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Allelopathic Effects of *Medicago sativa* L. and *Vicia cracca* L. Leaf and Root Extracts on Weeds

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Abstract: In this study, the allelopathic potential of different concentration (5, 25 and 50%) of *M. sativa* and *V. cracca* leaf and root extracts were evaluated on germination and radicle length of four weed species (*Amaranthus retroflexus* L., *Lolium perenne* L., *Ipomoea hederacea* L. and *Portulaca oleracea* L.) in laboratory condition. As a result, germination and radicle length of all species were reduced by the extract from *M. sativa* and *V. cracca* leaf and root at different percentage. Increasing the water extract concentrations from 5 to 50% of test plants parts significantly increased the inhibition of all weed species germination and radicle length.

Key words: Allelopathy, *Medicago sativa* L. *Vicia cracca* L., germination, seed

INTRODUCTION

Weeds cause considerable economic losses in crop growing areas. Weed management is necessary in all crops. However, we should keep the weeds below an economic threshold level, not kill them completely. Chemical methods are used routinely to control weeds in the world (Giudice, 1981). However, herbicides are often applied more than it is necessary and misapplication sometimes can occur. As a result, the environment can be damaged and cost and labour are wasted. Taking this into consideration alternative methods to chemical control are being investigated. One alternative method is the use of allelopathy. Allelopathy is defined as the direct or indirect harmful or beneficial effects of one plant on another through the release of chemical compounds into the environment (Rice, 1984). Allelopathic effects have been reported in many plant species including crops and weeds (Bhowmik and Doll, 1982; Curran *et al*., 1994; Gressel and Holm, 1964; Hedge and Miller, 1990; Kazincki *et al*., 1997; Narval and Tauro, 1994; Newman and Rovira, 1975; Rice, 1984; Turk and Tawaha, 2002; Uygur and Iskenderoglu, 1997). Recently allelopathic research has played an important role as allelochemicals may be used for integrated weed control and thus reducing environmental pollution. Allelopathy offers potential for biorational weed control through the production and release of allelochemics from leaves, flowers, seeds, stems and roots of living or decomposing plant materials. Under appropriate conditions, allelochemics often exhibit selectivity, similar to synthetic herbicides (Weston, 1996).

In this study, the allelopathic effects of two crops (alfalfa and bird vetch) leaf and root extracts on four weed species (*Amaranthus retroflexus* L., *Lolium perenne* L., *Ipomoea hederacea* L. and *Portulaca oleracea* L.) seed germination and radicle growth was investigated in bioassay experiments.

MATERIALS AND METHODS

Medicago sativa L. and *Vicia cracca* L. were selected test plants which contain secondary allelopathic compounds (White *et al*., 1989). The test crop species were collected from the near the North Grafton Massachusette, USA, at 50% of flowering stage in June 2004. In case of test plants were divided into roots and leaves. The roots and leaves were cut into small pieces in a grinder. After grinding, fresh leaves and roots of crops were soaked in distilled water through one week to give concentrations of 25, 125 and 250 g^{-1} of leaves and roots into 500 mL^{-1} water. After soaking, mixtures were filtered with pore size 0.22 μm. The experiment was conducted in sterilized petridishes with at 25°C in a dark incubator. 10 mL^{-1} of extract solution was added to 50 weed seeds (*Amaranthus retroflexus* L., *Lolium perenne* L., *Ipomoea hederacea* L. and *Portulaca oleracea* L.) which were surface sterilized with water:bleach (10:1) and were placed evenly on filter paper (Whatman 1, 9 cm) in petri dishes.

At the same time, distilled water was used as the control. The number of germinated seeds and radicle length were determined after one week. When seeds had produced seedlings 0.5 cm long, they were regarded as germinated and were removed the petri dish. Effectiveness of the extracts was calculated by Abbott's formula (Abbott, 1925). Each treatment was replicated four times in a completely randomized block design and experiment was repeated twice. Results were evaluated by analysis of variance. The means were separated by the Duncan's Multiple Range Test at the 0.05 level of probability.

