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PHYTOTOXIC POTENTIAL OF BARK EXTRACTS OF *ACACIA NILOTICA* AND *SYZYGIUM CUMINI* AGAINST *PARTHENIUM HYSTEROPHORUS*

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Abstract

Effect of aqueous and n-hexane bark extracts of allelopathic tree species viz., *Acacia nilotica* and *Syzygium cumini* (L.) Skeels was studied on germination and seedling growth of *Parthenium hysterophorus* L. In laboratory trials, all the concentrations of aqueous extract of both the test plants increased seed germination of target weed while by employing n-hexane concentrations of *A. nilotica* the number of germinated seeds remained the same as in control except 20%. However, among all the concentrations of n-hexane extract of *S. cumini,* significantly minimum seed germination (about 50%) was recorded at 20% concentration. There was a negative phytotoxic response on weed growth by aqueous extracts of both test plants. Conversely a pronounced effect on plant growth (shoot and root length) was exhibited by n-hexane extract of test plants. Similarly, the biomass was significantly reduced by both aqueous and n-hexane extracts of both test plants. In pot trials, all the n-hexane concentrations of test plants invariably suppressed the root and shoot growth of target weed. There was 30-35%, 20-27%, 50% and 50-55%, 80% and 80-82% reduction in shoot length, root length and fresh/dry biomass of parthenium by n-hexane extracts of *A. nilotica* and *S. cumini*, respectively.

Introduction

Parthenium hysterophorus, an alien invasive species, is annual ephemeral herb. In Pakistan, this weed is spreading aggressively in wastelands, degraded areas, rocky crevices, along water channels, road sides and railway tracks. It has also been reported in cultivated lands (Shabbir, 2002). This noxious weed affects crop production, animal husbandry, human health and biodiversity.

Among various weed control measures, herbicides are widely preferred by farmers for the control of *P. hysterophorus* due to cost- and time-effectiveness. However, the increased use of herbicides poses serious environmental and public health concerns (Mancini *et al*., 2008). Moreover, the evolution of resistance to several herbicides, including atrazine, fenoxaprop-p, flupyrsulfuron, isoproturon, and triazine, has been reported (Malik & Singh, 1995; Koeppe *et al*., 1997, Chhokar *et al.,* 2008). Therefore, alternative weed management systems are required which are based on naturally occurring compounds (Cuthbertson & Murchie, 2005). Thus plant derivatives are more environmentally safe than synthetic chemicals (Hashim & Devi, 2003). During the past two decades, much work has been done on plant-derived compounds as environmentally safe alternatives to herbicides for weed control (Duke *et al.,* 2002; Javaid *et al*., 2008a; Shafique *et al*., 2011). The present study, therefore, describes the phytotoxic potential of *A. nilotica* and *S. cumini* in controlling the wasteland weed *P. hysterophorus*.

Materials and Methods

Preparation of extracts: Fresh barks of *A. nilotica* and *S. cumini* were collected from University of the Punjab, Quaid-e-Azam Campus Lahore, Pakistan. The materials were thoroughly washed with tap water followed by washing with sterilized water and oven dried at 50°C. The dried material was crushed in pestle and mortar and soaked in distilled water and n-hexane @ 20 g 100 mL^{-1}. It was left for 2 days at 25°C. Afterward, extracts were filtered through muslin cloth followed by Whatman filter paper No. 1. Volume of organic solvent was reduced to 1 mL by evaporation at 40 ºC in continuous current of air and then diluted by adding appropriate quantity of sterilized distilled water to make the final volume 100 mL. Stock extracts were stored at 4ºC. In order to make n-hexane control solution, 1 mL of organic solvent was added to sterilized distilled water to make final volume 100 mL. Subsequently, appropriate quantity of distilled water was added to the stock solutions to get 5, 10 &15% (v/v) concentrations of different extracts (Javaid *et al.,* 2008b).

Laboratory bioassays: In a laboratory bioassay, the effect of different concentrations of 5–20% of aqueous and organic solvent extracts of *A. nilotica* and *S. cumini* was studied on germination and early seedling growth of parthenium. Ten seeds of parthenium were placed in 9-cm diameter Petri plates lined with Whatman No. 1 filter papers moistened with 3 mL of different concentrations of each extract. The respective control treatments for aqueous and n-hexane extracts received the same quantities of distilled water and n-hexane control solution, respectively. Each treatment was replicated thrice. Plates were incubated at 25 °C under 12 h light periods daily. After 7 days, seed germination, seedling root/shoot length and fresh biomass were determined.

