Phytotoxic Effects of Multi-purpose Tree Species on Germination and Growth of Parthenium hysterophorus L.

Mulatu Wakjira, Gezahegn Berecha and Befekadu Bulti
Department of Horticulture and Plant Sciences, College of Agriculture and Veterinary Medicine, Jimma University, P.O. Box 307, Jimma, Ethiopia

Corresponding Author: Mulatu Wakjira, Department of Horticulture and Plant Sciences, College of Agriculture and Veterinary Medicine, Jimma University, P.O. Box 307, Jimma, Ethiopia Tel: +251 (0) 471110102 Fax: +251 (0) 471110934

ABSTRACT
The current study was conducted to test the inhibitory potential of aqueous extracts and dry shoot residues of four multipurpose tree species (Albizia gummifera, Azadirachta indica, Melia azedarach and Sesbania sesban) on seed germination and seedling growth of an invasive alien weed Parthenium hysterophorus under laboratory and greenhouse conditions. Leaf aqueous extracts and dry residues of all the multi-purpose tree species drastically inhibited germination and seedling growth of parthenium. Parthenium radicle was more inhibited than plumule in Petri dishes. Inhibitory effects increased with increasing aqueous extract concentration and residue amount. Soil surface-placed leaf residues exerted strong reduction on germination and growth of parthenium than soil-incorporated residues. In contrast to the higher inhibitory effects in Petri dishes, lower inhibitory effects were observed by the aqueous extracts of the multi-purpose tree species in pot-culture. Therefore, further investigations will be required under greenhouse and field conditions for pragmatic recommendation of species selection in the frame of multi-purpose tree species-mediated ecological management of parthenium weed.

Key words: Aqueous extracts, dry leaf residues, germination, growth, invasive alien weed, Parthenium hysterophorus, phytotoxicity

INTRODUCTION
Invasive Alien Species (IAS) are widely recognized among the greatest threats to biodiversity and productivity. They are becoming major challenges for countries like Ethiopia striving for food security. Parthenium (Parthenium hysterophorus L., Asteraceae) is one of the IAS registered in Ethiopia. It is an aggressive noxious weed, native to the Americas but now widely spread in Asia, Africa and Australia (Evans, 1997). Parthenium weed was introduced accidentally into Ethiopia in the mid-1970s and it was first reported from Ethiopia in 1988 at Dire-Dawa and Harerge, eastern Ethiopia (Seifu, 1990). Subsequently it spreads rapidly in all regions of the country, along roads and railways, through grazing areas and arable lands, adversely threatening crop production, animal husbandry and biodiversity (Tefera, 2002). The successful spread of parthenium in so many parts of the world including Ethiopia has mainly been attributed to its allelopathic properties, due to which it outcompetes crops and pasture species (Singh et al., 2003, 2005).

Parthenium is a noxious weed because of its allelopathic effect (Wakjira et al., 2005; Kohli et al., 2006; Wakjira, 2009), its strong competitiveness with both crops and pasture for soil
moisture and nutrients (Singh et al., 2003, 2005) and the hazard it poses to humans and animals (Wiesner et al., 2007) and biodiversity (Tefera, 2002). Currently, parthenium is considered as the most serious weed in both arable and grazing lands as it caused severe crop losses (Tamado and Milberg, 2000). In Ethiopia, grain yield losses from 40 to 97% were recorded when parthenium is left uncontrolled throughout the growing season of sorghum (Tamado et al., 2002b).

Currently, parthenium is one of the noxious weeds threatening crop production in Ethiopia. The rapid spread of parthenium in Ethiopia would be a bigger risk to the expansion and sustainable production of many crops (Tefera, 2002; Wakjira et al., 2005; Wakjira, 2009; Wakjira et al., 2009). Control of parthenium is therefore crucial to boost productivity of crops and maintain the integrity of both agro-biodiversity and ecosystem complex of the country. Various control methods attempted worldwide in controlling parthenium have their own pros and cons. Hand-weeding mostly used by small-scale farmers is more difficult due to the allergic effects of parthenium on human body (Tefera, 2002; Wiesner et al., 2007). Mechanical control on the other hand is rather costly in terms of machinery, labour and time requirement. Furthermore, resource poor farmers of Ethiopia cannot afford the purchase of herbicides and the use of herbicides is unsafe in terms of health and environmental considerations and not allowed in organic farming systems. Therefore, other options must be sought for ecological management of parthenium weed in Ethiopia. One such option is the use of allelopathic plants that suppress the germination and growth of parthenium.

The use of phytotoxic plants as an ecological weed management tool will play an important role in future weed control and crop productivity. The phytotoxic/allelopathic compounds can be used as natural herbicides and they are less disruptive of the global ecosystem than are synthetic agrochemicals (Khalid et al., 2002). Recent investigations indicated the potential use of allelopathic plants in parthenium weed management (Anjum et al., 2005; Javaid et al., 2005; Shafique et al., 2005; Javaid and Anjum, 2006).

