Evaluating implementation success for seven seed head insects on *Centaurea solstitialis* in California, USA

M.J. Pitcairn,1 B.Villegas,1 D.M. Woods,1,2 R. Yacoub3 and D.B. Joley4

Summary

A stage-based invasion model proposed by Colautti and MacIsaac (2004) is used to evaluate the success of implementation efforts for seven seed head insects introduced on *Centaurea solstitialis* L. (Asteraceae) in California, USA. Six insects were introduced intentionally; one was introduced unintentionally. Establishment of initial foreign material (stage III invasion) at field nursery sites was very successful (86%) with six of seven species establishing (five of six intentional, one of one unintentional). For the intentional species, initial releases occurred at multiple locations, and establishment success among locations was 100% for four of the five species that established. Statewide distribution of seed head insects introduced intentionally (stage IV invasion) was performed through the network of County Agricultural Commissioners. Training workshops provided knowledge and insects to county biologists for release in their county. Hundreds of releases occurred in this way, and follow-up monitoring estimated establishment success above 87% for four of six species. Regional spread and population increase to high density (stage V invasion) was examined in an independent statewide survey of *C. solstitialis* seed heads in 2001 and 2002. Results showed that only one of the intentional species, *Eustenopus villosus* (Boheman) and the unintentional species, *Chaetorellia succinea* (Costa), appear to have achieved high densities over large areas. The fly, *C. succinea*, appears to be the most successful seed head insect, while *E. villosus* was second in abundance statewide. The other seed head insects were recovered in low numbers statewide and appear to contribute little to the herbivore pressure on this weed.

**Keywords:** invasion, stage-based implementation model.

Introduction

Development of a biological control organism may be grouped into pre-release and post-release research efforts. Pre-release efforts include the foreign exploration and host-specificity testing of candidate natural enemies, among other efforts (Harley and Forno, 1992; Briese, 2000). Post-release efforts begin with the approval to release the control organism from quarantine and consist of release and establishment efforts, field impact studies and non-target surveys (Hansen, 2004). The multi-year release and establishment effort is the ‘implementation phase’ of a new biological control organism and is commonly carried out by state and local government entities. This phase occurs in several steps: initial establishment of foreign material in field nursery sites; distribution of domestic material from nursery sites to satellite locations throughout the region; and eventual spread of the natural enemy away from release sites into the surrounding infestations of the target weed. The goal is to obtain self-sustaining populations of the biological control agent throughout the area infested by its host plant.

Several researchers have pointed out the similarity between the intentional introduction of a biological control organism and the invasion of an unintentionally introduced exotic organism (Crawley, 1989; Simberloff, 1989; Grevstad, 1999; McEvoy et al., 2000). Colautti and MacIsaac (2004) and Colautti (2005)
proposed a stage-based description of the invasion process over large geographic areas for invasive species. Here, we modify their model to describe the stages of the implementation phase of a biological control organism and use this model to evaluate the success of implementation efforts for a guild of exotic seed head insects introduced against *Centaurea solstitialis* L. (Asteraceae), commonly named yellow starthistle, in California (USA).

**Methods**

**The invasion model**

Colautti and Maclsaic (2004) and Colautti (2005) proposed a model of six stages to describe the invasion of an exotic organism into a new region. Stage 0 is the organism in its area of origin; in stage I, the organism is taken up by a transport vector (ship ballast, contaminant in a seed shipment, etc.); in stage II, the organism is introduced into the new region; in stage III, the organism establishes a small, incipient population; in stage IV, the organism spreads throughout the new region; and in stage V, the organism has spread over a large region and generally occurs in high density. Stage IV can be achieved through two processes: formation of several small satellite populations away from the initial infestation (stage IVa) or simple expansion of the initial established population (stage IVb). Both stages IVa and IVb expand to stage V.

