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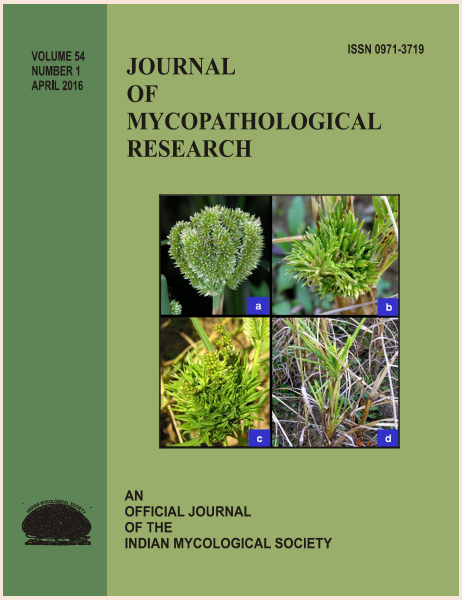

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Onion (*Allium cepa*) is a most popular as well as commercial vegetable crop grown in India. Purple blotch caused by *Alternaria porri* (Ellis) Cif. is one of the most destructive diseases which causes extensive damage to bulbs as well as seed crop. An *in vitro* experiment was undertaken to evaluate some plant extracts, bioagents and fungicides for managing the disease. The extracts of neem (*Azadirachta indica*), tulsi (*Ocimum sanctum*), eucalyptus (*Eucalyptus globus*), garlic (*Allium sativum*) and ginger (*Zingiber officinale*) were found to be the most effective in inhibiting the mycelial growth. Maximum inhibition was found in neem i.e. 90.4% and 82.6% followed by tulsi 86.8% and 78.4% at 20% and 10% concentration respectively. *Trichoderma harzianum* (88%) and *Trichoderma viride* (83%) were found most effective biocontrol agent against *Alternaria porri.* All the fungitoxicants inhibited the mycelial growth of *Alternaria porri*. Carbendazim + mancozeb and difenconazole showed complete inhibition (100%) followed by hexaconazole (94.2%), flusilazole (92.5%) and propiconazole (90.1%).

Key words: *Alternaria porri,* bioagents, fungicides, onion, plant extracts

INTRODUCTION

Onion (*Allium cepa* L.*)* is one of the oldest known and an important vegetable crop grown in India. It is commonly used for cooking purposes by almost all the people for its excellent taste to dishes and possess numerous therapeutic properties such as antibacterial, antifungal, antihelmintic, anti inflammatory, antiseptic and antispasmodic etc. Indian onions are famous for their pungency and are available round the year. India is the second largest producer of onion in the world after China. There is a lot of demand of Indian onion in the world, the country exported 1.48 MT of fresh onion accounting for Rs. 3,169.63 crores during the year 2013-14. Major export destinations (2013-14) are Bangladesh, Malaysia, Sri Lanka, United Arab Emirates, Indonesia, Pakistan and Singapore. The major onion producing states are Maharashtra, Karnataka, Madhya Pradesh, Gujarat, Bihar, Andhra Pradesh, Rajasthan, Haryana and Tamil Nadu. Maharashtra ranks first in onion production with a share of 27.72%. Being the eleventh highest producer of onion, Odisha produced 0.41 MT in 2012-13 (National Horticulture Board). Though there is huge production, still Odisha is unable to meet the onion demand. Many times it is found that diseases are important cause of yield reduc-

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tion. Onion is subjected to the attack by several fungal, bacterial and viral diseases. Among the diseases, Purple blotch caused by *Alternaria porri* (Ellis) Cif. is a destructive disease and causes extensive damage to bulbs as well as seed crop. The Alternaria leaf blight of onion is a widespread disease in the state of Odisha. Considering the importance of the disease, the research work has been undertaken to study the purple blotch pathogen, particularly the management aspect.

