
Charles Darwin University

Acacia holosericea (Fabaceae) litter has allelopathic and physical effects on mission grass (Cenchrus pedicellatus and C. polystachios) (Poaceae) seedling establishment

Quddus, Muhammad Salman; Bellairs, Sean; Wurm, Penelope

Published in:
Australian Journal of Botany

DOI:
[10.1071/BT13294](https://doi.org/10.1071/BT13294)

Published: 24/06/2014

Document Version
Peer reviewed version

[Link to publication](#)

Citation for published version (APA):

Quddus, M. S., Bellairs, S., & Wurm, P. (2014). Acacia holosericea (Fabaceae) litter has allelopathic and physical effects on mission grass (Cenchrus pedicellatus and C. polystachios) (Poaceae) seedling establishment. *Australian Journal of Botany*, 62(3), 189-195. <https://doi.org/10.1071/BT13294>

General rights

Copyright and moral rights for the publications made accessible in the public portal are retained by the authors and/or other copyright owners and it is a condition of accessing publications that users recognise and abide by the legal requirements associated with these rights.

- Users may download and print one copy of any publication from the public portal for the purpose of private study or research.
- You may not further distribute the material or use it for any profit-making activity or commercial gain
- You may freely distribute the URL identifying the publication in the public portal

Take down policy

If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

Download date: 21. May. 2022

1 ***Acacia holosericea* (Fabaceae) litter has allelopathic and physical effects on mission**
2 **grass (*Cenchrus pedicellatus* and *C. polystachios*) (Poaceae) seedling establishment**

3 Muhammad S. Quddus*, Sean M. Bellairs, Penelope A.S. Wurm.
4 Research Institute for the Environment and Livelihoods, Charles Darwin University,
5 Australia.

6 *Corresponding author Email: Muhammad.Quddus@cdu.edu.au

7 **Abstract**

8 Invasion of grass weeds is a major threat for ecosystems. Mission grass (*Cenchrus*
9 *pedicellatus* and *C. polystachios*) vigorously competes with native vegetation and has
10 become a serious problem in northern Australian savanna. A lower density of mission grass
11 has been observed under the canopy of stands of native *Acacia holosericea*. We used a series
12 of laboratory and shade house experiments to assess the potential for allelopathy and the role
13 of litter on germination, emergence and seedling growth of these two species of mission
14 grass. Different concentrations of aqueous leaf extract of *A. holosericea* were used to assess
15 allelopathic effects on germination. Various depths and types of litter were used to investigate
16 the allelopathic and physical effects of litter on emergence and growth of mission grass
17 seedlings in the shade house. Results indicate that extracts did not affect germination of either
18 species of mission grass but root growth of seedlings was affected. Emergence of seedlings in
19 the shade house was affected by physical litter treatments but not by allelopathy. After
20 emergence no negative effects on seedling growth were detected. Overall we found that there
21 was no allelopathic effect on germination and that the negative effect on emergence was due
22 to the physical properties of the litter. This effect on emergence increased with increasing
23 depth of litter. Allelopathy slightly inhibited root growth but once seedlings emerged, litter
24 tended to facilitate growth. This has implications for the ecological management of mission
25 grass on disturbed lands, using strategies such as manipulation of litter cover through *Acacia*
26 establishment.

27

28 **Key words:** allelopathy, germination, emergence, suppression, grass weed

29 **Introduction**

30 Leaves of the Australian native species *Acacia holosericea* A.Cunn. ex G.Don may have an
31 allelopathic or physical impact on establishment of two exotic species of mission grass
32 (*Cenchrus pedicellatus* Trin and *C. polystachios* L. Morrone). These two African grass
33 species were introduced to Australia as fodder crops and have become environmental weeds
34 (Douglas *et al.* 2006; Miller 2006). These exotic grass species have the ability to change the
35 ecosystem by altering the fire regimes and nutrient dynamics (Miller 2006; Brooks *et al.*
36 2010). Woodlands of the Northern Territory are dominated by *Eucalyptus* and *Acacia* trees.
37 Field observations show that mission grass understorey is reduced below stands of *A.*
38 *holosericea* trees in northern Australia. This may be due to allelopathy or physical effects of
39 the leaf litter underneath these stands. If so then *A. holosericea* could potentially be used to
40 control mission grass establishment on managed disturbed sites, such as on topsoil stockpiles
41 of mine sites.

42

43 Plant allelopathy is defined as the effects of the chemical compounds involved in plant-plant
44 interactions (Rice 1984; Kruse *et al.* 2000). Plants may favourably or adversely affect other
45 plants through allelochemicals, which may be released directly or indirectly from living or
46 dead plants (Putnam and Duke 1978; Rice 1979; Rice 1984; Putnam and Tang 1986; Kruse,
47 Strandberg *et al.* 2000; Zimdahl 2007). Allelopathy is defined as any process involving
48 secondary metabolites produced by plants, algae, bacteria and fungi which influence the
49 growth and development of agricultural and biological systems (Romeo 2000; Macias *et al.*
50 2007). Most plants have the potential to produce chemicals that can inhibit or stimulate other
51 plants. Allelopathy works in three steps. In the first step phytotoxic chemicals are produced,
52 in the second step these chemicals are transported from donor to recipient, and in the last step
53 target plants are exposed to chemicals for a sufficient time and concentration to have an
54 effect (Aldrich 1984; Lovett and Ryuntyu 1992). Many types of allelochemicals are released
55 from plants, affecting germination and growth of target plants. Plant parts that have been
56 implicated in the production of phytotoxic chemicals include roots rhizomes, stems, leaves,
57 flowers, inflorescences and seeds, and their impact varies from species to species. These
58 chemicals are released to the environment through root exudation and leachates from litter
59 (Rice 1984). For example, extracts from leaves of *Acacia nilotica* and *Eucalyptus rostrata*
60 reduced the growth and germination of *Zea mays* and *Phaselulus vulgaris* (El-Khawas and
61 Shehata 2005). Aqueous leachates of *Eucalyptus globulus* leaves reduce the chlorophyll
62 content in leaves of *Eleusine coracana* (Padhy *et al.* 2000). *Eucalyptus baxteri* releases
63 allelochemicals which suppress understorey vegetation (Lovett 1986). Mousavi *et al.* (2013)
64 compared extracted solutions from different organs of *Melilotus indicus* and found that the
65 leaf extract had the highest inhibitory effect on germination and seedling growth of *Triticum*
66 *aestivum*. Foliar leachates of many Australian woody plants inhibit the germination of *Avena*
67 *fatua* (Hobbs and Atkins 1991). Leaf extracts typically have greater inhibitory properties than
68 root extracts (Chon 2010).

