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The effect of St Johnson wort (*Hypericum perforatum*) extract on the weeds of corn (*Zea mays* L.) under laboratory condition

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ABSTRACT

The allelochemicals or secondary plant metabolites when are released to environment, influence the germination and seedling growth of neighbors' weeds. The aim of this study was to investigate the effect of the extract of St Johnson wort (Hypericum perforatum) on the number and mass of weed seedlings and their seed germination under laboratory conditions. The St Johnson wort extracts were considered with different concentrations (1.25 to 20%) on the germination and seedlings growth of Zea maize, Johnsongrass (Sorghum halepense), Common lambsquarter (Chenopodium album) and Redroot pigweed (Amaranthus retroflexus) for 5 days. Results showed that with increasing concentration of St Johnson wort extracts, their effects on reducing seed germination and seedling growth of the above mentioned plants were more severe. Comparison of extract effects with polyethylene glycol, revealed that reduction of Johnsongrass, corn seed germination and length of plumules were attributed to the osmotic pressure of the extract, where, inhibition of seed germination of Redroot pigweed and Common lambsquarter was due to allelochemicals.

Key words: Allelopathy, *hypericum perforatum*, extract, weeds, corn.

INTRODUCTION

Environmental effects of chemical herbicides and the limitations of their implementation have led to the increasing importance of non-chemical alternatives in the management of weeds [3]. Allelopathy is a biochemical reaction between two or more plants or cultivars in which the release of natural chemical materials (Allelopathins) by a herb affects the physiological processes of neighboring plants or organisms [3]. Using this phenomenon, one might be able to control the weeds in farms without implementing chemical herbicides. For example, the inhibitor effect of walnut leaves on neighboring herbs is an instance of allelopathic effects [15]. Jung et al. [2004] reported that various types of rice have different inhibitor effects on the emersion of Barnyardgrass seedlings [14]. In addition, in their green house experiments, Abdul- Rehman and Habib [1989] demonstrated that alfalfa and its decomposed remains have allelopathic effects on Gramine (*Imperata cylindrica*) [1].

Observations conducted in Pakistan indicated that comixing the remains of forage sorghum with soil and cultivating wheat in it is 40-50 percent effective in controlling weeds and leads to a better performance of the seed up to 15 percent [4]. With its allelochemicals, Rye (*Secale cereale*) has been used as a covering plant and mulch in cultivations such as corn, cotton and soya in the U.S [13, 2].

Medicinal and aromatic plants have ingredients that pose inhibitor effects on germination and growth of other plants [12]. Inhibitory capability of allelopathins of shaddock medicinal plant on the growth of some plants indicates that the extract of this plant has inhibitory effects on the growth of weeds [8]. Effect of tea leaf extract on the growth of linen showed that this plant is affected by the tea leaf extract [11].

St. Johnson wort (*Hypericum perforatum*) is a several-years-old plant from Hypericum family which is used as medical plant due to its multiple essenses and alkaloid and glycoside ingredients [3]. Known allelopathing in plants which have toxic properties mostly includ alkaloid, glycoside and phenolic ingredients [9, 18].

The objectives of this research were (i) to study the allelopathic effects of St. Johnson wort medicinal plant on the germination of the seeds and the growth of seedlings of weeds including Reedroot pigweed (*Amaranthos retroflexus*), Common lambsquarter (*Chenopodium album*) and Johnsongrass (*Sorghum halepense*) as well as cultivation plant corn (*Zea mays*) in laboratory circumstances and (ii) to segregate the allelopathic effects of the extracts, from osmotic pressure.

