



Original Research

Use of essential oils from local plants as potential bio-herbicides to deplete *Striga hermonthica* seedbank

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ABSTRACT

The parasitic weed *Striga hermonthica* (Del.) Benth, is a major biotic constraint to the production of cereal crops, particularly sorghum in Africa. The research was aimed to evaluate the allelopathic properties of local plants to inhibit *Striga hermonthica* weed from Burkina Faso. In this respect, the bio-herbicidal effect of essential oils from 10 local aromatic plants on the germination of *S. hermonthica* seeds, was evaluated. The research was conducted in 2017 at the phytopathology and weed science laboratory of INERA (Institut de l'Environnement et de Recherches Agricoles) located at Kamboinsé, Burkina Faso. The research using essential oils with 10% dilutions from 10 local aromatic plants were evaluated to inhibition of *S. hermonthica* germination. Dilutions of 0.2% and 0.4% were applied on conditioned *Striga* seeds to test their ability to induce *Striga* germination. Parameters were analyzed using the ANOVA, and the means were compared using DMRT at level of 5%. Germination data of *Striga* seeds were analyzed using the GenStat Release 12.1 software. The results showed that oils from *Cymbopogon citratus* (10%) inhibited completely the germination. The ones from *Cymbopogon nardus* L. and *Lippia multiflora* Moldenke also inhibited significantly the germination rate compared to the untreated control. Dilutions of 0.2% from four plants essential oils significantly stimulated germination by 5.26% to 60.64% (*C. citratus*). As we can observe from the above, the oil some essential oils contain metabolites that can be used as bio-herbicides to control *S. hermonthica* and improve cereal yield. Their use in field conditions can limit environmental pollution and contribute to fight against climate change.

Introduction

Striga genus is found in at least 50 countries in Africa (Rodenburg et al. 2016). There are about 40 *Striga* species, including 11 food crop pests (Elzein and Kroschel, 2003). All *Striga* species

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require host stimulant for seed germination to start plant growth (Qasem, 2006). Germination stimulants are specific chemicals present in the rhizosphere of host plants and some non-host plants (Wigchert, 1999). *Striga aspera* (Willd.) Benth., *Striga asiatica* (L.) Kuntze and *Striga hermonthica* caused an annual economic loss of US \$117 million in rain-fed upland rice in sub-Saharan Africa whose US \$ 1.56 million in Burkina Faso (Rodenburg et al. 2016). *S. hermonthica* which is the most economically significant among species, spreading in western, eastern and central Africa (Gressel et al. 2004). Sorghum growing area infested by *Striga* in Burkina Faso has been estimated at 1,319,000 ha caused 35-40% yield losses (Gressel et al. 2004). A healthy and well-developed *Striga* plant may produce up to 500,000 seeds that can stay dormant in the soil for 10-14 years (Qasem, 2006). The control of parasitic weeds is very difficult because of their ability to produce many seeds as well as the strong physiological interaction with their hosts (Elzein and Kroschel, 2003). Since 1960, one of control options of *S. hermonthica* in West Africa is based on chemical herbicides use (Hoffmann et al. 1997). However, excessive use of synthetic pesticides caused problems such as environmental pollution, pest resistance and human diseases (Anjarwalla et al. 2016). Chemical pesticides are global human rights concern (Mehdizadeh, 2019) and their use leads to detrimental consequences on nutritional supply (United Nations, 2017). In many countries, death rate from pesticide poisoning even exceeds those from infectious diseases (Eddleston, 2002). Problems caused by synthetic pesticides led to the development of botanical pesticides (Anjarwalla et al., 2016). Nevertheless, the use of pesticidal plants has the advantage of protecting the environment and being effective against pests (Mkenda et al. 2015; Stevenson et al. 2014; Mushtaq et al. 2020). Their application offers considerable potential for small farmers but remains underutilized (Isman, 2006, 2008). Their negative effects on targeted and non-targeted species are very low, compared to synthetic pesticides (Amoabeng et al. 2013; Charleston et al. 2006; Mkenda et al. 2015). Pesticidal plants are often used in the form of extracts (Mehdizadeh and Mushtaq, 2019).

