

Characterizing the Defense Hierarchy of *Populus trichocarpa* and its Hybrids.

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Project Goals: Plants host a diverse array of endophytes that can affect plant disease severity either by interacting directly with pathogens or by modulating the plant defense response. Plants also possess genes for pathogen resistance. Thirdly, competition among pathogens can reduce overall damage. Our ongoing project seeks to characterize the contingency rules of the defense hierarchy that includes all three mechanisms: genes for resistance to leaf rust and eriophyid mite pathogens, the mite/rust competition, and fungal and bacterial endophytes for which we first developed methodologies (Brown et al, 2018). Our model system involves the leaves of *Populus trichocarpa*. We are also seeking to determine whether the contingency rules of the foliar defense hierarchy apply to other tissues (i.e., fruits and seeds) of the host. We still need to complete seven manuscripts in addition to the two listed that are in press.

Abstract.

Genes for resistance to the leaf-bronzing, eriophyid mite, *Schizoempodium mesophyllincola*, were identified in both the GWAS common garden of *Populus trichocarpa* genotypes, and in *P. trichocarpa* x *P. deltoides* (TxD) hybrid pedigrees (Newcombe et al, 2018). In the former case, a cutin/suberin gene has been identified; in the latter an exapted gene inherited from *Populus deltoides* was associated with a ‘sapsucker’ or aphid resistance QTL. In this latter case resistance is exapted because the mite has a restricted, endemic distribution in the maritime Pacific Northwest, a region in which *Populus deltoides* is not native. Exapted resistance has been the norm in past studies of resistance to various pathogens of *Populus trichocarpa* and its hybrids, so this new finding fits the overall pattern. In terms of larger significance, exapted resistance poses a challenge to our understanding of the evolutionary basis of genes for resistance. The discovery of the cutin/suberin gene provides a possible answer to this evolutionary question. The cutin/suberin gene confers exapted resistance to *Sphaerulina musiva*, a non-native pathogen of the Pacific Northwest, but its evolutionary persistence could be due to adapted resistance to the mite that is native to the region. Genetic resistance is, however, only part of the resistance hierarchy that our research is revealing. We have also found that rust and mite pathogens can directly compete for leaf tissue and that endophytes can antagonize the rust fungus. These three mechanisms form a hierarchy: first genetic resistance, then competition between mites and rust, and lastly endophyte-mediated resistance. What this means is that endophytes mediate resistance only when genetic resistance and competition are absent. In spite of these advances in understanding of the defense hierarchy in

poplar leaves, we also have uncovered new complexity. For example, we have found a fungus that may attack and regulate the mite. Secondly, we have found strong geographic structuring of foliar endophytes that has not yet been integrated into our understanding of the hierarchy. Finally, we have discovered that hierarchies may be tissue-specific. Our exploratory research with fruits (capsules) and seeds of *Populus trichocarpa* hints at a hierarchy unlike that of poplar leaves. This reproductive hierarchy involves a new disease (*Marssonina* capsule blight), a previously unknown seedling pathogen of poplar (a pathovar of *Pseudomonas syringae*), and another bacterium that antagonizes *Pseudomonas syringae*. The main distinguishing feature is that pathogen competition has not been observed between *Marssonina* and *Pseudomonas*. In its place is facilitation of one pathogen by another.

References

Brown, PS, Leopold, D, and Busby PE. 2018. Protocols for investigating the foliar mycobiome using high throughput DNA sequencing. *Methods in Molecular Biology: Plant Pathogenic Fungi and Oomycetes*. Springer Nature. In press.

Newcombe, George, Muchero, Wellington, and Posy Busby. 2018. Exapted resistance to an eriophyid mite in a hybrid pedigree of *Populus*. *PLOS ONE*: accepted pending suitable revision.

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