Foliar spray bioassay: Parthenium seeds were sown in pots of 14 cm diameter and 18 cm deep each containing 4 Kg of sandy loam soil. Initially 10 seeds were sown in each pot, which were thinned to 3 uniform seedlings one week after germination and were further thinned to one at the time of photography. Only n-hexane extract was prepared as for foliar spray bioassays. Freshly prepared extracts were sprayed on the surface of 15 days old Parthenium plants with a hand sprayer. Three subsequent sprays were similarly carried out with 5 days intervals each. Control plants were similarly sprayed with water. Plants were harvested 5 days after last spray. Data regarding length and fresh biomass of both root and shoot were recorded.

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Statistical analysis: Data regarding germination, root/shoot length and plant fresh and dry weight were subjected to analysis of variance (ANOVA) followed by Duncan's Multiple Range Test to delineate mean differences (Steel & Torrie, 1980) using computer software COSTAT.

Results

Laboratory bioassays: An experiment comprising the evaluation of phytotoxic effect of *A. nilotica* and *S. cumini* on germination and growth response of Parthenium was carried out and data were recorded. The results of the study reveal that all the concentrations of aqueous extracts of bark of *A. nilotica* and *S. cumini* exhibited an erratic pattern of increase in germination as well as seedling growth of parthenium (Fig. 1 A-C). Whereas the highest concentration (20%) of *A. nilotica* significantly reduced plant growth, up to 50-60%, in terms of shoot and root length (Figs. 1B & C). In contrast to that, biomass of Parthenium was markedly reduced by all the concentrations of *A. nilotica*. (Fig. 1D). In general *S. cumini* proved ineffective in suppressing the growth of parthenium.

