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Evaluation of botanicals for managing leaf rust of castor caused by Melampsora ricini

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Castor plant is severely infected by leaf rust disease caused by (Melampsora ricini) which is often responsible for drying of the leaves. The use of botanicals is cost effective and environmentally safe and is potentially useful for resource poor small-scale farmers as it is locally available. The present study was undertaken to investigate the potential efficacy of various botanicals for the control of the disease. The present study was conducted with the objectives to evaluate the efficacy of plant extract against leaf rust of diseases (Melampsora ricini). Botanicals were tested against rust disease on castor plants in greenhouse and field conditions at Melkassa Agricultural Research Center (MARC). The treatments were arranged with Completely Randomized Design (CRD) for greenhouse and Randomized Complete Block Design (RCBD) in the field experiments in three replications. Significant differences were observed among botanicals in the green house as well as field experiments. In green house, next to mancozeb maximum disease reduction (52 %), fresh leaf weight (40gm) and dry leaf weight (8gm) were recorded from neem (Azadirachta indica). In field trial, among considered botanicals, minimum incidence (26.36%) and disease index (29.45%) were recorded from neem which was at par with garlic (33.33%), pyrethrum (40.56 %), lantana weed (47.22%) and mancozeb. Findings illustrated that among the tested botanicals neem was found effective for managing disease of leaf rust. Thus, it seems to be effective for the management of leaf rust and has the potential to be a vital component of integrated management for leaf rust of castor (Melampsora ricini). Therefore, it could be concluded that plant extracts (neem) may be useful to control leaf rust disease as a safe alternative option to chemical fungicides.

Key words: plant extracts, rust disease, castor plant

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INTRODUCTION

Castor *(Ricinus communis L.)* is the major feed plant for the production of eri silkworm, the one among the commercially exploited silkworm species which produce eri silk. Castor serves as primary food plant however, feeding of infected leaf has been found to adversely affect the growth and development of the silkworm, cocoon yield and silk quality (Joshi, 1992). Lakra and Saharan 1989 reported castor is known to be attacked by

many pathogens which include mostly fungi, bacteria and nematodes. However, only few pathogens can cause diseases of economic importance, at different crop growth stages, depending upon the seasonal conditions. Rust *(Melampsora ricini)* is recorded as important diseases on castor which causes substantial yield losses. Rust diseases are adversely affects the quality of the leaves and 89.9 % yield losses has been recorded by rust disease of castor. Abiy, *et al.,* 2014 *reported* rust *(Melampsora ricini)* is recorded as important diseases on castor and it causes 35-41 % of yield losses and it is a serious disease of castor inflicting considerable qualitative as well as quantitative losses.

The use of chemicals against disease outbreak has been one of common practices. However, problems associated with the use of hazardous chemicals for plant disease control causes health hazards, toxicity to silkworm, environmental pollution, pathogens' resistant to chemical (Meena P.D. *et al.,* 2008). Botanical extracts have been reported to be effective against diseases of many crops (Chattopadhyay, C., *et al.,* 2005). These are natural products that can even be used in their crude forms. They are far low toxic to non-target organisms, biodegradable and environmentally safe . Research on botanical control of plant pathogens has received much attention in recent years as a means of increasing crop production by avoiding a number of problems related to chemical control. Among different strategies for plant growth promotion and disease suppression, botanical control approaches are very useful and an ominous choice in managing plant diseases (Mesta, R.K., *et al.,* 2009). Therefore, there is a need to explore the possibility of using eco-friendly and environmentally safer formulations such as plant extracts which can fit into Integrated Disease Management (IDM) programme. Hence, this study is conducted to generate information on the efficacy of different plant extracts against rust diseases ***(Melampsora ricini)*** by testing the effects in greenhouse and field conditions.

MATERIALS AND METHODS

Description of the study area

The experiment was conducted at Melkassa Agricultural Research Center (MARC) from 2006-2010 E.C. MARC is found 117 kms away from Addis Ababa and 17 km to South-East of Adama in the East-Shewa Zone of Oromia Region. It is located 8°24'N latitude and 39°12'E longitude at elevation of 1500 meters above sea level and a mean annual rainfall of 770 mm.

