Target Invasive Plants and Biocontrol Agents Undergoing Screening

Ministry of Forests
Range Branch
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The BC Ministry of Forests (MFOR), through contributed funding, enables research by the Centre for Agriculture and Biosciences International (CABI) (https://www.cabi.org/what-we-do/invasive-species/biocontrol/) and Agriculture and Agri-Food Canada (AAFC)'s Weed Biological Control Program (https://profils-profiles.science.gc.ca/en/research-centre/lethbridge-research-and-development-centre) into potential new biocontrol agents for the province on an annual basis. Research on these agents is also funded by a consortium of interested partners, including other Canadian provincial and federal government departments, USA county, state and federal agencies, USA and Canadian Universities, and NGOs. Specific funding and research partners vary with each plant targeted.

The invasive plants currently targeted by British Columbia, as of **June 2022**, and their consortia objectives, are:

- Common reed (*Phragmites australis*)
- Common tansy (*Tanacetum vulgare*)
- Dalmatian and yellow toadflax (*Linaria dalmatica* and *L. vulgaris*)
- Field bindweed (*Convolvulus arvensis*)
- Flowering rush (*Butomus umbellatus*)
- Hawkweed complex (*Pilosella* spp., syn. *Hieracium* spp.)
- Himalayan balsam aka: Policeman's helmet (*Impatiens glandulifera*)
- Hoary cresses (*Lepidium draba, L. chalepense* and *L. appelianum*)
- Knotweed (*Fallopia/Polygonum spp.*)
- Oxeye daisy (*Leucanthemum vulgare*)
- Parrot's feather (*Myriophyllum aquaticum*)
- Russian olive (*Elaeagnus angustifolia*)
- Tree of heaven (*Ailanthus altissima*)

Introduced common reed (*Phragmites australis australis*)

Two biological control agents exist in Canada for the invasive genotype of this perennial wetland grass. Investigation into potential biological control for invasive common reed in North America began in 1998. Participation in the project was initiated by agencies from the USA and eastern Canada. In 2013, BC joined the project to assist with completion of work on the two candidate biological control agents as the province has several populations of this invasive plant. Laboratory and field tests of host range in Europe confirmed that the two shoot-mining noctuid moths, *Lenisa* (*Archanara) geminipuncta* and *A. neurica* are specialists on invasive *Phragmites* (CABI pers. comm.).

Both moth species were approved by CFIA for release in Canada in April 2019. Rearing colonies of both moths are kept at CABI in Switzerland, at AAFC in Lethbridge, Alberta and at the University of Toronto. Following the permission for release, experiments were initiated to assess the best insect stage: moths vs larvae vs eggs in order to get successful establishment of the biocontrol agents. Both moth species have been released in eastern Canada starting in 2019. As of 2022, monitoring of release locations indicates successful overwintering of moth eggs and development of the larvae in the field. Monitoring is ongoing to confirm if the biocontrol agents persist and their populations become established (AAFC pers. comm. June 6, 2022).

Common tansy (*Tanacetum vulgare*)

No biological control agents exist in North America to date for this perennial herbaceous flowering plant. The chemical variability of common tansy populations in Europe and North America has been described, particularly with respect to the varying proportions of the species' toxic compounds; the essential oil components such as alpha-thujone. This variability may influence host plant acceptance by biocontrol agent candidates.

Recent tests focussed on three insects: the shoot and flower-head mining moth *Platyptilia ochrodactyla;* the stem-mining weevil *Microplontus millefollii*; and the leaf-feeding chrysomelid *Chrysolina eurina*. Studies on *P. ochrodactyla* in Switzerland have shown under natural field conditions the moth has a narrow host range, however, work has been suspended as it has proven difficult to synchronize the agent with the various test plant life cycles (CABI pers. comm., Sept. 1, 2021). Tests began at CABI in Switzerland for *M. millefollii*, however, the studies are now conducted in Russia, the weevil's place of origin, to better synchronize the collection of weevils to be used in testing with their natural oviposition period. To date the weevil is showing a narrow host range (CABI pers. comm., Sept. 1, 2021). A preliminary impact experiment showed reduced stem length, plant biomass and number of flower heads on the target common tansy. Plans are underway if the political situation in Russia allows to attempt establishment of a *M. millefollii* colony in quarantine (AAFC-Lethbridge) for study in preparation for its petitioning for release (AAFC pers. comm. May 24, 2022). Studies on *C. eurina* are continuing. Single-choice tests completed in 2020 revealed a limited attack on the native *Tanacetum camphoratum* and were followed up with open-field tests in 2021 which will be evaluated in 2022 (CABI pers. comm., Sept. 1, 2021).

