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HERBICIDAL ACTIVITY OF *Datura metel* L. AGAINST *Phalaris minor* Retz.

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ABSTRACT

Present study was undertaken to investigate the herbicidal potential of aqueous and organic solvent (methanol and n-hexane) extracts of shoot and root of Datura metel L. (Syn, Datura alba Nees.) against Phalaris minor Retz., one of the most problematic weeds of wheat in Pakistan. In laboratory bioassays, aqueous, methanol and n-hexane extracts of 0, 5, 10 and 15% (w/v) were applied. Extracts in different solvents exhibited markedly variable herbicidal activities against the target weed species. Germination was significantly reduced by methanol root extract of 5-15% concentrations. Similarly, 15% n-hexane root extract also significantly decreased germination. Root length was significantly suppressed by all the employed concentrations of aqueous as well as organic solvent extracts. Shoot length was significantly declined by all the concentrations of methanol and n-hexane root extracts. The highest concentration of 15% of aqueous and methanol shoot extracts also reduced shoot length significantly. Aqueous shoot extracts were proved highly toxic to plant biomass where all the employed extract concentrations significantly reduced biomass.. The present study concludes that root and shoot extracts of D. metel contain herbicidal constituents. Further studies are suggested to isolate and identify the effective herbicidal constituents from D. metel which can be used as structural lead for the preparation of natural product based herbicide for the management of P. minor.

Key words: Aqueous extracts, *Datura metel*, herbicidal, organic solvents, *Phalaris minor.*

INTRODUCTION

Littleseed canarygrass (*Phalaris minor* Retz.), a native of the Mediterranean region, has spread to many parts of the world (Singh *et al.*, 1999). It has been identified as a serious weed of wheat and barley fields in India, USA, Canada, Africa, Australia, France, Pakistan, Iran, Iraq and Mexico (Holm *et al.*, 1979). It became aggravated weed due to the introduction of high-yielding dwarf wheat varieties where irrigation and fertilizers inputs were favorable (Chhokar *et al.*, 2008). Dwarf varieties, being less competitive with weeds, provide conditions conducive to growth and development of *P. minor*. Furthermore, a rice–wheat crop rotation also stimulated its emergence, growth and development (Chhokar and Malik, 1999). Weed seeds of many weed species may be killed during the summer or by rainy season flooding under rice cultivation, *P. minor* remains unaffected due to an impermeable seed coat. The weed is highly competitive and can cause significant yield losses under favourable conditions. Depending upon density of *P. minor*, 10-65% yield losses in wheat have been reported by various workers (Mehra and Gill, 1988; Dhaliwal *et al.*, 1997; Dhima and Eleftherohorinos, 2003; Chhokar *et al.*, 2008).

Due to morphological similarity of *P. minor* with wheat, manual weeding at early stages is difficult. Among various weed control measures, herbicides are widely preferred by wheat farmers for the control of *P. minor* due to cost- and time-effectiveness (Qureshi *et al.*, 2002; Bibi *et al.*, 2005; Marwat *et al.*, 2005). However, the increased use of herbicides poses serious environmental and public health concerns (Mancini *et al*., 2008). Increasing public concern on environmental issues requires alternative weed management systems, which are less pesticide dependant or based on naturally occurring compounds (Cuthbertson and Murchie, 2005). Botanical derivatives are more environmentally safe than synthetic chemicals (Hashim and 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*., 2006a, 2008a).

Datura metel L (syn. *Datura alba* Nees) is popular all over the world for its medicinal uses. It is known for its use in fever with catarrh, cerebral complications, diarrhea, skin diseases, antiseptic, animal bites, anti helmenthic and in herpetic diseases, and also has healing potential on burn wounds (Satyavati *et al.*, 1976; Priya *et al.*, 2002). It is also known for its antibacterial activity against burn pathogens (Gnanamani *et al*., 2003) and antifungal activity against phytopathogens (Dabur *et al.*, 2004; Kagale_*et al.*, 2004). However, research regarding herbicidal activity of *D. metel* is scarce. The present study was, therefore, designed to investigate the herbicidal activity of aqueous and organic solvent (methanol and *n*-hexane) extracts of *Datura metel* against germination, root and shoot growth and plant biomass of *P. minor.*

