United States Department of Agriculture

BIOLOGICAL CONTROL OF INVASIVE NATIVE AND NON-NATIVE PLANTS
NEW PROJECT PROPOSAL

Form Instructions: When copy and pasting into text fields please paste as "plain text".

Project Information
* The Submission (FY) refers to the current Fiscal Year.

	Program	Admin Region	Submission (FY)*	Additional Project ID	Report Type New Proposal (NP)
Project Number:	**BCIP**	**R1**	**19**	4	**NP**

Project Title: The dispersal of eriophyid mites with implications to host specificity testing and field releases.

Principle Investigator: Jeffrey Littlefield

Proposed Budget Summary (this table is auto-generated from Budget Information sections)

Year 1 BCIP Total	Year 2 BCIP Total	Year 3 BCIP Total	Total BCIP Funds	Total Funds (All)
$ 31,640	$ 32,080	$ 18,666	$ 82,386	$ 164,772

Subject Description

Target Invasive Plant:

Non-Native Invasive

Common and Scientific Names:

Hoarycress, Lepidium draba

Rush skeletonweed, Chondrilla juncea

Russian knapweed, Rhaponticum repens; Canada thistle, Cirsium arvense

Biological Control Agent(s):

Aceria drabae

Aceria chondrillae

Aceria sobhiani

Aceria anthocoptes

BCIP Priorities Addressed (check all that apply)

- [x] Developing improved rearing, host range testing, distribution and post-release monitoring techniques for a biological control agent.
- [ ] Development and/or implementation of technologies for monitoring/assessing plant trends and quantitative assessment of biological control impacts.
- [ ] Integrated weed management with a biological control component that is part of a methods technology development approach to determine efficacy and is not considered an operational treatment.
- [ ] Development of biological control strategies through funding of pilot projects.

Define the project being developed (e.g., what is the technology, goals and objectives, and who will maintain the tool if maintenance is necessary?) (250 words):

An overlying prerequisite of a weed biological control agent is that it is a relatively host specific organism. Eriophyid mites (Acari) have been considered as potential agents due to their intimate and specific interaction with their host plant, making many species host specific. Although a number of mite have or are currently being considered as potential agents; to date only three eriophyid mites have been intentionally introduced into the U.S. for weed control. One reason why more mites have not been introduced may lie with problems in their host testing. Unlike insects where individuals are able to actively disperse and select host plants, eriophyid mites differ in their behavior and dispersal capabilities making typical testing protocols problematic. As a result they appear to be less specific in no-choice tests. Eriophyids are microscopic and dispersal is usually passive through wind currents; although some may employ phoracy or are ambulatory as a means of dispersal. However very little is known about the dispersal nature of these mites. This study will consist of three objectives to determine: 1) the effects of environmental conditions, mite population and wind velocity on passive mite dispersal; 2) dispersal longevity of mites with differing feeding behaviors (rust mites vs gall inducing species); and 3) the effects of wind direction and vegetational structure on mite dispersal in the field. We hope that this knowledge may improve upon the host testing of these mites and how to strategize potential field releases.

Provide background, justification, and urgency (e.g., describe the management problem, impacts of the invasive plant, supportive research) (250 words):

Eriophyid mites are often host specific obligate plant feeders. Skoracka et al. (2010) indicated that about 80% of eriophyid mite species are recorded from a single host. This apparent host specificity makes them appealing as possible biocontrol agents of invasive weeds. Although many eriophyids have been identified as feeding on weeds, only three species have been intentionally introduced into the U.S.: Aceria chondrillae (rush skeletonweed), Aceria malherbae (field bindweed) and Floracarus perrepae (Old World climbing fern). A fourth mite Aceria drabae (hoarycress) has recently been approved for release (USDA-APHIS 2018). Currently eriophyids are being considered for the biocontrol of Russian knapweed, Russian thistle, medusahead, Russian olive, cheatgrass, among others (Smith et al. 2010, BBCA 2017).

