



A new report of natural infestation of *Fusarium solani* on banana rhizome weevil, *Cosmopolites sordidus* germar from Kerala

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Abstract

Banana rhizome weevil, *Cosmopolites sordidus* Germar is a common pest in banana plantations of southern Kerala. *Fusarium solani* (Mart.) Sacc. was isolated from the weevil collected from College of Agriculture, Vellayani, Thiruvananthapuram and is the first report of the infestation of *F. solani* on *C. sordidus*. The fungus was isolated and proved pathogenicity on grubs and adults of *C. sordidus*. Two days after treatment mortality was observed in grubs adults mortality was observed 5 days after treatment. Symptoms of infestation on grubs of the weevil were the initial brownish discoloration and shrinkage of body, later white creamy mycelial growth developed. On adults white creamy mycelia developed but not covering the entire body.

Keywords: *Cosmopolites sordidus*, *Fusarium solani*, monophialide, macroconidia, microconidia

Introduction

Banana rhizome weevil, *C. sordidus* is the borer pest of banana causes considerable loss in the production of banana all over the world. Yield loss caused by the attack of weevil can range from 40 to 100 per cent, if the infestation is severe (Seshu *et al.*, 1998) [8]. Tunnels created by the grubs inside the rhizome causes reduced nutrient uptake by the plant, root growth gets hampered and the plant becomes susceptible to pests and diseases (Rukazambuga *et al.*, 1998; Gold and Messiaen, 2000) [7].

Natural infestation of entomopathogenic fungi on *C. sordidus* can occur due to the abundance of fungal fauna in soil. Adults of *C. sordidus* can get infection of these fungi because of their close contact with the soil (Gold *et al.*, 2001) [4].

Fusarium sp. can be seen associated with the roots of crop plants as endophytes. Some of the isolates of *Fusarium* were found to be phytopathogenic, especially attacking the roots of many plants. They were known to cause infection on the soil inhabiting pests of crops (Majumder *et al.*, 2006). Shukla (2010) [9] isolated endophytic *Fusarium* strains from banana plants and were found to cause 30-48 per cent mortality of grubs of *C. sordidus*.

Materials and Methods

The study was conducted in College of agriculture, Vellayani, at Thiruvananthapuram district of Kerala during 2015-17. The adults and grubs of *C. sordidus* were collected from banana fields of Thiruvananthapuram district and also from College of Agriculture, Vellayani. Pseudostem traps were used for the collection of adult weevils. Pseudostems of 10-15cm were placed in different parts of the banana fields, near the rhizome region of the plant. Pseudostem traps were observed daily for the collection of weevils.

Adults and grubs were also collected from the decaying rhizomes of uprooted banana plants. Grubs were collected by cutting the rhizomes without damaging the grubs. Collected

weevils and grubs were maintained in rhizomes inside rearing jars of height of 25cm and width 15cm. The jar was covered with muslin cloth.

The collected insects were observed daily and dead insects were separated. They were kept in moist chamber for development of mycosis. Cadavers were kept one to two days for the development of fungal mycelia.

Cadavers with mycelial growth was surface sterilized using 2% sodium hypochlorite for three minutes followed by washing in sterile water and removed the excess moisture. The whole process were done inside a laminar airflow chamber. The cadavers were then placed in petri plates containing Potato Dextrose Agar (PDA) media for further development of mycelia. The pure culture was prepared after repeated hyphal tip culturing of the specific entomopathogenic fungi. Spore count of the fungus was determined using Neubauer hemocytometer.

Morphological and molecular identification of the fungal isolate was done. Morphological identification was done by observing the spores and conidiophores of the fungi under stereomicroscope. Molecular identification was done by DNA barcoding at Rajiv Gandhi Centre for Biotechnology (RGCB).

Morphological and molecular Identification

The morphological identification of the fungi was done by preparing the slide cultures of the fungi from 14 day old culture (Harris, 1986) [5]. Using a Motic BA 120 microscope, the characters of conidiophores along with the spore size and shape were studied. The radial growth of the fungi was measured by cutting a mycelial disc of 5 mm from the culture using a cork borer and was placed at the centre of the PDA fig. Three replications were maintained and the mean radial growth was recorded daily. The colour of the mycelia was also observed daily.

