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Full Length Research Article

PHYTOTOXIC EFFECT OF PLANT EXTRACTS FROM ASTERACEAE ON GERMINATION OF ECHINOCLOA CRUS-GALLI GROWTH

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ABSTRACT

In Egypt *Echinocloa crus-galli* is one of the most successful yield-limiting weeds in the rice fields that its control mostly relies on herbicides. Plant species *Nauplius graveolens, Picris asplenioides, Reichardia tingitana* and *Urospermum picroides* were collected from naturally growing population in coastal (Deltaic Mediterreanean coast) and inland desert WadiHagul of Egypt. The phytochemical analysis of the selected species in the present study showed that, they contain relatively high contents of tannins, saponins, flavonoids, alkaloids and phenols. At 500 μ g/ml the scavenging activity of *Nauplius graveolens, Picris asplenioides, Reichardia tingitana* and *Urospermum picroides* extracts were 14.3%, 6.25%, 7.73% and 4.14, respectively. The allelopathic effect of the extracts from the different tested species exhibited reduction of *Echinochloa crus-galli* seed germination. Also, the allelopathic activities of different extracts of the four species significantly inhibited shoot and root growth of *Echinochloa crus-galli* at both low and high concentrations.

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INTRODUCTION

Agriculture sector especially crop production is under immense pressure. Weeds are the most stubborn competitors of crops causing substantial reduction in yield by sharing light, air, water, nutrients and space (Semenov and Halford, 2009; McDonald et al., 2009). The phenomenon of allelopathy, where a plant species chemically interferes with the germination, growth or development of another plant species has been known for over 2000 years. Allelopathy refers to the beneficial or harmful effects of one plant on another plant, both crop and weed species, from the release of biochemicals, known as allelochemicals (Fraenkel 1959; Stamp 2003). Allelochemicals are diverse in nature and structure and thus lack common mode of action as well as direct and indirect effects on plants. Most allelochemicals are classified as secondary metabolites such as phenolic compounds, flavonoids, terpenoids, alkaloids, steroids, carbohydrates, and amino acids, with mixtures of different compounds sometimes having a greater allelopathic effect than individual compounds alone (Willis, 2010). Also, they are produced as off shoots of the primary metabolic pathways of the plant.

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These allelochemicals interfere with the cell division, hormone biosynthesis, mineral uptake and transport and membrane permeability (Rizvi et al., 1992; Kruse et al., 2000). Many plant species are most susceptible to allelochemicals at the seedling stage, thus phytotoxic activity of allelochemicals is responsible for growth suppression of weeds (Inderjit and Olofsdotter, 1998; Narwal et al., 1998). The family Asteraceae is one of the largest families of vascular plants. It is an advanced and botanically highly specialized family of mainly herbaceous plants. According to Funk et al. (2005), the Asteraceae is the richest vascular plant family in the world, with 1.600-1.700 genera and 240.00-300.00 species. This family is distributed over most of the earth and in almost all habitats particularly in semiarid region of the tropics, subtropics and warm temperate regions of South, Southeast and East Asia, Africa and Central South America (Rahman et al., 2008). The genera: Nauplius, Picris, Reichardia and Urospermum attracted the attention of many scientists to study their uses (El-Marsy et al., 1980; Abdel Salam et al., 1982; Amer et al., 1984; Akssira et al., 2006; Zidorn et al., 2007; Znini et al., 2012; Ramdani et al., 2014; El Alfy et al., 2015). In Egypt Echinocloa crus-galli is one of the most successful vield-limiting weeds in the rice fields which its control mostly relies on herbicides. Increasing use of herbicides might lead to enhance environmental pollution and human exposure to toxic materials (Zimdahl, 2004).

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Several workers have shown that allelopathy plays an important part in weed and weed interaction (Tajuddin *et al.*, 2002; Abd El-Gawad, 2014) and weed crop interaction (Olofsdotter *et al.*, 1999; Naderi and Bijanzadeh, 2012; El-Amier and Abdullah, 2014). In the present study, the phytotoxic effect of some wild Asteraceae species on germination and seedling growth of *Echinocloa crus-galli* growing in rice fields are evaluated. The phytochemical screening of the study species was investigated.