Although eriophyids appear to be relatively host specific, their release has been hindered by inconsistencies and difficulties with host specificity tests. Eriophyid are thought to rely primarily on passive dispersal by wind, or limited phoretic and ambulatory dispersal. This passive or limited dispersal makes them more prone to false positive feeding in laboratory host specificity tests (i.e. no-choice) and is also problematic in conducting choice tests under field conditions (Smith et al. 2010). However, little is known about the dispersal capabilities of eriophyids and how it is related to the mite's behavior, population dynamics, host interactions (i.e. gall inducing vs vagrant species).

By investigating the aerobiology of these potential biocontrol agents we hope to design better host tests and determine release strategies to promote their natural dispersal in the environment.

Methods/Approach (750 words):

Test species and colony maintenance: Proposed test species will include two gall inducing mites – Aceria drabae (hoarycress) and Aceria chondrillae (rush skeletonweed) and two vagrant mites – Aceria sobhiani (Russian knapweed) and Aceria anthocoptes (Canada thistle). Mite colonies will be maintained on their respective plant (target) hosts either at the Montana State University Containment Facility or at the BBCA or BBCA affiliate laboratories. Mite populations will be periodically observed for specific behaviors indicating dispersal tendencies (see Kiedrowicz et al. 2017) which will be used to further refined methods stated below.

Objective 1. Effects of environmental conditions, mite population and wind velocity on passive mite dispersal.

Wind speed: Experiments to determine optimal wind speed to promote dispersal will be conducted in a wind tunnel constructed of acrylic tubing fitted with a fan with a controller to adjust wind speed. Constant wind speeds of 0, 8, and 24 kph (typical average wind speeds for Montana in June, Montana DEQ) will be maintained. A sample stage will be connected to the wind tunnel which will allow for infested plant material to be introduced. Mites will be observed using a stereoscope and a glass slide coated with silicon grease will be situated at the end of the tunnel to catch mites as they dispersed. Initial trials will be conducted for 5 minute durations and the number of mites (separated by life stage) leaving the tunnel will be recorded per interval.

Population: To determine if the level of mite populations play a role in dispersal a wind tunnel (above) will be utilized using two mite population levels; a low and high population level. Numbers are to be determined by previous observations. An optimal wind velocity (determined above) will be utilized. Experiments will be conducted as above.

Temporal dispersal: Mites may disperse temporally during the day (Sabelis and Bruin 1996, Bergh 2001). Plants infested with mites will be grown on a greenhouse bench under ambient conditions. A fan will be directed on to the plant at four temporal times during the day: nautical twilight (dawn and dusk), noon, and mid-night. Air flow of 3.5 m/s will be maintained for a duration of 10 minutes during which time mites will be collected and counted on a 96 cm2 acetate sheet covered with silicon grease set 20 cm from the plant.

Environmental conditions: The effects of temperature and relative humidity on mite dispersal will be tested with wind tunnels set up in environmental chambers maintained at constant temperatures of 20 and 30 C. A relative humidity of 40 and 60% will be maintained by passing air through a flask containing a glycerol solution approximating the required RH (Braun and Braun 1959). Experiments will be conducted as above using a 14:10 L:D photoperiod.

Objective 2) Dispersal longevity of mites with differing feeding behaviors (rust mites vs gall inducing species).

The longevity of mites will be determined at four temperatures (15, 20, 25 and 30 C) and three humidities (5, 20 and 60%). Twenty mites will be placed in a small, escape proof petri dish placed in a plastic chamber maintained at a specific %RH and held at a constant temperature with an environmental chamber. Mite deaths (morbidity) will be counted periodically until all individuals are dead. Trails will be replicated five times on two separate days.

Objective 3) Effects of wind direction and vegetational structure on mite dispersal in the field.

