RESEARCH NOTE



Screening of Indian borage[*Plectranthus amboinicus* (Lour) Spreng], bitter weed [*Andrographis paniculata* (Burm.f.) Nees] and Southern cone marigold (*Tagetes minuta* L.) for allelopathic potential against weeds

K.K. Shakkira, P.V. Sindhu* and Meera V. Menon

Received: 16 January 2022 | Revised: 5 June 2022 | Accepted: 9 June 2022

ABSTRACT

Several plants express the allelopathic phenomenon through release of allelochemicals. Plants rich in allelochemicals can be used for controlling weeds in organic crop production. Current study was aimed at screening of *Andrographis paniculata (Burm.f.)* Nees (bitter weed), *Plectranthus amboinicus* (Lour) Spreng (Indian borage) and *Tagetes minuta* L.(Southern cone marigold) for allelopathic potential against upland weeds. This study was conducted from February to May 2021 in the Department of Agronomy, College of Agriculture, Vellanikkara, Kerala Agricultural University, Thrissur. The methanol extracts of *Tagetes minuta* and *Andrographis paniculate* at 25-30% concentration as pre-emergence application exhibited allelopathic effect on broad-leavedweeds.

Keywords: Allelopathy, Andrographis paniculata, Plectranthus amboinicus, Tagetes minuta, Weed management

Allelopathic plants could be a source of new potential herbicidal molecules for the chemical industry, which could be utilized to overcome the negative impacts of synthetic molecules. The term allelopathy generally refers to the stimulatory and inhibitory action of plants due to the direct or indirect release of some chemical compounds (Rice 1984). These plants synthesize and accumulate numerous components in the leaves, roots, fruits, flowers, and bark with various allelochemicals, including phenols, terpenoids, alkaloids, and flavonoids (Rizvi and Rizvi 1992). However, the pattern of germination inhibition and the suppression of earlier planted seedling growth have to be adequately studied.

Medicinal and aromatic plants are considered as sources of new natural allelopathic plant products (Azizi and Fuji 2006). The present experiment was conducted to assess the allelopathic potential of bitter weed (*Andrographis paniculata*), Indian borage (*Plectranthus amboinicus*), and Southern cone marigold (*Tagetes minuta*) to manage upland weeds. The experiment, on screening of selected plants for their allelopathic potential, was conducted inside the green house during February to May 2021 in the Department of Agronomy, College of Agriculture, Vellanikkara, KAU, Thrissur situated at 10°32'58" N latitude and 76°17'00" E longitude, and an altitude of 40.3 m above mean sea level. The experiment was laid out in a completely randomized design (CRD) in a factorial arrangement with three factors and three replications. Factor A consisted of three allelopathic donor plants *Plectranthus amboinicus, Andrographis paniculata* and *Tagetes minuta*. Factor B consisted of the method of extraction (cold water extraction, hot water extraction, and methanol extraction). Concentrations of extracts were included as third factor [5%, 10%, 15%, 0%, 25%, 30% and Control (distilled water)].

The allelopathic effect of selected medicinal plant donors on weeds was studied using 189 plastic trays (of size 25 x 20 x 5 cm) that were filled up to three-quarters with uniform quantity of soil (1.5 kg) collected from an open area in which Chinese potato (Solenostemon rotundifolius) was cultivated during previous years at the Agronomy Crop Museum, College of Agriculture, Vellanikkara, Thrissur. The texture of the experimental soil was sandy clay loam and was acidic in reaction with a pH of 4.74. The trays were separated into three groups of 63 trays, each group for a donor plant *i.e.*, three group of 9 trays; within the donor plants, the trays were grouped into three groups of 27, each for each type of extract. Within each method of extraction, three groups of 21 trays were randomly assigned, each for a concentration of extract, including water in sterilised soil as control treatment and one extra control treatment for each concentration (6 trays). The quantity of water required for attaining field capacity

Department of Agronomy, College of Agriculture, Kerala Agricultural University, Thrissur, Kerala 680656, India

^{*} Corresponding author email: pv.sindhu@kau.in

was tested before treatment application and calculated to be 350 ml for each tray. Extracts were prepared in appropriate quantities for each concentration for three replications. The treatments were imposed to assess the allelopathic effect of selected plants on weeds germination and growth. The treatments were applied uniformly to the plastic trays immediately after filling the trays with upland soil. Trays were irrigated at two days interval starting from 3^{rd} day after treatment application in order to maintain field capacity. Trays were examined daily for germination for one month, and observations on weed growth parameters were also recorded.

