

Inhibitory and stimulative effects of *Lantana camara* L. leaves on *Lycopersicum esculentus* and *Lactuca sativa* seedlings

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ABSTRACT

The study was carried out to determine the allelopathic effects of these mulches on germination and early growth of *Lycopersicum esculentus* and *Lactuca sativa* which are common vegetables in Zimbabwe. Laboratory and greenhouse experiments were conducted. A laboratory experiment with six treatments: 0, 1, 2, 3, 4 and 5 % aqueous concentration of *Lantana camara* leaf extracts was set up in the laboratory and was replicated three times and repeated twice over time. A greenhouse experiment in which *L. camara* dry matter was incorporated into the soil with 0, 1, 10, 20, 30, 40 and 50 grammes per pot of dry leaf biomass replicated three times and repeated twice over time was also set up. The results indicated that *Lantana* leaf extracts significantly reduced ($P < 0.05$) the germination, radicle length and plumule length of tomatoes but not for plumule length for lettuce. The level of inhibition was positively correlated to the concentration of the *Lantana* extracts on both vegetables. However, in the greenhouse, biomass of both vegetables was not affected by increase in *Lantana* concentration up to 40 g/pot. In this study, inhibitory effects of *Lantana* were observed in the laboratory but not in the green house where ground leaf biomass was mixed with red clay soil with a clay content of 33.5 %. The dry matter of lettuce increased with increase in *L. camara* up to 30 g/pot and then decreased. For tomatoes, it increased up to 40g/pot and then decreased raising the possibility of using reduced *lantana camara* rates to stimulate dry matter accumulation for both tomatoes and lettuce.

Keywords: *Lantana camara*, allelopathy, lettuce, tomato, seed germination

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1. Introduction

Lantana camara L. is a notorious global land invader that has spread rapidly in many of the 60 regions of the world to which it has been introduced by humans and is listed among the world's top 100 invasive species (Lowe et al., 2000; Vardien et al., 2012; El-Kenany et al., 2013). It is one of the most conspicuous invaders in the savanna ecosystems world-wide (Foxcroft et al., 2010). *Lantana* grows in a wide range of environmental conditions, infesting millions of hectares of natural ecosystem and cultivated lands causing great damage to biodiversity (Kathkayan et al., 2014). The

plant is also strongly allelopathic, having several compounds which repel or toxify vegetation thereby preventing their growth (Ahmed et al., 2007). Allelochemicals are present in all parts of the shrub and upon their release in the surrounding; these chemicals interfere with the germination and establishment of many species (Ambika et al., 2003; Bais et al., 2004; Kumar et al., 2011). Concurrent with this feature, *L. camara* has potential to prevent natural regeneration of some plants, blocks succession and replaces native species and thus it eventually threatens natural biodiversity (Ambika et al., 2003).

Tomato (*Lycopersicon esculentus*) is one of the most grown horticultural crops worldwide and the daily consumption of the crop is high in most households (Zivenge et al 2012). In Zimbabwe tomato production improved since recording by FAO in 1961, from 6 500 to over 23 744 tonnes, with significant drops between 1991 and 1992 from 15 000 to 9000 tons (FAOSTAT, 2014). Most of the tomato growers in Zimbabwe are located in Mutoko, 140 Km north east of Harare and 30km north of Harare in Chinamhora (Dobson *et al.*, 2001). They are mainly located within a radius of 50km from Harare and other major cities as these provide a good market for the tomatoes. Lettuce (*Lactuca sativa* L.) is an important vegetable grown in Zimbabwe for making sandwiches. It has gained countrywide acceptance and it is being grown both commercially and by small scale rural farmers who target urban markets. It is therefore imperative that the production of tomatoes and lettuce be optimized in Zimbabwe.

In agro-systems, *Lantana* allelopathy may affect the economical outcome of crop production. Suitable manipulation of the allelopathy towards improvement of crop productivity and environmental protection through eco-friendly control of weeds, pests, crop diseases and synthesis of novel agrochemicals based on natural products have gained attention of scientists (Bojovi and Jakovljević (2015). Ahmed et al., (2007) and Sahid and Sugau (1993) found that different concentrations of aqueous leaf extracts of *Lantana camara* caused significant inhibitory effect on germination, root and shoot elongation and development of lateral roots of some agricultural crops. It has been established that *L. camara* contain toxic compounds which include cyanogenic compounds, alkaloids, benzoxazinones (Putnam, 1988). However, in many cases it has also been reported that the responses to allelochemicals demonstrate stimulation at low concentrations and inhibition at higher concentrations (Fischer, 1986; Liu de and An, 2005); Belz, 2008; Hong et al.,

2008). Chang and Nelson (2010) reported that plant growth is stimulated below allelopathic thresholds with severe reductions above the threshold concentration and that depends on the sensitivity of the receiving species. Studies by Cheng et al., (2016) found that low concentrations of garlic had no negative effect on tomato seedlings and it was because the low concentrations tested were below the minimum inhibitory concentrations. In another study by Haq et al., (2010), post emergent sprays using mulberry leaf leachates promoted the growth of wheat and this raises the possibility that allelochemicals could be exploited for the benefit of agriculture.