Dalmatian and yellow toadflax (*Linaria dalmatica and L. vulgaris*)

There are five species of biological control agents that were purposely introduced into the province for Dalmatian and yellow toadflax, two of which attack both these perennial herbs with snap-dragon-like flowers. The agent *Mecinus janthinus*, introduced in 1991, has been genetically differentiated from *M. janthiniformis* which is morphologically very similar and was unknowingly mixed with *M. janthinus* during screening and first introductions into Canada (AAFC pers. comm. August 17, 2021). The former mainly feeds on yellow toadflax, the latter on Dalmatian, however, *M. janthinus* has not been confirmed to date to be established in the province.

The most recent agent, *Rhinusa pilosa*, was successfully released and established on its host yellow toadflax in BC in 2014. Studies on additional candidate agents continue in Serbia. Studies of the stem-boring weevils *Mecinus laeviceps* associated with *L. genistifolia/L. dalmatica, M. heydeni* targeting *L. vulgaris* and *M. peterharrisi* for control of *L. dalmatica* in northern regions are near completion, however, a key trial was interrupted, and rearing colonies were lost or heavily decreased by COVID-19 related restrictions, and requires repetition once weevil colonies are re-established (CABI pers. comm., Sept. 1, 2021). Studies of the *L. dalmatica* stem-galling weevil *R. rara* are completed and a petition for field release is being developed.

Field bindweed (*Convolvulus arvensis*)

Screening efforts to study and obtain biological control agents for field bindweed in Canada occurred several decades ago and resulted in one biological control agent approved for introduction into Canada from Europe (i.e., the leaf-galling mite, *Aceria malherbae*) to be released in the province in 1992. *A. malherbae* did not initially survive, however it has since established from populations obtained from Alberta in 2016. The leaf beetle *Deloyala guttata*, which is native to North America, was attempted for use by BC in field bindweed control in 1969. The beetle did not establish. In 2021, BC began contributing to the screening project that was resurrected by the USA starting in 2009. Out of six potential agents studied to date, four were removed due to lack of host specificity or inconclusive results while two remain in the project: *Melanagromyza albocilia* (root- and shoot-mining agromyzid fly) and *Microsphecia brosiformis* (root-mining clear-wing moth). Host specificity tests are ongoing for both species. Mainly open-field tests with *M. albocilia* are being conducted in southern Germany due to difficulties in synchronizing male and female populations of the fly. Studies on *M. brosiformis* are being conducted in laboratory

conditions in Serbia along with development of rearing methods as the moths have proven difficult to collect from their native sites (CABI pers. comm. May 24, 2022).

Flowering rush (*Butomus umbellatus*)

Currently no biological control agents exist in Canada for this perennial rush-like aquatic plant, the single genus/species within the Butomaceae family. Investigations into potential biological control of flowering rush began in 2013. To date there are only four documented infested water bodies in the province, however, surveys for the plant in BC lakes are ongoing. At this time, there are no additional infestations.

Currently manual treatments are used to control this extremely aggressive and damaging plant. The long-term effectiveness of this method is not yet known, and biological control is an additional potential management tool.

