The Bioscan 9(1): 355-357, 2014 (Supplement on Plant Pathology)
AN INTERNATIONAL QUARTERLY JOURNAL OF LIFE SCIENCES
www.thebioscan.in

A NEW REPORT OF TIP BLIGHT AND LEAF SPOT OF *ACORUS CALAMUS* CAUSED BY *NIGROSPORA ORYZAE* FROM WEST BENGAL, INDIA

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KEYWORDS
Acorus calamus
Nigrospora oryzae
Tip blight and Leaf spot

Received on :
04.11.2013

Accepted on :
14.02.2014

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ABSTRACT
Acorus calamus is known as sweet flag (Family: Acoraceae) is a perennial medicinal plant. Found in both temperate and subtemparate zones. Roots and rhizomes are stimulants, emetic, nauseant, stomachic, aromatic, expectorant, carminative, antispasmodic and nervine sedative. Gas chromatography had revealed the presence of two components isolated in pure state *i.e.* α-asarone and β- asarone. Diseases or pathogens cause deterioration of the active chemicals of the plants which affect the quantitative and qualitative loss of medicinal plants. During a study in the year 2010-2012 at three different locations of West Bengal, India, a new tip blight and leaf spot disease was observed in *Acorus calamus* caused by *Nigrospora oryzae*. In *Acorus calamus,* maximum disease incidence and disease index were recorded during November to February and minimum disease incidence and index of leaf spot or blight by *Nigrospora oryzae* during May - July, thereafter gradually increased and again reached to the peak during December - January.

INTRODUCTION

Currently in India Mints (1, 50,000 ha), Senna (20,000 ha), Tulsi (5,000 ha), Safed Musli (5,000 ha) etc are grown commercially and providing employments in terms of man-days by cultivation and post harvest processing in Mint (4.0 Cr), Tulsi (4.0 Cr), Safed Musli (1.3 Cr) and several others (Khanuja *et al*., 2006). So the challenge before country is not only preparation of a model GAP documents but also its implementation in its true spirit. West Bengal exhibits a varied range of topography and agro climatic conditions, which enormously contribute on its vegetation and floristic consumption. In spite of this, information on area under medicinal plants and production are not available (Das, 2002). Several biotic and abiotic factors limit the production of these crops. Those pathogenic disease cause significant damage of the crops as well as reduce the quality of the produces and acceptability to the market. The disease may reduce the active chemicals components in the plant parts used for medicinal purpose (Mukherjee, 2009). A survey was carried out to study the diseases of medicinal plants in West Bengal, India. Three locations i.e. Kalyani, Narendrapur and Krishnanagar were selected to study the different foliar diseases of different medicinal plants (Balai *et al.,* 2013). Among all the medicinal plants, a new disease was observed in three locations. Later, diseased leaf samples were collected from fields and isolated the pathogen. The pathogen was confirmed by pathogenicity test and the symptoms of the disease and morphomatric characters of the pathogen were studied very carefully.

Therefore the main objective of this study was to identify the actual cause of diseases of medicinal plant like *Acorus calamus* and on the basis of identification the control measures can be opted infuture.

MATERIALS AND METHODS

Study of the disease symptoms

Disease conditions in the plants were recognized according to the symptoms produced by the pathogens. The maladies observed on the plants were recorded. The plants were carefully studied for fixed and targeted spots on the leaves of *Acorus calamus*.

Potato Dextrose Agar media

Peeled potato	200 g
Dextrose	20 g
Agar	20 g
Distilled Water	1000 mL
Chloramphenicol	100ppm

Isolation of the fungi causing foliar diseases on the *Acorus calamus*

Method of isolation

The leaves showed characteristic symptoms were collected from the field and brought to the laboratory for isolation of the pathogen. Isolation was carried out on a sterilized zone of the laminar air low. The diseased specimens were washed with running tap water. The washed leaves were taken into laminar air flow chamber and cut into small pieces by a sterilized scissor which contained the half diseased portion and half



