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Demonstrative effects of crude extracts of *Desmodium* spp. to fight against the invasive weed species *Tagetes minuta*

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ABSTRACT

Threats on native flora and fauna by invasive plant species represent one of the main conservation and management challenges in rangelands. Methods that are both effective and ecologically safe to suppress invasives are urgently needed but have rarely been used, thus, highlighting the need to devise and test ones. In our study, we used a completely randomized design to assess the allelopathic effects of *Desmodium uncinatum* and *Desmodium intortum* leaf (DuL, DiL respectively) and root (DuR, DiR respectively) extracts on germination and seedling vigor of the invasive weed *Tagetes minuta*. We also assessed seedling germination, height, fresh weight and chlorophyll content after fourteen days and thirty days of treatment separately. The mean percentage germination per treatment in and across each group differed significantly ($P < 0.05$), with DuL having the highest suppressive effect ($P = 0.003$). Likewise, the mean germination per treatment was more strongly negatively correlated with treatment concentration in DuL treatments in both laboratory and screen house experiments ($r = -0.48$, $P = 0.0003$ and $r = -0.91$, $P < 0.0001$ respectively) than in DuR, DiL and DiR. Seedling height, fresh weight and chlorophyll content (Chl) differed significantly ($P < 0.05$) between the four treatment groups. Seedlings treated with higher concentrations were observed to be shorter, having lower fresh weights and Chl content than those treated with lower concentrations. The DuL higher concentration showed a trend of shorter seedlings with lower fresh weights and Chl content than other groups. Our findings suggest that *D. uncinatum* may probably be used to control invasive species, *T. minuta* and should be integrated into the management practices in the affected areas. Also our data suggest that a potential exists in devising an innovation that is both ecologically safe and effective by using *Desmodium* spp. making it possible to improve rangelands production through planting them in the areas affected by invasive species.

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1. Introduction

Tagetes minuta or Mexican marigold (Asteraceae) is an unpalatable plant to both human and animals that originates from South America and has been reported as an invasive weed across the tropics, subtropics and several temperate countries [25,23,9]. In Kenya, *T. minuta* was first recorded as an exotic weed during the 1920s, originally restricted to higher altitudes but increasingly spreading to lower ones [26]. In Tanzania, the species has been reported to invade most of the Serengeti ecosystem and the Ngorongoro crater. This weed has been extensively studied for medicinal purposes [22], therefore, has been introduced to various areas to the extent that it became a weed in most rangelands and agricultural areas of Tanzania [29].

Roots of *T. minuta* produce exudates that are polyacetylene derivatives and are allelopathic [18], which delays germination, suppress local plants and reduces crop yield in *T. minuta* infested soils. Mechanical removal, traditional and chemical controls have been recommended to be applied whenever the weed appears in farmlands [22]. The provision of shade in agricultural fields has been reported to be effective in controlling *T. minuta* [22] although new seedlings will normally rapidly germinate as soon as shading is not present. Moreover, chemical control by using herbicides has also been reported to be effective in rangelands [26], but care should be taken because according to the World Health Organization, the negative effects of chemical pesticides to the environment can be devastating [16], and in some instances this might not be applicable in natural ecosystems including protected rangelands.

Understanding germination of a particular weed is crucial as it can play a great role in its timely control. Effects of light, temperature and various plant hormones like Gibberellins on the germination of *T. minuta* have been well studied by different scholars [4,15,27]. On the contrary, little research has been conducted on the effects that other plant species might have on *T. minuta* performance. While *Desmodium* species are known to have suppressive effects on the growth of other plants [11,20], no efforts have been done to study the

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effects of *Desmodium uncinatum* and *Desmodium intortum* root and leaf crude extracts on germination and growth of *T. minuta.* This information, however, is highly important as it might lead to an effective control of this weed especially in nature reserves where the use of herbicides is strictly prohibited. Further, *Desmodium* spp. are abundant and their extracts are easy to generate. Thus, using the extracts of this species might be a highly successful and cheap weed management tool. This study, therefore, seeks to determine the effects of root and leaf crude extracts of *D. uncinatum* and *D. intortum* on the seed germination, seedling length, leaf chlorophyll content and biomass of *T. minuta*. We expected that the crude extracts will reduce germination rate and seedling height of *T. minuta*, which are important measures of seedling vigor, and that it will also reduce seedling biomass and leaf chlorophyll content. We further expected that higher concentrations of the extracts will lead to stronger reduction of the four parameters and *D. uncinatum* and *D. intortum* extracts will have similar effects. We chose to investigate these two *Desmodium* species as they are most preferred by herbivores [6,7] in such a way that they can be inter-planted in invaded areas and suppress the invasive *T. minuta* while providing feed for the animals.

