Allelochemicals of *Phragmites karka* Enhanced the Growth of Rice Seedlings

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Abstract: The aquatic macrophytes are well known to exhibit allelopathy. Phragmites karka (Retz.) Trin. ex Steud. was screened for their allelopathic potential to the growth of rice seedlings. The 3% aqueous leachate was tested for seed germination of rice (variety PR-106) in petriplates as well as pot culture. The P.karka was found to enhance the growth of rice seedlings. The rice seedlings were planted in pots with soil mixed with 3% (w/w) dried matter (Ag and Bg parts) of P. karka. The growth of rice seedlings was highly promoted. This study suggested the future prospects of the integrated management of rice crop using the potential of allelopathic potential of P.karka.

Keywords: Allelopathic potential, leachate and aquatic macrophytes

1. Introduction

Allelochemicals produced by the plants endogenously are chemicals, which after being released into the environment, subsequently alter and modify the growth and development of neighbouring plants (Rice, 1984). These are biomolecules released from various plant parts by means of volatilization, leaching, decomposition of residue and root exudation.

The *P.karka* is emergent weed growing in the downstream areas of the reservoirs. The dominating nature of emergent macrophytes is normally associated with the accumulation of allelochemicals in the marshy places particularly in monospecific stands over a long period (Gopal and Sharma, 1990; Sharma *et al* 1990; Saxena *et al.*, 2007). The *P.karka* is known to release phenolic compounds and exhibit allelopathic interactions. Thus, the use of phytochemicals of *P.karka* in the form of dried plant material in the field of rice can replace the hazardous chemical fertilizer for better ecofriendly grain yield (Vyvyan, 2002).

2. Materials and Methods

2.1 Collection of Plant Material

Above ground (Ag) and below ground (Bg) plant portions of *P. karka* was collected from the Botanical Garden, Dr. Bhim Rao Ambedkar Govt. College Sri Ganganagar. The above ground and below ground plant samples were cleaned with soft brush. These samples were washed gently with tap water and distilled water thereafter and dried on absorbent paper.

2.2 In Vitro Seed Germination and Seedling Growth Lab Bioassay

The *in vitro* seed germination and seedling growth bioassay experiments were performed in petriplates to explore the phytotoxicity of aqueous leachates of *P.karka* at 3% concentration on the growth of test crop rice under laboratory conditions. The leachate of three percent concentration (dry weight/volume) of *P.karka* was prepared by soaking 3g plant material in 100 ml of distilled water, each for 24h and then filtered. The seeds of crops were

surface sterilized with 0.1 % HgCl₂ solution for one minute and washed thereafter 4-7 times with sterilized distilled water and dried with filter paper. The pre-sterilized petriplates (9 cm) were lined with two filter papers. Ten healthy seeds of rice crop (rice variety RP-106) were placed at equidistance on top of the filter paper in petriplates. Each treatment was replicated 3-5 times for each test species.As per treatment, each petriplate received 5 ml of leachate on first day and 3 ml leachate on 2, 4 and 6 days after sowing (DAS). The petriplates were kept in BOD incubator at 20-22⁰C at 35-40^oC. The seedlings were harvested 7 days after sowing and germination of seeds, lengths of shoot and root of seedlings were measured. Subsequently, these were kept in an oven for drying at 80^oC for 24 h and weighed thereafter for total dry weight.

2.3 Pot Culture Experiment

The pot culture experiment was conduct to meet the objectives of in vitro seed germination and seedling growth lab bioassay. The experiment was set up in earthen pots (14 x 9cm) with aim to investigate the allelopathic impact of P.karka on the growth of rice. The pots were filled with about 4.5 kg garden soil. Three treatments with control set were made for experiment. In first treatment (impact of dry matter), around 5cm layer of the top soil of each pot was removed (approximately 2 kg) and mixed with dry matter (60 g) of P. karka in ratio of 3 % (w/w) and refilled in earthen pots. In second treatment (impact of aqueous leachate), the 3% leachate of P. karka was used. In third treatment, the soil was treated with chemical fertilizer. The chemical fertilizer was applied in combination with NPK as 20kg N, 22 kg P and 42 kg K per hectare. The N was applied in the form of Urea (45 % N) at the rate of 540 mg/ 9 kg soil, P in form of single super phosphate (16 % P) at the rate of 99 mg / 9 kg soil and K in form of muriate potash (60 % K_2O) at the rate of 198 mg/ 9 kg soil. Five replicates were

taken for each set of treatment and control set. Ten seeds of rice (test crop) were sown in each pot. Three harvests were made at the interval of 40, 80 and 120 days after sowing (DAS). The parameters such as length of root, shoot, dry weight and leaf area were measured.