In Ethiopia, most of the research work regarding parthenium concentrated on its distribution (Tamado and Milberg, 2000), germination ecology (Tamado et al., 2002a), impacts on sorghum (Tamado et al., 2002b), allelopathic effects (Tefera, 2002; Wakjira et al., 2005, 2009), biological control using fungi (Tessema, 2002) and management by utilization (Wakjira et al., 2009). Little attempts have been made to investigate and exploit the ecologically-based management methods like the use of allelopathic plant species including the multi-purpose tree species in Ethiopia. Therefore, the objective of the present study was to test the inhibitory potential of aqueous extracts and dry shoot residues of four multipurpose tree species (Albizia gummifera, Azadirachta indica, Melia azedarach and Sesbania sesban) on seed germination and seedling growth of an invasive alien weed parthenium under laboratory and greenhouse conditions.

MATERIALS AND METHODS
General experimental set-up: Laboratory and greenhouse experiments were executed at Jimma University, Jimma, Ethiopia from January 2007 to December 2009. The study site is located at about 7°, 41' N latitude and 38°, 50' E longitude at an altitude of 1710 m above sea level. The mean minimum and maximum temperatures in the laboratory and greenhouse were 20 and 23°C and 12 and 25°C during the experimental period. The relative humidity ranged from 31 to 90%. The soil was clay-loam in texture with an organic matter content of 3-5% and a pH of 5.6-6.5.

Fresh leaf samples of tree species commonly grown in Ethiopia (Albizia gummifera, Azadirachta indica, Melia azedarach and Sesbania sesban) were collected at their vegetative stage. Except A. indica, leaves of all other tree species were collected from Jimma area. The A. indica leaves were collected from Ziway, Ethiopia. The leaves were air-dried and ground in a mill of 2 mm sieve size.
Mature parthenium seeds were collected from naturally growing parthenium plants around the Jimma University campus. The seeds were air-dried and threshed by hand and their viability was selected. Seeds were surface-sterilized by shaking them for five minutes in 1% sodium hypochlorite (NaOCl) solution and washed with distilled water for three minutes immediately before use. For the laboratory experiment, 9 cm diameter Petri dishes were used. In the case of lathhouse experiments, 1 L pots were used. Sieved agricultural soil obtained from Eladale Research Farm of Jimma University was used for the pot experiments.

Experiments 1 and 2 were carried out to evaluate the inhibitory effects of aqueous extracts of the four multi-purpose tree species on parthenium germination and growth under laboratory and greenhouse conditions. Three, 6 and 9 g of each of the ground material of the test species were soaked in 100 mL distilled water (w/v) for 24 h at room temperature (21 to 22°C). The extracts were filtered through three layers of cheesecloth and designated as 3, 6 and 9% leaf aqueous extracts, respectively and stored in dark conical flasks until use.

Experiments 3 and 4 were carried out with dry leaf residues incorporated into the soil (Experiment 3) and placed on the soil surface (Experiment 4) of the four multi-purpose tree species under greenhouse conditions. The aims of these experiments were to investigate the variations in inhibitory potential of the ground leaf materials from the four tree species when incorporated into the soil and placed on the soil surface on the emergence and growth of parthenium.

**Experiment 1: Effects of leaf aqueous extracts in the laboratory:** Fifty sterilized parthenium seeds were placed in Petri dishes lined with double layers of filter paper, a thick filter paper (T415) was placed on the bottom and a thin filter paper (Whatman No. 1) on the top. The filter paper was moistened with 10 mL of the 3, 6 or 9% leaf aqueous extracts and distilled water (control). The treatments were replicated four times in a Completely Randomized Design (CRD). The Petri dishes and pots were rotated every third day to avoid unforeseen variations within the laboratory and greenhouse, respectively. The experiment was conducted twice.

**Experiment 2: Effects of leaf aqueous extracts in pot-culture:** Fifty sterilized parthenium seeds were sown per pot and 10 mL of the 3, 6 or 9% leaf aqueous extracts were added per pot. The control treatments received 10 mL distilled water per pot. The treatments were laid down in a Randomized Complete Block Design (RCBD) with four replications. The experiment was executed twice.

**Experiment 3: Effects of soil-incorporated dry leaf residues:** Ground leaves were thoroughly mixed in one litre pots filled with 1 kg sieved agricultural soil. A pot filled with same soil without dry leaf residues was used as control. Therefore, the treatments include 3, 6 and 9 g dry leaf residues of the four multi-purpose tree species and control (without dry leaf residue). The treatments were laid down in RCBD with three replications. The experiments were executed twice.

**Experiment 4: Effects of soil surface-placed dry leaf residues:** Three, 6 and 9 g of the dry leaf residues of the four tree species were placed on the surface of pots filled with sieved agricultural soil. A pot filled with the same volume of sieved agricultural soil without dry leaf residues was used as control. The treatments were laid down in RCBD with four replications. The experiments were executed twice.
Measurements: In the laboratory experiment with aqueous extracts, germinated seeds were counted daily from 10th to 14th days after sowing (DAS). Emerged seedlings were counted daily from 8th day to 17th DAS in the case of greenhouse experiments with aqueous extracts. In the greenhouse experiment with dry leaf residues, emerged seedlings were counted from 7th to 13th DAS. Germination/emergence percentages were determined from the number of germinated seeds or emerged seedlings. Germination/emergence rate (G/ER) is the number of seeds germinated or seedlings emerged from the seeds sown per Petri dish or per pot and was calculated by Maguire (1962) as:

\[ G/ER \text{ (seeds day}^{-1}) = \frac{\text{N.o. of normal seedlings}}{\text{Days of first count}} + \ldots + \frac{\text{N.o. of normal seedlings}}{\text{Days of final count}} \]

