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Control of Late Leaf Spot of Groundnut (*Arachis hypogaea*) by Extracts from Non-Host Plant Species

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The effects of leaf extracts of 14 different non-host plant species on *in vitro* conidial germination of *Phaeoisariopsis personata*, the causal organism of late leaf spot (LLS) of groundnut were evaluated. Aqueous and ethanol leaf extracts of *Datura metel*, *Lawsonia inermis* and aqueous leaf extracts of *Sphaeranthus indicus* at 25%(w/v) concentration completely inhibited the conidial germination of *P. personata* both at 24h and 48h after incubation. Aqueous leaf extracts of *Blumea bifoliata*, *Eucalyptus globules*, *Ocimum sanctum* and *Pongamia pinnata*, and ethanol leaf extracts of *Azadirachta indica* and *S. indicus* inhibited the conidial germination by >90%. Aqueous and ethanol leaf extracts of *L. inermis* and *S. indicus* wrer highly inhibitory to conidial germination up to 1% concentration. Aqueous and ethanol leaf extracts of *D. metel* and ethanol leaf extract of *A. indica* were highly inhibitory to *P. personata* even at 0.01% (100 ppm) concentration. Ethanol leaf extract of *A. indica* up to 80°C, aqueous leaf extracts of *D. metel* and *S. indicus* up to 100°C, and *L. inermis* up to 60°C, were highly stable and retained their fungitoxic effects. Extract of *D. metel* was antifungal even after 180 days when it was stored both at room temperature and 4°C. Aqueous leaf extract of *D. metel* at 2% concentration effectively reduced the development of LLS by >60%, under greenhouse conditions both in prophylactic and simultaneous applications. Extracts of *D. metel* could be a potential economical and an eco-frendly alternative for control of late leaf spot, and its efficacy under field conditions is further being evaluated.

Keywords : antifungal activity, *Arachis hypogaea*, *Datura metel*, late leaf spot, *Phaeoisariopsis personata*.

Groundnut is an important legume crop in tropical and subtropical countries of the world and is used as a food, oil and cash source. Late leaf spot (LLS) caused by *Phaeoisariopsis personata* (Berk. & Curt.) v. Arx [= *Cercosporidium personatum* (Berk. & Curt.) Deighton] is an economically important foliar disease of groundnut wherever the crop is grown. LLS causes severe defoliation and reduces both haulm and pod yields by more than 50% (McDonald et al., 1985). The disease can be effectively managed by a combination of fungicides and host plant resistance (Pande et al., 2001). Increasing concerns about environmental hazards caused by excessive usage of fungicides, development of fungicide-tolerant pathogen stains, non-availability of both fungicides and their application technology to resource-poor farmers, necessitates the development of more economical and eco-friendly alternative components of disease management.

Plants produce several secondary metabolite compounds including alkanoids, cyanogenic glycosides, glucosinolates, flavanoids, saponins, steroids and terpenoids to protect themselves from the continuous attack of naturally occurring pathogens, insect pests and environmental stresses (Ebel, 1986). These compounds with antimicrobial activity may be specific against a particular pathogen or may have a broad spectrum and can be used for control of fungal disease in crop plants. The activity of these compounds also depends on the method and solvent used for extraction, its concentration and structure. With advances in analytical instrumentation, bioassay techniques and recombinant DNA technology, the scope of using these antimicrobial compounds for disease control was further enhanced. The use of plant extracts with antifungal activity offers an economical, safe and easily available alternative method for the management of LLS in groundnuts. In the recent past, several plant species have been screened for antifungal activity and extracts/purified compounds from these plants were found to have a broad spectrum of antimicrobial activity (Grayer and Harborne, 1994) and control pre-harvest (Tewari, 1995) and post-harvest diseases of several plant species (Mishra and Dubey, 1994). These reports drew our attention to develop plant-based products for control of LLS which may be beneficial to resource-poor groundnut farmers.