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3. Observations

3.1 In Vitro Seed Germination and Seedling Growth Lab Bioassay

The data presented in Table 1 shows the impact of aqueous leachate of 3% (w/v) concentration of above ground (Ag) and below ground (Bg) plant parts of *P. karka* on germination and growth of rice seedlings. The aqueous leachate of both Ag and Bg parts of *P. karka* effectively increased the germination and growth of rice seedlings. The promotion was more pronounced in Bg part than Ag part.

The % germination was 104% of control in both Ag and Bg parts. The root length remained only 75% of control in Ag part on the other hand it sharply increased to 150 % of control in Bg part. In case of shoot, the length was 122 and 145% of control in Ag and Bg parts respectively. Like wise, total dry weight also showed an increase and observed 127% of control in Ag and 162% of control in Bg part at 7 DAS.

Table 1: Allelopathic impact of 3 % (w/v) aqueous leachate of above ground (Ag) and below ground (Bg) plant parts of *Phragmites karka* on test crop rice

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Growth	P. karka					
parameters	Control	Ag	Bg	LSD		
Germination (%)	90.33 ± 3.33	93.3 ± 3.33	93.33 ± 3.33	12.3		
GIR	-	-3.7	-3.7	-		
Root length (cm)	1.81 ± 0.12	$1.35\pm0.02\texttt{*}$	$2.71 \pm 0.12*$	0.21		
Shoot length (cm)	1.56 ± 0.17	$1.90\pm0.03*$	$2.26\pm0.17\texttt{*}$	0.28		
Total dry weight						
(g)	0.026 ± 0.00	$0.033\pm0.00*$	$0.042\pm3.33^{\boldsymbol{*}}$	0		

 $\begin{array}{l} \mbox{Mean}\pm\mbox{SE, GIR -Germination inhibition rate, LSD -Least} \\ \mbox{significant differences, * Significant at 0.05 \% level by} \\ \mbox{Dunnett's test applied} \end{array}$

after ANOVA, Ag=Above ground, Bg=Below ground

3.2 Pot Culture Experiment

Root length

The data in Table 2 shows the successive stages of growth in root length of rice. The treated sets (aqueous leachate and soil incorporated with dry matter of P.karka) showed an enhancement in root length. The best promoting results were observed in sets incorporated with (3% w/w) dry matter of P.karka in soil at 40, 80 and 120 DAS. In chemical treatment, it was only 117% of control in first harvest whereas it was 140 and 137 % of control in dry matter and aqueous leachate of *P.karka* respectively. The root length was 120 % of control in chemical treatment whereas it increased upto 150 and 138% of control in dry matter and aqueous leachate of P.karka respectively. At 120 DAS, the root length of rice seedlings was in the following order as dry matter > aqueous leachate > chemical treatment (Fig.2).The ANOVA was found significant in all parameter. The Dunnett's test proved that aqueous leachate and dry matter of P. karka were highly significant for root length of rice plants at 40, 80 and 120 DAS.

 Table 2: Allelopathic impact of chemical fertilizer, 3% (w/w) dry matter and 3% aqueous leachates of *Phragmites karka* on root length (cm) of test crop rice

Harvest	Day after sowing (DAS)	Control	Chemical fertilizer	P. karka (dry matter)	P. karka (Aqueous leachate)	LSD
Ι	40	4.44 ± 0.73	$5.18 \pm 0.74*$	$6.23 \pm 0.45*$	$6.06 \pm 0.67 *$	0.06
II	80	9.20 ± 1.20	$11.57 \pm 0.80*$	$13.77 \pm 0.65*$	$12.66 \pm 0.61 *$	0.05
III	120	9.50 ± 0.57	$11.85 \pm 0.48*$	$13\;3.91\pm0.45*$	$12.78 \pm 0.48 *$	0.14

