The effects of Phragmites Karka on Invasion in Plant Communities

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Abstract

Phragmites karka, a ubiquitous wetland plant, has been considered one of the most insidious species in the world. Allelopathy appears to be one of the invasion mechanisms; however, the effects could be masked by resource competition among target plants. The difficulty of distinguishing allelopathy from resource competition among plants has hindered investigations of the role of phytotoxic allelochemicals in plant communities. This has been addressed via experiments conducted in both the greenhouse and laboratory by growing associated with other plants. This study investigated the potential interacting influences of allelopathy and resource competition on plant growth–density relationships. The results demonstrated that plant–plant interference is the combined effect of allelopathy and resource competition with many other factors but this experimental design, target-neighbor mixed-culture in combination of plant grown at varying densities with varying level of phytotoxins, mono-culture, can successfully separate allelopathic effects from competition.

Keywords - Allelopathy, Density-dependent phytotoxin, Ecosystems, Invasion, *Phragmites karka*, Resource competition.

Introduction

Allelopathic interference by invasive plant species has potential to impact seed germination, seedling growth, development and establishment of neighboring plant species, as well as of the same species, in both natural and agricultural systems (Bich and Kato-Noguchi 2014). Allelopathy has been considered an important attribute to the success of an invasive species in natural ecosystems (Kimura et al. 2015; Lorenzo et al. 2010). The sources of allelochemicals released into the rhizosphere include leaching from leaves and other aerial parts, volatilization, root exudation and litter decomposition (Hussain and Reigosa 2012; Uddin et al. 2012).

Phragmites karka, a ubiquitous wetland plant, is considered one of the most invasive species in the world (Uddin et al. 2012) however; the origin of the species is still unclear (Plut et al. 2011). A perennial graminaceous plant, to 3 m tall, it reproduces mainly through rhizomes and, at low frequency, through seeds. Due to the impacts of *P. karka* invasions, habitats have been diminished or altered significantly for other flora and fauna causing loss of biodiversity and ecosystem functions. Several studies have identified chemicals within *P. karka* organs which have antialgal, antifungal or antibacterial effects (Li and Hu 2005). Previous allelopathic studies have shown that water extracts, decomposed materials, root exudates and specific identified chemicals of *P.karka* organs have strong phytotoxic effects on germination, growth, and establishment of other plant species (Kettenring et al. 2011; Rudrappa et al. 2007, 2009; Uddin et al. 2014 a) and thus, it is assumed that *P. karka* achieves its competitive advantages over invasion process into wetlands through allelopathy (Bains et al. 2009; Rudrappa et al. 2007).

While *P.karka* has clearly shown phytotoxic potential, the effects should be considered in more ecologically realistic ways by differentiating allelopathic interactions from resource competition. The allelopathic effects might be masked by resource competition among target plants (Barto and Cipollini 2009; Weidenhamer et al. 1989). A better understanding of dose–response relationships of allelochemicals would help to clarify this issue. Toxin dilution is thought to occur because plants share and compete not only for resources but also for toxin (Hansi et al. 2014). As the study of allelopathic interactions may be hindered by the lack of proper experimental methods, it may be more productive to first demonstrate explicit interference, by allelochemicals rather than rely solely on explanations that involve resource competition or other mechanisms.

Therefore, this study has been designed to determine the occurrence and magnitude of potential allelopathic effects mediated by *P. karka* root exudates, its litter and extracts of litter with a wide range of doses through a density-dependent approach. This method might be effective in distinguishing the allelopathic interactions of *P.karka* with neighbouring plant species from resource competition. We hypothesized that phytotoxic effects of allelochemicals depend on the neighbouring plant density, due to phytotoxins dilution among individual plants.

Methods

Fallen leaves of *P. karka* were collected from natural stands in Dr. Bhim Rao Ambedkar Govt. College Sri Ganganagar. Plant samples were sorted from other plant residue and debris, then kept at room temperature to air dry until constant dry weight. After desiccation, sorted samples were cut into small pieces and preserved in plastic ziplock bags until use. Soil samples were collected from the top layer of *P. karka* free areas of the same study site, separated from other organic materials, dried at room temperature and kept in ziplock bags after passing through a 2 mm sieve.

Choice of Target Species

Seeds of several species were used to determine any differential response in native, introduced and model species to allelopathy. Seeds of a model species, *Lantana camara* were purchased from a commercial source. Model species is easily grown, minimizing the risk of observed growth differences, due to factors other than treatments applied in the experiments.

Greenhouse Experiments

Un Burnt Versus Burnt Litter Extracts Mediated Effects on *L. Camara* Seedlings

Phragmites karka litter was placed in a furnace at 300 °C for 1 h to produce burnt litter and a phyto-toxicity test was conducted with extracts of both type of litter (un burnt and burnt). Extracts were made by mixing unburnt and burnt litter powder passed through 0.5 mm sieve with distilled water to a concentration of 10%. After a 24 h period, it was filtered with cheese cloth and centrifuged at 3000 rpm. The resultant filtrate then used as the extract for the experiment with pH adjustment at 6.5 with 1 N NaOH and 1 N HCl, to avoid non-relevant effects of extracts due to pH. Extract (5 mL) at four different concentrations (0, 2.0, 4.0, and 6.0%) was placed into a sterile 9 cm Petri dish containing two sterile sheets of filter paper (Whatman No. 1). At least, three replicates were used for each treatment with density of one, two, four and eight pre-germinated

L.camara seedlings. Petri dishes were sealed with parafilm and then placed in polyethylene bags to prevent water loss by evaporation and to avoid contamination by fungi and bacteria. The prepared dishes were placed in a growth chamber according to above mentioned conditions. The Petri dishes were randomized each day to minimize the spatial effect. After 7 days of experiment, phenotypic characteristics of the grown of the plants were measured.