2. Research methods

2.1. Laboratory study design

The effects of *D. uncinatum* and *D. intortum* leaf and root crude extracts on the seed germination, seedling height, leaf chlorophyll content and biomass of *T. minuta* were studied using a completely randomized design. Fifteen seeds of *T. minuta* were placed in each of eight petri dishes (70.84 cm^2 surface area) lined with cotton wool, and subjected to eight different concentration treatments. The experiment was replicated three times; therefore, a total of 96 petri dishes were involved. Distilled water was added regularly when required to moisten the seeds. Seeds were observed every day and the number of germinated seeds were recorded and counted for fourteen days. After fourteen days, seedlings were harvested and fresh weight, seedling height, chlorophyll content and dry weight were determined for each germinated seedling.

2.2. Screen house study design

The effects of leaf and root crude extracts of *D. uncinatum* and *D. intortum* on the seed germination, seedling height, leaf chlorophyll content and fresh weight of *T. minuta* were studied using a completely randomized design in a screen house under field conditions. Ten seeds of *T. minuta* were placed in each of six pots (763.82 cm^2 surface area) with soil, under six different concentration treatments. The experiment was replicated three times; therefore, a total of 72 pots were involved. Normal tap water was added regularly when required to moisten the soil. Seeds were observed every day and the number of germinated seeds were recorded and counted for thirty days. After thirty days, seedlings were harvested and fresh weight, seedling height and chlorophyll content were determined for each germinated seedling.

2.3. Root and leaf crude extract preparation

Fresh roots and leaves from young *D. uncinatum* and *D. intortum* were collected from Livestock Training Institute (LITI) Tengeru demonstration plots in early January 2015. Extracts were prepared according to [19] with some modifications as follows: roots and leaves were air dried under room temperature for fourteen days and were later ground into powder. For each species, 100 g of root and leaf powder was soaked separately in 1 l of distilled water and left for 72 h, after which the crude extracts were filtered using Watsman filter paper no. 1 to obtain a final volume of 100 ml each. Both crude extracts were diluted with distilled water to obtain different proportions of 0 g/l (0%), 0.0125 g/l (12.5%), 0.025 g/l (25%), 0.0375 g/l (37.5%), 0.05 g/l (50%), 0.0625 g/l (62.5%) and 0.075 g/l (75%) of the original concentration (0.1 g/l (100%)).

2.4. T. minuta seed preparation and treatment

Seeds of *T. minuta* were collected from Gomba Estate farms in Arusha in late August 2014. Prior to the experiment, the seeds were air dried and stored in plastic bags. *T. minuta* seed viability tests were performed, in which 100% of seeds planted in petri dishes lined with cotton wool in early January 2015 were germinated. Seeds were washed using tap water and sterilized with 5% NaOCl for two minutes then rinsed with distilled water before planting. Each petri dish/pot was irrigated once with 10 ml/100 ml respectively, of the different solution treatments, i.e., $T_1 = 0.0125$ g/l (12.5%), $T_2 = 0.025$ g/l (25%), $T_3 = 0.0375$ g/l (37.5%), $T_4 = 0.05$ g/l (50%), $T_5 = 0.0625$ g/l (62.5%), $T_6 = 0.075$ g/l (75%) and $T_7 = 0.1$ g/l (100%). The seeds that were treated with distilled water (T_0) were taken as a control.