Figure 1, 2 & 3: Tip blight and leaf spot of *Acorus calamus* caused by *Nigrospora oryzae*

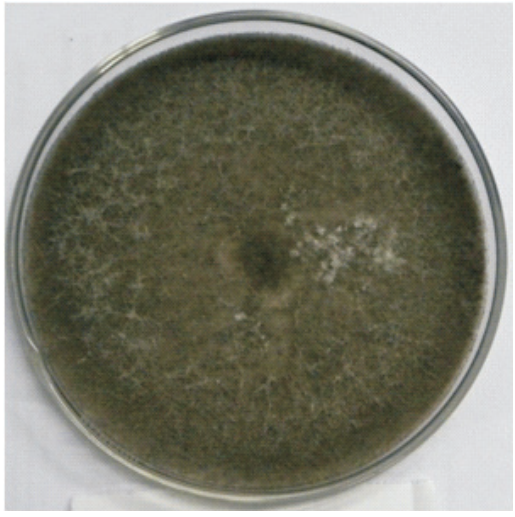

Figure 4: Culture of *Nigrospora oryzae* in PDA media

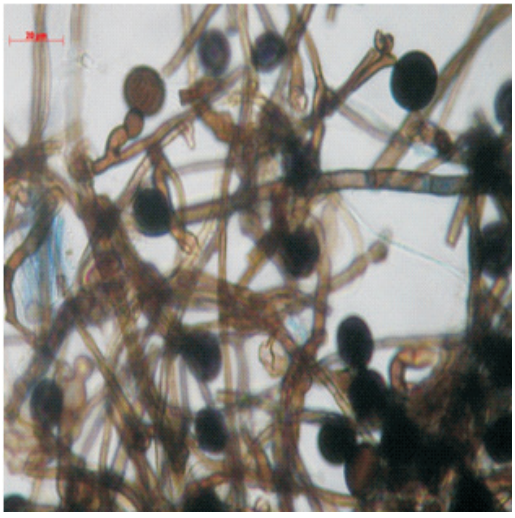

Figure 5: Conidia and conidiophores of *Nigrospora oryzae* (40X)

healthy portion. The pieces were dipped into 0.1% aqueous mercuric chloride solution for 30 seconds, followed by 3-4 times washings with sterile distilled water. With the help of a sterilized forcep, each piece was placed on the solidified PDA. About 3-4 such pieces were placed on each plate by maintaining some distance from each other. All the plates were kept into BOD incubator at 28 ± ºC for 4 days. Then growing hyphal tips were transferred into PDA slant (Junaid *et al.,* 2014).

Maintenance of pure cultures of the isolated fungi

The cultures were maintained in PDA slants and kept in refrigerator at 5ºC. Sub culturing of the isolated pathogens were made at 15 days interval.

Preparation of slides of the fungal cultures to be observed under the phase- contrast microscope

The slides of the selected fungal cultures or colony were prepared in order to study the fungal morphology such as the characteristics of the hyphae, conidiophores, spores etc. for easy identification of the fungal species. The prepared slides were observed under phase-contrast microscope.

Pathogenicity test

Preparation of spore suspension and artificial inoculation of the plants

For artificial inoculation, the culture of inoculums was prepared in PDA media poured petri plate. The plate was incubated for 4 days in BOD incubator at 28 ± ºC. After getting the culture of the test fungi, spore suspension was prepared with sterile distilled water. Spore suspension was sprayed on the 30 days old healthy plants, grown in the earthen pot, control plants also maintained by spraying only sterile distilled water and then covered by the poly-propylene packet to maintain the humidity and to maintain the favorable condition for disease development.

Confirmation of pathogens

15-20 days after inoculation on the test plants, disease symptoms were developed. The diseased leaves were collected and again re-isolated the pathogens to compare with the previous isolated pathogens and to get confirm about the disease causing pathogens.