69 Accumulation of litter is an important factor affecting the establishment of plants (Facelli and
70 Pickett 1991b). Litter of different depths can create different micro sites for plant
71 establishment (Molofsky and Augspurger 1992), which affects the structure and composition
72 of plant communities (Facelli and Pickett 1991b; Baker and Murray 2010). The effect of leaf
73 litter on plant recruitment can be positive, negative or neutral. Negative effects of leaf litter
74 other than allelopathy can be due to a mechanical barrier limiting radicle or shoot growth, or
75 due to modification of the quality and quantity of light reaching seeds. Negative effects of
76 litter on germination and seedling growth often increase with increased depth of litter (Xiong
77 and Nilsson (1999). Leaf litter modifies the soil micro climate by changing the radiation
78 reaching the soil surface, as well as moisture, temperature and nutrient levels in the soil
79 (Facelli and Pickett 1991b). Litter can increase seedling establishment by improving soil
80 moisture (Facelli and Pickett 1991b) and by protecting the seeds from predators (Cintra
81 1997). The impact of litter on seedling establishment is more likely to be positive when it is
82 present in small quantities (Loydi *et al.* 2013). The effect of litter also varies with species and
83 seasons (Facelli and Pickett 1991a; Ruprecht *et al.* 2010).

84

85 We evaluate the impact of the leaf litter of *A. holosericea* on *C. pedicellatus* and *C.*
86 *polystachios* germination in the laboratory and on seedling emergence and growth in a shade
87 house. We address the hypothesis that leaves of *A. holosericea* suppress the germination,
88 emergence and growth of mission grass via allelopathy and physical mechanisms. The
89 laboratory study investigated the allelopathic potential of *A. holosericea* on seed germination
90 and seedling growth of two species of invasive mission grass. The shade house study
91 investigated the effect of different types and depths of litter on seedling emergence and

92 growth. Implications are discussed for using *A. holosericea* for the ecological control of
93 mission grass in disturbed sites to assist in rehabilitation. Bioassays are the most common and
94 widely accepted technique used to investigate the effect of allelopathy in the laboratory
95 (Lovett and Ryuntyu 1992). Laboratory bioassays need to be supplemented with nursery
96 trials, as laboratory trials may create chemical conditions which do not occur in field.

97 **Material and methods**

98 *Plant sampling and preparation of aqueous extract*

99 Mature seeds of annual *C. pedicellatus* and perennial *C. polystachios* were collected in May
100 2013, seven months before the start of this study. Seeds were stored at a constant air
101 temperature of 18°C in paper bags. Germinability immediately before the experiment was
102 99%.

103
104 Mature leaves of *A. holosericea* were picked from plants growing on a disturbed site at the
105 Charles Darwin University, Darwin, Australia (12°22'S, 130°52'E). These leaves were rinsed
106 with distilled water to remove dust particles (Sarkar *et al.* 2012) and then placed on a 0.5 cm
107 layer of soil in a dry shaded area. They were kept there for four weeks to let them dry
108 thoroughly and to allow interactions with soil bacteria and fungi. After drying, the leaves
109 were crushed gently by hand to allow leaching of compounds from the leaf. Leaves were not
110 ground, as grinding of plant material can disrupt cellular integrity which impacts on the
111 release of allelochemicals (Putnam and Duke 1978; Inderjit and Dakshini 1995).

112
113 Leaf material was soaked in distilled water at the rate of 10, 20, 40 and 80 g of dry leaf L⁻¹, at
114 29 ± 1°C for three days as described by Al-Humaid and Warrag (1998) and Warrag (1995).
115 This mixture was then filtered through Whatman filter paper no. 4 (Heisey 1990). The pH of
116 these solutions was measured by using a TPS digital pH meter (LC80A). The electrical
117 conductivity was measured using a Hanna HI-8733 conductivity meter. The pH ranged from
118 5.46 to 5.27 and the EC ranged from 0.00 mS cm⁻¹ to 1.19 mS cm⁻¹ (40 g leaf L⁻¹) and 2.4 mS
119 cm⁻¹ (80 g leaf L⁻¹). Solutions were then refrigerated in plastic bottles at 5°C.

120
121 Plastic Petri dishes (90 mm diameter) were lined with two sheets of Whatman No. 1 filter
122 paper. Twenty five seeds were placed onto the filter papers in each Petri dish, and then
123 moistened with 5 ml of one of the four aqueous solutions of *A. holosericea* leaf extract along
124 with a control moistened with distilled water (Rejila and Vijayakumar 2011). Four replicate
125 Petri dishes were arranged in a randomized block design for the bioassay for both grass
126 species. The Petri dishes were sealed in plastic bags to reduce evaporation and placed in
127 growth chambers at 25-32°C and 12/12 hours dark/light photoperiods.

128
129 Seeds were considered germinated where the radicle of the hypocotyl exceeded 1 mm. The
130 number of germinated seeds was counted daily and the root and shoot length of all seeds that
131 had germinated were measured with a ruler at five days after sowing. Germination rate was
132 calculated as the average time until germination occurred using the following equation.

133
134 Germination rate = $G_1/t_1 + G_2/t_2 + G_3/t_3 + G_4/t_4 + G_5/t_5$
135

136 Where G was the percentage of seeds that germinated on that day and t was the number of
137 days into the germination period.

138 *Shade house study of leaf litter*

139 The effect of the different litter types on seedling emergence and growth of *C. pedicellatus*
140 and *C. polystachios* was investigated in germination trays in a shade house at Charles Darwin
141 University. Litter treatments comprised fresh *A. holosericea* leaves, dried *A. holosericea*
142 leaves and synthetic litter (Detpak brown paper bags A2317).

143
144 To select the appropriate materials to create synthetic litter, a pilot study was carried out to
145 evaluate a selection of materials similar to those used in other studies (Barritt and Facelli
146 2001; Harris *et al.* 2003; Rotundo and Aguiar 2005). Packing strips (Signode polypropylene
147 strapping), paper folders (cardboard file folder), shade cloth (90% universal shade cloth),
148 polypropylene bags (woven polypropylene bags) and paper bags (Detpak brown paper bags
149 A2317) were tested. Paper bags showed similar results to natural litter in colour, volume,
150 packing structure and physical response to wetting and drying.

151
152 Fresh leaves were obtained by cutting mature leaves from trees the day before the seeds were
153 sown. Leaves were rinsed with deionised water before being placed on the trays. Dry leaves
154 were prepared by collecting fresh leaves, which were then rinsed and placed as a 0.5 cm layer
155 on soil for four weeks in a dry shaded area, before being placed on the trays. Synthetic litter
156 was made by cutting the paper bags into strips the same size as a typical *A. holosericea* leaf,
157 3.5 cm wide by 12.5 cm long.

158
159 Treatments were: 1 cm depth of fresh leaves, 1 cm and 3 cm depths of dried leaves, 1 cm and
160 3 cm depths of synthetic litter, two concentrations of *A. holosericea* leaf extract aqueous
161 solutions (40 g leaf L⁻¹ and 80 g leaf L⁻¹) and a control with no litter or extract treatment. Leaf
162 litter depths of 1 cm and 3 cm were chosen as they reflect the depth of litter typically found
163 under field conditions (unpublished data). Field measurements of mass per unit area of dry
164 litter were used to calibrate the amount of litter used to create 1 cm and 3 cm treatments.

