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Preliminary phytoconstituents screening of some weeds and their potential toxicity on rice variety- Tarom via decomposition bioassay

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Abstract. The preliminary phytoconstituents screening of the mature root, stem and leaves of the dominance rice weeds like *Cyperus difformis* and *Echinochloa crusgalli* as well as second prominent weeds like *Paspalum paspaloides* and *Sagittaria trifolia* and their potential toxicity on local rice variety – Tarom was investigated in the paddy field of Babol (North of Iran) at 2010. Different phytochemicals like tannins, saponins, anthroquinones and terpenoides in water extract as well as flavonoids, glycoside and steroids in alcoholic extract are detected from some parts of these weeds. To evaluate the allelopathic effect of the various concentrations of the decomposed plant residue i.e. 2, 4, 8, 16, 32g/ 250g soil from selected weeds on the test crop, an experiment was conducted to form factorial examination in randomized of complete block design with three replications. This study indicated that seedling growth of Tarom variety was more hampered due to the various concentrations of *Sagittaria trifolia* except lowest concentration (2g) as compared with other studied weeds. The greatest inhibitory effect was found at the rate of 32g plant material in the soil about 55.99% over control. Unlike of seedling growth, seed germination of the test crop was more sensitive to decayed plant parts of Paspalum paspaloides than other selected weeds in all treatments. In general, seedling growth of the test crop was more hampered than seed germination. It might be due to the some allelochemicals which were present in the selected weeds and retards more seedling growth of the paddy seeds than seed germination.

Keywords: Paddy weeds, Preliminary phytoconstituents, Potential toxicity, Plant part residues, Various concentrations.

1. Introduction

Allelopathy differs from resource competition. Competition involves the removal or diminution of a shared resource, while allelopathy involves addition of a chemical compound/s to the environment through different processes (Rice, 1984 and Putnam, 1985). Allelochemical substances produced by plants and released from the plant by four general routs which are including leaching, decomposition of plant residues, root exudation and volatilization**.** The decomposition of plant residues usually contains the large quantity of allelochemicals that may be entering into the rhizosphere. According to Aldrich (1984), allelochemical must be concentrated in the root, stem or leaves rather than in the flowers or fruit. If it is concentrated in these organs it is unlikely that could be available in time to interfere with nighbouring plant.

After death and decay of plant materials the phytochemicals present in the cells are usually released in the surrounding environment by weathering and soil micro – organisms known as natural plant residues. The impregnation of soil with plant residues is phenomenon as decomposition of soil. Bioassay is a simple and highly sensitive for determining the toxicity of a wide range of phytochemicals and does not require sophisticated equipments and highly trained personnel. It is particularly useful for those phytochemicals, for which either specific methods are not available or are too much complicated (Narwal *et al*., 2004).

In the present study, to detect some active phytoconstituents and their allelopathic potential from some decomposed weeds like *Cyperus difformis* L., *Echinochloa crusgalli* (L.) P. Beauv and second prominent

weed like *Paspalum paspaloides* (Michx.) Scribner and *Sagittaria trifolia* L. on local paddy variety – Tarom, preliminary phtoconstitute screening and decomposing of the plant residues was undertaken.

2. Material and methods

Mature root, stem and leaves of paddy weed species viz. *Cyperus difformis* and *Echinochloa crusgalli Paspalum paspaloides* and *Sagittaria trifolia* were collected from different farm of paddy fields of Babol (Iran) at 2010. These parts were washed in distilled water for a minimum period to avoid leaching losses of water soluble allelochemicals and air- dried in shade (to prevent breakdown of chemicals) for a week and was chopped into small pieces in a grinder [fine powdered samples are more homogenous, provide greater precision and accuracy and fine powdered samples are easily dissolved/ in solvents owing to large surface area (Narwal *et al*., 2004)]. Then preliminary phytochemical tests were carried out for the confirmation of tannins, saponins, anthroquinones and terpenoids on water extracts, while flavonoids, glycosides and steroids in alcoholic extracts in the ratio of 1: 20 w/v (plant material: distilled water or methanol) by Trease and Evans, 1972 & 1989 method. For studding laboratory bioassay dried powdered material of whole studied weeds mixed with loamy soil (250g) at the rate of 2g, 4g, 8g, 16 & 32g and allow withering away for 40 days in the pot. The pots containing soil and grounded plant parts were maintained in wet condition by adding equal amount of distilled water. After 40 days of withering these mixtures was exposed to direct sunlight for evaporating allelochemicals present in it and placed in sterilized petridishes at the rate of 20g per petridish to form factorial examination in randomized of complete block design (RCBD) with three replications. Then to each petridish containing weighed soil, 10 ml of distilled water is added. Surface sterilized 10 certified seeds of local variety of rice – Tarom which is used as a test crop were sown in the soil. A petridish containing 20g of soil free from decaying plant parts were performed as a control. Each petridishs were wrapped by brown paper to avoid direct sunlight. After seven days germination % and root and shoot length were measured (Avchar and Deokule, 2007). Analysis of variance (ANOVA) of decomposed plant part of studied weeds was done in confidence level 95% by SPSS software. The mean treatments which had been significances were compared by using Duncan's Univariete multiple comparisons. Inhibition percentage (%) was calculated as [(control value- treatment value) / control value] × 100.

