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Meeting the Challenge: Invasive Plants in Pacific Northwest Ecosystems



ABSTRACT

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During September 19-20, 2006, a conference was held at the University of Washington Botanic Gardens, Seattle, WA, with the title "Meeting the challenge: invasive plants in Pacific Northwest Ecosystems." The mission of the conference was to create strategies and partnerships to understand and manage invasions of non-native plants in the Pacific Northwest. The audience included over 180 professionals, students, and citizens from public and private organizations responsible for monitoring, studying, or managing non-native invasive plants. This proceedings includes twenty-seven papers based on oral presentations at the conference plus a synthesis paper that summarizes workshop themes, discussions, and related information. Topics include early detection and rapid response; control techniques, biology, and impacts; management approaches; distribution and mapping of invasive plants; and partnerships, education, and outreach.

KEYWORDS: Non-native plants, invasive, exotic, weeds, vegetation management, early detection/rapid response, biological control, integrated management.

ENGLISH EQUIVALENTS

When you know:	Multiply by:	To find:
Degrees Celsius (°C)	$(C*9/5) + 32$	Degrees Fahrenheit (°F)
Centimeters (cm)	.3937	Inches (in)
Meters (m)	3.2808	Feet (ft)
Kilometers (km)	0.6214	Miles (m)
Square meters per hectare (m ² /ha)	4.3560	Square feet per acre (ft ² /ac)

FUNGAL ENDOPHYTES IN SPOTTED KNAPWEED: DO THEY AFFECT ITS INVASIVENESS?

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ABSTRACT

To what extent do biotic interactions affect plant competition and fitness? This question relates to the general problem of understanding invasions by some exotic plants. Without the benefit of any evolutionary period that would allow for local adaptation to environmental conditions, some exotic plants are able to enter a plant community and outcompete the natives. The Novel Weapons Hypothesis, developed by Ray Callaway and his colleagues, explains this phenomenon in terms of biotic, plant-to-plant interactions that favor the novel invader. Developed from earlier ideas about allelopathy, the hypothesis appears to explain the phenomenal success of spotted knapweed, *Centaurea stoebe*, in western North America. Spotted knapweed roots have been reported to exude a chemical, (-)-catechin, that is thought to be a novel weapon suppressing the growth of naïve neighbors in the native plant community. Questions were raised last year, however, by a report of an unsuccessful attempt to repeat some of the research on (-)-catechin. Our interest in spotted knapweed as a model system for plant invasions is also based on biotic interactions. But, in our case, we are hypothesizing that fungal endophytes are influencing plant competition, as well as other plant behaviors. There is precedent, at least by analogy, for this hypothesis. John Klironomos showed that soil microbiota may promote plant invasions because invasive species avoid or fail to elicit negative feedback with soil pathogens. This negative feedback reduces the competitiveness and abundance of many native plants. More specifically, knapweed's interactions with native plants are affected by soil fungi, and soil microbes in its native range inhibit knapweed, as predicted by Klironomos.

KEYWORDS: *Centaurea stoebe*, spotted knapweed, endophytes, competition.

ROLE OF ENDOPHYTES

Soil microbes are notoriously diverse (Curtis et al. 2002, Fierer and Jackson 2006); some may be 'residents,' whereas others may be 'transients' that disappear from the soil community as their specific plant substrate degrades. It is not yet clear whether fungal endophytes of aerial plant parts are generally soil residents or transients; the latter seems more likely. Apart from a brief soil phase, even non-systemic endophytes would thus spend most of their poorly understood life cycle in their host plants. In any event,

endophytes are associated with a growing list of ecological roles: thermotolerance of plants growing in geothermal soils (Redman et al. 2002); community biodiversity (Clay and Holah 1999); enhancement of plant growth (Ernst et al. 2003). Specific endophytes may play specific roles, and many plants host very diverse arrays of endophytes (Ganley et al. 2004; Ganley and Newcombe 2006).

Endophytes in introduced plants like knapweeds must either have been co-introduced in seeds of their host, or have 'jumped' from other plants in the invaded range of

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their host. Because we are interested in the nativity or origin of fungi, we have been sampling fungal endophytes from knapweed in both its native range in Europe, and in its invaded range in North America. From 2004 to 2006, we have obtained isolation frequencies from seeds of the native range in Europe varying from 13 to 100 percent; some sites in the Pacific Northwestern portion of the invaded range have yielded relatively high isolation frequencies also, but more often samples of 100, surface-sterilized seeds have yielded no endophytes at all. The reasons for the variation in isolation frequency may emerge in analyses that we will be performing with our in-progress, extensive, 2006 survey.

So far, most of our endophytes from knapweed seed have tended to belong to a few fungal genera: *Alternaria*, *Fusarium*, and *Botrytis*. There are also rarer occurrences of endophytes belonging to *Ulocladium*, *Cladosporium*, *Aureobasidium*, *Epicoccum*, *Phoma*, and *Nemania*. Within many of these genera, we are also discovering diversity in ITS haplotypes; for *Alternaria*, various Alt a1 haplotypes are emerging. Some haplotypes have been found in knapweed in both of its ranges, and these are suggestive of co-introduction. Differences in community composition between native and invaded ranges also appear likely, and these differences may affect behavior, including invasiveness.