Radicle and plumule lengths (mm) were measured on five randomly selected seedlings per Petri dish in laboratory experiments. In greenhouse experiments seedlings were thinned to ten per pot at 20 DAS and five seedlings were taken at random from five different positions from the middle of the pot four weeks after sowing. The five sample seedlings were separated into root and shoot parts and root and shoot lengths (mm) were measured for both experiments with aqueous extracts and dry leaf residues. In the case of dry leaf residues, the numbers of true leaves per plant were counted and leaf area per plant (cm²) was determined by scanning the leaves using a portable leaf area meter (ADC AM300). Root and shoot fresh weights were determined per plant using a sensitive balance. Root and shoot dry weight per plants was determined after drying in an oven at 70°C for 24 h. The inhibition (-) or stimulation (+) percentage compared to control was calculated using Chung et al. (2001):

\[
\text{Inhibition (-) or stimulation (+) percentage (%) =} \left\{ \frac{\text{Treatment} - \text{Control}}{\text{Control}} \right\} \times 100
\]

Data analysis: For each of the response variables, the validity of model assumptions was verified by examining the residuals. The average data were analyzed using Analysis of Variance (ANOVA). Two phases of statistical analysis were employed. First, whether the interaction and/or the main effects of the fixed factors of interest were significant was analyzed. Second, treatment combinations having significant interactions and main effects were further compared. Multiple comparisons of means were performed using Fisher’s protected Least Significant Difference (LSD) test. All statistical procedures were performed using GenStat statistical package, GenStat for Windows 12th Edition (VSN International Ltd., 2009).

RESULTS

Experiment 1 Effects of leaf aqueous extracts in the laboratory: In Petri dishes, leaf aqueous extracts of the four multi-purpose tree species reduced germination percentage, germination rate, radicle length and plumule length of parthenium (average reductions of 59, 60, 63 and 55%, respectively) (Fig. 1). Leaf aqueous extracts of A. indica, M. azedarach, S. sesban and A. gummifera reduced germination percentage (by 67, 59, 56 and 54%, respectively), germination rate (by 65, 63, 62 and 49%, respectively), radicle length (by 67, 64, 71 and 50%, respectively) and plumule length (by 60, 65, 55 and 41%, respectively). Though the multi-purpose tree species

152
Fig. 1: Main effects of leaf aqueous extracts of four tree species (CON: Control, AG: Albizia gummifera, AI: Azadirachta indica, MA: Melia azedarach and SS: Seshania sesban) on germination percentage (a), germination rate (b), radicle length (c) and plume length (d) in Petri dishes. Means followed by different letters differ significantly (p<5%) as established by Fisher's protected LSD-test (LSD = 5.37, 0.49, 4.401 and 1.56, respectively). Vertical bars represent mean values ±SE

Table 1: Main effects of leaf aqueous extract concentrations of four tree species on Germination Percentage (GP), Germination Rate (GR), Radicle Length (RL) and Plume Length (PL) in Petri dishes

<table>
<thead>
<tr>
<th>Aqueous extract concentration (%)</th>
<th>GP (%)</th>
<th>GR (seeds day⁻¹)</th>
<th>RL (mm)</th>
<th>PL (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>60.0a</td>
<td>4.6a</td>
<td>20.4a</td>
<td>8.4a</td>
</tr>
<tr>
<td>3</td>
<td>23.8 (-60)b</td>
<td>1.7 (-62)b</td>
<td>6.4 (-59)b</td>
<td>4.7 (-44)b</td>
</tr>
<tr>
<td>6</td>
<td>10.5 (-83)c</td>
<td>0.8 (-82)c</td>
<td>3.6 (-83)c</td>
<td>1.9 (-77)c</td>
</tr>
<tr>
<td>9</td>
<td>4.3 (-90)d</td>
<td>0.2 (-90)d</td>
<td>0.03 (-100)c</td>
<td>0.1 (-90)d</td>
</tr>
<tr>
<td>LSD</td>
<td>5.370</td>
<td>0.4911</td>
<td>4.401</td>
<td>1.556</td>
</tr>
</tbody>
</table>

1 Values in parenthesis indicate percentage reduction (-) or stimulation (+) compared to control. Means followed by different letters within one column differ significantly (p<5%) as established by Fisher's protected LSD-test.

provided strong reduction on germination and growth of parthenium compared to control, the differences between the tree species were not large.

Compared to control, leaf aqueous extract concentrations from the four multi-purpose tree species drastically reduced germination percentage (-79%) and rate (-80%) and radicle (-84%) and plume lengths (-74%) on average basis (Table 1). For all the response variables, there was an increase in inhibitory effect with an increase in aqueous extract concentration. Consequently, at the highest concentration (9%), nearly complete failure of germination and growth of parthenium was observed (97%).

