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Full Length Research Paper

Scientific Communication Allelopathic influence of *Ipomoea batatas* (L.) Lam. commercial clone 'CEMSA 78-354' on weeds

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The allelopathic potential of extracts and residue of sweet potato (*Ipomoea batatas* L.) Lam. plant clone 'CEMSA 78-354' on weeds seeds germination was examined. Plant residues inhibited the weed emergence and weed seeds germination. The negative Response Index indicated that monocot weeds were less sensitive than dicot weeds. *I. batatas* plant parts extracts inhibited seed germination and stimulate radicle length of *P. oleracea* and *A. spinosus*. Many phytochemicals (fatty acids, triterpenes, steroids, alkaloids, quinones, phenols, tannins, flavonoids, saponins etc.) with allelochemical potential were detected in *I. batatas* plant.

Keywords: Allopathic effect, sweet potato, extracts, residue, *Portulaca oleracea*, *Amaranthus spinosus*.

INTRODUCTION

Ipomoea batatas is cultivated for food in > 100 countries (FAO, 2010) and is major crops in Cuba and developing countries. It ranks fourth after rice, wheat and yam (Macías *et al.*, 2008). The plants of genus *Ipomoea* (Convolvulaceae), suppresses the weeds growth (Rodríguez *et al.*, 1985). *Ipomoea* species are very aggressive and competitive, due to their strong propagative potential and high allelopathic interference (Anaya, 1989). This effect has been observed against *Portulaca oleracea*

L. and *Amaranthus spinosus* L. in tropical and subtropical countries (Blum *et al.*, 2002, Rodríguez *et al.*, 1985). Though *I. batatas* has been studied as an invasive species with allelochemicals potential (Chon and Boo, 2005; Hernández, *et al.*, 1999; Mbaeyi and Emejulu, 2013), but the commercial clone (CEMSA 78-354) has not been evaluated as bio-herbicide in integrated weed management programme (Hu *et al.*, 2004).

The sweet potatoes cultivars vary in the total phenolic content and the content of each phenolic component. Most of the phenolic components were of chlorogenic acid and other caffeoylquinic acids. Caffeic acid was < 10% of total phenolic content in four cultivars but was 36% in Jewel

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cultivar (Harrison *et al.*, 2003). Invasive plant species owe part of their success as invaders to the release of phytotoxic compounds that are toxic to the native species Kato *et al.*, 2015). This study aimed to assess the allelopathic potential of extracts and residues from *I. batatas* (L.) Lam. commercial clone 'CEMSA 78-354' and its effect on the germination of weed seeds.

MATERIALS AND METHODS

Bioassay:

The experimental treatments consisted of 3 factors: (i). Plant parts: 5 (Leaves, Inflorescence, Foliage, Stems, Tuber cortex), (ii). Extract concentrations 2 (0,1 %) and (iii) test weeds *Portulaca oleracea* L. (purslane) and *Amaranthus spinosus* L. (spin amaranth). The treatments were replicate five times in Completely Randomized Design.

Field Experiment

The experimental treatments consisted of 1 factor (dry leaves of *I. batatas* commercial clone 'CEMSA 78-354') with four levels (0, 1.4, 2.8, 4.2, 5.6 t/ha). The treatments were replicate five times in Completely Randomised Design. The dry leaves (105 days old) of *I. batatas*, were uniformly incorporated 5 cm deep in soil in plots (26 x 32 cm). The field soil was brown silaitic (Hernández *et al.*, 1999) and irrigated daily with the same volume of water that the weight lost in the parcels, irrigated initially at the 80 % field capacity. Each treatment was replicated 5 times in completely randomized design. The numbers of weeds in each plot were recorded at 30 days after the start of the experiment. The experiments were developed from October to November 2011, in the Faculty of Agricultural Sciences, Villa Clara, Cuba.

Weed species were identified as dicotyledonous and monocotyledonous using botanical descriptions according to Rodriguez *et al.* (1985). The allelopathic Response Index (RI) was calculated according to Richardson and Williamson cited by Hu and Zhang (2013). A positive RI value indicated a stimulatory effect and negative RI value indicated an inhibitory effect. The absolute value of RI reflected the intensity of allelopathic effect.