165
166 To estimate the amount of paper leaves required for the synthetic litter treatment, we used the
167 equivalent area of leaves used in the dry litter treatment. Leaf area was measured with a leaf
168 scanner (Epson Perfection V33) and Image J software (developed by Wayne Rasband
169 National Institutes of Health, Maryland). On the basis of this measurement, the weight of
170 synthetic litter required to create the two depth treatments was calculated.

171
172 Topsoil was collected from the field away from the *Acacia* plants and was sieved through a
173 10 mm sieve to remove gravel from the shallow lateritic soil. This sieved soil was then mixed
174 with 20% coco peat for better drainage to avoid water logging and placed in 34 x 28 x 5 cm
175 seedling trays. Each tray was divided into halves using a plastic barrier; one half sown with
176 50 seeds of *C. pedicellatus* and the other sown with 50 seeds of *C. polystachios*. Four
177 replicate trays were used per treatment. Germination trays were arranged randomly in a shade
178 house.

179
180 Seeds were sown into each germination tray prior to the placement of the leaf litter treatments
181 on top of the soil. Germination trays were watered manually daily and emergence was
182 recorded on a daily basis for three weeks. At the end of the experiment, three weeks after
183 sowing, the number of tillers were counted and root and shoot length were measured using a
184 ruler. Root and shoot dry weights were also recorded after harvest. Soil was carefully washed
185 from the roots and the seedling samples were dried in an oven at 65°C for 24 hours.

186

187 The effect of leaf extracts and litter treatments on germination, emergence and growth were
188 analysed separately for each species using one way ANOVA. Analyses were performed using
189 STATISTICA 11 (StatSoft, Tulsa, USA). Data were transformed where necessary and
190 Tukey's test was performed for *post hoc* comparison of means.

191 Results

192 *Seed germination and seedling growth bioassay*

193 For the laboratory bioassay using the aqueous leaf extracts, seed germination was high in all
194 treatments. There were no significant effects of the aqueous solutions of *A. holosericea* leaf
195 extract on germination of *C. pedicellatus* or *C. polystachios* in the laboratory experiment
196 ($P>0.05$; Fig. 1). Germination for *C. pedicellatus* ranged from 95-99% and *C. polystachios*
197 from 87-100%.

198

199 Seedling root growth in the laboratory was significantly affected by the treatments for both
200 species ($P<0.05$; Fig. 2a and b). The shortest mean root length for *C. pedicellatus* occurred in
201 the 80 g leaf L⁻¹ solution treatment (15.2 ± 0.4 mm) and the longest mean root length in the
202 control (33.6 ± 0.5 mm). *C. polystachios* also had longest mean root length in the control
203 (25.7 ± 1.7 mm) and shortest (4.3 ± 0.2 mm) in the highest extract concentration (80 g leaf
204 L⁻¹). Differences in shoot length were significant for *C. pedicellatus* ($P<0.05$) but not *C.*
205 *polystachios* ($P>0.05$). Shoot length of *C. pedicellatus* was smallest when exposed to the
206 most concentrated extract solution (80 g leaf L⁻¹) and this was significantly different to that
207 in the 20 g leaf L⁻¹ and 40 g leaf L⁻¹ treatments.

208

209 *Seedling emergence through leaf litter*

210 In the shade house trial there was a slight allelopathic effect on one species. Leaf extracts of
211 *A. holosericea* significantly affected emergence of *C. polystachios* ($P<0.05$) at the highest
212 concentration but there was no effect on the emergence of *C. pedicellatus* ($P>0.05$), which
213 had 80 – 90% emergence in the extract treatments and control (Fig. 3a). Only 74%
214 emergence of *C. polystachios* occurred when exposed to the most concentrated extract
215 solution (80 g leaf L⁻¹) and this was significantly different to the 90% germination occurring
216 in the control (Fig. 3b).

217

218 Different litter treatments affected the emergence of *C. pedicellatus* and *C. polystachios*
219 seedlings and the effects were greater than the allelopathic responses to treatments involving
220 application of aqueous leaf extracts. Increases in litter depth significantly decreased
221 emergence ($P<0.05$). Emergence of *C. pedicellatus* was 88% in the control, 38% in trays with
222 1 cm of dry leaf litter and only 10% in trays with 3 cm of dry leaf litter (Fig. 3a). Emergence
223 in trays with 1 cm of fresh leaf litter was significantly lower as compared to control and
224 significantly higher than the dry litter treatments. The effects of the synthetic litter treatments
225 were very similar to those of the equivalent depths of dry leaf litter.

226

227 Effects of the litter treatments on emergence of *C. polystachios* seedlings were similar to *C.*
228 *pedicellatus* ($P<0.05$; Fig. 3b). However, the degree of suppression of *C. polystachios*
229 emergence was greater. The suppression of emergence by fresh leaves was significantly
230 different to that under dry and synthetic litter. There were no significant differences in
231 emergence between the dry leaf litter and the same depth of synthetic litter. Mean emergence
232 in the 3 cm litter treatments was about half that of the 1 cm litter treatments but the

233 differences were not significant. Emergence was reduced from 90% in the control to just 4%
234 in the treatment with 3 cm depth of dry leaf litter.

235

236 *Allelopathic effect on growth of seedlings in the shade house*

237 The aqueous leaf extracts of *A. holosericea* had no effect on shoot length of *C. pedicellatus* or
238 *C. polystachios* or on root length of *C. pedicellatus* ($P>0.05$; Fig. 4a and b). Root length of *C.*
239 *polystachios* was significantly shorter ($P<0.05$) in the highest extract concentration of 80 g
240 leaf L⁻¹ as compared to the control, although the 40 g leaf L⁻¹ treatment was not significantly
241 different to the control or 80 g leaf L⁻¹ extract treatment (Fig. 4b).

242

243 The different litter treatments had no significant effect on shoot or root growth of *C.*
244 *pedicellatus* or *C. polystachios* ($P>0.05$) but results were very variable between plants. Mean
245 shoot length of *C. pedicellatus* was 104 ± 11 mm in the control and a mean maximum of 293
246 ± 24 mm occurred in the 1 cm dry leaf litter treatment (Fig. 4a). Root lengths varied
247 consistently with shoot lengths. Similarly the *C. polystachios* treatment with the longest mean
248 shoot length (171 ± 38 mm) was the dry leaf litter to 1 cm depth and mean shoot growth in
249 the control was 109 ± 29 mm (Fig. 4b).

250

251 Discussion

252 Leaf extracts of *A. holosericea* had some limited effects on *C. pedicellatus* and *C.*
253 *polystachios* in the laboratory. The leaf extracts did not affect the proportion of seeds that
254 germinated. However seedlings had reduced root growth. Chon *et al.* (2002), Olson and
255 Wallander (2002) and Kelsey and Locken (1987) also found that root growth was more
256 sensitive to toxicity as compared to germination percentage or shoot length. This effect was
257 not due to pH differences as there was little difference in pH between the strongest solution
258 and the control. EC differences between the extracts were also small but the EC of the
259 strongest solution may just be sufficient to affect the growth of sensitive species (Landon
260 1991). However, bioassay trials are used only to determine that plant to plant interactions
261 occur. Whether this is ecologically important needs to be confirmed in more natural
262 conditions (Inderjit and Moral 1997; Inderjit and Nilsen 2003) and so the findings of the
263 shade house experiment must also be considered.