The development and utilization of a biological control organism may be described in a similar way. Stage 0 is the natural enemy in its host’s area of origin; in stage I, a sample of the natural enemy population is collected and held in quarantine for host-specificity testing; in stage II, a sample of the material in quarantine is released on the target weed in the new region; in stage III, the organism has established small populations in field nursery sites; in stage IV, biological control workers collect the organism from the initial nursery sites and distribute it throughout the region infested by the target weed. The goal is stage V, where the biological control organism forms self-sustaining populations of high density throughout the regions infested by the target weed. In this model, the implementation phase of a biological control organism begins with its approval to release (stage II) and ends with the biological control organism occurring in high density throughout the area infested by the target weed (stage V).

**Biology of yellow starthistle in California**

Yellow starthistle is an invasive exotic weed from the Mediterranean region of Europe and was likely introduced into California as a contaminant in shipments of alfalfa seed (DiTomaso and Gerlach, 2000). It was first recorded near the San Francisco Bay in 1859 and now infests over 5.7 million ha in California alone (Pitcairn et al., 2006). Yellow starthistle is a winter annual that invades rangelands, orchards, vineyards, pastures, parks and natural areas. It is favored by soil disturbance but can invade areas that have not been disturbed by humans or livestock. Germination begins with the onset of the winter rains; flowering begins in early summer and continues into the fall. Individual plants may produce from one to 100 capitula (hereafter, seed heads); infested areas commonly produce 650 to 700 seed heads per square metre, but densities as high as 3000 seed heads per square metre have been reported (DiTomaso et al., 2003). In uninfested heads, approximately 30 to 40 achenes (hereafter, seeds) per head are produced. Yields of 50 million seeds/ha are common (M. J. Pitcairn, unpublished data), but estimates of 120 to 500 million seeds/ha have been recorded in heavily infested areas (Maddox, 1981; DiTomaso and Gerlach, 2000).

Yellow starthistle is the target of an ongoing biological control effort in the western United States. Research efforts resulted in the release approval of six exotic insects that oviposit in the immature seed heads, and their larvae feed on developing seeds. The first insect released was the gall fly, *Urophora jaculata* (Rondani) (Dipt.: Tephritidae), in 1969, but it failed to establish (Turner et al., 1994). Five other insects were released from 1984 through 1992, and all have established: *Bangasterus orientalis* (Capiomont) (Col.: Curculionidae), *Eustenopus villosus* (Bohemian) (Col.: Curculionidae), *Urophora sirunaseva* (Hering) (Dipt.: Tephritidae), *Chaetorellia australis* (Hering) (Dipt.: Tephritidae) and *Larinus curtus* Hochhut (Col.: Curculionidae) (Turner et al., 1994). In California, three species, *B. orientalis*, *E. villosus* and *U. sirunaseva*, are now widespread. The two other insects, *C. australis* and *L. curtus*, occur in low numbers at a limited number of locations. A seventh insect, *Chaetorellia succinea* (Costa) (Dipt.: Tephritidae), was accidentally introduced into southern Oregon in 1991 and is also widely established throughout California (Balciunas and Villegas, 1999).

**Evaluation of the implementation stages**

**Stage III - releases of foreign material:** The six insect species intentionally released as biological control organisms were collected from their area of origin (Greece, Italy and Turkey) as larvae in seed heads, sent to the United States Department of Agriculture, Agricultural Research Service quarantine facility in Albany, CA, USA and held for emergence in sleeve cages (Turner et al., 1996). Emerged adults were collected, identified and a subsample killed and examined for internal parasites and diseases. When ready, the remaining adults were released into field nursery sites (Turner et al., 1996). Most nursery sites were monitored annually for recovery of the insect and, when population levels were considered
high enough, the sites were used for training and a source of adults for distribution in stage IV. **Stage IV - releases of domestic material**: Stage IV involves the transfer of knowledge and biological control organisms from state control to local control. The State of California is divided into 58 counties for the purposes of supporting local governmental activities. Each county supports an Agricultural Commissioner who maintains a small staff of biologists that assist with issues concerning local agriculture. The California Department of Food and Agriculture performs workshops and training sessions for the county biologists on the biology of biological control organisms and the damage they cause to the target weed (Villegas, 1998). Training occurs through oral presentations at field nursery sites and through active participation of the attendees in the collection, counting and packaging of biological control organisms. Afterwards, workshop attendees return to their counties and release the organisms at new field sites. This network of trained county biologists has been very effective in the distribution of biological control organisms throughout California.

