ISSN: 1991-2951 (Print)
ISSN: 2091-2781 (Online)

Nature Conservation & Health Care Council

Our Nature

Journal homepage: http://nepjol.info/index.php/ON

Growth performance of Sal seedlings in sterilized and unsterilized soils infested by Siam weed (*Chromolaena odoarata*)

Muna Baral[1,a], Chandra Bahadur Thapa[2.b], Sanjay Kumar Jha[1,c], and Lal B. Thapa[1,d,*]

[1]*Central Department of Botany, Tribhuvan University, Kirtipur, Kathmandu, Nepal*
[2]*Department of Botany, Butwal Multiple Campus, Tribhuvan University, Butwal, Nepal*

**E-mail: lal_thapa25@yahoo.com*
[a]*https://orcid.org/0009-0000-2745-2654*, [b]*https://orcid.org/0000-0001-5113-9043*, [c]*https://orcid.org/0000-0001-9737-3214*, [d]*https://orcid.org/0000-0003-3513-2276*

Abstract

The role of alien invasive plants on the interaction between native plants and soil has been a critical concern for understanding invasion mechanism and response of native plants towards invasion. This study aims to analyze the effect of invaded soils by an invasive Siam weed (*Chromolaena odorata* (L.) R.M. King & H. Rob.) under sterilized and unsterilized conditions on growth performance of seedlings of a valuable native Sal tree (*Shorea robusta Gaertn. f.*). For the analysis, seedlings of *S. robusta* were grown in pots and growth parameters were measured. Results showed that the *C. odorata* invaded soil reduces the root biomass, leaf length and leaf breadth of *S. robusta* seedlings. Sterilization of the invaded increased root and shoot and leaf size of the seedlings. In conclusion, soil sterilization can promote early-stage growth for *S. robusta* seedlings, adjusting with altering the shape and size of leaves in response to invasion. This suggests that soil microbes play an important role in negatively impacting native plants with invasion of *C. odorata*.

Keywords: Invasiveness, seedling growth, *Shorea robusta,* soil microbes, sterilization

DOI: https://doi.org/10.3126/on.v23i1.74963
Manuscript details: Received: 25.07.2024 / Accepted:20.11.2024
Citation: Baral, M., Thapa, C.B., Jha, S.K. and Thapa, L.B. 2025. Growth performance of Sal seedlings in sterilized and unsterilized soils infested by Siam weed (*Chromolaena odoarata*). *Our Nature* 23(1): 7-13. DOI: https://doi.org/10.3126/on.v23i1.74963

Introduction

Chromolaena odorata (L.) R.M. King & H. Rob. is one among the worst invasive alien species aggressively growing throughout tropical and subtropical regions in the countries of Old world (Zachariades *et al.*, 2009). It has invaded a wide range of habitats including roadsides, agricultural fields, disturbed grasslands, forests, and abandoned fields resulting in significant economic and biodiversity losses (Rusdy, 2016; Shrestha, 2019).

This plant is known to alter native plant communities and suppress the germination, growth and development of several native plants (De Rouw, 1991; Uwalaka and Muoghalu 2021; Poudel*et al.*, 2024). These negative impacts are further intensified by the alteration of soil properties and microbial interactions by the invasion of this weed (Tiébré and Gnanazan, 2018; Zhang *et al.*, 2024). In Nepal, tropical Sal (*Shorea robusta Gaertn. f.*) forest have been severely impacted by *C. odorata* invasion, threatening this vital ecosystem (Thapa *et al.*, 2016; Bhatta *et al.*, 2020).

Field studies highlighted adverse impacts of *C. odorata* on native species composition or seedling regeneration (Thapa *et al.*, 2016; Gbètoho *et al.*, 2018). And, experiments to evaluate potential allelopathic impactof *C. odorata* on seed germination and seedling growth of native wild or crop plants have used extracts of most commonly leaves (Julio *et al.*, 2019; Poudel *et al.*, 2024). Still, there has been limited studies on evaluating potential effects of invasion of this weed, especially by growing native seedlings in both weed invaded and uninvaded soil to observe how the seedlings respond invasion. Such method closely simulates natural condition and may help to better understand the real impact of invasion.

