

Searching for fungal mycoherbicides effective against climbing asparagus (*Asparagus scandens*) in New Zealand

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Background

Climbing asparagus (*Asparagus scandens*) is a widespread invasive species in New Zealand and endemic to South Africa (Timmins & Reid, 2000) (Figure 1). It is harmful to biodiversity due to its coverage of the forest floor. *A. scandens* is distributed by birds and other animals.

Current chemical herbicides such as glyphosate can be toxic to non-target plants and cause disturbance to nearby native plant species. *A. scandens* may develop resistance to glyphosate in the future. New control approaches against *A. scandens* are essential.

Fungal bioherbicides can control weeds and minimize toxicity to native plants (Kakhaki et al. 2017). Some fungal phytopathogens can cause severe symptoms on *A. scandens*. *Colletotrichum* spp. and *Fusarium* spp. are two of many fungal species isolated from *A. scandens* in New Zealand.

It is hoped that some fungal phytopathogens may cause severe symptoms on *A. scandens*, but a systematic survey has not been done previously (Waipara et al. 2009), and only *Colletotrichum* species have been isolated from *A. scandens* in New Zealand before our study.



Figure 1: *A. scandens* cultured at Unitec lab (left) and collected from an Auckland sampling site (right).

Project aims

1. Survey for the presence of fungal pathogens of *A. scandens* from selected New Zealand sites.
2. Demonstrate the pathogenicity of isolated fungal pathogens against *A. scandens*.
3. Establish baselines for future fungal mycoherbicide application *in planta*.

Methods

1. Sixteen sites have been visited across Auckland and other regions (Figure 2). *A. scandens* samples were collected if symptoms were identified on leaves or stems.
2. Fungal strains were isolated from symptomatic *A. scandens* tissues and cultured on Potato Dextrose Agar (PDA) plates at 18°C for at least 7 days.
3. DNA was collected from fungal isolations and amplified by PCR for sequencing, sequences were compared with GeneBank of U.S. National Library of Medicine for identification.
4. Pure fungal isolates of interest were inoculated into *A. scandens* leaves (Figure 3) with 1 mL water and cultured at 18°C with 12 h light daily. Another 1 mL water was added at 3dpi. Plant symptoms were observed and recorded at 7dpi.

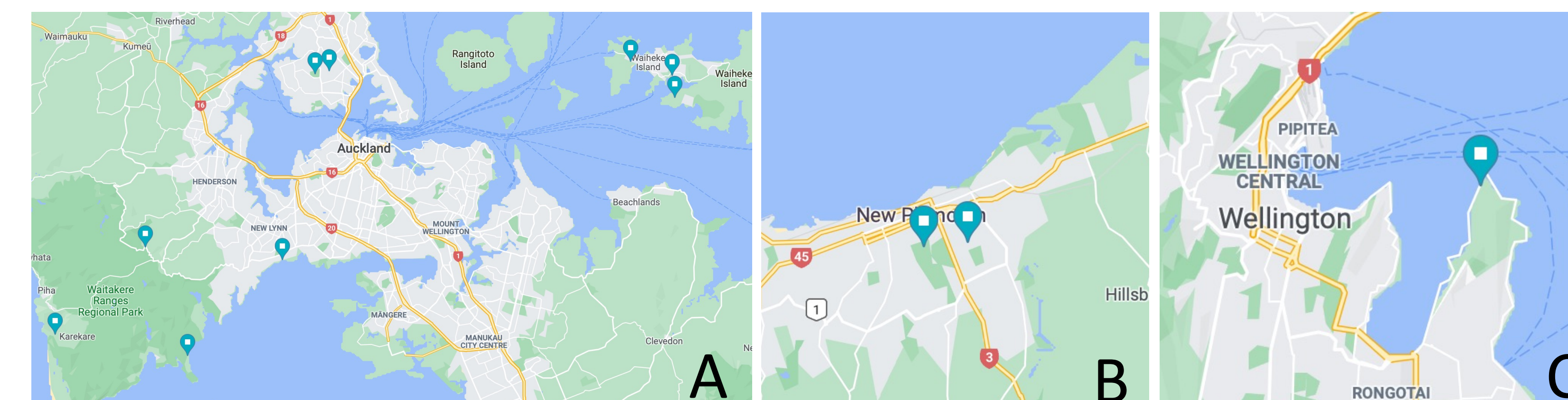


Figure 2. Indications of site locations (until November 2021). Each point may reference to one or multiple sub-locations. A: Collection sites at Auckland; B: Collection sites at Taranaki; C: Collection site at Wellington