MATERIALS AND METHODS

This research was conducted in the laboratory of Iranian Phytopathology Research Institute. After drying in shaddow and 25ºC temperature, aerial parts of St Johnson wort was grinded and powdered. Fifty grams of this powder was added to 500 ml water containing 20' alchohol and then put on shaker with rotational speed of 130 tpm in room temperature (22+2ºC) for 24 hours. The extract was passed through filter paper and 500 ml water containing 20º alchohol and then put on shaker with rotational speed = 130 tpm in room temperature for 24 hours. The resulted solution was passed through filter paper. The extracts obtained from this two processes were then mixed and kept in temperature -5ºC for the next steps of the experiment. In order to eliminate alchohol, the extract was dried in room temperature under vacuum using rotary evaporator. Resulting remains from drying the extract was dissolved in 50 ml water containing 20º alchohol and various densities of the medicinal plant extract were provided from it. Densities of 1.25, 2.5, 3.75, 5, 10, 15 and 20 microliters of this alchoholic extract were added to Petri dishes containing filter paper [19] (Table 1). Polyethylene glycol tests were conducted to determine the standard of osmotic pressure in exploring the effects of Yarrow extract on the germination and growth of weeds and differentiating it from allelopathic effect [19]. For this purpose, osmotic potential of the extracts with various densities was determined by osmometer and then, PEG solutions with similar potential were provided. Evaluation of indicators including percentage of germination, length of radicles and plumules were examined five days later [6].

Table 1. Osmotic potential of various densities of St Johnson wort

Density of extract (%)	0	1.25	2.5	3.75	5	10	15	20
Osmotic potential (meg a passal)	0	4.12	8.25	12.37	16.50	33.00	49.50	66.00

To evaluate the effects of St Johnson extract and polyethylene glycol, tests were conducted separately under similar conditions. After evaporating the organic resolvent, sterile aquapura was added to each Petri dish to create various densities of the extract in terms of milligram/ liter [6]. In laboratory, 50 aseptic seeds of weeds and corn were placed in each Petri dish. Viability of the tested seeds for corn, Redroot pigweed, Common lambsquarter and Johnsongrass were determined 90, 98, 40 and 60 percent, respectively. In order to break the sleep of the seeds in Common lambsquarter case Clorox solution (5.25 percent hypochlorite sodium) was applied in temperature 25'C for 10 minutes and in Johnsongrass case it was done by scratching the seeds. Petri dishes containing filter paper and water with 20ºC alchohol were used as control. Petri dishes containing seeds were placed in germinator in thermal and lighting cycles of 25ºC and 14 hours of lighting and 20ºC with 10 hours of darkness. Each of the two experiments on the extracts and polyethylene glycol were conducted as balanced and random models with four replications for each treatment in laboratory conditions including 16 hours of lighting with 25ºC and 8 hours of darkness with 22ºC.

Statistical analysis

Transformation of data was performed using the formula $\sqrt{x+o.5}$ [10]. Analysis of variance and mean comparison with Duncan,s multiple range test were performed by SAS software.

RESULTS

Results indicated that St Johnson wort extract has the capacity of inhibitory effects on the germination and growth of wild Redroot pigweed, Common lambsquarter, Johnsongrass and corn seedlings. This effect, however, is different on the germination and length growth of the radicles and plumules of the species studied. In this regard, the effects of St Johnson wort extracts with various densities were compared with those of polyethylene. Since polyethylene glycol does not have phototoxic effect and the inhibition of germination and growth of corn seedlings and weeds takes place due to osmotic pressure, it can differentiate between allelopathic effect of the extract and osmotic pressure [19].

It was also observed that the allelopathic effect of St Johnson wort extract on the germination and length of plumules of Johnsongrass and corn is low and due to osmotic pressure, while its effect on the germination of Redroot pigweed and Common lambsquarter and length of plumules of Common lambsquarter is evaluated as allelopathic effect.

Effect of St Johnson wort extract

Effect of St Johnson wort extract on the percentage of germination and length of radicles and plumules of Redroot pigweed, Common lambsquarter and Johnsongrass as weeds and corn as a cultivation plant is demonstrated in Table 2. Comparison of means exhibited that densities of 2.5, 3.75 and 10 percent and higher significantly reduced the percentage of germination of Common lambsquarter, Johnsongrass and Redroot pigweed. Its extract with densities of 15 and 20 percent decreased percentage of germination of corn significantly. St Johnson wort extract significantly decreased the radicle length of Common lambsquarter and Johnsongrass in all densities (Table 2).