Based on studies of natural enemies in Europe, five potential biocontrol agents have been identified (AAFC pers. comm. August 4, 2021). This includes two weevil species, *Bagous nodulosus* and *B. validus*; two fly species, *Phytoliriomyza ornata* and *Hydrellia concolor*, all of which feed on the stems and leaves of flowering rush; and a fungus, *Doassansia niesslii*. Host range testing work by CABI has focussed on *B. nodulosus*, *P. ornata* and *D. niesslii*. Testing of adult *B. nodulosus* has demonstrated the weevil to be very host specific (CABI pers. comm., Sept. 2021). Research was completed in 2021 and a joint Canada/USA petition was submitted for approval within each country in April 2022. Results are pending. To date, host range testing of *P. ornata* has found the fly also to be very host specific to flowering rush (CABI pers. comm., Sept. 2021) and capable of causing a 50% reduction of above-ground biomass of flowering rush (CABI pers. comm., May 24, 2022). Studies of the fungal pathogen have also been initiated. Different fungal life stages affect both the submerged and emerged portions of the plant. Attack of the emerged plant parts has led to significant infection and plant death. Three strains of fungus have been tested on the multiple North American flowering rush genotypes and one fungal strain was effective against the unique BC genotype that occurs in Bouchie Lake, BC (CABI pers. comm., Sept. 2021). However, this strain did not attack the other most common genotypes found in Canada and the USA A new fungal strain was found in Romania in 2021 on the two most common genotypes of flowering rush It is currently being tested to determine if it will be effective against the more common invasive flowering rush genotypes in Canada (CABI pers. comm. May 24, 2022).

Hawkweed complex (*Pilosella* spp.; syn. *Hieracium* spp.)

To date, two biological control agents have been imported into BC for several species of these invasive perennial herbs. Studies of additional biocontrol agents for several hawkweed species are ongoing. Since 2000, seven biocontrol candidates have been considered for potential release in Canada, of which three were immediately dropped due to lack of specificity or effectiveness. The stolon-tip gall wasp *Aulacidea subterminalis* was the first agent to be approved by CFIA for release in Canada, and first released in BC in 2011. Although the wasp has only been reported on mouse-ear hawkweed under field conditions in Europe (*Pilosella officinarum*), lab testing showed it can potentially accept several invasive hawkweeds as hosts. It currently is being used by BC to target whiplash hawkweed (*Pilosella flagellaris*) (a stable hybrid of mouse-ear (*P. officinarum*) and meadow hawkweed (*P. caespitosa*)), kingdevil hawkweed (*P. floribunda*), and the less preferred host orange hawkweed (*P. aurantiaca*) (AAFC pers. comm. August 17, 2021). Its establishment on whiplash hawkweed was initially minimal, however in 2020 larger numbers of galls were found at two sites. In 2021, a significant number or galls were found at an additional site, allowing a collection and release to a new location. There has been no establishment confirmed on kingdevil or orange hawkweed.

The root-feeding hoverfly *Cheilosia urbana* was approved for import in spring of 2016 and first released in BC in 2017. A hoverfly matching the description of *C. urbana* was observed on hawkweed plants at the release site in spring 2018, but its identity was not confirmed. A small number of adult flies were subsequently reared from some additional 2017 infested plants that were overwintered at AAFC-Lethbridge, and these were shipped to BC for release at the same site in June 2018 and May 2020. Eggs of the fly shipped to AAFC-Lethbridge by CABI also were immediately hand transferred to field hawkweed plants at the same site in May 2018. No flies have been recovered from this site to date. The fly primarily targets meadow and orange hawkweeds.

Work on the rosette-feeding hoverfly *C. psilophthalma* overseas has been postponed; studies on the stem-galling wasp *A. hieracii* have been terminated due to it rarely being found in Europe on the hawkweed species being targeted for control in North America; and attempts to maintain a colony of the rust, *Puccinia hieracii* var. *piloselloidarum* for testing have proven difficult, thus work on the rust has ceased. Research by CABI Switzerland and AAFC-Lethbridge continues on two genetically and biologically divergent forms (biotypes) of another gall wasp *A. pilosellae*; one biotype is only found on mouse ear hawkweed in Europe, and the second occurs on several hawkweed species, including meadow, yellowdevil (*P. glomerata*), kingdevil, tall (*P. piloselloides*), infrequently on orange, but not on mouse ear under field conditions. AAFC-Lethbridge has been conducting studies on the impact of *A. pilosellae* on BC's invasive hawkweeds, which is part of the screening process for candidate biocontrol agents. In tests completed so far, both biotypes look to be very host specific on the invasive hawkweeds, versus on native North

American hawkweeds. Studies on both biotypes of *A. pilosellae* will continue at CABI and AAFC-Lethbridge in 2022.