2.5. Leaf chlorophyll determination

Chlorophyll of *T. minuta* seedling was extracted according to [8] with some modifications: 50 mg of fresh leaves of *T. minuta* was immersed in 3 ml of Dimethyl Sulfoxide (DMSO) and incubated at 65 °C for 12 h. The extract was transferred to glass cuvettes for absorbance determination. The absorbance of blank liquid (DMSO) and samples were recorded at 645 and 663 nm [8] and the total leaf chlorophyll (Total Chl) calculated according to [1] using the following equation:

Total Chl $= 0.0202\ A_{663} + 0.00802\ A_{645}$.

Where: A_{663} and A_{645} are absorbance readings at 663 and 645 nm respectively.

2.6. Statistical analysis

For the laboratory experiment, one-way ANOVA was carried out to reveal the differences in seedling mean germination rate while Kruskal–Wallis Rank Sum test was performed on seedling height, fresh weight and chlorophyll contents of *T. minuta* under *D. uncinatum* and *D. intortum* root and leaf crude extracts. Spearman's rank-order correlation analysis was performed to reveal the relationship between (a) seedling biomass, (b) seedling height and (c) total seedling chlorophyll contents while Pearson's product–moment correlation analysis was performed on mean percentage germination rate to concentration treatments applied. For the screen house experiment, one-way ANOVA was carried out to reveal the differences in seedling mean germination rate, seedling height, seedling fresh weight, root length and chlorophyll contents of *T. minuta* under *D. uncinatum* and *D. intortum* root and leaf crude extracts. Spearman's rank-order correlation analysis was performed to reveal the relationship between mean germination rate to concentration treatments applied while Pearson's product–moment correlation analysis was performed on (a) seedling height, (b) seedling fresh weight and (c) total seedling chlorophyll contents. The statistical software used was STATISTICA version 8 and the level of significance was set at $P < 0.05$.

3. Results

3.1. Allelopathic effects on seed germination

3.1.1. Seed germination number

Generally, higher concentrations (≥0.0625 g/l) of *D. uncinatum* leaf treatments in both laboratory and screen house experiments were effective in suppressing *T. minuta* seed germination (Fig. 1). A concentration of 0.1 g/l of *D. uncinatum* leaf (DuL) extract was observed to be the most effective of all, with as much as twice the suppressive effect to those of DuR, DiL and DiR (Tables 1 & 2).

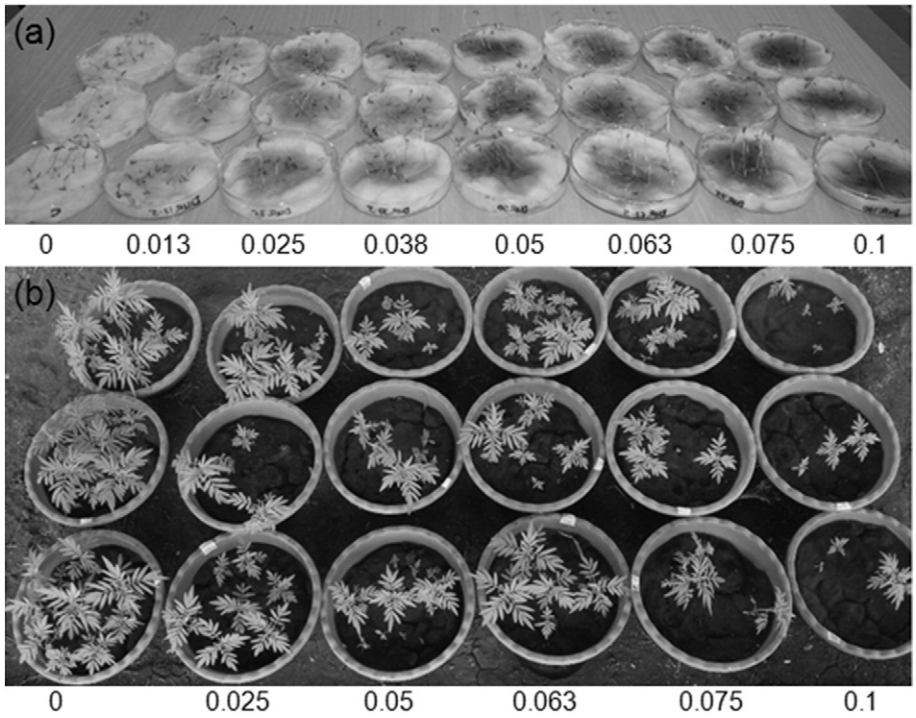

Fig. 1. *T. minuta* seedling germination trends in (a) laboratory experiment and (b) screen house experiment under DuL treatment (numbers represents concentration in g/l).