It appears that if invasives could be used gainfully it may offset the costs of mechanically removing them and also exercise some control over their spread (Karthikeyan et al., 2014). Leaves of *L. camara* have been used as mulch in communal gardens and nurseries in Zimbabwe for moisture conservation and provision of shade in nurseries. By using mulch rates below the allelopathic threshold, crop production could be increased with subsequent benefits of lowering the spreading of the invasive *Lantana camara*. Despite its recognition as one of the worst invasive alien species in the world, information on the ecological interference of *L. camara* on the germination and early growth of horticultural crops is scanty in Zimbabwe. In this present study, the allelopathic effects of leaf extracts and leaf incorporation were evaluated on tomatoes and lettuce which are common vegetable crops for the small holder sector.

2. MATERIALS AND METHODS

2.1 Experimental site

The study was carried out at the University of Zimbabwe (UZ) in the Crop Science Department in Harare. UZ (17.78° S, 31.05° E) has an altitude of 1 523 meters above sea level and has an annual rainfall of 1

000 mm and temperature range of 25-30 °C. The germination experiment was carried out in the Weed Science Laboratory and the pot experiment was carried out in the glasshouse. UZ red clay soil (33.5 % clay, 9.5 % silt and 57 % sand) was used in the pot experiment.

2.2 Materials

Fresh leaves of *L. camara* were collected from the Faculty of Agriculture fields at the University of Zimbabwe between December 2013 and January 2014. *L. camara* leaves were washed thoroughly with tap water to remove any dirt and dust. The leaves were air dried at room temperature (approximately 25-30 °C) for three weeks in the Weed Science Laboratory. During drying, the leaves were continuously turned over after every two days to facilitate uniform drying. After drying the leaves were ground into a fine powder using a Thomas-Wiley Laboratory Mill (model 4) with 0.5 mm sieve size. Moneymaker: an indeterminate variety of tomato was used in the assays and the lettuce variety used was Commander.

2.3: Effect of *L. camara* leaf extracts on the germination, radicle and plumule growth of tomato and lettuce

2.3.1 Extract preparation

L. camara leaf biomass powder of 10 g, 20 g, 30 g, 40 g and 50 g was soaked in 1000 ml distilled water respectively to give the following aqueous extracts 1, 2, 3, 4 and 5 % w/v respectively. The aqueous leaf extracts were left to soak for 24 hours at room temperature before being filtered through a 0.045 mm Rahmen Analysensieb Retscher sieve to remove solid materials. The resultant filtrate was then filtered using whatman no. 2 filter paper. The filtrate was collected in 1000 ml beakers, labeled and stored at 4 °C.

2.3.2 Tomato and lettuce germination test

The germination bioassays were laid out as randomized complete block designs with the six treatments replicated six times. Germination tests were done in 9 cm diameter Petri dishes lined with whatman paper number 2 filter paper. Ten seeds of each assay plant (tomato and lettuce) were placed in separate petri dishes. The seeds were moistened by adding two millilitres of the different *L. camara* filtrate concentrations to each respective Petri dish using a five millilitres syringe. Two millilitres of the filtrate were added once a day as the moisture levels decreased so as to keep the filter paper moist. Furthermore, the Petri dishes were kept closed to conserve moisture. The controls were prepared using two millilitres of distilled water instead of the extract.

Germination counts were done daily for five days after germination. Germination was considered to have occurred when the radicle emerged from the seed coat. Five seedlings were chosen at random in each Petri dish and the radicle and plumule lengths were measured using a 30 cm ruler at the termination of the experiment at thirteen days from setting up the experiment. Germination percentages were calculated for the ten seeds. The formula used for calculating the germination percentage is given below:

$$\text{Germination \%} = \frac{\text{germinated seeds} \times 100}{\text{Total number of seeds (10)}}$$

2.4 Effect of *L. camara* leaf biomass applied at different rates on the days to emergence and biomass of tomatoes and lettuce

2.4.1 Experimental design and experimental Procedures

The glasshouse experiment was arranged in a randomized complete block design with six treatments replicated six times. The treatments were laid out in six blocks and

the blocking factors were the effects of shade and the position from the windows and doors.

Pots measuring 30 cm in diameter and 32 cm height were three quarter filled with the UZ red clay soil. 10 g, 20 g, 30 g, 40 g and 50 g of *Lantana* dry leaf biomass were mixed with the top two centimeters of the soil. In the control treatment, no powder was mixed with the soil. Ten seeds of each assay plant i.e. Moneymaker tomato variety and Commander lettuce variety were planted in each pot at a depth of one centimeter. The pots were watered daily using tap water using a watering cane fitted with a fine rose and no fertilizer applications were done.