Planting of castor seeds in the green house

A mixture of sterilized sandy clay loam soil, decomposed animal dung and sand (3:2:1 ratio) was autoclaved at 121°C for 2 h and filled into plastic pots (20 cm ×15 cm). Susceptible variety of castor (Hiruy) was used for the study. Two Seeds of castor per pot was planted on sterile soil in plastic trays containing 1200 cc of sterilized soil with 1:2:3 proportions of sand, compost and clay, respectively in greenhouse and kept on benches. The plants were allowed to grow for 6 weeks in a greenhouse while maintaining the soil field capacity at 26 ± 2°C and 50 to 60 % relative humidity.

Preparation *of plant extracts*

Leaves, seeds, flower and bulb of 8 plants, namely Neem (*Azadirachataindica),*Garlic (*Allium sativum*), Pyrethrum *(Pyrethrum Parthenium),*Lantana weed *(Lantana camara L.*) , Australian Fever Tree (*Eucalyptus globules), Stinking Roger* (*Tagetes minuta),* Congress weed (*Parthenium hysterophorus L)* and *Jatropha* (*Jatropha curcus)(Table 1) were* collected from different parts of the country and test for their efficacy in reducing the mycelia growth of *in vitro* using , the poisoned food technique *(Schmitz* 1930).

They were sun dried for 3 days then crude extract of each plant was prepared as a solvent. The extraction technique is used as a modification of Ruch and Worf (2001) method. One hundred and fifty gram of the dried material of each plant was soaked in 500 ml of water with constant stirring for 30 min and then maintained at room temperature for 24 h before being filtered. The soaked plant material was filtered with the help of a very fine and clean piece of cheese cloth separately for each plant. The filtrates were preserved in glass bottles in a refrigerator at 4°C for further use. The plant extract will be further diluted to have 20 % concentration.

Isolation and preparation of spore suspension of *a* leaf rust disease (*Melampsora ricini)*

The diseased leaves having distinct symptoms *with Melampsora ricini* was collected from the castor farm at MARC, in sterilized polythene bags and brought to Crop Protection Laboratory, MARC for culturing, isolation and identification. The diseased leaf having distinct leaf spot symptoms was collected and the identification of causal pathogen was observed with microscope.

For preparation of inoculum (spore suspension), sterile distilled water was added and dislodging spores gently with a sterile glass rod. The suspension was subsequently filtered through 4 layers of cheese cloth in order to remove the mycelia and the concentration of spores is determined by means of haemocytometer. The concentration of spores was adjusted to $1x\ 10^5$ spores per ml with sterile distilled water prior to the inoculation of castor leaves.

Table 1. Plant species screened in the experiment

Botanical name (Plant Species)	Common name	Family	Part to be used
Azadirachta indica	Neem	*Meliaceae*	Seed
Allium sativum	*Garlic*	*Liliaceae*	Bulb
Pyrethrum parthenium	*Phyrethrum*	*Compositae*	Flower
Lantana camara L.	Lantana Weed	*Verbenaceae*	Leaf
Eucalyptus globules	Australian Fever Tree	*Myrtaceae*	Leaf
Tagetes minuta	*Stinking Roger*	Asteraceae	Leaf
Parthenium hysterophorus L	Congress weed	Asteraceae	Leaf
Jatropha curcus	*Jatropha*	Euphorbiaceae	Seed

Experimental design and Treatments application

A total of 10 treatments were used, namely; crude extracts of Neem (*Azadirachata indica),* Garlic (*Allium sativum*), Pyrethrum *(Pyrethrum Parthenium)*, Lantana weed *(Lantana camara L.*), Australian Fever Tree (*Eucalyptus globules), Stinking roger* (*Tagetes minuta),* Congress weed **(***Parthenium hysterophorus L)***,** and *Jatropha* (*Jatropha curcus)* were evaluated in greenhouse conditions. There were also two controls. Treated control/ standard check (mancozeb) and untreated control (neither rust nor the botanicals was applied. The spore suspension of the pathogen containing 1x 10^5 spores/ml was sprayed on one and half months of castor seedlings. Then, the botanicals each at 20 % concentration was sprayed on infested plants after 72 h of pathogen inoculation and continued for two months at 15 day interval. The treatments were arranged in randomized complete block design (RCBD) with three replication.

