

Full Length Research Paper

Phytotoxic characterization of various fractions of *Launaea procumbens*

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Allelopathic screening of various fractions of *Launaea procumbens*, collected from Wah Cantt (Punjab) Pakistan, was conceded to identify potent allelopathic fraction for future phytochemical analyses. For this purpose, radish root inhibition method was used to test allelopathic potential. Two different concentrations of 100 ppm and 1000 ppm of *Launaea procumbens* were used in this study. Methanolic and ethyl acetate fraction potently inhibited root and radical growth; comparative to other fractions, it might be due to the presence of bioactive allelochemicals. Based on this screening, both of these fractions were recommended for future bioassay guided isolation of allelochemicals.

Key words: *Launaea procumbens*, radish seeds, root inhibition, *Launaea procumbens* methanolic fraction.

INTRODUCTION

Every year, about 13% of the world's crops have been lost due to damages caused by weeds. The progress of weed control technology such as transgenic crops and synthetic herbicides has made a great input to the enhancement of crop yields through the years (Bridges, 1994). Allelopathy means the inhibition of plant growth through the production of phytochemicals/phytotoxins released by another plant. This incident represents competition between neighboring plants for light, water, and nutrients (Inderjit, 2003). There are several crop plants in which allelopathic effects have been observed. These include rice, wheat, oats, reddish seed, sunflower, barley, and sorghum, with rice being the most studied case. Medicinal plants plays a crucial role in improving various pathogenesis (Sahreen et al., 2010; Khan et al., 2009; Khan et al., 2010a,b) as well as allelopotency (Khan et al., 2010c). Therefore, extensive research has

been conducted to develop allelopathic compounds which improve the production of agricultural ecosystems through different ways, including using the natural products from plants as pesticides instead of synthetic chemicals. Medicinal plants are screened for their allelopathic and or medicinal potentials and to select the most bioactive ones for chemical analyses (Fujii et al., 2003; Khan et al., 2010c).

Launaea procumbens is traditionally used in the treatment of rheumatism (Parekh and Chanda, 2006), kidney and liver dysfunctions (Ahmad et al., 2006; Qureshi and Raza, 2008), eye diseases and as food (Wazir et al., 2007). Nutritional analysis showed that *L. procumbens* is composed of salicylic acid, vanillic acid, synergic acid, 2-methyl-resercinol and gallic acid (Shaukat et al., 2003) which have antioxidant, liver disorders, anticancer, allelopathic and anti-inflammatory properties. Therefore, this study was established to evaluate the allelopathic properties of the various fractions of *L. procumbens* versus the radish root and seedling growth under controlled conditions.

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Abbreviations: DMSO, Dimethyl sulfoxide; LPME, *Launaea procumbens* methanolic extract; LPHE, *Launaea procumbens* n-hexane extract; LPEE, *Launaea procumbens* ethyl acetate extract; LPCE, *Launaea procumbens* chloroform extract; LPBE, *Launaea procumbens* butanolic extract; LPWE, *Launaea procumbens* water extract.

MATERIALS AND METHODS

Plant collection and extraction

Launaea procumbens was collected at maturity from Wah Cantt District Rawalpindi, Pakistan. It was identified and its ariel parts

Root inhibition (mm)

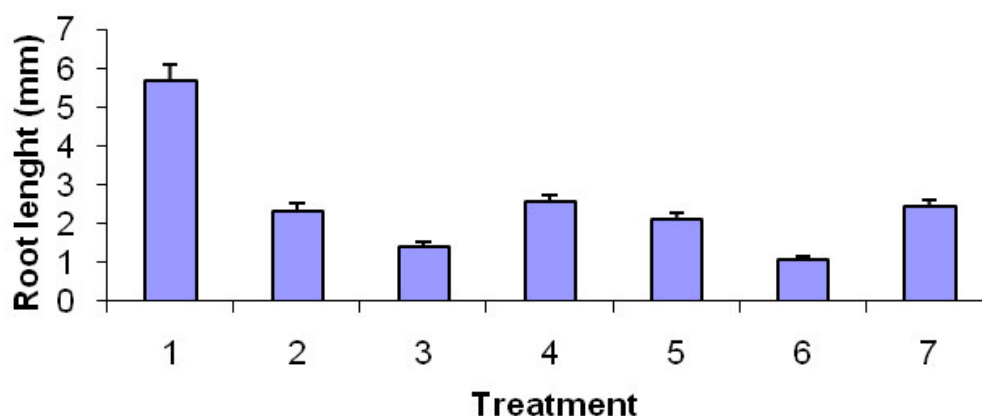


Figure 1a. Representation of the root inhibition at 100 ppm concentration of various fractions of *Launaea procumbens*. 1, non treated control; 2, n-hexane fraction; 3, ethyl acetate fraction; 4, chloroform fraction; 5, butanol fraction; 6, methanol fraction; 7, water fraction.