3. Results and Discussion

The preliminary phytoconstitute screening of the root, stem and leaves of studied weed species for the confirmation of tannins, saponins and anthroquinones in water extract as well as flavonoids, glycoside and steroids in alcoholic extract as presented in Tale 1 and 2. From these Tables, studied weeds contain different phytochemicals in the studied parts except incase of terpenoids which was detected in all suited weeds from different parts distinctly. These results are consistent with findings Lin *et al.* (2004) who reported allelochemicals activity is highly species- specific and changes among tissues.

Table 1. Results of phytochemical screening of *Cyperus difformis* and *Echinocola crusgalli*

Test	***Cyperus difformis***			***Echinocola crusgalli***		
	Root	Stem	Leaves	Root	Stem	Leaves
A) Water extracts						
Tannins	+ve	-ve	+ve	-ve	+ve	+ve
Saponins	-ve	+ve	+ve	-ve	+ve	+ve
Anthroquinones	-ve	-ve	-ve	-ve	-ve	-ve
Terpenoids	+ve	+ve	+ve	+ve	+ve	+ve
B) Alcoholic						

extracts						
Flavonoids	-ve	-ve	-ve	+ve	+ve	+ve
Glycosides	+ve	-ve	+ve	-ve	-ve	-ve
Steroids	+ve	+ve	+ve	+ve	+ve	+ve

+ve Presence of constitute
-ve Absence of constitute

Table 2. Results of phytochemical screening of *Paspalum paspalodes* and *Sagittaria trifolia*

Test	***Paspalum paspaloides Sagittaria trifolia***			***Paspalum paspaloides Sagittaria trifolia***		
	Root	Stem	Leaves	Root	Stem	Leaves
A) Water extracts						
Tannins	+ve	+ve	+ve	+ve	+ve	+ve
Saponins	-ve	-ve	+ve	-ve	+ve	+ve
Anthroquinones	-ve	+ve	-ve	-ve	-ve	-ve
Terpenoids	+ve	+ve	+ve	+ve	+ve	+ve
B) Alcoholic extracts						
Flavonoids	+ve	+ve	+ve	-ve	-ve	-ve
Glycosides	-ve	-ve	+ve	+ve	-ve	+ve
Steroids	+ve	-ve	+ve	-ve	-ve	-ve

+ve Presence of constitute
-ve Absence of constitute

The results of decomposition bioassay of *Cyperus difformis* on Tarom variety showed in (Table 1) indicated that the decaying plant materials at higher rates of 16 and 32 / 250g of the soil significantly decreased shoot length of test crop over control about (35.83%) and (47.35%) respectively. On the contrary, radicle length of Tarom seedling was not significantly affected in lower and higher amount of plant material allowed for decomposing. However, incorporation of dried plant parts in the soil at higher rates i.e. 16 and 32g of plant parts showed significant inhibition on seedling growth about 18.66% and 25.35% over control respectively. It was also observed that germination percentage of Tarom seeds was significantly hampered with amount of the plant material at the rates of 16 and 32g of the soil from that of control with same level of inhibitory effect statistically. It might be due to more allelochemicals release during withering processes by increasing plant material in the soil which retards stem length of seedling as well as seed germination of paddy seeds.

By the treatments of *Echinochola crusgalli* (in the same Table 1) radical length of test crop was significantly hampered at higher values (8, 16 and 32g). Shoot length of test crop was significantly inhibited at all values except 2g of plant part admixed into soil as compared with control. The maximum inhibitory effect was caused at concentration of 32g (73.52%). However, the decayed plant parts of studied weed at higher rates (8, 16 and 32g) inhibited seedling growth of test crop. The greatest inhibitory effect was

recorded at the rates of 16 and 32g of plant material added in the soil with same level of inhibitory effect about 36.15% and 32.39% respectively. It was also observed that there were no significant inhibitory effects on seed germination of test crop at lower and higher rates of plant materials added in the soil.

The results of decomposition bioassay of *Paspalum paspaloides* on seeds of Tarom variety (in the same Table 3) also revealed that decomposed plant parts of this weed at higher amounts (8, 16 and 32g) into soil significantly suppressed both radicle and shoot length over control. The major toxicity was caused by 32g plant materials allowed for decomposing. Perhaps, it maybe due to the large quantities of harmful chemicals was accumulated in the soil during withering processes. It was also recorded that seed germination percentage of the test crop was significantly decreased at lower and higher rates of incorporation dried plant parts into soil. However, incorporation of plant material admixed into soil at the higher rates (16 and 32g) caused maximum reduction on germination percentage of the test variety over control about (28.57%) and (32.14%) respectively.

Table 3. Decomposition bioassay of selected weeds on seedling growth and seed germination of Tarom variety.