In our quest to identify eco-friendly and economical components and to use them in the integrated management of LLS, we initiated investigations with leaf extracts of 14

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non-host plant species of *P. personata*. The main purpose of this study was to evaluate leaf extracts of the selected 14 non-host plant species for their *in vitro* antifungal activity against *P. personata* and to evaluate the potent extracts for the disease control in a greenhouse. Thermal stability and longevity of promising extracts were also determined.

Materials and Methods

Plant extracts. Aqueous and ethanol leaf extracts of 14 different non-host plants species of *P. personata*, *Azadirachta indica* A. Jurs, *Blumea bifoliata* DC., *Calotropis procera* Ait. R. Br., *Datura metel* L., *Eucalyptus globules* Labill, *Lantana camara* L., *Lawsonia inermis* L., *Ocimum sanctum* L., *Parthenium hysterophorus* L., *Pongamia pinnata* L., *Sphaeranthus indicus* L., *Tridax procumbens* L., *Vernonia albicans* DC. and *Vitex negundo* L., were evaluated for their antifunagl activity against *P. personata*.

Preparation of leaf extracts. Young leaves from healthy plants of each plant species were collected, washed thoroughly under running tap water followed by sterile distilled water (SDW). Aqueous leaf extracts (ALE) were prepared by homogenizing 25 g of leaves in 100 ml of SDW using a warning blender. The homogenized solutions were filtered twice through a cheese cloth and centrifuged at 5,000 rpm for 10 min at 4°C. The supernatant collected was treated as 25%(w/v) ALE. Ethanol leaf extracts (ELE) were prepared by homogenizing 25 g of leaves in 100 ml of 95% ethanol, homogenized solution was filtered twice through a cheese cloth and centrifuged at 5,000 rpm for 10 min at 4°C. The supernatant was subjected to vacuum evaporation at 50 and the residue was dissolved in 100 ml of 5% ethanol to obtain 25%(w/v) ELEs. Further dilutions of ALEs and ELEs were made as per requirement with SDW and 5% ethanol respectively.

Fungal strain. Conidia of *P. personata* were collected from infected leaves from an experimental field at International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru and pure culture was maintained in laboratory on detached leaves of susceptible groundnut variety TMV2 (Wadia and Butler, 1994).

Preparation of *P. personata* inoculum. Fresh conidia of *P. personata* harvested with a cyclone spore collector (Gast Manufacturing Corporation, USA) were equally suspended in SDW containing a few drops (0.01%) of Tween 20 (polyoxyeythlene sorbitan minolaurate). The concentration of conidia was measured and adjusted with SDW by observing them under light microscope using a haemocytometer. Conidial suspension containing 50,000 conidia/ml for *in vitro* antifungal assay and 20,000 conidia/ml for greenhouse experiments were used.

***In vitro* antifungal assay.** To assay the antifungal activity of the leaf extracts, 50 µl of each conidial suspension and leaf extracts were mixed well on a cavity slide. Each well of the cavity slide was considered as one replication and three replications were maintained throughout the experiments. Conidial suspension mixed with an equal volume of SDW and 5% ethanol served as controls for aqueous and ethanol leaf extracts, respectively. All slides were kept in humid chambers prepared by lining 90 mm diameter petri dishes with wet blotting paper, and incubated in dark at 24 ± 1°C. The slides were directly observed under light microscope for conidial germination at 24 h and 48 h after incubation. Immediately after incubation, a drop of lactophenol-cotton blue was added to each well to prevent further germination of the conidia. Two hundred conidia were observed in each replication and the number of germinated conidia was counted. Percentage inhibition of conidial germination in each treatment was calculated from the formula, percentage inhibition = {(number of conidia germinated in control – number of conidia germinated in treatment)/number of conidia germinated in control} × 100.

Thermal stability of leaf extracts. ALEs of *D. metel*, *L. inermis* and *S. indicus*, and ELE of *A. indica*, at 2% concentration were highly antagonistic against condial germination of *P. personata* and were evaluated further for their thermal stability and longevity. Though the ELEs of *D. metel*, *L. inermis* and *S. indicus* were also highly antagonistic to *P. personata* at the same concentration, thermal stability and longevity of these extracts were not evaluated, since use of ALEs rather than ELEs will be more convenient for use at farmer's field. To determine the thermal stability, 1 ml aliquots of 2% leaf extracts were exposed to various temperatures for 30 min by incubation them in a water bath at temperatures ranging from 30 to 100°C with an increment of 10°C and the heat treated extracts were evaluated for fungitoxicity against *P. personata*.