The mean percentage germination per treatment in and across each group differed significantly ($P < 0.05$), with DuL suppressing the germination of *T. minuta* by over 70% in both laboratory and screen house experiments ($F_{(7,31)} = 7.96$, $P = 0.003$ and $F_{(5,18)} = 19.49$, $P = 0.00002$ respectively), (Tables 1 & 2). Likewise, the mean germination per treatment was strongly negatively correlated with increasing DuL concentration but less strongly so with DuR, DiL and DiR concentrations (Tables 3 & 4). Fisher LSD revealed higher significance differences in mean percentage germination ($P < 0.05$) between lower and higher treatment concentration of DuL than those of DuR, DiL and DiR.

3.1.2. Seedling height

Seedling height differed significantly across all four groups in laboratory experimentation ($F_{(3,7)} = 3.42$, $P = 0.02$) (Table 3) but in a screen house experiment they were only significant in DuL and DiL treatments ($F_{(5,18)} = 11.13$, $P = 0.0004$ and $F_{(5,18)} = 7.89$, $P = 0.002$ respectively) (Table 4). Seedlings treated with higher treatment concentrations especially of DuL in both laboratory and screen house experiments were observed to be twice shorter than those treated with lower concentrations (Fig. 2). The seedling height per treatment were negatively correlated with treatments in DuL, DuR and DiL treatments with DuL having a strong negative correlation than the others in both laboratory and screen house experimentations ($\rho = -0.39$, $P < 0.05$ and $r = -0.89$, $P = 0.1 \times 10^{-5}$ respectively) (Tables 3 & 4).

3.1.3. Seedling fresh weight

Average seedling fresh weight differed significantly with DuL treatment in both laboratory and screen house experimentations ($H = 14.89$, $P = 0.002$ and $F_{(5,18)} = 7.31$, $P = 0.002$ respectively). DuL and DiL as well as DuR and DiR treatments were significantly negatively correlated with seedling fresh weight in laboratory and screen house experimentations (Tables 3 & 4). Seedlings treated with higher concentrations in both laboratory and screen house experimentations were observed to have lower fresh weights than those treated with lower concentrations which were most strongly observed for DuL (Fig. 3).

Table 1
Mean percentage germination of *T. minuta* seeds per treatment of *D. uncinatum* and *D. intortum* leaf and root extracts after 14 days of treatment in a laboratory experiment.

	D. uncinatum		*D. intortum*	
Concentration (g/l)	Leaves	Roots	Leaves	Roots
0.0000	91 ± 5c	80 ± 1c	88 ± 4a	82 ± 5
0.0125	60 ± 1b	80 ± 3c	84 ± 5a	75 ± 1
0.0250	60 ± 3b	64 ± 2abc	73 ± 3abc	60 ± 1
0.0375	44 ± 8b	55 ± 5ab	71 ± 2abc	62 ± 9
0.0500	60 ± 7b	55 ± 8ab	68 ± 1abc	57 ± 5
0.0625	28 ± 5a	73 ± 6bc	62 ± 8ab	64 ± 8
0.0750	31 ± 9a	62 ± 5abc	80 ± 1a	51 ± 8
0.1000	28 ± 8a	51 ± 5a	53 ± 3b	62 ± 8
Statistics	$F_{(7,24)} = 7.96^{**}$	$F_{(7,24)} = 2.93^{*}$	$F_{(7,24)} = 2.76^{*}$	$F_{(7,24)} = 1^{*}$

Values represent mean ± SE, values with dissimilar letter(s) in a column are significant by Fisher LSD at $P = 0.05$.
* $P \leq 0.05$.
** $P \leq 0.003$.

Table 2
Mean percentage germination of *T. minuta* seeds per treatment of *D. uncinatum* and *D. intortum* leaf and root extracts after 30 days of treatment in the screen house experiment.

