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Original Article

Impacts of invasive *Chromolaena odorata* on species richness, composition and seedling recruitment of *Shorea robusta* in a tropical Sal forest, Nepal

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Abstract

Chromolaena odorata is an invasive species known to have its adverse impacts on native diversity. We studied its impacts on plant species richness, composition and *Shorea robusta* seedlings in a tropical Sal forest of Nepal. We conducted field experiment along transects established in invaded and uninvaded understory vegetation. Our results show differences in native species richness and *S. robusta* seedling density between invaded and uninvaded plots. The invaded plots are associated with fewer species than uninvaded plots. Plot type (invaded and uninvaded), *C. odorata* density and cover show an effect on vegetation composition. Moreover, some of the native species are found replaced from *C. odorata* invaded sites. Overall, the Sal forest protects native diversity and Sal seedling recruitment besides having its high economic value and beneficial impacts on people's livelihood. Therefore, control and proper management of *C. odorata* is needed for conserving native vegetation and preventing future problems associated with invasion.

Keywords: *Chromolaena odorata*, *Shorea robusta*, invasive species, species richness, species composition

1. Introduction

Many of invasive alien species have caused various ecological, environmental, economic and human health problems (D'Antonio *et al.,* 2002; Pimentel *et al.,* 2001, 2005; Vilà *et al.,* 2011). They have been considered as one of the greatest threats to native diversity, ecosystem structure and functioning (Ehrenfeld, 2003; Gurevitch and Padilla, 2004; Higgins *et al*., 1999). Common characteristics of invasive alien species include capacity for high seed production, high germination rate, strong vegetative growth and phenotypic plasticity (Davidson *et al.,* 2011; Tiwari *et al.,* 2005). Beside these characteristics they bring changes in soil properties such as pH, moisture, organic matter content, nutrient concentrations (Ehrenfeld, 2003; Kourtev *et al.,* 1998; Timsina *et al.,* 2011) and microbial community (Chacon *et al.,* 2009; Klironomos, 2002; Mangla and Callaway, 2008).

Chromolaena odorata (L.) King & Robinson (Asteraceae), native to tropical America, is one of the serious weeds growing rapidly and invading a wide range of natural vegetation in Asia, Africa and Europe (McFadyen and Skarratt, 1996; Olaoye and Moody, 1986; Zachariades *et al.,* 2009). It had become prevalent in South Asia (India, Nepal and Sri Lanka) and South East Asia (Myanmar, Thailand, Vietnam, and Laos) by early twentieth century (Zachariades *et al.,* 2009). It has invaded tropical ranges in Asia up to 1,000

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meters above sea level (m.a.s.l.) (McFadyen, 1989). In Nepal, *C. odorata* is called "Seto banmara" which is becoming aggressive and invasive for four decades from eastern to central part of the country (Tiwari *et al.,* 2005). Disturbed areas, fallow land, road side, degraded forest margins and tree canopy gaps in the forests are the sites of its colonization (Tiwari *et al.,* 2005; Zachariades *et al.,* 2009). Native vegetation is found to be impacted by this species through altering soil microbial community (Mangla and Callaway 2008), changing soil quality (Mboukou-Kimbatsa *et al.,* 2007; Mandal and Joshi, 2014) and allelopathy (LiJun *et al.,* 2010; Hu and Zhang, 2013).

Tropical forests (< 1,000 m.a.s.l.) in Nepal is widely dominated by *Shorea robusta* C.F. Gaertn., commonly called 'Sal forest'. Native distribution of *S. robusta* (Sal) is found in Nepal, India and Myanmar (Orwa *et al.*, 2009). The Sal forest is extended into subtropical region or foothill of the Himalayas up to 1,500 m.a.s.l. (Gautam, 1990; Tewari, 1995). The understory vegetation in Sal forest is rich in species diversity (Webb and Sah, 2003) and plant communities in the forests represent unique entities in terms of their structure and composition as well (Timilsina *et al.,* 2007). Concerning conservation of forests, community forestry has becoming one of the best practices and successful methods in Nepal but anthropogenic disturbances and human intended management efforts have caused negative impacts on them (Shrestha *et al.,* 2010). As an example, alien species invasion has become one of the serious problems in the natural ecosystems (Dogra *et al.*, 2010).

