

Full Length Research Paper

Herbicidal effects of *Datura stramonium* (L.) leaf extracts on *Amaranthus hybridus* (L.) and *Tagetes minuta* (L.)

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Evolution of weeds resistant to herbicides demands new solutions to cope with the problem since economic losses generated by weeds can be higher than those caused by insect pests. Bioactive compounds known as allelochemicals have the potential to act as natural herbicides in weed management in agro-ecosystems. Laboratory, pot and greenhouse experiments were carried out to investigate the herbicidal effects of *Datura stramonium* aqueous leaf extracts on the germination and early growth of *Tagetes minuta* and *Amaranthus hybridus*. The laboratory and greenhouse experiments were arranged as completely randomised design, and the open field pot experiment was arranged as a randomised complete block design. Four concentrations of 2, 4, 6 and 8%, respectively of *D. stramonium* aqueous leaf extracts were used. Distilled water was the control. Data for germination, radicle and plumule length was collected within the first 10 days for the laboratory experiment. Root length, shoot length and biomass yield was collected 30 days after germination for both the greenhouse and field experiments. Results indicated that germination percentage, radicle length, plumule length and dry matter significantly decreased ($P < 0.001$) as concentration of *D. stramonium* leaf extracts increased in all the experiments. This implies that *D. stramonium* has pre-emergence and early post emergence herbicidal effects on the two weeds. This study revealed that allelopathic sprays of *D. stramonium* can be used by resource poor small scale farmers or organic farmers for the control of *Amaranthus hybridus* and *T. minuta* in Zimbabwe.

Key words: Allelopathy, aqueous leaf extract, *D. stramonium*, *Amaranthus hybridus*, *Tagetes erecta*, herbicidal effects, germination, early growth.

INTRODUCTION

Allelopathy is defined as any process involving secondary metabolites produced by plants, algae, bacteria and fungi

that influence the growth and development of agricultural or biological systems (Macías et al., 2007; Thi et al., 2015). Allelopathy involves synthesis of bioactive compounds known as allelochemicals which are capable of acting as natural pesticides. Plants produce these compounds as a mechanism to defend themselves in the course of co-evolution. The fact that allelopathy is a crucial defense and attack weapon of the plant to gain a foothold on the community can not be ignored (Marcias et al., 2007).

According to Asaduzzaman et al. (2014), Casimiro et al. (2017) and Farooq et al. (2011), the wise exploitation of allelopathy in the cropping systems may be an effective, economical and natural method of weed management. These compounds are usually degraded easily in the environment due to their short half life as they contain fewer halogen constituents in their structures. Due to their impure nature, they usually contain a number of active compounds which can act on more than one site like a mixture of herbicides and may control a wider spectrum of weeds (Solts et al., 2013). This discourages the development of resistance. Abandoning of chemical control with current agriculture is rather impossible, it is therefore necessary to create new classes of herbicides with new mechanism of action and target site not previously exploited. Natural compounds pose as a potential source for the discovery of eco-friendly herbicides, so called bio herbicides (Solts et al., 2013).

The herb, *D. stramonium* is an annual upland weed that is widely distributed throughout the world. In Mexico, the plant inhabits open, cultivated and disturbed sites where they attain an average height of 1 m (Valverde et al., 2002). According to Fatoba et al. (2001), the plant is characterised by solitary white trumpet shaped flowers. Weed surveys done in Zimbabwe by Thomas (1971) and Chivinge (1983, 1988) classified the weed as aggressive and difficult to control. The plant has been increasing in the cropping systems and farmers cut it and use the leaves as mulch.