Mean ± SE, GIR -Germination inhibition rate, LSD -Least significant differences, * Significant at 0.05 % level by Dunnett's test applied after ANOVA

Shoot length

The data presented in Table 3 shows the impact of *P.karka* (dry matter and aqueous leachate) and chemical fertilizer on shoot length of rice plants. All the three treatments significantly increased the shoot length. The soil incorporated with dry matter registered highest length followed by aqueous leachate and chemical fertilizer. At 40 DAS, it was 131, 123 and 109% of control in dry matter, aqueous leachate and chemical fertilizer respectively. The

length gradually increased at each harvest in all the treatments in comparison to control. It was 127, 123 and 111% of control at 80 DAS and 124, 119 and 107 of control at 120 days respectively (Fig.3).The ANOVA was found significant in all the harvest. Further Dunnett's test proved that aqueous leachate; dry matter of *P. karka* and chemical fertilizer significantly increased the shoot length at different stages of plant growth.

 Table 3: Allelopathic impact of chemical fertilizer, 3% (w/w) dry matter and 3% aqueous leachate of *Phragmites karka* on shoot length (cm) of test crop rice

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Harvest	Day after sowing (DAS)	Control	Chemical fertilizer	P. karka (dry matter)	P. karka (Aqueous leachate)	LSD
Ι	40	12.96 ± 0.62	$14.20 \pm 0.74 *$	$17.01 \pm 0.45 *$	$15.91 \pm 0.87*$	0.1
II	80	45.45 ± 1.20	$50.41 \pm 1.25*$	$57.85 \pm 2.75*$	$55.84 \pm 1.45*$	0.14
III	120	$9.\;48.92\pm 0.57$	$52.35 \pm 0.68*$	$60.51 \pm 1.05*$	$58.19 \pm 1.01*$	0.3

Mean±E, GIR -Germination inhibition rate, LSD -Least significant differences, * Significant at 0.05 % level by Dunnett's test applied after ANOVA

Total dry weight

The data summarized in Table 4 shows the total dry weight of rice plants at three different harvests. The soil incorporated with dry matter of *P. karka* enhanced the total dry weight in all the three harvests. All the three treatments viz. chemical fertilizer, dry matter and aqueous leachate of *P.karka* significantly enhanced (126, 169 and 158% of control, respectively) the total dry weight at first harvest in comparison to control. At 80 DAS the total dry weight was found highest in soil incorporated with dry matter and lowest in chemical fertilizer treatment. At final harvest (120 DAS) the dry weight was recorded as 128, 119 and 109% of control in dry matter, aqueous leachate of *P.karka* and chemical fertilizer respectively.

Table 4: Allelopathic impact of chemical fertilizer, 3% (w/w) dry matter and 3% aqueous leachates of *Phragmites* karka on dry weight (g) of test crop rice

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KL	arka on dry weight	(g) of test crop ric	e			
Harvest	Day after sowing	Control	Chemical fertilizer	P. karka (dry	P. karka (Aqueous	LSD
	(DAS)			matter)	leachate)	
Ι	40	$0.549{\pm}0.05$	0.690±0.03*	0.923±0.02*	0.868±0.02*	0
II	80	$0.952{\pm}0.00$	$1.060 \pm 0.00*$	$1.147 \pm 0.01*$	$1.111 \pm 0.00*$	0
III	120	1.051 ± 0.04	1.142±0.04*	$1.351 \pm 0.04*$	$1.251 \pm 0.04*$	0.02

Mean ±SE, GIR - Germination inhibition rate, LSD - Least significant differences, * Significant at 0.05 % level by Dunnett's test applied after ANOVA

4. Discussion

Phragmites karka was screened for investigation of their allelopathic potential rice seed germination and seedling growth bioassay in laboratory as well as pot experiment. In the present study, *P.karka* has shown significant positive impact on the growth of rice in pot culture experiments. The sets with dry matter (3% w/w) and aqueous leachate (3% w/v) of *P.karka* significantly enhanced the growth of rice seedling up to 147 and 125% of control, respectively. The shoot length measured up to 124 and 119% of control in sets treated with dry matter and aqueous leachate of *P.karka*, respectively. The total dry weight was also found 128 and 118% of control respectively. Hence, the present study confirmed that the *P. karka* plant has favorable impact on rice.

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