(leaves, stem, flowers and seeds) were shade, dried at room temperature, grinded mechanically and extracted with methanol to get crude methanolic extract. Crude methanolic extract was further fractionated using n-hexane, ethyl acetate, chloroform, butanol and distilled water with increasing order of polarity. Each fraction was evaporated through rotary evaporator and screened for phytotoxic potency.

Phytotoxicity bioassay

This test was performed according to the modified protocol of McLaughlin and Rogers, (1998). Various fractions of *L. procumbens* were incur-porated at different concentrations; that is, 100, 1000 ppm in respective solvents in replicates. Radish seed was washed with distilled water (dH₂O) and with 1% mercuric chloride. Filter paper was put in each autoclaved petri plates. 5 ml of each fraction was poured in each plate and the respective solvent was evaporated. 10 seeds were placed in each plate and incubated in a growth room for five days. After 5 days, the root and shoot inhibition was noted. Fresh and dry weight was also recorded.

RESULTS

Phytotoxicity assessment

Root growth inhibition

Phytotoxic (allelopathic) effects of *Launaea procumbens* were evaluated against radish seed growth under controlled environmental condition in the growth room. Radish root inhibition was shown in Figures 1a and b. *Launaea procumbens* methanolic extract (LPME) and *Launaea procumbens* ethyl acetate extract (LPEE) showed significant ($P < 0.01$) inhibition comparative to the control of non treated seed root growth at the fifth

day of the treatment; both at 100 and 1000 ppm respectively.

Shoot growth inhibition

L. procumbens also effected the inhibition of shoot growth. Data revealed that methanolic as well ethyl acetate fractions have marked significance ($P < 0.01$) on the growth of radish seeds at the fifth day of the treatment, both at 100 and 1000 ppm respectively as shown in Figures 2a and b.

Effect of the fractions on fresh and dry weight

At the fifth day of the treatment, fresh weight was calculated and it was observed that, methanolic and ethyl acetate fractions of *L. procumbens* significantly reduced ($P < 0.01$) the weight; confirming the presence of the bioactive allelochemicals in the fractions. At the end of the experiment, the mass of the experimental seedlings were dried under controlled condition and it was found that, the weights of LPME and LPEE group were effectively less ($P < 0.01$); comparative to the non treated controlled group (Table 1).

DISCUSSION

The results of these screening assays justified the use of the investigated plants against herbicides in Pakistan. The phytotoxic results of all the fractions of *Launaea procumbens* showed that, the growth of radish root as

Root inhibition (mm)

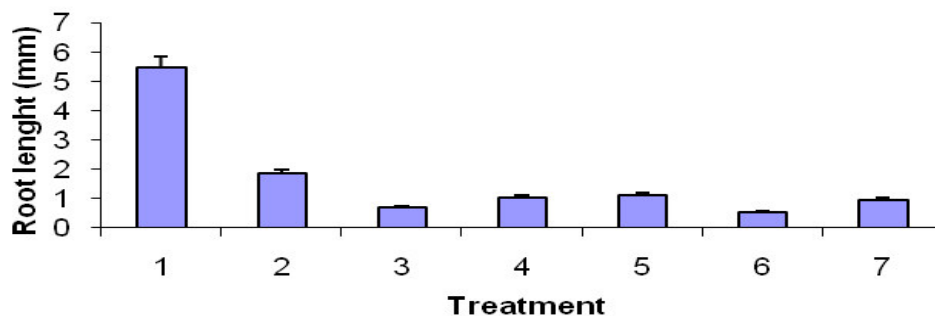


Figure 1b. Representation of the root inhibition at 1000 ppm concentration of various fractions of *Launaea procumbens*. 1, non treated control; 2, n-hexane fraction; 3, ethyl acetate fraction; 4, chloroform fraction; 5, butanol fraction; 6, methanol fraction; 7, water fraction.