Litter Mediated Effects on *L. Camara* Seed Germination

In seed germination bioassay four different concentrations of air dried litter by weight (0, 2.0, 4.0, and 6.0%) were placed in between two layers of 0.5% agarose (total 10 mL) in a container of 4.5 by 5.5 cm. Agarose was autoclaved at 121 °C for 15 min and subsequently cooled at room temperature. In addition, microbial community in substrate convert phytotoxins to more active or less active compounds but autoclaving may eliminate the possibility (Duke et al. 2009). Using autoclaved agarose with litter, the seeds of *L. camara*at a density of four, eight, and 16 were placed on each container and incubated for 7 days as above condition with three replicates. The germination and biometric parameters were measured at the end of the experiment.

Discussion

Analysis of growth–density relationships is useful tool for understanding the resource competition and allelopathic interference between plants of the suspected invasive species (Weidenhamer et al. 1989). The issue 'separating allelopathy from resource competition' is controversial in natural ecosystems but it is important in plant–plant interactions to evaluate the relative contribution and identify the mechanisms involved in their biological invasion processes.

Density-dependent phytotoxicity stands in contrast to resource competition as increased growth of plants at low density is dependent on large part to the amount of resources available. Despite the allelopathic potential of *P. karka* on associated and model plant species, as shown by the growth of *L. camara* observed in this study was masked by the resource competition but the allelopathic effects of *P.karka* are well supported (Rudrappa et al. 2007; Uddin et al. 2014a). These studies showed that water extracts of different organs, residue decomposition and root secreted phytotoxins had negative effect on germination, growth, and development of other plant species.

In greenhouse experiments of this study, the strongest growth inhibition was observed in high density treatments when compared to low and medium density. This demonstrates resource competition is the dominating factor, consistent with other studies (Uddin et al. a).In addition, the laboratory experiments showed a clear density-dependent phytotoxic effect, a result well aligned with other studies (Hansi et al. 2014; Lambertini et al. 2012) where allelochemicals, herbicides and inorganic compounds such as copper showing phytotoxicity is density-dependent.

The inhibition increased as the concentration increased at lower density but stimulation was observed with intermediate density in most of the cases. This suggests that lower seed density increases the availability of phytotoxin per seed. In general, allelopathy research is more concerned with using concentrations, the introduction of soil microbes, and an autotoxicity test, involving a wide range of associated plant species in the bioassays, by questioning whether those involved are ecologically relevant. Despite this, it may be difficult to determine with some precision as to those occurring naturally in the field, but this has importance in determining causal relationships and minimizing effects due to unnaturally occurring situations. Therefore, allelopathy studies should consider more complex bioassays involving soil microbial communities, field concentrations of allelochemicals, multiple test species and using native leachate as a control that might represent the ecological phenomena in the field. For example, soil

Table – 1

Allelopathic Impact of 3% (W/V) Aqueous Leachate of Above Ground (Ag) And Below Ground (Bg) Plant Parts of Phragmites Karka On Lantana Camara

Growth parameters	Control	P. karka		LSD
		Ag	Bg	
Germination (%)	96.00 ± 2.44	88.00 ± 3.74	96.00 ± 2.44	9.07
GIR	-	8.33	0.00	-
Root length (cm)	5.45 ± 0.10	3.55 ± 0.01*	6.11 ± 0.41*	0.21
Shoot length (cm)	4.38 ± 0.08	3.45 ± 0.12*	4.65 ± 0.08	0.31
Total dry weight (g)	0.151±0.01	0.070 ± 0.01*	0.142 ± 0.01	0.05

Mean ± SE, GIR -Germination inhibition rate, LSD -Least significant differences,* Significant at 0.05 % level by Dunnett's test applied after ANOVA

For example, soil microorganisms might play an important role in influencing the bioavailability of allelochemicals in soil (Bauer et al. 2012), which could be achieved by the addition of microbial inoculums in experimental soil substrate collected from test species grown in the field. So, further research related to incorporation of soil microorganisms might be imperative to advance the allelopathy as one of its invasion mechanisms.

An effort has been made to overcome the concern through measuring the concentration of allelochemicals in the *P. karka* rhizosphere soil, considering the osmotic potential of higher concentrations (Uddin et al. 2014a), adjusting pH, and measuring the quantity of litter biomass produced per unit of soil or covered area. These criteria in our previous studies have been taken into consideration in this current study. Moreover, separation of allelopathic effects from resource competition is a vital point in allelopathy research which has been addressed in this study, indicating phytotoxins secreted by different means from *P.karka* are responsible for invasion process except root exudations. On the other hand, plant–plant allelopathic interactions may be explained by species-specific.

Conclusions

The overall observation of growth reductions in test plant species at low densities was inconsistent with the standard resource competition hypothesis and provides support for the hypothesis of chemical interference by *P.karka*. Although, the growth response of test species did not follow the consistency in all experiments, in most cases, the results demonstrate the density-dependent phytotoxicity concept. Therefore, these studies may provide an understanding of plant–plant allelopathy interactions and may distinguish the mechanisms involved in plant interference i.e.

resource competition and allelopathy. Our findings may be useful to evaluate the response of agricultural plants such as *L. camara* to weed residues, and may also provide insight evidence of allelopathic potential in *P.karka* invaded wetlands. In addition, the density-dependent phytotoxicity phenomenon may bring important ecological implications as a methodological approach in allelopathy (Weidenhamer and Romeo 1989).

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