

A search to reducing *Striga hermonthica* (Del.) Benth propagation using plant aqueous extracts with bio-herbicide properties on the seeds in vitro

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Abstract

Background

The genus *Striga* includes 11 parasitic plants species of food crops in at least 50 African countries. *Striga hermonthica* (Del.) Benth. is a major biotic constraint to the cereal crops production in Africa. It is the most widespread species in fields in Burkina Faso and grows on all types of soil inducing losses estimated at 35–40% on sorghum and millet. The significant reductions in yields result in insufficient food for the populations.

Methods

This study aims to identify local plants with bio-herbicidal properties for the management of *Striga hermonthica*. The inhibiting and stimulating effect of aqueous extracts from 13 local plant species on the germination of *Striga* seeds was assessed in vitro.

Results

The aqueous extracts from the leaves of *Azadirachta indica* A. Juss, *Jatropha curcas* L., *Jatropha gossypifolia* L., *Lawsonia inermis* L. and those from the leafy stems of *Cassia obtusifolia* L., *Crotalaria retusa* L., *Phyllanthus amarus* L. completely inhibited germination of *Striga*. However, those from five plants significantly stimulated germination of which the highest germination rate (60%) was recorded with the extract from *Euphorbia hirta* L. leafy stems.

Conclusions

The plant extracts thus constitute an ecological avenue for *Striga*. control. Further experiments could lead to the formulation of bio-herbicides against the parasitic plant to improve cereal production while limiting environmental pollution.

Background

Striga hermonthica, an obligate root-parasitic weed, is one of the most economically important parasitic plants among the 42 known *Striga* species. Its problem has become a major threat to food security, exacerbating hunger, and poverty in many African countries (Khan et al., 2014). A few estimates have indicated that *Striga* is causing enormous yield losses with a value ranging from 7 to 10 billion US\$ annually affecting the life of more than 300 million people in Africa (Gressel et al., 2004; Rodenburg et al., 2010).

Finding suitable *Striga* control strategies is crucial in order to reduce the extent of damage and also to retain further spread into the non-contaminant fields (Berner et al., 1995). A multitude of control methods such as chemical or mechanical means, the use of resistant varieties, cultural measures have been developed and proposed to address the challenge presented by *Striga* (Teka, 2014, Jamil et al., 2021). These approaches have been used in isolation or in integrated way to improve soil fertility or directly target the pest. Use of herbicides and synthetic suicidal germination agents to deplete *Striga* seed bank in infested soils has recently gained a lot of attention (Zwanenburg et al., 2016; Kountche et al., 2019).

However, the excessive use of synthetic pesticides in agriculture poses risks of environmental contamination and health issues (Anjarwalla et al., 2016). Thus, chemical pesticides are a global human rights concern (UN, 2017). In some countries, deaths from pesticide poisoning even exceed deaths from infectious diseases (Eddleston, 2002). Issues with synthetic pesticides have led to more targeted research and development of botanical pesticides (Anjarwalla et al., 2016). The use of plant pesticides has the advantage of respecting the environment while being effective in controlling pests (Stevenson et al., 2014; Mkenda et al., 2015). Their effect on non-target species is negligible compared to synthetic pesticides (Charleston et al., 2006; Amoabeng et al., 2013; Mkenda et al., 2015). Using of plants offers considerable potential for smallholder farmers but remains underexploited (Isman, 2006, 2008). Many pesticide plant compounds are present in foods and medicines (Anjarwalla et al., 2016). They are often used in the form of aqueous extracts or extracts made with solvents. The present study aims to identify Burkina Faso's local plants whose aqueous extracts have bio-herbicide properties for *Striga hermonthica* control. The specific objectives are: to identify aqueous plant extracts effective for the inhibition of *S. hermonthica* seed germination and, to identify the ones that are effective in *Striga* suicidal germination.

Material And Methods

Material

The plant material was *S. hermonthica* seeds harvested in 2015 from a sorghum field in the Kouaré village, in the Burkina Faso's eastern region of located between 11°55'03" North and, 0°30'58" East.

To obtain the plant extracts, thirteen (13) local plant species (Table 1) were harvested in 2016 in the vicinity of Ouagadougou, the Burkina Faso's Center region. The choice of these species was made on the basis of their availability, their bitter taste, their toxicity, or their properties.