A sample of stage IV release sites \((n = 60–120)\) were monitored 3 to 4 years following their initial releases. The sites were chosen to represent the different climatic regions where yellow starthistle grows. Plants were swept with a sweep net at late bud stage or early flowering to collect any active adult seed head insects, and any evidence of oviposition and feeding damage was recorded.

**Stage V - local spread and population increase**: It was expected that each of the seed head insects would increase their densities locally and spread throughout the populations of yellow starthistle located nearby. The regional spread of the seed head insects away from their release sites was evaluated by a survey performed in 2001 and 2002 where plant samples were collected along roads throughout areas infested with yellow starthistle. Samples occurred approximately every 16 km. Over 100 seed heads from at least ten plants were collected, and the date and latitude and longitude coordinates for each sample location were recorded. All seed heads were returned to the laboratory, and a minimum of 100 heads from each sample was dissected, and the presence of insects was recorded by species. Each biological control organism produces a characteristic type of feeding damage that is easily recognized upon dissection of the head. The exception was the damage caused by the two species of Chaetorellia. A second survey where adult flies were reared from seed heads showed that C. australis was infrequently recovered, and its infestation rate was very low (<5%). Thus, for the analysis reported here, we consider all heads damaged by Chaetorellia spp. to be infested with C. succinea.

Typically, immature seed head production begins in May with peak production in early July (unpublished data). Seed heads accumulate on plants and provide a record of the cumulative attack by insects throughout the season. Survey collections began after peak seed head production (late July). A total of 421 samples were collected during the two summers of this study. A data file of insect abundance based on the proportion of seed heads infested was plotted using a geographic information system (GIS), and values between points were interpolated using an inverse-distance weighting (IDW) algorithm that covered the entire state of California.

### Results

**Stage III – releases of foreign material**

Five of the six seed head insects released as approved biological control organisms established (Table 1). For each species, releases of foreign material occurred in three to seven locations within California (additional releases occurred in Oregon, Washington and Idaho; Turner et al., 1994). Establishment rates among locations were 100% except for U. jaculata, which failed to establish, and for C. australis, which failed to establish at all five release sites in California. An overall establishment rate of 27% for C. australis is estimated from its rate of establishment among all locations in the other western states (Turner et al., 1996).

The unintentional release of C. succinea occurred when foreign material from quarantine consisting of a mixture of C. succinea and C. australis adults was released at a field site near Merlin, Oregon (Balciunas and Villegas, 1999). Both flies apparently established. Examination of the voucher specimens retained from the overseas shipments received in quarantine suggested that the accidental release of C. succinea occurred just once (Balciunas and Villegas, 1999). Thus, the establishment rate of C. succinea in stage III is 100% (one of one).

**Stage IV – releases of domestic material**

Three insects, U. sirunaseva, B. orientalis and E. villosus, built up high populations at field nursery sites and were distributed statewide through a distribution program. The program operated primarily through a series of workshops designed to train county biologists in the identification, collection and release of the biological control organisms. Based on the training received at the workshops, county biologists collected available biological control organisms from their release sites in stage III then returned to their county and released the insects at their own nursery sites. The new county sites would serve as sources for further distribution of insects within their counties. Training workshop sites began 2 to 3 years following initial release of foreign material in stage III. A total of 41,380 domestically produced U. sirunaseva was released at 163 locations, 80,290
**Table 1.** Summary for the implementation of seven seed head insects on yellow starthistle (Centaurea solstitialis L.) in California (USA). The values for stages III and IV represent the proportion of releases that successfully established.