MATERIALS AND METHODS

Plant material

Plant species (*Nauplius graveolens, Picris asplenioides, Reichardia tingitana* and *Urospermum picroides*) were collected from naturally growing population in coastal (Deltaic Mediterreanean coast) and inland desert WadiHagul of Egypt. The plant material was handly cleaned, washed several times distilled water to remove dust and other residues, dried at room temperature in shaded place for several day till complete dryness and ground into powder, then preserved in well stopped bottles (AOAC, 1990).

Phytochemical analysis

Studied species were collected and prepared as previously mentioned. The phenol content was determined spectrophotometrically (Sadasivam and Manickam, 2008). Tannin was estimated according to the method of Van Burden and Robinson (1969). Saponin content was estimated by the method adopted by Obdoni and Ochuko (2001). Flavonoid content was estimated by Bohm and Kocipai-Abyazan, (1994), while alkaloids were determined according to Harborne (1973).

DPPH free radical scavenging activity

Antioxidant activity was determined by using a stable free radical (1,1-diphenyl-2-picrylhydrazyl) DPPH (Lim and Quah, 2007) as follows: 2 ml of 0.15 mM DPPH was added to 1 ml of various plant extracts (about 20 g of powdered samples were extracted with 200 ml of methanol 50 %) in different concentrations. A control was prepared by adding 2 ml of DPPH to 1 ml solvent (methanol 50 %). The mixture was incubated at the room temperature for 30 min. The absorbance was recorded at 517 nm and the antioxidant activity was expressed as:

% Radical scavenging activity = $[1 - (A_{sample}/A_{control})] \times 100$

Preparation of extracts

For bioassay tests, stock extracts (10% w/v) were diluted with distilled water to obtain various concentrations of 10%, 20%, 30% and 40% (w/v).

The solutions were filtered through double layers of muslin cloth followed by Whatman No. 1 filter paper. The pH of the mixtures was adjusted to 7 with 1 M HCl, and then mixtures were stored in a refrigerator at 4 °C until further use (Rice, 1972).

Allelopathy bioassay

Twenty seeds were placed on each filter paper in addition to 10 mL of plant extract for each petri dish. Five replicate samples were placed at 25 °C in a dark growth chamber for 15 days. A control sample was assigned with distilled water, a seed with a radical of 0.5 cm was considered to be germinated. Daily readings of the germinated seeds were recorded during the experimental period and final the measurements of the shoot and root growth were recorded.

RESULTS AND DISCUSSION

Phytochemical analysis

The phytochemical analysis of selected species (Nauplius graveolens, Picris asplenioides, Reichardia tingitana and Urospermum picroides) indicated that almost all plants are important and there is a need to develop techniques for judicious utilization and sustainable development (Table 1). Naupliusgraveolens contained highest contents of flavonoids (5.51mg/g dry weight) and alkaloids (4.67mg/g dry weight). Reichardia tingitana and Picris asplenioides contained highest contents of tannins (12.54 and 13.69 mg/g dry weight), saponins (7.13 and 5.27 mg/g dry weight) and phenolic (8.17 and 8.77 mg/g dry weight), respectively. Urospermum picroides low contents of active constituents, except phenolics (9.31 mg/g dry weight). The phytochemical analysis of the wild plants in this study are relatively comparable except to that obtained by El-El-Amier et al. (2015) on some wild Aizoaceae species (Aizoon canariense, Mesembryanthemum crystallinum, M. forsskaolii and M. nodiflorum) and Ramez (2015) on Launaea species (L. capitate, L. mucronata, L. nudicaulis and L. spinosa) growing in Egyptian Desert, but lower than those reported by El-Amier and Abdullah (2014) on Calligonum polygonoides, Cakile maritima and Senecio glaucus

Antioxidant activity

The evaluation of the antioxidant activity between the four plant extracts is showed in Figure 1. By increasing the plant extract concentration there was a corresponding continuous increase in scavenging activity. In case of *Nauplius graveolens, Picris asplenioides, Reichardia tingitana and Urospermum picroides* extracts the increase was up to 500 μ g/ml where the scavenging activity was 14.3%, 6.25%, 7.73% and 4.14, respectively.