The natural environment is a multifaceted system, where complex interactions exist among plant-soil and microbes including invasions (Rai, 2015). This make difficult to predict how these factors interact and influence each other. Though challenging, include or include microbes in experimental design helps to explain their role in native and invasive plant interactions. Excluding them allows observation of how their absence affect plant competition and including them exposes their beneficial or harmful impact on plant growth, nutrient cycling, and competition (Kumar and Verma, 2018). One option of exclusion of microbes in pot experiments is soil sterilization (Cortois *et al.*, 2016). This study aims to analyze the effect of *C. odorata* invaded soil on growth and development of native *S. robusta* seedlings. Simultaneously, the study evaluates whether soil sterilization could reduce any potential toxic effects of the invaded soil on seedling growth.

Materials and methods

Seed and soil collection

Seeds of *S. robusta* and invaded and uninvaded soils were collected from Indreni Community Forest of Chitwan district, Bagmati Province, Nepal (84°28'16"E, 27°44'65"N, elevation 220-290 m asl) during July 2020. The seeds, which were freshly dropped on the ground from mother tree (*S. robusta*) and homogeneous in size, were collected. The seeds were transported to the laboratory and sown in pots prepared.

Invaded soil was collected from *C. odorata* invaded site (cover >50%) and uninvaded soil was collected from the site where *C. odorata* was absent from the depth 0-15 cm by removing surface litters and debris. The seed and soil samples were transported to the laboratory.

Seedling preparation, soil sterilization and pot experiment

Wings of the seeds were removed and the seeds were washed by distilled water and treated with the insecticide Carbine (10 mL per 100 mL of water) to protect seeds from being destroyed by pests. The seeds were spread on moist muslin cloth on a tray in dark to allow germination. The soaked seeds started to germinate after 5th days of soaking.

The polyethylene pots of size 14 cm diameter and 26 cm height were filled with the collected soil samples from the field. One thousand and five hundred grams of each invaded and uninvaded soils were kept in each pot separately. One set of each invaded and uninvaded soils samples were sterilized and another set was unsterilized. For the sterilization, soils were autoclaved at 121°C for 35 minutes.

The germinated seeds of *S. robusta,* (having radicle length 1 cm) were transplanted to the pots containing respective soils (inserting the radicle into moist soil) and allowed to grow into seedlings. The seedlings were exposed to altogether four treatments (i) uninvaded unsterilized soil (ii) uninvaded sterilized soil (iii) invaded unsterilized soil (iv) invaded sterilized soil. There were 10 replicated plots for each treatment and each pot contained a single seedling. The pots were kept in greenhouse of Central Department of Botany, Tribhuvan University, Kirtipur, Kathmandu, Nepal. The temperature and humidity were recorded from 17°C to 45°C and 40% to 80%, respectively during the period of experiment. The pots were randomized in each alternate day to minimize the positional effect.

In each sterilized and unsterilized pot, 20 mL of distilled water was poured daily.

Measuring parameters

The seedlings were harvested on 86th days after transplantation of germinating seeds into pots. Shoot length and biomass, root length and biomass, leaf size (lamina length and breadth) were measured. Leaf lamina length was measured from the base of the lamina to the leaf tip and breadth was measured from the widest middle part of the blade. The root samples were washed to remove adhered soil particles. After that, the root and shoot were dried in hot air oven at 80°C for 72 hours for measuring biomass. The shoot biomass included the leaves.

Statistical analysis

The growth parameters of *S. robusta*, including shoot and root length, shoot and root biomass, leaf size (length and breadth), root to shoot and leaf length to breadth ratios were analyzed using a Two- way Analysis of Variance (ANOVA) to evaluate the effect of soil type, soil sterilization and their interaction. Root length and shoot biomass data were log-transformed as the data were not normally distributed. Statistical significance was considered at a significance level of $p<0.05$. The data were analyzed using the software R (R Core Team, 2023).

Results

Root and shoot length

Analysis of root length revealed a significant effect of soil sterilization (P=0.050) and interaction between sterilization and soil type (P =0.026) (Table 1). Roots were longer in sterile invaded soil, while the root length was similar in both sterile and unsterile uninvaded soils (P = 0.523, Table 1, Fig. 1). Similarly, shoots of seedlings varied with soil sterilization (P=0.002, Table 1). Shoots were longer in sterilized soil condition (Fig. 1). Effect of soil type and interaction of soil type and sterilization did not show significant impact on shoot length (P > 0.05, Table 1).