Currently, the weed has turned invasive, thereby making available its leaves for mulch placement in gardens and agronomic fields. Other farmers have reported that it reduces weed germination. It has been said that several chemicals have been identified and phytochemical investigators believe that there are still many other chemicals in *D. stramonium* which have not been identified to be exploited as bioherbicides (Elisante et al., 2014). Allelochemicals found in *D. stramonium* have allelopathic effects on survival of native plants. *D. stramonium* contains a series of allelochemical in form of

alkaloids, atropine, hiosciamine and scopolamine (Butnariu, 2012), which inhibits the growth and development of root and shoots of *Trigonella* and *Lepidium* in a concentration dependent manner (EL-Shora and Abd EL-Gawad, 2014; 2015a; An et al., 1996). Currently, there is no basic information of the allelopathic effects of *D. stramonium* on *A. hybridus* and *T. minuta* which are seriously problematic arable weeds in Zimbabwe. The objective of this current study was to determine the multi-herbicidal effects or mode of actions of *D. stramonium* leaf extracts on the germination and early establishment of the *A. hybridus* and *T. minuta*.

MATERIALS AND METHODS

Experiment 1: Effect of *D. stramonium* concentration on the germination and early establishment of two weeds in the laboratory experiment.

Study site

The laboratory experiment was carried out at Midlands State University, located in Midlands province of Zimbabwe. The geographical location is 19°45' S (line of latitude) and 29°85' E (line of longitude). It experiences mean annual temperature of 18°C. The site is in agro-ecological region III, at an altitude of 1428 m (Vincent and Thomas, 1960; Mugandani et al., 2012).

Experimental design

The experiment was arranged as a complete randomised design with five treatments replicated three times. Treatments were 20 ml of distilled water (control) and aqueous *D. stramonium* leaf extracts applied at 2, 4, 6 and 8% concentration as a ratio of plant extract powder to 100 ml distilled water. 2 g of extract powder was added to 100 ml of distilled water to give 2% concentration of aqueous and the same was done for 4, 6 and 8% concentrations.

Preparation of *D. stramonium* aqueous leaf concentrations for the three experiments

Leaves of fully grown plants collected from the wild were washed to remove soil particles. The material was then cut into pieces and shed dried for one month. After drying, the material was crushed into powder form manually using a traditional mortar and pestle. Further grinding was done by using an electric mortar. The material (powder and distilled water) was mixed and poured into a conical flask with its mouth closed and kept for 24 h in the dark at room temperature according to the method used by Dhawan and Narwal (1994). The four flasks were marked with stickers according to the *D. stramonium* concentrations (2, 4, 6 and 8%, respectively). This was followed by filtration process in two steps. In the first step, muslin cloth was used, and later the filtrate was allowed to pass

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through Whatman filter paper no.1. The prepared aqueous concentrations were kept in a refrigerator for the duration of the experiment to prevent conversions of some of the compounds upon exposure to light and high temperature.

Experimental procedure

Two hundred and twenty five seeds of the selected weeds were surface sterilized with 0.1 % mercuric chloride solution for two minutes and washed twice with distilled water. The petri dishes were labelled with a permanent marker in relation to concentration level. Fifteen seeds of each weed were placed in petri dishes on Whatman filter paper no.1. Twenty millilitres of each *D. stramonium* aqueous concentration (2, 4, 6 and 8%, respectively) was added to each petri dish. The same amount of distilled water was used as a control. Watering was done after every three days, and the petri dishes were kept in an incubator at 24°C room temperature for 10 days.

Experiment 2: Pot experiment: Effects of different *D. stramonium* aqueous concentrations on germination and early seedling growth of weeds in the field.

Study site

The field experiment was carried out at Midlands State University, located in Midlands province of Zimbabwe. The geographical location is 19°45' S (line of latitude) and 29°85' E (line of longitude). It experiences mean annual temperature of 18°C. The site is in agro-ecological region III, at an altitude of 1428 m.

Experimental design

The experiment was arranged as a complete randomised design with five treatments replicated three times and two weeds were tested.

Experimental procedure for field and green house experiments

Two hundred and twenty five seeds of the selected crops were surface sterilized with 0.1% mercuric chloride solution for two minutes, and washed twice with distilled water. Five litre pots were used and they were filled with mixtures of soil (loamy sand). Fifteen seeds of each of the tested weeds were sown in each pot at 0.5 cm, and then irrigated with various solutions to field capacity every three days.