Table 1
List of local plant species on which aqueous extractions have been performed

Plant species	Plant families	Plant material used
<i>Acacia nilotica</i> var. <i>adansonii</i> (L.) Willd. ex Delile	Fabaceae-Mimosoideae	Bark (B)
<i>Azadirachta indica</i> A. Juss.	Meliaceae	Bark and leaves (B + L)
<i>Balanites aegyptiaca</i> (L.) Delile	Balanitaceae	Bark (B)
<i>Cassia obtusifolia</i> L.	Fabaceae-Caesalpinioideae	Stem and leaves (St + L)
<i>Crotalaria retusa</i> L.	Fabaceae-Faboideae	Stem and leaves (St + L)
<i>Eucalyptus camaldulensis</i> Dehnh.	Myrtaceae	Leaves (L)
<i>Euphorbia hirta</i> L.	Euphorbiaceae	Stem and leaves (St + L)
<i>Jatropha curcas</i> L.	Euphorbiaceae	Leaves (L)
<i>Jatropha gossypifolia</i> L.	Euphorbiaceae	Leaves (L)
<i>Khaya senegalensis</i> (Desr.) A. Juss.	Meliaceae	Leaves (L)
<i>Lawsonia inermis</i> L.	Lythraceae	Leaves (L)
<i>Moringa oleifera</i> Lam.	Moringaceae	Leaves (L)
<i>Phyllanthus amarus</i> L.	Phyllanthaceae	Leaves (L)

Methods

Aqueous extraction

The extractions and tests were carried out in 2017 *in vitro* at the CREAM (Environmental, Agricultural and Training Research Center) in Kamboinsé in the Laboratory of Phytopathology and Weeds. The plant samples harvested were dried in the shade at ambient laboratory temperature (25–30°C) for at least 30 days and then ground to powder. Aqueous extracts were prepared by introducing 10 grams of the powder from each sample into an Erlenmeyer flask containing 100 ml of sterile distilled water. The mixture was placed on a stirrer and stirred for 24 h. The decoctions thus obtained were filtered and the filtrates which are aqueous extracts concentrated at 10% were used for the *Striga* seeds germination tests. Two samples of *Azadirachta indica* were used for the extractions: the first one on the leaves and the second one on the bark. This gives a total of 14 aqueous extracts used for the evaluation of inhibiting or stimulating properties on the germination.

Evaluation Of The Plant Extracts Inhibitory Effect On The Germination

Striga seeds were disinfected and packaged in Petri dishes (9 cm in diameter) containing filter paper with 3 ml of extract per dish. Seeds were placed on 6 mm diameter Wattman GF/A filter paper discs whose 20–30 seeds per disc. Four (4) disks were placed equidistantly in each Petri dish. The treatments in comparison were: 1) *Striga* seeds conditioned with sterile distilled water (control); 2) *Striga* seeds conditioned with each of the 14 plant extracts.

Petri dishes were sealed and wrapped with aluminum foil and then incubated at 28°C for 14 days. At the end of 14 days, 25 µl of the GR24 (0.0001%), synthetic germination stimulant was applied to each disc bearing the *Striga* seeds. The dishes were sealed and rewrapped for a 72 h incubation after which they were opened to observe the germinated seeds under a binocular microscope. Germination rates were calculated to determine the effectiveness of each treatment in inhibiting seed germination. For each test, three Petri dishes, each corresponding to one repetition, were used per treatment and the test was repeated three times. This corresponds to 9 repetitions per treatment.

Evaluation of the plant extracts stimulatory effect on the germination.

Disinfected seeds were again placed on 6 mm diameter discs (20–30 seeds/disc) and then in Petri dishes (9 cm diameter) with sterile distilled water by the reason of four (4) discs per dish. Dishes were sealed, wrapped with aluminum foil, and incubated at 28°C for 14 days. Then, 25 µl of extract (10%) or GR24 (0.0001%) was applied to the seeds of each disc. The dishes were further sealed and wrapped for another 72 hours of incubation after which they were observed under a binocular microscope to calculate the induced germination rates. Three dishes were used per extract or per control treatment (GR24). The test was repeated 3 times and the efficacy of the extracts to stimulate the *Striga* seed germination was evaluated.

Data Statistical Analysis

The data collected were subjected to one-way analysis of variance (ANOVA) through GenStat Release 12.1. software (PC/Windows Vista), VSN International Ltd. A comparison of germination rates averages was performed by the Newman Keuls Student test at the 5% level.