Longevity of leaf extracts. Longevity of ALEs of *D. metel*, *L. inermis* and *S. indicus*, and ELE of *A. indica*, both at room temperature and 4°C was determined. To determine the longevity of these extracts, 1 ml aliquots of 2% extracts were stored in glass vials, one set at room temperature and another at 4°C. Three vials of each extract were taken out periodically at different time intervals up to 180 days and evaluated for inhibition of conidial germination. Each vial was considered as one replication.

Control of LLS under greenhouse environment. Susceptible groundnut variety TMV2 was used for greenhouse studies. Thirty-day-old seedlings grown in 15 cm diameter plastic pots filled with a mixture of red soil (alfisols), sand and farm yard manure in 3 : 1 : 1 ratio under greenhouse environment were used for artificial inoculation. Five seeds were planted in each pot and thinned to four before inoculation. ALE of *D. metel* and *L. inermis*, and ELE of *A. indica* at 2% (w/v) concentration were tesed for control of LLS by applying them as a foliar spray on to the plants in two different treatments. 24h before the pathogen inoculation and simultaneously along with the pathogeh. Control pots were maintained for both the treatments by spraying the plants with SDW. The plants were evenly sprayed with conidial suspension using a hand sprayer. Immediately after inoculation, the plants were shifted to a dew chamber (Clifford, 1973) to maintain leaf wetness during nights. The plants were removed from the dew chamber on the morning of the following day and returned to greenhouse to provide dry period. This alternate wet (16h) and dry (8h) period treatments were repeated up to 10 days after inoculation (DAI) (Butler et al., 1994), and then the pots were left in the greenhouse untill end of the experiment. Temperature of 24 ± 2°C was maintained both in greenhouse and in dew chamber during the entire period of experimentation. Though the leaf extract of *S.*

indicus was highly antagonistic against *P. personata* conidaial germination, it was not tested under greenhouse conditions since it was not available at the time of experimentation. Five pots were considered as one replication and triplicates were maintained for each treatment and the experiment was repeated once.

Disease scoring. Severity of LLS was measured as a) lesion frequency (number of lesions/cm^2 leaf area) at 15 DAI and b) disease severity score on 1-9 rating scale (1 = no disease, and 9 => 80% disease) at 20 and 30 DAI. In each plant, third and fourth quadrifoliate leaves from the top were tagged before pathogen inoculation and these leaves were used to measure the lesion frequency.

Statistical analysis. All the experiments were conducted in triplicates and repeated twice. The experiments were conducted in a randomized complete block design and the data were subjected to analysis of variance (ANOVA) and the mean values in each treatment were compared using least significant differences at 1% level of significance ($P = 0.01$)

Results and Discussion

***In vitro* antifungal activity of leaf extracts.** Among the ALEs tested, except *P. hysterophorus* and *T. procumbens*, all the ELEs tested at 25% concentration significantly inhibited *P. personata* conidial germination both at 24 h and 48 h after incubation (Table 1). All the plant species selected for this study were earlier reported to have antifungal activity against one or more phytopathogenic fungi and in the present study they were observed to be antifungal even to *P. personata*. ALEs of *D. metel, L. inermis* and *S. indicus*, and ELEs of *D. metel* and *L. inermis* at 25% concentration completely inhibited conidial germination of *P. personata*. At the same concentration, ALEs of *B. bifoliata, E. globules, O. sanctum* and *P. pinnata*, and ELEs of *A. indica* and *S. indicus* inhibited *P. personata* conidial germination by >90% up to 24 h after incubation (Table 1).