<table>
<thead>
<tr>
<th>Species</th>
<th>Stage III (%)</th>
<th>Stage IV (%)</th>
<th>Stage V</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urophora jaculata (Rondani)</td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Urophora sirunaseva (Hering)</td>
<td>100</td>
<td>87</td>
<td>No</td>
</tr>
<tr>
<td>Bangasterus orientalis (Capionmont)</td>
<td>100</td>
<td>92</td>
<td>No</td>
</tr>
<tr>
<td>Chaetorellia australis (Hering)</td>
<td>27</td>
<td>19</td>
<td>No</td>
</tr>
<tr>
<td>Eustenopus villosus (Boheman)</td>
<td>100</td>
<td>92</td>
<td>Yes</td>
</tr>
<tr>
<td>Larinus curtus Hochhut</td>
<td>100</td>
<td>25</td>
<td>No</td>
</tr>
<tr>
<td>Chaetorellia succinea (Costa)</td>
<td>100</td>
<td></td>
<td>Yes</td>
</tr>
</tbody>
</table>

*Turner et al. (1994).*<sup>a</sup>  
*Turner et al. (1996).*<sup>b</sup>  
*Balcinius and Villegas (1999).*<sup>c</sup>

_B. orientalis_ at 371 locations, and 316,000 _E. villosus_ at 1024 locations. Post-release monitoring at a subset of these locations estimated establishment at 87% (45 of 52, _U. sirunaseva_), 92% (118 of 128, _B. orientalis_) and 92% (119 of 129, _E. villosus_ Table 1).

Stage IV distribution of _C. australis_ and _C. succinea_ in California resulted from a release effort during 1994 through 1996 (Balcinius and Villegas, 1999). Encouraged by high populations of what was thought to be _C. australis_ in southern Oregon, 9463 Chaetorellia adults were collected and released at 21 locations in California. It was during this release effort that the presence of _C. succinea_ was discovered, and releases were discontinued. Post-release surveys of the 21 release sites recovered _C. australis_ at four locations (establishment rate = 19%) and _C. succinea_ at 21 locations (establishment rate = 100%).

The performance of the weevil, _L. curtus_, following establishment in stage III was disappointing as populations failed to increase and were recovered in low numbers several years afterward. A small number of field-collected adults from several release sites were found to be infested with a species of _Nosema_, a microsporidian parasite of the alimentary canal. A population of _L. curtus_ in northern Oregon had readily established and built up high population numbers. Field samples later revealed no infection by _Nosema_ at this location. It was thought that the microsporidian hindered population growth, so over 5600 adult _L. curtus_ were collected from northern Oregon and released at 23 locations in California from 1997 to 1999. Post-release monitoring showed that the weevil established at five of 20 locations (establishment rate = 25%), but densities of the established populations remained extremely low.

**Stage V – local spread and population increase**

Four species of seed head insects were recovered during an independent survey of yellow starthistle plants away from release sites in 2001 and 2002: _B. orientalis_, _U. sirunaseva_, _E. villosus_ and _C. succinea_. All were found widespread throughout the state but in varying levels of abundance. Both _B. orientalis_ and _U. sirunaseva_ were recovered from 62% of the locations but occurred mostly in low numbers (usually <15% of seed heads attacked). In contrast, _E. villosus_ was recovered at 80% of the locations, and its abundance ranged from 0% to 93% of the heads attacked. The fly, _C. succinea_, was recovered at 99% of the locations, and its abundance ranged from 0% to 96% of the heads attacked. The weevil, _L. curtus_, was not recovered. The fly, _C. australis_, was recovered in a follow-up survey where adult flies were reared from sampled seed heads; however, adult emergence rarely exceeded five flies per 100 heads.