Leaf length and breadth

Both soil type and sterilization showed significant change in leaf length (lamina) of *S. robusta* (P=0.020 and P < 0.001, respectively) but their interaction did not show the change in leaf length (P = 0.894) (Table 1). Leaves were longer in the sterile soil in both invaded and uninvaded soils (Fig. 3). Soil type had no effect on leaf breadth (P = 0.069), however, sterilization and interaction of soil type and sterilization showed significant change in the breadth (P < 0.001 and P = 0.013, respectively, Table 1). Leaf breadth was narrow in case of unsterile invaded soil (Fig. 3).

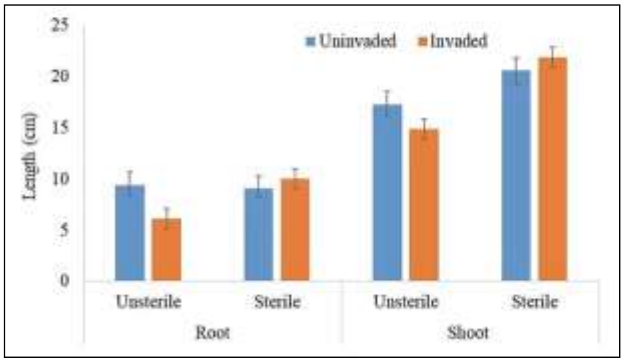

Figure 1. Effect of soil type and sterilization on root and shoot length of *S. robusta*. Bar graph shows the mean value ± standard error.

Root and shoot biomass

Root biomass of *S. robusta* differed significantly with soil type and interaction of soil type and sterilization (P = 0.046) but sterilization effect on the biomass was not significant (P = 0.329, Table 1). In unsterile condition, the root biomass was high in uninvaded soil, while the biomass was high in the sterile invaded soil than unsterile invaded soil (Fig. 2). Shoot biomass did not show variation due to soil type, sterilization and their interactions (P > 0.05, Table 1).

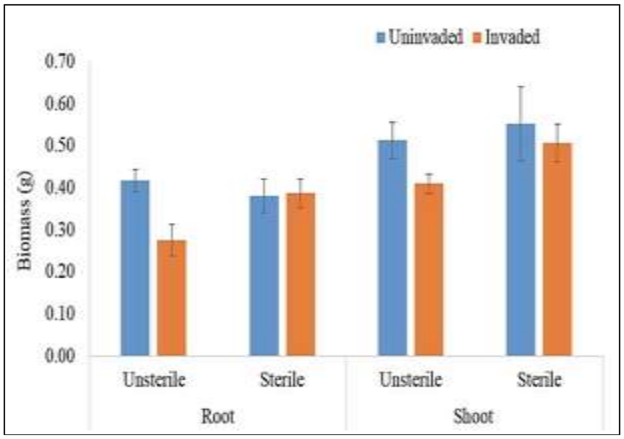

Figure 2. Effect of soil type and sterilization onroot and shoot biomass of *S. robusta*. Bar graph shows the mean value ± standard error.

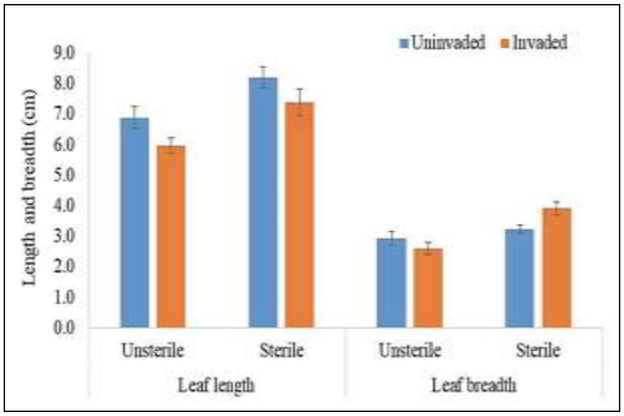

Figure 3. Effect of soil type and sterilization on leaf length and leaf breadth. Bar graph shows the mean value ± standard error.