Table 1. Percentage inhibition of *Phaeoisariopsis personata* conidial germination *in vitro* by 25%(w/v) leaf extracts of fourteen non-host plant species. Conidia of *P. personata* were taken on to a cavity slide and incubated in presence of leaf extracts at 24±1°C in dark and then were observed for germination

Plant species	Aqueous leaf extracts		Ethanol leaf extracts	
	24h[a]	48h	24h	48h
Azadirachta indica	88.2*	76.1*	97.5*	87.5*
Blume bifoliata	96.2*	93.9*	86.4*	87.0*
Calotropis procrera	83.4*	81.7*	55.3*	29.4*
Datura metel	100.0*[c]	100.0*	100.0*	100.0*
Eucalyptus globules	91.5*	84.0*	88.2*	86.5*
Lantana camara	87.5*	80.7*	69.4*	64.8*
Lawsonia inermis	100.0*	100.0*	100.0*	100.0*
Ocimum sanctum	92.5*	73.9*	74.8*	68.7*
Parthenium hysterophorus	22.4*	5.8	88.4*	86.5*
Pongamia pinnata	90.6*	86.8*	72.6*	54.1*
Sphaeranthus indicus	100.0*	100.0*	91.4*	75.9*
Tridax procumbens	9.9	5.1*	41.8*	43.7*
Vernonia albicans	75.5*	65.5*	80.7*	69.6*
Vitex negundo	86.1*	82.7*	61.0*	49.1*
Control	0.0	0.0	0.0	0.0
LSD (p=0.01)	10.4	6.2	20.1	22.8

[a] Conidia were observed of germination at 24 h and 48 h after incubation.

[c] Mean values are significantly higher than control at P=0.01. All the mean values presented are the mean of three replications in two sets of experiments

To identify plant species with potent antifungal activity against *P. personata*, leaf extracts which are inhibitory to *P. personata* conidial germination by >90% at 25% concentration were further evaluated at lower concentrations up to 0.01%. ALEs of *B. bifoliata, E. globules, O. sanctum* and *P. pinnata* lost most of their antifungal activity at 2% and lower concentrations, and hence were not used in further experiments. ALEs *of L. inermis* and *S. indicus* up to 1% concentration, and *D. metel* up to 0.01% concentration were highly effective against *P. personata* (Table 2). Among ELEs, *L. inermis* and *S. indicus* up to 1%, *A. indica* and *D. metel* up to 0.01% concentration were highly antagonistic against *P. personata* conidial germination (Table 3). The antifungal activity of these extracts may be attributed to the presence of active principles such as alkaloids, steroids, terpenes, tannins and phenols, The antimicrobial activity of the four plant species, *A. indica, D. metel, L. inermis* and *S. indicus* was well known. ELEs of *A. indica* at a concentration of 1000 µg/ml was highly inhibitory to the growth of *Alternaria brassicola, Colletotrichum capsici, Fusarium*

Table 2. Percentage inhibition of *Phaeoisariopsis personata* conidial germination *in vitro* by aqueous leaf extracts of selected non-host plant species. Conidia of *P. personata* were taken on to a cavity slide, incubated in presence of leaf extracts at 24±1°C for 24h and then observed for germination under a light microscope

Plant species	Concentration (w/v) of aqueous leaf extracts					
	10%	5%	2%	1%	0.1%	0.01%
Blume bifoliata	69.7*	37.8*	9.3*	2.2	0.0	0.0
Datura metel	100.0*	100.0*	99.2*	98.9*	97.6*	93.9*
Eucalyptus globules	63.2*	44.4*	31.8*	14.3*	0.0	0.0
Lawsonia inermis	98.9*	97.8*	92.6*	83.4*	19.8*	0.0
Ocimum sanctum	74.9*	31.8*	18.9*	9.8*	0.0	0.0
Pongamia pinnata	68.9*	39.8*	8.9*	0.0	0.0	0.0
Sphaeranthus indicus	98.9*	97.7*	92.8*	84.9*	46.8*	18.9*
Control	0.0	0.0	0.0	0.0	0.0	0.0
LSD (p= 0.01)	5.2	26.8	5.7	4.0	3.1	1.8

* Mean values are significantly higher than control at P=0.01. All the mean values are the mean of three replications in two sets of experiments.