Data to be collected

Estimation of disease severity is made by visual disease assessment key. Disease intensity was assessed using 0 – 4 points rating scale, where 0 = no disease (healthy); 1 = 1 to 25%, 2 = 26 to 50%; 3 = 51 to 75%; 4 = 76 to 100% leaf areas infected, using the following formula:

Where, PDC: percentage disease control, DC: disease in control, DT: disease in treated plants.
PDC= DC-DT/DC

***Field* evaluation of effective botanicals against leaf rust disease *(Melampsora ricini)* of castor plants**

Field preparation

Experiment was conducted in a field with a history of leaf rust disease (*Melampsora ricini)* at MARC; susceptible variety of castor (Hiruy) was planted in row to row and plant to plant distance of 75cm and 50cm, respectively in the field of sericulture for this experiment. The fertilizer, DAP and Urea were applied as recommended whilst, watering and hand weeding was done as recommended.

Experimental design and Treatments application

Only those effective plant extracts i.e., *Azadirachta indica* (Neem), *Allium sativum (garlic), Pyrethrum Parthenium (phyrethrum), Lantana camara L. (*Lantana Weed) during greenhouse experiment were further investigated under field condition. Untreated control and the standard chemical were applied in the field as in the green house. The experiment is laid down in a randomized complete block design (RCBD), with three replications. The treatments will be applied at an interval of 15 days starting from the first appearance of the disease. A guard row was kept in between the treatments to prevent the drift while spraying.

Data to be recorded

Percentage of disease incidence and severity were taken and calculated based on the following formulae

$$Incidence\ (I) = \frac{Number\ of\ affected\ plant\ units}{Total\ number\ of\ plant\ units (healthy\ and\ affected\ units\ assessed)} x100$$

$$Severity\quad (S) = \frac{Area\quad of\quad plant\quad tissues\quad affected\quad by\quad pest}{Total\quad area\quad of\quad plant\quad (tissue)} x100$$

Other agronomic data such as: plant height, production of leaves, fresh leaf weight and dry leaf weight were assessed

Data Analysis

The data were subjected to analysis of variance (ANOVA) using Statistical Analysis Software (Gomez and Gomez, 1984) (version 9.00, SAS, Institute Inc., Cary,

NC, USA). Treatment means were separated using the least significant differences (LSD).

RESULTS AND DISCUSSION

Efficacy of plant extracts under greenhouse condition

The effect of plant extracts against leaf rust infestation in castor was studied in green house. The extracts were applied as spray method during inoculation. The results of the experiment showed that all the eight botanical extracts revealed significant differences ($P<0.05$) as compared to the control (Table 2).

Among botanicals, minimum disease incidence and severity on leaves (18.7% and 24%) were observed in neem, which was at par with garlic (20% and 26%), Pyrethrum (23 % and 30%) and Lantana weed (21% and 27%), respectively. However, similar results of incidence was recorded from Australian fever tree (30%), stinking rogus (32 %), congress weed (33 %) and jatropha (31 %). The extracts able to reduce the severity of the disease ranged from 10 to 52 % (Table 3). The standard fungicide (Mancozeb) showed highest reduction in disease incidence (63%) and severity (64%). On this study applying a foliar spray showed high efficacy in reducing the leaf rust infection and neem extract was the most effective treatment as compared to botanical extracts and it reduced disease incidence and severity by 46.6 % and 52 % respectively (Table 3). The increasing serious problems of resistance and residue to pesticides and contamination of the biosphere associated with large-scale use of broad spectrum synthetic pesticides have led to the need for effective biodegradable pesticides with greater selectivity. This awareness has created a worldwide interest in the development of alternative strategies, including the discovery of newer pesticide (Dayan F.E*. et al.,* 2009).

There were significant differences ($P < 0.05$) among treatments in affecting, fresh and dry leaf weight as compared to treated control. Maximum fresh leaf weight (48 gm) and dry leaf weight (9.6gm) per plant were recorded from mancozeb. Among botanicals neem recorded highest fresh (40 gm) and dry leaf weight (8 gm) per plant. John and Hebs (2000) also reported that bare root dip of brinjal seedlings in neem leaf extract significantly reduced the gall index and increase the plant growth parameters significantly. Soil application of neem and caltropis leaf extract reduced the root knot nematode infestation in cowpea.