The amount of reduction on germination and growth of parthenium provided by the main effects of leaf aqueous extract concentration (-80%) was higher than that provided by the main effects of multi-purpose tree species (-60%) (Fig. 1 and Table 2). In both cases, comparable reductions were observed for emergence and growth whereas radicles were reduced more than plumeles.
Table 2: Main effects of leaf aqueous extracts of four tree species on emergence percentage (EP), emergence rate (ER), root length (RL) and shoot length (SL) in pot-culture

<table>
<thead>
<tr>
<th>Tree species</th>
<th>EP (%)</th>
<th>ER (seeds day⁻¹)</th>
<th>RL (mm)</th>
<th>SL (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>46.1a</td>
<td>30.8a</td>
<td>34.8a</td>
<td>6.11a</td>
</tr>
<tr>
<td>Albizia gummifera</td>
<td>41.1 (-11)bc</td>
<td>25.0 (-17)b</td>
<td>27.7 (-20)b</td>
<td>4.97 (-19)b</td>
</tr>
<tr>
<td>Azadirachta indica</td>
<td>41.9 (-9)b</td>
<td>24.5 (-21)b</td>
<td>28.3 (-19)b</td>
<td>4.78 (-22)bc</td>
</tr>
<tr>
<td>Melia azedarach</td>
<td>40.4 (-13)bc</td>
<td>23.7 (-20)b</td>
<td>25.7 (-20)b</td>
<td>4.34 (-20)bc</td>
</tr>
<tr>
<td>Sesbania sesban</td>
<td>38.4 (-17)c</td>
<td>24.4 (-21)b</td>
<td>26.0 (-20)b</td>
<td>4.16 (-23)c</td>
</tr>
<tr>
<td>LSD</td>
<td>3.133</td>
<td>2.619</td>
<td>4.174</td>
<td>0.778</td>
</tr>
</tbody>
</table>

Values in parenthesis indicate percentage reduction (-) or stimulation (+) compared to control. Means followed by different letters within one column differ significantly (p< 0.05) as established by Fisher's protected LSD-test.

Table 3: Main effects of leaf aqueous extract concentrations of four tree species on Emergence Percentage (EP), Emergence Rate (ER), Root Length (RL) and Shoot Length (SL) in pot-culture

<table>
<thead>
<tr>
<th>Aqueous extract concentration (%)</th>
<th>EP (%)</th>
<th>ER (seeds day⁻¹)</th>
<th>RL (mm)</th>
<th>SL (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>46.1a</td>
<td>30.8a</td>
<td>34.8a</td>
<td>6.11a</td>
</tr>
<tr>
<td>3</td>
<td>41.4 (-10)b</td>
<td>25.0 (-19)b</td>
<td>29.0 (-17)b</td>
<td>5.19 (-15)b</td>
</tr>
<tr>
<td>6</td>
<td>30.2 (-15)b</td>
<td>22.0 (-25)b</td>
<td>23.9 (-32)c</td>
<td>3.90 (-36)c</td>
</tr>
<tr>
<td>9</td>
<td>34.9 (-24)c</td>
<td>19.4 (-37)c</td>
<td>20.1 (-42)c</td>
<td>3.04 (-50)d</td>
</tr>
<tr>
<td>LSD</td>
<td>3.133</td>
<td>2.619</td>
<td>4.174</td>
<td>0.778</td>
</tr>
</tbody>
</table>

Values in parenthesis indicate percentage reduction (-) or stimulation (+) compared to control. Means followed by different letters within one column differ significantly (p<0.05) as established by Fisher's protected LSD-test.

Experiment 2 Effects of leaf aqueous extracts in pot-culture: In contrast to the results obtained under laboratory conditions in Petri dishes, lower inhibitions were observed on emergence and growth of parthenium under greenhouse conditions in pot-culture. Evidently, lower effects were observed by the main effects of the multi-purpose tree species and aqueous extract concentrations under greenhouse conditions (Table 2, 3). Aqueous extracts from the four multi-purpose tree species reduced emergence percentage (-12%), emergence rate (-20%), root length (-23%) and shoot length (-25%) of parthenium compared to control on average (Table 2).

Though the multi-purpose tree species reduced germination and growth of parthenium compared to control, the differences between the inhibition effects of the tree species were very small. The amount of reduction provided by the multi-purpose tree species (20% on average) on emergence and growth of parthenium was relatively lower compared to the reduction provided by the aqueous extract concentrations (27% on average) (Table 2 and 3). Compared to control, aqueous extract concentrations from the four multi-purpose tree species reduced parthenium emergence percentage (-17%), emergence rate (-27%), radicle length (-30%) and plumule length (-34%) (Table 3). Similar to the results under laboratory conditions, for all the variables, there was an increase in inhibitory effect with an increase in aqueous extract concentration.

Experiment 3 Effects of soil-incorporated dry leaf residues: Soil-incorporated dry leaf residues of the four multi-purpose tree species provided a significant reduction on emergence percentage (-23%), emergence rate (-28%), root length (-10%), leaf area (-15%) and root fresh weight (-25%) of parthenium (Table 4). Shoot fresh weight was inhibited by A. indica and S. sesban (by 44 and 4%, respectively) while M. azedarach and A. gummifera stimulated shoot fresh weight by 46 and 16%, respectively (Table 4). In relation to inhibition of emergence, the four tree species ranked in the order of S. sesban > A. indica > M. azedarach > A. gummifera. However, the effects on growth of parthenium were inconsistent.

