

California Department of Food and Agriculture
Integrated Pest Control Branch
Biological Control Program
3288 Meadowview Road
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WORK PLAN FOR COLONIZATION RELEASES OF THE YELLOW STARHISTLE RUST, *Puccinia jaceae* VAR. *SOLSTITIALIS* FOR THE BIOLOGICAL CONTROL OF YELLOW STARHISTLE IN CALIFORNIA

I. INTRODUCTION

The rust, *Puccinia jaceae* var. *solstitialis* is a natural enemy of yellow starthistle in Europe. In 1978, a sample collected from Turkey was brought to a USDA quarantine facility in Maryland for host specificity testing. Testing was completed by Dr. William Bruckart, USDA-ARS demonstrating that the rust was highly host specific. The CDFA Biological Control Program, in co-operation with Dr. Bruckart, received a limited release permit for field release in selected sites in California in 2003.

The rust was field released at a single isolated site in Napa County in July 2003. A second release was made in a raised planting box at our Meadowview facilities in Sacramento also in July 2003. In both cases the rust successfully infected the inoculated plants. The Napa release was made when the plants were fully mature. As of January 2004, the rust is in an overwintering mode at this site. The Meadowview release has plants that were watered daily and the fungus has continued to be active through the winter and up to the present time.

Proposal for the 2004 calendar year – If sufficient inoculum can be prepared, 20 releases are established as a goal for the spring/summer of 2004. An additional 20 or more releases will be made the following year. Releases are contingent on approval by USDA-APHIS of a pending amendment of our field release permit. If the proposed amendment is approved, we will proceed with this plan. If approval cannot be obtained, releases will be limited to the counties in the original permit (Napa, Placer, Sonoma and Shasta).

Proposed release sites – Twenty counties were selected to a) represent the diversity of climates where yellow starthistle is found, b) distribute the rust geographically over the state and c) fulfill the need to accumulate data on the fungus that will improve future biological control efforts. The following counties (listed south to north), are proposed for release: Santa Barbara, San Luis Obispo, Tulare, Monterey, Fresno, Merced, Santa Clara, Tuolumne, Contra Costa, Solano, Sonoma, El Dorado, Yolo, Nevada, Glenn, Mendocino, Plumas, Tehama, Shasta, Siskiyou. A subset of counties (Monterey, Solano, Yolo and Sonoma) will be used by the Biological Control program for intensive post-release monitoring to examine spread and impact of the rust on

yellow starthistle. Releases in the other counties will be monitored only for establishment and spread.

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Expected time frame – The 2004 releases should begin late March or early April in the southern portion of the state and will progress northward in the following weeks.

Release procedure – Each release will be a one square meter plot. The ground should be moistened prior to release of the fungus. A spore sample will be mixed with water along with a wetting agent in a hand-held plant mister. The spore suspension will be sprayed on the square meter area of plants, and then covered with a plastic dew tent. Inoculation will be in the late afternoon or evening (to avoid sun exposure). The dew tent will be removed the next morning. A second night of dew is preferred and may improve the likelihood of successful establishment.

Expected follow-up – Each release will be followed up with infield monitoring to determine establishment and spread. Under ideal conditions, the rust may readily infect and new pustules may appear in 10 days. Since these releases are being made under much colder conditions than in our laboratory tests, the first evidence of infection may take as long as three weeks. A detailed procedure will be provided at the workshop covering methods to accumulate evidence of establishment and spread from the release site.

II. OBJECTIVES

1. To establish county nursery sites that will serve as natural distribution centers for further movements of the rust in California.
2. To monitor and determine establishment of the rust at each county nursery site.
3. To provide training to county staff and cooperating agencies in the release of a microbial biological control agent and its subsequent monitoring and movement methods.

III. PERSONNEL

This biological control field implementation project is being organized by the CDFA Biological Control Program under the general supervision of Baldo Villegas. Dr. Dale Woods, Ms. Viola Popescu and Dr. Mike Pitcairn will assist in all aspects of the rust collections, field applications, and subsequent monitoring. Following is their contact information:

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If the selected counties would like to participate, they are expected to designate a staff biologist to work on this biological control project. The designated county biologists will be given 1) an informational packet that will include information on the rust fungus along with quarantine and pesticide use permits, and 2) a "Rust Application Kit" which will include materials for a setting up a one square meter dew tent, misting bottle, wetting agent, and a sample of the rust fungus in order to apply the fungus at a county nursery site. County biologists MUST attend a scheduled training workshop in order to receive the informational packets and the Rust Application Kits. The rust fungus must be applied at a pre-selected field nursery site within 48 hours after attending the training workshop.

IV. PROCEDURES

A. Source of the Rust Fungal Spores

The Biological Control Program received its original supply of the yellow starthistle rust fungus from Dr. Bill Bruckart, USDA-ARS, located at Fort Dietrick, Maryland. There the rust was host tested and found to attack biotypes of yellow starthistle grown in California and not to attack other, closely related plant species, including safflower, artichoke, and native *Cirsium* thistles. Individual spores of the rust fungus were received by Dr. Dale Woods at the BC Program and they were applied to YST plants in a greenhouse at the Meadowview Facility in Sacramento County. Dr. Bruckart also supplied the aliquots of rust applied at the initial release site in Napa County during July 2003. Rust spores from infected YST plants at the Meadowview greenhouse have been massed collected since summer 2004 and stored frozen for the 2004 field applications throughout California.