Table 1. The concentration of the active constituents in mg/g dry weight for the selected plant species

Bioactive constituent	Reichardia tingitana	Nauplius graveolens	Picris asplenioides	Urospermum picroides
	mg/g dry weight			
Tannins	12.54±0.05	10.01±0.07	13.69±0.06	9.20±0.04
Saponins	7.13±0.06	6.08 ± 0.06	5.27±0.04	5.01±0.03
Flavonoids	5.06±0.10	5.51±0.07	4.80±0.06	4.24±0.04
Alkaloids	4.18±0.06	4.67±0.05	3.87±0.03	3.14±0.05
Phenolics	8.17±0.03	9.15±0.04	8.77±0.09	9.31±0.04

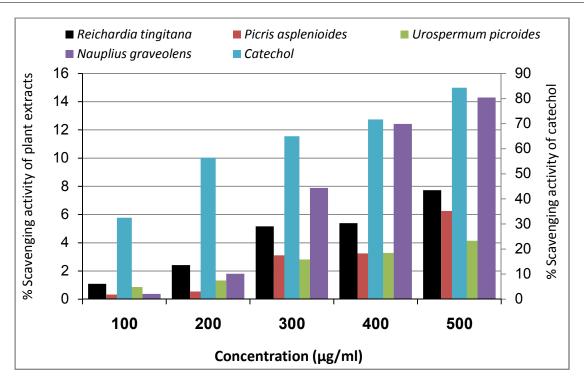


Figure 1. Scavenging activity of methanolic extract of tested species

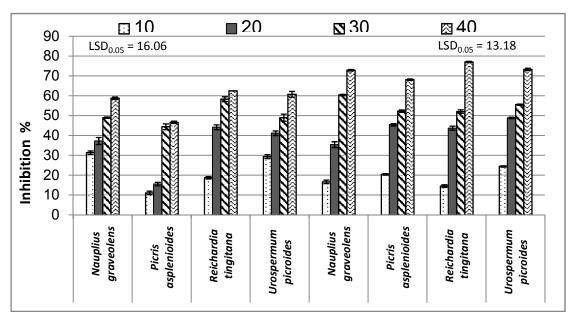


Figure 2. Effect of four plant extracts on the germination inhibition percentage (mean value ± standard error) of the Echinochloa crus-galli after 4 DAT

Allelopathic potentiality

This experiment aims to study allelopathic effect of four wild Asteraceae species (*Nauplius graveolens, Picris asplenioides, Reichardia tingitana* and *Urospermum picroides*). The aqueous and methanol extract were prepared in 10, 20, 30 and 40 g/l. The allelopathic effect of four species extracts on the germination percentage of *Echinochloa crus-galli* after four days after treatment (DAT) are shown in Figure 2. After 4 days of treatment, all extracts significantly reduced the germination of *Echinochloa crus-galli*. *Urospermum picroides* recorded the greatest inhibition percentage of 60.78% and 73.33% at 40 g/l concentration under aqueous and methanol

extracts, respectively. The extract of *Reichardiatingitana* attained the highest effective (77.08%) at 40 g/l under methanol extract, while under aqueous extract is 62.5% at 40g/l. *Nauplius graveolens, Picris asplenioides* attained the moderateallelopathic potential at different concentrations (Figure 2). The different extracts of four wild plants were also significantly inhibited shoot growth of *Echinochloa crus-galli* are showed in Figure 3, where the highest inhibition percentage was also recorded at 40 g/l, especially *Reichardia tingitana* (45.81% and 69.92%) under aqueous and methanol extracts, respectively. *Urospermum picroides* expressed less effect as it inhibited shoot growth (31.58%) at 40 g/l concentration under aqueous extract, while

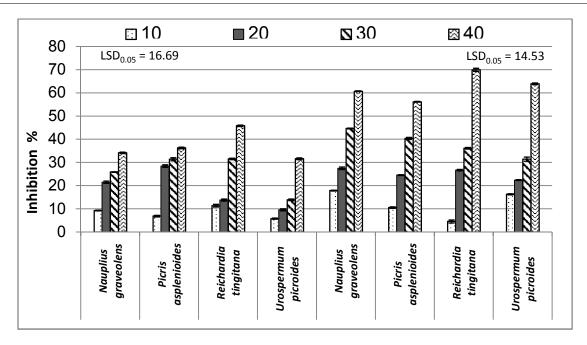


Figure 3. Effect of four plant extracts on shoot growth inhibition percentage (mean value ± standard error) of the Echinochloa crus-galli after 4 DAT