The numbers of emerged tomato and lettuce seedlings were recorded from 5 to 27 days after planting. The percentage emergence was calculated using the formula shown below:

$$\text{Emergence\%} = \frac{\text{emerged seeds}}{\text{Total number of seeds planted (10)}} \times 100$$

3. Results

3.1 Lettuce germination, plumule and radicle length

The relationship of *L. camara* concentration and lettuce germination was significantly linear ($p < 0.01$) and the correlation ($r^2 = 0.909$) was high. The control treatment had

Dry biomass of the ten seedlings of each vegetable crop were harvested on the last day of the experiment, i.e. at day 27. Harvesting was done by uprooting all the plants and washing the roots with water to remove the soil. Plants from the same pot were placed in the same labeled envelope and oven dried for 48 hours at 80 °C. The dry matter of each crop was weighed using a sensitive Mettley digital scale.

2.4.2 Data analysis

Regression analysis for the germination assay data was done to determine the relationship of *L. camara* concentrations on the seed germination, radicle and plumule length of each crop using Genstat version 14. Final emergence counts from the glasshouse experiment data were subjected to Analysis of Variance (ANOVA) using Minitab version 14. Regression analysis was done to relate *L. camara* biomass rates (g/pot) to tomato and lettuce parameters (emergence counts and dry matter) using Minitab version 14, Sigma Plot was used for the graphs.

high lettuce germination as compared to the other treatments. Increasing the concentration of *L. camara* extract from 0 to 5 g per litre decreased the germination of lettuce seeds, nearly completely inhibiting it at the highest concentration (Figure 1).

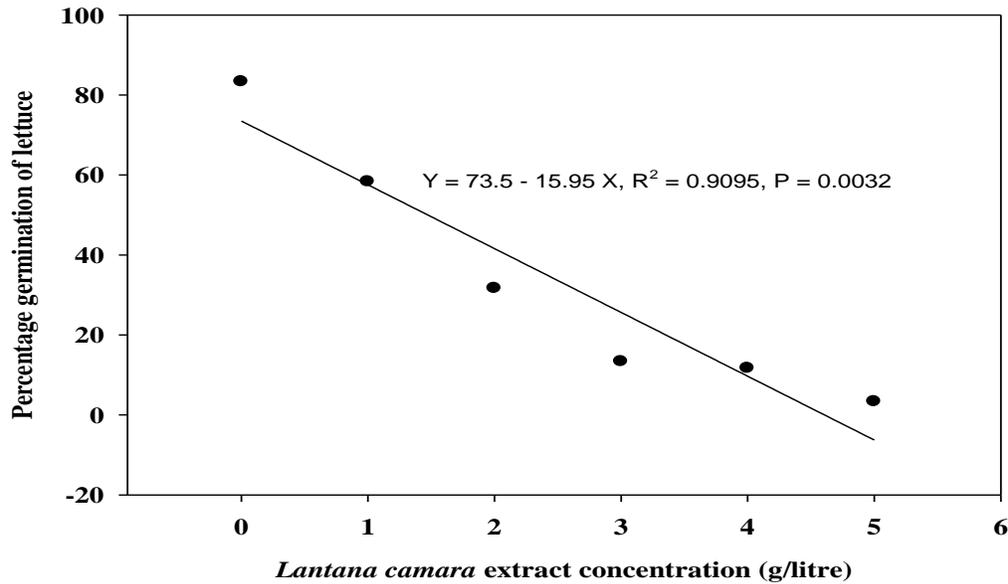


Fig. 1. The relationship of the percentage germination of lettuce with different concentrations of *L. camara* leaf extracts.

Germination of lettuce in treatments 1 g, 2 g, 3 g and 4 g per litre started on the sixth day after planting. There were significant treatment differences ($p < 0.001$) from 6 to 13 days after planting (Fig. 2). For the concentration of 5 g per litre germination was recorded at 13 days from planting. There was a steep increase in the rate of

germination in the concentration of 1g/litre at 10 days to 13 days from planting. Higher germination was achieved in the control treatment. Plant germination continued until the termination of the experiment at 13 days from planting. Generally, the germination of lettuce decreased as the concentration of *Lantana* increased.