Results

Inhibition of germination by the plant aqueous extracts

The results from the analysis of variance of the data collected by test are recorded in Table 2. The ANOVA thus indicated significant differences ($P < 0.001$) between the average germination rates of the *Striga* seeds obtained under the effect of the control and the 14 aqueous plant extracts. In all the tests, the

extract from *Euphorbia hirta* followed by those from *Eucalyptus camaldulensis*, *Azadirachta indica*, *Khaya senegalensis*, *Moringa oleifera* are those which inhibited germination less. The other nine extracts completely inhibited germination.

Table 2
Germination rates at the end of the three inhibition tests

Treatments	Test 1 (%)	Test 2 (%)	Test 3 (%)
H ₂ O	92.84a	82.79a	84.80a
<i>Euphorbia hirta</i> (L + St)	77.87b	51.50c	48.74bc
<i>Eucalyptus camaldulensis</i> (L)	57.23c	49.04c	54.77b
<i>Azadirachta indica</i> (B)	53.65c	38.98d	39.76c
<i>Khaya senegalensis</i> (B)	43.27d	74.10b	42.58c
<i>Moringa oleifera</i> (L)	33.86e	29.04e	39.68c
<i>Acassia nilotica</i> (B)	0.40f	0.00f	0.00d
<i>Azadirachta indica</i> (L)	0.00f	0.00f	0.00d
<i>Balanites aegyptiaca</i> (B)	0.00f	0.00f	0.00d
<i>Cassia obtusifolia</i> (L + St)	0.00f	0.00f	0.00d
<i>Crotalaria rotusa</i> (L + St)	0.00f	0.00f	0.00d
<i>Jatropha curcas</i> (L)	0.00f	0.00f	0.00d
<i>Jatropha gossypifolia</i> (L)	0.00f	0.00f	0.00d
<i>Lawsonia inermis</i> (L)	0.00f	0.00f	0.00d
<i>Phyllanthus amarus</i> (L + St)	0.00f	0.00f	0.00d
Mean	23.94	21.70	20.69
LSD (5%)	9.402	8.687	8.733
cv%	48.7	49.7	52.4
F pr	< 0.001	< 0.001	< 0.001
Key: LSD (5%) = Least significant differences of means (5% level); cv = coefficient of variation, F Pr = Fisher probability; L = leaves; St = stems; B = bark			

In a column, means followed by the same alphabet letter are not significantly different.

The comparison of the means showed that the germination rate (GR) of the control (86.81%) is significantly higher than those of all the plant extracts (Fig. 1). Five (5) extracts reduced the germination

of *S. hermonthica* seeds with inhibition rates ranging from 31.61% (leafy stems of *Euphorbia hirta*) to 60.60% (leaves of *Moringa oleifera*) compared to the control. Among these five extracts, two are not significantly different (those of *Eucalyptus camaldulensis* leaves and *Khaya senegalensis* bark) while the other three extracts are significantly different. The GR of *S. hermonthica* seeds conditioned with eight (8) extracts (those from the leaves of *Azadirachta indica*, *Jatropha curcas*, *J. gossypifolia*, *Lawsonia inermis*, leafy stems of *Cassia obtusifolia*, *Crotalaria retusa*, *Phyllanthus amarus*) were nil (0%). The GR average obtained with the *Acacia nilotica* bark extract (0.13%) is not significantly different from that of the eight extracts which each generated a zero GR (Fig. 1).

Figure 1

Inhibition effect of aqueous plant extracts on the *Striga* seeds germination.

Key

L = leaves; St = stems ; B = bark

The means followed by the same alphabet letter are not statistically different at the 5% level according to the Newman Keuls Student test

Stimulation Of Germination By The Plant Aqueous Extracts

The results of the analysis of variance (ANOVA) of the data obtained from the three stimulation tests are recorded in Table 3. For each of the tests, the means of the GR presented significant differences with P (probabilities) < 0.001. Control GR24 stimulated germination more than plant extracts. In test 1, extract from *Euphorbia hirta* was statistically identical to control GR 24 in terms of its ability to stimulate germination. In tests 2 and 3, the extract from *Euphorbia hirta* is the one that still stimulated the most germination among the 14 extracts but with rates significantly lower than that of the control.