264
265 When the *A. holosericea* leaf extracts were applied to soil the effect on mission grass was
266 minimal. Only a minor effect on *C. polystachios* emergence at the highest extract
267 concentration was observed and there was no significant effect on emergence of the annual *C.*
268 *pedicellatus*. This inhibition effect could be due to the osmotic concentration of the extract
269 rather than due to particular toxicity effects. For example, Chou *et al.* (1998) found for
270 *Acacia confusa*, that inhibition can occur both due to osmotic concentration of the extract and
271 phytotoxicity. The osmotic concentration of their 5% extracts ranged from 40 to 50 mosmol.
272 Normally when osmotic concentration exceeds 50 mosmol, it may cause inhibition of
273 emergence. Regardless, there is little evidence of allelopathy.

274
275 Both the laboratory and the shade house results in our study suggested that at most there is
276 minimal influence of allelopathy on germination and emergence. Allelopathic effects can
277 vary with time, as allelochemicals can be toxified or detoxified in soil by microorganisms
278 (Inderjit 2001; Bhadoria 2011). Gonzalez *et al.* (1995) suggested that continuous presence of
279 *Acacia* leaves on soil might be responsible for toxicity and that they are more toxic during the
280 early period of decomposition (Souto *et al.* 1994). If this were so for *A. holosericea* we would
281 have expected it to occur in the fresh leaf treatment.

282
283 Litter can affect seedling emergence due to physical, chemical or biological factors or a
284 combination of these factors (Facelli and Pickett 1991b; Cavieres *et al.* 2006). Litter
285 accumulation alters the physical environment by changing light conditions and soil
286 temperature. Light quality, light quantity or temperature conditions received by seeds can
287 inhibit seed germination and emergence. Chemical factors affecting emergence could be
288 through the release of nutrients, chemicals stimulating germination or toxic chemicals
289 (Facelli and Pickett 1991b). Biological impacts of litter can be through changes to the soil
290 biology influencing fungal and non-fungal diseases killing seeds and seedlings (Facelli and
291 Pickett 1991b; Rotundo and Aguiar 2005). However, our study determined the major effect
292 on mission grass was physical with a minor chemical impact.

293
294 Seedling emergence of both species of mission grass decreased considerably with increased
295 depth of litter. This effect was not via allelopathy, as the effect of synthetic litter was similar
296 to that of *Acacia* leaf litter. Barritt and Facelli (2001) documented that litter reduced seedling
297 emergence and that natural and artificial litter can have the same physical effect on seedling
298 emergence. They assessed the effects of *Casuarina pauper* litter on the emergence and
299 growth of an introduced annual forb, *Carrichtera annua* and a native grass *Danthonia*
300 *caespitosa*. They concluded that litter had strong and consistent negative effects on the

301 emergence of the seedlings of both species due to the physical barrier provided by litter.
302 Baker and Murray (2010) also found that increased leaf litter depth reduced emergence and
303 establishment. Hamrick and Lee (1987) observed that hypocotyl length of *Carduus nutans*
304 was longer under high litter conditions as compared to less or no litter and suggested that
305 mortality was higher due to the use of more stored energy used to penetrate the litter layers.
306 This extra use of energy weakened the seedlings and many died before reaching the surface
307 or soon afterwards. Our study concurs with these previous findings.

308
309 Small seeds are more susceptible to negative effects compared to large seeds. Mission grass
310 has a small seed and seed size can influence whether litter has a negative or positive effect on
311 emergence. In contrast to our findings, Molofsky and Augspurger (1992) trialled different
312 litter depths and found that *Gustavia* seedling emergence was greater under litter than on bare
313 ground. This positive effect on *Gustavia* emergence was due to the higher moisture and
314 humidity in soil with litter cover and the seedlings of *Gustavia* are shade tolerant. However
315 they also found that emergence of small seeded *Luehea*, *Ochroma* and *Ceiba* were negatively
316 affected by litter and this effect increased with increases in litter depths.

317
318 Subsequent to emergence, the positive effects of litter treatments on growth might be due to
319 moisture conservation (Xiong and Nilsson 1999). Favourable growth could be due to benefits
320 of reduced evaporation and increased water holding capacity. Litter may serve as a source of
321 nutrients and soil insulation from high temperatures (Cheplick and Quinn 1987; Facelli and
322 Pickett 1991c; Facelli and Brenton 1996).

323
324 While allelopathic effects of *A. holosericea* did not affect the proportion of seedlings that
325 emerged in the shade house, it inhibited the seedling growth of *C. polystachios* at the highest
326 concentration, affecting root growth. These findings are consistent with other studies. For
327 example, percent germination, shoot length and root length of rice and cow peas have been
328 shown to decrease due to *Acacia auriculiformis* leaf leachates, and root and shoot length were
329 affected more than germination (Hoque *et al.* 2003; Oyun 2006). *A. nilotica* and *E. rostrata*
330 released allelochemicals which reduced the growth of *Z. mays* and *P. vulgaris* (El-Khawas
331 and Shehata 2005; Bargali and Bargali 2009). Lorenzo *et al.* (2011) reported the inhibitory
332 effects of *Acacia dealbata* on understory *Dactylis glomerata*, and suggested that allelopathic
333 interference seems to contribute to this process. Many other Australian trees, and especially
334 *Eucalyptus* species, produce allelochemicals which affect the understory vegetation (Bowman
335 and Kirkpatrick 1986; May and Ash 1990). While not completely suppressing establishment of
336 seedlings, reduced root development in seedlings may contribute to a reduction in the
337 development of understory.

338
339 Field observations show that mission grass understory is reduced below *A. holosericea* trees
340 in northern Australia. From this study it is concluded that there is no effect of allelopathy on
341 germination, but that litter has a negative physical effect on emergence and this is greater for
342 thicker litter layers. Allelopathy may have a slight inhibitory effect on seedling root growth
343 but after emergence thin litter layers could later have a facilitative effect. The physical impact
344 of litter is more important than allelopathy on the establishment and growth. *A. holosericea*
345 has relatively thick robust leaves which provide more of a physical barrier than small and thin
346 leaved species. The slight effect of allelopathy in reducing grass seedling root length may
347 increase water stress which would become more critical in the field when combined with
348 competition with trees. Thus the findings of this study point to the control of mission grass
349 establishment in the field, by the physical impact of dense *A. holosericea* leaf litter, combined
350 with a mild allelopathic effect on seedling root growth and with tree-grass competition.

351 **Acknowledgments**

352 The authors wish to thank the Pakistan Higher Education Commission and Charles Darwin
353 University for providing funds to conduct research. We also thank Jayshree Mamtora for
354 assistance with library services.