Essential oils may also have potential pesticidal effects (Isman, 2000). Many of the pesticidal plant compounds are found in food and medicines (Anjarwalla et al. 2016). Essential oils from plants have been reported as anti-malarial, anti-inflammatory, anti-fungal, and anti-bacterial from Burkina Faso (Baba et al. 2012; Erler et al. 2006; Ntonga et al. 2014; Paranagama et al. 2003; Voundi et al. 2015). These extracts showed effective activities against crop pathogens (Bonzi et al. 2013; Zida et al. 2008). Essential plant oils showed also some allelopathic effect on seed germination of weed species (Angelini et al. 2003; Azirak and Karaman, 2008; Paudel and Gupta, 2008). The objective of this study is to evaluate in bioassay the efficacy of essential oils of local plants to stimulate or inhibit *Striga hermonthica* seed germination.

Materials and Methods

Plant material

Striga hermonthica seeds were harvested in 2015 in sorghum field in the Kouaré village (11°95'03" N and 0°30' 58" E) located in the Eastern Burkina Faso. Ten (10) local aromatic plants (Table 1) were harvested in 2016 in Plateau Central region of Burkina Faso. Their essential oils were extracted by steam distillation and tested on *S. hermonthica* seed germination. The choice of these plant species was made according to their aromaticity, their virtues and their availability across the country.

Table 1. Local plant species and materials used for essential oil extractions.

Scientific name	Plant family	material used
<i>Cymbopogon citratus</i> L.	Poaceae	stems + leaves
<i>Cymbopogon nardus</i> L.	Poaceae	stems + leaves
<i>Cymbopogon schoenanthus</i> (L.) Spreng.	Poaceae	stems + leaves
<i>Eucalyptus camaldulensis</i> Dehnh.	Myrtaceae	Stems + leaves + flowers + fruits
<i>Hyptis spicigera</i> Lam.	Lamiaceae	Stems+ leaves + flowers + fruits
<i>Hyptis suaveolens</i> L. Poit.	Lamiaceae	stems + leaves
<i>Lantana camara</i> L.	Verbenaceae	Stems + leaves+ flowers + fruits
<i>Lippia multiflora</i> Moldenke	Verbenaceae	Stems + leaves + flowers + fruits
<i>Ocimum americanum</i> L.	Lamiaceae	whole plant
<i>Ocimum gratissimum</i> L.	Lamiaceae	whole plant

Evaluation of essential oil effects on Striga hermonthica seed germination

Inhibition bioassay

Thirty to 40 surface sterilized *Striga* seeds were placed in Petri dishes (9 cm Ø) containing glass microfibre filter paper (GF/A) discs of 6 mm diameter. Each dish was lined with a double moistened Whatman filter paper. The dishes containing *Striga* seeds were sealed, wrapped in aluminum foil and incubated at 28 °C in darkness for 7 days for seed conditioning in sterile water. After the incubation, 0.7% agar solution was poured into other sterile Petri dishes (9 cm Ø). Four cavities were lightly made on the solidified agar gel in each dish. Twenty five (25) microliters of each diluted oil was applied into each cavity. Then, a disc bearing conditioned *Striga* seeds was placed on the top cavity containing diluted oil. The whole was sealed and incubated at 28 °C for 7 days and then, 20 microliters of GR 24 (0.0001%) were applied on *Striga* seeds to stimulate their germination and incubated for 72 h at 28 °C.

Then oils were assessed for their ability to inhibit the germination of *Striga* seeds. Acetone (80%) was used for each oil to prepare stock solutions (Paudel et Gupta, 2008). Then, a dilution of 10% oils concentration were prepared from each stock solution and used to condition the *Striga* seeds. Three treatments were compared: 1) *Striga* seeds conditioned with sterile distilled water (negative control); 2) *Striga* seeds conditioned in acetone (positive control); 3) *Striga* seeds conditioned in oil 10% diluted oil. Three Petri dishes were used for each treatment and the experiment was replicated three times.

The ability of each diluted oil to inhibit *Striga* seed germination was assessed by counting the number of germinated seeds under microscope and the germination rates were therefore derived.

Stimulation bioassay

Thirty (30) to forty (40) surface sterilized *Striga* seeds were placed in Petri dishes (9 cm Ø) containing Whatman GF/A filter paper discs (6 mm). Each Petri dish contained a moistened Whatman double filter paper. The Petri dish was packaged as previously described and *Striga* seeds were conditioned in sterile water for 14 days (Yonli et al., 2010). After the incubation, 25 µl of 0.2% and 0.4% diluted oils were introduced in each of the four cavities made on solidified Agar gel (0.7%) in sterile Petri dishes. A disc containing conditioned *Striga* seeds was placed on the top of each cavity. Diluted oils were compared to GR 24 (0.0001%) and the treatments were as follows: 1) *Striga* seed germination induced by sterile distilled water (negative control); 2) *Striga* seed germination induced by GR 24 (positive control); 3) *Striga* seed germination induced by acetone (Negative control) and; 4) *Striga* seed germination induced by each concentration of ten diluted oils. The sealed Petri dishes were wrapped in aluminum foil and black polyethylene and incubated in darkness at 28 °C for 7 days. After this incubation, the ability of each treatment to stimulate *Striga* seeds germination was evaluated by counting the number of germinated seeds under microscope. Three Petri dishes were used for each treatment and the experiment was replicated three times.