Table 3
Germination rates at the end of the three stimulation tests

Treatments	Test 1 (%)	Test 2 (%)	Test 3 (%)
GR24	69.31a	96.76a	97.96a
<i>Euphorbia hirta</i> (L + St)	66.75a	56.11b	57.34b
<i>Azadirachta indica</i> (B)	29.65b	25.15c	28.11c
<i>Khaya senegalensis</i> (B)	8.30c	7.13d	2.07 ^e
<i>Moringa oleifera</i> (L)	3.66cd	3.13e	7.62d
<i>Eucalyptus camaldulensis</i> (L)	3.48 d	3.29e	2.68 ^e
<i>Acassia nilotica</i> (B)	0.00d	0.00f	0.00 ^e
<i>Azadirachta indica</i> (L)	0.00d	0.00eg	0.00 ^e
<i>Balanites aegyptiaca</i> (B)	0.00d	0.00eg	0.00 ^e
<i>Cassia obtusifolia</i> (L + St)	0.00d	0.00eg	0.00 ^e
<i>Crotalaria rotusa</i> (L + St)	0.00d	0.00eg	0.00 ^e
<i>Jatropha curcas</i> (L)	0.00d	0.00eg	0.00 ^e
<i>Jatropha gossypiiifolia</i> (L)	0.00d	0.00eg	0.00 ^e
<i>Lawsonia inermis</i> (L)	0.00d	0.00eg	0.00 ^e
<i>Phyllanthus amarus</i> (L + St)	0.00d	0.00eg	0.00 ^e
Mean	12.08	12.77	13.05
LSD (5%)	5.537	2.652	3.908
cv%	56.9	25.8	37.1
F pr	< 0.001	< 0.001	< 0.001
Key: LSD (5%) = Least significant differences of means (5% level); cv = coefficient of variation, F Pr = Fisher probability; L = leaves; St = stems ; B = bark			

In a column, means followed by the same alphabet letter are not significantly different

ANOVA of the data from the three tests combined showed significant differences ($P < 0.001$) between the germination percentages caused by the 14 plant extracts and the control on the *Striga* seeds (Fig. 2). Comparison of means showed that GR24 (positive control) induced the highest *Striga* seed germination rate (88.01%). It is followed in descending order by those obtained with extracts from leafy stems of

Euphorbia hirta (60.06%), leaves of *Moringa oleifera* (27.64%), *Eucalyptus camaldulensis* (5.83%), *Azadirachta indica* bark (4.87%) and *Khaya senegalensis* (3.15%). The average germination rates of the last three extracts are not significantly different. However, those induced by the extracts of *Euphorbia hirta* and *Moringa oleifera* are different and superior to those obtained with the other extracts. As for the extracts from the leafy stems of *Cassia obtusifolia*, *Crotalaria retusa*, from *Phyllanthus amarus*, from the leaves of *Azadirachta indica*, *Jatropha curcas*, *Jatropha gossypifolia*, *Lawsonia inermis* and from the bark of *Acacia nilotica*, *Balanites aegyptiaca*, they were statistically identical with a zero rate (0%).

Figure 2 : Stimulation effect of aqueous plant extracts on the *Striga* seeds germination

Key

L = leaves; St = stems; B = bark

The means followed by the same alphabet letter are not statistically different at the 5% level according to the Newman Keuls Student test

Discussion

Aqueous extracts from eight (8) plants including *Azadirachta indica* (L), *Jatropha curcas*, *Jatropha gossypifolia*, *Lawsonia inermis*, *Cassia obtusifolia*, *Crotalaria retusa*, *Phyllanthus amarus*, *Balanites aegyptiaca* which completely inhibited germination, despite the application of the GR24 stimulant, would probably contain molecules with allelochemical properties on the *Striga* seed germ. The germination inhibition rate of the aqueous extract of *E. camaldulensis* leaves obtained in this study (38.16%) is lower than that recorded by Yonli et al. (2010) with the same extract (84.33%) at the same concentration of 10%. In contrast, *A. indica* leaf powder evaluated under greenhouse and field conditions in the Nigerian savannah by Marley et al. (2004) was not significantly different from the control in reducing *Striga* emergence. The inhibiting effect of leaf extracts from *A. indica*, *J. curcas* and *J. gossypifolia*, concentrated at 10%, had already been showed (Yonli et al., 2010). However, the inhibition was not total as is the case in the present study. The aqueous extract of the seeds of *Trigonella foenum-graecum* L., a medicinal plant of the Fabaceae family, also significantly inhibited the germination of *S. hermonthica* seeds (Hassan et al., 2013).