154

Table 4: Main effects of soil-incorporated dry leaf residues of four tree species on Emergence Percentage (EP) and rate (ER), Leaf Area (LA) and Root Fresh Weight (RFW) and Shoot Fresh Weight (SFW) in pot-culture

<table>
<thead>
<tr>
<th>Tree species</th>
<th>EP (%)</th>
<th>ER (seeds day$^{-1}$)</th>
<th>RL (mm)</th>
<th>LA (cm$^2$ plant$^{-1}$)</th>
<th>RFW (g plant$^{-1}$)</th>
<th>SFW (g plant$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>74.7a</td>
<td>12.4a</td>
<td>30.5a</td>
<td>235.1a</td>
<td>0.7ab</td>
<td>0.31ab</td>
</tr>
<tr>
<td>Albizia gummifera</td>
<td>59.0 (-21)b</td>
<td>10.3 (-17)b</td>
<td>28.7 (-6)ab</td>
<td>192.9 (-18)b</td>
<td>0.5 (-27)bc</td>
<td>0.35 (+16)a</td>
</tr>
<tr>
<td>Azadirachta indica</td>
<td>54.7 (-27)bc</td>
<td>8.2 (-34)d</td>
<td>36.6 (-13)c</td>
<td>144.7 (-30)c</td>
<td>0.4 (-41)c</td>
<td>0.17 (-45)b</td>
</tr>
<tr>
<td>Melia azedarach</td>
<td>58.7 (-21)bc</td>
<td>9.2 (-25)bc</td>
<td>26.7 (-15)bc</td>
<td>230.6 (-23)ab</td>
<td>0.8 (-10)a</td>
<td>0.45 (+46)a</td>
</tr>
<tr>
<td>Sesbania sebim</td>
<td>54.0 (-26)c</td>
<td>7.6 (-36)d</td>
<td>27.9 (-9)bc</td>
<td>227.8 (-3)ab</td>
<td>0.4 (-42)c</td>
<td>0.29 (-43ab)</td>
</tr>
<tr>
<td>LSD</td>
<td>4.382</td>
<td>1.395</td>
<td>2.039</td>
<td>41.95</td>
<td>0.2328</td>
<td>0.1576</td>
</tr>
</tbody>
</table>

$^1$Values in parenthesis indicate percentage reduction (-) or stimulation (+) compared to control. Means followed by different letters within one column differ significantly (p<0.05) as established by Fisher's protected LSD-test.

Table 5: Main effects of soil-incorporated dry leaf residues of four tree species on Emergence Percentage (EP) and rate (ER), Root Length (RL), Leaf Area (LA) and Root Fresh Weight (RFW) in pot-culture

<table>
<thead>
<tr>
<th>Residue amount (g)</th>
<th>EP (%)</th>
<th>ER (seeds day$^{-1}$)</th>
<th>RL (mm)</th>
<th>LA (cm$^2$ plant$^{-1}$)</th>
<th>RFW (g plant$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>74.7a</td>
<td>12.4a</td>
<td>30.5a</td>
<td>235.1a</td>
<td>0.71a</td>
</tr>
<tr>
<td>3</td>
<td>57.3 (-23)b</td>
<td>9.1 (-25)b</td>
<td>28.9 (-5)ab</td>
<td>222.2 (-6)ab</td>
<td>0.61 (-13)ab</td>
</tr>
<tr>
<td>6</td>
<td>52.0 (-30)c</td>
<td>7.9 (-37)b</td>
<td>25.0 (-15)b</td>
<td>187.8 (-30)bc</td>
<td>0.4 (-35)bc</td>
</tr>
<tr>
<td>9</td>
<td>42.3 (-43)d</td>
<td>6.1 (-51)c</td>
<td>24.5 (-20)b</td>
<td>150.9 (-36)c</td>
<td>0.35 (-51)c</td>
</tr>
<tr>
<td>LSD</td>
<td>4.382</td>
<td>1.395</td>
<td>2.039</td>
<td>41.95</td>
<td>0.2328</td>
</tr>
</tbody>
</table>

$^1$Values in parenthesis indicate percentage reduction (-) or stimulation (+) compared to control. Means followed by different letters within one column differ significantly (p<0.05) as established by Fisher's protected LSD-test.

Soil-incorporated residue amounts provided greater reduction compared to the main effects of the multi-purpose tree species (average reductions of 27 and 20%, respectively) (Table 4, 5). Compared to control, the amounts of soil-incorporated dry leaf residues from the four multi-purpose tree species reduced emergence percentage (-32%), emergence rate (-38%), root length (-13%), leaf area (-21%) and root fresh weight (-33%) on an average (Table 5). For all the response variables, there was an increase in inhibitory effect on parthenium emergence and growth with an increase in the amount of soil-incorporated dry leaf residue. Evidently, the highest residue amount (9 g) provided reduction on emergence percentage, emergence rate, root length, leaf area and root fresh weights of 43, 51, 20, 36 and 51%, respectively.

**Experiment 4 Effects of soil surface-placed dry leaf residues**: The analysis of variance for germination percentage and rate confirmed that there were significant two-way interactions between multi-purpose tree species and amounts of dry leaf residues placed on the soil surface (p<0.001) (Fig. 2). This clearly indicated that multiplicative effects occur between tree species and amounts of soil-surface placed dry leaf residues in inhibiting emergence of parthenium, both percentage and rate.