Table 3. Percentage inhibition of *Phaeoisariopsis personata* conidial germination *in vitro* by aqueous leaf extracts of selected non-host plant species. Conidia of *P. personata* were taken on to a cavity slide, incubated in presence of leaf extracts at 24±1°C for 24h and then observed for germination under a light microscope

Plant species	Concentration (w/v) of ethanol leaf extracts					
	10%	5%	2%	1%	0.1%	0.01%
Azadirachta indica	100.0*	98.9*a	99.5*	97.0*	84.5*	68.6*
Datura metel	100.0*	100.0*a	100.0*	99.4*	99.5*	98.4*
Lawsonia inermis	100.0*	99.2*a	86.8*	68.4*	19.8*	0.0
Sphaeranthus indicus	86.8*	72.8*a	32.3*	49.2*	0.0	0.0
Control	0.0	0.0	0.0	0.0	0.0	0.0
LSD (p=0.01)	3.9	3.5	3.4	14.7	4.0	3.9

*Mean values are significantly higher than control at P=0.01. All the mean values are the mean of three replications in two sets of experiments.

oxysporum, *Rhizoctonia solani* and *Sclerotinia sclerotiorum* (Shivpuri et al., 1997). Two limnoids and polar extract derived through solvent partitioning from *A. indica* were inhibitory to *Puccinia arachidis*, causal agent of rust in groundnut and reduced the pustule numbers and disease severity (Suresh et al., 1997). *Datura metel* completely inhibited *in vitro* conidial germination of *C. capsici* (Gomathi and Kannabiran, 2000). Kurucheve et al. (1997) reported that extracts of *L. inermis* were inhibitory to *R. solani* and Bambawale et al. (1995) reported that ethanol extracts of *L. inermis* were effective in control of the cotton pathogens *Alternaria macrospora*, *Myrothecium roridum* and *Xanthomonas compestris* in *in vitro* tests (Bambawale et al., 1995). Essential oil (Garg and Kasera, 1983) and alcoholic extracts of *S. indicus* flowers (Shaikh et al., 1986) were observed to be antibacterial. ALE of *D. metel* and ELEs of *A. indica* and *D. metel* offers a great hope for their use in control of LLS, since they are highly effective against *P. personata* even at 0.01% concentration.

Thermal stability of leaf extracts. The high antifungal activity of extracts of *A. indicia*, *D. metel*, *L. inermis* and *S. indicus* even at lower concentrations (Tables 2 and 3) prompted us to examine the thermal stability and longevity of these extracts. ALEs of *D. metel* and *S. indicus* were stable and retained their antifungal activity against *P. personata* up to 100°C. ALE of *L. inermis* up to 60°C, ELE of *A. indica* up to 80°C were highly stable and the antifungal activity of these extracts decreased with further increase in temperature (Fig. 1). This could be due to the denaturation of the active principle present in *A. indica* and *L. inermis* extracts at higher temperatures. This is in contrast to the earlier reports that the fungitoxicity of *L. inermis* bark extracts against ring worm fungi was unaltered even after autoclaving and storage for longer periods (Singh and Pandey, 1989).