RESULTS AND DISCUSSION

Leaf and root water extracts from fresh *M. sativa* showed inhibitory effects on selected weed seed germination (Table 1). All aqueous extracts reduced seed germination compared with distilled water control. As a result, different leaf extracts of *M. sativa* (5, 25 and 50%) reduced germination of *A. retroflexus* 10.26, 46.25 and 98.72%; *L. perenne* 14.29, 60.00 and 100.00%; *Ipomoea hederacea* 19.01, 77.46 and 100.00%, *Portulaca oleracea* 20.27, 75.68 and 100.00%, respectively. Leaf extracts of *M. sativa* (50%) was affected, except *A. retroflexus*, 100% on studied all weeds. According to Sozeri (2003) the first time allelopathic plant of *M. sativa* was reported by Mishustin and Nauvoma (1955). Some researchers have determined that alfalfa plants contain water-soluble phytotoxic compounds that are released into the soil environment from fresh leaf, stem and crown tissue, as well as from dry hay, old roots and seeds (Nielsen *et al*., 1960; Klein and Miller, 1980). Chung and Miller (1995a) determined that allelochemicals are found in various alfalfa plant parts in differing concentrations. Nielsen *et al.* (1960) studied the effects of water extracts of alfalfa, timothy, corn, oat and potato on alfalfa, timothy, oat, soybean, pea and corn. They reported that alfalfa extracts caused the greatest delay in seed germination and seedling growth of all test plants. This report is in agree with my results. As seen Table 1, also root water extracts of *M. sativa* significantly inhibited germination of *A. retroflexus* 22.50, 58.75 and 100.00%; *L. perenne* 29.45, 71.92 and 100.00%; *Ipomoea hederacea* 27.78, 84.72 and 100.00%, *Portulaca oleracea* 37.50, 81.58 and 100.00%, respectively. This result is supported by Chung and Miller (1995b), who report that the degree of inhibition increased extract concentration. Root extracts was the more inhibitory than the leaf extracts. Miller (1983) reported that alfalfa contains water-soluble substances that toxic to alfalfa itself as well as to other species. Sozeri (2003) reported that water extracts of alfalfa roots and leaves in 1/10, 1/20 and 1/40 dilutions killed germinated seedlings of russian knapweed at incubated in the blotter 25, 32.50, 45% and 40, 42,50 and 37.50%, respectively. According to Li (1981), alfalfa root aqueous extracts that were incubated room temperature for one week still reduced alfalfa seed germination and root development. Ells and Mcsay (1991) reported that alfalfa (*M. sativa*) residue was toxic to *Cucumis sativus* seed germination and seedling growth. Aqueous extracts of *M. sativa* herbage inhibited weed seed germination and seedling growth of dicotyledonous species more than monocotyledonous species (Miller, 1996). Leaf extracts of *V. cracca* (5, 25 and 50%) reduced germination of *A. retroflexus* 7.89, 42.11 and 100.00%; *L. perenne* 22.22, 46.53 and 100.00%; *Ipomoea hederacea* 12.82, 55.77 and 100.00%, *Portulaca oleracea* 23.61, 52.08 and 100.00%, respectively (Table 1). The degree of reduction increased as the extract concentration progressively increased from 5% to 50%. As seen Table 1, root water extracts reduced germination of *A. retroflexus* by 9.21, 56.58 and 100.00%; *L. perenne* 28.47, 68.06 and 100.00%; *Ipomoea hederacea*

Table 1: Effects of *M. sativa* L. leaf and root extracts on germination percentage and radicle length of weed species

	Weed species							
	Amaranthus retroflexus		*Loliumperenne*		*Ipomoea hederacea*		*Portulaca oleracea*	
Treatments	Ger.	Rad.	Ger.	Rad. (cm)	Ger. (%)	Rad. (cm)	Ger. (%)	Rad (cm)
M. sativa leaf extracts								
Control	97.50a*	5.95a	87.50a	5.85a	88.75a	7.10a	92.50a	6.30a
5%	87.50b	3.75b	75.00b	4.10b	71.88a	4.50b	73.75a	3.95b
25%	52.50c	1.80c	35.00c	2.25c	20.00b	2.80c	22.50b	2.95c
50%	1.25d	0.13d	0.00d	0.00d	0.00c	0.00d	0.00c	0.00d
M. sativa root extracts								
Control	100.00a	5.70a	91.25a	6.05a	90.00a	8.00a	95.00a	6.70a
5%	77.50b	2.90b	64.38b	3.10b	65.00b	2.75b	86.25b	2.95b
25%	41.25c	1.00c	25.63c	1.25c	13.75c	1.35c	41.25c	1.55c
50%	0.00d	0.00c	0.00d	0.00d	0.00d	0.00d	0.00d	0.00d
V. cracca leaf extracts								
Control	95.00a	4.78a	90.00a	4.85a	97.50a	5.88a	90.00a	5.10a
5%	87.50a	3.36b	70.00b	2.58b	85.00b	3.15b	68.75b	2.55b
25%	55.00b	1.88c	48.13c	1.38c	43.13c	2.05c	43.13c	1.13c
50%	0.00c	0.00d	0.00d	0.00d	0.00d	0.00d	0.00d	0.00d
V. cracca root extracts								
Control	95.00a	5.00a	90.00a	5.50a	97.50a	5.38a	90.00a	5.95a
5%	86.25b	2.73b	64.38b	1.98b	79.38b	1.90b	64.25b	2.38b
25%	41.25c	1.43c	28.75c	1.05c	25.13c	1.05c	27.00c	0.88c
50%	0.00d	0.00d	0.00d	0.00d	0.00d	0.00d	0.00d	0.00d*