However, its effect on the radicle length of Redroot pigweed and corn in densities of 2.5 and 3.75 was significant while compared to that of the control. Length of plumules of Redroot pigweed, Common lambsquarter and corn in densities of 3.75 percent and higher significantly decreased, while its effect on the plumules length of Johnsongrass in densities of 1.25 percent and higher was significant while compared to that of the control.

Table 2. Effect of St Johnson wort on Germination and Growth of weeds and corn seedlings

Treatment (density of extract)	Common lambsquarter			Redroot pigweed			Johnsongrass			Corn		
	Radicals (mm)	Plumule (mm)	Germination (%)	Radicals (mm)	Plumule (mm)	Germination (%)	Radicals (mm)	Plumule (mm)	Germination (%)	Radicals (mm)	Plumule (mm)	Germination (%)
0%	7.1^{a*}	7.7^{a}	39^{a}	13.4^{a}	11^{a}	45^{a}	21.7^{a}	27.9^{a}	42.5^{a}	36.9^{a}	11.9^{a}	43.5^{ab}
1.25%	4.4^{b}	8^{a}	32^{ab}	12.4^{a}	10.1^{a}	42.7^{a}	4.5^{b}	18.02^{c}	43.7^{a}	35.4^{a}	10.5^{ab}	45.5^{ab}
2.5%	2.2^{c}	8.2^{a}	26^{bc}	5.8^{b}	8.7^{ab}	40.7^{a}	2.7^{c}	17.8^{c}	38.7^{ab}	37.1^{a}	10.4^{ab}	46^{a}
3.75%	1.4^{c}	2.8^{b}	13^{dc}	3.9^{b}	6.5^{bc}	42^{a}	2.3^{c}	18.1^{c}	33.7^{bc}	27.6^{b}	7.4^{bc}	45.5^{ab}
5%	1.2^{dc}	2.6^{b}	21^{c}	3.3^{b}	5.3^{c}	43.5^{a}	1.5^{de}	21^{b}	32.7^{c}	17.2^{c}	7.1^{bc}	43.5^{ab}
10%	1^{de}	1.3^{bc}	7.5^{d}	0.05^{c}	0.8^{d}	0.75^{b}	1.1^{e}	9.6^{d}	30.5^{c}	17.5^{c}	7^{bc}	45^{ab}
15%	0.2^{de}	0.7^{cd}	3.5^{e}	0^{c}	0^{d}	0^{b}	1.02^{e}	7.6^{e}	23.7^{d}	14.5^{c}	4.3^{dc}	39^{bc}
20%	0^{e}	0^{d}	0^{f}	0^{c}	0^{d}	0^{b}	1.1^{e}	7.9^{ed}	19.7^{d}	14.5^{c}	4.1^{d}	34^{c}

Table 3. Effect of Polyethylene Glycon on germination and growth of weeds and corn seedlings

Treatment (desity of extract)	Common lambsquarter			Redroot pigweed			Johnsongrass			Corn		
	Radicals (mm)	Plumule (mm)	Germination (%)	Radicals (mm)	Plumule (mm)	Germination (%)	Radicals (mm)	Plumule (mm)	Germination (%)	Radicals (mm)	Plumule (mm)	Germination (%)
0%	14.65^{a}	8.65^{a}	36.2^{a}	23^{a}	14.65^{a}	45.5^{a}	18.4^{a}	35.32^{a}	33.7^{a}	38.2^{a}	17.95^{a}	45.5^{a}
1.25%	14.05^{a}	8.2^{a}	34^{ab}	20.97^{b}	14.2_{ab}	44.2^{a}	18^{ab}	34.65^{a}	32.7^{a}	33.8^{b}	17.85^{a}	44.5^{a}
2.5%	12.7^{b}	8.35^{a}	32.7^{b}	19.05^{c}	13.8^{ab}	42.7^{ab}	18.65^{a}	33.65^{a}	31.2^{a}	32.15^{b}	15.67^{b}	43.5^{ab}
3.75%	12.55^{b}	8^{a}	32.2^{bc}	19.45^{c}	13.65^{b}	40.7^{b}	17.92^{ab}	33.85^{a}	28.5^{b}	30.75^{bc}	15.45^{b}	42.5^{abc}
5%	11.6^{b}	7.15^{b}	29.7^{c}	13.9^{d}	9.3^{c}	40.7^{b}	16.7^{bc}	33.8^{a}	27.7^{b}	28.5^{cd}	15.45^{b}	42.5^{abc}
10%	9.25^{c}	6.7^{b}	27^{d}	12.6^{e}	8.45^{d}	33.5^{c}	19.9^{c}	30.65^{b}	20.2^{c}	28^{cd}	13.65^{c}	42^{abc}
15%	7.2^{d}	4.45^{c}	22^{e}	12.25^{ef}	7.55^{e}	25.2^{d}	12.45^{d}	24.5^{c}	16^{d}	26.05^{d}	13^{c}	41^{bc}
20%	6.8^{d}	3.75^{d}	13.7^{f}	11.5^{f}	5.6^{f}	17.7^{e}	10.1^{e}	21.9^{d}	13.5^{e}	17.3^{e}	4.4^{d}	40^{c}

