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Allelopathic Effect of Aqueous Extracts of Weeds on the Germination and Seedling Growth of Rice (*Oryza sativa* L.)

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Abstract

Germination and seedling growth of rice (*Oryza sativa*) seeds soaked in distilled water and water extracts of some weeds were investigated in two laboratory experiments. The root, shoot, leaf, seed and whole plants of *Trianthema portulacastrum* L., *Dactyloctenium aegyptium* L. *and Eleusine indica* L. were soaked in distilled water (1:20 w/v) individually for 24 hours at room temperature to obtain extracts. The germinated rice seeds were counted and removed from petri dishes every day in first experiment. Whereas, in second experiment, rice seeds were sown in sand culture to assess the seedling growth. Significantly the maximum mean germination time and time taken to 50 % germination was observed where rice seeds were soaked in water extracts of roots of *Trianthema portulacastrum* L. The rice seeds soaked in leaf extracts of *T. portulacastrum* L. showed the maximum inhibitory effects on root and shoot length of rice whereas, root and shoot dry weight per seedling as a result of interactive effect of weeds and water extracts was significantly minimum when rice seeds were soaked in root extract of *Dactyloctenium aegyptium* L. Pre sowing soaking in water extract of seed of *T. portulacastrum* increased shoot length of rice over control. Pre sowing soaking by leaf extract of *T. portulacastrum* proved to be the most injurious to germination and seedling growth of rice.

Keywords: Allelopathy, *Dactyloctenium aegyptium, Eleusine indica,* rice, seedling growth, *Trianthema portulacastrum*.

Introduction

Direct or indirect stimulatory or inhibitory effects of one plant on another through release of chemical compounds into the environment is referred to as allelopathy. Root exudation, leaching by dews and rains, and volatilization or decaying plant tissue from allelopathic plants results in release of compounds into the environment (Rice, 1984). Allelopathic efficacy of weeds on germination and seedling growth of crops vary from weed to weed (Hamayun et al., 2005). The allelopathic effects of various parts of same weed also differ for their effects on germination and initial growth of plants (Aziz et al., 2008; Economou et al., 2002).

Trianthema portulacastrum L. is a serious weed worldwide (Balyan and Bhan, 1986). *Dactyloctenium aegyptium* L. is a weed of the tropics and is among the 20 most globally widespread weeds (Simpson 1990). Whereas *Eleusine indica* is reported in 46 crops in 60 countries and is widely distributed in tropics and subtropics (Holm et al., 1977). These weeds may have either positive or negative effects on the growth of nearby plants. Information about the allelopathic effects of water extracts of different parts of these weeds on the germination and seedling growth of rice seeds soaked in water extracts of weeds is scarcely available. Therefore the allelopathic effects of root, shoot, leaf, seed and whole plant extracts of the above mentioned weeds on the germination and early growth of *Oryza sativa* soaked in water extracts of weeds were investigated in the laboratory.

Materials and Methods

Allelopathic effects of water extracts of various parts of *T. portulacastrum, D. aegyptium* and *E. indica* on the germination and seedling growth of rice seeds soaked in water extracts of weeds for 24 hours were investigated in two laboratory experiments. The experiments were laid out in completely randomized design with factorial arrangement having three replications. In first experiment, germination of rice seeds soaked in water extracts of weeds for 24 hours was assessed in petri dishes. Whereas in second experiment, rice seeds soaked in water extracts of weeds for 24 hours, were sown in sand culture to evaluate the seedling growth in the laboratory. Three above mentioned weeds were harvested at maturity from the Agronomy Research Farm, University of Agriculture, Faisalabad. Plants were separated into

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root, shoot, leaf, seed and whole plants and cut into small pieces. These pieces were dried for one month at room temperature. The various plant parts were then soaked in distilled water (1:20 w/v) individually for 24 hours at room temperature to obtain water extracts. The water extracts were filtered through a sieve to separate solid materials.

Experiment 1: Germination of rice in response to pre sowing soaking in water extracts of weeds.

In this experiment, rice seeds were soaked in distilled water (control) and water extracts of various parts of *T. Portulacastrum, D. aegyptium* and *E. indica* for 24 hours. Then fifty seeds of rice were placed on Whatman # 42 filter paper in 9 cm petri dishes. Four mL of the distilled water was added to each petri dish. The petri dishes were placed in a germinator at 30 °C for 10 days. Distilled water was applied when required during the course of experiment.