Values in the columns followed by same letter(s) are not significantly different at the 0.05 according to Duncan's Multiple Range Test

18.59, 74.23 and 100.00%, *Portulaca oleracea* 28.61, 70.00 and 100.00%, respectively. Leaf and root extracts of *V. cracca* (50%) was affected 100% on studied all weed species. Generally most studies have been conducted to identify allelopathic effect of *V. villosa* and *V. sativa*. Research with hairy vetch extracts and incorporated residue has demonstrated the potential for this species to inhibit several weed and crop species (White *et al*., 1989). Patrick *et al.* (1963) using lettuce (*Lactuca sativa* L.) as a bioassay, found phytotoxic substance in the decomposition products of hairy vetch extracts. White *et al.* (1989) showed that germination inhibition of some crops from residues and leachates of crimson clover and hairy vetch. They also reported that *V. villosa* residue may also suppress weed germination through allelopathy. Chung and Miller (1995a) reported that aqueous extracts of alfalfa and hairy vetch reduced corn and soybean seed germination. Bradow and Connick (1990) demonstrated that *V. villosa* residue exudes allelochemicals that inhibit germination of some weeds. Although allelopathic potential of *V. villosa* and other leguminous has been demonstrated, some studies have shown that weed suppression by a legume cover crop was generally less than grass cover crops (Wallace and Bellinder, 1992). Radicle length of selected weed species were affected by *M. sativa* leaf and root extracts (Table 1). All extracts caused a marked reduction in radicle length of *A. retroflexus*, *L. perenne*, *Ipooea hederacea* and *P. oleracea*. As shown Table 1, root water extracts reduced germination of *A. retroflexus* by 36.97, 69.75 and 97.90%; *L. perenne* 29.91, 61.54 and 100.00%; *Ipomoea hederacea* 36.62, 60.56 and 100.00%, *Portulaca oleracea* 37.30, 53.17 and 100.00%, respectively. Guenzi *et al.* (1964) demonstrated that extracts of alfalfa containing water-soluble saponins inhibited the shoot and root length of corn seedlings. Karaaltin *et al.* (1999) reported that aqueous root, stem, leaf, flower and seed extract of *Elçi* of *M. sativa* was no effect on wheat, corn and barley seed growth. Miller (1996) found that aqueous extracts of separated alfalfa plant parts significantly inhibited alfalfa germination, as well as seedling length and weight. Same researcher reported that radicle length was more sensitive to aqueous extracts than seed germination or hypocotyl length. The leaf and root water extracts of *V. cracca* reduced radicle length of *A. retroflexus* by 29.58, 60.73 and 100%, 45.50, 71.50 and 100%; *L. perenne* 46.91, 71.65 and 100.00%; 64.09, 80.91 and 100.00%; *Ipomoea hederacea* 46.38, 65.11 and 100.00%, 64.65, 80.47 and 100.00%; *Portulaca oleracea* 50, 77.94 and 100.00%, 60.08, 85.29 and 100%, respectively (Table 1). According to Chung and Miller (1995a) aqueous extracts of *V. villosa* reduced corn and soybean seedling radicle length and seedling weight.

In conclusion, further experiments are also needed to investigate the allelopathic effective of these plants and to identify the active compounds involved in *M. sativa* and *V. cracca* allelopathy and to search their application under field condition.

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