	D. uncinatum		*D. intortum*	
Concentration (g/l)	Leaves	Roots	Leaves	Roots
0.0000	87 ± 0.2c	77 ± 0.2	93 ± 0.2	83 ± 0.2
0.0250	73 ± 0.2ac	77 ± 0.2	87 ± 0.2	87 ± 0.2
0.0500	63 ± 0.1a	73 ± 0.2	77 ± 0.2	73 ± 0.2
0.0625	60 ± 0.2a	70 ± 0.2	67 ± 0.2	80 ± 0.2
0.0750	37 ± 0.1b	67 ± 0.2	73 ± 0.2	80 ± 0.2
0.1000	27 ± 0.1b	60 ± 0.1	83 ± 0.2	87 ± 0.2
Statistics	$F_{(5,18)} = 19.49^{*}$	$H(5, N = 18) = 8.5^{**}$	$H(5, N = 18) = 7.2^{**}$	$H(5, N = 18) = 7.5^{**}$

Values represent mean ± SE, values with dissimilar letter(s) in a column are significant by Fisher LSD at $P = 0.05$.
* $P \leq 0.00002$.
** $P \geq 0.05$.

Table 3
Kruskal Wallis and one-way ANOVA test of seedling parameters (mean seedling fresh weight, height, chlorophyll content and % germination) per treatment after 14 days of treatment in a laboratory experiment ($H = H(7, N = 24)$).

Parameters	*D. uncinatum*		*D. intortum*	
	DuL	DuR	DiL	DiR
% germination	$F = 7.96^{***}$	$F = 2.93^{*}$	$F = 2.76^{*}$	$F = 1.00^{*}$
	$r = -0.48^{*}$	$r = -0.04$	$r = -0.19$	$r = -0.07$
Seedlings height	$H = 22.18^{**}$	$F = 6.38^{**}$	$H = 21.69^{**}$	$F = 143.79^{***}$
	$\rho = -0.39^{*}$	$r = -0.22$	$\rho = -0.57^{*}$	$r = -0.33$
Fresh weight	$H = 21.86^{**}$	$H = 22.30^{**}$	$H = 22.88^{**}$	$H = 22.60^{**}$
	$\rho = -0.24$	$\rho = -0.59^{*}$	$\rho = -0.39$	$\rho = 0.57^{*}$
Chl content	$H = 22.79^{**}$	$H = 22.58^{**}$	$H = 22.58^{**}$	$H = 22.58^{**}$
	$\rho = -0.20$	$\rho = -0.43^{*}$	$\rho = -0.20$	$\rho = -0.23^{***}$

r = Pearson's correlation coefficient and ρ = Spearman correlation coefficient at 95% C.I. DuL (*D. uncinatum* leaf), DuR (*D. uncinatum* root), DiL (*D. intortum* leaf) and DiR (*D. intortum* root).
* $P \le 0.05$.
** $P \le 0.003$.
*** $P \le 0.0003$.

3.1.4. Seedling total chlorophyll content

Seedling total leaf chlorophyll (Chl) differed significantly in both laboratory and screen house experimentations under DuL treatment ($H = 22.79$, $P = 0.003$ and $F_{(5,18)} = 16.39$, $P = 0.00005$ respectively) (Tables 3 & 4). Unlike in laboratory experimentation where only DuR treatment was having a significant negative correlation to Chl content, in screen house experimentation both DuL and DiL were significantly negatively correlated to Chl content (Tables 3 & 4). Seedlings treated with higher concentrations in all four groups were observed to have lower Chl contents than those treated with lower concentrations especially in DuL treatments (Fig. 4).

4. Discussions

Our results showed that high leaf and to some extent root crude extract concentrations of *D. uncinatum* can successfully be used as an alternative bioherbicide for controlling the exotic weed *T. minuta*, this method can be used instead of synthetic chemical herbicides. The latter can have a number of negative effects on the environment, particularly in protected areas and to human health [12,14,16,24]. The success of suppression is likely due to the strong allelopathic effects displayed by *D. uncinatum* [20]. In our study, one single application of 10 ml (petri dish) and 100 ml (pot) of leaf and or root crude extracts of this species to *T. minuta* seedlings suppressed seedling survival significantly by over

Table 4
Kruskal–Wallis rank sum and one-way ANOVA test of *T. minuta* seedling parameters (% germination, height, fresh weight and chlorophyll content) per treatment after 30 days of treatment in a screen house experiment ($H = H(5, N = 18)$ and $F = F_{(5,18)}$).