Data collection for field and green house experiments

Data on seed emergence, shoot, and root length; seedling fresh and dry weight was recorded. Seed emergence was determined by physically counting the number of seedlings on the 8th day after planting. During the experiment period (after 30 days after planting), shoot and root length was also measured using a 30 cm ruler. The dry weight was determined by placing the tested samples in the oven to a temperature of 110°C for 48 h until a constant weight was realised.

Experiment 3: Effects of different *D. stramonium* aqueous concentrations on germination and early seedling growth of weeds in the greenhouse.

Study site

The greenhouse experiment was carried out during the 15/16 summer season at Morningside suburb in Masvingo Province of Zimbabwe at a geographical location of latitude 20° 7' 17S and longitude 30° 49' 58 E. The site is in agro-ecological zone 4, at an altitude of 1034 m above the sea level. It receives an average of 600 mm of rain annually with a mean annual temperature of 28°C.

Experimental design

The experiment was arranged as a complete randomised design with five treatments replicated three times.

Data analysis

Collected data was subjected to Analysis of Variance at 5% significance level using Genstat 4.0 version 2013. Fishers protected least significance test at 5% was used to separate the means where significant differences were noted.

RESULTS

Germination and emergence

The results showed that the germination percentage as affected by *D. stramonium* aqueous leaf extracts was significantly ($P < 0.001$) lower than the control at all levels in the laboratory percentage compared to the rest of the treatment (Figure 1) in the laboratory. As concentrations increased, germination percentage decreased. The highest germination (100%) was recorded where distilled water was applied in all tested species whilst 8% concentration significantly ($p < 0.001$) decreased germination. The same trend was observed in the field (Figure 2) and in the greenhouse (Figure 3) where the emergence percentage decreased with increase in the concentration.

Radicle and root length

Results indicated that as the concentration decreased from 8 to 0%, the radicle and root length increased with a decrease in the concentration of *D. stramonium*. Results showed highly significant effects ($p < 0.001$) of *D. stramonium* on *A. hybridus* and *T. minuta* as shown on Table 1 across all the environments.

Plumule and shoot length

Results indicated that aqueous concentrations of thorn

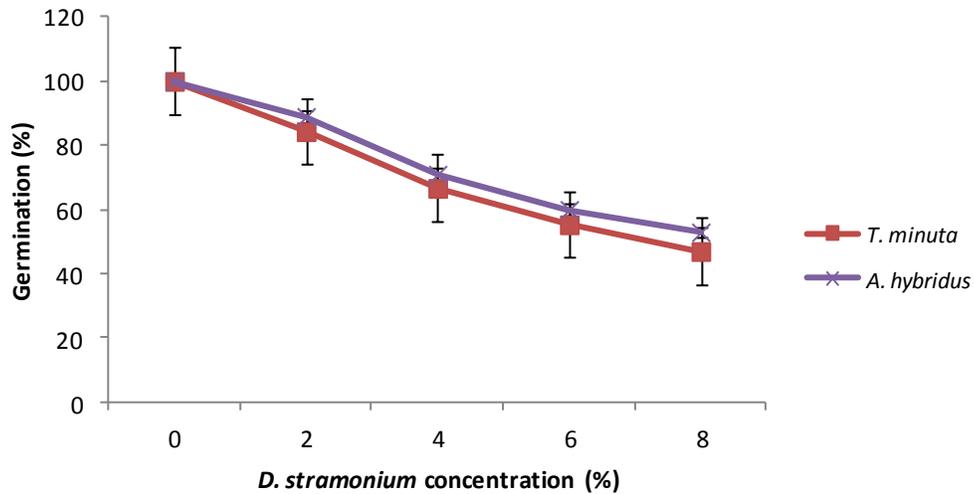


Figure 1. Effect of *Datura stramonium* concentration on the germination of the two weeds in the laboratory.