Shoot inhibition (mm)

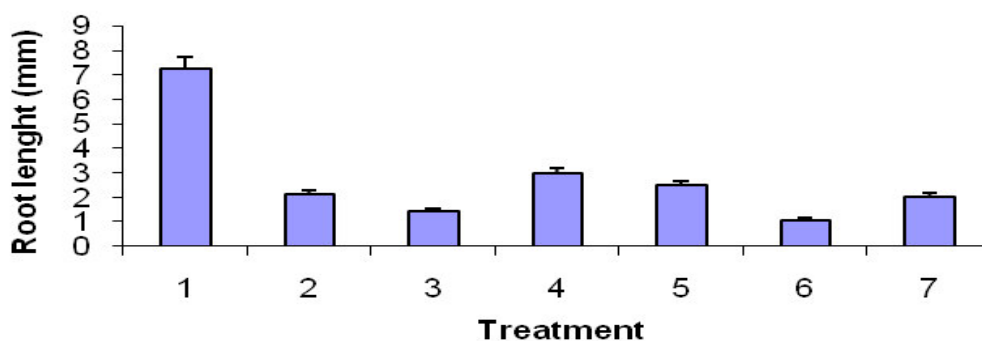


Figure 2a. Representation of the shoot inhibition at 100 ppm concentration of various fractions of *Launaea procumbens*. 1, non treated control; 2, n-hexane fraction; 3, ethyl acetate fraction; 4, chloroform fraction; 5, butanol fraction; 6, methanol fraction; 7, water fraction.

shoot inhibition (mm)

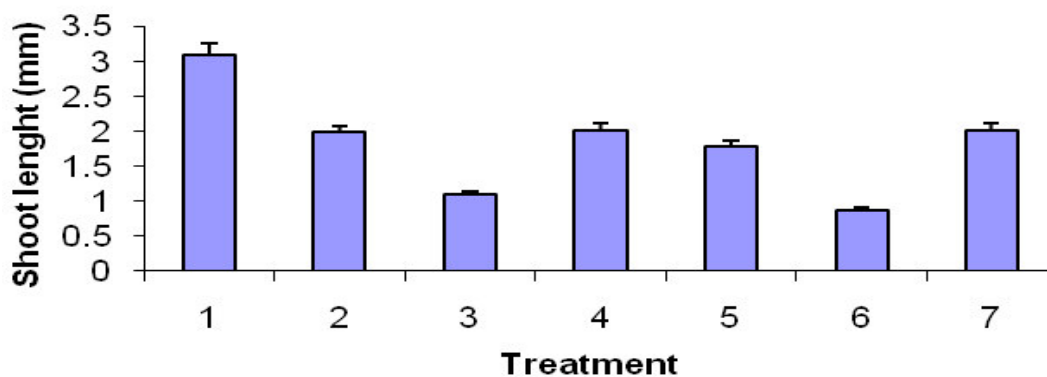


Figure 2b. Representation of the shoot inhibition at 1000 ppm concentration of various fractions of *Launaea procumbens*. 1, non treated control; 2, n-hexane fraction; 3, ethyl acetate fraction; 4, chloroform fraction; 5, butanol fraction; 6, methanol fraction; 7, water fraction.

Table 1. The fresh and dry weight of various fractions of *Launaea procumbens*.

Treatment	Fresh weight (g)		Dry weight (g)	
	100 ppm	1000 ppm	100 ppm	1000 ppm
Control	5.67 ± 0.6a	5.45 ± 0.2a	7.23 ± 0.4a	3.09 ± 0.02a
LPHE	2.34 ± 0.2b	1.87 ± 0.05b	2.12 ± 0.02b	1.98 ± 0.09b
LPEE	1.41 ± 0.02c	0.69 ± 0.001c	1.45 ± 0.006c	1.09 ± 0.001c
LPCE	2.56 ± 0.1b	1.02 ± 0.01b	3.09 ± 0.1b	2.01 ± 0.1b
LPBE	2.12 ± 0.5b	1.1 ± 0.2b	2.54 ± 0.2b	1.78 ± 0.3b
LPME	1.09 ± 0.06c	0.54 ± 0.002c	1.08 ± 0.003c	0.87 ± 0.001c
LPWE	2.45 ± 0.1b	0.97 ± 0.3b	2.02 ± 0.1b	2.01 ± 0.12b