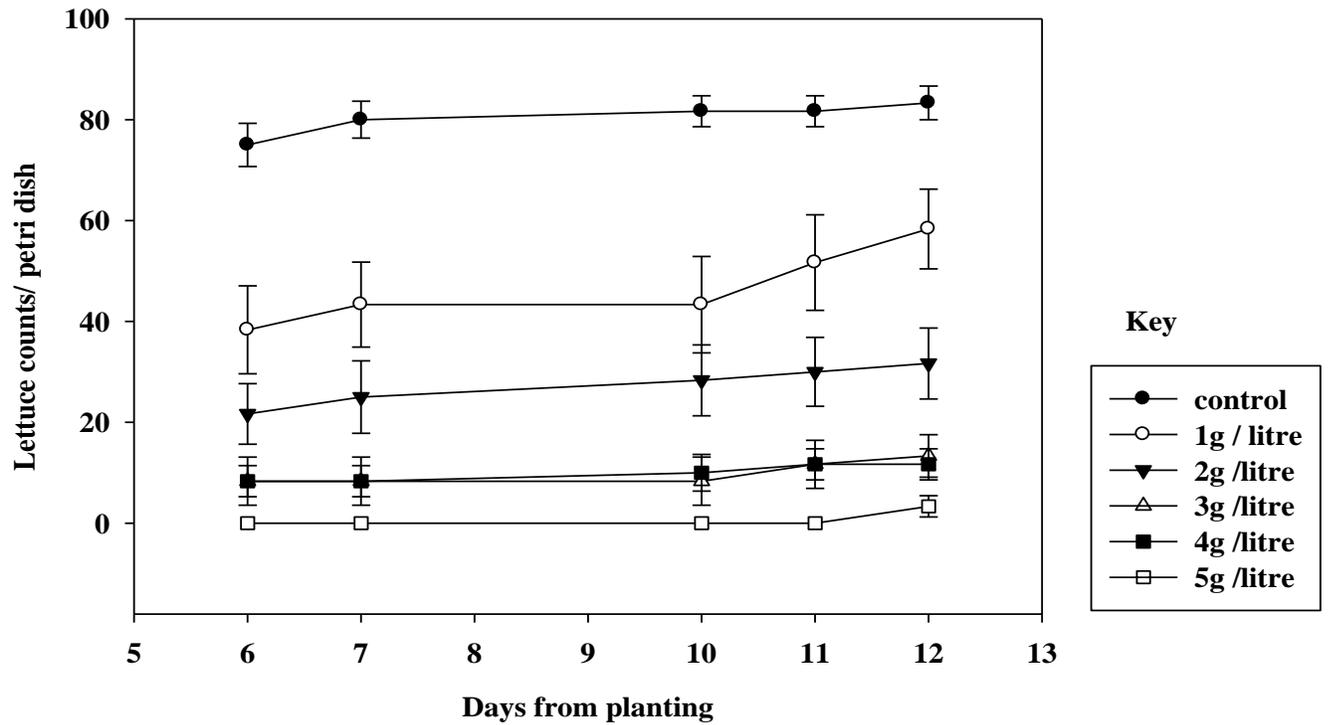


Fig. 2. Lettuce germination in different *L. camara* concentrations.
Vertical bars show standard errors.

The lettuce radicle length was significantly ($p < 0.05$) linearly related to *Lantana* leaf extracts concentration with a high r^2 value. The effect of *Lantana* leaf extracts on lettuce radicle length is shown on Fig. 3.

When the concentration of the *Lantana* leaf extracts was increased from 0 to 5 g / litre the radicle length was reduced, being almost completely inhibited by 5 g/litre.