Data statistical analysis

Germination data of *Striga* seeds were analyzed using the GenStat Release 12.1 software. An analysis of variance (ANOVA) followed by a comparison of germination rates averages was performed by the Newman Keuls Student test at the 5% level.

Results and Discussion

Inhibition effect of essential oils on Striga hermonthica seed germination

The germination rates of *Striga hermonthica* seeds conditioned in essential oils dilutions (10%) recorded with the three inhibition tests are shown in Table 2. The ANOVA results showed an average of 39.92% with significant differences ($P < 0.0001$) between the germination rates in the 10% dilutions from the 10 essential oils and the controls.

Table 2. Germination rates of *Striga hermonthica* seeds conditioned in 10% essential oils dilutions.

Treatments	Test 1 (%)	Test 2 (%)	Test 3 (%)
H ₂ O	90.09 a	87.58 ab	89.61 a
Acetone	90.08 a	87.28 ab	89.32 a
<i>L. camara</i>	55.74 b	90.96 a	48.65 b
<i>E. camaldulensis</i>	42.26 c	84.13 ab	43.34 bc
<i>H. spicigera</i>	35.83 cd	83.29 ab	36.79 cd
<i>C. nardus</i>	31.29 de	5.63 f	31.11 d
<i>H. suaveolens</i>	24.95 e	69.48 cd	23.41 e
<i>C. schoenanthus</i>	7.17 f	74.97 bc	8.33 f
<i>O. americanum</i>	1.17 f	62.47 d	1.15 f
<i>L. multiflora</i>	0.79 f	19.54 e	0.92 f
<i>O. gratissimum</i>	0.61 f	18.63 e	0.46 f
<i>C. citratus</i>	0.00 f	0.00 f	0.00 f
Mean	31.67	57.00	31.09
cv%	30.6	21.4	29.3
F pr	<.001	<.001	<.001

Key: cv = coefficient of variation; F Pr = Fisher probability. Means followed by the same alphabet letter are not significantly different

Both negative controls (H₂O and acetone) induced about 90% of germination and there was not significant difference between them (Figure 1). Only the 10% dilution from *C. citratus* essential oil completely inhibited the germination of *Striga* seeds (100%). It was followed by *O. gratissimum*, *L. Multiflora* and *C. Nardus* with respectively inhibition rates of 92.60%, 92.12% and 74.54% in comparison to the untreated control (H₂O). A significant inhibition of high concentration of essential oil from *C. citratus* in a laboratory bioassay, on germination and seedling growth of the weed *Echinochloa crus-galli* is reported by Poonpaiboonpipat et al. (2013). This ability could be due to the presence of the citral the main compound of this essential oil. Likewise, allelochemical effects of essential oils were shown (Dudai et al. 1999; Sulastri et al. 2019).