Extracts Preparation

Two types of extracts [aqueous and ethanolic extracts] were prepared, from the leaves, inflorescence, stems, tuber cortex and foliage of sweet potato. These tissues were dried and powdered with a mill of hammers (Veb Nosser 8225 Nossen). The aqueous extract was prepared

in dark, macerating 5 g of powder of each part of *I. batatas* in 150 mL of distilled water (1:30 w/v). The mixture was stirred on a magnetic stirrer for 5 min and stored for 24 h at room temperature. It was filtered under reduced pressure and the resulting solution (0.033 g mL⁻¹) was diluted in sterile distilled water to obtain 0.01 g mL⁻¹ (1.0 % w/v) concentration. The ethanolic extract was obtained in same way like aqueous extract except that; it was dried by roto-evaporation at 35 rpm/45°C. The pellets were resuspended in 50 mL of methyl cellulose (0.4% w/v), to get final solution (0.01 g mL⁻¹) (Takemura *et al.*, 2013).

Bioassay with extracts

The bioassay was conducted in Petri dishes (Ø 9cm) with Whatman No.1 filter paper. The *P. oleracea* seeds were disinfected with 1% sodium hypochlorite for 3 min and then rinsed many times with sterile water. *A. spinosus* seeds were scarified with 50% sulfuric acid for 5 min as per Aliero (2004). and then washed with sterile water for 10 min. Two concentration of extract [0 (control) and 0.01 g mL⁻¹ (1% w / v)] were applied at 2.5 mL per petri plate. The petri plates without extract were used as control. The treatments were replicated five times in completely randomized design. Fifty seeds each of *P. oleracea* and *A. spinosus* were sown/ Petri plate and then kept in humid chamber (diffused day light 12 h, 100 % relative humidity and temperature 26± 2°C).

After 18 h, the number of seeds germinated were recorded every 6 h. Total germination (TG) was calculated as per Bradbeer cited by Anjum and Bajwa (2005). The Germination Speed Index (GSI) was determined as per Maguire cited by Chiapusio *et al.* (1997) and allelopathic Response Index (RI) was calculated as per Richardson and Williamson cited by Hu and Zhang (2013).

Phytochemical in plant tissues

For this, 5 g dried leaf, tuber cortex and foliage of plant were immersed in 30 mL of Hexane, ethanol or distilled water (24 h). Aliquots of 1 mL were taken for different identification reactions and evaluated by categories: Absence (-), present in low concentration (+), Present in moderate concentration (++) and Present in high concentration (+++) (30).

Testing of various extracts for various chemicals was as per the Public Health Branch Standard 311/98 (MINSAP, 1998). Specific tests were done to detect the presence of each metabolites e.g. fatty acids, lipids or essential oils (Sudan's test), triterpenes and steroids (Liebermann-Burchard's test), lactones and coumarins (Baljet), alkaloids (Dragendorff's test), quinones (Borntrager's test), cardiac glycosides (Kedde's test), phenols and tannins (Ferric Chloride test), flavonoids (Shinoda's test), reducing sugar

(Fehling's test), free amino acids and amines (Ninhydrin test), saponins (Foam and Mucilage test), according to Bukar and Mudi (2011).

Total phenolic acids

The total phenolic acids in various plant parts were estimated as per Medina *et al.* (2009) with Gallic acid as the standard. To 80 μL sample, we added undiluted Folin-Ciocalteu-reagent (2.0 N). After 5 min, 20% (w/v) aqueous Na_2CO_3 (800 μL) were added, and the volume was made up to 4.0 mL with "nanopure" water. The control contained all the reaction reagents only without the extract. The solution was incubated at room temperature (25 $^\circ\text{C}$) in dark for 90 min. The absorbance was measured at 760 nm and compared to gallic acid equivalents, using a gallic acid (5, 10, 15 and 20 mg L^{-1}) standard curve. Additional dilution was done, if the absorbance was measured over the linear range of the standard curve. One g dry residue (45 $^\circ\text{C}$ / 7 days) of each organ was taken and following the procedure described earlier, the total amount of phenols was quantified in each plant organ. The results were expressed as mg of gallic acid equivalents (GAE)/g dry weight of plant material. All measurements were made in triplicate and the mean values were calculated.

Statistical analysis

The data were processed in the Statgraphics program Ver. 5.0. All results were tested for normality and homoscedasticity with the Shapiro-Wilk and Levene tests, respectively. Normal and homoscedastic data were analyzed with ANOVA followed by post hoc analysis with Fisher's Least Significance Difference (CD) at 5%.