Effect of polyethylene glycol

Effect of polyethylene glycols (Table 3) on germination percentage and length of radicles and plumules of Redroot pigweed, Common lambsquarter and Johnsongrass as weeds and corn as cultivation plant is demonstrated in Table 3. Results revealed that polyethylene glycol in densities of 3.75 percent and higher significantly decreased the germination percentage of Redroot pigweed and Johnsongrass. On the other hand, its effect on the germination of Common lambsquarter in densities of 2.5 percent and higher and in corn, in densities of 15 and 20 percent was significant. Polyethylene glycol significantly decreased the radicle length of Redroot pigweed and corn in all densities. Its effect on the length reduction of Common lambsquarter and Johnsongrass in densities of 2.5 and 5 percent and higher was significant while compared to the control. Comparison of the means showed that densities of 2.5, 3.75, 5 and 10 percent and higher significantly reduced the plumule length of corn, Redroot pigweed Common lambsquarter and Johnsongrass.

Comparison of figures

Comparing the effect of St Johnson wort extract with that of PEG diagram of the effects of St Johnson wort extract on the reduction of germination percentage in Common lambsquarter and Redroot pigweed has a sharper slope than that of polyethylene glycol (Fig. 1). This reduction is due to osmotic pressure as well as the allelopathic effects of St Johnson wort extract. The results suggested that due to parallelism and similarity between the graphs of PEG effects and those of St Johnson wort on the reduction of germination percentage of corn and Johnsongrass, one can attribute this reduction to osmotic pressure. Since equal densities of PEG and St Johnson wort extract demonstrated different effects, hence increase in the effects of St Johnson wort extract was attributed to the allelopathic effect (Fig. 1).