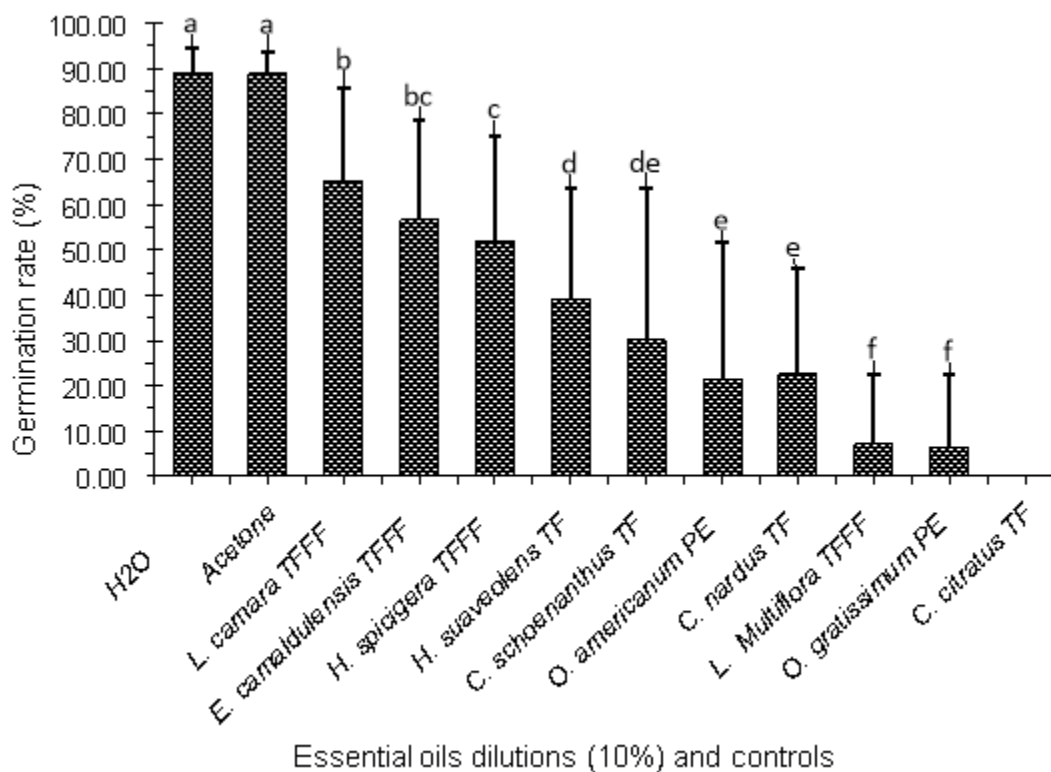


Figure 1. Inhibition effect of local plants essential oils on *Striga hermonthica* seeds germination. Means followed by the same alphabet letter are not significantly different.

These results made it possible to know some herbicide properties of the ten plants, on the parasitic weed, *Striga hermonthica*. The oils from *C. nardus* and *L. multiflora* diluted to 10% which significantly reduced the germination rates would contain some molecules that inhibit germination. The aqueous extracts from leaves and stems of *H. Spicigera* and *L. camara* and roots of *E. camaldulensis*, concentrated at 10%, nevertheless significantly inhibited the germination of *S. hermonthica* seeds by 59.5%, 71.7% and 85.7% respectively, compared to the untreated control. Essential oil from *Cymbopogon nardus* which inhibited *Striga* germination, also showed inhibitory activity on fungal strains including dermatophytes at 75 to 200 $\mu\text{g.ml}^{-1}$ (Koba et al. 2004). The inhibitory properties of *C. nardus* could be due to a toxicity of its essence. Essential oils from *C. schoenanthus*, *H. spicigera*, *L. camara* and *O. americanum* tested *in vitro* significantly reduced the contamination of sorghum and millet seeds by fungi while *C. nardus* significantly inhibited the mycelial growth of all the fungi tested (Zida et al. 2008).

Stimulation effect of essential oils on Striga hermonthica seed germination

The germination rates of *Striga hermonthica* seeds induced by 0.2% and 0.4% essential oils dilutions recorded with the three stimulation tests are presented in Table 3.

Table 3. Germination rates of *Striga hermonthica* seeds induced by essential oils dilutions.

Treatments	0.2% dilutions			0.4% dilutions		
	Test 1	Test 2	Test 3	Test 1	Test 2	Test 3
GR24	89.82 a	91.51 a	82.14 a	88.84 a	88.41 a	85.942 a
<i>C. citratus</i>	79.76 b	36.31 b	65.84 b	61.28 b	47.35 b	39.151 b
<i>O. gratissimum</i>	31.71 c	2.63 d	0.00 c	0.00 c	0.00 c	0.00 c
<i>H. suaveolens</i>	14.10 d	3.55 d	0.00 c	0.00 c	0.00 c	0.00 c
<i>O. americanum</i>	4.83 e	9.29 c	1.67 c	0.19 c	0.00 c	1.328 c
<i>C. schoenanthus</i>	1.13 e	0.29 d	1.49 c	0.00 c	0.00 c	0.00 c
Acetone	0.00 e	0.00 d	0.68 c	0.00 c	0.00 c	0.00 c
<i>C. nardus</i>	0.00 e	0.00 d	0.00 c	0.00 c	0.00 c	0.00 c
<i>E. camaldulensis</i>	0.00 e	0.00 d	0.00 c	0.00 c	0.00 c	0.00 c
<i>H. spicigera</i>	0.00 e	0.00 d	0.00 c	0.00 c	0.00 c	0.00 c
H ₂ O	0.00 e	0.00 d	0.00 c	0.00 c	0.00 c	0.00 c
<i>L. camara</i>	0.00 e	0.00 d	0.00 c	0.00 c	0.00 c	0.00 c
<i>L. multiflora</i>	0.00 e	0.00 d	0.00 c	0.00 c	0.00 c	1.807 c
Mean	17.03	11.04	11.68	11.56	10.44	9.864
cv%	55.0	32.9	62.7	63.5	37.0	75.2
F pr	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