RESULTS AND DISCUSSION

Plant residues and weed emergence

The effect of varying levels of plant residues on the emergence of mono and dicot weeds under field conditions is given in Figure 1.

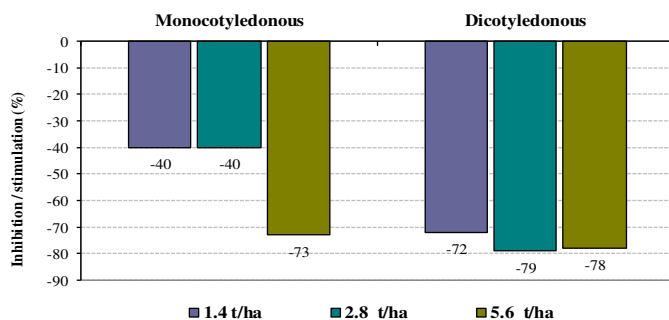


Figure 1. Effects of levels of *I. batatas* plant residues on germination of mono and dicotyledonous weed.

The differences between the doses of residues used for both the mono and dicotyledonous weeds, show a negative values of inhibition of the response index (RI). The highest activity was exerted on dicots with 72 – 79% of inhibition, while for monocots a significant inhibitory effect was noticed only at the highest dose (5.6 t ha^{-1}).

Similar results are reported Blum *et al.*, (2002) for incorporation of wheat (*Triticum aestivum* L.) residues for controlling germination of dicotyledonous weed species. Since the allelopathic effect depends largely on the concentration of allelochemicals released, it follows that lower doses produce little inhibition (An, 2005). Earlier, allelopathic activity has been attributed to allelochemicals present in cortex of sweet potato tuber (Chon and Boo, 2005; Dini *et al.*, 2009; Einhellig, 2002).

Effects of extracts on germination

Germination of both weed species was inhibited by the extracts of the vegetative parts of the sweet potato plant (Figure 2). The inhibitory activity depended on the plant part and the extract. Both leaf and root extracts showed inhibitory effects independent of the weed species and the type of solvent used in the extract preparation. Maximum inhibition of *A. spinosus* germination was seen when ethanolic extracts of leaves (100% of inhibition) and aqueous extracts of stems (92% of inhibition) were applied. Whereas for *P. oleracea*, maximum inhibition (9% of inhibition) was seen only when ethanol extracts of foliage and tuber cortex were used.

The extracts stimulated the growth of the radicle of *A. spinosus*, until 53% and 42% with aqueous and ethanolic extracts of the foliage of *I. batatas* (CEMSA 78-354), followed by inflorescence, leaves, tuber cortex and stems. It didn't happen this way in *P. oleracea*, where radicle grew more with the aqueous extract (33-40%), while with the ethanolic extract there was only stimulation about 24% with the leaves, but with the other organs it was imperceptible or lightly inhibitory (1-6%).

Inhibition of germination and growth of *Amaranthus hypochondriacus* L. and *Echinochloa crus-galli* (Beauv.) by aqueous extracts of *Ipomoea tricolor* L. is reported earlier with the former being affected more than the latter (Macías *et al.*, 2008). Takao *et al.* (2011) have also reported similarly with extracts of *Ipomoea cairica* (L.) Sweet.

Bigger sensibility has been demonstrated to the extracts of *I. batatas* (CEMSA 78-354) in *A. spinosus* that in *P. oleracea*. They have also been able to detect stimulating effects on the growth and inhibitory in the germination with same concentration of extracts. A bigger sensibility of the receiver species and the biggest accumulation of allelochemicals in the leaves of donor plant have been treated by An (2005) to describe the factors that determine the manifestation of the allelopathic phenomenon. Although the work carried out on sweet potatoes makes more emphasis in the allelopathic activity of the roots and