Neem, *Azadirachta indica*, is one of the most versatile trees with regard to germicidal activity against an array of microorganisms. Its leaves, bark, and seed kernels can be used in nematode management. Various compounds such as nimbin, nimbidin, azadirachtin, salannin, thionemon, and meliantriol occur in the seeds, leaves, and bark of neem in high concentrations and are responsible for the tree's antimicrobial and nematicidal activity. Powder from the seed kernels and leaves has been found to be suppressive against some diseases (Alam, 1993).

Sindhan and Hooda (2004), reported bulb extracts of *Allium sativum* (garlic) and *A. cepa* (onion) more effective than eucalyptus leaf extract for the control of white rust of castor. However, all plant extracts improved fresh and dry weight of leaves as compared as treated control. It is evident from several reports that plant extracts are effective bio control agents against a wide range of plant pathogens (Golob P. and I. Gudrups I., 1999).

Promising botanicals verified during greenhouse evaluation i.e neem, garlic, pyrethrum and lantana weed were selected and further evaluated under field condition.

Efficacy of plant extracts under field condition

For the field experiment, only those effective botanicals during greenhouse experiment were investigated. All the treatments were significantly superior to untreated control for rust severity on leaf (Table 4). All botanical extracts applied on rust infested plots significantly ($P \leq 0.05$) decreased the severity of rust disease over untreated control plots (Table 4).

Among considered botanicals, minimum incidence (26.36 %) and disease index (29.45 %) were recorded from neem. Minimum disease index on leaves (29.45 %) was observed in neem, which was at par with garlic (33.33%), pyrethrum (40.56 %), lantana weed (47.22 %) and mancozeb. The result indicates spore germination was affected by some of the plant extracts but complete inhibition was not achieved by any of them. The neem plant is the most well-known example and its various parts, namely, leaves, crushed seeds, powdered fruits, oil, and so forth, have been used to protect stored grain from infestation (Talukder F.A. *et al.,* 2004). The neem oil and kernel powder gave effective grain protection against stored grain insect pests like Sitophilus oryzae, Tribolium cataneum, Rhyzopertha dominica, and Callosobruchus chinensis at the rate of 1 to 2% kernel powder or oil (J. Pereira and R. Wohlgemuth, 1982). Yadava and Bhatnagar (1987) reported that a dried leaves of *Azadirachta indica* have been mixed with stored grains for protection against insects. Azadirachtin is an active principle from the neem plant, which is an effective grain protectant against insect infestation.

The present study has demonstrated the antifungal potential of plant extract that corresponded with the work done by Lakshmi BS *et al.,* 2010 who reported methanolic extract of *P. juliflora* leaves to have excellent in vitro antifungal activity against soil mycoflora. Consecutive sprays of plant extract at 15 days interval

Table 2. Mean effect of different botanicals on incidence and severity of rust diseases of castor under greenhouse condition in Melkassa Agricultural Research Center

Treatment	Fresh leaf weight(gm)	Dry leaf weight(gm)
Neem	40b	8b
Garlic	33c	7c
Pyrethrum	33c	7c
Lantana weed	30cd	6cd
Australian fever tree	20f	4f
Stinking rogus	25def	5def
Congress weed	27de	5.4de
Jatropha	25def	5def
Mancozeb	48a	9.6a
TC (Treated control)	24ef	4.8ef
LCD	**5**	**1**
CV	**9.8**	**10**

Table 3. Mean effect of different botanicals on fresh and dry leaf weight of castor under greenhouse condition in Melkassa Agricultural Research Center

Treatment	Disease incidence % (DI)	Reduction in Incidence % (RI	Severity %	Reduction in Severity % (RS)
Neem	18.7bc	46.6	24f	52
Garlic	20b	43	26ef	48
Pyrethrum	23b	34	30de	40
Lantana weed	21b	40	27ef	46
Australian fever tree	30a	14	45ab	10
Stinking rogus	32a	9	35cd	30
Congress weed	33a	6	43b	14
Jatropha	31a	11.4	40bc	20
Mancozeb	13c	63	18g	64
TC	35a	-	50a	-
LCD	**6.3**		**6**	
CV	**14.3**		**10.3**	

Table 4. Efficacy of different botanicals for the control of leaf rust disease *(Melampsora ricini)* on castor plant