Mean Germination Time (MGT) was calculated as per equation of Ellis and Roberts (1981).

$$MGT = \frac{\sum Dn}{\sum n}$$

Where n is the number of seeds that had germinated on day D and D is the number of days counted from the beginning of germination.

Time taken to 50% germination (T_{50}) was calculated by using the formula given by Coolbear et ai., (1984) as modified by Farooq et ai., (2004).

$$T_{50} = \frac{ti + (N/2 - ni)(tj - ti)}{(nj - ni)}$$

Where N is the final number of germinated seeds while nj and ni are the cumulative number of seeds germinated by adjacent counts at times tj and ti, respectively, where $ni < N/2 < nj$. Germination index (GI) was calculated as given by Association of Official Seed Analysis (1990).

Germination was calculated by counting and removing the germinated seeds. Germination was observed daily in accordance with the methods of Association of Official Seed Analysis (1990).

Experiment 2: Seedling growth of rice in response to pre sowing soaking in water extracts of weeds.

In this experiment, ten seeds of rice soaked in distilled water and water extracts of weeds for 24 hours, before the start of experiment, were placed in beakers filled with sand for each treatment of each replication. The beakers were placed in an incubator at 30 °C. Distilled water was added subsequently when required. Root and shoot length of seedlings were measured at the end of experiment (after 16 days). Root and shoot dry weight per seedling of rice soaked in water extracts of weeds was recorded by cutting the seedlings into roots and shoots and oven drying till constant weight. Coefficient of uniformity of emergence (CUE) was calculated according to the following formula of Bewley and Black (1985).

$$CUE = \sum n / \sum [(t' - t)^2 n]$$

Where t is the time in days, starting from day 0, the day of sowing and n is the number of seeds completing emergence on day t and t' is equal to MET. Seedling vigor index (SVI) was calculated by using the following formula of Abdul-baki & Anderson (1973)

SVI = germination/emergence% × radicle length

Statistical analysis

Analysis of variance of all the data was conducted. The treatment means were grouped on the basis of least significant difference at the 0.05 level of probability.

Results and Discussion

Germination of rice seeds was significantly affected by soaking in distilled water and water extracts of weeds under test. Maximum mean germination time (MGT) was found where rice seeds were soaked in root and leaf extracts of *T. portulacastrum*. Whereas the least mean time taken for germination of rice seeds was observed where rice seeds were soaked in distilled water before sowing. Soaking of rice seed in water extracts of root, leaf and whole plant of *D. aegyptium* and *E. indica* were similar but different from respective water extracts of *T. portulacastrum.* Maximum time taken for 50% germination (T_{50}) was found where seeds were soaked in root and leaf extracts of *T. portulacastrum* prior to sowing. Minimum T_{50} was observed in distilled water treatment. Rice seeds soaked in shoot and seed extracts of all the weeds showed same results while root and leaf extracts of *D. aegyptium* and *E. indica* were also statistically alike but significantly different from same water extracts of *T. portulacastrum*. The maximum MGT and T_{50} in treatment where rice seeds were soaked in root extract of *T. portulacastrum,* indicate that there might be inhibitory compounds in water extracts of roots of *T. portulacastrum* which delayed the germination process of rice seeds. Results are supported by the findings of Babar et ai., (2009) who stated that chickpea seeds soaked in root extract of *Asphodelus tenuifolius* Cav. took more time for germination than seeds soaked in stem and fruit extracts of *A. tenuifolius*. Furthermore, they also reported higher T_{50} of chickpea seeds soaked in the root, stem and fruit extracts of *A. tenuifolius* before sowing, when compared with control. Non-significant differences in pre sowing soaking with water extracts of *E. indica* except its seed extract over control, might indicate that there were little inhibitory compounds in these parts as far as their effects on T_{50} are concerned.

Table 1 Effect of pre sowing soaking in water extracts of different weeds on germination and seedling growth of rice.