The results recorded with the extracts of *C. obtusifolia*, *C. retusa* and *P. amarus* have particular advantages due to the fact that these plants are wild herbaceous and often weeds of crops that can invade plots. A large-scale application of these results would also make it possible to control these weeds. Virtues of some of these plants had been reported by other authors. For example, the inhibitory activity of aqueous and ethanolic extracts from *P. amarus* on the in vitro growth of seven strains of *Mycobacterium ulcerans*, responsible for ulcers, has been reported in Côte d'Ivoire (Coulibaly et al., 2011).

The stimulatory effect of five (5) aqueous plant extracts (*Euphorbia hirta*, *Azadirachta indica*, *Khaya senegalensis*, *Moringa oleifera*, *Eucalyptus camaldulensis*) concentrated at 10% revealed in this study,

promises efficient management of *Striga*. Indeed, the induction of the germination of *Striga* seeds whose dormancy had been previously lifted, is of great agronomic interest. Extracts from the leafy stems of *Euphorbia hirta* and from the bark of *A. indica* were the most effective in stimulation, resulting in germination rates of 60.06% and 27.64%, respectively. These extracts could contain chemical elements present in strigolactones and their analogues. Tests involving higher and lower concentrations than 10% of the extracts could result in higher germination rates. This will allow to identify the optimal concentration of each extract. The germination stimulating property would be even more beneficial for an ecological management. It could be exploited to reduce or even eliminate the stocks of seeds of the parasitic plant and to clean up crop plots by causing suicidal germinations. Furthermore, Van Mourik (2007) reported that the germination of *Striga* seeds is the main factor in the reduction of the seed -bank in the soil.

The ethyl acetate and butanol fractions of *Euphorbia hirta* exhibited antifungal activity against *Phoma sorghina*, a parasite of sorghum through inhibition of mycelial growth (Karanga et al., 2017). The use of *E. hirta* would therefore be promising because it will make it possible to control both the fungal pathogen *P. sorghina* and *S. hermonthica* without risk of environmental pollution. The aqueous extract from the leaves of *Eucalyptus camaldulensis* concentrated at 1% significantly stimulated the germination of *Striga* seeds at 38.9% (Yonli et al., 2010). Compared to the stimulation rate of 3.15% of the same extract at the concentration of 10% in this study, one could say that the lower dose stimulates germination more effectively. The results on the leaf extracts of *A. indica*, *J. curcas*, *J. gossypiiifolia* perfectly corroborate those of the same authors where no stimulation was obtained with these extracts. Irrigation of a maize field sown on soil infested by *S. hermonthica* with an aqueous extract of *Desmodium uncinatum* plants resulted in a highly significant reduction in infestation (Qasem, 2006). The efficacy of seed from *Azadirachta indica* (neem) seeds, and fruit pod powders from *Parkia biglobosa* (Jacq.) R.Br. ex G. Don in reducing infestation of *Striga* was evaluated under greenhouse and field conditions in the Nigerian Savannah (Marley et al., 2004). Neem seed powder was the most effective, with only 16.5% *Striga* emergence. It is followed by fruit powder and *P. biglobosa* pod powder, with an emergence of 29.1% and 38.8% of *Striga* respectively. In addition, the application of the powder of the pods of *P. biglobosa* in peasant fields in the center of Burkina Faso, allowed a reduction in the emergence of *S. hermonthica* and an increase in the contents of macro elements of the soil, with a surplus of maize yield (Kambou et al., 2000). Endogenous plants whose effect of stimulating or inhibiting the germination of *S. hermonthica* seeds has been revealed can thus be evaluated under natural conditions with a view to transferring green technology.

Conclusion

The results obtained in this study on the inhibition and induction of the germination of *Striga hermonthica* seeds confirm that in addition to the multiple virtues in the medical and cosmetic fields, the plants constitute an alternative in this parasitic plants management. Aqueous extracts of *Euphorbia hirta*, through stimulation of germination and of *Azadirachta indica*, *Cassia obtusifolia*, *Crotalaria retusa*, *Jatropha curcas*, *J. gossypiiifolia*, through inhibition, are potential bio-herbicides against *Striga*.