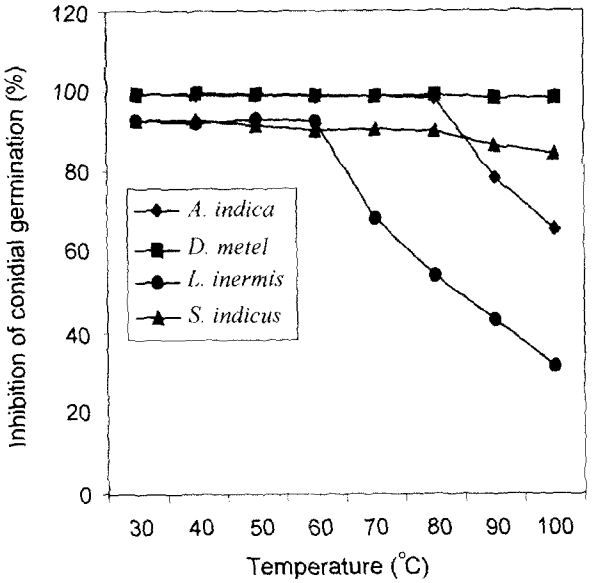

Fig. 1. Thermal stability of 2% ethanol leaf extract of *Azadirachta indica* and aqueous leaf extracts of *Datura metel*, *Lawsonia inermis* and *Sphaeranthus indicus*. The leaf extracts were subjected to temperatures from 30-100°C for 30 min and then were tested for their inhibitory activity on *in vitro* conidial germination of *Phaeoisariopsis personata*. All the mean values presented are the mean of three replications in two sets of experiments.

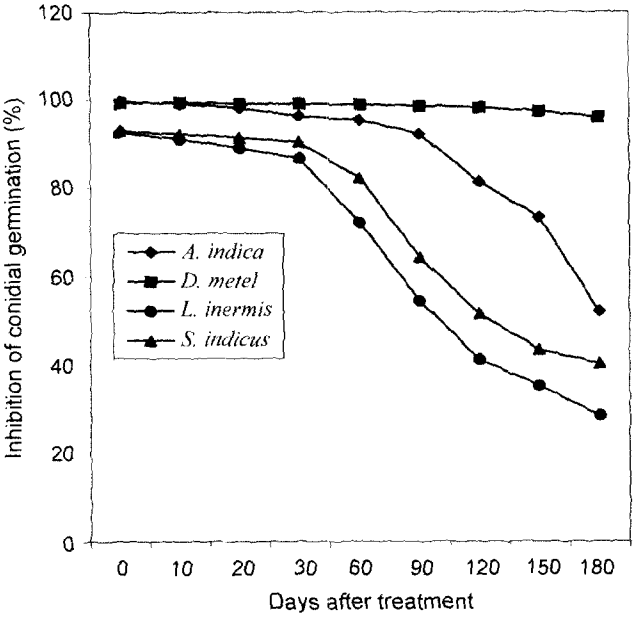

Fig. 2. Longevity of ethanol leaf extracts of *Azadirachta indica*, and aqueous leaf extracts of *Datura metel*, *Lawsonia inermis* and *Sphaeranthus indicus*. Aliquots of leaf extract were stored at room temperature (28°C) and were evaluated for their inhibitory activity on in vitro conidial germination of *Phaeoisariopsis personata* at different time intervals. All the mean values presented are the mean of three replications in two sets of experiments.

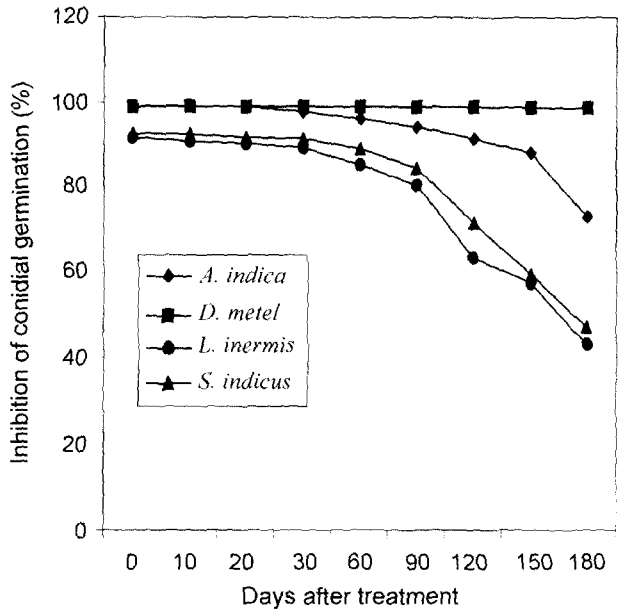

Fig. 3. Longevity of ethanol leaf extracts of *Azadirachta indica*, and aqueous leaf extracts of *Datura metel*, *Lawsonia inermis* and *Sphaeranthus indicus*. Aliquots of leaf extract were stored at 4°C and were evaluated for their inhibitory activity on *in vitro* conidial germination of *Phaeoisariopsis personata* at different time intervals. All the mean values presented are the mean of three replications in two sets of experiments.