	MGT	T_{50}	GI	Germination (%)	Root Length (cm)	Shoot Length (cm)	Root dry wt. (mg)	Shoot dry wt. (mg)
Weed								
W_1= *T. portulacastrum*	6.73A	5.49A	26.63B	82.61B	5.22B	5.42B	12.45B	11.41C
W_2= *D.aegyptium*	5.41B	3.87B	32.74A	84.94A	6.32A	6.12A	12.36B	12.07B
W_3= *E. indica*	5.09B	3.51B	34.65A	84.39A	6.17A	5.44B	13.09A	12.79A
LSD 5%	0.445	0.416	2.055	0.991	0.339	0.109	0.127	0.143
Extract								
E_1= Distilled water	4.30C	2.70E	51.99A	97.44A	7.59A	5.77C	14.56A	13.58A
E_2= Root extract	6.55A	5.29A	23.36D	77.00D	4.78C	5.09D	11.91D	11.06D
E_3= Shoot extract	5.57B	3.87D	33.33B	85.89B	6.44B	6.28B	13.31B	12.79B
E_4=Leaf extract	6.34A	4.99AB	25.43CD	78.22D	5.23C	4.97D	11.96D	11.67C
E_5= Seed extract	5.67B	4.25CD	28.31C	85.67B	6.29B	6.72A	12.18C	11.66C
E_6 = Whole plant extract	6.06AB	4.62BC	25.63CD	79.67C	5.10C	5.12D	11.86D	11.80C
LSD 5%	0.630	0.588	2.906	1.401	0.479	0.154	0.179	0.203
Interaction								
W_1E_1	4.26g	2.60e	53.57a	98.00a	7.57ab	5.68gh	14.79a	13.49a
W_1E_2	9.00a	8.08a	15.20e	75.00g	4.22jk	4.52ij	12.07g	10.57g
W_1E_3	5.36cdef	3.86c	34.09b	90.67b	6.55cdef	6.65b	13.48c	12.87bc
W_1E_4	8.55ab	7.62a	13.20e	68.33i	2.78l	3.32k	11.39hi	10.32g
W_1E_5	5.60cde	4.33c	27.92cd	91.33b	6.65cde	7.86a	10.90j	9.620h
W_1E_6	7.61b	6.44b	15.81e	72.33h	3.57kl	4.46j	12.06g	11.62e
W_2E_1	4.30fg	2.73de	50.62a	96.67a	7.31abc	5.88efg	14.65a	13.69a
W_2E_2	5.92cd	4.37c	29.25bcd	83.00def	4.76ij	6.24cd	10.93j	9.870h
W_2E_3	6.24c	4.11c	33.40b	81.33ef	6.75bcde	6.45bc	12.89de	12.46d
W_2E_4	5.19cdefg	3.79c	29.94bcd	84.00cd	7.13abcd	5.53h	11.52h	11.70e
W_2E_5	5.58cde	4.31c	25.31d	81.00f	6.43def	6.47bc	13.09d	13.50a
W_2E_6	5.25cdefg	3.88c	27.95cd	83.67cde	5.54ghi	6.14de	11.09ij	11.21f
W_3E_1	4.32fg	2.75de	51.77a	97.67a	7.89a	5.73gh	14.25b	13.55a
W_3E_2	4.72efg	3.43cde	25.62d	73.00gh	5.35hi	4.50ij	12.73ef	12.73bcd
W_3E_3	5.12defg	3.64cd	32.49bc	85.67c	6.01efgh	5.73gh	13.57c	13.04b
W_3E_4	5.28cdefg	3.57cde	33.15b	82.33def	5.78fgh	6.05def	12.97de	12.99b
W_3E_5	5.83cd	4.12c	31.70bc	84.67cd	5.79fgh	5.84fg	12.54f	11.86e
W_3E_6	5.31cdefg	3.55cde	33.13b	83.00def	6.20efg	4.76i	12.44f	12.57cd
LSD 5%	1.091	1.018	5.034	2.427	0.830	0.267	0.310	0.351

MGT= Mean germination time, T_{50}= Time taken for 50% germination, GI= Germination index,
Means sharing the same letters in a column do not differ significantly at 0.05 probability level according to Least Significant Difference (LSD) test.