355 **References:**

- 356 Al-Humaid AI, Warrag MOA (1998) Allelopathic effects of mesquite (*Prosopis juliflora*)
357 foliage on seed germination and seedling growth of bermudagrass (*Cynodon dactylon*).
358 *Journal of Arid Environments* **38**, 237-243.
- 359
360 Aldrich RJ (1984) 'Weed-crop ecology: Principles in weed management.' (Breton Publishers:
361 Massachusetts).
- 362
363 Baker AC, Murray BR (2010) Relationships between leaf-litter traits and the emergence and
364 early growth of invasive *Pinus radiata* seedlings. *Weed Research* **50**, 586-596.
- 365
366 Bargali K, Bargali S (2009) *Acacia nilotica*: A multipurpose leguminous plant. *Nature and*
367 *Science* **7**, 11-19.
- 368
369 Barritt AR, Facelli JM (2001) Effects of *Casuarina pauper* litter and grove soil on emergence
370 and growth of understorey species in arid lands of South Australia. *Journal of Arid*
371 *Environments* **49**, 569-579.
- 372
373 Bhadoria PBS (2011) Allelopathy: A natural way towards weed management. *American*
374 *Journal of Experimental Agriculture* **1**, 7-20.
- 375
376 Bowman D, Kirkpatrick J (1986) Establishment, suppression and growth of *Eucalyptus*
377 *delegatensis* R.T. Baker in multiaged forests .III. Intraspecific allelopathy, competition
378 between adult and juvenile for moisture and nutrients, and frost damage to seedlings.
379 *Australian Journal of Botany* **34**, 81-94.
- 380
381 Brooks KJ, Setterfield SA, Douglas MM (2010) Exotic grass invasions: Applying a
382 conceptual framework to the dynamics of degradation and restoration in Australia's tropical
383 savannas. *Restoration Ecology* **18**, 188-197.
- 384
385 Cavieres LA, Chacon P, Penaloza A, Molina-Montenegro M, Arroyo MTK (2006) Leaf litter
386 of *Kageneckia angustifolia* D. Don (Rosaceae) inhibits seed germination in sclerophyllous
387 montane woodlands of central Chile. *Plant Ecology* **190**, 13-22.
- 388
389 Cheplick GP, Quinn JA (1987) The role of seed depth, litter, and fire in the seedling
390 establishment of amphicarpic peanutgrass (*Amphicarpum purshii*). *Oecologia* **73**, 459-464.
- 391
392 Chon SU (2010) Allelopathy in Compositae plants. A review. *Agronomy for Sustainable*
393 *Development* **30**, 349-358.
- 394

395 Chon SU, Choi S-K, Jung S, Jang H-G, Pyo B-S, Kim S-M (2002) Effects of alfalfa leaf
396 extracts and phenolic allelochemicals on early seedling growth and root morphology of
397 alfalfa and barnyard grass. *Crop Protection* **21**, 1077-1082.

398

399 Chou C-H, Fu C-Y, Li S-Y, Wang Y-F (1998) Allelopathic potential of *Acacia confusa* and
400 related species in Taiwan. *Journal of Chemical Ecology* **24**, 2131-2150.

401

402 Cintra R (1997) Leaf litter effects on seed and seedling predation of the palm *Astrocaryum*
403 *murumuru* and the legume tree *Dipteryx micrantha* in Amazonian forest. *Journal of Tropical*
404 *Ecology* **13**, 709-725.

405

406 Douglas MM, Setterfield SA, O'Connor RA, Ferdinands K, Rossiter NA, Brooks KJ, Ryan B,
407 Parr C (2006) Different weeds, different habitats, same effects: exotic grass invasion in
408 tropical woodlands and wetlands. In '15th Australian Weeds Conference Managing Weeds in
409 a Changing Climate', 24-28 September 2006, Adelaide. (Eds C Preston., JH Watts. and ND
410 Crossman.), pp. 811-814

411

412 El-Khawas SA, Shehata MM (2005) The allelopathic potentialities of *Acacia nilotica* and
413 *Eucalyptus rostrata* on monocot (*Zea mays* L.) and dicot (*Phaseolus vulgaris* L.) plants.
414 *Biotechnology* **4**, 23-34.

415

416 Facelli JM, Brenton L (1996) Germination requirements and responses to leaf litter of four
417 species of eucalypt. *Oecologia* **107**, 441-445.

418

419 Facelli JM, Pickett STA (1991a) Indirect effects of litter on woody seedlings subject to herb
420 competition. *Oikos* **62**, 129-138.

421

422 Facelli JM, Pickett STA (1991b) Plant litter: Its dynamics and effects on plant community
423 structure. *The Botanical Review* **57**, 1-32.

424

425 Facelli JM, Pickett STA (1991c) Plant litter: Light interception and effects on an old-field
426 plant community. *Ecology* **72**, 1024-1031.

427

428 Gonzalez L, Souto XC, Reigosa MJ (1995) Allelopathic effects of *Acacia melanoxylon* R. Br.
429 phyllodes during their decomposition. *Forest Ecology and Management* **77**, 53-63.

430

431 Hamrick JL, Lee JM (1987) Effect of soil surface topography and litter cover on the
432 germination, survival, and growth of musk thistle (*Carduus nutans*). *American Journal of*
433 *Botany* **74**, 451-457.

434

435 Harris MR, Lamb D, Erskine PD (2003) An investigation into the possible inhibitory effects
436 of white cypress pine (*Callitris glaucophylla*) litter on the germination and growth of
437 associated ground cover species. *Australian Journal of Botany* **51**, 93-102.

438
439 Heisey RM (1990) Allelopathic and herbicidal effects of extracts from tree of heaven
440 (*Ailanthus altissima*). *American Journal of Botany* **77**, 662-670.

441
442 Hobbs RJ, Atkins L (1991) Interactions between annuals and woody perennials in a Western
443 Australian nature reserve. *Journal of Vegetation Science* **2**, 643-654.

444
445 Hoque A, Ahmed R, Uddin M, Hossain M (2003) Allelopathic effect of different
446 concentration of water extracts of *Acacia auriculiformis* leaf on some initial growth
447 parameters of five common agricultural crops. *Pakistan Journal of Agronomy* **2**, 92-100.

448
449 Inderjit (2001) Soil: Environmental effects on allelochemical activity. *Agronomy Journal* **93**,
450 79-84.

451
452 Inderjit, Dakshini KMM (1995) On laboratory bioassays in allelopathy. *Botanical Review* **61**,
453 28-44.

454
455 Inderjit, Moral R (1997) Is separating resource competition from allelopathy realistic? *The*
456 *Botanical Review* **63**, 221-230.

457
458 Inderjit, Nilsen ET (2003) Bioassays and field studies for allelopathy in terrestrial plants:
459 Progress and problems. *Critical Reviews in Plant Sciences* **22**, 221-238.

460
461 Kelsey R, Locken L (1987) Phytotoxic properties of cnicin, a sesquiterpene lactone from
462 *Centaurea maculosa* (spotted knapweed). *Journal of Chemical Ecology* **13**, 19-33.

463
464 Kruse M, Strandberg M, Strandberg B (2000) 'Ecological effects of allelopathic plants—a
465 review technical report No. 315.' (National Environmental Research Institute: Silkeborg,
466 Denmark).