Root to shoot and leaf length to breadth ratios

Root to shoot ratio of *S. robusta* seedlings showed no variations due to soil type, sterilization and their interaction (P > 0.05, Table 3). Leaf length to breadth ratio varied with soil type (P = 0.002) but not due to sterilization and interaction of soil type and sterilization (P > 0.05, Table 1). The invaded soil reduced the leaf length to breadth ratio in both sterilized and unsterilized soils (Fig. 4).

Table 1. Two-way ANOVA results on the effect of soil type, sterilization and their interaction on *S. robusta* seedling growth parameters

		P values							
	df	**Length**			**breadth**	**Biomass**		**Ratio**	
		Root	**Shoot**	**Leaf**	**Leaf**	**Root**	**Shoot**	**R:S**	**LL:LW**
Soil		0.523	0.377	0.020	0.069	0.046	0.125	0.530	0.002
Sterilization	1	0.050	0.002	<0.001	<0.001	0.329	0.153	0.777	0.344
Soil × sterilization		0.026	0.141	0.894	0.013	0.046	0.466	0.588	0.067

Boldface are the significant values. R : S = root to shoot ratio, LL:LW = leaf length to breadthratio

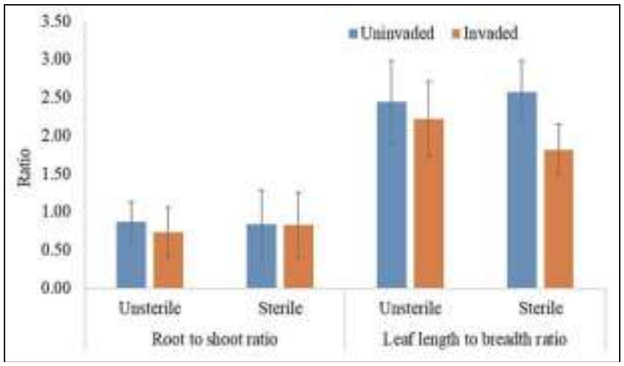

Figure 4. Effect of soil type and sterilization on root to shoot and leaf length to breadth ratios. Bar graph shows the mean value ± standard error.

Discussion

This study analyzed the effect of *C. odorata* invaded soil and its sterilization on growth and development of native *S. robusta* seedlings, comparing the growth parameters with uninvaded and unsterile soils. The toxic effect of invaded unsterile soil is evident in the reduced root biomass and smaller leaf size observed in *S. robusta* seedlings (Fig. 3 and 4). This suggests that the invasive *C. odorata* contributes detrimental residues for native species in its invaded sites. Previous studies have identified several allelochemicals from *C. odorata* such as phenolic and flavonoid compounds that impairs growth and development of native species (Thapa *et al.*, 2021; Budha Magar *et al.*, 2023). Additionally, detrimental impact on native plants is due to alteration in soil qualities and microbial communities (Tondoh *et al.*, 2013; Koné *et al.*, 2021).

Results also showed that there was no significant difference in both root and shoot length due to soil type (invaded and uninvaded) but soil sterilization significantly changed the root and shoot length of *S. robusta* (Table 1). Shoots are likely to be longer in both invaded and uninvaded soil after sterilization, while roots are likely to increase in length, especially in sterilized invaded soil (Fig. 1). Sterilization effect was also found to be significant on the leaf expansion with increased leaf length and breadth under sterilized soils (Fig. 3). As indicated by increased root and shoot length, soil sterilization stimulated root and shoot elongation as well as leaf expansion.

Soil sterilization eliminates both the useful and harmful microbes present in soil. Such negative effects have some consequences on the growing seedling development. Beneficial microbes, such as nitrogen-fixing bacteria or mycorrhizal fungi, help plants access nutrients and act as antagonists (Dellagi *et al.*, 2020). Firstly, if useful microbes are killed, there might be change in nutritional dynamics in soil where microbes contribute potentially reducing nutrient availability in the long term. However, in the short term, seedlings may have taken available nutrients in the sterile soil, as it contains residual nutrients.

Despite the lack of microbial activity on the soil nutrients, the readily available forms of nutrients might have supported the seedlings. Serrasolsas and Khanna (1995) have concluded that the heating soils to 120°C increases extractable nitrogen in the organic and mineral forms.