Rice seed soaking in distilled water showed maximum germination index (GI) while the minimum was observed where rice seeds were soaked in leaf, root and whole plant extracts of *T.portulacastrum*. Seed and shoot extracts of *D. aegyptium* applied to rice seeds before sowing were statistically different from each other. Pre sowing soaking in root extract of *E. indica* was significantly different from all other water extracts of the same weed. Root and leaf extracts of *D. aegyptium* and *E. indica* were same but different from same extracts of *T. portulacastrum*. Shoot extract of all weeds showed non-significant differences with each other. The greatest inhibition of germination percentage of rice was recorded by rice seed soaking in leaf extract of *T. portulacastrum* followed by whole plant extract of the same weed while the distilled water treatment showed higher germination percentage. Root and shoot extracts of *E. indica* were significantly different from each other and also with other extracts of the same weed. Pre sowing soaking in leaf extract of *T. portulacastrum* showed least GI and

germination percentage indicating the likely presence of allelochemicals which might have reduced the vigor of rice seeds and the germination process. Tanveer et ai., (2008) also reported minimum GI and germination percentage of rice when applied with leaf leachate of *Xanthium strumarium*. Shoot and seed extracts of all the weeds differed significantly from one another. Inhibition in seedling growth of rice by pre sowing soaking in water extracts of various parts of three weeds may indicate the allelopathic potential of these three weeds. These results are in accordance with the findings of Meihua et ai., (2006) who reported inhibitory effects of water extracts of *Lactarius hatsudake* on seedling growth of rape (*Brassica campestris*) and radish (*Raphanus sativus*). The seedling growth of rice was significantly affected by soaking in water extracts of various parts of weeds under study (Table 1). Significantly minimum root length of rice seedlings was observed by pre sowing soaking in leaf and whole plant extracts of *T. portulacastrum*. Higher root length was found in distilled water treatment while the higher shoot length was observed in seed extract of *T. portulacastrum*. Root length of rice seeds subjected to various water extracts of all the weeds differed significantly from control. Leaf extracts of all the weeds when applied pre sowing showed significantly different results from one another. Maximum reduction in root length over control was found where rice seeds were soaked in leaf extract of *T. portulacastrum* prior to sowing. Whereas among water extracts of *D. aegyptium* and *E. indica,* the maximum reduction was found by pre sowing soaking in root extracts of the respective two weeds (Figure 1). Minimum and significantly lower shoot length of rice seedling was found where rice seeds were soaked in leaf extract of *T. portulacastrum.* Seed and shoot extracts of *T. portulacastrum*, all the water extracts of *D. aegyptium* except its leaf extract and leaf extract of *E. indica* showed increase in shoot length of rice seeds soaked in water extracts of weeds, over control. Maximum reduction in shoot length of rice was observed where water extract of leaf of *T. portulacastrum* was applied to rice before sowing. Soaking rice seeds in water extracts of seed of *T. portulacastrum* caused maximum increase in shoot length over control. Shoot extract of *T. portulacastrum*, all water extracts of *D. aegyptium* except leaf extract, leaf and seed extracts of *E. indica* also caused increase in shoot length of rice seedlings over control (Figure 2). Significantly lowest root and shoot length of rice caused by pre sowing soaking in leaf extract of *T. portulacastrum* and reduction in root length by soaking in whole plant extract of *T. portulacastrum* could be attributed to presence of inhibitory chemicals in leaves and whole plants of *T. portulacastrum* respectively. Water soluble inhibitors could be the reason of reducing the root and shoot length of rice significantly (Kil and Yun 1992). Cell division might have been affected which reduced the root and shoot lengths of rice seedlings as allelopathic compounds are known to inhibit functioning of gibberellin and indole acetic acid (Tomaszewski and Thimann 1966). Pre sowing soaking in water extracts of seed, shoot of *T. portulacastrum*, all the water extracts of *D. aegyptium* except its leaf extract and water extract of leaf of *E. indica,* increased shoot length of rice which indicates the presence of compounds that stimulated the shoot length of rice seedlings compared with control.