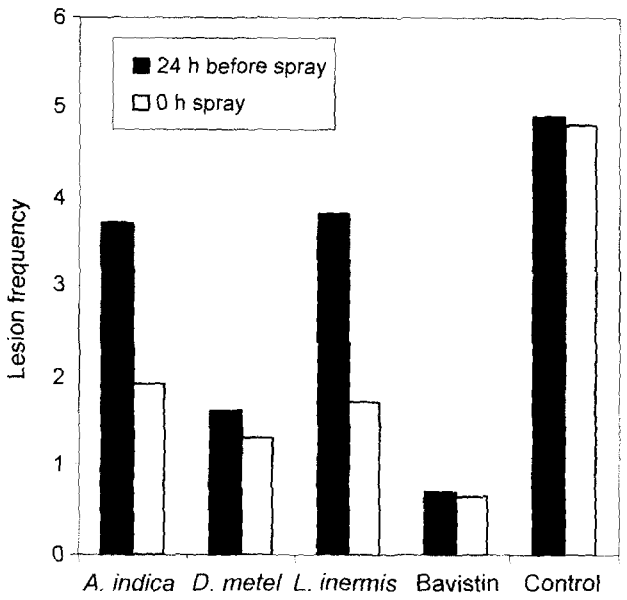

Fig. 4. Effect of ethanol leaf extract of *Azadirachta indica*, and aqueous leaf extracts of *Datura metel* and *Lawsonia inermis* on development of late leaf spot under greenhouse conditions. Leaf extracts of 2% concentrations were used as a foliar spray on to the groundnut plants at 24 h and 0 h before pathgen inoculation and the lesion frequency (number of lesions/cm^2 leaf area) was measured at 15 days after pathogen inoculation. All the mean values presented are the mean of three replications in two sets of experiments.

Longevity of leaf extracts. When stored at room temperature, ALE of *D. metel* up to 180 days, *L. inermis* and *S. indicus* up to 60 days, and ELE of *A. indica* up to 90 days retained their high antifungal activity (Fig. 2). When stored at 4°C, ALE of *D. metel* up to 180 days, *L. inermis* and *S. indicus* up to 90 days, and ELE of *A. indica* up to 150 days retained their antifungal activity to higher significant levels (Fig. 3).

Control of LLS under greenhouse environment. ALE of *D. metel* and *L. inermis* and ELE of *A. indica* were effective in reducing the severity of LLS under greenhouse conditions when applied as a foliar spray at the same time as pathogen inoculation. ALE of *D. metel* was effective in controlling LLS both at simultaneous and 24 h before pathogen inoculation. Reduction in disease severity was >60% when compared with control in both the components measured – lesion frequency and disease severity score on 1-9 rating scale (Fig. 4 and 5). Disease protection offered by *D. metel* extract was comparable with the commercial fungicide Bavistin. Earlier, a few attempts have been made by various research groups to control LLS by using extracts from different plant species. ALEs of *A. indica* and *L. inermis* were found to be effective in controlling both LLS and rust diseases of groundnut, and increase the pod yield by 15-40% under field conditions (Ghewande, 1989). Foliar spray of seed kernel extract of *A. indica* alone (Usman et al., 1991), and along with potash significantly reduced the incidence of LLS (Chandrasekar et al., 1994). A small reduction of LLS was obtained by foliar spray of 4% aqueous leaf extract of *A. indica* (Adiver et al., 1995). Neem oil from *A. indica* and leaf extract from *Nerium odourm* reduced the incidence of LLS (Ganapathy and Narayanasamy, 1990). Foliar spray of 5% leaf extracts of *C. procera* at 70 days after sowing proved to be highly significant in reducing the incidence of both early and late spot diseases and increase the yield of groundnut (Srinivas et al., 1997). Though, the reductions in disease severity and increase in yield obtained in these experiments were significantly higher when compared with unsprayed controls, they were far less than that obtained with the use of chemical fungicides.