Soil surface-placed dry leaf residues of all the multi-purpose tree species provided strong reductions on emergence percentage (-43%) and emergence rate (-46%) compared to control (Fig. 2). Application of A. gummifera, A. indica, M. azedarach and S. sebim on the soil surface reduced emergence percentage by 33, 40, 80 and 74%, respectively (Fig. 1a). Emergence rate was reduced by surface application of dry leaf residues of A. gummifera, A. indica, M. azedarach and S. sebim by 34, 46, 86 and 80%, respectively (Fig. 2b). Except for A. gummifera on emergence rate, there was an increase in inhibitory effect on the emergence of parthenium with an increase in the amount of leaf residue placed on the soil surface. For A. gummifera, there were no significant
Fig. 2: Effects of soil surface-placed dry leaf residues of four tree species (CON: Control, AG: Albizia gummierea, AI: Azadirachta indica, MA: Melia azedarach and SS: Sesbania sesban) at different amounts: 0 g (black), 3 g (grey), 6 g (stripes) and 9 g (white) on parthenium emergence (%) and rate (seeds per day) in pot-culture. Means followed by different letters differ significantly (p<5%) as established by Fisher’s protected LSD-test (LSD = 13.09 and 1.81, respectively). Vertical bars represent mean values ±SE

Table 6: Main effects of soil surface-placed dry leaf residues of four tree species on Root Length (RL), Leaf Number (LN), Root Fresh Weight (RFW) and Root Dry Weight (RDW) in pot-culture

<table>
<thead>
<tr>
<th>Tree species</th>
<th>RL (mm)</th>
<th>LN</th>
<th>RFW (g plant⁻¹)</th>
<th>RDW (g plant⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>30.7±1</td>
<td>8.6±a</td>
<td>0.70±a</td>
<td>0.12±2</td>
</tr>
<tr>
<td><em>Albizia gummifera</em></td>
<td>49.3±(4)</td>
<td>8.3±(3)</td>
<td>0.45±(3)</td>
<td>0.05±(19)</td>
</tr>
<tr>
<td><em>Azadirachta indica</em></td>
<td>47.4±(4)</td>
<td>8.2±(4)</td>
<td>0.67±(4)</td>
<td>0.13±(12)</td>
</tr>
<tr>
<td><em>Melia azedarach</em></td>
<td>73.0±(5)</td>
<td>7.8±(9)</td>
<td>0.36±(6)</td>
<td>0.07±(41)</td>
</tr>
<tr>
<td><em>Sesbania sesban</em></td>
<td>58.6±(19)</td>
<td>6.4±(26)</td>
<td>0.36±(50)</td>
<td>0.05±(40)</td>
</tr>
<tr>
<td>LSD</td>
<td>3.9±4</td>
<td>0.8±8</td>
<td>0.2±12</td>
<td>0.04±772</td>
</tr>
</tbody>
</table>

Values in parenthesis indicate percentage reduction (-) or stimulation (+) compared to control. Means followed by different letters within one column differ significantly (p<5%) as established by Fisher’s protected LSD-test.

There were no significant differences between all amounts of soil surface-placed dry leaf residues in reducing parthenium emergence rate (Fig. 2b). The amounts of soil surface-placed dry leaf residues reduced emergence percentage by 57% whereas emergence rate by 62%.

Soil surface-placed dry leaf residues from the four multi-purpose tree species reduced root length (-15%), leaf number (-10%) and root fresh weight (-34%) of parthenium on average compared to control (Table 6). Root dry weight was reduced by M. azedarach, *S. sesban* and *A. gummifera* (by 41, 40 and 19%, respectively) whereas stimulated by A. indica (by 12%). The amounts of surface-placed dry leaf residues exerted more reduction (-39%) on emergence and growth of parthenium compared to the main effects of the multi-purpose tree species (-29%). Compared to control, the amounts of soil surface-placed dry leaf residues from the four multi-purpose tree species provided reductions on root length (-17%), leaf number (-14%), leaf area (-31%), root fresh weight (-46%) and shoot dry weight (-42%) (Table 7). For all the response variables, there was an increase in inhibitory effect on emergence and growth of parthenium with an increase in the amount of dry leaf residue placed on the soil surface.

**DISCUSSION**

Plant invasions are threatening biodiversity and productivity worldwide and are becoming the major challenges for developing countries like Ethiopia striving to achieve food security.
Table 7: Main effects of soil surface-placed dry leaf residue amounts of four tree species on Root Length (RL), Leaf Area (LA), Leaf Number (LN), Root Fresh Weight (RFW) and Shoot Dry Weight (SDW) in pot-culture

