

Biodiversity Congress 2018: Metabarcoding and culturing to investigate the diverse fungi associated with cabbage in and their bio-control potential

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Abstract

Statement of the Problem: Agricultural crops like cabbage (*Brassica oleracea* var. *capitata*) are vulnerable to a plethora of pests and diseases. Endophytic fungi are increasingly used in biological control against these pests and diseases as agricultural producers plan to reduce their environmental footprint.

Methodology & Theoretical Orientation:

During this study, putative endophytic fungi were isolated from cabbage grown in three regions in New Zealand so as to spot isolates with potential to help crop production. Isolation method was optimised in an exploratory study, and subsequently large scale sampling took place, where a culture-independent method was also used to detect fungi associated with cabbage. Spatial patterns in fungal communities in various plant tissues and geographical regions were investigated based on data obtained by culture-dependent and culture-independent methods. Selected fungal isolates were tested in bioassays to work out their potential for biological control of a fungal disease caused by *Sclerotinia sclerotiorum* and an insect pest of cabbage

- Diamondback moth (*Plutella xylostella*). In addition to isolation from plants, patterns in fungal communities were also described using metabarcoding data acquired by high-throughput sequencing and compared with data on direct isolation through culturing.

Findings: Over 100 species were identified using culturing approaches, while metabarcoding approaches suggest the amount of fungal species related to cabbage tissues might be over 200. Fungal communities differ in above and belowground parts of cabbage. There does not seem to be a significant difference between fungal communities in different regions of New Zealand. Fewer species were recovered in both years than would be expected, nevertheless, these taxa stand out as ubiquitous and abundant; fungi such as *Alternaria*, *Boeremia*, *Cladosporium*, *Fusarium*, *Monographella*, *Plectosphaerella*, *Pyrenochaeta* and *Stemphylium* were isolated in most of the sampled plants and usually in high frequencies. However, most of the species isolated in this study were rare. Such distribution of fungi is similar to that in a study describing mycorrhizal and phyllosphere fungal communities [139], or

to the one describing potential of endophytic fungi isolated from oilseed rape [153]. It is not possible to directly compare 2015 and 2016 studies, given differences in plant ages, isolation approaches, and sampled fields. Nonetheless, the relatively large number of species found in only one year or the other is consistent with a very high total diversity of fungi in these systems. Increased mortality of Diamondback moth larvae feeding on cabbage leaves treated with spore suspensions of several isolates of *Lecanicillium* sp. was observed. None of the tested isolates used as seed inoculants significantly reduced disease symptoms caused by *Sclerotinia sclerotiorum* during a bioassay on cabbage seedlings.

Conclusion & Significance: During this study several isolates were identified as promising biological control agents of an insect pest of cabbage. However, these isolates are members of a large consortium of fungi. The uncovered patterns within the fungal communities underline the importance of understanding of the microbial communities related to agricultural crop for us to enhance it. Endophytic fungi isolated from internal tissues of crop plants such as cabbage, have potential for improving crop production through biological control of pests. Diverse fungal communities can be detected, not only by isolation, but also culture independent methods such as

Illumina high-throughput sequencing. This study is far from being exhaustive in describing microbial communities associated with cabbage but gives a solid description of fungi associated with different plant tissues across different geographical regions. Three out of over fifty isolates tested, showed potential for biological control of Diamondback moth, however, further research needs to be conducted in order to formulate a biopesticide based on these isolates. In addition, more pests could be targeted by fungi isolated in this study.

This work is partly presented at [7th International Conference on Biodiversity Conservation and Ecosystem Management](#) July 26-27, 2018, Melbourne, Australia