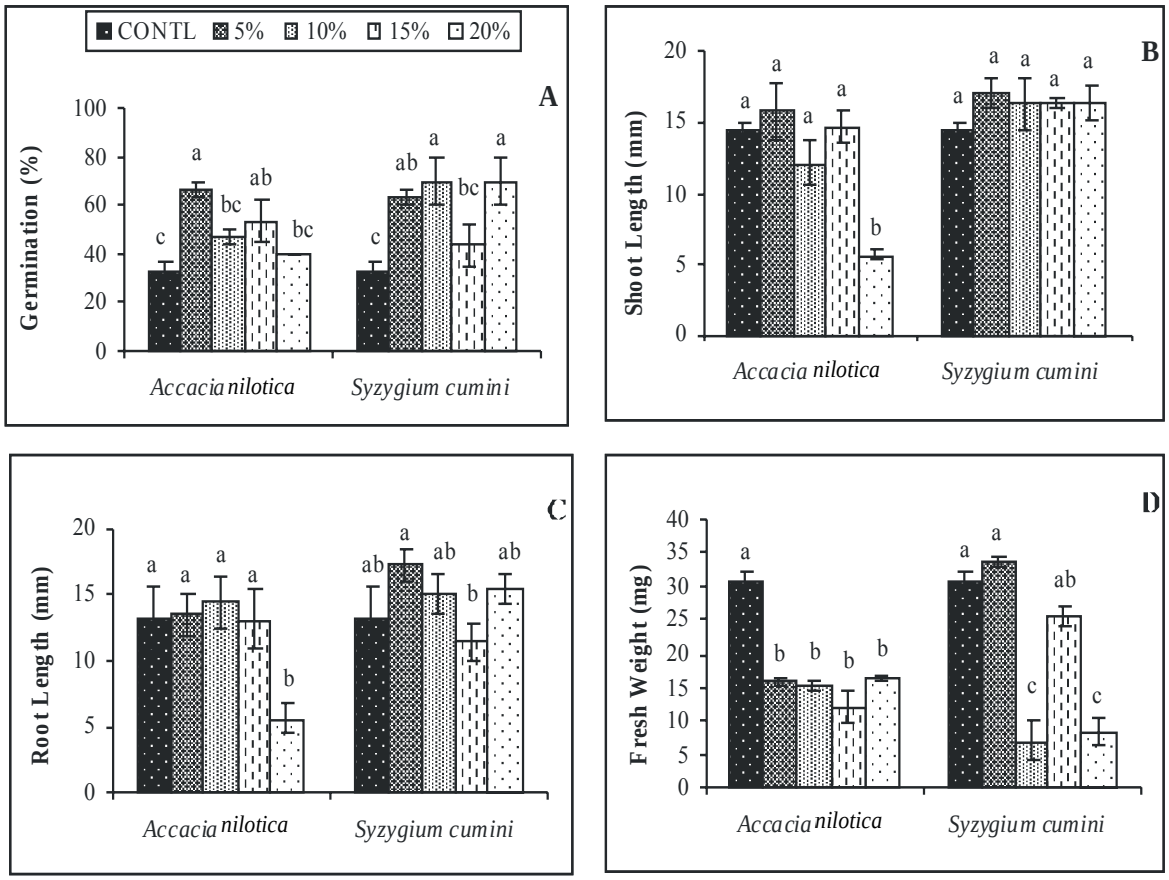

Fig. 1. Effect of different concentrations of aqueous extract of *Acacia* and *Syzygium cumini* on germination and seedling growth of *Parthenium* in laboratory bioassays.
Vertical bars show standard errors of means of three replicates. Values with different letters at their top show significant difference ($p \leq 0.05$) as determined by Duncan's Multiple Range Test.

On the other hand, all the n-hexane extract concentrations significantly reduced the germination and seedling growth except concentrations of *A. nilotica* which depicted no effect on germination (Fig. 2 A). Over all, n-hexane extract of *A. nilotica* was more toxic than *S. cumini* in reducing shoot and root length (Fig. 2 B-C). However, biomass was found to be the most susceptible to all the employed concentrations of *A. nilotica* and *S. cumini* (Fig. 2 D).

The comparative analysis of aqueous and n-hexane extracts of *A. nilotica* and *S. cumini* reveal the significant phytotoxic potential of n-hexane extracts over the aqueous extracts so was selected for subsequent studies.

Foliar spray bioassay: Data regarding the effect of foliar spray of n-hexane extracts of bark of *A. nilotica* and *S. cumini* on shoot and root growth of parthenium plants is presented in Figs. 3 and 4. All the concentrations of both test plant extracts significantly suppressed shoot (30-35% by *A. nilotica* and 20-27% by *S. cumini*) as well as root length (approx. 50% by *A. nilotica* and 50-55% by *S. cumini*) of parthenium plants (Figs. 3 A & B). Likewise, all the employed extract types significantly reduced (about 80%) the shoot and root fresh as well as dry biomass of parthenium whereas there was not any pronounced difference between effectiveness of different concentrations of extracts of employed test plants (Figs. 3 C-F).