B. County Nursery Site Selection Criteria.

1. A previous biological control site can be chosen, especially if a partnership has been developed with the property owner.
2. The site should contain AT LEAST ONE ACRE OF YST and should be UNDISTURBED by farm equipment, vehicular traffic, livestock (no grazing), rodents (major infestation), mowing, and pesticide use.
3. The site should have a moderately dense infestation of YST. However, the YST population should not be so dense that plants are stressed and stunted or the site is impenetrable or difficult to survey.
4. The site should have mixed habitats nearby (e.g., full sun, partial shade, etc.) that are infested with YST.
5. The owner of the property HAS TO BE CONTACTED and the property owner has

to give his/her permission for the application of the rust fungus to take place. Access to the release site for five years would be ideal. There will be periodic visits to the release site to assess the status of the rust fungus and the yellow starthistle plants by county, BC Program, and USDA personnel. Also, it must be OK with the property owner to mark the release area with flagged stakes.

C. YST Rust Collection and Distribution

Due to the conditions of the permits, only BC Program staff and designated county personnel will be allowed work with this microbial biological control agent at the county release sites. The current plan concerns releases in 2004-05 and counties should not attempt to distribute beyond the initial release site as it would be a violation of our EPA permit. Long term distribution programs to aid natural spread have not been formalized yet.

D. Training Workshops

The 20 selected counties have been subdivided into six groups of 3-4 counties each. A training workshop will be arranged with a county within each of the county subgroups. Adjacent counties to the selected county will send their county representative on the scheduled date and time. The duration of the workshop will be approximately 2-4 hours depending on the location of the site. Directions to the field site will be either faxed or emailed to each participating county approximately one week prior to the scheduled workshop.

Week 1	Week 2	Week 3
Santa Barbara San Luis Obispo (1) Monterey (2)	Tulare (1) Fresno Merced (2)	Tuolumne El Dorado (1) Nevada (2)
Week 4	Week 5	Week 6
Contra Costa (1) Santa Clara Solano (2) Sonoma	Yolo (1) Glenn (2) Mendocino	Plumas Tehama (2) Shasta (1) Siskiyou

NOTE: Number after the county indicates the possibility of a demonstration plot in that county subgroup.

Counties wishing to participate in a workshop should contact Baldo Villegas by March 15, 2004 to reserve space in one of the sessions. The workshops will cover all aspects of this protocol as well as all phases of the yellow starthistle biological control program with the rust fungus. At the end of the workshop, each participating county will be given a "Rust Application Kit" which will include all the materials needed to apply the rust fungus at a county nursery site. Application of the fungus at the pre-selected county site is expected to take place within 48 hours after attending the demonstration

workshop. Details of the application process are contingent upon the developing permit.

E. YST RUST Monitoring

Each county nursery site should be monitored starting two weeks after inoculation. Visual inspection every week for 4 weeks may be necessary for detection of infection. After evidence of infection is detected, a survey of surrounding plants will be initiated. Spread will be monitored every 2 weeks in 4 directions as the rust moves. The rust may move very little in the first year or may jump miles within a few weeks. Continued communication of field observations to BC Program staff will determine the distance to monitor. The actual time on site will be minimal and this request of commitment of county biologist time to repeat visits their site will hopefully be rewarded by indications of rapid spread.

V. TIMETABLE. Following is an outline of the proposed year-to-year activities per county nursery site:

A. First Year (Spring/Summer/Fall 2004) Activities

1. BC Program will check with State and Federal agencies to make sure that the permitting process for making 20 releases in the proposed counties is on track. It is hoped that approval of the permit will arrive soon in order to provide counties with lead time to locate and secure a possible release site.
2. Each participating county is expected to designate a staff biologist to work on this biological control project. Designated county staff will attend a workshop, be given informational materials and a "Rust Application Kit," and apply the rust fungus in a one square meter area at a pre-selected release site within two days (48 hours) of the workshop. A field release form including a map of the release site, description of the site, and latitude and longitude coordinates, must be filled out for every release made in order to satisfy the conditions of the permits. A copy of the form should be returned to the BC Program within two weeks of the release.
3. A field visit to the release site must be made starting two weeks after inoculation in order to determine pustule formation (colonization of the rust). Further monitoring visits at 1-2 week intervals are needed in order to monitor the progress of the rust colonization, infection levels and movement of the rust. The information gathered should be relayed to the BC Program.

B. Second Year and Following Years' Activities

- 1 The goal of the second and subsequent years activities is to determine successful infection of the rust in the application plot and to determine off site movement.
- 2 The BC Program will determine additional data gathering surveys that can be accomplished during the spring-fall monitoring period.
- 3 Efforts to monitor the movement of the rust to other sites will be investigated by the BC Program and determine the most effective methods.
- 4 Related plant species will be monitored for potential non-target impacts

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