *et al.* 2004). It terminates its diapause in late spring when adults can be seen feeding on young buds on the newly bolted yellow starthistle plants. Collection of the hairy weevil for distribution to new areas is best during late June and early July when females are beginning to deposit eggs into the seed heads. In California, the hairy weevil are available at no cost to the user from each County Agricultural Commissioner's Office. For each release, only 100 weevil adults are necessary to establish a viable population. If more weevils are available, it is best to distribute them to as many different locations as possible, rather than concentrating them at one or two release sites.

Monitoring Seed Head Insects

HAIRY WEEVIL

The presence of the seed head insects is best determined by looking for adult insects sitting on the flower buds or by observing damage caused by each of the insects "(Pitcairn *et al.* 2004). The hairy weevil is very destructive to the seed head and its damage is distinctive. Both males and females feed on the young undeveloped flower buds by chewing a small hole in the base of the bud and eating away the developing tissue (Connett and McCaffrey 2004). This feeding damage causes the whole young bud (buds with diameters less than 1/8 inch) to die and turn brown. At locations with high populations of hairy weevils, most of the young flower buds may be killed by their feeding damage. Following destruction of its early flower buds, the plant responds by developing flowers along the stems. This substantially changes the architecture of the plant. Undamaged plants have the flowers located at the top of the plant on long stems, while plants damaged by *E. villosus* are less bushy with flowers located close to the branches on short stems. Later, the female weevil oviposits by chewing a hole in the side of the flower head and depositing an egg inside the head. The hole is then filled with a black plug by the female to protect the egg. The plant responds to the chewing damage by emitting a dark sap that fills in around the damaged area of the flower head. This type of damage can be seen in July and August. The black plug is easily seen on the outside of the flower head. Sometimes the area around the plug is distorted and the dark sap oozes out of the head. Adult hairy weevils are active during the day and can be observed sitting on the seed heads and stems of yellow starthistle plants. Adults can be captured with a sweep net passed through the plants.

FALSE PEACOCK FLY

False peacock flies can be detected by looking for ovipositing adults or by tearing apart seed heads and observing the larvae and pupae. The adult flies are slightly smaller than a housefly, and have blond bodies with brown stripes on clear wings. They are easily seen sitting on the seed heads during the day. The female oviposits by inserting her ovipositor between the bracts of the unopened flower bud and depositing several eggs. After hatching, the larvae burrow throughout the seed head and feed on the developing seeds. When ready to pupate, the larva becomes a swollen pupal capsule that is blond in color and approximately 1/10 inch long. The pupae are usually located near the base of the bracts. They can be seen by breaking open the seed head. Adults can be captured with a sweep net passed through the plants.

OTHER SEED HEAD INSECTS

The larvae of the gall fly, *U. sirunaseva*, produce hard, woody galls inside the seed head (Pitcairn *et al*. 2004). They occur like small hard nuts inside the head, approximately 1/10 inch in diameter. The adult flies frequently forage among the seed heads. The adult gall fly is approximately half the size of the false peacock fly and their bodies are black with yellow legs while their wings are clear-colored with black marks across the surface.

Presence of the bud weevil, *B. orientalis,* at a site is best indicated by the presence of eggs on or directly below the flower buds. The eggs are round, black ball-like structures glued to the stem. Within the black structure is a single yellow egg. The female secretes the black material covering the egg to adhere the egg to the plant and to protect it from desiccation.

Economics

The major advantage of weed biological control is that it is considered to be environmentally safe, cost-effective, and self-sustaining. The high cost of developing biological control is borne upfront in the foreign exploration, host testing, and permitting of candidate biological control agents. However, the significant long-term benefits of a successful biocontrol program make it very cost-effective. Once approved and released, distribution of the agents is generally conducted by federal and state agencies. In California, the California Department of Food and Agriculture and the Offices of the County Agricultural Commissioners distribute biological control agents at no cost to the land manager. Ideally, if biological controls are successful, weed populations will slowly decline and become much easier to manage using conventional control methods. In very successful programs, biological controls may eliminate the need for additional control efforts altogether. Some costs may result from the delay between release of the agents and the time when their populations have increased sufficiently to cause a reduction in the plant populations. This delay may be substantial. For yellow starthistle, 4-6 years elapsed before reduction in starthistle populations was observed at the two long-term monitoring sites.

Risks

Despite the overwhelmingly positive aspects of biological control, some risks do exist. These risks are associated with the introduction of an exotic organism and can result in direct or indirect impacts to non-target species. Direct impacts occur with feeding on non-target plant species. Indirect non-target impacts consist of changes in abundance of endemic predators (such as field mice) that may alter foraging behavior and exploit a new resource. This can lead to changes in the community food web.

Host-specificity testing of candidate biological control agents has been shown to be a good indicator of host use in the exotic habitat. A review of insects introduced into North America for use as biological control agents showed that all have performed as expected, and that no plants identified as unsuitable during host testing became targets after release of the agents in the field (Pemberton 2000). Approximately 10% of the control agents examined do attack some native plant species, but these were predicted by the host specificity testing. All of these agents were released prior to 1970 when attack on weedy native plants was considered beneficial. Today, attack on native plants is undesirable and the required level of host-specificity of biological control agents has increased.

For yellow starthistle, none of the seed head insects has been observed to attack any native non-target plant species. Based on genetic similarity, yellow starthistle is most closely related to other species in the tribe Cardueae. Within this tribe are safflower (*Carthamus tinctorius*), artichoke (*Cynara scolymus*), sunflower (*Helianthus annuus*) and *Cirsium*, a genus of native thistles (Stevens *et al*. 1990, Keil 2004). Many surveys of these potential non-target species have been performed (Villegas *et al*. 1999, 2000b; Balciunas and Villegas 1999), and no evidence of non-target use of agricultural and native plants has yet been observed. Some use of other exotic plants by yellow starthistle bioagents has been observed. For example, the hairy weevil will attack several exotic *Centaurea* species, including Sicilian starthistle (*C. surphurea*), Malta starthistle or tocalote (*C. melitensis*), and spotted knapweed (*C. maculosa [=C. biebersteinii]*). All are exotic noxious weeds. Thus, the risk of direct non-target attack by the yellow starthistle insects is extremely low.

The risk of indirect impacts also appears to be very low for the biological control agents of yellow starthistle. Pearson *et al*. (2000) found that gall flies used as biocontrol agents on spotted knapweed (*Centaurea maculosa*), caused indirect increases in populations of deer mice by providing a food source over the Montana winter. However, a similar scenario is unlikely with yellow starthistle, because yellow starthistle favors mild-winter areas and is an annual plant which dies by winter.

Hairy weevil damage. *This yellow starthistle bud will never open due to damage from the hairy weevil. (Photo: B. Villegas)*