MATERIALS AND METHODS

Fresh root and shoot materials of mature *D. metel* were collected at flowering stage from a waste land in University of the Punjab, Lahore, Pakistan during January 2008. Plant materials were thoroughly washed with tap water followed by washing with sterilized water. Fresh plant materials were crushed in pestle and mortar and soaked in distilled water, methanol and *n*-hexane @ 15 g 100 ml^{-1}. Materials were soaked for 3 days at 25°C. After 3 days extracts were filtered through muslin cloth followed by Whatman filter paper No. 1. Volume of organic solvents was reduced to 2 ml by evaporation at 40 °C in a continuous supply of air in an electric oven and then diluted by adding appropriate quantity of sterilized distilled water to make the final volume 100 ml. These stock extracts (15% w/v) were stored at 4 °C. In order to make methanol and *n*-hexane control, 2 ml of each of the two organic solvents were added to sterilized distilled water to make the final volume 100 ml. These were referred as 2% methanol and 2% *n*-hexane solutions. Afterwards, appropriate quantities of distilled water, methanol and *n*-hexane were added to stock solutions to get 5 and 10% w/v concentrations of different extracts.

In a laboratory bioassay, the effect of 5, 10 and 15% of aqueous and the two organic solvent extracts was studied on germination and early seedling growth of *P. minor*. Ten seeds of test weed species, collected during May 2007 and stored in paper bags at room temperature, were placed in a 9-cm diameter Petri plates lined with Whatman No. 1 filter paper moistened with 3 mL of different concentrations of each extract. The respective control treatments for aqueous, methanol and n-hexane extracts received the same quantities of distilled water, 2% v/v methanol and 2% v/v *n*-hexane, respectively. Each treatment was replicated thrice. Plates were arranged in a completely randomized design in a growth room at 18 °C under 12 h light periods daily. After 10 days, seed germination, seedling root and shoot length, and plant fresh biomass were determined.

All the data were subjected to analysis of variance (ANOVA) followed by Duncan's Multiple Range test to delineate mean differences (Steel & Torrie, 1980) using COSTAT computer software.

RESULTS AND DISCUSSION

Analysis of variance shows that the effect of extracts in different solvents (S) was significant for root and shoot length, and plant biomass but not for germination. Effect of plant parts assayed (P) was significant only for root length. However, effect of extract concentration (C) was significant for all the studied parameters. The interactive effects of S×P, S×C, P×C and S×P×C were significant for all the studied parameters except germination (Table-1).

Table-1. Mean Squares of ANOVA for effect of different concentrations of aqueous, methanol and n-hexane shoot and root extracts of *D.metel* on germination and growth of *P.minor.*

Trait	df	Mean squares			
		Germination	Shoot length	Root length	Fresh biomass
Treatments	23	479**	301***	360***	22***
Solvent (S)	2	429 ns	615***	179***	46***
Plant part (P)	1	5.6 ns	82 ns	83*	0.4 ns
Concentration (C)	3	1750***	336***	2101***	34***
S × P	2	185 ns	275**	93**	15**
S × C	6	257 ns	342***	101***	28***
P × C	3	446 ns	144**	81**	14**
S × P × C	6	275 ns	262***	81***	12**
Error	48	190	38.4	18	3.1
Total	72				

*, **, ***, significant at P≤0.05, 0.01&0.001 ns: Non significant.