Extract of *D. metel* was effective in controlling LLS both in prophylactic and simultaneous applications and the disease protection obtained was greater when compared with *A. indica* and *L. inermis* extracts with were earlier reported to have disease control under field conditions. Extract of *D. metel* reduced the severity of LLS up to 65% in both the measured components, lesion frequency and disease rating on 1-9 scale. Hence, it can be further evaluated under field conditions for control of LLS to minimize the usage of fungicides. In a similar study, prophylactic spray of crude

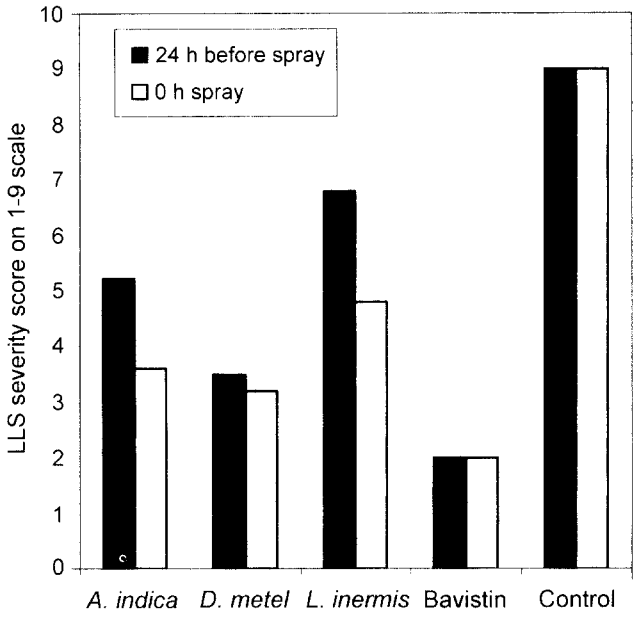

Fig. 5. Effect of ethanol leaf extract of *Azadirachta indica*, and aqueous leaf extracts of *Datura metel* and *Lawsonia inermis* on development of late leaf spot under greenhouse conditions. Leaf extracts of 2% concentrations were used as a foliar spray on to the groundnut plants at 24 h and 0 h before pathgen inoculation and the disease severity score on 1-9 rating scale was measured at 30 days after pathogen inoculation. All the mean values presented are the mean of three replications in two sets of experiments.

extract of bulbs of *Allium sativum* controlled ergot of sorghum by 98-100% under greenhouse conditions (Singh and Navi, 2000). The disease control obtained by prophylactic application of *D. metel* extract could be due to the stable persistence of its antifungal compound(s) present in it, on groundnut leaf surface. Also there is a possibility that extract of *D. metel* contain a signal molecule which activate a series of defense mechanisms – induction of phytoalexins and other antimicorbial compounds, or induction of systemic resistance in groundnut before pathogen infection takes place. Earlier, it was reported that extracts from *Reynoutria sachalenensis* reduced the incidence of powdery mildew in cucumber and induced the synthesis of phenolic compounds p-coumaric, caffeic and ferulic acids and p-coumaric acid methyl ester (Daayf et al., 2000). *Reynoutria sachalenensis* also increased the activities of chitinase, β-1,3-glucanase, peroxidase, polyphenol oxidase and phenyl alanine ammonia lyase in tobacco and cucumber leaves (Schneider and Ullrich, 1994).

In the present study, leaf extract of *D. metel* was found to have potent antagonistic activity against conidial germination of *P. personata* even at minute concentrations (100 ppm) and was stable at higher temperatures and for longer periods at room temperature. All these properties, coupled with its high efficacy to control LLS under greenhouse conditions suggests that it may be a reliable alternative for use of chemical fungicides. Under field conditions, the disease pressure and environmental conditions conducive for pathogen multiplication also determines the efficacy of *D. metel* extracts. The toxicity of groundnut haulm sprayed with *D. metel* extract when it is fed to animals is to be determined. However, the use of *D. metel* extract offers an economical and easily available alternative approach to fungicide usage for control of LLS. Further investigations on evaluation of *D. metel* extract for control of LLS under field conditions are under progress.

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