Molecular identification was done through ITS (Internal Transcribed Sequencing).

Pathogenicity

Pathogenicity of the isolate was tested on adults and grubs of *C. sordidus* by spraying the spore suspension prepared from 14 days old culture on the insect and then they have been given fresh rhizome. Mortality of *C. sordidus* along with the symptoms developed after infection were observed daily.

Results and Discussion

Fusarium species are mainly the endophytes which are associated with the roots of many plants. This may cause infection on the subterranean insect pests (Majumdar *et al.*, 2006) [6]. Similarly the natural infestation of *F. solani* was observed by Anitha, (2000) [2] on pseudostem weevil and Majumder *et al.* (2006) observed its natural infestation on the root maggot of sugarbeet, *T. myopaeformis*. The fungus isolated from the mycosed cadaver of the weevil produced creamy white floccose colonies on the upper surface of the PDA fig (fig 1) and cream color on the reverse side (fig 2). Slide culture of the fungi was prepared in order to identify the fungi at its genus level (Auichi *et al.*, 2013).

Within 14 DAI (Days After Inoculation), the fungus attained radial growth of 9 cm. The macroconidia was 3 to 4 septate fusiform with blunt end having a mean length of $15.91 \pm 5.20 \mu\text{m}$ and width of $4.09 \pm 0.93 \mu\text{m}$. Ovoid aseptate microconidia was also observed with a mean length of $11.10 \pm 2.6 \mu\text{m}$ and width of $3.7 \pm 0.93 \mu\text{m}$ (fig 3). Microconidia and macroconidia have a length /width ratio of 3.88. The phialides were produced as single from unbranched conidiophores (fig 4). Spore count of the fungus was found to be 1.5×10^7 spore's mL^{-1} .

The fungus was identified at Rajiv Gandhi Centre for Biotechnology, Thiruvananthapuram and obtained a 532 base pair (bp) sequence. The nucleotide sequence was analysed using BLAST (Basic Local Alignment Searching Tool) showed cent per cent similarity with *F. solani*.

F. solani was found to be pathogenic to grubs and adults of *C. sordidus*. Its pathogenicity was proved on grubs of pseudostem weevil and pupae of sugarbeet root maggot (Anitha, 2000; Majumdar *et al.*, 2006) [2, 6]. Grubs were noticed with reduced feeding and movement after 2 days of treatment. Adults were not showed any changes in feeding behavior. The fungus penetrate the insect cuticle by the production of enzymes which along with the mechanical pressure causes the invasion of fungus into the insect body (Shykh *et al.*, 1977) [10].

The symptom development occurred after death of the insect. Grubs after two days of treatment found dead and started developing a brownish discoloration at the ventral thoracic region and became shrunken. Initially white creamy mycelial growth was appeared in the ventral thoracic region later the entire body was covered with mycelial growth (fig 5). Grubs became hard and stiffy (fig 6).

Adult weevils has got infection after 5 days of treatment and the creamy white growth initiated from the intersegmental region. The growth was cream colored and was not covering entire body (fig 7).



Fig 1: Upper side of PDA plate



Fig 2: Reverse side of PDA plate



Fig 3: Macroconidia and microconidia of *F. solani*



Fig 4: Monopialide produced from unbranched conidiophore



Fig 5: Grub with Brownish discoloration



Fig 6: Grub with hard and stiffy body

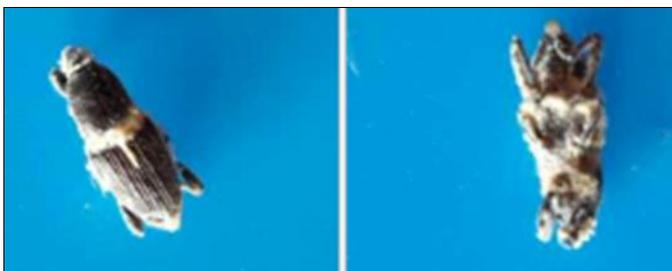


Fig 7: Adults with mycelial growth of *F. solani*

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