IDENTIFYING SOURCE AND MOVEMENT OF ENDOPHYTES

Greenhouse experiments have been performed with seedlings that were either E⁺ [endophyte-infected] or E⁻ [endophyte-free]. Flowers of both E⁺ and E⁻ plants, after pollination with bee abdomens, yielded E⁻ seed. Apparently, endophytes are not systemic in spotted knapweed. An endophyte cannot be vertically transmitted in seed of a knapweed plant unless it has infected that plant at some point. In the greenhouse, infection events are unlikely as foliage is never wet for the period of time that most fungi require for infection.

Re-isolation experiments in the greenhouse have revealed that if flowers are pollinated and then inoculated 48 hours later, the inoculated endophyte can be re-isolated

from the surface-sterilized, mature seed at a rate that varies with the endophyte. In other words, infection can take place during flowering, but it is likely that it can also occur before flowering.

Some endophytes produced sesquiterpenoid volatiles; these same isolates repelled seedhead weevils, *Larinus minutus*, when inoculated into pollinated flowers 12 hours prior to weevil introduction. Many knapweed endophytes significantly reduced germination of seed of Idaho fescue, *Festuca idahoensis*. Significantly fewer knapweed endophytes reduced germination of seed of knapweed itself. One *Fusarium* isolate caused significant knapweed seedling mortality.

In model competition experiments that were patterned after Callaway's groundbreaking work (Ridenour and Callaway 2001), endophytes in knapweed affected Idaho fescue plants grown in the same pots. E⁺ knapweed plants were significantly bigger (i.e., greater dry biomass) than their E⁻ counterparts. The opposite was true of fescue: E⁺ plants were significantly smaller than the E⁻. This result, in particular, is difficult to reconcile with the emerging differences in isolation frequency in the two ranges. Where spotted knapweed is native and non-invasive, endophytes occur at high frequency. Where spotted knapweed is non-native and invasive, endophytes are frequently absent. Resolution of this conundrum will likely involve distinctions among specific endophytes, their distributions, isolations from leaves and roots in addition to seeds, and possibly other factors.

In conclusion, our experimental research to date suggests that endophytes influence the invasiveness of spotted knapweed by affecting competition and relationships with herbivorous insects. But endophyte diversity in spotted knapweed is substantial, and effects on competition and other biotic relationships may very well depend on specific endophytes. Our understanding of the distributions and life cycles of specific endophytes in knapweed's native and invaded ranges is still fragmentary. Explaining invasiveness in simple terms has proven challenging for ecologists (Blair et al. 2006), but no hypothesis to date has included the contributions of endophytes.

LITERATURE CITED

- Bais, H.P.; Vepachedu, R.; Gilroy, S.; Callaway, R.M.; Vivanco, J.M. 2003.** Allelopathy and exotic plant invasion: from molecules and genes to species interactions. *Science*. 301: 1377–1380.
- Blair, A.C.; Nissen, S.J.; Brunk, G.R.; Hufbauer, R.A. 2006.** A lack of evidence for an ecological role of the putative allelochemical (\pm)-catechin in spotted knapweed invasion success. *Journal of Chemical Ecology*. 32: 2327–2331.
- Blair, A.C.; Hanson, B.D.; Brunk, G.R.; Marrs, R.A.; Westra, P.; Nissen, S.J.; Hufbauer, R.A. 2005.** New techniques and findings in the study of a candidate allelochemical implicated in invasion success. *Ecology Letters*. 8: 1039–1047.
- Callaway, R.; Thelen, G.C.; Barth, S.; Ramsey, P.W.; Gannon, J.E. 2004a.** Soil fungi alter interactions between the invader *Centaurea maculosa* and North American natives. *Ecology*. 85: 1062–1071.
- Callaway, R.M.; Thelen, G.C.; Rodriguez, A.; Holben, W.E. 2004b.** Soil biota and exotic plant invasion. *Nature*. 427: 731–733.
- Clay, K.; Holah, J. 1999.** Fungal endophyte symbiosis and plant diversity in successional fields. *Science*. 285: 1742–1744.
- Curtis, T.P.; Sloan, W.T.; Scannell, J.W. 2002.** Estimating prokaryotic diversity and its limits. *Proceedings of the National Academy of Sciences*. 99: 10494–10499.
- Ernst, M.; Mendgen, K.W.; Wiersel, S.G.R. 2003.** Endophytic fungal mutualists: seed-borne *Stagonospora* spp. enhance reed biomass production in axenic microcosms. *Molecular Plant-Microbe Interactions*. 16: 580–587.
- Fierer, N.; Jackson, R.B. 2006.** The diversity and biogeography of soil bacterial communities. *Proceedings of the National Academy of Sciences*. 103: 626–631.
- Ganley, R.J.; Newcombe, G. 2006.** Fungal endophytes in seeds and needles of *Pinus monticola*. *Mycological Research*. 110: 318–327.
- Ganley, R.J.; Brunsfeld, S.J.; Newcombe, G. 2004.** A community of unknown, endophytic fungi in western white pine. *Proceedings of the National Academy of Sciences*. 101: 10107–10112.
- Klironomos, J.N. 2002.** Feedback with soil biota contributes to plant rarity and invasiveness in communities. *Nature*. 417: 67–70.
- Redman, R.S.; Seehan, K.B.; Stout, R.G.; Rodriguez, R.J.; Henson, J.M. 2002.** Thermotolerance generated by plant/fungal symbiosis. *Science*. 298: 1581.
- Ridenour, W.M.; Callaway, R. 2001.** The relative importance of allelopathy in interference: the effects of an invasive weed on a native bunchgrass. *Oecologia*. 126: 444–450.