Secondly, the beneficial microbes like bacteria and fungi compete with plants for nutrients such as nitrogen and phosphorus by reducing the amount accessible to plants (Zhu *et al.*, 2016). Eliminating soil microbes by sterilization might have reduced competition for nutrients (Kuzyakov and Xu, 2013) between *S. robusta* seedlings and microbes. Hence, without microbial interference, the seedlings might have accessed more available nutrients, supporting elongation of root, shoot and expansion of leaves. Thirdly, elimination of harmful microbes supports growth and development of seedlings, enabling plants to make healthy with stronger root system and allocate more energy towards growth rather than defense mechanism (Janvier *et al.*, 2007; Gleeson and Tilman, 1992).

Lastly, microorganisms play a crucial role in minimizing the toxic effects of allelochemicals produced by invasive plants through degradation or transformation (Vidal and Bauman, 1997; Jilani *et al.*, 2008). In such cases, plant growth performance can be expected better in the unsterile soils. However, in some cases, microbial transformation of allelochemicals can produce more toxic compounds for non-native plants (Nair *et al.*, 1990). Therefore, if that is the case, predicting the actual mechanism is difficult without knowing the specific microbes and allelochemicals of *C. odorata* present in the soils.

despite longer roots and shoots in sterile soils, particularly in invaded sterile soils (fig. 1), sterilization did not show a significant change on root and shoot biomass (table 1). it suggests that the sterile soil promote initial growth in terms of root and shoot length as explained above but lack of significant effect on biomass may indicate that the sterile soil condition do not support seedlings to accumulate biomass. nevertheless, elongation of roots, shoots and leaves of seedlings benefits them by improving resource acquisition, enhancing competition and increasing stress tolerance (grossnickle and macdonald, 2018; tajima, 2021).

Therefore, one of the mentionable benefit of having longer shoots and roots and broader leaves in sterile conditions is the improved resource acquisition, especially for light and water. This adaptation of *S. robusta* seedlings can promote early-stage survival and improve plant's resilience in *C. odorata* invaded soils.

Unchanged root to shoot ratio of *S. robusta* seedlings by sterilization and soil type (Table 3) suggests that the seedlings may allocate resources in a similar manner in the conditions where they were grown. However, whether increased leaf length to breadth ratio (longer and narrower leaves) in uninvaded soils or decreased the ratio in invaded sterile soil condition (wider leaves) (Table 3, Fig. 4) benefit the seedlings for light capturing (Poorter and Rozendaal, 2008). Such changes indicate that native plants adapt differently by modifying the shape and size of leaves in response to invasion. Further, presence of soil microbes and available nutrients are other crucial factors that influence these modifications in leaf shape and size.

Conclusion

In conclusion, invaded soil reduces the root biomass, leaf length and leaf breadth of *S. robusta* seedlings. Sterilization of invaded soil stimulates root and shoot elongation, and enhances the leaf length and breadth of the seedlings. It indicates that the sterilization can promote early-stage survival for *S. robusta* seedlings and improve their resilience in *C. odorata* invaded soils. Modification in the leaf size of the seedlings suggest that the native plants adapt differently by altering the shape and size of leaves in response to invasion. Additionally, presence of soil microbes and available nutrients in soil are other factors influencing the growth parameters. Characterization of microbial communities and identifying specific microbes, along with allelochemicals of invasive plants in invaded soils, can help to understand invasion-microbe interactions and native plants' response against the interactions.

Acknowledgements

President Chure Terai Madhesh Conservation Development Board, Government of Nepal is acknowledged for the research grant for the first author.

References

Bhatta, S., L. R. Joshi, and B. B. Shrestha 2020. Distribution and impact of invasive alien plant species in Bardia National Park, western Nepal. *Environmental Conservation*, 47(3): 197-205. https://doi.org/10.1017/S0376892920000223

Budha Magar, A., D. Shrestha, S. Pakka, and K. R. Sharma 2023. Phytochemistry, biological, and toxicity study on aqueous and methanol extracts of *Chromolaena odorata*. *The Scientific World Journal*, 2023(1): 6689271. https://doi.org/10.1155/2023/6689271

Cortois, R., T. Schröder-Georgi, A. Weigelt, W. H. van der Putten, and G. B. De Deyn 2016. Plant–soil feedbacks: role of plant functional group and plant traits. *Journal of Ecology*, 104(6): 1608-1617. https://doi.org/10.1111/1365-2745.12643