For preparing aqueous extract, 5 kg of each plant was collected and washed to remove the adhering soil. Cleaned samples were crushed, and 5 L of distilled water was added. These samples were shaken for one hour continuously in an electric shaker. The mixture was left to stand for 48 hours at room temperature, and the extracts were obtained through filtration using Whatman No. 1filter paper having a concentration of 100% w/v used as stock solution. These extracts were diluted to desired concentration of 5%, 10%, 15%, 20%, 25% and 30% using distilled water.

For preparing hot water extract, fresh and clean samples weighing 5 kg were crushed and transferred into a beaker containing 5 L distilled water and boiled for five minutes. The room cooled extract was filtered through Whatman No. 1filter paper and these extracts, having 100% concentration (w/v), were used as stock solutions. From these stock solutions, solutions of concentrations 5%, 10%, 15%, 20%, 25% and 30% were prepared using distilled water. Methanol extracts were prepared by soaking 5 kg crushed whole plant samples in analytical grade methanol of 5 L and boiled for five minutes, then shaking in an electrical shaker for one hour at room temperature. The extracts were filtered through

The extracts were characterized biochemically. The pH and EC of extracts were measured using a pH meter and electrical conductivity meter. The total alkaloids, flavonoids, phenols, and tannins were determined using the method of Harborne (1973). Observations on germination count of weeds at weekly intervals, and weed density and dry weight (biomass) at one month after extract application were recorded. The data were analyzed statistically using analysis of variance (ANOVA) with the statistical package 'OP Stat' (Sheoran *et al.* 1998). The data on weed density which showed wide variation, were subjected to square root transformation to make the analysis of variance valid (Gomez and Gomez 1984).

Biochemical characterization of extracts

The pH of extracts ranged from 7.62 to 4.3, and the EC ranged from 0.21 to 0.49. All the three donor plants were rich in secondary metabolites like alkaloids, flavonoids, phenols, and tannins (**Table 1**). The content of alkaloids was comparatively higher than other secondary metabolites. Higher content of alkaloids was observed in *Tagetes minuta* (mean value of 0.485%), followed by *Andrographis paniculata* (mean value of 0.417%). Among different extraction methods, methanol was more efficient in extracting the secondary metabolites.

Weed germination

distilled water.

Major weeds observed during the experimentation were Panicum sp., Boerhavia diffusa, Alternanthera bettzickiana, Emilia

Medicinal plants	Method of	pН	EC	Alkaloids	Flavonoids	Phenols	Tannins
	extraction		(dSm)	(%)	(%)	(%)	(%)
A. paniculata	Cold water	6.53	0.32	0.541	0.023	0.001	0.0007
	Hot water	7.62	0.23	0.149	0.020	0.001	0.0006
	Methanol	5.82	0.43	0.562	0.026	0.002	0.0009
	Mean	6.66	0.33	0.417	0.023	0.001	0.0007
P. amboinicus	Cold water	6.19	0.47	0.154	0.037	0.004	0.0002
	Hot water	6.70	0.49	0.156	0.027	0.003	0.0002
	Methanol	4.47	0.42	0.237	0.053	0.006	0.0003
	Mean	5.79	0.46	0.182	0.039	0.004	0.0002
T. minuta	Cold water	6.18	0.21	0.386	0.030	0.003	0.0005
	Hot water	7.03	0.49	0.218	0.024	0.003	0.0001
	Methanol	4.3	0.32	0.851	0.040	0.004	0.0007
	Mean	5.84	0.34	0.485	0.031	0.003	0.0004
LSD (p=0.05)		1.24	0.24	0.32	0.01	0.001	NS

 Table 1. Biochemical properties of leaves extracts of three donor medicinal plants selected

sonchifolia, Cleome viscosa and Euphobia hirta.

Among three plants screened for their allelopathic potential, *Tagetes minuta* exhibited highest allelopathic potential in delaying germination of weeds followed by *Andrographis paniculata* and the lowest was by *Plectranthus amboinicus* (**Table 2**). Better allelopathic effect of *Tagetes minuta* and *Andrographis paniculata* can be correlated with their higher contents of total alkaloids. Inhibitory effect of *Tagetes minuta* on sun spurge (*Euphorbia helioscopia*) and Johnson grass (*Sorghum halepense*) was reported by Sadia *et al.* (2015).