The Sal forest habitat in Nepal is facing problem with the detrimental invader, *C. odorata* (Tiwari *et al.,* 2005). Luxuriant growth of *S. robusta* seedlings associated with number of native species is found in uninvaded sites and contrasting to this aggressively growing *C. odorata* has dominated over native understory vegetation along the invaded sites of the study area. We anticipated that *C. odorata* might have created severe ecological problems on species richness, composition and *S. robusta* seedling recruitment in the invaded Sal forests. Previous studies have focused on the effect of *C. odorata* on agroforestry and agricultural systems (e.g. Roder *et al.,* 1995; Ngobo *et al*., 2004). We compared species richness and density of *S. robusta* seedlings in *C. odorata* invaded and uninvaded plots as well as analyzed the effect of plot types, *C. odorata* density and cover on species composition in a Sal forest in Nepal to understand the potential impacts of *C. odorata* in field.

2. Methods

2.1 Study sites

The study was carried out in Panchakanya community forest located at Chitwan district of central Nepal. The community forest lies in tropical zone (170-216 m.a.s.l.) having dry-wet climate in winter and hot-humid in summer. Two study sites were selected in the community forest for the investigation. Site 1 was located at the north-west part (27°38′35″N 84°28′53″E) of the forest and site 2 was located at the south-east part (27°38′19″N 84°29′01″E). Each site was characterized by the dominance of *S. robusta* and both of them were homogenous in topography, soil type and vegetation.

2.2 Vegetation sampling

Vegetation sampling was done using belt transect method (Timsina *et al.,* 2011) from August to September 2014. Fifty quadrats (plots), each of size 1×1 m, were placed along 10 transects (100 m long) made at both study sites. A total of five quadrats were placed in each transect and the distance between two quadrats was at least 10 m. Each transects included at least two quadrats in *C. odorata* invaded and two quadrats in uninvaded parts. A quadrat was considered as uninvaded if the cover of *C. odorata* was less than 10%. Growing tree species with height < 1 m were considered as seedlings and counted in each plot. All species encountered in each quadrat were recorded and identified using standard literature (Hara *et al.*, 1982; Polunin and Stainton, 1984; Press *et al.*, 2000). Herbarium specimens were deposited at National Herbarium and Plant Laboratory (KATH), Godawari, Lalitpur, Nepal.

2.2.1 Data analysis

Canonical correspondence analysis (CCA) was used to analyze the effect of *C. odorata* on species composition in the study area as the gradient length in the data yielded by *detrended correspondence analysis (*DCA) was 3.129, and thus unimodal technique of analysis was applied. Effects of *C. odorata* density (number of *C. odorata* ramets), *C. odorata* cover and plot type (invaded and uninvaded) were tested on species composition. Species less than four occurrences and *C. odorata* were excluded from the species composition data. The data were down weighted to reduce the effect of the occurrences of rare species in the results. We used permutational multivariate analysis of variance (PERMANOVA) to test the significance of the relationships.

Independent sample t-tests and Mann-Whitney U tests were used to compare the means of total species richness, native species richness and number of *S. robusta* seedlings between invaded and uninvaded plots. Data on number of species at site 2 were square root transformed before analysis to meet the assumptions of the analyses. All tests were carried out in R Software, version 3.0.0 (R Development Core Team, 2013).

3. Results

3.1 Species richness

Thirty species were recorded at site 1 and 36 species at site 2 from both invaded and uninvaded plots. They are included in 31 genera and 17 families. The species at site 1 were also common at site 2. Among them, 26 species were native and 10 species were non-native (Table 1 and 2). Out of the non-native species 4 species were invasive (*Senna occidentalis* (L.) Link*, Senna tora* (L.) Roxb.*, C. odorata,* and *Mikenia micrantha* Kunth).

Number of native species (native species richness) at both sites was significantly different between invaded and uninvaded plots. The invaded plots were associated with fewer native species. At site 1, numbers of native species recorded were 3.25±0.27 in invaded plots and 4.15±0.31 in uninvaded plots (mean/m^2±SE; t-test, t=2.15, df=48, p=0.036). At site 2, numbers of native species were 3.92±0.3 in invaded plots and 4.85±0.34 in uninvaded plots (mean/m^2±SE; t-test, t=2.22, df=48, p=0.030). Total species (native and non-native) per plot was approximately 5-6 which was not significantly different between invaded and uninvaded plots for both sites.

3.2 Species composition

C. odorata invaded and uninvaded plots showed different species composition. The plot types (invaded and uninvaded) explained 2.42% of the total variation in CCA analysis in the species composition data set which is 7.44% of the variation which could be explained by one ordination

Table 1. Numbers of native and non-native species found at both study sites.

Type of species	Site 1	Site 2
Native species	21	26
Non-native species	9	10
Total	30	36

Table 2. Plant species reported from the study plots, their abbreviation, family, growth habit and status.