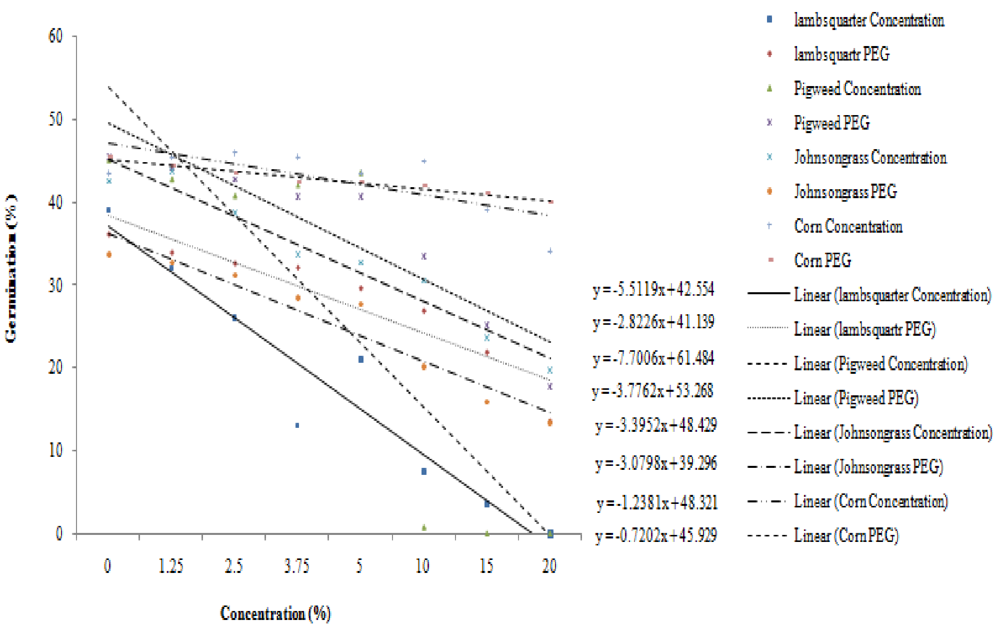

Fig. 1. effects of St Johnson wort extract and PEG on germination percentage in Common lambsquarter, Redroot pigweed, Johnsongrass and corn

Effects of St Johnson wort extract on the reduction of length growth of radicles and plumules in Redroot pigweed, Common lambsquarter, Johnsongrass and corn can be attributed to osmotic pressures because their graph is almost parallel with that of PEG (Fig. 2, 3).

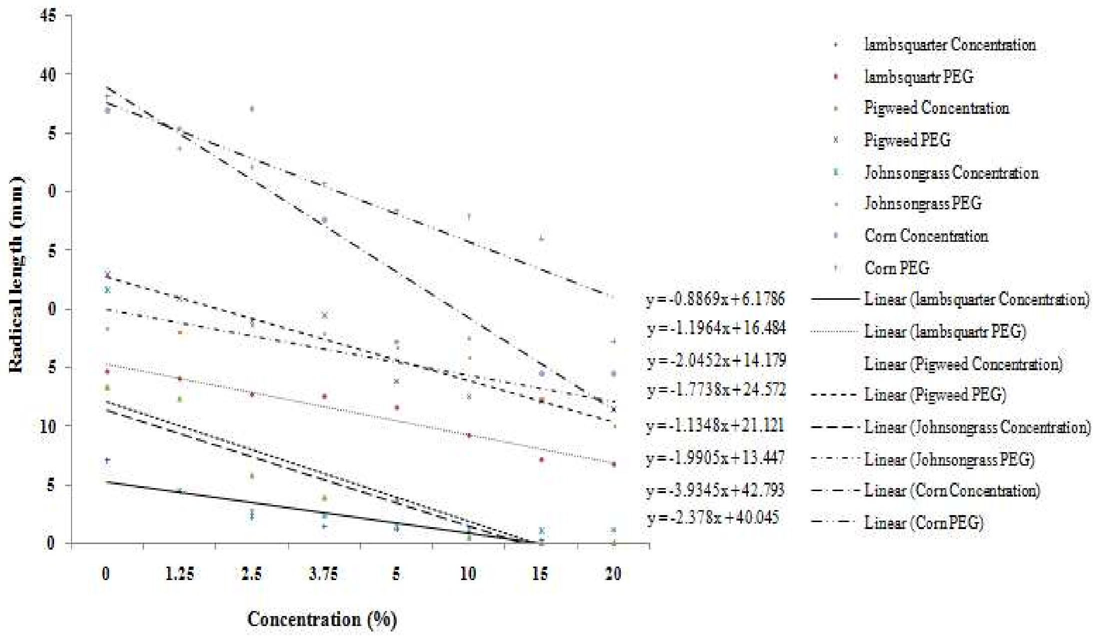

Fig. 2. Effects of St Johnson wort extract an PEG on the plumule length in Common lambsquarter, Redroot pigweed, Johnsongrass and corn

