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Allelopathic effect of *Populus deltoides* and *Ulmus wallichiana* on biochemical constituents of maize, bean and sunflower

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Abstract : The extracts of *P. deltoides* and *U. wallichiana* were tested for their allelopathic effect on biochemical constituents of *Zea mays* L. (Maize), *Phaseolus vulgaris* L. (Kidney bean) and *Helianthus annus* L. (Sunflower). The findings revealed that total chlorophyll, total soluble sugars and soluble proteins were reduced in the recipient species by all the extracts used. The chromatographic investigations revealed that the secondary metabolites identified in *P. deltoides* were dominated by phenolic glycosides and few phenolics and those identified in the extracts of *U. wallichiana* belong to the group of aliphatic hydrocarbons and triterpenes. Maximum number of allelochemicals were recovered in extracts obtained after 24 hrs long soaking time. Prolongation of soaking time beyond 48 hrs did not yield any component from the extracts of both the species. The compounds identified in both the species under investigation are known to induce inhibition in plant growth and development and hence their direct exposure could lead to reduction in overall productivity.

Key words: Agroforestry, Allelopathic effect, Biochemical constituents, Field crops

Introduction

The agroforestry systems have been designed to fulfill the economic, social and cultural needs of the local population, while keeping an eye on the ecological balance. The three potential causes of interference in agroforestry systems include allelospoly- competition, allelomediation- selective harboring of a herbivore and allelopathy- interaction through chemicals (Tomer and Srivastava, 1986). These chemical interactions, which take place near the tree-crop interface, are having significant effect on the total productivity (Muzika, 1993 and Rhodes, 1994). Numerous allelochemicals are released to the environment either by volatilization, leaching and exudation through roots or by decomposition of plant parts (Chou, 1983 and Rice, 1984). These compounds have either deleterious or beneficial effects on other plants growing in the vicinity of trees. The inhibitory effects attributed to these allelo-chemicals include suppression of cell elongation, cell division, membrane permeability, hydraulic conductivity, mineral uptake and enzymatic activity (Sahar *et al.,* 2005). Among the beneficial aspects of allelopathy many workers have demonstrated that some of these compounds can stimulate photosynthesis and absorption of p32; besides, imparting disease resistance and acting as herbicides and insecticides (Rakhteenko *et al.,* 1973; Netzly *et al.,* 1988 and Dzyubenno and Petrenko, 1971). The knowledge of these chemical interactions is very essential in deciding the best possible tree-crop combinations in agroforestry systems (Beart *et al.,* 1985).

The *P. deltoides* and *U. wallichiana* assume a great importance in plantations of Kashmir. Due to multiple uses and high net returns, these species are planted wherever the land is available. Although considered as first choice by the farmers in many traditional agroforestry systems, the allelopathic potential of these species has not received any attention. Hence, the present study was undertaken to determine the allelopathic effect of aqueous extracts of *P. deltoides* and *U. wallichiana* on biochemical constituents of *Zea mays* L. (maize), *Phaseolus vulgaris* L. (kidney bean) and *Helianthus annus* L. (sunflower).

Material and methods

The present investigation was conducted in the Forest Nursery at Faculty of Agriculture, Wadura, Sopore, Kashmir during the year 2002-05. The experimental site is situated at a latitude of 34°16' and 34°42' N and longitude of 74°20' and 74°32' E at an elevation of 1510m above MSL. The mean maximum and minimum temperature of the study site varies between 29.8 to -1.92° C with July and January as hottest and coldest months respectively. The average precipitation is about 710 mm, most of which is received in the form of snow during winter.

The seeds of maize, bean and sunflower were sown in poly bags (12"x18") in 2nd week of April having four seeds/ polybags. The aqueous extracts of fresh leaves were prepared by using plant material collected from eleven to thirteen year old experimental plantations of *P. deltoides* and *U. wallichiana.* The extracts were prepared (in the ratio of 1:5 w/v basis) by soaking plant material in distilled water for a period of 72 hours. The treatments are:-T_1= No treatment, T_2= treatment with fresh leaf extract of *P. deltoides* and T3= treatment with fresh leaf extract of *U. wallichiana*. These treatments were applied twice a week (for full crop period) to the test crops *viz.*, maize, bean and sunflower, replicating four times with 60 polybags/ treatment under RBD design. The experiment was repeated for three years.

For biochemical analysis, 1 kg fresh samples of leaves of *P. deltoides* and *U. wallichiana* were oven dried at 50° ± 5° C for 24 hrs and then crushed to powder to prepare extract in distilled water. The seeds of selected crops were soaked in extracts and distilled water (as control) for 1 hour and sown in plastic pots. The leaf samples for biochemical analysis were collected 30 days after germination. Total chlorophyll, total soluble sugars and soluble proteins were estimated by procedures followed by

Holden (1976), Irigoyen *et al.* (1992) and Bradford (1976) respectively.

Paper chromatographic procedure of Lodhi and Rice (1971) modified by Kil (1992) was used to identify the compounds present in the aqueous extracts of selected tree species. Two types of extracts were prepared from fresh leaves of *P. deltoides* and *U. wallichiana*. For this, samples were soaked in distilled water (1:20 w/v basis) or crushed to prepare extracts. The extract was filtered and acidified with 2N HCl to make pH 2.0 and two fractions- water and ether were made. The two fractions prepared were chromatographed in two dimension on Whatmans filter paper with n-butanol-acetic acid - water (BAW 63: 10: 27; v/v/v) followed by 6% aqueous acetic acid (AA6%). The chromatographs were inspected under ultra violet light (2537 Å) and compounds were marked. The data collected on different parameters were analyzed using suitable statistical tests.