Effect of aqueous extracts

Aqueous shoot extract at different concentrations viz. 5, 10 and 15% (w/v) insignificantly affected germination of *P. minor* (Fig.-1A). Shoot length of *P. minor* showed variable response to different concentrations of *D. metel* shoot extract. The lowest concentration of 5% significantly enhanced shoot length by 38%. By contrast, 15% extract significantly declined the shoot length by 22% while effect of 10% shoot extract was insignificant (Fig.-1B, Table-2). Earlier studies also show that lower concentrations of plant extract may be stimulatory to the test plant growth (Javaid *et al.*, 2006b). Root length was more sensitive to shoot extract of *D. metel* than shoot length where all the employed extract concentrations significantly suppressed root length by 22–72%. There was a gradual decline in root length with the increase in extract concentration (Fig.-1C, Table-2). Greater sensitivity of root growth than shoot growth to the plant extracts have also been demonstrated in other plant species (Javaid *et al.*, 2008a). Since roots are the first to absorb chemical compounds from the environment, and thus may show their abnormal growth in response to chemicals present in the extracts, resulting in reduced length (Javaid and Shah, 2007). All the shoot extract concentrations of *D. metel* significantly reduced the plant biomass as compared to control. However, difference among the various extract concentrations of 5–15% was insignificant (Fig.-1D). *D. metel* is well known for tropane alkaloids and asteroids which could be responsible for its herbicidal activity (Sahai *et al.*, 1999).

Aqueous root extract of *D. metel* was less toxic to germination and seedling growth of *P. minor.* None of root extract concentration exhibited significant effect on germination, shoot length and plant biomass. Root length was, however, significantly declined by all the employed extract concentrations (Fig.-1A-D).

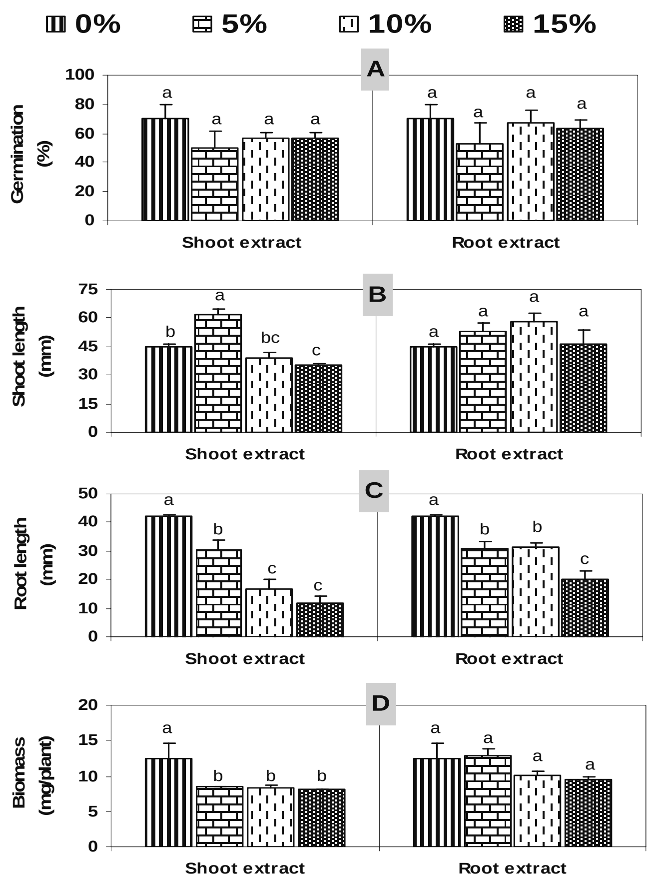

Fig. 1. Effect of aqueous shoot and root extracts of *D. metel* on germination and growth of *P. minor.*Vertical bars show standard errors of means of three replicates.Bars with different letters show significant differences (P≤0.05) by DMRT

Table-2. Percentage increase/decrease in germination, root and shoot length, and plant biomass of *Phalaris minor* due to different concentrations of aqueous, methanol and *n*-hexane shoot and root extracts of *Datura metel.*

Extracts	Percentage increase/decrease over control							
	Shoot extract				Root extract			
	Germi-nation	Shoot length	Root length	Plant biomass	Germi-nation	Shoot length	Root length	Plant biomass
Aqueous								
5%	−29	+38	−27	−32	−24	+22	−27	+4
10%	−19	−13	−60	−33	−4	+29	−25	−18
15%	−19	−22	−72	−35	−10	+2	−52	−24
Methanol								
5%	−9	−16	33	+9	−50	−39	−45	−12
10%	−38	−16	50	+61	−31	−38	−57	+66
15%	−29	−50	77	−44	−50	−24	−28	−52
n-hexane								
5%	−25	−19	−57	−20	−15	−42	−66	−24
10%	−36	−36	−61	−38	−25	−26	−57	−4
15%	−25	+15	−56	+22	−42	−40	−68	−60