467
468 Landon J (Ed.) (1991) 'Booker tropical soil manual: A handbook for soil survey and
469 agricultural land evaluation in the tropics and subtropics.' (Addison Wesley Longman Ltd:
470 Harlow).

471
472 Lorenzo P, Palomera-Perez A, Reigosa MJ, Gonzalez L (2011) Allelopathic interference of
473 invasive *Acacia dealbata* Link on the physiological parameters of native understory species.
474 *Plant Ecology* **212**, 403-412.

475

476 Lovett J, Ryuntyu M (1992) Allelopathy: Broadening the context. In 'Allelopathy: Basic and
477 applied aspects. (Eds SJH Rizvi and V Rizvi) pp. 11-19. (Chapman & Hall: London).

478
479 Lovett JV (Ed.) (1986) 'Allelopathy: the Australian experience.' The science of allelopathy
480 (John Wiley & Sons: New York).

481
482 Loydi A, Eckstein RL, Otte A, Donath TW (2013) Effects of litter on seedling establishment
483 in natural and semi-natural grasslands: A meta-analysis. *Journal of Ecology* **101**, 454-464.

484
485 Macias FA, Molinillo JMG, Varela RM, Galindo JCG (2007) Allelopathy—a natural
486 alternative for weed control. *Pest Management Science* **63**, 327-348.

487
488 May F, Ash J (1990) An assessment of the allelopathic potential of *Eucalyptus*. *Australian*
489 *Journal of Botany* **38**, 245-254.

490
491 Miller I (2006) 'Management of mission grass (*Pennisetum polystachion*).' (Biosecurity and
492 Product Integrity, Department of Primary Industry, Fisheries and Mines, Northern Territory
493 Government: Darwin).

494
495 Molofsky J, Augspurger CK (1992) The effect of leaf litter on early seedling establishment in
496 a tropical forest. *Ecology* **73**, 68-77.

497
498 Mousavi S, Alami-Saeid K, Moshatati A (2013) Effect of leaf, stem and root extract of alfalfa
499 (*Melilotus indicus*) on seed germination and seedling growth of wheat (*Triticum aestivum*).
500 *International Journal of Agriculture and Crop Sciences* **5**, 44-49.

501
502 Olson BE, Wallander RT (2002) Effects of invasive forb litter on seed germination, seedling
503 growth and survival. *Basic and Applied Ecology* **3**, 309-317.

504
505 Oyun M (2006) Allelopathic potentialities of *Gliricidia sepium* and *Acacia auriculiformis* on
506 the germination and seedling vigour of maize (*Zea mays L.*). *American Journal of*
507 *Agricultural and Biological Sciences* **1**, 44-47.

508
509 Padhy B, Patnaik PK, Tripathy AK (2000) Allelopathic potential of *Eucalyptus* leaf litter
510 leachates on germination and seedling growth of finger millet. *Allelopathy Journal* **7**, 69-78.

511
512 Putnam AR, Duke WB (1978) Allelopathy in agroecosystems. *Annual Review of*
513 *Phytopathology* **16**, 431-451.

514
515 Putnam AR, Tang C-S (1986) 'The science of allelopathy.' (John Wiley & Sons Inc.: New
516 York).

517
518 Rejila S, Vijayakumar N (2011) Allelopathic effect of *Jatropha curcas* on selected
519 intercropping plants (green chilli and sesame). *Journal of Phytology* **3**, 1-3.

520
521 Rice EL (1979) Allelopathy: An Update. *Botanical Review* **45**, 15-109.

522
523 Rice EL (1984) 'Allelopathy ' (Academic Press: London).

524
525 Romeo J (2000) Raising the beam: Moving beyond phytotoxicity. *Journal of Chemical*
526 *Ecology* **26**, 2011-2014.

527
528 Rotundo JL, Aguiar MR (2005) Litter effects on plant regeneration in arid lands: A complex
529 balance between seed retention, seed longevity and soil–seed contact. *Journal of Ecology* **93**,
530 829-838.

531
532 Ruprecht E, Jozsa J, Olvedi TB, Simon J (2010) Differential effects of several “litter” types
533 on the germination of dry grassland species. *Journal of Vegetation Science* **21**, 1069-1081.

534
535 Sarkar E, Chatterjee SN, Chakraborty P (2012) Allelopathic effect of *Cassia tora* on seed
536 germination and growth of mustard. *Turkish Journal of Botany* **36**, 488-494.

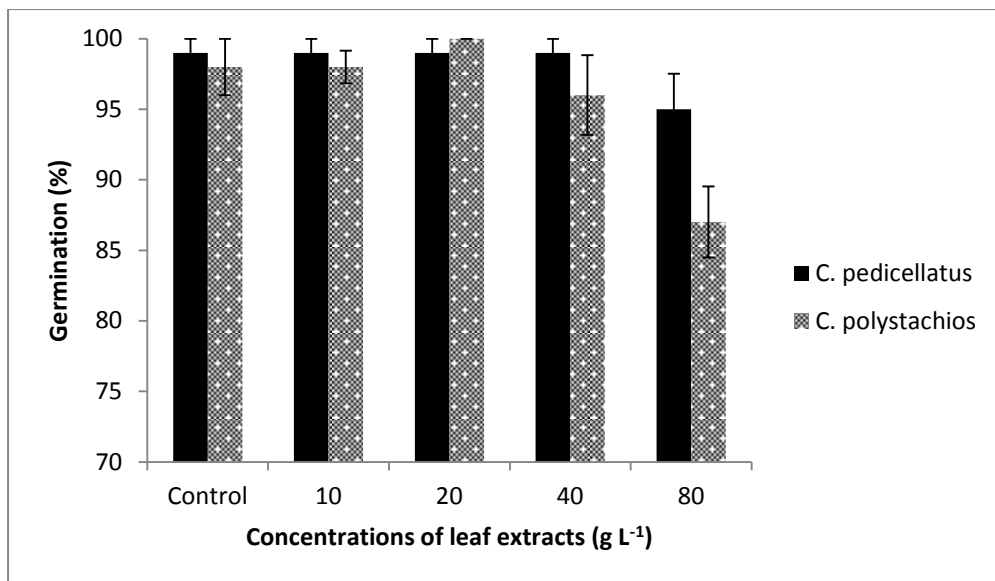
537
538 Souto X, Gonzales L, Reigosa M (1994) Comparative analysis of allelopathic effects
539 produced by four forestry species during decomposition process in their soils in Galicia (NW
540 Spain). *Journal of Chemical Ecology* **20**, 3005-3015.

541
542 Warrag MOA (1995) Autotoxic potential of foliage on seed germination and early growth of
543 mesquite (*Prosopis juliflora*). *Journal of Arid Environments* **31**, 415-421.

544
545 Xiong S, Nilsson C (1999) The effects of plant litter on vegetation: a meta-analysis. *Journal*
546 *of Ecology* **87**, 984-994.