Key: cv = coefficient of variation; F Pr = Fisher probability. Means followed by the same alphabet letter are not significantly different

The ANOVA showed a mean of 13.25% of germination with significant differences ($P < 0.0001$) between the 0.2% and 0.4% dilutions from plant essential oils and the controls, about the stimulation assay. The 0.2% dilutions of the essential oils from *Cymbopogon citratus*, *Ocimum gratissimum*, *Hyptis suaveolens*, *Ocimum americanum* and *Cymbopogon schoenanthus*, from the highest to the lowest, stimulated seed germination at rates from 60.64% to 0.97% (Figure 2). There was a significant difference between the germination rates of the extract from *C. citratus* (0.2%) and GR24 (positive control). On the one hand, the average of *C. citratus* extract was significantly higher than those of the other four extracts mentioned above. On the other hand, the means of the germination rates recorded with *H. suaveolens* and *O. americanum* oils were not statistically different. Only the 0.4% concentration from *C. citratus*, *O. gratissimum* and *O. americanum* extracts, stimulated the *Striga* seeds germination at the respective rates of 49.36%, 0.60% and 0.51%

(Figure 2). The average of the germination rates induced by the essential oils diluted to 0.4% was 10.62% with significant differences among them ($p < 0.0001$).

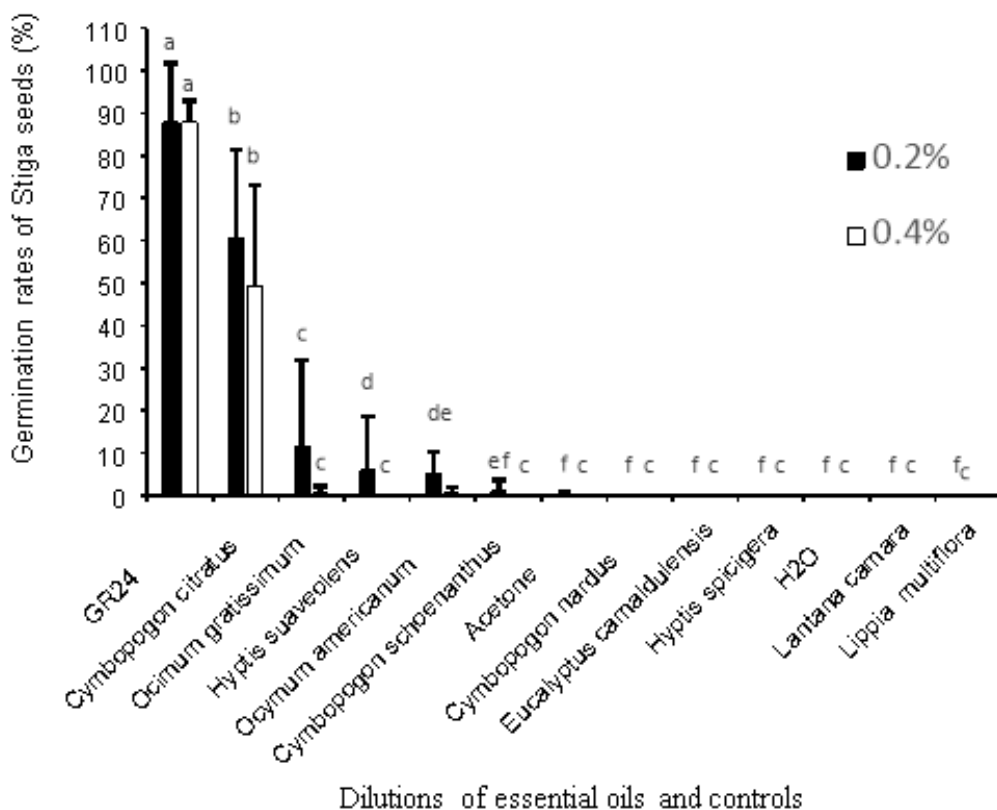


Figure 1. Stimulation effect of local plants essential oils on *Striga hermonthica* seeds germination. Means followed by the same alphabet letter are not significantly different