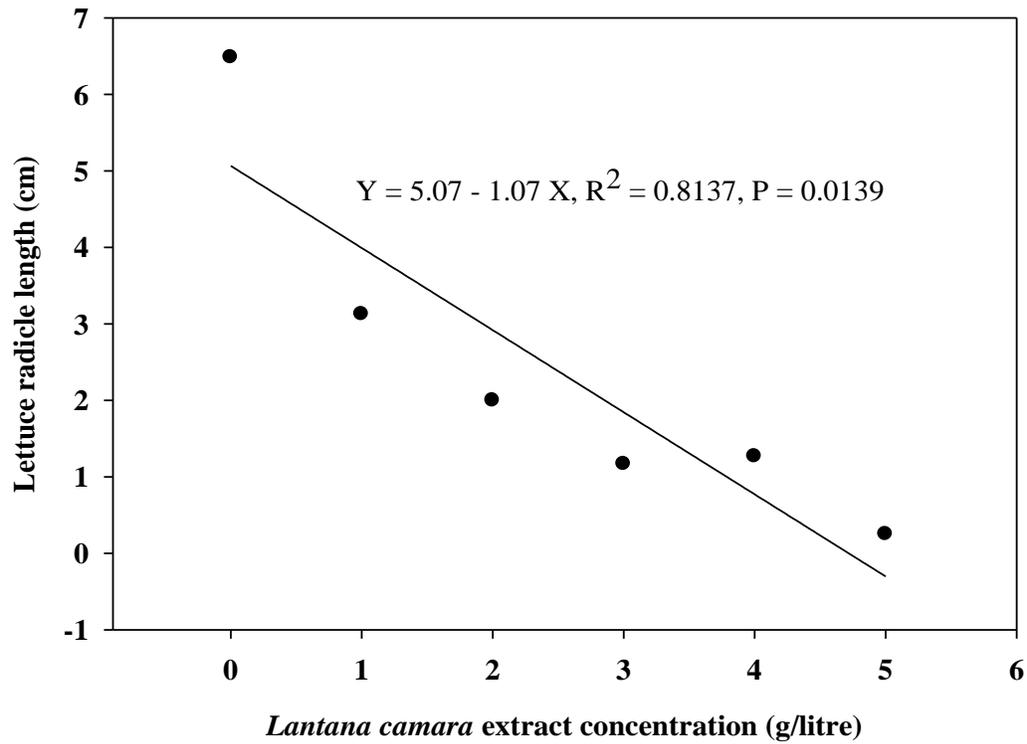


Fig. 3 The relationship of lettuce radicle length with the different concentrations of *L. camara* leaf extracts

The linear relationship of *Lantana* leaf extracts and lettuce plumule length was not significant ($p > 0.05$) and the r^2 value was low ($r^2 = 0.4038$). Low concentrations of *L. camara* leaf extracts did not have an

inhibitory effect on the plumule length of lettuce. The effect of *Lantana* leaf extracts concentration were clearly noted in 5 g/ litre treatment (Fig. 4).

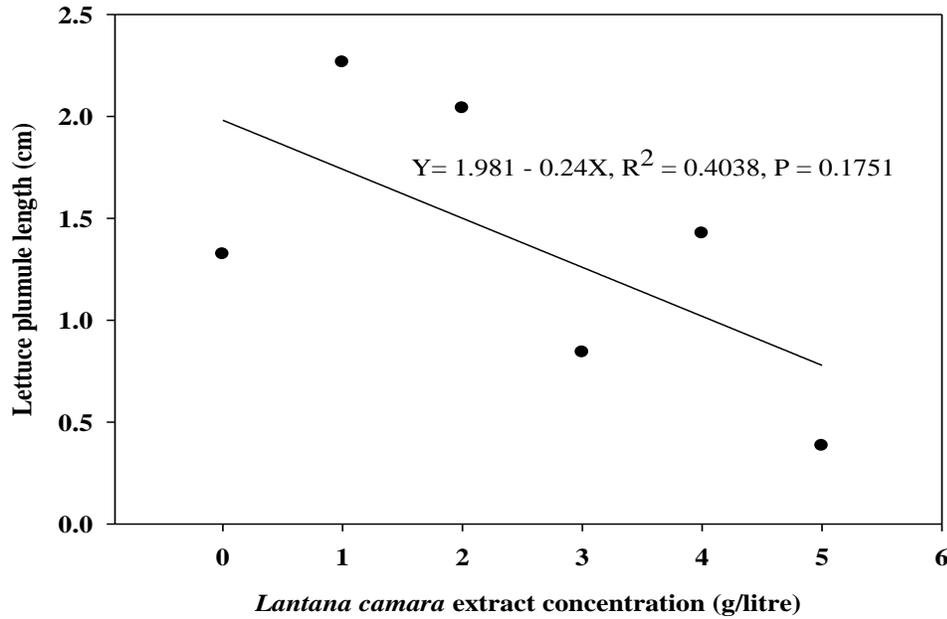


Fig. 4 The relationship of lettuce plumule length with different concentrations of *L. camara* leaf extracts.

3.2 Tomato germination, radicle and plumule length

The relationship of *L. camara* extract and tomato germination was significantly linear ($P < 0, 05$) and the r^2 was high. The effect of lantana leaf extract concentration is shown

in Fig. 5. Increasing the concentration of lantana leaf extract from 0 to 5 g per litre tended to decrease the germination of tomato seeds, with the highest concentration decreasing it to about 30 % from about 70 %.