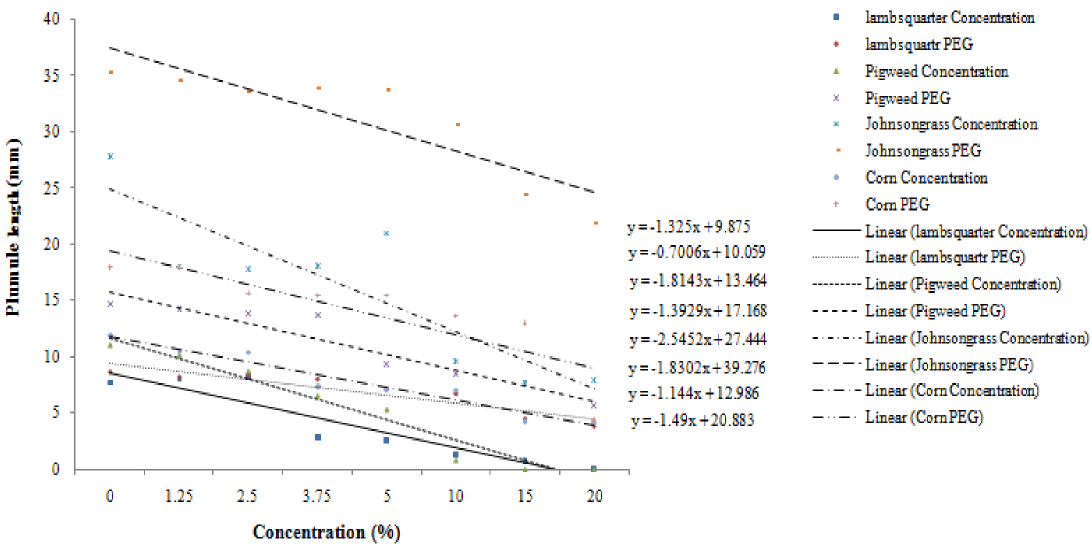

Fig. 3. Effects of St Johnson wort extract and PEG on radicle length in Common lambsquarter, Redroot pigweed, Johnsongrass and corn

DISCUSSION

Allelochemicals affect physiological processes in neighboring plants or mechanisms. Ingredients in medicinal of aromatic plants contain materials which have inhibitory effects on the germination and growth in other plants [12, 20, 21]. Results of this research and Cheng findings [5] indicated that the extract of aerial parts of medicinal plants had a significant effect on the reduction of germination and growth of the neighboring plants seedlings. These indicators demonstrated that in some plants, inhibitory chemicals exist mainly in the growing parts comparing to their under ground parts.

Our findings also showed that ingredients in the aerial parts of St Johnson wort medicinal plant have inhibitory effect on the germination and growth of seedlings of wild Redroot pigweed, Common lambsquarter and Johnsongrass as weeds and Corn as a cultivar, though this effect is different on seed germination and length growth of radicles and plumules. These results as well as those of researches conducted by Romangi and Tworkoski [24, 25] suggest that ingredients in the extract and essence of medicinal plants have strong herbicidal effects and in densities higher than 1 percent can prevent weeds from germinating (Fig. 1, 2, 3).

In consistence with Hartman et al. [12] and Rizvi and Rizvi reports [21], results of this research showed that St Johnson wort extract affects the length of radicle more than that of plumule in the plants study. This more effect on the radicle length is evaluated in terms of its direct contact with the extract. In higher densities, a reduction in the radicle growth comparing to plumule growth was observed which is due to the extract type, plant species and chemical properties of the allelochemicals.

Results of Fuji's experiments [8] on 239 medicinal plants displayed that ingredients in various parts of these plants have strong inhibitory effects. Intensity of this effect on radicles and plumule depends on the density of the extract as well as ingredients that form it. Moreover, with the osmotic pressure that exists in such extracts, their inhibitory effects can be attributed to the allelopathy phenomenon [19]. This result along with findings of Dayan et al. [7] showed that effect of St Johnson wort extract on the growth of seedling of cultivation plants is mostly due to the allelopathic phenomenon.

Studying the allelopathic effects of medicinal plants on controlling weeds is useful for organic agriculture and can be a proper guide for selecting alternation programs [14]. Cultivating medicinal plants which are resistant against cold weather in mild and cold regions has the advantages of autumn sowing. The advantage of raining during the growth season, decrease in illnesses and weeds are among advantages of placing medicinal plants such as camomile, oxtongue and maritigal in alternation programs [5].

Since weather conditions and genetic characteristics of plants are considered among the most important factors that affect the formation and diffusion of allelopathic materials, controlling weeds using this method in alternation systems requires supplementary experiments in farm conditions and on other cultivating types of corn [17].

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