\+ indicates increase over control
– indicates decrease over control

Effect of methanol extracts

Different concentrations of methanol shoot extracts of *D. metel* exhibited insignificant adverse effect on germination of *P. minor* (Fig. 2A). Shoot extract adversely affected the shoot length of *P. minor.* However, only the highest concentration of 15% showed significant effect resulting in 50% reduction in the shoot length (Fig.-2B). Shoot extract was highly toxic to root growth of *P. minor* where different employed extract concentrations significantly reduced root length by 33–77% (Fig.-2C). The lowest concentration of 5% did not show any pronounced effect on plant biomass while 10% extract significantly enhanced plant biomass by 61%. Further increase in extract concentration to 15% adversely affected the plant biomass resulting in 45% suppression in plant biomass (Fig.-2D, Table-2). Five withanolide glycosides named daturametelins H-J, daturataturin A and 7,27-dihydroxy-1-oxowitha-2,5,24-trienolide, have been isolated from the methanol extract of the aerial parts of *D. metel* (Ma *et al*., 2006), which may be responsible for herbicidal activity of this plant. Methanol root extract was more toxic to *P. minor* than shoot extract. All the employed root extract concentrations significantly declined germination by 31-50%. Similarly, shoot and root length was significantly reduced by 24-38% and 28-60%, respectively, due to various root extract treatments. Response of plant biomass to root extract was similar to that of response to shoot extract where 10% root extract significantly promoted while 15% extract significantly declined plant biomass (Fig.-2A-D, Table-2).

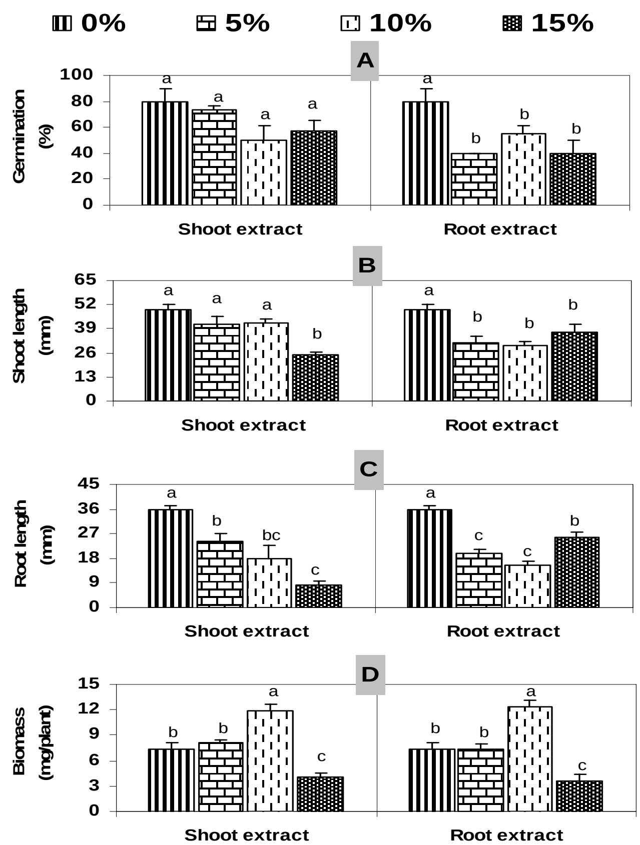

Fig. 2. Effect of methanol shoot and root extracts of *D. metel* on germination and early seedling growth of *P. minor.* Vertical bars show standard errors of means of three replicates. Bars with different letters show significant difference (P≤0.05) by DMRT

Effect of *n*-hexane extracts

Different concentrations of *n*-hexane shoot extract reduced germination of *P. minor* seeds by 25-36%. However, the effect was insignificant statistically (Fig. 3A, Table 2). Both shoot length and plant biomass showed similar response to different concentrations of shoot extract. There was a gradual decrease in these parameters up to 10% concentration and then a significant enhancement (Fig.-3B&D). Root length was more sensitive to various employed extract treatments than shoot length. All the concentrations of *n*-hexane shoot extract significantly suppressed root length by 55-61% (Fig.-3C, Table-2).