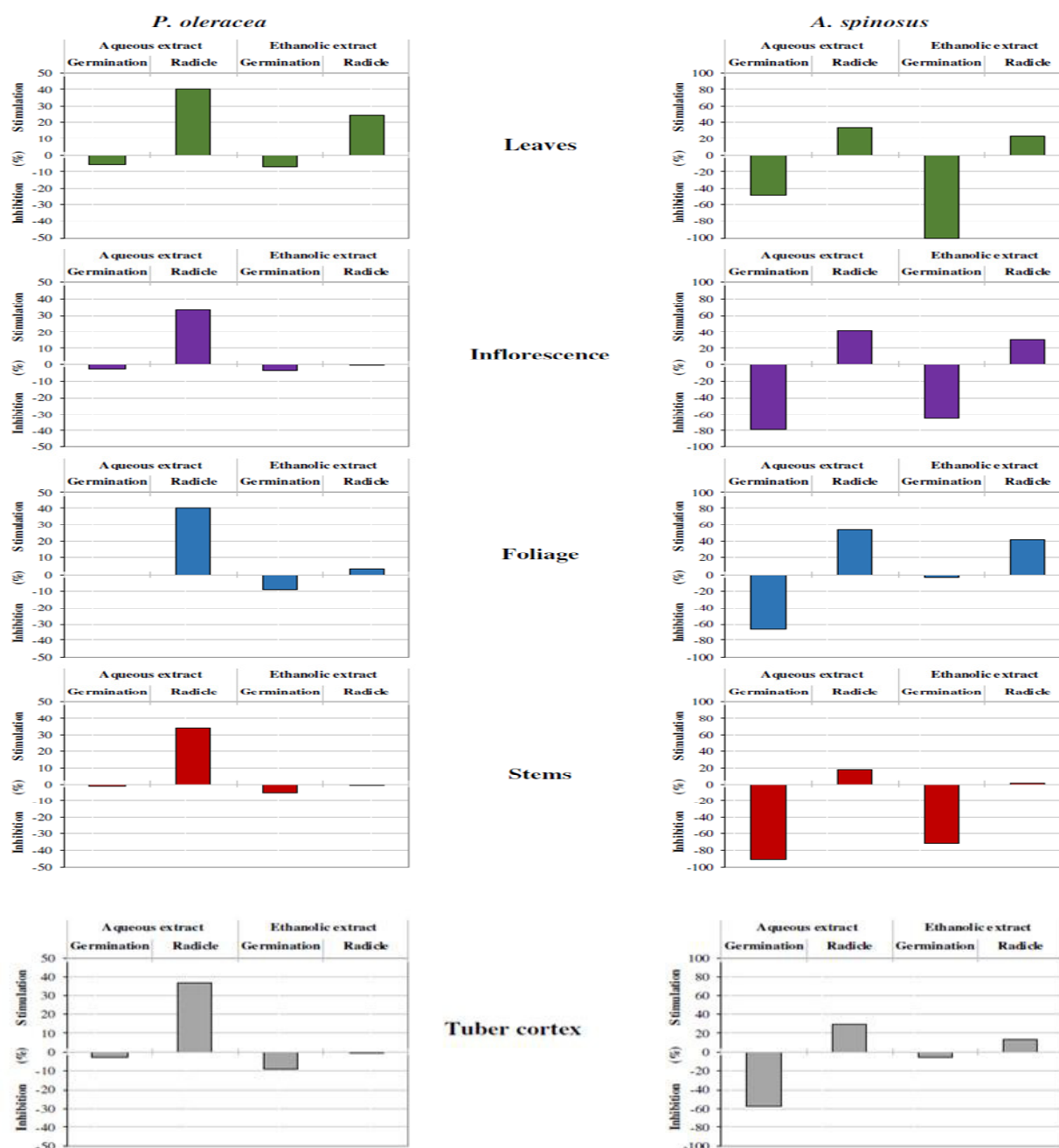


Figure 2. Inhibitory/Stimulatory effects of *I. batatas* (CEMSA 78-354) extracts on germination and growth of *P. oleracea* and *A. spinosus*.

tubers of this species (Rodríguez *et al.*, 1985; Harrison *et al.*, 2003; Harrison *et al.*, 2008).

Phytochemical screening of plant tissue

The results of qualitative screening of the various tissues for the presence of allelopathic substances is given in table 1.

Phytochemicals detected include fatty acids, steroids, triterpenes, alkaloids, quinones, phenols, tannins, flavonoids, reducing sugars, free amino acids and saponins.