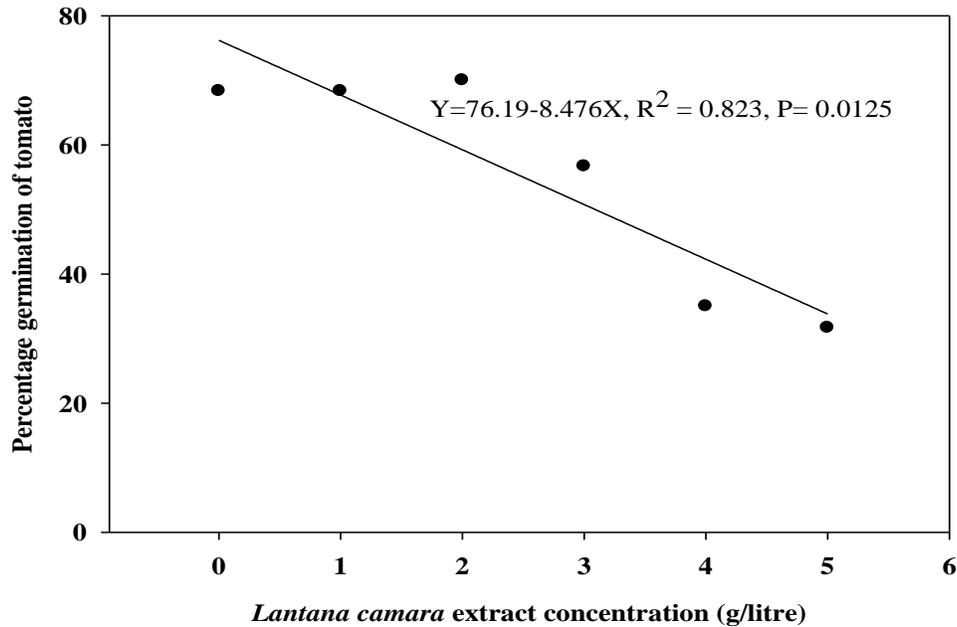


Fig. 5 The relationship of the percentage germination of tomatoes with the different concentrations of *L. camara* leaf extracts.

Fig.6 shows that the different *L. camara* concentrations had a significant ($p < 0.05$) effect on the germination of tomatoes after 13 days from planting. Tomato germination did not occur in all the concentrations. At six days from planting treatments with 3, 4 and 5 g/ litre of *L. camara* leaf extract recorded no germination. Germination was recorded

from seven days onwards and in all other treatments as shown in Fig. 6. Higher germination was achieved in the control treatment. Plant germination continued until the termination of the experiment at 13 days from planting. Generally, the germination of tomatoes decreased as the concentration of lantana increased.

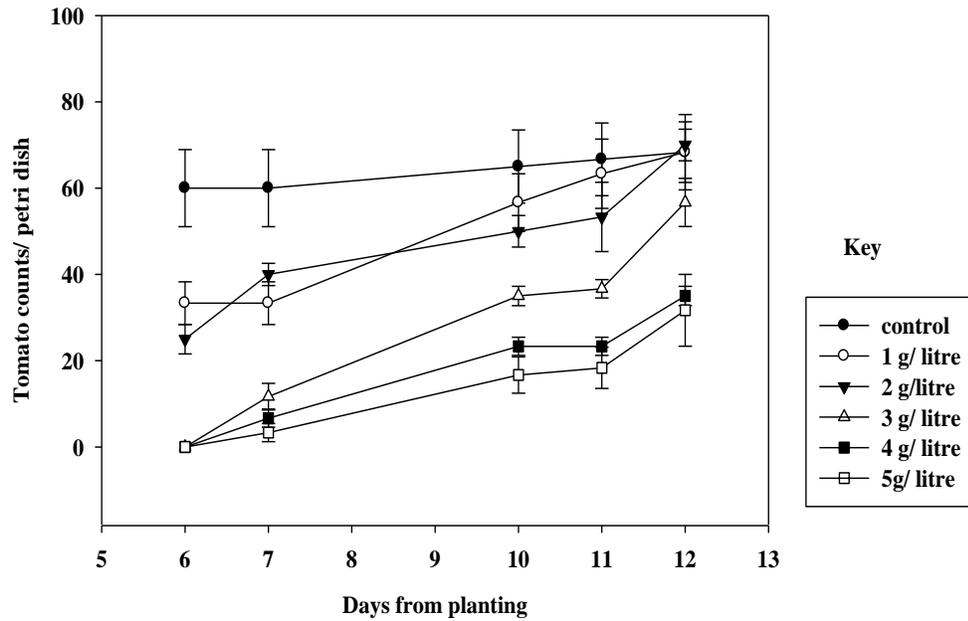


Fig. 6 Tomato germination in different *L. camara* concentrations. Vertical bars show standard errors

There was a significant ($p < 0.05$) linear relationship between lantana leaf extracts and tomato radicle length. The effects of lantana leaf extracts on tomato radicle

length are shown on in Fig. 7. Increasing the concentration of lantana decreased tomato radicle length.

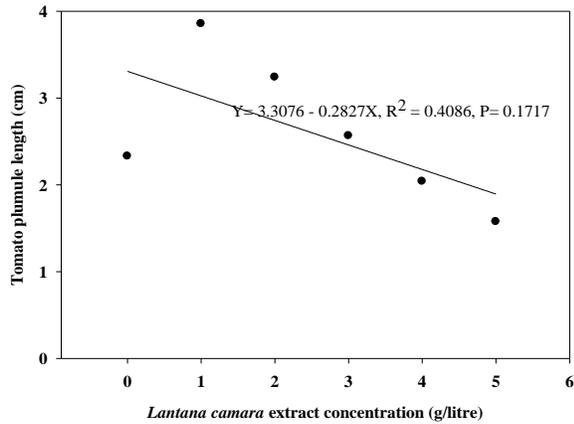


Fig. 8. The relationship of tomatoes plumule length with the different concentrations of *Lantana camara* extracts.