De Rouw, A. 1991. The invasion of *Chromolaena odorata* (L.) King & Robinson (ex *Eupatoriumo doratum*), and competition with the native flora, in a rain forest zone, south-west Cote d'Ivoire. *Journal of Biogeography*, 18: 13-23. https://doi.org/10.2307/2845241

Dellagi, A., I. Quillere, and B. Hirel 2020. Beneficial soil-borne bacteria and fungi: a promising way to improve plant nitrogen acquisition. *Journal of Experimental Botany*, 71(15): 4469-4479. https://doi.org/10.1093/jxb/eraa112

Gbètoho, A. J., A. Kingbo, P.B. Kiki, D. Gnanguènon-Guéssè, A. K. Aoudji, K. Koura, S. B. Kakpo, Y. Togni, and J. C. Ganglo 2018. Impacts of *Chromolaena odorata* on native trees' regeneration in the Lama secondary forests in Benin, West Africa. *Bois et Forets des*

Tropiques, 338(4): 5-14. https://doi.org/10.19182/bft2018.338.a31675

Gleeson, S. K. and D. Tilman 1992. Plant allocation and the multiple limitation hypothesis. *The American Naturalist*, 139(6): 1322-1343.

Grossnickle, S. C. and J. E. MacDonald 2018. Why seedlings grow: influence of plant attributes. *New Forests*, 49: 1-34. https://doi.org/10.1007/s11056-017-9606-4

Janvier, C., F. Villeneuve, C. Alabouvette, V. Edel-Hermann, T. Mateille, and C. Steinberg 2007. Soil health through soil disease suppression: which strategy from descriptors to indicators? *Soil Biology and Biochemistry*, 39(1): 1-23. https://doi.org/10.1016/j.soilbio.2006.07 .001

Jilani, G., S. Mahmood, A. N. Chaudhry, I. Hassan, and M. Akram 2008. Allelochemicals: sources, toxicity and microbial transformation in soil - a review. *Annals of Microbiology*, 58: 351-357. https://doi.org/10.1007/BF03175528

Julio, A., W. Carven Tandoc, H. Daniel Tipace, Y. Franzine Vendivil, Z. Yanesa, V. R. Tare, E. Jon Lactaoen, and K. Joseph Clemente 2019. Allelopathic effect of *Lantana camara* and *Chromolaenaodorata* leaf extracts on plant germination. *Asian Journal of Agriculture and Biology*, 7(2): 190-196.

Koné, A. W., S. P. A. Kassi, B. Y. Koffi, D. Masse, A. A. Maïga, J. E. Tondoh, O. M. Kisaka, and G. P. T. Touré 2021. *Chromolaena odorata* (L.) K and R (Asteraceae) invasion effects on soil microbial biomass and activities in a forest-savanna mosaic. *Catena*, 207: 105619. https://doi.org/10.1016/j.catena.2021.105619

Kumar, A., and J. P. Verma 2018. Does plant-microbe interaction confer stress tolerance in plants: a review? *Microbiological Research*, 207: 41-52. https://doi.org/10.1016/j.micres.2017.11.004

Kuzyakov, Y., and X. Xu 2013. Competition between roots and microorganisms for nitrogen: mechanisms and ecological relevance. *New Phytologist*, 198(3): 656-669. https://doi.org/10.1111/ nph.12235

Nair, M. G., C. J. Whitenack, and A. R. Putnam 1990. 2, 2′-OXO-1, 1′-azobenzene a microbially transformed allelochemical from 2, 3-benzoxazolinone: I. *Journal of Chemical Ecology*, 16: 353-364. https://doi.org/10.1007/BF01021770

Poorter, L., and D. M. Rozendaal 2008. Leaf size and leaf display of thirty-eight tropical tree species. *Oecologia*, 158: 35-46. https://doi.org/10.1007/s00442-008-1131-x

Poudel, S., R. R. Pant, L. B. Thapa, and M. K. Chettri 2024. Impacts of invasive weed *Chromolaena odorata* on growth and development of threatened native tree *Aegle marmelos* under water stress conditions. *Journal of Ecology and Environment*, 48: 42. https://doi.org/10.5141/jee.24.072

R Core Team 2023. *R: A Language and Environment for Statistical Computing*. Version 4.3.1, R Foundation for Statistical Computing, 2023. www.R-project.org.