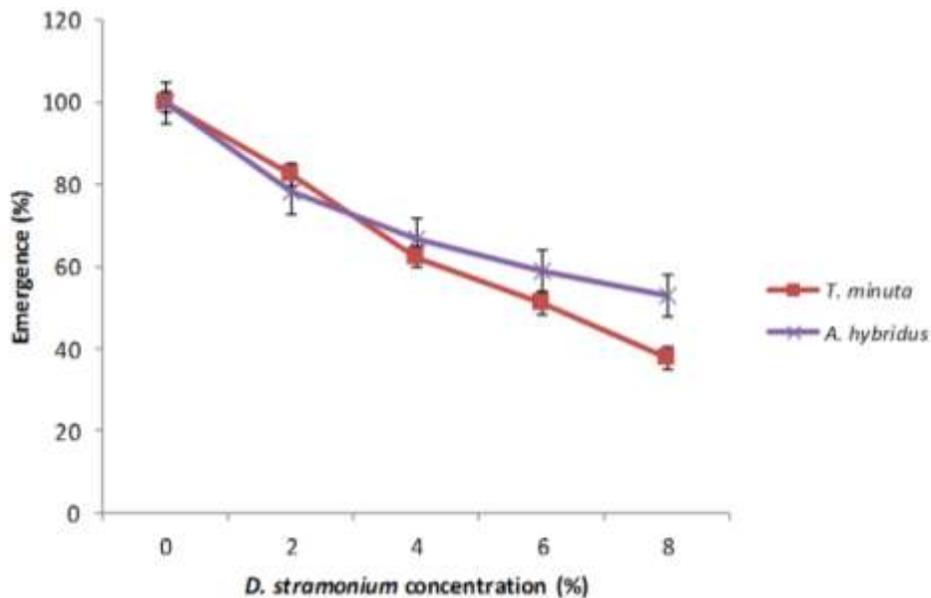


Figure 2. Effect of *D. stramonium* concentrations on the emergence percentage of the two weeds in the pot experiment in the field.

apple on plumule and shoot length was highly significant ($P < 0.001$). Distilled water recorded the highest plumule length and shoot length when all treatments were compared on all tested species. It was observed that the rate of percentage decrease in plumule and shoot length was concentration dependent across all the tested species. Shoot length decreased as the concentration of *D. stramonium* increased from 0 to 8% as presented on

Table 2.

Dry matter traits

Results indicated that the effects of aqueous concentrations of thorn apple on seedling dry weight was significant ($P < 0.001$). There was a general percentage