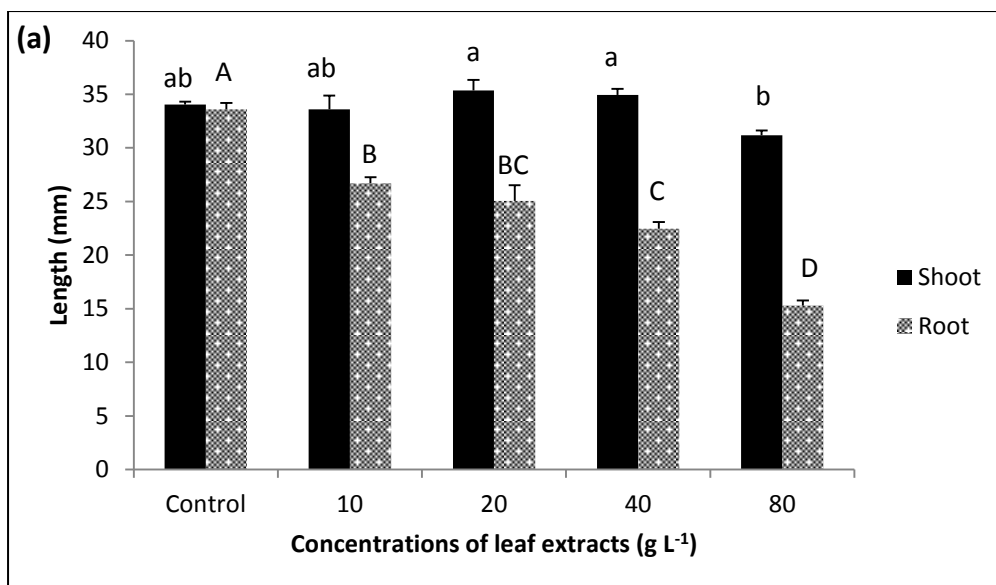
547
548 Zimdahl RL (2007) 'Fundamentals of weed science.' (Academic Press: San Diego).

549
550
551
552
553
554
555

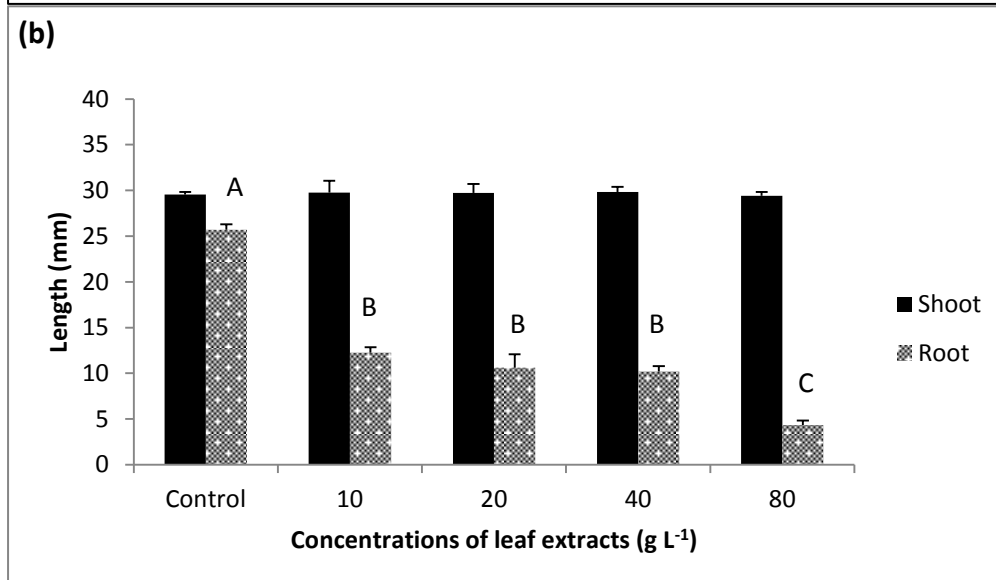


556

557 Fig. 1. Effects of leaf extract of *A. holosericea* on germination of *C. pedicellatus* (■) and *C.*
 558 *polystachios* (▨). Values are mean ± s.e. (n=4).



