

**COMPARATIVE PHYTOTOXIC EFFECTS OF AERIAL AND ROOT  
AQUEOUS EXTRACTS OF *SIDA CORDIFOLIA* L. ON GERMINATION  
AND SEEDLING VIGOUR PERFORMANCE OF LETTUCE,  
TOMATO AND CARROT**

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**Abstract**

A comparative study was conducted to see the effect of *Sida cordifolia* L. aerial and root extracts on seed germination and seedling vigour performance of *Lactuca sativa* (Lettuce), *Lycopersicon esculentum* (Tomato) and *Daucus carota* (Carrot). Four treatments were prepared 50, 25, 10 and 0 g/l (control). The result showed a significant decrease in germination and growth in all the treatments and regardless of concentrations lettuce was observed to be more susceptible in all extracts. Leaf extract exhibited the highest phytotoxicity followed by root while shoot extract produced the least effect. Inhibition index showed decrease in growth appeared to be concentration dependent. Liquid chromatography mass spectrum analysis of leaf and root extracts revealed presence of aliphatic acids notably stearic and palmitic which may act as inhibitory agents.

**Introduction**

Plant extracts are essential pool utilized for their medicinal and pharmaceutical importance and their secondary metabolites represent a large reservoir of structural moieties which work together thereby exhibiting a wide range of biological activity (Sharmila *et al.* 2007). Chemical interaction between plant species may hinder or promote growth through interference. It is a regular practice for control of weeds and could represent a good approach to reduce the use of pesticides (Nunes *et al.* 2015). Many of plant species have been reported to exhibit allelopathic activity (Khan *et al.* 2009) which inhibited germination (Haq *et al.* 2010). Plants compete for nutrients and important resources and these interactions are thereby involved in chemical signaling such as allelopathy. The use of plants with strong allelopathic properties has continued to show promising result. Allelochemicals are usually released by plant into the environment mainly via leachate or root exudates. Generally, plant residue contains a wide range of chemical compounds such as alkaloids, flavonoids, phenolics, coumarins, terpenoids etc. and many of these compounds are known to enhance the growth of certain plants while inhibit other (Visioli *et al.* 2011). Recently much attention has been focused to allelopathic effect of plants as a mean of biological control of weeds (Sahid and Yusoff 2014).

*Sida cordifolia* L. belongs to the family Malvaceae is an annual herb plant native to India, Australia, west and east Africa. It is extensively used in ethno-botany, medicinal and antimicrobial activity (Sivapalan 2015). Study regarding its potential application as bioherbicide is lacking, this study aimed at evaluating the effect of aerial and root aqueous extracts of *S. cordifolia* on germination and early seedling growth of the *Lactuca sativa* (lettuce), *Lycopersicon esculentum* (tomato) and *Daucus carota* (carrot).

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### Materials and Methods

Fresh foliage, shoot and roots of *Sida cordifolia* were collected at flowering stage between February and March, 2015 at Ladang 10 Faculty of Agriculture, Universiti Putra Malaysia (2°, 58', 65' N and latitude 101°, 42', 46' E and 52 m above sea level). The plant materials were shade-dried and pulverized to fine powder. Fifty g (T<sub>1</sub>), 25 g (T<sub>2</sub>) and 10 g (T<sub>3</sub>) and 0 g (T<sub>4</sub>) powder from each part of the plant was soaked in 1000 ml distilled water and shaken for 48 hrs using orbital shaker at room temperature then filtered through 1 mm sieve, and later passed through Whatman filter paper. Distilled water served as control. Lettuce, tomato and carrot seeds were obtained from Green Eagle Seed, Malaysia. These plants are commonly used species in the bioassay (Aslani *et al.* 2014) due to their sensitivity to phytotoxins and uniform germination potential. Seeds were sterilized with 1% sodium hypochloride for 5 min then rinsed severally with distilled water. Ten seeds were placed onto Petri dish on a double layer Whatman filter paper and 5 ml of different concentrations were added and placed in a growth chamber at 30°C/25°C, 70% humidity and 12 hrs photoperiod day/night. Distilled water was used for the control. Treatments were arranged in CRD with six replicates. Data were collected on germination, radicle and shoot lengths at 5 days after sowing, 50 g powder of leaves were soaked in 1000 ml of 70% aqueous methanol into a conical flask and then wrapped with paraffin and shaken in an orbital shaker at 25°C for 48 hrs (Aslani *et al.* 2013). Twenty mg crude powder was diluted into 20 ml 100% methanol (HPLC grade) and filtered with 0.2 µm nylon membrane prior to injection. The sample was analyzed on Acquity UPLC C18 100 mm × 2.1 mm × 1.7 µm at 30°C eluted with a 60 min 5-95% acetonitrile and Isopropanol at a flow rate of 0.3 ml/min. Water Single Quadrapule Detector (SQD) at 150°C at 3.9 kv capillary voltage and 3v extractor.