Himalayan balsam (aka: Policeman's helmet) (*Impatiens glandulifera*)

No biological control agents are currently available in North America for this annual non-native plant often purposely grown as an ornamental. Investigation into potential biological control agents for Himalayan balsam began in the U.K. in 2006. BC joined the project in 2012 with the development of a North American test plant list. Screening work is being conducted by CABI in the United Kingdom. Canadian test plant species collected in BC have been provided to the U.K. lab for host-range testing of the rust *Puccinia komarovii* originally collected in the Indian region of the Himalayas (AAFC pers. comm. August 4, 2021). In 2014, the rust was permitted for release in the U.K. and released on field sites in England, the first ever fungal biological control agent in Europe. Field monitoring in the U.K. indicated that U.K. populations of Himalayan balsam were variably susceptible to the rust. Research is continuing to match rust strains with Himalayan balsam genotypes in both the U.K. and British Columbia. None of the Canadian non-target species tested by CABI were affected by the available rust strains. Molecular analysis of seven BC Himalayan balsam populations has revealed the two most common haplotypes are similar to the most common ones invading the U.K. Further investigations of the U.K. populations will assist understanding of effectiveness of the rust. In 2021, an additional rust strain was collected from Northern Pakistan that may be better matched to the Canadian Himalayan balsam populations. If this strain infects the Canadian plants, host-specificity screening will commence (CABI pers. comm., May 24, 2022).

Hoary cresses (*Lepidium draba, L. chalepense and L. appelianum*)

No biological control agents exist in Canada to date for the three species of this perennial clonal mustard. Work on hoary cresses (or whitetops) was initiated by the USA in 2001 and BC joined the project in 2011.

Seven phytophagous insect species were chosen for study, four of which have undergone initial screening and have been removed because of a lack of specificity (CABI pers. comm.). Following completion of the research in 2012, the mite *Aceria drabae* was released in 2019 in the U.S.A on its host *Lepidium draba* (CABI pers. comm. May 24, 2022). Depending on the impact results in the USA, this agent may have potential for Canada (AAFC pers. comm. June 28, 2022) The subsequent focus narrowed to two species; the gall-forming weevil *Ceutorhynchus cardariae* and the seed-feeding weevil *C. turbatus*.

The USA and CABI submitted a petition to the USDA, APHIS TAG in 2011 to request permission to release *C. cardariae* in the USA. APHIS responded with a request for testing of additional non-target plant species and for additional clarification of existing data. Following the submission of a supplemental release petition in the USA in January 2020, TAG responded that at this point in time, they do not recommend the release of *C. cardariae* in the USA. This insect has not been petitioned for release in Canada (AAFC pers. comm. August 4, 2021). With this result the focus has shifted to ongoing host range testing studies at CABI Switzerland on *C. turbatus.* Tests to date show *C. turbatus* has a

narrow host range (CABI pers. comm., May 24, 2022).

Knotweed (*Fallopia/Polygonum*)

To date, a single biological control agent has been imported into Canada targeting three knotweed species in British Columbia: Japanese knotweed (*Fallopia japonica* or *Polygonum cuspidatum*); Giant knotweed (*F. sachaliensis* or *P. sachalinense*) and the hybrid between these two species, Bohemian knotweed (*F. bohemica*, hybrid). This invasive plant mainly infests Vancouver Island, the Lower Mainland, the Central Kootenays and is found sporadically elsewhere in the province. Based on climate modelling by AAFC, the knotweeds will likely spread further throughout BC. The sap-sucking psyllid, *Aphalara itadori* was permitted for release in 2014 and research for establishment and field impact studies by AAFC in BC, Alberta and Ontario and by CABI in the U.K. are ongoing. The selection of release sites in multiple provinces was to cover large variations in climatic conditions and to assess overwintering capabilities of the psyllid. Adult psyllids have successfully overwintered in all three provinces and there has been completion of at least one generation in the field for multiple years following releases. However, to date, no sustained psyllid populations at any single location across all release years have been confirmed. Collections of additional psyllid lines were conducted in Japan in 2019 to restore genetic diversity that may have been lost in long- term laboratory rearing (since 2004) of initial psyllid lines. These new lines show similar host specificity to the lines that had been in long-term laboratory culture and higher impact on knotweed leaves. Releases of the new lines started in 2020 in BC, Ontario and Alberta and monitoring of populations for establishment is continuing (AAFC pers. comm. August 4, 2021). Studies are continuing on these new lines to understand factors that contribute to leaf-curling damage on knotweeds and methods to improve initial survival of psyllids in the field (CABI pers. comm. May 24, 2022).