Root extract was more toxic than shoot extract to germination and seedling growth of *P. minor.* There was a gradual decrease in seed germination with an increase in extract concentration. The adverse effect of 15% extract concentration was significant where 40% reduction in seed germination was recorded as compared to control (Fig.-3A, Table-2). All the root extract concentrations significantly suppressed shoot and root length by 28-41% and 57-68%, respectively. Root length was more sensitive to extract than shoot length (Fig.-3B&C). In general, plant biomass was declined by *n*-hexane root extract, however, only the effect of 15% extract was significant where 60% reduction in biomass was recorded as compared to control (Fig.-3D, Table-2).

In the present study, extract of same part of the test plant species in different solvents exhibited variable herbicidal activity against the target weed species. This variation in herbicidal activity of the extracts in different solvents may be attributed to the different chemical nature of the three solvents. Both water and methanol are polar while n-hexane is non-polar in nature. It is likely that different types of chemicals were extracted in different solvents that resulted in variable activity of the extracts of same part of the plant (Javaid *et al.*, 2008a). The present study concludes that both root and shoot of *D. metel* contain herbicidal constituents. Further studies regarding the isolation and identification of these herbicidal constituents are in progress.

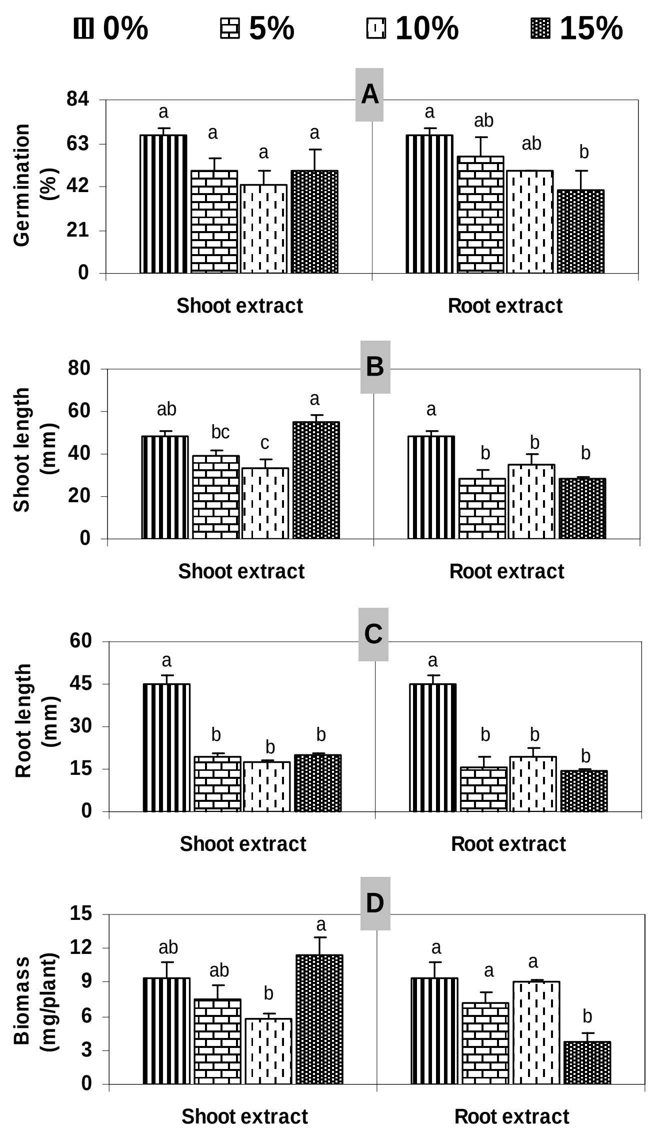

Fig.-3. Effect of n-hexane shoot and root extracts of *Datura metel* on germination and early seedling growth of *Phalaris minor.* Vertical bars show standard errors of means of three replicates. Bars with different letters show significant difference ($P \leq 0.05$) as determined by Duncan's Multiple Range test.

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