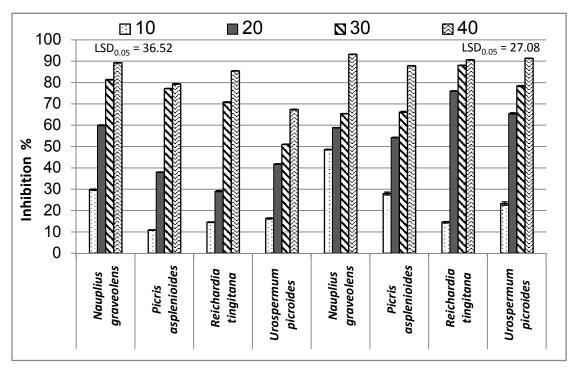


Figure 4. Effect of four plant extracts on root growth inhibition percentage (mean value ± standard error) of the Echinochloa crus-galli after 4 DAT

Picris asplenioides attained 56.14% at 40 g/l under methanol extract. Generally, all extracts of the tested wild plants significantly reduced the shoot growth of *Echinochloa crusgalli* at concentration-dependent manner. In many studies, it was found that root growth was inhibited more than shoot growth (Inderjit and Dakshini, 1995). On the other hand, root growth is sensitive to autotoxic chemicals at low concentrations, more than hypocotyl growth and seed germination (Chon *et al.*, 2000 and 2003). Also, the allelopathic activities of different extracts of the four species were significantly inhibited root growth of

Echinochloa crus-galli at both low and high concentrations (Figure 4). The extracts of *Nauplius graveolens* was the most effective as it inhibited root growth by about 89.23% and 93.22% at 40 g/l, followed by *Reichardia tingitana*, where extracts inhibited root growth by about 85.44% and 90.60% at 40 g/l and then *Picris asplenioides*, where extract inhibited root growth by about 79.35% and 87.83% at 40 g/l under aqueous and methanol extracts, respectively. *Urospermum picroides* was the least effective as it inhibited root growth by about 67.35% and 91.38% at 40 g/l under aqueous and methanol extracts, respectively.

The allelopathic inhibitions are the result of several compounds such as, phenolics, quinones, sesquiterpene lactones, alkaloids and others alter root morphology (Einhellig, 2002). Although, the cell membrane is an early interface with allelochemicals, relatively little attention has been given to membrane- related effects and their molecular targets (Inderjit and Mukerji, 2006). The phytochemical analysis of Nauplius graveolens, Picris asplenioides, Reichardia tingitana and Urospermum picroides in the present study showed that, they contain relatively high contents of tannins, saponins, flavonoids, alkaloids and phenols. Previous investigation on these plant species revealed that, the presence of sesqueterpine lactones and glycoside (Abdel-Mogib et al., 1993; Zidorn et al., 2007), phenolics (Recio et al., 1992) and essential oil (El Alfy et al., 2015) from Reichardia tingitana. Glucoside of urospermal A (Abdel Salam et al., 1982), sesquiterpene lactones and glycosides (Balboul et al., 1997) were isolated from Urospermum picroides.

Naupliolide, a sesquiterpene lactone (Akssira et al., 2006) and essential oil Znini et al. (2012) were isolated form Nauplius graveolens. Hence the allelopathic potential of the selected species could be attributed to these bioactive compounds. Aqueous extract of some plant species may contain some toxic substances (Habib and Abdul Rehman, 1988). These substances probably inhibit the germination and seedling growth of other plants species (Al-Charachafchi et al., 1987), which was due to their interference with indol acetic acid metabolism, or synthesis of protein and ions uptake by the plants (Hussain and khan, 1988). These phenolics inhibit the germination and seedling growth of same plant species or others by their effects on metabolic processes of germination and growth (Castro et al., 1984). Finally, it could be concluded that, the selected four wild species could be used as biocontrol or biofriend for controlling management of the nuisance weed Echinochloa crus-galli which intensively competing summer crops in Egypt.

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