The first field release of Aceria drabae for the biocontrol of hoarycress is scheduled for the spring of 2019 in Montana. Two release sites will be selected to determine the aerial dispersal of the mite under field conditions. At each site, two 1 m2 plots will be inoculated with mites. One plot will have the surrounding vegetation mowed to augment the effects of the wind and the other plot, vegetation will remain unaltered. An anemometer will record wind speed and prevailing direction at 1 m above ground. Approximately three weeks after inoculations adhesive and water traps will be placed 5, 15, and 25 m from the plot with and against prevailing wind directions. Adhesive traps will be similar as those used in laboratory studies and water traps will consist of a pan filled with water and 0.5 ml of detergent to break water tension. Water will be periodically filtered from the pan and mites of both traps counted and sub-sampled for identification.

Statistics: A combination of non-parametric and ANOVA tests will be performed depending upon the experiment.

Describe stakeholder involvement in development of the proposal and application of the new technology (e.g., how involved are managers in the project?) (150 words):

Through various biological control consortia groups, stakeholder involvement often occurs in the pre-release screening and prioritization of potential agents. Once these agents are approved for release, state, federal, county and private cooperators will assist in initial field release efforts and eventual redistribution.

Describe technology assistance/transfer, outreach, and the expected impacts to forest health/forest management (e.g., how will the technology be shared and how will it change current management?) (150 words):

Expected impacts are twofold. First, since existing host specificity testing protocols may be problematic in testing these specialized biocontrol agents we hope to develop more predictive tests thus providing stakeholders with more informative data to prioritize agents or for gaining their approval for release into the environment. Currently a number of eriophyid mites are being considered and improved testing may speed up the overall screening process resulting in a more rapid assessment and eventual utilization by land managers. Secondly, since biocontrol agent redistribution is often time consuming, improved knowledge on the behavior and aerobiology of the mites will lead to better release strategies that will enhance the natural dispersal of these eriophyid mites to more distant weed infestations. This may reduce inputs by land managers (i.e. redistribution efforts) allowing resources to be used for other activities.

Products/Publications/Technology transfer (e.g., provide timeline of expected project accomplishments) (150 words):

Products: Products include publications of results in scientific journals and presentations at various regional and scientific meetings. Indirectly we hope to utilize the knowledge gain in these studies to direct applications in host specificity testing of current eriophyid mites (e.g. for Russian knapweed, medusahead), or for new agents.

Timetable:
Year 1 (2019): Collect and establish colonies of mites for experiments. Conduct preliminary observations on mite dispersal and to modify or refine research protocols. Construct and conduct preliminary tests of a wind tunnel. Determine the utility of water pan vs sticky cards for eriophyid monitoring. Initiate field releases of Aceria drabae.

Year 2 (2020): Continue to maintain mite colonies or to establish new colonies. Laboratory studies will continue in Montana and at the BBCA. Aceria drabae sites in Montana will be monitored for mite establishment and spread.

Year 3 (2021): Studies will be concluded and data compiled for a final report/publication.

Citations:

Bergh, JC. 2001. Ecology and aerobiology of dispersing citrus rust mite (Acari: Eriophyoidae) in Central Florida. Environ Entomol 30:318-326.

Bergh, JC, McCoy, CW. 1997. Aerial dispersal of citrus rust mite (Acari: Eriophyoidae) from Florida citrus groves. Environ Entomol 26:256–264.

Braun, JV and Braun, JD. 1959. A simplified method of preparing glycerol and water solutions for humidity control. Corrosion 14: 117-118.

Cromroy, HL. 1977. The potential use of eriophyoid mites for control of weeds. In: Freeman TE (ed) Proceedings of the IV international symposium on biological control of weeds, University of Florida, Gainesville, Florida, pp 294–296.

Cromroy, HL (1979) Eriophyoidea in biological control of weeds. In: Rodriguez JG (ed) Recent advances in acarology, vol 1. Academic Press, New York, pp 473–475.

Cromroy, HL (1983) Potential use of mites in biological control of terrestrial and aquatic weeds. In: Hoy MA, Cunningham GL, Knutson L (eds) Biological control of pests by mites. Univ Calif Agri Exp Stn Special Publ 3304:61–66.