There was no significant relationship between lantana leaf extract concentration and tomato plumule length. The control treatment recorded a lower plumule length

3.3 Greenhouse Experiment

3.3.1 Lettuce and tomato emergence and dry matter

The effect of lantana biomass on percentage emergence of lettuce is shown on Fig. 9. There was no significant linear relationship between lantana biomass

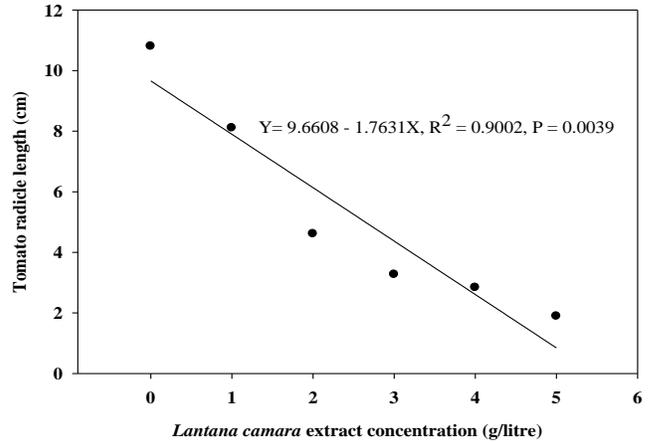


Fig. 7 The relationship of tomatoes radicle length with different concentrations of *L. camara* leaf extracts

as compared to the treatments with 1 to 3 % of *L. camara* leaf extract. The effects of lantana leaf extracts on tomato plumule length are shown in Fig. 8.

addition and the percentage emergence of lettuce ($P > 0.05$). This shows that an increase in lantana leaf biomass did not have an inhibitory effect on the emergence of lettuce. There was no significant linear relationship between lantana biomass additions and the emergence of tomato ($P > 0.05$). Lantana biomass did not affect percentage emergence of tomato plants.

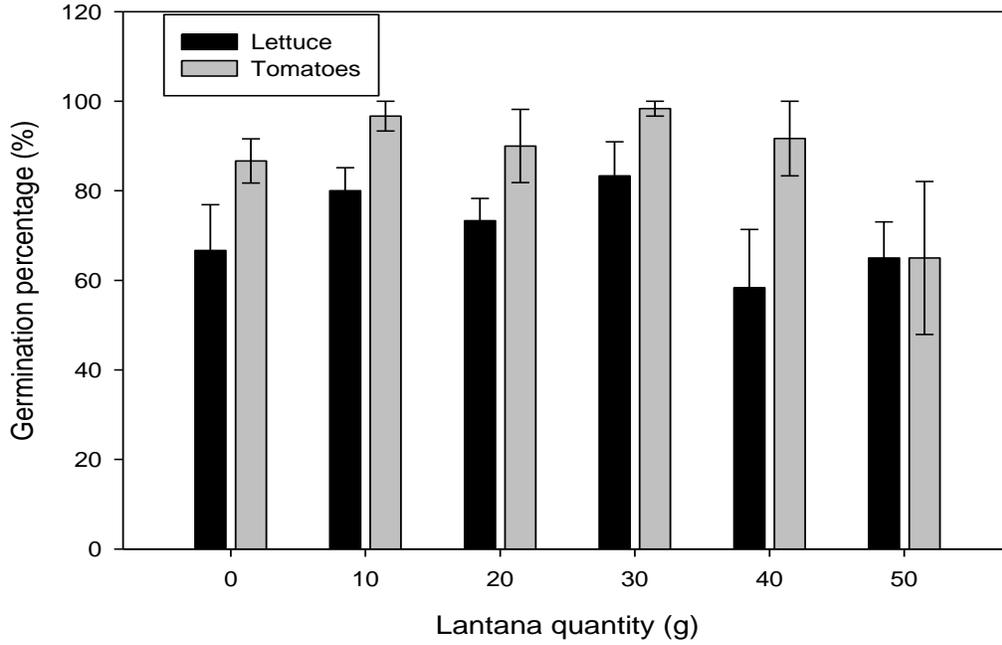


Figure 9 Effect of lantana camara quantity on the germination percentage of both lettuce and tomatoes

Increasing the *Lantana camara* leaf biomass from 0 to 30 grams per pot resulted in an increase in the lettuce biomass.

However, further increase in the concentration resulted in no such positive effect (Fig. 10).