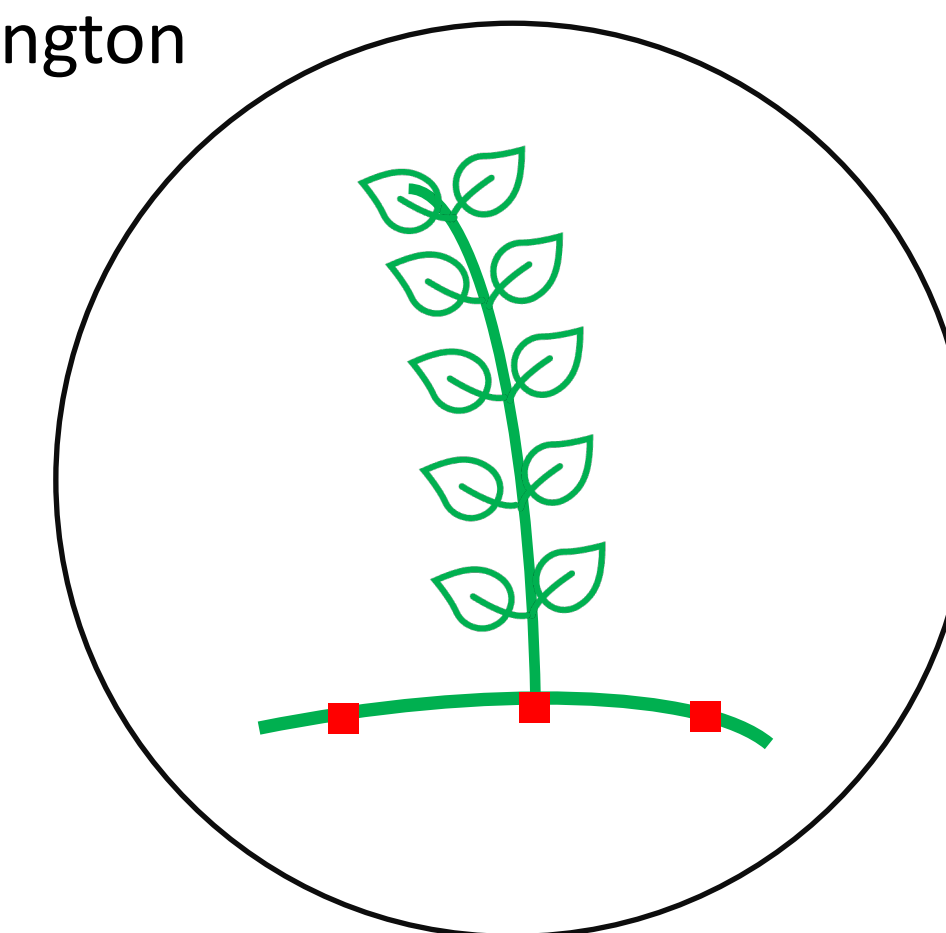


Figure 3. *In vitro* inoculation of pure fungal strains into *A. scandens* leaves. Each group contains needle-wounded and unwounded plants. Red squares indicate locations of culture plugs placed.



Figure 4. *In vitro* inoculation test sample in 7dpi. Plant sample has discolouration on leaves, dieback on tips and visible hyphae identified on top end of the sample.

Results

1. As of November, the most identified fungal species are: *Colletotrichum* spp.; *Fusarium* spp.; *Pestalotiopsis* spp. and *Penicillium* spp.
2. Most fungal species resulted in symptomatic samples (Figure 4). Discolouration on leaves and stem, visible hyphae on sample, leaf or/and stem dieback). *Fusarium* spp. and *Colletotrichum* spp. showed obvious damage *in vitro*.
3. Water deficiency (total water 1 mL vs 2 mL) and relatively high temperature (20°C vs 18°C) could accelerate symptom development.

Discussion

This study uncovered the potential application of local isolated fungal species as future mycoherbicides against invasive climbing asparagus. Following steps include collection of more fungal isolates, inoculation of pathogenetic isolates into whole *A. scandens* plants in combinations, and design application methods in fields.

This study could reduce the dependence on chemical sprays and avoid introducing novel micro-organisms into New Zealand. It is an early step of a bigger conservation programme with more studies in the near future. We would like to perform fungal tests on real plants soon and start to consider biocontrol applications in the field at a larger scale than in the Unitec laboratory.

References

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Acknowledgement

We are very grateful to the Auckland Council for funding this research programme.