Germinability was determined upon protrusion of the radicle by more than one millimeter while smaller size lengths in shoot and root were measured. All parameters were statistically analyzed using ANOVA and means were separated by least significant difference (LSD) using SAS software 9.4 version.

### Results and Discussion

The result of The present experimentation indicated that leaf, root and shoot leachates of *Sida cordifolia* contain germination and growth inhibitors that are proficient to reduce growth of lettuce, tomato and carrot. Lettuce seeds treated with leaf aqueous extract had significantly decreased ( $p < 0.05$ ) germination ranged from 100, 70, 36.6 to 16% in T<sub>4</sub>/T<sub>3</sub>/ T<sub>2</sub>/ T<sub>1</sub> treatments, followed by tomato 96.6 to 23.3%, then carrot 100 to 26.6% (Fig. 1a).

Similar decrease in germination was also significantly high in lettuce treated with root aqueous extract at T<sub>1</sub> and T<sub>2</sub> with 30 and 53.3% (Fig. 1b) compared to tomato (36.6 and 60%) and carrot (40 and 56.6%). Though all the crops recorded high germination value with shoot aqueous extract but lettuce at T<sub>1</sub> exhibited the highest germination of 50% (Fig. 1c) while at T<sub>2</sub> and T<sub>3</sub> tomato appeared less affected (70 and 86.6%) followed by carrot (73.3 and 86.6%). Germination and growth inhibition are substantial parameters used in bioassay studies (Ziebrahimi *et al.* 2007). The findings of this study was supported by Aslani *et al.* (2014, 2015) who reported application of both *Tinospora tuberculata* leaf and stem extracts significantly reduced percentage of germination of lettuce, cucumber, carrot, tomato, rice and weedy rice with increase in extract concentration as compared to control. The aqueous extract of *Parthanium hysterosphorus* leaf and flower seriously inhibited seed germination and seedling growth of lettuce (Mulatu *et al.* 2005).

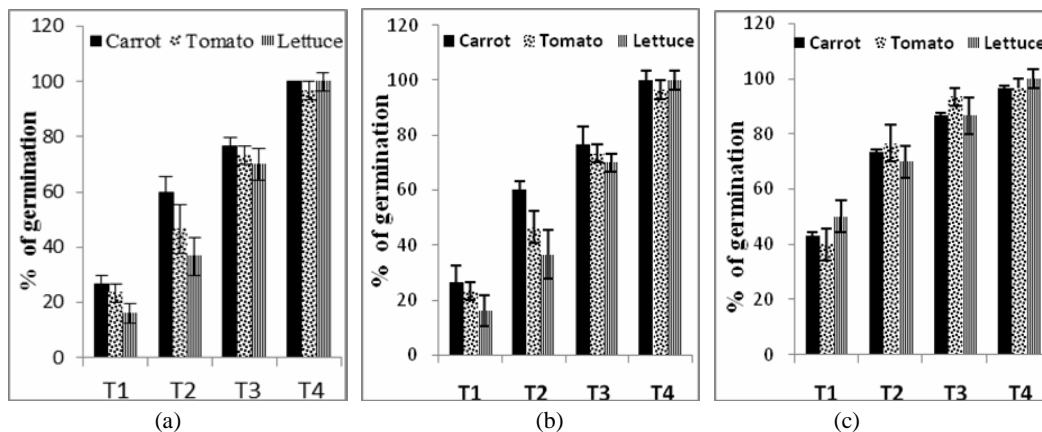


Fig. 1. Impact of *Sida cordifolia* (a) leaf, (b) root and (c) shoot aqueous extract on seed germination of carrot, tomato and lettuce.