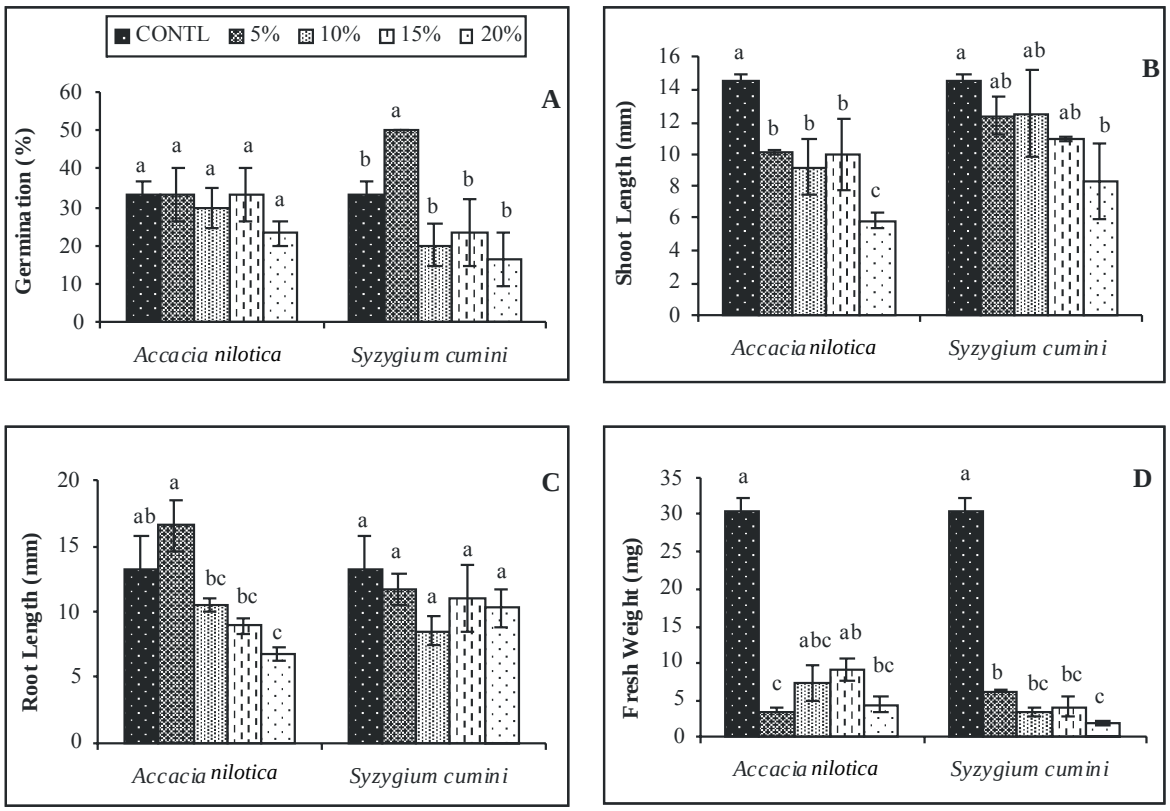

Fig. 2. Effect of different concentrations of n-hexane extract of *Acacia nilotica* and *Syzygium cumini* on germination and seedling growth of *Parthenium* in laboratory bioassays.
Vertical bars show standard errors of means of three replicates. Values with different letters at their top show significant difference ($p \leq 0.05$) as determined by Duncan's Multiple Range Test.

It is concluded from the results that the target weed parthenium was highly susceptible to foliar sprays of both the bark extracts of test plants *A. nilotica* and *S. cumini*.

Discussion

Aqueous bark extract of *A. nilotica* at different concentrations significantly enhanced germination, shoot and root length except 20% concentration which significantly declined shoot and root length. In general *S. cumini* proved unproductive in suppressing the growth of Parthenium. Earlier studies also show that lower concentrations of plant extract may be stimulatory to the test plant growth (Reigosa *et al*., 1999; Javaid & Anjum, 2006; Javaid *et al*., 2006b). Conversely, both extract types significantly reduced plant biomass as compared to control. These results corroborate to those of Khan *et al*., (2008) who reported reduction in dry weight of plants by *Eucalyptus* application. In general, n-hexane extract was more effective in suppressing the growth of Parthenium as compared to aqueous extract. This variation in herbicidal activity of the extracts in different solvents may be attributed to the different chemical nature of the solvents. Water is polar while n-hexane is non-polar in nature. The variation in activity of extracts of same part of the plant is attributed to different types of chemicals extracted in different solvents (Javaid *et al.,* 2008a).

Foliar sprays revealed that n-hexane extracts of both the test plants were highly toxic to germinating parthenium. Root length was more sensitive to n-hexane extract of both the test plants than shoot length where all the employed extract concentrations significantly suppressed root length. Pronounced sensitivity of root growth to the plant extracts have also been recorded in other plant species (Afzal *et al*., 2000; Javaid *et al*., 2008a; 2008b). Since roots are the first to absorb chemical compounds from the environment, so exhibit abnormal growth in response to chemicals present in the extracts, resulting in suppressive growth (Javaid & Shah, 2007). The extract of *A. nilotica* is known to contain gallic acid, m-digallic acid, catechin, chlorogenic acid, gallolyated flaven-3, 4-diol and rabidandiol (Malan, 1991). The inhibitory effect of *Acacia* species on seed germination and seedlings was related to the presence of allelochemical including tannins, wax, flavonoides and phenolic acids. Furthermore, the toxicity might cause due to synergistic effect of rather than single. Benherlal & Arumughan (2007) reported that the leaves, stems, flower buds, opened blossoms, and bark of *S. cumini* have some antibiotic activity. Extract from *S. cumini* consists mainly of mono- or sesqui-terpene hydrocarbons which are very common in essential oils and the bark is also reported to contain 8 to 19% tannin. The reduction in the growth rate of Parthenium could be attributed to the presence of such compounds in different extracts.

Further the studies can be extended regarding the efficacy of these crude extracts under field conditions and isolation and identification of allelochemicals responsible for germination and growth reduction of parthenium are requisite.