Current Results - Weather and Science Facts: https://www.currentresults.com/Weather/Montana/average-montana-weather.php

Galvao, AS, Melo, JWS, Vaneska B, Monteiro, VB, Debora B. Lima, DB, De Moraes, GJ, Gondim Jr., MGC. 2012. Dispersal strategies of Aceria guerreronis (Acari: Eriophyidae), a coconut pest. Exp Appl Acarol 57:1–13. DOI 10.1007/s10493-012-9527-z

Kiedrowicz, A, Kuczyński, L., Lewandowski, M., Proctor, H., Skoracka, A. 2017 Behavioural responses to potential dispersal cues in two economically important species of cereal-feeding eriophyid mites. Scientific Reports 7: 3890. DOI:10.1038/s41598-017-04372-7

Melo, JWS, Lima DB, Sabelis MW, Pallini A, Gondim Jr. MGC. 2014.Limits to ambulatory displacement of coconut mitesin absence and presence of food-related cues. Exp Appl Acarol 62:449–461. DOI 10.1007/s10493-013-9753-z

Montana Department of Environmental Quality: https://deq.mt.gov/Portals/112/Energy/EnergizeMT/EnergySites/WindAtlas/Chapter3.PDF
Navia, D, Ochoa, R, Welbourn, C, Ferragut, F. 2010. Adventive eriophyoid mites: a global review of their impact, pathways, prevention and challenges. Exp Appl Acarol 51: 225-255.

Ochoa R, Erbe EF, Wergin WP, Frye C, Lydon J. 2001. The presence of Aceria anthocoptes (Nalepa) (Acari: Eriophyidae) on Cirsium species in the United States. Int J Acarol 27(3):179–187.

Rosenthal SS (1996) Aceria, Epitrimerus and Aculus species and biological control of weeds. In: Lindquist EE, Sabelis MW, Bruin J (eds) Eriophyoid mites–their biology, natural enemies and control, vol 6. Elsevier, Amsterdam, pp 729–739. World Crop Pests

Sabelis MW, Bruin J. 1996. Evolutionary ecology: life history patterns, food plant choice and dispersal. In: Lindquist EE, Sabelis MW, Bruin J (eds) Eriophyoid mites–their biology, natural enemies and control, vol 6. Elsevier, Amsterdam, pp 329–366. World Crop Pests

Smith, L, de Lillo, E, Amrine Jr. JW. 2010. Effectiveness of eriophyid mites for biological control of weedy plants and challenges for future research Exp Appl Acarol 51:115–149. DOI 10.1007/s10493-009-9299-2

Skoracka A, Oldfield G, Smith L, Cristofaro M, Ochoa R, Amrine JW Jr. 2010. Host plant specificity and specialization in eriophyoid mites. Exp Appl Acarol 51: 93-113.

USDA-APHIS. 2018. Field release of the gall mite, Aceria drabae (Acari: Eriophyidae), for classical biological control of hoarycress (Lepidium draba L., Lepidium chalapense L., and Lepidium appelianum Al-Shehbaz) (Brassicaceae), in the contiguous United States. Environmental Assessment, January 2018. 41 pp.

Year 1 Budget Information (for a summary of total project costs, see page 1)

Fiscal Year (FY): 2019

Budget Type	Budget	Requested BCIP Funding	Non-Federal Match**	Leveraged Funds***	Match Source	Leveraged Source
ADMINISTRATION	Salary	$ 16,080	$ 16,418		MSU	
	Travel for Data Collection		$ 1,300		MTNWTF	
	Travel to Meetings*					
	Travel for Other					
PROCUREMENT	Contracting	$ 15,000				
	Equipment					
	Supplies	$ 560				
INDIRECT	Overhead		$ 13,922		MSU	
	Other					
	SUBTOTAL	$ 31,640	$ 31,640	$ 0		

Overhead Rate % (Describe in Notes): 44

Year 1 Total: $ 63,280

Year 1 Notes (2000 characters):

Budget - Salary 3.5 mo. Technician ($4561/mo. Salary & 38.8% benefits) to maintain mite colonies and conduct research. Supplies – for wind speed & direction monitor. Space fees for greenhouse bench/ environmental chamber MSU Containment Lab. Contracted service agreement with BBCA to cover labor, travel & supplies.