Numerous studies confirm the abundance of phenols, tannins, reducing sugars, flavonoids, alkaloids and saponins in plants in the genus *Ipomoea*, including: *I.*

Table 1. Phytochemicals detected in *I. batatas* clone 'CEMSA 78-354' plant parts

Metabolites	Foliage			Leaf			Tuber cortex		
	1	2	3	1	2	3	1	2	3
Fatty acids	+	NA	NA	+	NA	NA	+	NA	NA
Triterpenes and steroids	++	NA	NA	++	NA	NA	+	NA	NA
Cumarins	-	-	NA	-	-	NA	-	-	NA
Alkaloids	-	+	NA	-	+	NA	-	+	NA
Quinones	NA	+	NA	NA	-	NA	NA	-	NA
Cardiac glycosides	NA	-	NA	NA	-	NA	NA	-	NA
Phenols / tannins	NA	+	+	NA	+	NA	NA	+	NA
Flavonoids	NA	-	+	NA	-	+	NA	-	+
Reducing sugars	NA	+++	+++	NA	+++	+++	NA	+++	+++
Free amino acids	NA	+	+++	NA	+++	+++	NA	+	+
Saponins	NA	-	+	NA	-	+	NA	+	-
Gums and mucilages	NA	NA	-	NA	NA	-	NA	NA	-

Key: 1= Hexane extract; 2= Ethanol extract; 3= Aqueous extract. + present in low concentration; ++ present in moderate concentration,+++ present in high concentration; - absent; NA: Not analysed

Table 2. Total content of phenolic compounds in organs of *I. batatas* 'CEMSA 78-354' expressed in terms of gallic acid equivalents (mg of gallic acid/g of dry weight of plant organ).

Organs	Concentration (mg/g DW)
Leaves	421.4
Inflorescence	671.2
Stems	244.7
Foliage	438.1
Tuber cortex	202.6

batatas, *I. obscura* L., *I. asarifolia* R. et Schult., *I. pes-caprae* (Linn.) Roth., *I. mauritiana* Jacq., *I. cairica* (Chon and Boo, 2005; Matunog, and Bajo, 2013; Mbaeyi and Emejulu, 2013). Some of these chemicals may have a role in allelopathy.

Total phenolic acids

The highest concentration of phenolics was found in the inflorescences (671 mg g⁻¹ dry matter), followed by foliage, leaves, stems and tuber cortex (Table 2). The aqueous extract had higher phenol content, indicating the presence of this phytotoxic compound in different parts of the plant. However, there was no inhibition with extracts of flowers of *P. oleracea* and *A. spinosus*, which could be due to lower concentration of phenols present in different tissues of *I. batatas*.

The presence of phenolics in the leaves and flowers of *I. cairicia* also been reported by Ralte (2014) and these may be released naturally by leaching from incorporated residues. Mbaeyi and Emejulu (2013) have reported the presence of as lower amounts of phenol (552.7 mg g⁻¹), in the leaves of *I. batatas* 'Suioh' compared to other species of *Ipomea* and the amounts were higher in aqueous extracts than in ethanol and peptone extracts (Harrison *et al.*, 2003).

Harrison *et al.*, (2008) found higher concentration of caffeic acid in the tuber cortex of *I. batatas* (0.047 mg g⁻¹). This however varied between clones and growing seasons, indicating that genetic and environmental factors are involved in determining the levels of these phenolic substances. The concentration of Caffeic acid in the periderm was higher than in the tuber cortex. Other substances, such as glycosidic resins, chlorogenic acid,

caffeoyl quinic acids, p-coumaric acid, Scopolin, and scopoletin have also been found in the periderm. and Coumarins, p-hydroxybenzoic acid, cynamide, 2-benzoxazolinone and scopoletin have been reported as germination inhibitors (Williams and Bartholomew, 2011).

CONCLUSIONS

Vegetative residues of *I. batatas* clone 'CEMSA 78-354' applied to the soil inhibited emergence of weeds, mainly dicotyledonous. In addition, ethanolic and aqueous extracts of aerial parts of the plant also inhibited germination of *P. oleracea* and *A. spinosus*. The germination (GSI) of *A. spinosus*, was delayed when seeds were exposed to the leaves and tuber cortex extracts. A variety of Secondary metabolites were identified in sweet potato plant parts which may be involved in allelopathic activity of this invasive plant. The highest content of total phenols was detected in the inflorescences followed by leaves, stems and the tuber cortex. More studies are needed to identify the allelochemicals involved in the allelopathic activity of the sweet potato clone 'CEMSA 78-354'.

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