Insect abundance, estimated as the number of heads attacked per sample, was interpolated between sample locations using a GIS system, and the resulting maps showed regional differences in abundance. For each insect species, the frequency distribution of abundance values was sorted by magnitude and divided into ten quantiles. The top quantile consisted of values in the top 10% of those recorded for the species. Areas of highest abundance were identified as those areas with interpolated values within the top quantile (Fig. 1). For _B. orientalis_, locations with abundance values above 18% heads attacked represented the top quantile, and these occurred in the mountain ranges of the most northern areas of California. For _U. sirunaseva_, locations with values above 9% attack rate represented the top quantile, and these areas were scattered within the northern and southern regions of the Coastal Mountain Range. For _E. villosus_, its top quantile consisted of locations with values above 41% attack rate, and these were observed throughout the mountains of northern California and along the foothills of the Sierra Nevada Mountains. Areas with the lowest abundance of _E. villosus_ were observed in the San Joaquin and Sacramento Valleys. The fly, _C. succinea_, was the most common seed head insect recovered in the survey. Its top quantile consisted of abundance values above 70%. However, in comparison with the other seed head insects,
Evaluating implementation success for seven seed head insects on *Centaurea solstitialis* in California, USA

Abundance values above 50% are considered high. For *C. succinea*, areas with interpolated abundance values above 50% occurred throughout central and southern California (Fig. 1).

**Discussion**

The stage-based implementation model identified three main stages in the release and establishment of a biological control organism: establishment of foreign material from quarantine (stage III), establishment of domestic material (stage IV) and the increase and spread of the control organism throughout the region (stage V). Examination of establishment rates among stages shows that implementation of the six seed head insects intentionally introduced as biological control organisms was very successful. Five of six species established (83%) and establishment rates of the foreign material were 100% for four of five species. Collection and distribution of material produced domestically (stage IV) was also fairly successful with three of five species showing establishment rates above 87% throughout California. Interestingly, the accidentally introduced fly, *C. succinea*, also showed a high rate of establishment (100% in stages III and IV). However, the regional spread of the seed head insects at stage V was not as successful as only two species, *E. villosus* and *C. succinea*, appear to have built up populations approaching high densities.

**Figure 1.** Plot of interpolation values for the four seed head insects recovered from *Centaurea solstitialis* L. in California during 2001–2002. All plots show areas of highest abundance as indicated by seed head attack rates in the top 10% of observed values (top quantile). The weevil *Bangasternus orientalis* and the fly *Urophora sirunaseva* rarely exceeded attack rates of 18% and 9%, respectively. The weevil *Eustenopus villosus* and the fly *Chaetorellia succinea* occurred in higher numbers (>41% and 50%, respectively). Sample locations are indicated by the filled dots.
over large regional areas. The fly, *C. succinea*, appears to be the most successful organism with estimated attack rates greater than 50% over half of the area infested by yellow starthistle (Fig. 1). The weevil, *E. villosus*, was second in abundance with approximately 35% of the yellow starthistle infestation experiencing attack rates greater than 41%. None of the other insects appear to contribute much to the overall natural enemy pressure on this weed.

Several studies have identified propagule pressure as an important factor in the successful establishment of invading organisms. For example, Kolar and Lodge (2001) reviewed published studies on invasive species in an effort to identify attributes that might predict probable invaders. While the information they presented was limited to only a few taxa, one strong pattern was that successful establishment was positively related to propagule pressure. Propagule pressure, either number of individuals released or number of releases, is one factor that biological control workers have increasing control over due to improvements in rearing technology and faster, more efficient shipping abilities. For implementation efforts, establishment rates in stages III and IV would benefit from increases in the number of insects used. The only stage over which we appear to have little control is stage V. Reviews of past biological control projects (e.g. Hall and Ehler, 1979; Crawley, 1989; Simberloff, 1989; Coombs, 2004) have concentrated on establishment success in stages III and IV but provided little examination of projects in stage V. While establishment is an important component in the implementation of a biological control organism, ultimate success in controlling the target weed may reside in the attributes of control organisms that result in their ability to obtain high densities and regional spread. More research on the transition of biological control organisms to stage V may greatly improve our understanding of the attributes of an effective biological control agent and may lead to releases of more effective agents in the future.

**Acknowledgements**

We thank Kathy Chan for providing release information from unpublished quarantine records located at the USDA Agricultural Research Service facility in Albany, CA.

**References**


Evaluating implementation success for seven seed head insects on *Centaurea solstitialis* in California, USA