Parameters	*D. uncinatum*		*D. intortum*	
	DuL	DuR	DiL	DiR
% germination	$F = 19.49^{***}$	$H = 8.50$	$H = 7.19$	$H = 7.53$
	$r = -0.91^{*}$	$\rho = -0.47^{*}$	$\rho = -0.01$	$\rho = -0.31$
Seedlings height	$F = 11.13^{***}$	$F = 2.29$	$F = 7.89^{**}$	$F = 1.74$
	$r = -0.89^{*}$	$r = -0.65^{*}$	$r = -0.68^{*}$	$r = -0.34$
Fresh weight	$F = 7.31^{**}$	$F = 1.54$	$F = 2.74$	$F = 2.48$
	$r = -0.84^{*}$	$r = -0.45$	$r = -0.56^{*}$	$r = 0.13$
Chl content	$F = 16.39^{***}$	$F = 2.92$	$F = 1.23$	$F = 8.31^{**}$
	$r = -0.79^{*}$	$r = -0.17$	$r = -0.45$	$r = -0.66^{*}$

r = Pearson's correlation coefficient and ρ = Spearman correlation coefficient at 95% C.I. DuL (*D. uncinatum* leaf), DuR (*D. uncinatum* root), DiL (*D. intortum* leaf) and DiR (*D. intortum* root).
* $P < 0.05$.
** $P \le 0.008$.
*** $P \le 0.0003$.

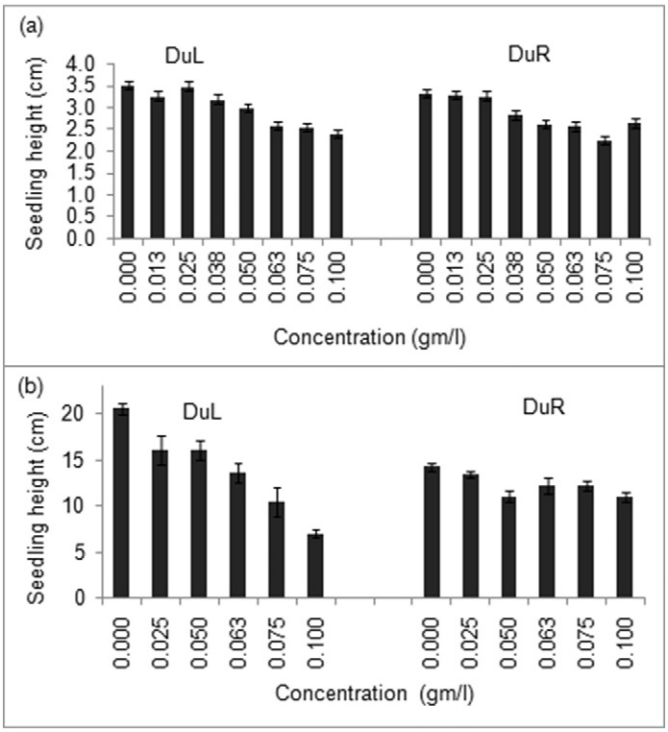

Fig. 2. Mean seedling heights (±S.E.) of germinated seeds in all groups (a) after 14 days in laboratory and (b) after 30 days in a screen house, DuL (*D. uncinatum* leaf), DuR (*D. uncinatum* root) treatments.