The germination rate of *C. citratus* essential oil was significantly higher than those of the other two plants. The oils from *C. nardus*, *E. camaldulensis*, *H. spicigera*, *L. camara* and *L. multiflora*, at a concentration of 0.4%, were statistically identical to the negative controls which did not stimulate the germination of *Striga hermonthica* seeds. Extracts from *C. citratus*, *O. gratissimum*, *H. suaveolens*, *O. americanum* and *C. schoenanthus* diluted to 10% significantly reduced the germination rate (by 0% for *C. citratus*); meanwhile, their 0.2% and 0.4% dilutions stimulated germination. These five essential oils would contain some molecules that stimulate seed germination only at low concentrations. Thus, the ten essential oils evaluated in the present study, on the basis of their effects, could be divided into three groups: 1) oils inhibiting germination of *Striga* seeds (*Cymbopogon nardus* and *Lippia multiflora*); 2) oils stimulating germination (*Cymbopogon citratus*, *Ocimum gratissimum*, *Hyptis suaveolens*, *Ocimum americanum* and *Cymbopogon schoenanthus*) and 3) oils without remarkable effect on the germination (*Lantana camara*, *Eucalyptus camaldulensis* and *Hyptis spicigera*).

Yonli et al. (2010) reported that the aqueous extracts from the leaves of *Eucalyptus camaldulensis* and bark of *Ceiba pentandra* (L.) Gaertn. concentrated at 1% significantly stimulated the germination of *Striga* seeds by 38.9% and 39.2% respectively. As a result of that, essential oil from *Eucalyptus* that inhibited germination of *Striga* seeds may not have the same chemical compositions as aqueous extracts. The highest stimulation rate in this study (60.64%), recorded with the lemongrass essence is much higher than that obtained by Babalola et al. (2007), 3.05%, with the bacterial isolate of *Pseudomonas* sp. *Striga* seeds germination stimulation by essential oils would be more reliable because the inhibition could be due to the fatty nature of the extracts and not linked to the action of particular molecule (s). In addition, stimulation would be more beneficial for an ecological weed management through suicidal germination. In fact, in the host plant absence, *Striga* germinated seedlings can't survive. This would allow a reduction or even elimination of the parasite seed bank and would result in the remediation of infested soils. Besides, seeds germination is the main cause of *Striga* seed bank depletion (Van Mourik, 2007).

The properties of the essential oils highlighted in this study increase the number of these substances virtues already studied by several authors. For example, essential oils from *C. citratus* and *L. multiflora* significantly reduced *in vitro* the infection level of the fungus *Phoma sorghina* on the seeds of five sorghum varieties compared to untreated seeds (Bonzi et al., 2013). The use of essential oils against *Striga* could therefore have another protective effect on sorghum through an action against pathogenic fungi. Oil of *C. citratus* has been an active insecticide against larvae and pupae of the housefly (*Musca domestica* L.) through contact toxicity assay (Kumar et al., 2013). Essential oils extracted from the leaves of this plant diluted to 8 ml l/1, from leaves of *Eucalyptus citrodora* and *Cinnamomum camphora* diluted to 12 ml l/1 completely inhibited seed germination and seedling length of the weed *Parthenium hysterophorus* L. (Paudel and Gupta, 2008). Results on essential oils bio-herbicidal effect could be applied in the natural conditions as well as the effectiveness of seed powder from *Azadirachta indica* A. Juss., fruit powder and fruit peel powder of *Parkia biglobosa* (Jacq.) R.Br. ex G. Don for *Striga* infestation reduction (Marley et al. 2004).

Conclusion

The results obtained from this study on *Striga hermonthica* seeds germination reveal further beneficial properties of essential oils on fighting against pests. Substances stimulating the germination of *Striga* seeds such as the essential oil from *Cymbopogon citratus* would have a double effect on the parasitic plant. It stimulates the germination in low concentrations and the high doses had an inhibitory effect. Metabolites produced by the local plant species in this study can be used for *S. hermonthica* management and improve the yield of cereal host crops of this parasitic

weed. This in vitro study is a first step in the research of bio-herbicides against *S. hermonthica*. Further experiments are needed to determine the exact concentration of each essential oil in the germination inhibition or stimulation in order to reach an effective application of results by farmers.

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Conflicts of Interest

No conflicts of interest have been declared.

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