

Site selection for the release of the rust fungus *Puccinia komarovii* var. *glanduliferae* as a biological control agent for *Impatiens glandulifera* in the UK

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Introduction and background

Himalayan balsam (*Impatiens glandulifera*, Ericales: Balsaminaceae), is an annual weed introduced into the UK, mainland Europe and North America from its native range, the Western Himalayas (India and Pakistan). Himalayan balsam was introduced into the UK in 1839 and is now the most commonly occurring non-native plant species on riparian systems, occupying over 13% of rivers in England and Wales. This species has an adverse impact on native biodiversity, ecosystem services, and in Canada, recent research has highlighted the negative impact of the weed on pollination of native plants. A study undertaken in Switzerland also demonstrated that riverbanks, laid bare by the annual weed after it dies back in the winter, renders them more prone to erosion. The negative environmental impact of this species, together with the high financial costs of control using traditional chemical and mechanical methods, resulted in the UK Government asking CABI to investigate the potential of classical biological control (CBC) of Himalayan balsam in the UK.

After extensive surveys for natural enemies attacking Himalayan in its native range, the rust fungus *Puccinia komarovii* var. *glanduliferae* (collected in the Kullu Valley in India) was selected as the prime candidate for the CBC of Himalayan balsam in the UK. A comprehensive, 4-year research programme was undertaken to assess the safety and suitability of the pathogen. The rust was tested against 74 plant species to ensure its specificity to Himalayan balsam. A Pest Risk Analysis (PRA) which fully detailed the research conducted on the host range, life cycle and ecology of the rust was submitted for official assessment by the Food and Environment Research Agency (FERA) (a Defra Agency) in 2014. The PRA also underwent public consultation, and further evaluation by the European Commission's Standing Committee on Plant Health. Following their feedback, Defra Ministers approved the release of the rust into the wild in England on the 27th July 2014.

Rust release programme

Since its approval for release in 2014, the rust has been released at more than 100 sites in England, Scotland and Wales. During this time release techniques, habitat type and environmental conditions have been studied and conclusions drawn concerning the optimum conditions required for the rust to establish and proliferate. The aim of the release programme is to achieve establishment of Himalayan balsam rust in selected populations of the weed in the UK and reach a level of infection that triggers a rust epiphytotic. Once this has been achieved, the rust spores will naturally spread within the weed population and to new populations potentially many kilometres away.

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The rust will cause significant damage on the current year's population of the weed, reducing plant seed production and potentially weed density in the following years. Equally important is that high level of spore production in a weed population will result in high levels of overwintering spore development late in the season (teliospores). These spores remain dormant in the soil leaf litter, germinating in the spring to infect seedlings and enable the rust to complete its lifecycle, and cause a significant reduction in seedling survival.

Variation in plant susceptibility to the rust

The results of the field releases in 2015 revealed that there was far more genetic diversity in the Himalayan balsam population in the UK than realised. This suggests that there have been multiple introductions of Himalayan balsam in to the UK from different parts of its native range. Not all populations of the plant were found to be susceptible to the strain of the rust that was released from India. The rust is highly selective, and tends to infect genetic types of the plant that originated from the area in the native range where the rust evolved. Fortunately, CABI has access to a number of rust isolates from different areas in the native range. A molecular study by CABI has identified at least three biotypes of the weed in the UK and identified where these biotypes originated. A second isolate of the rust from Pakistan was screened and released in 2017. This isolate infects many of the biotypes that the Indian isolate does not infect. Additional isolates which affect those biotypes which the Indian and Pakistani isolates do not, are currently being sought from the native range.

Because of the biotypic variation in the weed it is critical to check which rust isolate is most suited to release in a particular area, currently CABI is testing two isolates of the rust in the field (the original isolate from India and an additional isolate from Pakistan).

Seed collection

Prior to the release of the rust at a site, it is necessary for CABI to determine the susceptibility of the plants to establish which of the two rust strains is the most pathogenic. Seeds from each potential rust release site need to be sent to CABI, the preceding autumn; at least 250 seeds per site from multiple plants. Seeds need to be collected when mature (i.e. black). Collect the seed by carefully placing a plastic zip lock bag over a group of flower heads that have matured pods and then shaking the plant to encourage the pods to explode. The black seeds can be picked from the leaf material and allowed to dry.

Following the removal of the plant material and any white, immature seed, place the seeds on a tray and allow to air dry for approximately one week. The seeds will turn to grey in colour. Put seeds in a paper envelope per site, and label with site details, including grid reference or GPS location, collection date and collector's name. The best time to collect seed is during August and September. Please send the seeds as soon as you can and between rigid packaging (e.g. cardboard) to prevent crushing during transit.

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Site selection

Due to the biotypic variation in the weed, it is essential that the seed is collected, in the autumn, from the exact site where the rust is to be released the following spring. The following site selection criteria should be considered:

- sites that are stable i.e. unlikely to be washed away in the winter;
- have a high natural humidity i.e. near a watercourse, ditch, pond or in boggy ground;
- not liable to be submerged by water for extended periods during the winter;
- have a minimum of ~10m x 10m of dense, healthy, actively growing populations of Himalayan balsam;
- open sites or with low or no shade from trees/shrubs.

In addition, please preferentially select sites that:

- are away from easy public access;
- are safely accessible with nearby parking facilities;
- have agreement of land owner to allow access and rust release (or can be readily attained) for the duration of the trial;
- the landowner is happy to leave a small area unmanaged for the duration of the trial, with no liability to project partners resulting from non-management.

The rust will be inoculated (spraying spores suspended in water) onto all the plants in a sub-population of approximately 5m x 5m, 3 times over the growing season, in June, July and late August. The inoculated population will need to be marked with canes or ribbons so that the plants can be monitored for infection over the season. A data logger will be hidden at each site to record temperature and humidity. Normal Himalayan balsam management techniques should be continued outside of the 10m x 10m experimental zone.

On-going work can be found on the following web site, which is regularly updated:

<https://www.invasive-species.org/species/himalayan-balsam/>

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