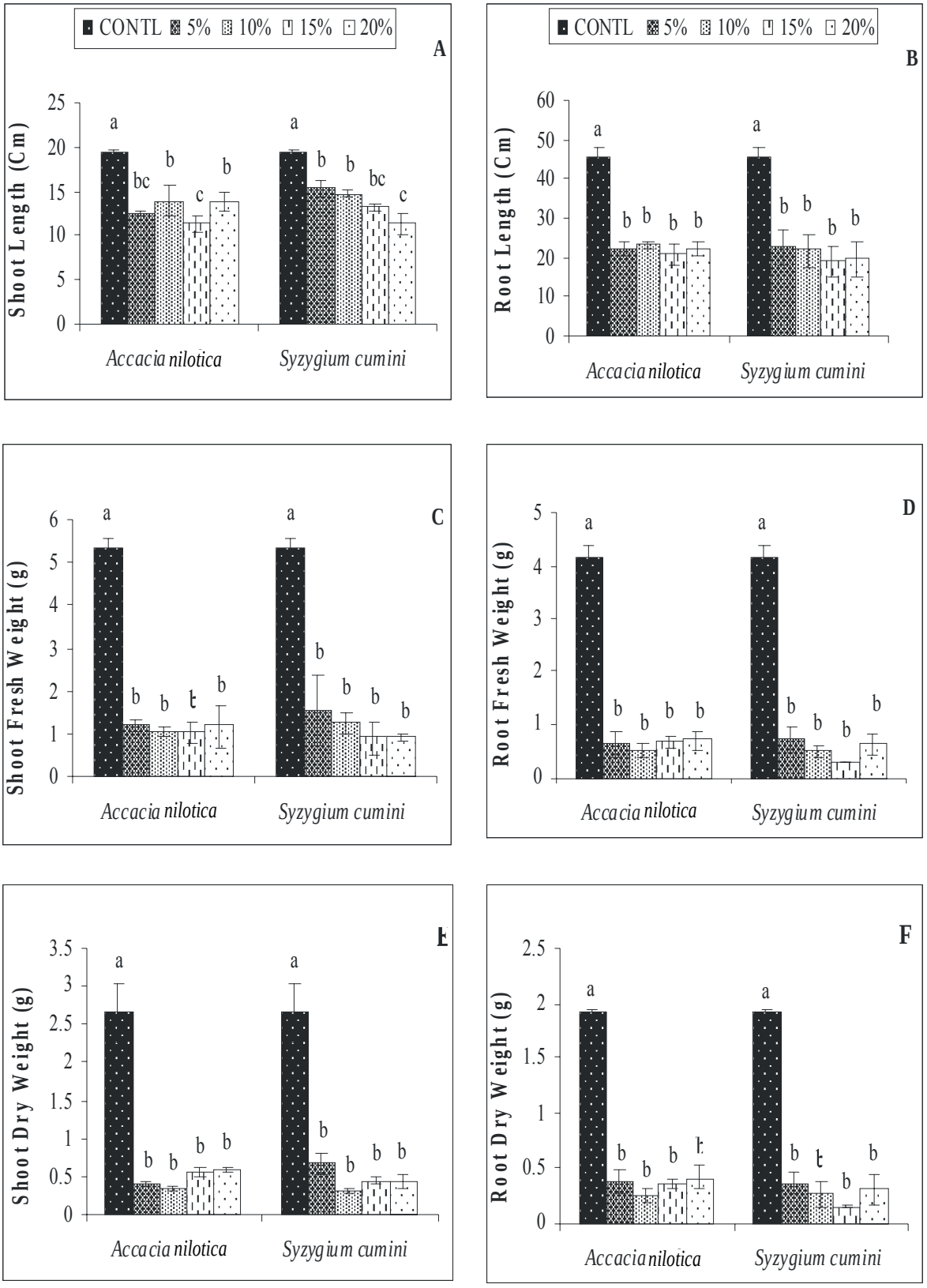

Fig. 3. Effect of different concentrations of n-hexane extract of *Acacia nilotica* and *Syzygium cumini* on growth of *Parthenium* in pot bioassays.

Vertical bars show standard errors of means of three replicates. Values with different letters at their top show significant difference ($p \leq 0.05$) as determined by Duncan's Multiple Range Test.

Fig. 4. Effect of foliar spray of n-hexane extract of bark of *Acacia nilotica* (A) and *Syzygium cumini* (B) on growth of *Parthenium hysterophorus* in pot trials.

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