Match Funds – Matching funds (in-kind) through MSU (Montana State University): 0.27 FTE for PI salary state funds and 44% Indirect Costs not recovered; plus MTNWTF (Montana Noxious Weed Trust Fund) match of technician's salary and benefits (~2 mo.) plus travel to field sites for mite release & collection, contracted service agreement for mite collection, and space fees for MSU Containment Facility for mite colony maintenance.

* Funds allocated for "Travel to Meetings" in year 1 will only be approved for proposals requesting a single year of funding.
** Non-Federal Matching (cost sharing) funds are raised from outside sources to increase the level of support provided by the Federal Government. This includes both cash and in-kind contributions.
*** Leveraged Funds are raised from outside sources to increase the level of support provided by the Federal Government (including cash and in-kind contributions) beyond the non-Federal matching requirements.

Year 2 Budget Information (for a summary of total project costs, see page 1)

Fiscal Year (FY): 2020

Budget Type	Budget	Requested BCIP Funding	Non-Federal Match	Leveraged Funds	Match Source	Leveraged Source
ADMINISTRATION	Salary	$ 16,080	$ 17,965		MSU	
	Travel for Data Collection					
	Travel to Meetings					
	Travel for Other					
PROCUREMENT	Contracting	$ 16,000				
	Equipment					
	Supplies					
INDIRECT	Overhead		$ 14,115		MSU	
	Other					
	SUBTOTAL	$ 32,080	$ 32,080	$ 0		

Overhead Rate %: 44

Year 2 Total: $ 64,160

Year 2 Notes (2000 characters):

Budget - Salary 4 mo. Technician ($4561/mo. Salary + 38.8% benefits) to maintain mite colonies and conduct research. Contracted service agreement with BBCA to cover labor, travel & supplies.

Match & Leverage funds – Matching funds (in-kind) through MSU (Montana State University): 44% Indirect Costs not recovered; plus 0.3 FTE for PI salary (state funds).

Year 3 Budget Information (for a summary of total project costs, see page 1)

Fiscal Year (FY): 2021

Budget Type	Budget	Requested BCIP Funding	Non-Federal Match	Leveraged Funds	Match Source	Leveraged Source
ADMINISTRATION	Salary	$ 9,166	$ 10,453		MSU	
	Travel for Data Collection					
	Travel to Meetings					
	Travel for Other					
PROCUREMENT	Contracting	$ 9,500				
	Equipment					
	Supplies					
INDIRECT	Overhead		$ 8,213		MSU	
	Other					
	SUBTOTAL	$ 18,666	$ 18,666	$ 0		

Overhead Rate %: 44

Year 3 Total: $ 37,332

Year 3 Notes (2000 characters):

Budget - Salary 2 mo. Technician ($4561/mo. Salary + 38.8% benefits) to maintain mite colonies and conduct research. Contracted service agreement with BBCA to cover labor, travel & supplies.

Match & Leverage funds – Matching funds (in-kind) through MSU (Montana State University): 0.2 FTE for PI salary (state funds), 44% Indirect Costs not recovered.

Project Contacts (The Funding Coordinator is the individual who manages project funds. A single funding coordinator must be selected.)