60%. Particularly high concentrations of *D. uncinatum* leaf extract displayed twice the suppressive effect on *T. minuta* seed germination and seedling vigor compared to *D. uncinatum* root or *D. intortum* leaf and root extracts. The suppressive effects of *D. uncinatum* were similarly reported by Pickett et al. [20] and Khan et al. [11] where the root exudes

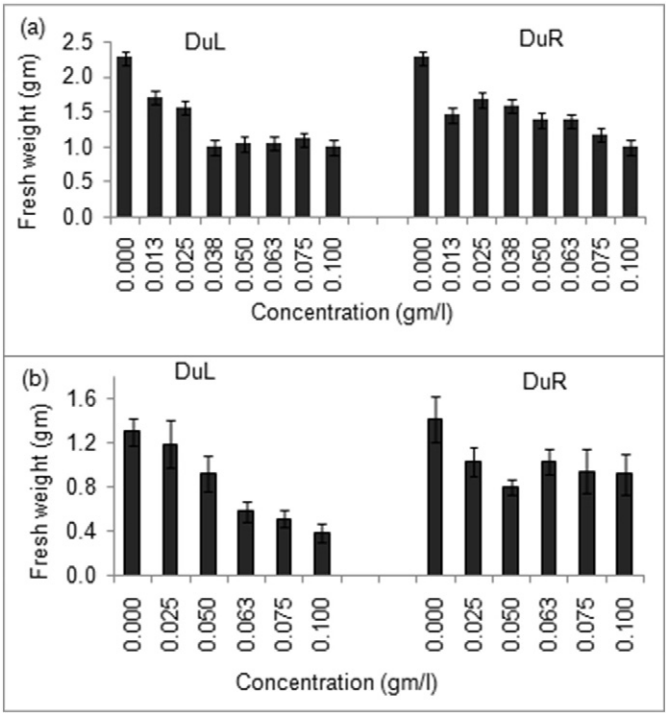

Fig. 3. Mean seedling fresh weight (±S.E.) of germinated seeds in all groups (a) after 14 days in laboratory and (b) after 30 days in a screen house DuL (*D. uncinatum* leaf), DuR (*D. uncinatum* root) treatments.

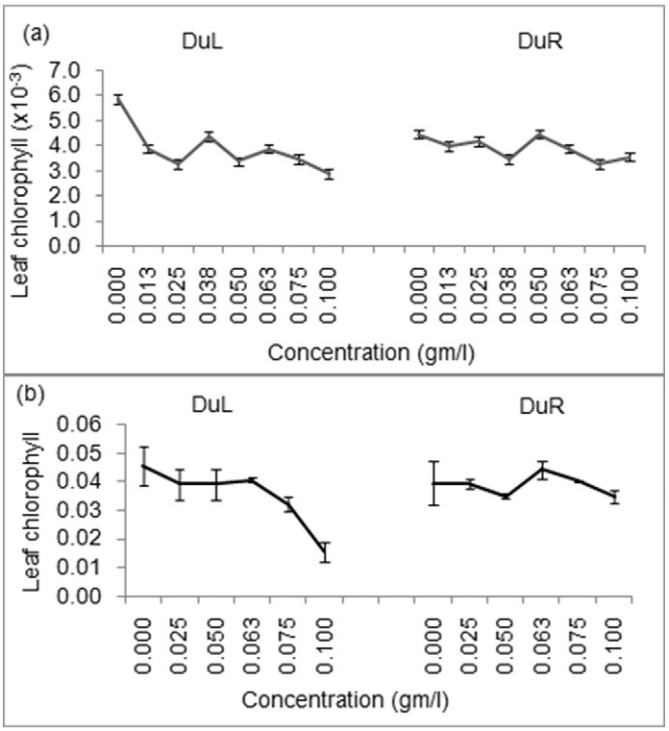

Fig. 4. Mean seedling chlorophyll content (±S.E.) of germinated seeds (a) after 14 days in laboratory and (b) after 30 days in a screen house, DuL (*D. uncinatum* leaf), DuR (*D. uncinatum* root) treatments.

of *D. uncinatum* successfully controlled *Striga hermonthica* and stemborers when planted as intercrop in a *Zea mays–Desmodium* species intercrop. Stronger allelopathic effects of *D. uncinatum* leaves compared to those of its roots could be due to its climbing nature as it has to outcompete other plants to ensure effective sun bathing for photosynthesis. A similar observation was reported by Carter et al. [2] where the adaptability of vines to low light environments was related to their climbing mechanics.