Table 1. Effect of extracts of tree species on biochemical constituents of maize, bean and sunflower

Treatment	Biochemical constituent				
	Test crop	Total chlorophyll content (mg/ 100F.wt)	Total soluble sugars (mg/ 100F.wt)	Soluble proteins (µg/g F.wt)	Total chlorophyll/ protein ratio
T1	Maize	13.88	292.17	2.13	6.54
	Bean	11.69	221.35	2.49	4.70
	Sunflower	7.92	98.08	1.95	4.07
T2	Maize	13.54	278.26	2.09	6.48
	Bean	11.07	206.26	2.39	4.53
	Sunflower	7.34	87.69	1.85	3.97
T3	Maize	13.08	264.42	2.03	6.44
	Bean	10.83	197.78	2.34	4.69
	Sunflower	7.20	82.07	1.82	3.95
C.D. at 5%		0.32	10.22	0.03	0.05

The data in Table represent pooled mean values for three years

Results and discussion

The results on biochemical constituents of the test crops are presented in table 1. The data clearly shows that the reduction exerted due to aqueous extracts in the concentration of recorded leaf biochemical parameters of treated plants. The reduction in total chlorophyll content of maize, bean and sunflower was 6, 7 and 9 % in plants treated with extracts of *U. wallichiana* and 2, 5 and 7 % in plants applied with extracts of *P. deltoides*. Total soluble sugar content of leaves was also reduced in all the test crops. The reduction by *P. deltoides* and *U. wallichiana* extracts was in order of 5 and 9 % for maize, 7 and 11 % for bean and 11 and 16 % for sunflower. The total soluble protein content was also reduced in all the plants treated with aqueous extracts. The extracts of *P. deltoides* and *U. wallichiana* reduced the total soluble protein content by 2 and 5 % in maize, 4 and 6 % in bean and 5 and 7 % in sunflower respectively. The decrease in the concentration of chlorophyll, soluble sugars and protein content could be related to the direct exposure of recipient crops to the extracts of selected tree species.

The results of the present study are in agreement with the studies made by Sahar *et al.* (2005) and Singh *et al.* (2003) who observed that the leaf leachates of *Eucalyptus citriodora* and *E. globulus* influenced the metabolism of seeds and seedlings and reduced the concentration of chlorophyll, sugars, amino acids, organic acids and nitrogen content.

The data pertaining to identification of allelochemicals in aqueous extracts of selected tree species are presented in table 2. The allelochemicals identified in extracts of *P. deltoides* obtained

Table 2. Allelopathic compounds identified in aqueous extracts of tree species

Sl. No.	Chemical constituent	Tree species	
		Populus deltoides	*Ulmus wallichiana*
1.	Alnulin	-	+
2.	Betulin	-	+
3.	Caffic acid	+	+
4.	Catechol	-	+
5.	Ferulic acid	+	+
6.	Docosane	-	+
7.	Fragilin	+	-
8.	Gallic acid	+	-
9.	Grandidentatin	+	-
10.	Heptacosane	-	+
11.	Isofraxidin	-	+
12.	Lupenol	-	+
13.	Naonasane	-	+
14.	Pentacosane	-	+
15.	Picein	+	-
16.	Populin	+	-
17.	Protocatechol	-	+
18.	Salicylic acid	+	-
19.	Salicin	+	-
20.	Salicortin	+	-
21.	Scopoletin	-	+
22.	Syringaldehyde	-	+
23.	Tremulacin	-	+
24.	Tremuloidin	+	-
25.	Tricosane	-	+
26.	Vanillin	-	+

+ = Present *- = Absent*

after 24 hrs soaking in distilled water included condensed tannins and phenolic glycosides like Salicin, Salicortin, Tremuloidin, Tremulacin, Populin, Fragelin, Grandidentatin and Picein. Among the phenolics, ferulic acid, caffic acid and gallic acid were identified in the extracts obtained after 48 hrs soaking. The increase in soaking time to 72 hrs did not result in capturing of any additional compound. In contrast to present findings, Julkunen-Thtto (1988) could not detect the presence of populin, grandidentatin and tricosane in *Populus tremula* and *Populus balsemifera*. On the basis of these results, it appears that the distribution of glycosides in *Populus* was relatively species-specific.

The chromatograms of *U. wallichiana* showed that the most abundant components in the extracts of this species are aliphatic hydrocarbons and triterpenes. The main aliphatic hydrocarbons identified included nanocosane, heptacosane, tricosane and pentacosane. Two of the triterpenes identified included lupenol and alnulin. The increase in soaking time to 48 hrs yielded in capturing of three additional chemicals including betulin, scopoletin and isofraxidin. Prolongation of soaking time to 72 hr did not yield any additional allelopathic components. Compared to our study Martin *et al.* (2004) have identified eleven aliphatic hydrocarbons and eight triterpenes from the bark extracts of *Ulmus minor.*

The allelochemicals identified in the extracts of *P. deltoides* and *U. wallichiana* are known to inhibit the basic physiological processes; including protein biosynthesis, disruption of mineral support, suppression of cell elongation, cell division and membrane stability, which in turn induce reduction in growth and development of recipient crops (Rice, 1984; Wink and Twardoski, 1992). Since the selected crops were subjected to direct exposure to the extracts, the phenolics present therein resulted in pronounced reduction in growth, yield and biochemical parameters of the recipient crops. Thus, the direct contact of extracts of one plant with other plants could result in the decrease of overall productivity.

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