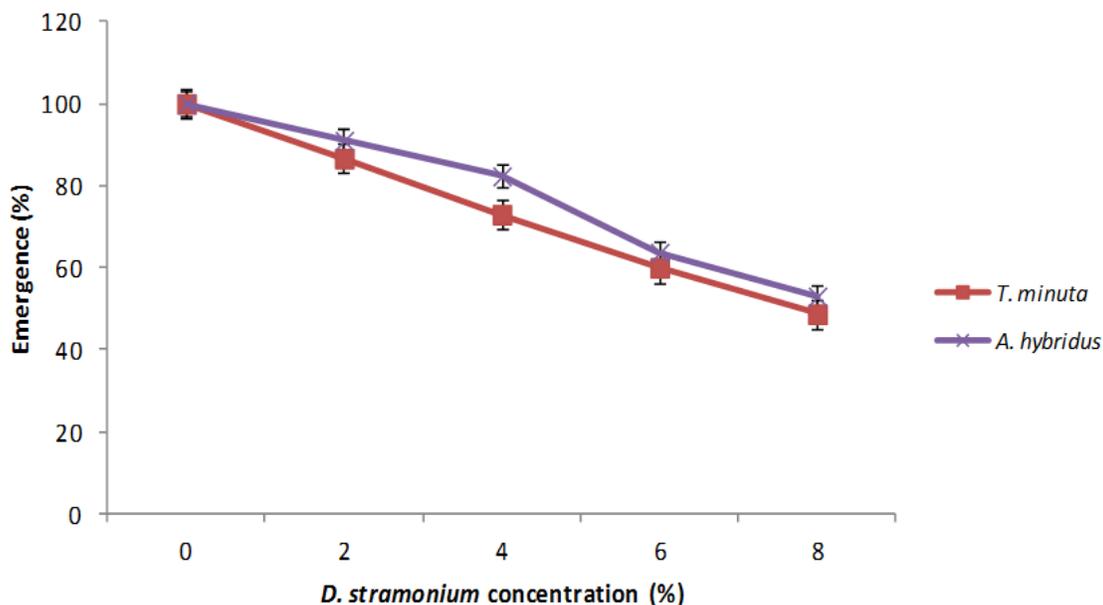


Figure 3. Effect of *D. stramonium* concentrations on the emergence of the two weeds in the greenhouse.

Table 1. Effect of the *D. stramonium* concentration on radicle and root length of *T. minuta*. and *A. hybridus* in laboratory, field and greenhouse conditions.

Concentrations	Laboratory		Field		Greenhouse	
	<i>T. minuta</i>	<i>A. hybridus</i>	<i>T. minuta</i>	<i>A. hybridus</i>	<i>A. hybridus</i>	<i>T. minuta</i>
0	26.97±0.14 ^a	28.67±0.491 ^a	44.17±2.09 ^a	80.4±1.21 ^a	80.13±1.07 ^a	44.17±2.09 ^a
2	25.1±0.14 ^b	24.97±0.491 ^b	38.37±2.09 ^b	75.2±1.21 ^b	74.50±1.07 ^b	38.37±2.09 ^b
4	22.1±0.14 ^c	22.83±0.491 ^c	34.3±2.09 ^c	64.9±1.21 ^c	64.43±1.07 ^c	34.30±2.09 ^{bc}
6	20.93±0.14 ^d	21.35±0.491 ^d	31.7±2.09 ^d	54.47±1.21 ^d	54.27±1.07 ^d	31.70±2.09 ^c
8	19.37±0.14 ^e	19.67±0.491 ^e	26.8±2.09 ^e	44.6±1.21 ^e	44.27±1.07 ^e	26.80±2.09 ^d
P-value	<0.001	<0.001	<0.001	<0.001	0.001	<0.001
CV (%)	0.7	2.3	6.6	2.1	1.8	6.6

*Means followed by the same letter in the same column are not significantly different.

Table 2. Effect of the *D. stramonium* concentration on plumule and shoot length of *T. minuta* and *A. hybridus* in laboratory, field and greenhouse conditions.

Concentration (%)	Laboratory		Field		Green house	
	<i>T. minuta</i>	<i>A. hybridus</i>	<i>T. minuta</i>	<i>A. hybridus</i>	<i>T. minuta</i>	<i>A. hybridus</i>
0	16.5±0.09 ^a	20.07±0.705 ^a	65.47 ±2.85 ^a	80.13±1.07 ^a	65.03±0.27 ^a	80.40±1.22 ^a
2	14.83±0.09 ^b	18.40±0.705 ^b	63.17±2.85 ^b	74.50±1.07 ^b	62.37±0.27 ^b	75.20±1.22 ^b
4	12.90±0.09 ^c	16.30±0.705 ^c	60.13±2.85 ^{ab}	64.43±1.07 ^c	55.43±0.27 ^c	64.90±1.22 ^c
6	11.53±0.09 ^d	15.40±0.705 ^{cd}	55.50±2.85 ^{bc}	54.27±1.07 ^d	50.93±0.27 ^d	54.47±1.22 ^d
8	10.17±0.09 ^e	14.60±0.705 ^d	50.27±2.85 ^c	44.27±1.07 ^e	49.90±0.27 ^e	44.60±1.22 ^e
P value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
CV (%)	0.8	4.6	5.3	1.8	0.5	2.1

*Means followed by the same letter in the same column are not significantly different.

Table 3. Effects of the various *Datura stramonium* concentrations on the dry matter properties of the *Targetis minuta* and *Amaranthus hybridus* in the greenhouse and the field experiments.