Study of the morphomatric characters of the pathogens and identification

To identify the pathogens and to know their taxonomic

position, morphomatric studies were carried out. To study the morphomatric characters of fungi, size of the spores, and different parts of the fungal fruiting bodies were measured under microscope with the help of stage and ocular micrometer and the length and breadth measurements of different fungal structures were recorded. Later fungal cultures were sent to National Center of Fungal Taxonomy, New Delhi for confirmation up to species level (Adhikary *et al.,* 2013).

RESULTS AND DISCUSSION

Symptom of the leaves was studied very carefully. First minute brown to black spots were appeared at the leaf tip, later it spread from leaf tip to lower part of the leaves. Spots coalesced with each other and appeared as a large spot. Blight symptom was shown from tip and drying of the leaves occurred from leaf margin (Fig. 1 and 2). Whole leaf becomes dry in severe infestation (Fig. 3). Pathogen was isolated in PDA media and confirmed by Koch's postulate test.

The pathogen was grown on PDA medium from Infected leaf and identified as *Nigrospora oryzae* on the basis of cultural and morphometric characters (Barkley and Broome, Petch, 1924). On PDA media the colony of the pathogen was whitish velvety mycelial growth which became grey to black in later stage. Submerged mycelia with black colour at centre of back side of the plate; later black colour was spread towards the periphery of the plate with the increase in formation of spore (Fig. 4). Hyphae of the fungus were hyaline, branched, septate with 2.68-5.44 μm width. Conidia were subglobose and reddish brown to black in colour, smooth surfaced, born on short pedicel. Pedicels were broader at the base and taper toward the attachment point of the spore, spore size ranging from 6.12 -15.00 $\times$ 5.06 - 13.31 μm (Fig. 5). The confirmation of the pathogen was done from National Centre of Fungal Taxonomy, New Delhi (Identification No. 4290.11). Some pathogens like *Gloeosporium* sp. (Singh and Gupta 1977), *Uromyces sporgoni* (Rangaswami *et al.,* 1970) have been found associated with diseases of the host plant i.e *Acorus calamus*. Therefore this is the first report of *Nigrospora oryza* on *Acorus calamus*.

REFERENCES

Adhikary, N. K. Dey, S. and Tarafdar , J. 2013. Studies on morphology of mango anthracnose disease causing fungus *Colletotrichum gloeosporioides* (penz.) penz. and sacc. and efficacy of azoxystrobin against the fungus under *in vitro* and *in vivo* condition. *The Bioscan*. **8(2):** 493-497.

Balai, L. P., Singh, R. B., and Yadav, S. M. 2013. Survey for the disease status intensity of alternaria blight of pigeonpea in eastern part of Uttar pradesh and adjoining districts of western Bihar. *The Bioscan*. **8(1):** 63-66.

(Berk. and Broome) Petch. **1924**. Nigrospora oryzae. *J. Indian Bot. Soc*. **4:** 24.

Chopra, R., Chopra, I., Handa, K. and Kapur, I. 1994. Indigenous Drugs of India. *Academic Publishers, Calcutta,* India. pp.

Junaid, J. M., Shah, T. A., Bhat, A. H., Bhat, N. A., Dar, N. A. and Ambardar, V. K. 2014. Morphology and status of occurrence of anthracnose of bean (*Phaseolus vulgaris* L.) caused by *Colletotrichum lindemuthianum* (Sacc. and Magn.) Scrib. *In kashmir valley. The Bioscan*. **9(1):** 235-241.

Khanuja, S. P. S., Kalra, A. and Singh, A. K. 2006. Export market: advantage India. *The Hindu Survey of Indian Agriculture*. pp. 208-211.

Mukherjee, M. 2009. Studies on Major Diseases and Biochemical Investigations of Some Important Medicinal Plants in West Bengal with Special reference to Senna (*Cassia angustifolia*). *Ph. D (Plant Pathology) Dissertation, BCKV,* Mohanpur. pp. 6-7.

Rangaswami, G., Seshadri, V. S. and Channamma, K. A. 1970. Fungi of South India. *University of Agricultural Sciences,* Bangalore. p.193

Singh, I. D. and Gupta, R. C. 1977. Two new leaf spot diseases. *Curr Sci*. **46:** 539.