Additionally to *T. minuta* seedling germination rate, seedling height, fresh weight and total chlorophyll content were significantly suppressed by our extracts. Germination rate and seedling height are highly important in determining seedling vigor [30,10,28]. Likewise, chlorophyll content is the crucial plant component that determines the plant's ability to perform photosynthesis [5], a process that is essential for seedling growth and development and, therefore, vigor. While a low chlorophyll content has been associated with a plant's failure to compete for light and, thus, to survive [13] seedling height and fresh biomass are as well important in ensuring seedling's competitiveness. Short seedlings cannot compete well for light while those with low fresh biomass are more susceptible to the effects of trampling and other physical factors within their environment. Therefore, affecting these three parameters negatively can help in controlling the spread of the exotic weed *T. minuta*. While these three parameters can be efficiently suppressed through chemical herbicides such as Isoproturon (Proton 50% WP) [17], our study suggests a novel way of using natural components leading to the same effect but with less harm to the environment. As the mode of action of most allelochemicals is similar to synthetic herbicides [24], we suggest using *D. uncinatum* leaf for *T. minuta* weed management as a possible bioherbicide.

As expected, germination inhibition was found to increase with increasing treatment concentration, which was also reported by Cipollini et al. [3] where the allelopathic effects of *Alliaria petiolata*, *Lonicera maackii* and *Ranunculus ficaria* on the germination of three native woodland plants increased with treatment concentration. Hence, our newly discovered natural herbicide could be sprayed in affected areas (farmlands, rangelands and protected areas). The spraying of extracts as bioherbicides, for example, from *Sorghum* (*Sorghum bicolor* (L.) Moench), has proved to be a successful weed management tool without affecting productivity in cotton, soybean, wheat and rice [24]. Based on our results, we are expecting a suppression of more than 60% of *T. minuta* seedlings in affected areas with a single spray application of ≥62.5 mg/l of *D. uncinatum* leaf extract. As most allelochemicals have a decreased chemical environmental half-life [24], the application should be done seasonal. During our experimental trials on *T. minuta* we ensured that the germination was not hampered through deprivation of light, water or oxygen. In the laboratory experimentation temperature was maintained at 25 °C, the optimal temperature for *T. minuta* seed germination [4] throughout the study period and water was provided ad libitum. Further, petri dishes and pots position were moved after every three days to avoid other confounding factors that could have been triggered through location to influence seedling emergency. Our method was tested both in the laboratory and in the screen house where field conditions were adopted. We, therefore, expect successful suppressive effects in the field since the reduction in seedling survival was so strong in both laboratory and screen house experiments. One of the reason for poor performance of *D. intortum* observed in our study could be due to lack of combination, as through combination of methods control of some troublesome weeds have been reported whereby 100% control of *Striga asiatica* was attained using a combination of 100 kg N ha^{-1}, 1:1 (*Sorghum–Desmodium* ratio [21]). Another reason could be low concentration of allelochemicals due to the location as *D. intortum* was harvested from plots that were covered to 100% by *D. intortum* while *D. uncinatum* was obtained from plots that were intermixed with other plants. Therefore, *D. uncinatum* was already found under a competitive environment where it might have to produce more allelochemicals to suppress other plants.

5. Conclusions

While several control methods of *T. minuta* including the use of chemical herbicides have been reported [22] we claim that a natural extract can be a large-scale remedy and environmental friendly management alternative. Our findings suggest that *D. uncinatum* may probably be used to control invasive species, *T. minuta* and should be integrated into the management practices in the affected areas. Also our data suggest that a potential exists in devising an innovation that is both ecologically safe and effective by using *D. uncinatum* making it possible to improve rangelands production through planting them in the areas affected by invasive species. Moreover, *D. uncinatum* is preferred by herbivorous animals such as cattle [6] and readily available at a lower cost as it can be grown easily. Using *D. uncinatum* leaves extract will help improving the availability of native plants that provide food for the wild animals. It will keep the non-palatable *T. minuta* abundance low as germination of most of its seeds in the soil seed bank will be suppressed. This provides a beneficial control mechanism using a bioherbicide to suppress an aggressive invasives species in the long run.

Competing interests

The authors declare that they have no competing of interests.

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