FHP/BCIP Regional/Station/Area Contact

Name: Carol Randall Title: Entomologist

Institution: USDA Forest Service Phone: 208-769-3051

Email: crandall@fs.fed.us

Technical Monitor: BCIP contact

Principal Investigators

Name: Jeffrey Littlefield Title: Research Scientist

Institution: Montana State University Phone: 406-994-4722

Email: JeffreyL@Montana.edu Time commitment: 02 FTE

Project Role: Project coordinator - lead researcher Montana Funding Coordinator: ✓

Name: Massimo Christofaro Title: Research Leader

Institution: Biotechnology and Biological Control Agency (BBCA) Phone: +39 0691132995

Email: m.cristofaro55@gmail.com Time commitment: 0.1 FTE

Project Role: Research coordinator overseas. Funding Coordinator: ☐

Name: Francesca Marini Title:

Institution: Biotechnology and Biological Control Agency (BBCA) Phone: +39 0691132995

Email: fra.rini@gmail.com Time commitment: 0.3 FTE

Project Role: Conducting research on overseas species. Funding Coordinator: ☐

Cooperators

Name: Title:

Institution: Phone:

Email: Time commitment:

Project Role: Funding Coordinator: ☐

Name: Title:

Institution: Phone:

Email: Time commitment:

Project Role: Funding Coordinator: ☐

Name: Title:

Institution: Phone:

Email: Time commitment:

Project Role: Funding Coordinator: ☐

List additional documents being sent in support of the project. (e.g., curriculum vitae, letters from stakeholders, spreadsheets, etc.) (not more than one page):

Jeffrey L. Littlefield
Department of Land Resources & Environmental Sciences
Montana State University
Bozeman, MT 59717
Telephone: (406) 994-4722
E-mail: JeffreyL@Montana.edu

EDUCATION
Ph.D. 1986. Entomology. University of Wyoming, Laramie, WY
M.S. 1980. Entomology. University of Idaho, Moscow, ID
B.S.F. 1975. Forestry. University of New Hampshire, Durham, NH
B.S. 1975. Entomology. University of New Hampshire, Durham, NH

ACADEMIC APPOINTMENT
Research Scientist/Quarantine Director

MAJOR ADMINISTRATIVE RESPONSIBILITIES
My position has both a service and a research component. As Quarantine Director I am responsible for the maintenance and daily operation of the Biological Containment Facility for the importation of weed feeding organisms (arthropods and pathogens); maintaining permit, importation and release records and voucher specimens of imported organisms; "pass-through screening" of organisms (including screening for pathogens, parasites and confirming species identity); insect rearing, developing standard operating procedures and training personnel. In addition, I maintain the budget for the facility and seek grant support for its operation. In this position I develop risk assessment documents, both BAs and EAs, for field release of exotic organisms for biological control. I work closely with state and regulatory personnel for maintaining the certification of the containment lab and for the importation of various biological agents.

CURRENT RESEARCH
For research my primary emphasis is biological control of weeds, including the determination of host specificity, bionomics, and field release of potential biocontrol agents. In the past I have worked on insects attacking a variety of other weed species; as well as, parasitoids of the Russian wheat aphid, alfalfa weevil and the wheat stem sawfly. Currently I am investigating the use of gall mites (Eriophyidae) and other arthropods as potential biocontrol agents of field bindweed, Russian knapweed, invasive hawkweeds, and whitetop. I also have cooperative projects involving the biological control of common tansy, ox-eye daisy various forest weeds including tansy ragwort and rush skeletonweed. Through these projects I work with several co-investigators and cooperators from various state, federal and international agencies or organizations.

GRANT SUPPORT
Since 2013 I have administered or co-authored 35 grants funded and received approximately $1.27 million in grant supported funding from a variety of sources, both federal and state, for the study, rearing and release of biological control agents for invasive hawkweeds; including hoary cress, tansy ragwort, field bindweed, Russian knapweed, rush skeletonweed, common tansy and ox-eye daisy. Funding supported overseas survey and screening of new biological control agents, and for domestic host testing, rearing, release and monitoring of these and other agents, and the study of other invasive species.

PUBLICATIONS, REPORTS & PRESENTATIONS
In the past four years I have authored or co-authored 7 reviewed or non-reviewed publications, technical reports (including TAG petitions) and abstracts.

Keywords (50 words):

Eriophyidae, dispersal, biological control, invasive weeds, host specificity testing