Weeds	Quantity of plant parts in decomposition (g/ 250g soil)	Rg (cm)	Sg (cm)	Tsg (cm)	germination percentage
Cyperus difformis	**Control (D.W)**	**5.31***a*	**3.21***a*	**8.52***a*	**93.33***a*
	2.00	5.70*a*	3.38*a*	9.08*a*	86.67*ab*
		(6.84)	**(5.295)**	**(6.57)**	**(-7.16)**
	4.00	5.39*a*	3.05*a*	8.44*a*	83.33*ab*
		(1.51)	**(-4.98)**	**(-0.94)**	**(-10.71)**
	8.00	5.40*a*	3.01*a*	8.41*a*	86.67*ab*
		(1.69)	**(-6.23)**	**(-1.29)**	**(-7.14)**
	16.00	4.87*a*	2.06*b*	6.93*b*	76.67*b*
		(8.26)	**(-35.83)**	**(-18.66)**	**(-17.85)**
	32.00	4.67*a*	1.69*b*	6.36*b*	76.67*b*
		(12.05)	**(-47.35)**	**(-25.35)**	**(-17.85)**
Echinocholoa crusgalli	**Control (D.W)**	**5.31***a*	**3.21***a*	**8.52***a*	**93.33***a*
	2.00	6.10*ab*	3.32*a*	9.42*a*	83.33*a*
		(14.88)	**(3.43)**	**(10.56)**	**(-10.71)**
	4.00	6.50*a*	2.57*b*	9.07*a*	80.00*a*
		(22.41)	**(-19.94)**	**(6.46)**	**(-14.28)**
	8.00	4.69*cd*	2.47*bc*	7.16*b*	76.67*a*
		(-11.68)	**(-23.05)**	**(-15.96)**	**(-17.85)**
	16.00	3.12*e*	2.32*bc*	5.44*c*	76.67*a*
		(-41.24)	**(-27.73)**	**(-36.15)**	**(-17.85)**
	32.00	3.91*de*	0.85*c*	5.76*c*	76.67*a*
		(-26.37)	**(-73.52)**	**(-32.39)**	**(-17.85)**
Paspalum paspaloides	**Control (D.W)**	**5.31***a*	**3.21***a*	**8.52***a*	**93.33***a*
	2.00	5.39*a*	2.78*a*	8.17*a*	80.00*b*
		(1.51)	**(-13.40)**	**(-4.11)**	**(-14.28)**
	4.00	5.82*a*	2.86*a*	8.68*a*	80.00*b*
		(9.60)	**(-10.90)**	**(1.86)**	**(-14.28)**
	8.00	4.13*b*	2.71*a*	6.84*b*	73.33*bc*
		(-22.22)	**(-15.58)**	**(-19.72)**	**(-21.43)**
	16.00	4.26*b*	1.87*b*	6.13*b*	66.67*c*
		(-19.77)	**(-41.74)**	**(-28.05)**	**(-28.57)**
	32.00	2.90*c*	1.33*c*	4.24*c*	63.33*c*
		(-45.39)	**(-58.57)**	**(-50.23)**	**(-32.14)**
Sagittaria	**Control (D.W)**	**5.31***a*	**3.21***a*	**8.52***a*	**93.33***a*

trifolia				
2.00	5.46*a*	3.89*a*	9.35*a*	83.33*a*
	(2.82)	**(21.18)**	**(9.74)**	**(-10.71)**
4.00	4.41*bc*	2.51*b*	6.92*b*	76.67*a*
	(-16.95)	**(-21.81)**	**(-18.78)**	**(-17.85)**
8.00	4.10*cd*	2.27*b*	6.37*b*	76.67*a*
	(-22.79)	**(-29.28)**	**(-25.23)**	**(-17.85)**
16.00	3.29*de*	1.04*c*	4.32*c*	83.33*a*
	(-38.04)	**(-67.60)**	**(-49.30)**	**(-10.71)**
32.00	2.90*e*	0.85*c*	3.75*c*	76.67*a*
	(-45.39)	**(-73.52)**	**(-55.99)**	**(-17.85)**

. Number with different letters within column refers significant difference with confidence level 95% according Duncan's Univariete- range test. The data in parenthesis indicate % inhibition (-) over control.

As shown in Table 1, different treatments of decayed plant parts of *Sagittaria trifolia* significantly hampered on shoot length of the test variety except at 2g. The plant materials at higher rates 16g (67.60%) and 32g (73.52%) per 250g of the soil showed much significant inhibition on shoot length of test crop with same level of toxicity effect. Approximately results of decayed plant parts on radicle length were similar with results of shoot length but rates of inhibitory effect at different treatments were less than shoot length of the test crop. It was also recorded that seed germination was not much sensitive to all treatments over control.

In general, this study indicated that seedling growth of Tarom variety was more hampered due to various concentrations of *Sagittaria trifolia* except at lowest concentration as compared with other studied weeds. This study also revealed that seed germination of the test crop was more sensitive to decayed plant parts of *Paspalum paspaloides* than other selected weeds. All the average, seedling growth of the test crop was more hampered than seed germination. It might be due to the some allelochemicals like terpenoids, tannins, saponins, glycosides flavonoids, anthroquinones and steroids which were present in the selected weeds as retards more seedling growth of paddy seeds than seed germination. Variation in allelochemical activity during the excrete process indicates that plant itself is also key controlling factor (Lin *et al.* 2004).

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