<table>
<thead>
<tr>
<th>Residue amount (g)</th>
<th>RL (mm)</th>
<th>LA (cm² plant⁻¹)</th>
<th>LN</th>
<th>RFW (g plant⁻¹)</th>
<th>SDW (g plant⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>30.7a¹</td>
<td>226.7a</td>
<td>8.6a</td>
<td>0.69a</td>
<td>0.076a</td>
</tr>
<tr>
<td>3</td>
<td>30.3 (1)a</td>
<td>198.7 (-12a)b</td>
<td>8.1 (-6a)b</td>
<td>0.46 (-4)b</td>
<td>0.051 (-4)b</td>
</tr>
<tr>
<td>6</td>
<td>24.8 (-19)b</td>
<td>151.9 (-39)bc</td>
<td>7.3 (-15)bc</td>
<td>0.31 (-7)b</td>
<td>0.046 (-4)b</td>
</tr>
<tr>
<td>9</td>
<td>21.5 (-30)b</td>
<td>121.5 (-46)c</td>
<td>6.7 (-22)c</td>
<td>0.36 (-60)b</td>
<td>0.036 (-52)b</td>
</tr>
<tr>
<td>LSD</td>
<td>3.914</td>
<td>55.45</td>
<td>0.969</td>
<td>0.2112</td>
<td>0.02251</td>
</tr>
</tbody>
</table>

¹Values in parenthesis indicate percentage reduction (-) or stimulation (+) compared to control. Means followed by different letters within one column differ significantly (p<0.05) as established by Fisher’s protected LSD-test.

Parthenium is one of the IAS spreading at an alarming rate in Ethiopia. Due to increased societal concerns about the environmental and human health impacts together with inability of the small-scale farmers to buy herbicides, efforts are being made worldwide to reduce the heavy reliance on synthetic herbicides through the use of alternative methods. The use of weed suppressive multi-purpose tree species in agro-ecosystem is among these alternatives as it has the potential to retard germination and growth of weeds. Inhibition of germination could minimize the number of weed plants that compete with the crop species and that grow and add seed to the seed bank. Furthermore, weed plants that germinated at slower rates are often smaller and less competitive. This could give an opportunity for the crop species to outcompete the weeds (Jefferson and Pennacchio, 2003). The aim of the present study was to evaluate different multi-purpose tree species for their inhibitory potential in suppressing germination and growth of an invasive alien weed parthenium. To achieve this objective, four sets of experiments were executed using leaf aqueous extracts and dry residues of four multi-purpose tree species under the laboratory conditions in Petri dishes and under the greenhouse conditions in pot-culture.

**Effects of multi-purpose tree species:** There were variations among the multi-purpose tree species in their inhibitory potential in the present study. This finding is in line with previous findings that indicated different plants have different inhibitory potential (Stoll et al., 2006; Adler and Chase, 2007; Price et al., 2008). The plausible mechanism for inhibition of germination and growth could be due to reduced rate of cell division and elongation attributed to allelochemicals (Javaid and Anjum, 2006) or could be physical interference by surface-placed residues with the upward movement of the emerging seedlings and the downward penetration of light to germinating seeds (Teasdale and Mohler, 2000).

Compared to control, leaf aqueous extracts of all of the multi-purpose tree species greatly reduced germination percentage and rate and radicle and plumule lengths of parthenium under laboratory conditions in Petri dishes (Fig. 1). However, variations were evident among the tree species in the extent of reduction they provided. Regarding reduction of germination, the four tree species ranked in the order of A. indica > M. azedarach > S. sesban > A. gummifera whereas in reduction of growth they were ranked in the order of M. azedarach > A. indica > S. sesban > A. gummifera. In the case of aqueous extracts in pot-culture, S. sesban and M. azedarach provided greater reduction of both emergence and growth compared to A. indica and A. gummifera (Table 2). In relation to inhibition of emergence provided by soil-incorporated leaf residues, the four tree species ranked in the order of S. sesban > A. indica > M. azedarach > A. gummifera. The effects on growth of parthenium were inconsistent though A. indica and S. sesban still played a significant role in reduction of growth. However, soil-incorporated residues from M. azedarach and
A. gummifera strongly stimulated shoot fresh weight (Table 4). In the case of soil surface-placed residues, M. azedarach and S. sesban provided more reduction on both germination and growth of parthenium than A. indica and A. gummifera (Fig. 2 and Table 6). Furthermore, A. indica slightly stimulated root dry weight (Table 6).

This finding is supported by previous reports that demonstrated about the allelopathic effects of A. indica (Xuan et al., 2004; Shafique et al., 2005; Al-Charchafchi et al., 2007; Rahnavard et al., 2008), M. azedarach (Hong et al., 2003, 2004) and S. sesban (Mubarak et al., 2009) on different plants. For A. gummifera, findings that indicate about its allelopathic effects are nonexistent. Hence, it was for the first time that we have systematically evaluated and demonstrated the allelopathic inhibitory effects of A. gummifera on germination and growth of parthenium.

Effects of quantity: aqueous extract concentrations and dry leaf residue amounts: Both under laboratory conditions in Petri dishes and under greenhouse conditions in pot-culture, there was an increase in inhibitory effect on both germination and growth of parthenium with an increase in extract concentration from the four multi-purpose tree species. This was in agreement with several previous authors for many different plants including A. indica (Teferra, 2002; Xuan et al., 2004; Wakjira et al., 2005; Al-Charchafchi et al., 2007; Jafari et al., 2007; Rahnavard et al., 2008; Mutlu and Atici, 2009; Wakjira, 2009). Similar to the aqueous extract concentrations, as the amount of dry shoot residue incorporated and placed on the soil surface increased, inhibitory effect also increased though the variation between the amounts was small. This was in agreement with previous reports for many plants including A. indica and M. azedarach (Hong et al., 2004; Xuan et al., 2004; Singh et al., 2005; Batlang and Shushu, 2007; Wakjira, 2009).