MATERIALS AND METHODS

Bioassay study of different plant extracts in vitro

Eleven plant species such as Papaya(*Carica papaya*), Ginger (*Zingiber officinale*), Bitter gourd (*Momordica charantia*), Garlic (*Allium sativum*), Tagar (*Tabernaemontana coronaria*), Tulsi (*Ocimum sanctum*), Eucalyptus (*Eucalyptus globulus*), Neem (*Azadirachta indica*), Marigold (*Tagetes patula*), milkweed (*Calotropis procera*) and Bel (*Aegle marmelos*) were selected basing on their local availability and medicinal values to evaluate the efficacy on the inhibition of mycelial growth of test fungus using Poison food technique. To obtain the extracts of all the plants the plant parts were cut in to small pieces and weighed up to 100 g, ground individually in 100 ml sterile water with the help of mortar and pestle. Then the material was passed through two layer muslin cloth. The material was homogenized for 5 minutes and filtered. This stock solution was further diluted to desired concentration. Two different concentrations such as 10% and 20% were prepared by aseptically adding plant extracts to sterilize molten potato dextrose agar medium in required concentration and poured into Petridishes aseptically. The medium without any plant extract served as control. The Petriplates were then inoculated with mycelial discs of 5 mm diameter cut out aseptically from the periphery of 15 days old culture of test fungus maintained on potato dextrose agar medium plates and incubated at room temperature (28±1°C). The experiment was set in completely randomized block design with triplicates. The activity was measured in terms of radial growth of mycelium till fungal growth in control covered the entire Petriplate. The per cent inhibition of growth of the fungus was calculated following the formula of Vincent (1947): I=C-T/C X 100 where C- Colony diameter in control, T- Colony diameter in treatment, I - Per cent inhibition

Efficacy of biocontrol agents against the growth of the test fungus

Five common bio control agents such as *Verticillium lecanii, Trichoderma viride, Trichoderma harzianum, Metarrhizium anisopliae* and *Beauveria bassiana* were collected from AICRP on Biological control, OUAT, Bhubaneswar and cultured individually. Some entomopathogenic fungus were also included in order to explore their activity against *Alternaria porri*. The effectiveness of bio- control agents were tested against the test fungus following dual culture technique. A set of sterilized Petriplates were taken and 20ml of sterilized potato dextrose agar was poured into each of the Petridishes under aseptic condition. After the media solidified, the test fungus of 5 mm disc was inoculated at one end of the Petriplate and same amount of cultured bio control agent on the opposite end. Each set of treatment was replicated four times and incubated at (28±1°C) for 10 days. A set of control was maintained without any biocontrol agent. Observations were taken regarding the inhibitory effect of the bio control agent over the growth of the test fungus and per cent inhibition of the mycelial growth was calculated by using 'Vincent formula' as described earlier.

Bioassay of fungitoxicants

In order to determine the relative efficacy of eleven new generation fungitoxicants on the inhibition of the mycelial growth of the test fungus, poison food technique was followed. The efficacy of the fungicides at their recommended concentrations were assessed. Required quantity of individual fungicide was added separately into molten potato dextrose agar medium so as to get dissolved in the substratum. Later 20 ml of the poisoned medium was poured into the sterile Petriplate. Medium without any chemicals served as control. Three replications were maintained for the purpose of study. Each Petriplate was inoculated at the centre with mycelial disc of 5 mm diameter cut out aseptically from 15 days old culture of the test fungus and incubated at room temperature (28±1°C) for 7-10 days. The colony diameter was measured in each case. Inhibition of mycelial growth was calculated by using Vincent's formula (1947).

RESULTS AND DISCUSSION

Bioassay of plant extracts in inhibiting the growth of pathogen

All the plant extracts inhibited the mycelial growth

Table 1: Bioassay of plant extracts against *Alternaria porri*

Common Name	Botanical name	Family	Plant parts used	Mean percent inhibition at 10% conc.	Mean percent inhibition at 20% conc.
Papaya	*Carica papaya*	Caricaceae	Leaf	56.2	63.2
Ginger	*Zingiber officinale*	Zingiberaceae	Rhizome	68.4	75.2
Bitter gourd	*Momordica charantia*	Cucurbitaceae	Leaf	58.2	64.5
Garlic	*Allium sativum*	Amaryllidaceae	Bulb	72.1	79.5
Tagar	*Tabernaemontana Coronaria*	Apocynaceae	Leaf	52.0	59.4
Tulsi	*Ocimum sanctum*	Lamiaceae	Leaf	78.4	86.8
Eucalyptus	*Eucalyptus globulus*	Myrtaceae	Leaf	72.1	79.5
Neem	*Azadirachta indica*	Meliaceae	Leaf	82.6	90.4
Marigold	*Tagetes patula*	Asteraceae	Leaf	52.4	60.5
Milk weed	*Calotropis procera*	Apocynaceae	Leaf	58.4	62.6
Bel	*Aegle marmelos*	Rutaceae	Leaf	62.4	68.5
SE(m)±				1.37	1.30
CD(0.05)				4.02	3.8