Concentration (%)	Field			Greenhouse	
	<i>T. minuta</i>	<i>A. hybridus</i>		<i>T. minuta</i>	<i>A. hybridus</i>
0	0.786±0.06 ^a	2.013±0.069 ^a		0.803±0.051 ^a	2.037±0.076 ^a
2	0.736±0.06 ^a	1.86±0.069 ^b		0.723±0.051 ^a	1.887±0.076 ^a
4	0.413±0.06 ^b	1.673±0.069 ^c		0.406±0.051 ^b	1.697±0.076 ^b
6	0.266±0.06 ^c	1.49±0.069 ^d		0.273±0.051 ^c	1.513±0.076 ^c
8	0.116±0.06 ^d	1.26±0.069 ^e		0.123±0.051 ^d	1.28±0.076 ^d
P value	<0.001	<0.001		<0.001	<0.001
CV (%)	14.3	4.6		12.4	5

*Means followed by the same letter in the same column are not significantly different.

decrease in seedling dry weight as aqueous concentration increased from 0 to 8% on all tested species. *Targetis minuta* recorded the highest decrease of 54.8% whilst wheat and *A. hybridus* recorded seedling dry weight decreases of 22 and 15.3% respectively as concentration increased from 6 to 8%. The concentration of 2 % was not significantly different from the treatment watered by distilled water except for *A. hybridus* in the field (Table 3).

DISCUSSION

The results showed a reduced germination percentage with increasing concentration of allelochemicals from *D.stramonium* across all the measured weeds. These results concur with the findings of many authors (Hassannejad and Ghafarbi, 2013; Yu et al., 2003; Elisante et al., 2013; Levitt et al., 1984; Oyun, 2006; Alam and Islam, 2002). *D. stramonium* allelochemicals contains chemicals that retard the metabolism of food reserves in the seed (Levit et al., 1984) and the secondary effects of these processes include reduced germination and early growth of radicles (Levitt and Lovetti, 1984).

Altikat et al. (2013), Ullah et al. (2015) and Alam and Islam (2002) concur with these findings and reported that allelochemicals disturb the activities of the peroxidase alpha amylase enzyme and acid phosphatases which aid the breaking down of starch for successful germination to occur. Another assertion by EL-Shora *et al.* (2015a) and Oyun (2006) posits that allelochemicals inhibit water absorption which is a precursor for physiological processes that should occur before germination is triggered. All this help to support the assertion that *D. stramonium* has pre-emergence herbicidal effects.

Both shoot and root lengths of the two weeds were reduced by leaf extracts and the level of decrease depended on the concentration of the allelochemicals. Hussain and Reigosa (2011) found similar results on *D. glomerata*, *L. perenne* and *R. acetosa*. Gholami et al.

(2011) concluded that *D. stramonium* alkaloids (hiosciamine and scopolamine) can reduce cell division or interferes with the auxin that induces growth of shoots and roots. Findings by EL-shora et al. (2015a) found that *D. stramonium* inhibit cell division. This can serve as a confirmation of the existence of the early post emergence effects of the allelochemicals. This further confirms the existence of more than one mode of action of herbicide which is critical in developing herbicides that are not prone to resistance development.

Total dry matter for all the weeds was reduced as concentration increased. Total dry matter is the function of the ability of the whole plant to obtain edaphic resources (minerals and water). Whilst all parameters were analysed individually, the cumulative contributions of the small differences has bigger effects on the metabolism of the whole plant (Robeiro, 2011). Any inhibition at each stage in the growth of the plant contributes towards reduced ability of the plant to capture resources for its survival. The various concentrations are therefore able to reduce dry mass of both weeds which indicated the presence of herbicidal effects.

Conclusion

We conclude that *D.stramonium* leaf extracts have both pre-emergence and early post emergence herbicidal effects towards the weeds studied. This study therefore recommends the use of *D. stramonium* leaf extracts at high concentrations as cheap bio herbicides to control *T. minuta* and *A. hybridus* in Zimbabwe. However, there is need for further research on the efficacy of other plant parts like the roots and fruits and solvent extraction method (ethanol and aqueous) of *D. stramonium*.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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