Trt	INC%	Reduction over UC (%)	Percent disease Index % (PDI)	Reduction over UC (%)
Neem	26.363 cd	40	29.447bc	42
Garlic	30.197 bcd	25	33.333 abc	35.2
Pyrethrum	35.437bc	35.3	40.557abc	21.7
Lantana weed	41.260b	25.7	47.220ab	5.8
Mancozeb	20.6d	52	23.003 c	55.8
UC	55.190a	-	53.553 a	-
CV	**21**		**30**	
LSD	**13.45**		**21.096**	

Table 5. Mean effect of botanicals on leaf rust on agronomic and yield characters of castor

Trt	PH	NL	PB	SB	IL	LA	FLW	TFLW	DLW	TDLW	NCPP
Neem	265a	20.5a	5.2a	1.9a	8.1a	2813a	320a	8533a	70a	1867a	68a
Garlic	248ab	14.9a	6.5a	1.9a	7a	2563.5 a	265.7b	7081b	58b	1555b	59.7ab
Pyrethrum	250ab	20.5a	6.9a	0.43a	7.7a	2253.8 a	234.7b	6261b	47.7b	1273b	64a
Lantana camara	223b	16a	4.8a	0.7a	8.7a	2529.9a	243b	6480 b	50.7b	1360.7b	78a
Mancozeb	256ab	17.4 a	5.5a	1.7a	7.2a	2766.7a	330a	8800a	85a	2133a	56ab
UC	250ab	12a	5.4a	0.33a	7.3a	2160.9a	210c	5600c	38c	1013c	33b
CV	**7.7**	**35**	**36**	**25**	**18**	**22**	**33**	**33**	**31**	**35**	**27**
LSD	**34.9**	**10.9**	**3.86**	**2.5**	**2.5**	**1024.5**	**23**	**655**	**9**	**250**	**29.434**

Means within the same column with a common letter are not significantly different (P<0.05), the same letters in the same column means there is no significant difference, PH= Plant height, NL= Number of leaves, PB= Primary branch, SB= secondary branch, IL=Internode length, LA=Leaf area, FLW=Fresh leaf weight, TFLW= Total fresh leaf weight, DLW= Dry leaf weight, TDLW=Total dry leaf weight, NRPP=number of racemes per plant, NCPP= Number of capsule per plant

starting from the initiation of the disease have significantly reduced the rust disease significantly. Our finding was in conformity with the report of (Hussain *et al.,* 1992) who reported that leaf extract of *D. stramonium* reduced the development of the rust pustules on leaves of wheat. The standard fungicide (Mancozeb) showed the best efficacy compared to all botanical extracts this result was similar with green house experiment. According to Umar *et al.,* 2010 the use of botanicals is one of the alternative methods suggested by nematologist for nematode control. Botanicals such as Azadirachta, Eucalyptus, Chrommelina, Sida acuta and Targetis have been found to be effective in the control of nematodes in tomato, cowpea and eggplant fields. These botanicals not only control nematodes but also improve the soil productivity and crop yield by several folds.

There were no significant differences (P<0.05) in plant height, number of primary branches, stem thickness, number of leaves, leaf area as well as internode length. However, significant differences in fresh and dry leaf weight and number of capsule were observed between treatments. Highest fresh and dry leaf weight were recorded from mancozeb (330gm and 85 gm neem) an neem (320gm and 70 gm) respectively. Whilst, the least fresh and dry leaf weight were recorded from control (210 gm and 38 gm) respectively (Table 5). Javed *et al*., (2006) reported that neem is one of the most effective agent against root knot nematode on the roots of tomato, neem formulations have been applied against root knot nematodes development of the second stage juveniles (J2) as soil drenching . Fadzirayi *et al.,* (2010) stated that garlic disrupts nematode mobility, feeding and reproduction, the latter being a plausible explanation for the low reproduction rate.

CONCLUSION AND RECOMMENDATION

The results of the study revealed that botanicals have the potential to regulate the infestation of leaf rust of disease *(Melampsora ricini)* of castor by suppressing, growth of mycelia of fungi in green house and field conditions. Due to exorbitant cost of chemicals and the environmental hazards they cause, it is high time to modify the rust management options like botanicals which are safer and cost-effective. Findings illustrated that the tested botanicals were effective for managing *leaf rust, (Melampsora ricini).* However, Neem seed *(Azadirachta indica)* revealed more potential than other botanicals for managing rust. *Thus, it* seems to be effective for the management of leaf rust disease and has the potential to be a vital component of integrated pest management for rust diseases of castor plant *(Ricinus communis).*

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