The lowest root (RDWt) and shoot dry weight (SDWt) per seedling of rice was obtained by pre sowing soaking in seed extract of *T. portulacastrum.* Root dry weight per seedling was also lowest by soaking in root and whole plant extracts of *D. aegyptium*. Leaf and seed extracts of *D. aegyptium* were significantly different from each other and also from other extracts of the same weed. Shoot extract of *E. indica* differed significantly from other water extracts of *E. indica* for root dry weight per seedling of rice soaked in weeds water extracts for 24 hours prior to sowing. Water extracts of root, seed and whole plants of all weeds differed significantly with one another for their effect on root dry weight per seedling of rice. Whereas the maximum root and shoot dry weight per seedling was noted where rice was soaked in distilled water. Seed extract of *T. portulacastrum*, resulted in minimum and significantly lower root and shoot dry weight per seedling. Whereas rice seeds when soaked in root extract of *D. aegyptium* caused higher reduction in shoot dry weight. It can be owed to inhibitory compounds present in water extract of seed of *T. portulacastrum*. The reduction in root dry weight per seedling was due to reduction in root length and root thickness. Chou and Lee (1991) found that aqueous extracts of *Miscanthus transmorrisonensis* showed significant allelopathic effects on seed germination and radicle growth of rye grass (*Lolium perenne* L.). Increase in shoot dry weight per seedling over control was due to increase in thickness and length of shoot. Minimum co efficient of uniformity of emergence (CUE) was recorded where rice seeds were soaked in shoot extract of *D. aegyptium*. Maximum CUE was found where rice seeds were soaked in root extract of *E. indica* (Figure 3). Significantly minimum seedling vigor index (SVI) was found where rice seeds were soaked in leaf extract of *T. portulacastrum* before sowing while maximum was recorded in distilled water control (Figure 4).

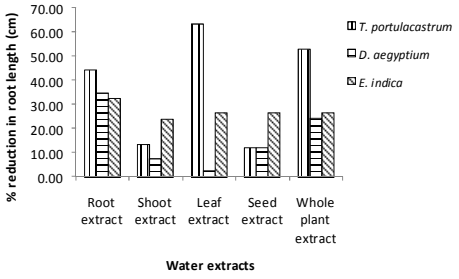

Fig 1 Effect of water extracts of various parts of weeds on % reduction in root length over control of rice seeds soaked in water extracts of weeds for 24 hours.

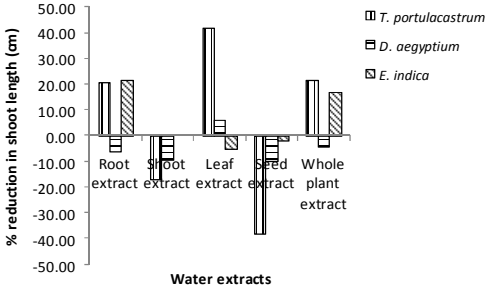

Fig 2 Effect of water extracts of various parts of weeds on % reduction in shoot length over control of rice seeds soaked in water extracts of weeds for 24 hours.

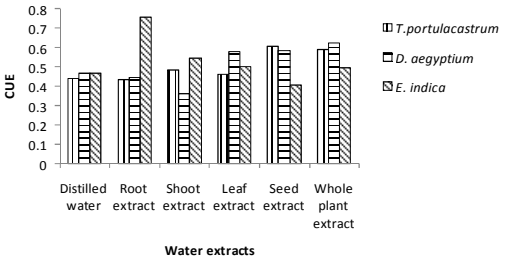

Fig 3 Effect of water extracts of various parts of weeds on CUE of rice soaked in water extracts of weeds for 24 hours.

Allelopathic activity of water extracts of *T. portulacastrum* was more pronounced on germination and seedling growth of rice. An overall powerful allelopathic inhibition in germination and seedling growth of rice occurred by pre sowing soaking in leaf extract of *T. portulacastrum*. We may conclude on the basis of these results, that most of the extracts of various parts of weeds under test contains water soluble compounds to a varying degree. These compounds may be released by rain or irrigation and dissolved in water under field conditions, therefore *T. portulacastrum*, *D. aegyptium* and *E. indica* present in farm fields should be controlled at early stage to avoid phytotoxic allelopathic effects of these weeds.

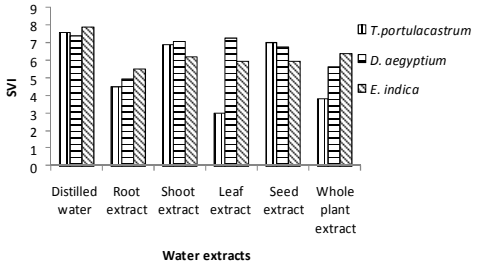

Fig 4 Effect of water extracts of various parts of weeds on SVI of rice soaked in water extracts of weeds for 24 hours.

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