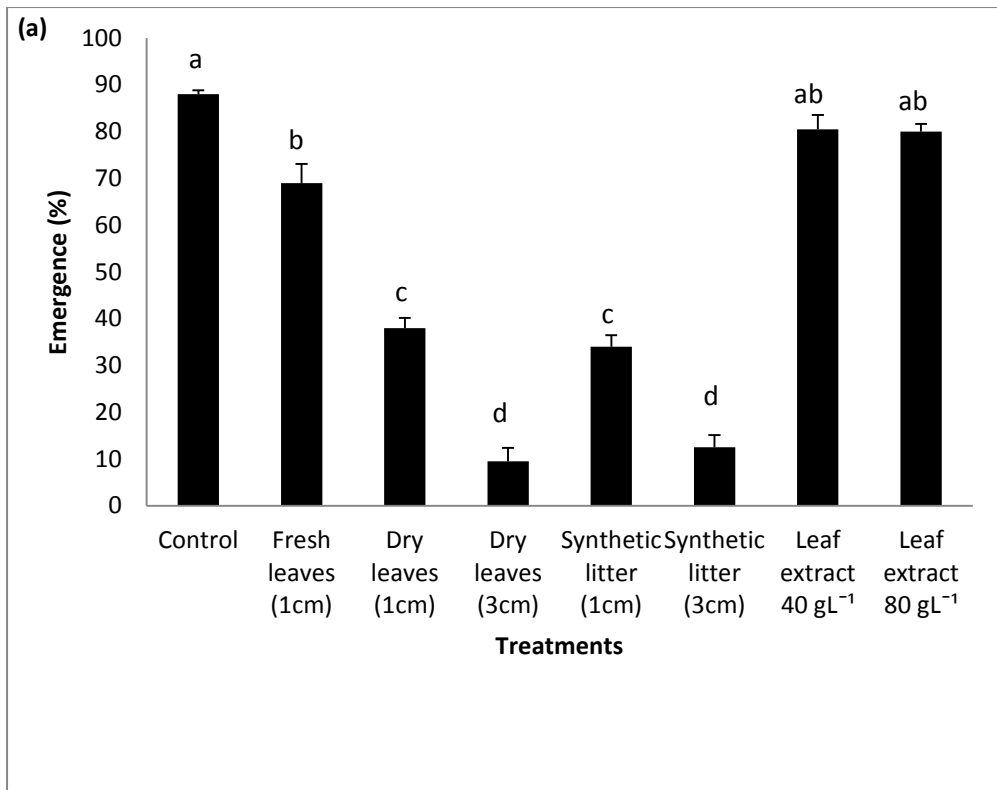
559



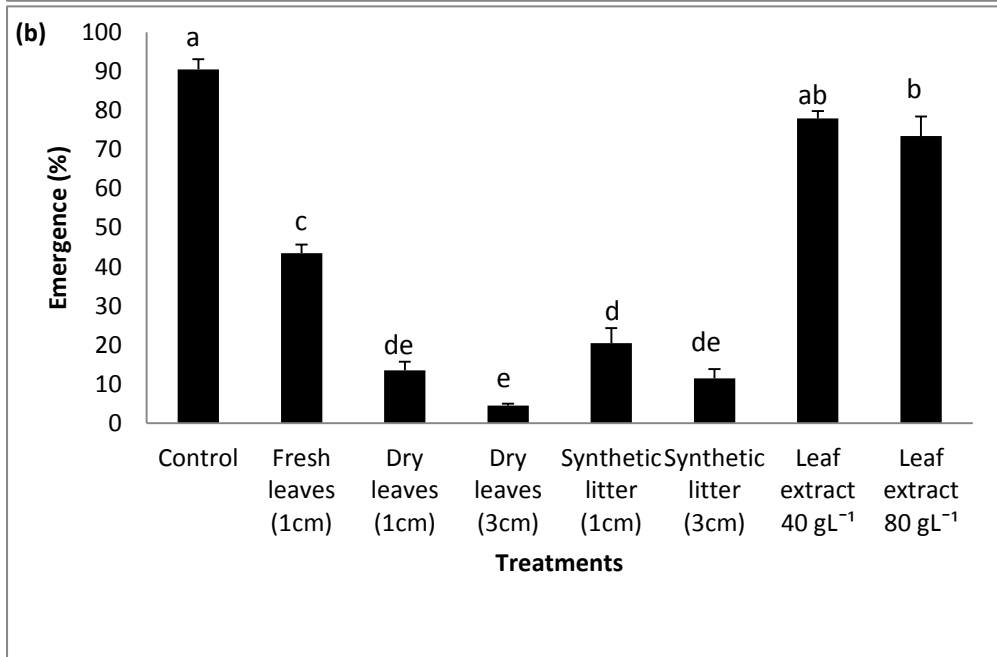
560

561 Fig. 2. Effects of leaf extracts of *A. holosericea* on shoot lengths and root lengths of (a) *C.*
 562 *pedicellatus* (b) *C. polystachios*. Values are mean \pm s.e. (n=4). Different letters indicate
 563 significant differences determined by Tukey's HSD within root or shoot values only.
 564 Significant differences of shoot length are denoted by lower case letters whereas significant
 565 differences of root length are denoted by upper case letters ($P < 0.05$).

566



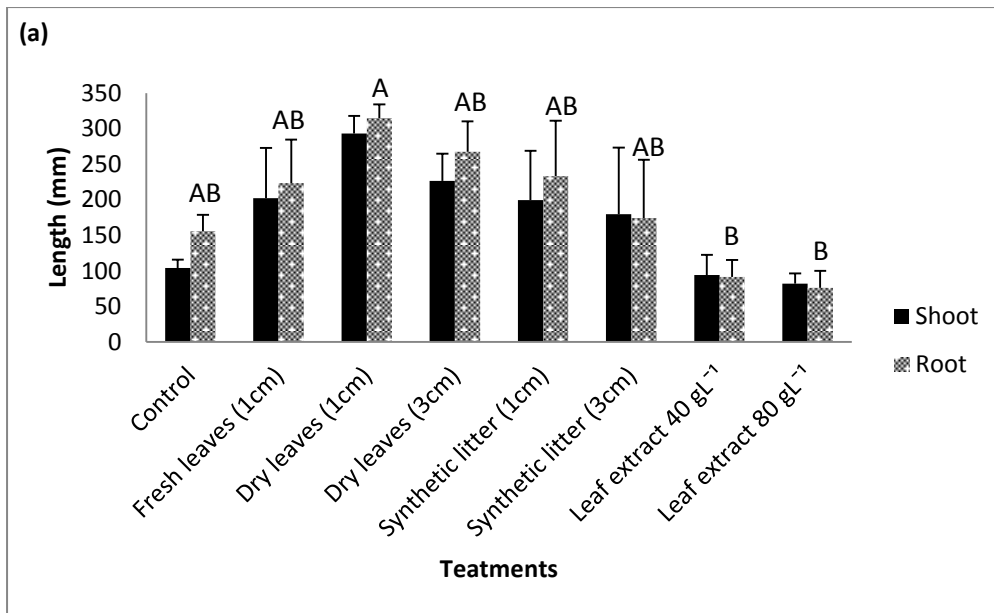
567



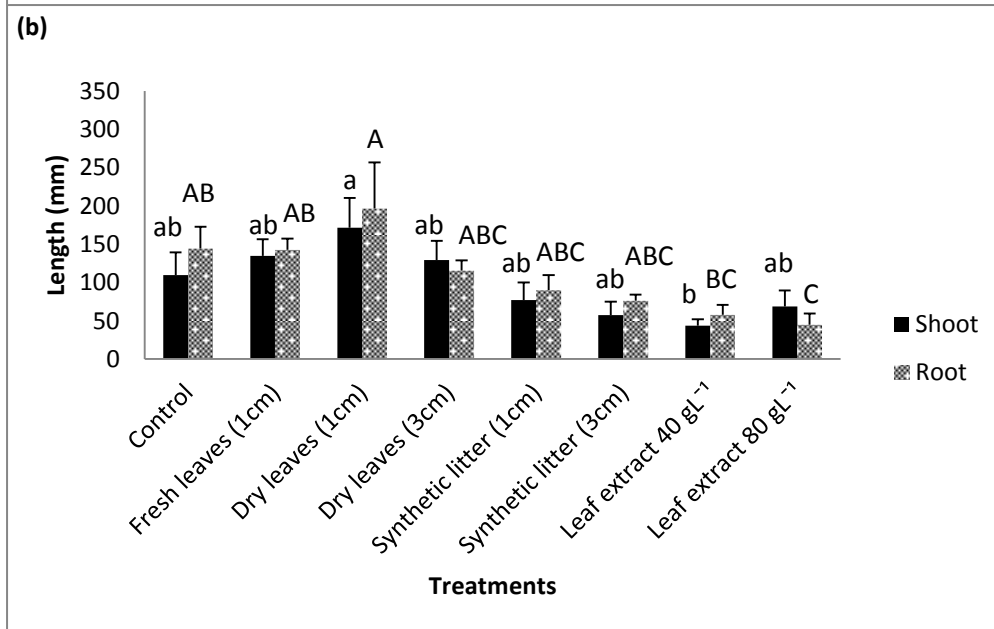
568

569 Fig. 3. Effects of different litter treatments and leaf extracts of *A. holosericea* on emergence
 570 of (a) *C. pedicellatus* (b) *C. polystachios*. Values are mean \pm s.e. (n=4). Different letters
 571 indicate significant differences determined by Tukey's HSD ($P < 0.05$).

572



573



574

575 Fig. 4. Effects of different litter treatments and leaf extracts of *A. holosericea* on shoot length
 576 and root length of (a) *C. pedicellatus* (b) *C. polystachios*. Values are mean \pm s.e. (n=4).
 577 Within either root or shoot values, bars that do not share the same letter are significantly
 578 different as determined by Tukey's HSD. Significant differences in shoot length are denoted
 579 by lower case letters and significant differences in root length are denoted by upper case
 580 letters ($P < 0.05$).