Metabolites from these local plants can be used in the control of *S. hermonthica* and improve the yield of the cereal crops host. This experiment carried out in vitro presents a first step in the search for bio-herbicide products to control *Striga hermonthica*. Additional tests will determine the optimal concentration of each extract for effective action. This would lead to an application of the results in natural conditions to improve food security.

Abbreviations

GR

Germination Rate

GR24

3[2,5-dihydro-3-methyl-2oxo-5-furanyl] oxymethylene-3,3a, 4,8b-tetrahydroindeno-[1,2-b] furan-Z-one

H₂O

Dihydrogen monoxide

UN

United Nations

DY

Djibril YONLI

HT

Hamidou TRAORE

JIB

Joseph Issaka BOUSSIM

TCSI

Tinkoudougou Cathérine SAWADOGO/ILBOUDO

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Declarations

Ethics approval and consent to participate

Not applicable

Consent for publication

Not applicable

Competing interests

The authors declare that they have no competing interests.

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Author contributions

DY, PZ, HT, JIB conceived and designed the project. TCSI applied the project for funding. TCSI and SS performed the laboratory assays. DY and TCSI conducted the data analysis. TCSI prepared the manuscript with substantial input and review from DY, HT, JIB. All authors read and approved the final manuscript.

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Availability of data and materials

The datasets used and analyzed during the current study are available from the corresponding author on reasonable request.