of the test fungus. However, there was significant variation in inhibition of mycelial growth at different concentrations of plant extracts. At 10 % concentration, maximum inhibition was found in neem (82.6%) followed by tulsi (78.4%). The extracts of neem (*Azadirachta indica*), tulsi (*Ocimum sanctum*), eucalyptus (*Eucalyptus globulus*), garlic (*Allium sativum*) and ginger (*Zingiber officinale*) were found to be the most effective in inhibiting the mycelial growth (>75% mean inhibition) at 20% concentration recording 90.4%, 86.8%, 79.5%, 78% and 75.2 % respectively. Therefore, 20% of stock solution of plant extracts can be treated as standard. In both the concentrations *Tabernaemontana coronaria* gave least inhibition of pathogen. The neem leaf and neem products possessing antifungal activity have been demonstrated earlier against different diseases attacking a wide range of crop plants. The potentiality of Nimbicidin and neem leaf extract have been proved successfully by Pramod Kumar (2007) against *A. porri and A. alternata*. Patilkulkarni (2013) also reported the effectiveness of *Azadirachta indica* against *Alternaria porri* which are in agreement to the present findings. Further Patilkulkarni (2013) conducted experiment on *Ocimum sanctum* against *Alternaria porri* infecting onion which is also in support with our findings. The role of garlic clove extract and NSKE extracts were found highly inhibitory to *Alternaria porri*, which is corroborating the present investigation (Ravichandran,2012). Therefore, the botanicals such as neem, eucalyptus, tulsi and ginger were proved effective in inhibiting the growth of *Alternaria porri* in laboratory condition as reported by Tiwari and Srivastava (2004) which is in corroboration with present finding (Table 1).

Efficacy of bio control agents on the growth of the test fungus

Among the bio control agents evaluated against *Alternaria porri,* the higher percentage of growth inhibition was recorded in *Trichoderma harzianum* (88%) followed by *Trichoderma viride* (83%).). The effectiveness of *Trichoderma harzianum* against *A. porri* was previously reported by Patilkulkarni (2013) and Ravichandran (2012), which supported the present finding. *Trichoderma harzianum* iso-

late (TH-3) expressed high level of disease reduction and growth promotion when different methods viz; seed treatment, seedling dip and three foliar sprays were evaluated on onion bulb crop under glass house and field conditions. The antagonistic effect of *Trichoderma viride* against leaf blight of onion (*Alternaria porri*) is in line of conformity with the findings of Mohan *et al,* (2001), Pramod kumar (2007) and Bhosale *et al.,* (2008) (Table 2).

Bioassay of fungitoxicants

All the fungitoxicants appreciably inhibited the mycelial growth of *Alternaria porri*. Carbendazim + mancozeb and difenconazole showed complete inhibition (100%) followed by hexaconazole (94.2%),

Table 2 : *In vitro* study of antagonists on the growth of *Alternaria porri*

Antagonists	Mean diameter of test fungus in control(mm)	Mean colony diameter in dual culture (mm)	Per cent growth inhibition
Metarhizium anisopliae	75	30.2	59
Verticillium lecanii	75	12.6	83
Beauveria bassiana	75	15.8	78
Trichoderma viride	75	12.4	83
Trichoderma harzianum	75	8.5	88
SE (m)±			1.082
CD (0.05)			3.26