Regarding the method of extraction, a significant result was noticed for methanol extract and cold water extraction. Allelopathic efficacy of plants was found to decrease when they were extracted by the hot water extraction method. Better

 Table 2. Mean main effect of treatments on total germinated weeds seedling density

	Total weed density (no./m ²)			
Treatment	1 st week	2 nd week		
Allelopathic medicinal plant				
Andrographis paniculata	8.74(86.5)	11.46(130.6)		
Plectranthus amboinicus	12.44(154.9)	11.49(131.4)		
Tagetes minuta	8.04(72.4)	11.58(134.2)		
LSD (p=0.05)	0.21	NS		
Method of extraction				
Cold water extract	9.78(103.0)	11.52(132.7)		
Hot water extract	10.48(114.5)	11.52(132.8)		
Methanol extract	8.95(92.1)	11.43(130.7)		
LSD (p=0.05)	0.21	NS		
Concentration				
5%	10.49(113.0)	11.58(134.3)		
10%	10.11(105.9)	11.61(135.4)		
15%	9.55(96.1)	11.68(137.2)		
20%	9.20(89.8)	11.65(136.5)		
25%	8.66(82.0)	11.22(126.1)		
30%	7.19(67.2)	11.09(123.3)		
Control	12.98(168.3)	11.47(131.7)		
LSD (p=0.05)	0.32	0.398		

** $\sqrt{x+0.5}$ transformed values, original values are given in parentheses

allelopathic performance of methanol extracts can be attributed to the better extraction efficiency of secondary metabolites from plant samples. As compared to cold water (Waris *et al.* 2016) and hot water extraction methods, the contents of alkaloids and flavonoids were higher in the methanol extracts.

Among different concentrations tested, the best results were obtained with higher concentrations of 30 and 25%. Azambuja (2010) and Arora et al. (2015) also found a reduction in the allelopathic effect with a decrease in the concentration. With respect to the combined effect of all the three factors studied, the interaction was significant only in the first week after the application of treatments. In the 1st week, the lowest weed germination and weed density was observed with 30% methanol extract of T. minuta (6.67 no./m²) compared to the highest (168.33 no./m²) with control treatment (Figure 1). As compared to the control treatment 96.04% suppression in germination count was observed at 1st week by the application of 30% methanol extract of T. minuta. It was at par with methanol extract of A. paniculata at 30% (8.33 no./m²) concentration. Methanol extract of A. paniculata at 3% concentration resulted in weed suppression of 95.05% as compared to control. P. amboinicus extracts at different concentrations did not exhibit any effect on the germination of weeds. As compared to P. amboinicus, the per cent content of total alkaloids was higher in T. minuta and A. paniculata (0.851 and 0.562%, respectively) which might have contributed to their better allelopathic performance.

Weed density and biomass at one month after application

Weed density (Figures 2a and 2b) and biomass (Figures 3a and 3b) recorded one month after application of treatments indicated significant difference in weed density and biomass of broadleaved weeds and total weeds but not on grass weeds due to combined effect of allelopathic plants, methods of extraction and concentrations. Aslani *et*



Figure 1. Interaction effect of allelopathic plants, methods of extraction and concentrations on weed count at 1st week after application







Figure 2b. Interaction effect of three allelopathic plants, methods of extraction and concentrations on total weeds density at one month after application



weed biomass at one month after application



Figure 3b. Interaction effect of allelopathic plants, methods of extraction and concentrations on total weed biomass at one month after application

al. (2014) also reported that the dicot weeds were affected more severely than the monocots when treated with allelopathic plant extract.

Lower broad-leaved weed density and total density were observed with methanol and cold water extracts at 30 and 25% concentrations of T. minuta and A. paniculata. All the treatment combinations with these plants considerably reduced both weed density and biomass as compared to control. However, all treatment combinations with P. amboinicus could not succeed in reducing either density or dry weight of weeds. Owing to the richness of allelochemicals, Tagetes minuta might play a very important role in weed management through allelopathic interactions (Batish et al. 2007, Arora et al. 2015). Similarly, Li et al. (2010) and Kumar et al. (2018) reported inhibitory effect of A. paniculata on dicot plants. Effect of extracts on germination of weeds persisted only up to one week, indicating lack of residual action for the selected plant extracts. The germinated weed seedling density at 12 and 25 DAS did not differ significantly. In this preliminary screening study, it was observed that allelopathic plants T. minuta and A. paniculata could be effectively utilized for reducing the emergence of broad-leaved weeds. Many scientists (Bhadoria 2011, Ihsan et al. 2015) recommended the use of allelochemicals for the production of environment friendly herbicides since they caused few environmental problems in the soil due to the fairly high degradability.

It was concluded that maximum inhibitory effect on weeds germination and growth was observed with 30% methanol extract of *T. minuta* followed by it's 25% and the broad-leaved weeds were more sensitive to allelopathic extracts than grass weeds. The persistence of allelopathic effect of plants on weeds was significant only for a short period of time *i.e.* up to one week after the application.

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