Species	Abbreviation	Family	Growth habit	Status
Ageratum houstonianum Mill.	age.hau	Asteraceae	Perennial	Non-native
Cassia fistula L.	cas.fis	Fabaceae	Perennial	Native
Senna occidentalis (L.) Link	cas.occ	Fabaceae	Perennial	Non-native
Senna tora (L.) Roxb.	cas.tor	Fabaceae	Perennial	Non-native
Chromolaena odorata (L.) King & Robinson	chr.odo	Asteraceae	Perennial	Non-native
Clerodendrum viscosum Vent.	cle.vis	Verbenaceae	Perennial	Native
Colacasia sp.	col.sp	Araceae	Perennial	Native
Commelina sp.	com.sp	Commelinaceae	Annual	Native
Costus speciosus (Koenig) Sm.	coc.spe	Zingiberaceae	Perennial	Native
Crotalaria sp.	cor.sp	Fabaceae	Perennial	Native
Cyanotis vaga (Lour.) J. A. & J. H. Schult.	cyn.vag	Commelinaceae	Annual	Native
Cyanotis sp.	cyn.sp	Commelinaceae	Annual	Native
Cyperus rotundus L.	cyp.rot	Cyperaceae	Perennial	Native
Dalbergia sissoo Roxb. ex DC.	dal.sis	Fabaceae	Perennial	Native
Desmodium caudatum DC.	des.cau	Fabaceae	Annual	Native
Desmodium sp.	des.sp	Fabaceae	Annual	Native
Desmodium pulchellum (L.) Benth.	des.pul	Fabaceae	Perennial	Native
Dioscorea bulbifera L.	dio.bul	Dioscoreaceae	Perennial	Native
Elephantopus scaber L.	ele.sca	Asteraceae	Perennial	Non-native
Evolvulus nummularius (L.) L.	evo.num	Convolvulaceae	Perennial	Non-native
Galinsoga parviflora Cav.	gal.par	Asteraceae	Annual	Non-native
Helicteres isora L.	hel.iso	Sterculiaceae	Perennial	Native
Imperata cylindrica (L.) P. Beauv.	imp.cyl	Poaceae	Perennial	Native
Kyllinga sp.	kyl.sp	Cyperaceae	Perennial	Non-native
Leea crispa Royen ex L.	lee.cri	Leeaceae	Perennial	Native
Lindernia sp.	lin.sp	Scrophulariaceae	Annual	Native
Mikania micrantha Kunth	mik.mic	Asteraceae	Perennial	Non-native
Ophioglossum petiolatum Hooker	oph.pet	Ophioglossaceae	Annual	Native
Oplismenus burmannii (Retz.) P. Beauv.	opl.bur	Poaceae	Annual	Native
Phyllanthus urinaria L.	phy.uri	Phyllanthaceae	Annual	Non-native
Pogostemon benghalensis (Burm. f.) Kuntze	pog.ben	Lamiaceae	Perennial	Native
Saccharum spontaneum L.	sac.spo	Poaceae	Perennial	Native
Setaria pumila (Poir.) Roem. & Schult.	set.pum	Poaceae	Annual	Native
Shorea robusta C.F. Gaertn.	sho.rob	Dipterocarpaceae	Perennial	Native
Tadehagi triquetrum (L.) Ohashi	tad.tri	Fabaceae	Perennial	Native
Typhonium sp.	typ.sp	Araceae	Annual	Native

axis (p=0.003) (Figure 1).

Similarly, the factors (both *C. odorata* cover and density) explained 3.12% of the total variation in the species composition data set that is 9.58% of the total variation which could be explained by two ordination axis (p=0.038) (Figure 2). The *C. odorata* cover and density separately explained 2.01% (p=0.011) and 2.21% (p=0.006) of the total variation respectively in the data set. The variations are 6.19% and 6.79% respectively of the total variation that could be explained by one ordination axis (not shown).

3.3 Shorea robusta seedling recruitment

Numbers of *S. robusta* seedlings were significantly different between invaded and uninvaded plots. Uninvaded plots were found to be associated with greater number of seedlings. At site 1, numbers of seedlings were 2.73±0.50 in invaded plots and 9.0±1.03 in uninvaded plots (mean/m^2± SE; Mann-Whitney U test, p=0.036). Similarly, at site 2, the numbers of seedlings were 5.08±0.95 and 10±1.33 in invaded and uninvaded plots, respectively (mean/m^2±SE; t-test, t= 3.04, df=48, p=0.003).