Table 3 : Bioassay study of fungitoxicants

Fungicide	Trade name	Fomulation	Concentration (%)	Mean per cent mycelial growth inhibition
Carbendazim(12%WP) +Mancozeb (63% WP)	Saaf	75%WP	0.2	100
Thiophanate Methyl	Roko	75%WP	0.15	86.5
Metalaxyl (8%) + Mancozeb (64%)	Ridomyl-MZ	72%WP	0.2	82.4
Chlorothalonil	Kavach	75%WP	0.2	80.6
Copper hydroxide	Kocide 101	77%WP	0.1	87.2
Fenamidione (10%) + Mancozeb (50%)	Sectin	60%WG	0.1	72
Cyamoxanil (8%) + Mancozeb (64%)	Curate M-8	72%WP	0.1	78
Hexaconazole	Contaf	5%SC	0.2	94.2
Flusilazole	Cursor	40%EC	0.15	92.5
Propiconazole	Tilt	25%EC	0.15	90.1
Difenconazole	Score	25%EC	0.05	100
SE(m)±				1.15
CD(0.05)				3.37

flusilazole (92.5%) and propiconazole (90.1%) which found at par with each other. Fenamidione (10%) + mancozeb (50%) was reported to be least effective chemical (72%). Deshmukh *et al,* (2007) observed maximum disease control (79.58%) by foliar application of a mixture of hexaconazole (0.005%) + mancozeb (0.3%) followed by difenconazole (0.025%) + Mancozeb(0.3%) in reducing disease intensity 78.58% and 70.72% as well as avoiding yield loss by 26.16% and 22.67% respectively. Difenconazole, a new molecule found to be highly effective against *Alternaria porri* which inhibited spore germination of by 87.38% followed by Mancozeb alone (85%) as reported by Abdul *et al.* (2012) supports the present finding. However, the role of chlorothalonil in inhibiting the spore germination of *Alternaria porri* was not enchouraging, but in the present investigation chlorothalonil proved efficacious against *Alternaria porri* (Table 3).

Onion is an important vegetable, can be taken as raw as well as cooked. Due to its growing demand there is need to reduce the loss due to disease in field, storage as well as in transit. Excessive use of chemicals may leave residue or may cause environmental pollution. Biocontrol agents and phytoextracts may not give total control of the disease, however found efficacious. Therefore, an integrated disease management schedule may be devised keeping in view the vegetable productivity and safety to environment.

REFERENCES

Abdul, M.K., Murthy K.V.M. Krishna, Waseem M.A., Ahamed P.M. and Nadaf ,H. 2012. Effect of fungicides on spore germination and growth of *Alternaria porri*. *Journal of life sciences*, **9** : 292-294.

Bhosale, M.V., Perne, R.R.; Shete, M.H. and Pawar, N.B. (2008) Efficacy of botanicals, bioagents and fungicides on management of leaf blight (*Alternaria porri*) of onion. *Journal of Plant Disease Sciences,* **3:** 88-89.

Deshmukh V.S., Dhruj. V., and Chavan, R.V. 2007. Chemical control of purple blotch (*Alternariaporri*) (Ellis) Cif. Of onion. *Plant Disease Reasearch*, **22**: 32-34.

Mohan, K., Ebenezer, E. G., and Seetharaman, K. 2001. Management of leaf blight disease of onion caused by *Alternaria porri* by plant extracts, plant oils and bio control agents. *National Horticultural Research and Development Foundation,* **1** :11-14.

Pramod Kumar,T. 2007. *Biological management of Alternaria blight of onion* . M.Sc (Agri) Thesis, University of Agriculture Science, Dharwad, Karnataka

Patilkulkarni, V.G. 2013.*Organic management of purple bloth of onion caused by Alternaria porri (ellis) cif..* M.Sc (Agri) Thesis, University of Agriculture Science, Dharwad, Karnataka

Ravichandran, S. 2012 *Epidemiology and management of purple blotch of onion caused by Alternaria porri (ellis.) cif. M.Sc (Agri) Thesis,* University of Agriculture Science, Dharwad, Karnataka.

Tiwari, B.K and Srivastava, K.J. 2004. Studies on bioefficacy of some plant extracts against pathogens of onion. *National Horticultural Research and Development Foundation*, **24** : 6-10.

Vincent, J.M.,1947.Distortion of fungal hyphae in presence of certain inhibitors.*Nature*,**150**:850.