Shoot length in carrot treated with leaf extract showed greater reduction 0.63 cm at T<sub>1</sub> followed by lettuce 0.66 cm then tomato 2.03 cm. Lettuce shoots significantly ( $p < 0.05$ ) indicated low growth with the root extract at T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> (2.01, 3.33 and 3.86 cm) as compared to the control (5.43 cm), similar trends were also observed in carrot and tomato (Table 1). Shoot extract appeared to induce more decreased in carrot shoot length among the crops while in lettuce no difference was observed among the treatments except with the control. Similarly, Guang *et al.* (2009) reported root exudate of hot pepper had strong allelopathic effect on lettuce and lettuce radicle length was found to be more sensitive to the allelochemicals than watermelon root growth (Hao *et al.* 2007). Generally, lettuce appeared more sensitive and susceptible to different extracts. Similar findings were reported by Santos *et al.* (2015). Root length appeared minimum in lettuce treated with leaf extract with significant reduction at highest concentration followed by carrot then tomato. All the treatments are significantly different at 5% (Table 1). Minimum radicle elongation with shoot extract was observed in the lettuce followed by carrot then tomato. Carrot radicle length was decreased by application of the root extract at different treatment levels ranged from 4.02 to 1.60 cm at T<sub>3</sub> and T<sub>1</sub> with the control having 4.25 cm. Significant difference was observed between the treatments across the crop species (Table 1).

Reduction in root growth was severe perhaps due to its direct contact with the extract. During the study it was observed that the root tips swelled, darken and seized to advance upon contact with the leachate concentrations thereby suggesting induced phytotoxicity. Visible effect of allelopathy includes decrease in germination, shoot and root inhibition, discoloration, swelling of root tips, necrosis and lower reproductive capacity (Niakan and Saberi 2009). Generally the extract had demoted shoot and root length as the concentrations increased. Growth reduction across the extracts on the species followed the order: leaf extract > shoot extract > root extract.

Phytochemical analysis of leaf and root of *Sida* revealed presence of aliphatic acids (stearic and palmitic) (Fig. 2). Stearic and palmitic acids are important allelochemicals that caused significant inhibition in growth of rice seedling (Tsuzuku *et al.* 1987) and exert cytotoxic effect on the plasma membrane and alter membrane permeability (Wu *et al.* 2006). The aerial part of *Sida* is famous to constitute malvic, fatty acids and many cytotoxic alkaloids compounds (Nagashayana *et al.* 2000). Present findings indicated the existence of aliphatic fatty acids allelochemicals in the extracts and revealed *S. cordifolia* aptitude towards allelopathic inhibition. Reduction in root growth was severe perhaps due to its direct contact with the extract. During the study, it was

**Table 1. Shoot and radicle length of lettuce, tomato and carrot treated with difference aqueous concentrations of root leaf, root and shoot extract of *S. cordifolia*.**

Treatment	Leaf extract		Root extract		Shoot extract	
	Shoot length (cm)	Radicle length (cm)	Shoot length (cm)	Radicle length (cm)	Shoot length (cm)	Radicle length (cm)
<b>Carrot</b>						
T1	0.63 ± 0.35c	0.50 ± 0.11c	2.53 ± 0.29c	1.60 ± 0.22b	2.05 ± 0.22c	1.83 ± 0.20c
T2	3.00 ± 0.32b	2.36 ± 0.27b	3.93 ± 0.35b	3.73 ± 0.44a	3.50 ± 0.28b	2.66 ± 0.17b
T3	3.95 ± 0.36a	3.05 ± 0.23b	4.05 ± 0.24b	4.02 ± 0.79a	3.66 ± 0.31b	3.13 ± 0.36b
T4	4.46 ± 0.16a	4.25 ± 0.42a	4.46 ± 0.16a	4.25 ± 0.42a	4.46 ± 0.16a	4.25 ± 0.42a
<b>Tomato</b>						
T1	2.03 ± 0.36b	2.00 ± 0.20d	2.81 ± 0.28d	2.46 ± 0.21c	2.48 ± 0.11c	2.50 ± 0.55b
T2	5.06 ± 0.48a	4.25 ± 0.50c	4.13 ± 0.58c	3.70 ± 0.25c	5.68 ± 0.64b	5.41 ± 0.62a
T3	5.71 ± 0.39a	6.96 ± 0.49b	5.28 ± 0.37ba	5.28 ± 0.37b	6.40 ± 0.53a	7.85 ± 0.50a
T4	6.66 ± 1.08a	9.4 ± 0.80a	6.66 ± 1.08a	9.40 ± 0.80a	6.66 ± 1.08a	9.40 ± 0.80a
<b>Lettuce</b>						
T1	0.66 ± 0.42c	0.08 ± 0.05c	2.01 ± 0.27c	1.85 ± 0.24c	2.83 ± 0.36b	1.63 ± 0.40c
T2	2.83 ± 0.49b	3.30 ± 0.50b	3.33 ± 0.25b	3.91 ± 0.58b	3.00 ± 0.18b	4.06 ± 0.49a
T3	3.86 ± 0.25b	5.28 ± 0.48ab	3.68 ± 0.29b	5.21 ± 0.28ab	3.36 ± 0.23b	5.11 ± 0.86a
T4	5.43 ± 0.45a	6.33 ± 1.31a	5.43 ± 0.45a	6.33 ± 1.31a	5.43 ± 0.45a	6.33 ± 1.31a