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Figures

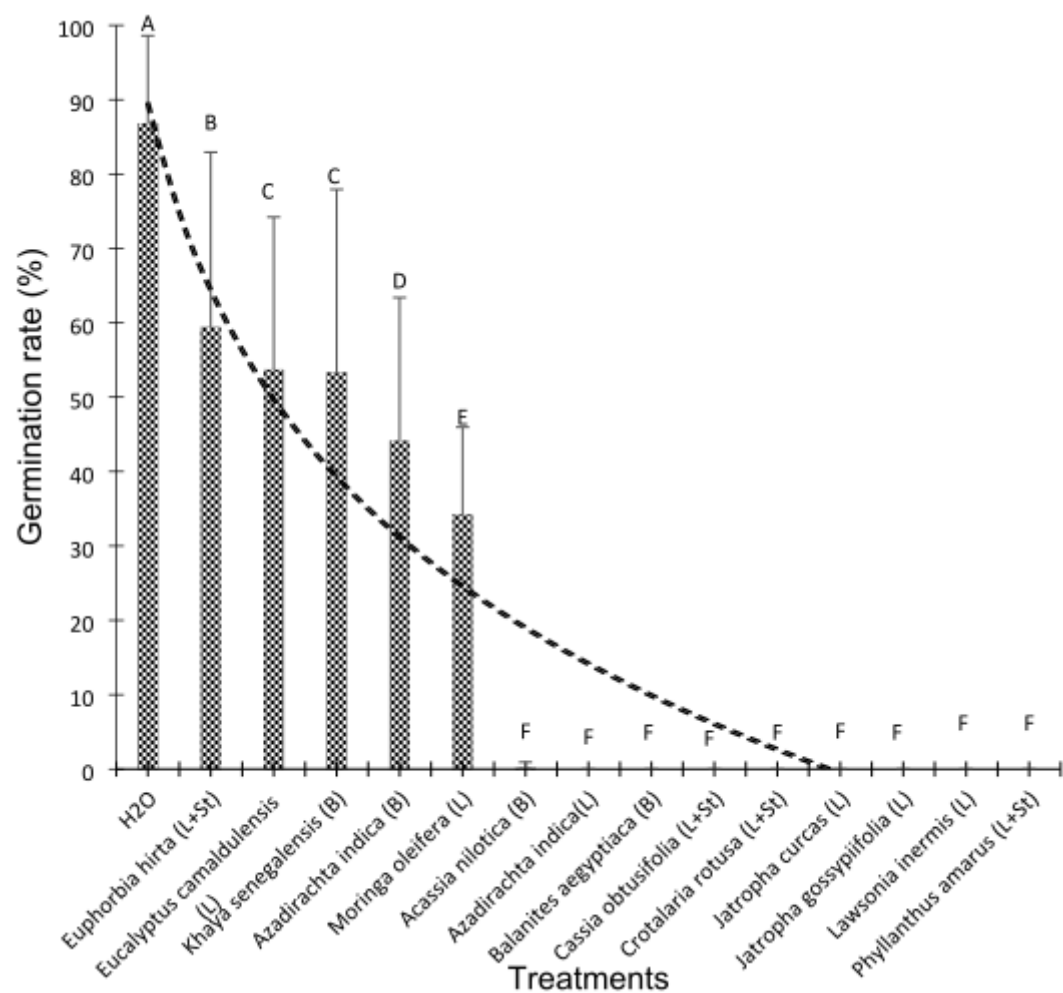


Figure 1

Inhibition effect of aqueous plant extracts on the *Striga* seeds germination.

Key: L = leaves; St = stems ; B = bark

The means followed by the same alphabet letter are not statistically different at the 5% level according to the Newman Keuls Student test

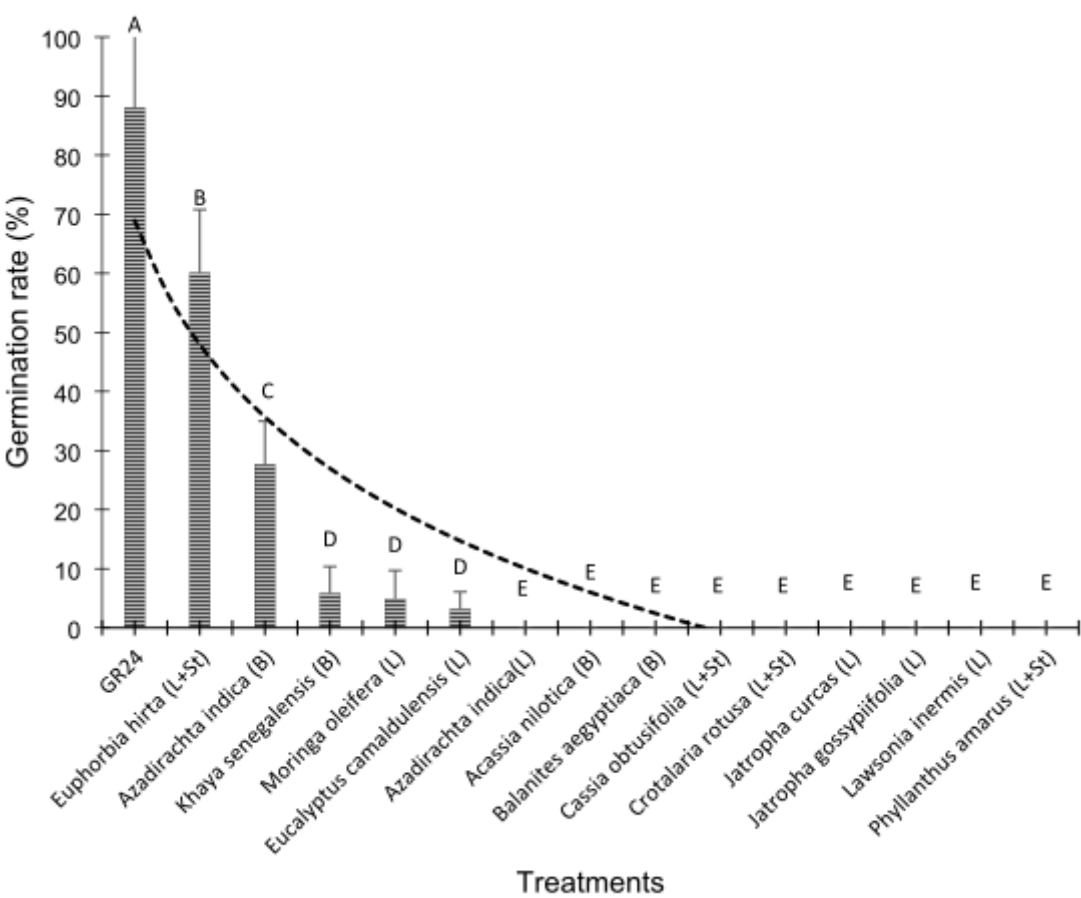


Figure 2
Stimulation effect of aqueous plant extracts on the *Striga* seeds germination

Key: L = leaves; St = stems; B = bark

The means followed by the same alphabet letter are not statistically different at the 5% level according to the Newman Keuls Student test