4. Discussion

The results show that *C. odorata* has effects on species richness and composition in a tropical Sal forest. The vegetation composition is different between invaded and uninvaded plots. CCA analysis indicates that non-native herbaceous plants such as *Evolvulus nummularius* (L.) L., *Phyllanthus niruri* L. and *Ageratum houstonianum* Mill. and native shrub *Clerodendrum viscosum* Vent. are associated with *C. odorata* while certain monocots *Oplismenus burmannii* (Retz.) P. Beauv., *Cyanotis* sp., *Costus specious* (Koenig), *Dioscorea bulbifera* L., and dicot shrub species *Helicteres isora* L. and *Desmodium* sp. are replaced by *C. odorata*. CCA analysis suggests that *C. odorata* density and cover affect vegetation composition in the invaded plots. This result is similar to the results of Norgrove *et al.* (2008) who found reduction of certain monocots (*Aframomum* sp. and *Murdannia simplex*) and increase of abundance of some alien dicot weeds (*Stachytarpheta cayennensis* and *Ageratum conyzoides*) with the invasion of *C. odorata* in the study of plant community composition in invaded and uninvaded savannah sites in north-west Cameroon. This lends further support to the hypothesis that *C. odorata* invasion causes a shift in native species abundance with an increase of non-

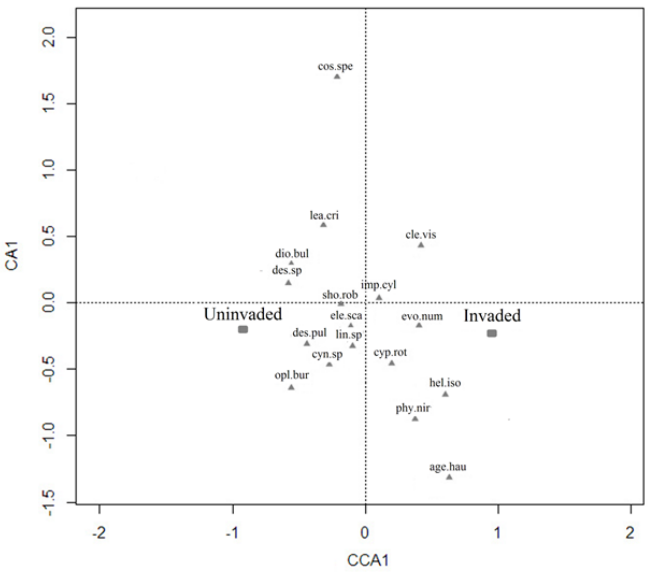

Figure 1. Results of CCA analysis showing the effect of plot type on species composition in ordination space. Full names and authors of each species are given in Table 2.

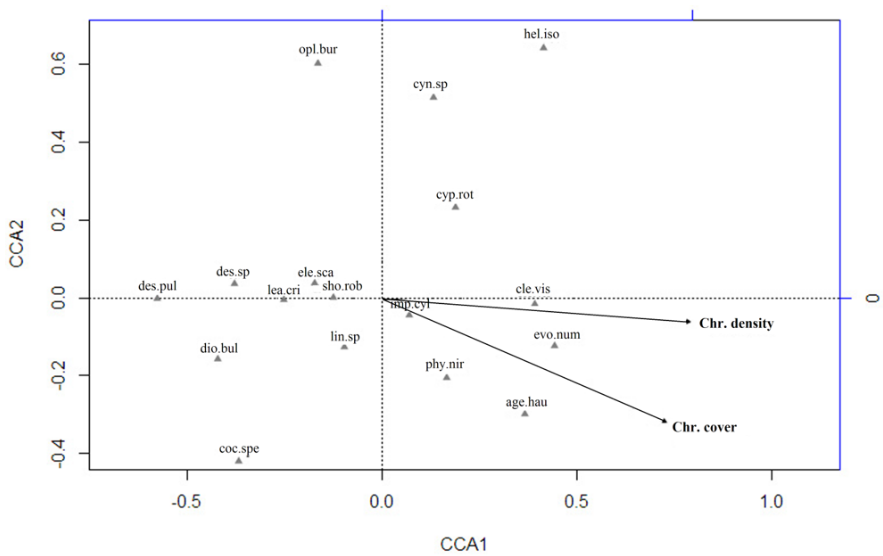

Figure 2. Results of CCA analysis showing the effect of *C. odorata* density and cover on species composition. (Chr.: *Chromolaena odorata*; Full names and authors of each species are given in Table 2).

native species.