Each value in the table is represented as mean ± SD ($n = 3$). Means not sharing the same letter are significantly different (LSD) at $P < 0.01$ probability level in each column. LPHE (n-hexane fraction), LPEE (ethyl acetate fraction), LPCE (chloroform fraction), LPBE (butanol fraction), LPME (methanol fraction), LPWE (water fraction).

well as the shoot was inhibited when compared to the non treated controlled plant. Water, methanolic and butanolic fractions showed marked growth inhibition of root and shoot while n-hexane and ethyl acetate fraction of both plants showed moderate effects. These findings showed similarity with the results reported by Javaid (2009) that, water extract of *Withania somnifera* and *Datura alba* possessed some bioactive compounds which significantly inhibited the growth of root and shoot of *Rumex dentatus* L.; highly competitive weed in wheat during allelopathic screening. Similar investigations were found by Kordali et al. (2008) that, essential oil isolated from Turkish *Origanum acutidens* and their phenolic compounds completely inhibited the growth of seedlings and roots, and possessed antifungal activity when compared to the standard compounds. Other investigations of Khan et al. (2010c) showed that, methanolic and butanolic fractions of *Sonchus asper* significantly controlled the shoot and root growth of radish seeds under controlled environmental condition which strongly supports these investigations. The investigation of Hussain et al. (2010) was also in accordance to the results obtained during the Phytotoxic screening of some selected medicinal plants of the family Polygonaceae.

Conclusion

From these data, it was inferred that, *Launaea procumbens* methanolic and ethyl acetate fractions have significant herbicidal potency, which might be the presence of allelochemicals. Therefore, further study on the isolation and purification of these allelochemicals are suggested.

REFERENCES

- Ahmad M, Khan MA, Manzoor S, Zafar M, Sultana S (2006). Check list of medicinal flora of Tehsil Isakhel, District Mianwali Pakistan. *Ethnobot. Leaflets*, 10: 41-48.
- Bridges DC (1994). Impact of weeds on human endeavours. *Weed Technol.* 8: 392-395.
- Fujii Y, Parvez SS, Parvez MM, Ohmae Y, Iida O (2003). Screening of 239 medicinal plant species for allelopathic activity using sandwich method. *Weed Biol. Manage.* 3: 233-241
- Hussain F, Hameed I, Dastagir G, Shams-un-Nisa, Khan I, Ahmad B (2010). Cytotoxicity and phytotoxicity of some selected medicinal plants of the family Polygonaceae. *Afr. J. Biotechnol.* 9(5): 770-774.
- Inderjit DSO (2003). Ecophysiological aspects of allelopathy. *Planta*, 217: 529-539.
- Javaid A (2009). Role of Effective Microorganisms in Sustainable Agricultural Productivity. In: *Advances in Sustainable Agriculture*. Springer Publishers. (in Press).
- Khan MR, Rizvi W, Khan GN, Khan RA, Shaheen S (2009). Carbon tetrachloride induced nephrotoxicity in rats: Protective role of *Digera muricata*. *J. Ethnopharmacol.* doi:10.1016/j.jep.2008.12.006.
- Khan RA, Khan MR, Sahreen S (2010a). Evaluation of *Launaea procumbens* use in renal disorders: A rat model. *J. Ethnopharmacol.* 128: 452-461.
- Khan RA, Khan MR, Sahreen S, Bokhari J (2010b). Prevention of CCl₄-induced nephrotoxicity with *Sonchus asper* in rat. *Food Chem. Toxicol.* doi:10.1016/j.fct.2010.06.016
- Khan RA, Khan MR, Sahreen S, Bokhari J (2010c). Antimicrobial and Phytotoxic screening of various fractions of *Sonchus asper*. *Afr. J. Biotechnol.* 9(25): 3883-3887.
- McLaughlin JL, Rogers LL (1998). The use of biological assays to evaluate botanicals. *Drug Inform. J.* 32: 513-524.
- Parekh J, Chanda S (2006). Screening of aqueous and alcoholic extracts of some Indian medicinal plants for antibacterial activity. *Indian J. Pharm. Sci.* 68: 835-838.
- Qureshi R, Raza BG (2008). Ethnobotany of plants used by the Thari people of Nara Desert, Pakistan. *J. Fitoter.* 79: 468-473.
- Sahreen S, Khan M R, Khan RA (2010). Evaluation of antioxidant activities of various solvent extracts of *Carissa opaca* fruits. *Food Chem.* 22: 1205-1211.