Additional screening studies of a knotweed pathogen as a potential biocontrol agent have been conducted by CABI in the U.K. Research has been focussed on two strains of the leaf spot fungus *Mycosphaerella polygoni-cuspidati* and shown the fungus affects all three knotweed species.

In host range testing of non-target species, however, the pathogen was found to infect important related U.K. and North American test plants. Thus, screening of these pathogen strains as classical biocontrol agents was stopped.

Oxeye daisy (*Leucanthemum vulgare*)

No biological control agents exist in North America to date for this rhizomatous perennial herb. In 2008, BC initiated funding on oxeye daisy with CABI in Switzerland and a test plant list was drafted. Studies have also been conducted into oxeye daisy's phylogeny and its potential relationship to other desirable daisies. Neither native North American species nor ornamental daisies are found within the same subtribe *Leucantheminae* as oxeye daisy with the exception of Shasta daisy. Ploidy analysis has found a diploid and a tetraploid species of oxeye daisy in North America. The tetraploid *Leucanthemum ircutianum* is more common in Europe while the diploid *L. vulgare* is more common in North America. Both are found in BC. The potential biological control agents investigated to date can be found on both species of oxeye daisy in Europe. Initial literature and field surveys prioritized eight potential biocontrol agents, but four were discontinued due to lack of host specificity or impact (CABI pers. comm.). For example, host-specificity tests with the root-mining weevil *Cyphocleonus trisulcatus* showed that the weevil would attack ornamental Shasta daisy varieties to a similar degree as *L. vulgare* and the weevil was subsequently not further considered as an agent for North America (CABI pers. comm. May 24, 2022). Work has continued at CABI in Switzerland on: the root-mining tortricid moth *Dichrorampha aeratana*; the root-galling tephritid fly *Oxyna nebulosa*; and the shoot-mining moth *D. consortana*. Tests on *D. aeratana* are complete and the moth appears to be host specific and effective as shown in an impact study that resulted in a significant decrease of flowers and below ground biomass of oxeye daisy. A petition for release of the moth was submitted November 2021 for review by Canadian and USA regulatory authorities. A shipment of the moths was sent to the AAFC quarantine facility in Lethbridge for use in a host-specificity test involving a native Arctic daisy, and for rearing in anticipation of approval to release (AAFC pers. comm. August 17, 2021). No-choice tests and efficacy experiments continue on *O. nebulosa*. This fly was found to decrease flower heads and reduce height and above-ground biomass of attacked oxeye daisy plants. Similar to *D. aeratana*, a population of *O. nebulosa* was shipped to the AAFC Lethbridge quarantine by CABI Switzerland in 2019 for testing with the native Arctic daisy and to initiate a rearing colony for biological studies. Studies to date on *D. consortana* have resulted in low larval survival and attack rates. The current focus is to further study the biology of this moth and improve rearing techniques. An additional agent, the root-mining moth *D. baixerasana*, may also be studied in the future if a collection site can be located (CABI pers. comm. May 24, 2022).

Parrot's feather (*Myriophyllum aquaticum*)