Our results also demonstrate that native plant species richness is negatively affected by *C. odorata* invasion. A significant difference was found in native species richness between invaded and uninvaded plots but no significant differences in total species richness. This suggests that non-native plant species replace native species in invaded areas or *C. odorata* invasion supports introduction of non-native species during replacement of native plant species. Moreover, significantly lower *S. robusta* seedling density in invaded plots is an evidence of inhibitory impact of *C. odorata* on *S. robusta* seedlings in nature. These findings are in accordance with previous findings by De Rouw (1991), Goodall and Erasmus (1996), Mangla and Callaway (2008), Norgrove *et al.* (2008), and Hu and Zhang (2013).

Negative feedback mechanism by soil microbial community in invaded areas might have essential role in the replacement of native species. An important regulator of plant community structure is the ability of plants to change soil microbial communities (Klironomos, 2002). Mangla and Callaway (2008) had reported that *C. odorata* is able to accumulate high concentration of the generalist soil born fungal pathogen (*Fusarium* spp.) responsible for creating a negative feedback for native plant species and concluded that the impact of *C. odorata* is due to biotic interactions among native plants and soil biotic community.

Allelochemicals released from *C. odorata* could also play an inhibitory role for native species growth in invaded areas. Novel weapons hypothesis proposes that the invasive species can produce novel biochemical weapons as powerful allelopathic agents in exotic range to harm associated vegetation (Callaway and Ridenour, 2004). Allelopathic property of this species has been described previously (eg. Ivens, 1974; Sahid and Sugau, 1993). Hu and Zhang (2013) observed more inhibitory effects of *C. odorata* leaf extract on native species (*Rottboellia exaltata*, *Digitaria sanguinalis, Hemistepta lyrata, Youngia japonica and Dicliptera chinensis*) than non-native species (*Bidens pilosa, Ageratum conyzoides, Amaranthus spinosus, Conyza sumatrensis* and *Chenopodium ambrosioides*). These indicate that the native plant species are more susceptible to allelopathic compounds released by *C. odorata* which is further supported by our findings.

C. odorata is also found suppressing the growth of tree species, pine and eucalypt (Matthews and Brand, 2004) similar to our study, i.e. there is significantly lower number of *S. robusta* seedlings in invaded plots than uninvaded plots. *C. odorata* produces masses of rapidly germinating seeds and grows aggressively forming a thick cover over neighboring plants which is another mechanism to suppress native plants (De Rouw, 1991; Goodall and Erasmus, 1996; Awanyo *et al.*, 2011). The *S. robusta* seedlings and other native species might be more susceptible to any above mentioned mechanisms developed by *C. odorata* rather than non-native species such as *Evolvulus nummularius*, *Phyllanthus niruri* and *Ageratum houstonianum*.

Interestingly, it is suggested that *C. odorata* increases soil nutrients (NPK, magnesium) and soil organic content, which is likely to support the growth of several non-native species in invaded areas (Mandal and Joshi, 2014). As *C. odorata* prefers open canopy of forest in non-native ranges (Kluge, 1991; Malahlela*et al.,* 2015; Tiwari *et al.,* 2005), it would be interesting to assess its invasion success in deep Sal forests. Joshi *et al.* (2013) has described an important mechanism of its successful invasion through its underground stem (corm) which is responsible for vegetative growth. The corm cannot be affected by forest fire while upper parts of this plant and native vegetation get destroyed. Later on the corm regenerates shoots in rainy season and dominates over other species in the next growing season (Joshi *et al.*, 2013).

Currently, the controlling and management activities of *C. odorata* have not been taken seriously by community people and managers which indicate that the level of *C. odorata* invasion would be more severe in future. As long term impact of *C. odorata,* native plant diversity would be negatively affected with eradication of important elements from understory vegetation of Sal forest. The soil ecology and dynamics would be altered and only a pure stand of *C. odorata* could be expected in Sal forests in future. Results of this study could be helpful to raise some questions about certain mechanisms adopted by *C. odorata* for its invasion in the Sal forest which would replace native species and having negative impacts on *S. robusta* seedling establishment.

In conclusion, *C. odorata* has great impact on native species diversity, change community composition and create problems with *S. robusta* seedling recruitment in tropical Sal forests in Nepal. It is capable of replacing native species by supporting non-native species in invaded areas. Importantly, its impact on *S. robusta* seedlings indicates threats to natural regeneration of high valuable tree. It is expected that the level of *C. odorata* invasion would be increased more severely as there is no more activities of controlling and management of this weed. Therefore, managers, responsible authorities and community people should be aware to implement appropriate measures for controlling and management of *C. odorata* to conserve native ecosystem and diversity.

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