Rai, P. K. 2015. What makes the plant invasion possible? Paradigm of invasion mechanisms, theories and attributes. *Environmental Skeptics and Critics*, 4(2): 36.

Rusdy, M. 2016. The spread, impact and control of *Chromolaena odorata* (L.) RM King and H. Robinson in grassland area. *Journal of Agriculture and Ecology Research International*, 5(1): 1-8. https://doi.org/10.9734/ JAERI/2016/16391

Serrasolsas, I., and P. K. Khanna 1995. Changes in heated and autoclaved forest soils of SE Australia. I. Carbon and nitrogen. *Biogeochemistry*, 29: 3-24. https://doi.org/10. 1007/BF00002591

Shrestha, B. B. 2019. Management of Invasive Alien Plants in Nepal: Current Practices and Future Prospects. In *Tropical Ecosystems: Structure, Functions and Challenges in the Face of Global Change* (Eds. S. Garkoti, S. Van Bloem, P. Fulé, and R. Semwal). Springer, Singapore. pp. 45-68. https://doi.org/10.1007/978-981-13-8249-9_4

Tajima, R. 2021. Importance of individual root traits to understand crop root system in agronomic and environmental contexts. *Breeding Science*, 71(1): 13-19. https://doi.org/10.1270/jsbbs.20095

Thapa, L. B., K. Kaewchumnong, A. Sinkkonen, and K. Sridith 2016. Impacts of invasive *Chromolaena odorata* on species richness, composition and seedling recruitment of *Shorea robusta* in a tropical Sal forest, Nepal. *Songklanakarin Journal of Science and Technology*, 38(6): 683-689. https://doi.org/10.14 456/sjst-psu.2016.86

Thapa, L. B., S. Pathak, K. B. Pal, T. M. Miya, T. B. Darji, G. Pant, and R. R. Pant 2021. Chemical constituents of the essential oil of invasive *Chromolaena odorata* leaves in Central Nepal. *Journal of Nepal Chemical Society*, 42(1): 132-137. https://doi.org/10.3126/jncs.v42i1.35364

Tiébré, M. S., and Z. R. G. Gnanazan 2018. Impact of *Chromolaena odorata* (L.) RM King & H. Rob. (Asteraceae) on the floristic composition and the physico-chemical properties of the soil of a coastal relict forest. *International Journal of Innovation and Applied Studies*, 24(2): 773-788.

Tondoh, J. E., A. W. Koné, J. K. N'Dri, L. Tamene, and D. Brunet 2013. Changes in soil quality after subsequent establishment of *Chromolaena odorata* fallows in humid savannahs, Ivory Coast. *Catena*, 101: 99-107. https://doi.org/10.1016/j.catena.2012.10.005

Uwalaka, N. O., and J. I. Muoghalu 2021. Ecological impact of *Chromolaena odorata* (L.) king and Robinson on native plant communities and soil seed bank in Ile-Ife, Nigeria. *Afr. J. Ecol.* 59(2): 572–581. https://doi.org/10.1111/aje.12848

Vidal, R. A., and T. T. Bauman 1997. Fate of allelochemicals in the soil. *Ciência Rural*, 27: 351-357. https://doi.org/10.1590/S0103-84781997000200032

Zachariades, C., M. Day, R. Muniappan, and G. V. P. Reddy 2009. *Chromolaena odorata* (L.) King and Robinson (Asteraceae). In *Biological control of tropical weeds using arthropods* (Eds. R. Muniappan, G. V. Reddy, and A. Raman). Cambridge University Press, Cambridge pp. 130-162.

Zhang, M. Z., W. T. Li, W. J. Liu, and Y. L. Zheng 2024. Rhizosphere microbial community construction during the latitudinal spread of the invader *Chromolaena odorata*. *BMC Microbiology*, 24(1): 294. https://doi.org/10.1186/s12866-024-03450-x

Zhu, Q., W. J. Riley, J. Tang, and C. D. Koven 2016. Multiple soil nutrient competition between plants, microbes, and mineral surfaces: model development, parameterization, and example applications in several tropical forests. *Biogeosciences*, 13(1): 341-363. https://doi.org/ 10.5194/ bg-13-341-2016