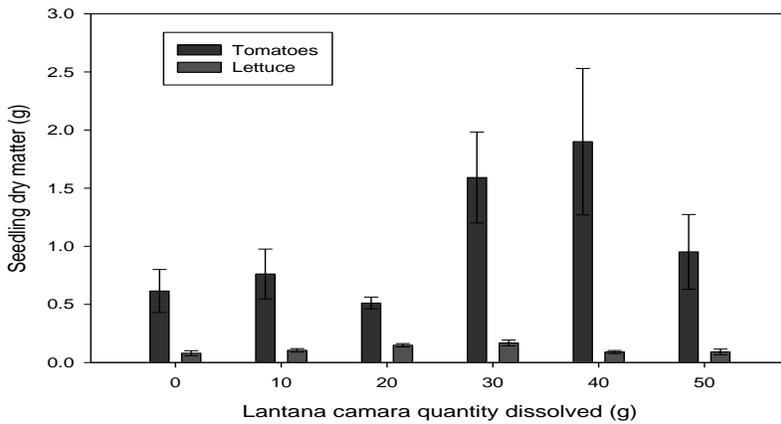


Figure 10 Effects of *Lantana camara* leaf biomass concentrations on the biomass of both lettuce and tomatoes.

There was no significant linear relationship ($P > 0.05$) between lantana biomass and the dry matter of tomato plants (Figure 12). It

was noted that the dry matter of tomato plants tended to increase with the addition of 30 and 40 g / pot of lantana biomass.

4. Discussion

In the laboratory, *Lantana camara* extracts concentrations were allelopathic on the germination and radicle length of both tomatoes and lettuce compared to the control. These results support the notion that *Lantana camara* is inhibitory to the growth of other plants. These results agree with Prasad and Shrivastva (1991) who observed that *Lantana camara* extracts reduced the germination and growth of rice seedlings. Also, Batlang and Shushu (2007) found inhibitory effects of *Lantana* extract on sunflower seedlings. Initial growth processes like germination are controlled by cell division processes such as mitosis. In this respect studies done by Rice (1984) and Hussein et al., (1984) *Lantana camara* inhibited both cell division and elongation and this further reinforces our results. Wollen et al., (1994) extracted flavonoid aglycon and triterpenoids from the leaf extracts of *L. camara* and the same compounds have been reported to inhibit cell division and elongation hence reduced germination and radicle and plumule growth. In support of the same fact, Casado (1995) stated that seeds exposed to *Lantana camara* leaf leachates in the laboratory in a confined environment such as a petri dish were more likely to be affected by allelochemicals in the growth media.

In the greenhouse, germination was not inhibited by the four concentrations of *L. camara* leaf extracts. The suppressive magnitude of the lantana leaves on germination was not apparent in this experiment. The expression of allelopathy could have been limited in the greenhouse due to biotic and abiotic factors which could have had an influence on the phytotoxicity

levels of allelochemicals which include temperature, light soil conditions, microflora which influence the release of allelochemicals from the leaves (Bhadoria, 2011). The results supports the notion that that soil chemicals, biological and physicochemical properties of the soil may influence the activity and toxicity of allelochemicals. The results coincided with Sahid and Sugau (1993) and Achhireddy et al., (1984) who reported that *Lantana camara* did not influence crop growth in the soil although literature is awash with reports that *Lantana* is allelopathic to crops.

On the contrary, biomass of lettuce increased with addition of 0, 10, 20 and 30g of *Lantana* biomass and decreased at 40 and 50g/pot. The results align to Sahuid and Sugau (1993)'s assertion that in some cases the toxin must accumulate to the physiologically active level for inhibition to take place through allelopathy. The results also show that it was the lower concentration of *Lantana spp.* extracts that showed stimulatory activity and a further increase causes biomass accumulation to decrease. Hossain and Alam (2010) mentioned that the inhibitory effects of *L. camara* were proportional to the concentration of the extracts on most field crops and forest plants with an estimation of about 1,2 tonnes per hectare. Lower concentrations of lantana leaf biomass showed some stimulatory effect as there was increase in crop emergence and biomass.

This study has demonstrated that *L. camara* has no significant allelopathic effects on crops such as tomato and lettuce under glasshouse conditions in a clay soil. On the other hand *L. camara* has been reported to have allelopathic effects on weeds such as

Bidens pilosa and *Ipomoea tricolor* (Kwembeya *et al.*, 2013; Casado, 1995) and it could be useful to include these weeds as a basis for comparison on the effects of *L. camara* in future experiments. Therefore, there is an opportunity to use lantana leaf residues for weed control in horticultural crops. Furthermore, Chikuvire *et al* (2013) reported that the addition of lantana extracts to the soil increased soil nitrogen leading to improved yields in horticultural crops. The study can help with the use and management of the locally available resources of *L. camara* with the prunings being used as live fences.

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5. Conclusions

In the laboratory, both tomatoes and lettuce had germination percentage and radicle length inhibited by *Lantana camara* leachates. Also germinations counts showed increased inhibition at the highest lantana concentration. In the greenhouse pot experiment the different concentration had no effect on germination. Rates of up to 30g and 40g/pot increased lettuce and tomato dry matter respectively showing that the rates could be applied to the soil and dry matter accumulation increase compared to the control. This raises the possibility of using *Lantana camara* as a dry matter accumulation stimulant in lettuce and tomatoes to control the invasive weed.

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