Native to South America, parrot's feather has been imported to many countries, likely as a desirable aquatic plant. It has since escaped controlled settings into natural water bodies where it creates dense mats, obstructs water flow, and degrades water quality and habitat for fish and wildlife. South Africa completed biological control screening research into a leaf-feeding beetle (*Lysathia* sp.) and a stem-mining weevil (*Listronotus marginicollis*) and released the *Lysathia* beetle. No further agents were needed as the beetle sufficiently controlled parrot's feather in South Africa. BC began funding research by CABI into potential biocontrol agents for this plant species in spring 2020. The initial focus is on host specificity testing of the beetle and weevil agents against other *Myriophyllum* species, of which there are 13 in Canada (including *M. aquaticum* and the invasive *M. spicatum*), and other plants of concern to Canada (AAFC pers. comm. August 10, 2021). In September 2021, initial host specificity testing began with the *Lysathia* beetle that to date has shown a preference for the Canadian parrot's feather. A potential third agent, *Phytobius vestitus*, a weevil native to North America, has been identified and documented to cause significant damage to some parrot's feather populations in Louisiana, USA. Studies will continue on *Lysathia* sp. and if possible, *L. marginicollis* and/or *P. vestitus* will be obtained and studies will begin (CABI pers. comm. May 24, 2022). With input from AAFC, potential parrot's feather biocontrol will be informed by climate matching between agent source populations in South Africa and Argentina and that of infested locations on Vancouver Island and in the Lower Mainland (AAFC pers. comm. August 10, 2021).

Russian olive (*Elaeagnus angustifolia*)

No biological control agents exist in North America to date for this flowering shrubby tree. The Russian olive biological control screening project was initiated in 2007 and has been funded by USA agencies. BC began contributing to the project in 2014. This tree has been purposefully grown and sold in North America for a variety of purposes including as an ornamental tree, a source of nectar for honeybees and a prairie windbreak, but it has since spread aggressively, causing particular problems in riparian habitats (AAFC pers. comm. August 17, 2021). Therefore, the focus has been on finding biological control agents that attack the tree's reproductive capacity to not disrupt use of existing trees.

Three potential agents have been investigated by CABI in Switzerland: the shoot-galling mite *Aceria angustifoliae*; the fruit-attacking moth *Anarsia eleagnella*; and the shoot-galling mite *Aceria eleagnicola*. Host specificity tests have been completed for *Aceria angustifoliae*

and a petition for import and release was submitted in the USA and Canada in November 2019 with submission of additional supporting information in 2021. Approval for release in Canada was given in April 2022, and first field releases of the mite are anticipated in 2023 after a generation is reared in quarantine at AAFC Lethbridge for the purpose of colony purification (AAFC pers. comm. April 13, 2022).

Initial studies on effectiveness of *Anarsia eleagnella* looked promising with a significant reduction in seed production. However, initial host specificity tests may indicate a broader host range than required. A lack of survival of some test plant species and difficulty in obtaining an export permit from Iran for further moths has caused a suspension of this work. The impact of *Aceria eleagnicola* on Russian olive is considered mild and ineffective. Thus, no further work is planned for this mite. Surveys for additional potential agents are on-going (CABI pers. comm.).

Tree of heaven (*Ailanthus altissima*)

Tree of heaven, a deciduous tree species originating from China, was imported to several continents as an ornamental and has since become invasive as it spreads vegetatively and produces a significant number of seeds. Tree of heaven is also a primary host for two devastating invasive generalist insect species, the spotted lanternfly (*Lycorma delicatula*), and the brown marmorated stink bug (*Halyomorpha halys*). Both these insect species impact a diversity of crops including grapes and tree fruits, numerous forest tree species, and a range of native plant species. While spotted lanternfly is not yet present in Canada, the occurrence of tree of heaven has been identified as at high risk of facilitating the spread of this invasive insect into Canada and across North America (AAFC pers. comm. August 30, 2021).

No biological control agents exist in Canada to date for tree of heaven. Investigations into potential biological control agents began in 2004 between China and the USA and permission is currently being sought to release the trunk-boring weevil, *Eucryptorrhynchus brandti*, in the USA (AAFC pers. comm. May 26, 2022; Technical Advisory Group 2022). A parallel investigation into potential biological control agents is being conducted in Europe through CABI in collaboration with the Biotechnology and Biological Control Agency (BBCA) in Italy (CABI pers. comm. May 24, 2022). In spring of 2020, BC began to contribute funding to this project with a focus on a test plant list for closely related species in Canada and initiation of host specificity tests on the eriophyid mite *Aculus mosoniensis* by BBCA in Italy. *Aculus mosoniensis* affects young shoots of tree of haven through its feeding, causing malformations which can even lead to death of young trees (AAFC pers. comm. August 30, 2021). In 2022, studies on the host specificity of the mite will continue and a study on its temperature dependent development will be completed (CABI pers. comm. May 24, 2022).

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