Effects of residue placement method: The amount and quality of the residue material, soil biological, chemical and physical factors, environmental conditions, extent of tissue disruption and methods of residue placement influence the release of allelochemicals and the consequent residue-mediated inhibitory or stimulatory effects (Mohler, 1996; Liebman and Mohler, 2001; Morra and Kirkegaard, 2002; Kruidhof et al., 2009). In the present study, dry and ground leaves were used both in the incorporation and surface-placed experiments that provided uniform tissue disruption. Compared to soil-incorporated dry leaf residues, the soil surface-placed dry leaf residues drastically inhibited germination and growth of parthenium (Table 4-7, Fig. 2). Our current finding was in agreement with previous findings for other plant species (Ismail and Chong, 2002; Wakjira, 2009). The inhibitory effect as a result of soil incorporation might be explained by the fact that the more inhibitory chemicals were available that caused inhibitory effect (Chon et al., 2005). The possible explanation for the reduction provided by the surface placed residues could be physical interference with the upward movement of the emerging seedlings and the downward penetration of light to germinating seeds (Teasdale and Mohler, 2000) or due to the release of inhibitory allelochemicals.

Effects on radicle and plumule growth: In the laboratory experiment we have demonstrated that radicle growth is more reduced than plumule growth (Fig. 3). This is in line with several other reports on many plants including (Teferra, 2002; Wakjira et al., 2005; Al-Charchafchi et al., 2007; Rahnavard et al., 2008; Wakjira, 2009). The plausible reason could be the radicles are in direct
contact with the leaf aqueous extracts and hence with the inhibitory chemicals (Tefera, 2002; Rietjens and Alink, 2003; Wakjira et al., 2005; Al-Charchafchi et al., 2007; Wakjira, 2009).

Comparison of laboratory and greenhouse experiment results: In the present study, clear, greater and fast inhibition effect by the multi-purpose tree species was observed in Petri dishes compared to pot-culture. Experiments with aqueous extracts in Petri dishes are easy and fast but do not prove the existence of allelopathy under field conditions. Pot soil with residue material is less convenient and more laborious, but presumed to be closer to reality. From the present study the multi-purpose tree species provided a strong reduction on germination and growth of parthenium under laboratory conditions in Petri dishes. This however does not mean that they are also maintained under greenhouse and field conditions. Laboratory screening however helps to select promising species for greenhouse and field evaluation and field studies are pertinent to provide strong evidence for existence of allelopathy-mediated inhibitory potentials (Inderjit and Weston, 2000; Wu et al., 2001; Morgan and Overholt, 2005). In contrast to the results obtained under laboratory conditions Petri dishes, lower inhibitory effects of aqueous extracts of the four multi-purpose tree species were observed under greenhouse conditions in pot-culture. This is congruent with findings of previous workers (Morgan and Overholt, 2005). This could be explained by the complex reactions taking place in the soil. Allelochemicals are subject to various biotic and abiotic processes that reduce their persistence, concentrations, availability and biological activities after they are added into the soil (Huang et al., 1977; Wang et al., 1986; Makino et al., 1996; Blum et al., 1998; Okumura et al., 1999).

Practical considerations and prospects: The rapid spread of parthenium in Ethiopia would be a bigger risk to the expansion and sustainable production of many crops (Tamado et al., 2002b; Tefera, 2002; Tessema, 2002; Wakjira et al., 2005, 2009; Wakjira, 2009). Control of parthenium is therefore crucial to boost productivity of many crops and maintain the integrity of both agro-biodiversity and ecosystem complex of in the country. Various control methods attempted world-wide in controlling parthenium have their own pros and cons. Hand-weeding mostly used by small-scale farmers is more difficult due to the allergic effects of parthenium on human body (Tefera, 2002; Wiesner et al., 2007). Mechanical control on the other hand is rather costly in terms of machinery, labour and time requirement. Furthermore, resource poor farmers of Ethiopia cannot afford the purchase of herbicides and the use of herbicides is unsafe in terms of health and environmental considerations and not allowed in when the farmers want to certify for organic production. Therefore, other system-oriented options must be sought for ecological management of parthenium weed in Ethiopia. One such option is the use of phytotoxic plants that suppress the germination and growth of parthenium. In the current study, the four multi-purpose tree species drastically reduced the germination and growth of parthenium. This could suggest that these multi-purpose tree species produce allelochemicals responsible for inhibitory effects or provide physical suppression on germination and growth of parthenium. Though the trend in pot-culture followed that of the Petri dish result, in contrast to the higher inhibitory effects obtained in Petri dishes, lower inhibitory effects by the multi-purpose tree species were observed in pot-culture. Therefore, further investigations will be required under greenhouse and field conditions for pragmatic recommendation of species selection in the frame of multi-purpose tree species-mediated ecological management of parthenium weed.
ACKNOWLEDGMENTS

We thank Ethiopian Institute of Agricultural Research (EIAR) for funding and Jimma University College of Agriculture and Veterinary Medicine (JUCAVM) for laboratory and greenhouse facilities. We are greatly indebted to Daniel Damtew for his assistance during execution of the laboratory and greenhouse experiments and Arafat Kassahun for data entry.

REFERENCES