Values are expressed as mean ± S.E. Mean with the same letter in the column are not significant at 5%.

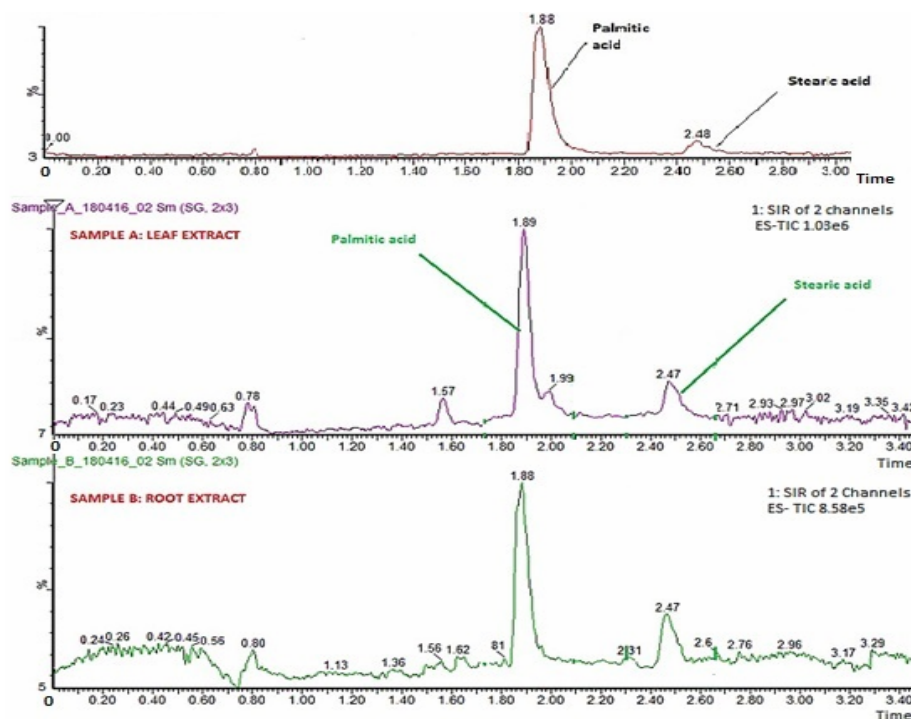


Fig. 2. Chromatogram of major fatty acids compounds detected in the leaf and root extracts of *Sida cordifolia* (top) standards peaks of stearic and palmitic acid, (bottom) leaf and root samples analyzed.

observed that the roots tips swelled, darken and severely seized to advance upon contact with the leachate concentrations thereby suggesting induced phytotoxicity. Visible effect of allelopathy includes decrease in germination, shoot and root inhibition, swelling of root tips and necrosis (Niakan and Saberi 2009). Stearic and palmitic acids are important allelochemicals that caused significant inhibition in growth of rice seedling (Tsuzuku *et al.* 1987).

The observed variability in growth indicated that *Sida* leaf has great phytotoxicity on germination, radicle and shoot growth with inhibition order lettuce > tomato > carrot. LC- MS analysis confirmed the presence of aliphatic acids in *Sida* extract which might be capable of inducing allelopathic effect on the tested crops. The present investigation revealed *S. cordifolia* aptitude towards allelopathy and potential use in natural weed management. Further identification of allelochemicals and mechanism of activity is needed.

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