

cropping beds in a mushroom farm in Shimla. Although pathogenic, it is less destructive than *A. composticola*, *A. agarici* and *A. myceliophagus*.

***Aphelenchoides sacchari*:** This species, is one of the destructive pests of mushrooms in India. It has a fast multiplication rate and one generation from egg to egg takes 12 days to complete. This is the first Aphelenchoid spp. that has been grown on the artificial medium.

Mode of feeding: Myceliophagous nematodes damage the mushroom mycelium directly as they feed upon the mycelia sap of the hyphal cell. The nematodes pierce the cell wall by to and fro movement of their needle like hollow stylet and suck the cell sap. After sucking the sap of the cell, the nematode shifts to the next cell and in the process, the whole mycelium is damaged due to devitalisation of the cells. Shifting of the nematode from one cell to another is aided by either the moist film already present in the compost or by the film produced by the bleeding of the damaged hyphal cells.

Symptomatology: Symptoms of the nematode attack are generally not evident initially, mainly because of the reluctance of the grower to disturb the beds after spawning. The first clear-cut symptom of the nematode attack is visible when whiteness of the spawn starts turning brown.

Nematode management

Chemical control: Thionazin has been found to be fit for recommendation. This nematicides when spreaded on the bed surface at 80 ppm during the spawn run whereas increased the button production manifold, did not pose residual problem on the sporophores. Unfortunately, this chemical is not available in India.

Biological control: Some botanical agents like neem, coconut, castor and groundnut cakes were quite effective and left no residual effect on the crop. Different formulations were available yet sanitation and good hygiene remain the first priority.

Neem (*Azadirachta indica*) leaf, Neem cake and Dazomet treatment of composed beds) effectively increased number of fruiting bodies as well as yield of *Agaricus bisporus* by reducing the population of *A. composticola*. Among the treatments, neem cake @ 20 g and 10 g and neem leaf @ 26 g dose showed manorial effect on *A. bisporus* in terms of yield potential, but at higher doses proved toxic to fungi.

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Collar Rot: A Severe Seedling Disease of Groundnut

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Introduction: The cultivated groundnut (*Arachis hypogaea* L.) belongs to family *Fabaceae*, subfamily *Papilionaceae*. Groundnut is grown in nearly 100

countries. In India, it is cultivated on 5.31 million ha with production of 6.93 million tonne (Anonymous, 2012). It is mainly grown in Gujarat, Maharashtra,

as was evidenced from number of fruiting bodies and yield data.

Predatory fungi: Nematode trapping fungi, *Arthrobotrys robusta* has been recommended and is commercially available as "Royal 300". Besides, *Arthrobotrys oligospora* and *A. superba* have also been known to reduce the nematode count and increase the yield of mushroom. In India, the fungus, *Arthrobotrys irregular* has shown the promising result against myceliophagous nematodes without showing any inhibiting effect on the development of *A. bisporus* mycelium.

Nematodes have no sense of sight; they depend on chemical clues to track down food and avoid danger. Carnivorous fungi take advantage of that by exuding chemicals that attract nematodes, then snaring them. The net fungus produces a protein that combines on contact with a sugar in nematode surface cuticle to produce instant superglue. The chemical bond cannot be broken by physical force; no matter how violently the worm wiggles, it can't get loose after just a few seconds contact. Hyphae strands invade its body, first releasing paralyzing toxins, then digesting it from the inside out.

The bacterium, *Bacillus thuringiensis* has also been reported to suppress the population of *Ditylenchus myceliophagus*.

Physical control: Steaming of compost packed rooms at air as well as bed temperature of 60°C for at least 2 hours before spawning eliminate the nematodes. Casing soil also needs similar heat treatment. Heating of cropping rooms at 70°C for 5-6 hours or at 80°C for 30-60 minutes checks out nematode dissemination to next crop.

Conclusion: Nematode once entered into the cropping system cannot be eliminated with any of the management strategy without destroying the running crop. Since various control measures have their own limitations as for as their use in mushroom cultivation is concerned, "Precaution is better than cure" holds true in this case. Thus various under given precautions and practices if adopted raise a successful crop. Fertilizers and soils used in the preparation of mushroom composts form the main source of infestation. Therefore the manure should be broken thoroughly 3 times and its layers allowed to decay. This together with strict agrotechnical practices should be observed in the cultivation of mushrooms.

Andhra Pradesh, Uttar Pradesh, Tamil Nadu, Rajasthan, Karnataka and Madhya Pradesh. The groundnut contains high oil (46-54%) and protein (25-36%) content. There are several soil-borne fungal diseases affecting yield of groundnut but collar rot or crown rot is most serious and widely spread disease of groundnut in sandy loam and medium black soils. This disease is caused by *Aspergillus niger* Van Tieghem, is a filamentous fungus growing aerobically on organic matter. In nature, it is found in soil and litter, in compost and on decaying plant material. This fungus is able to grow on wide range of temperature but optimum growth at 32-37 °C. Besides causing collar rot of groundnut, this fungus causes black mold on fruits and vegetables such as grape and onion.

Distribution and economic importance: Collar rot has got economic importance as the fungus causes rot of both seed and seedling which drastically reduces the plant stand (Kishore et al. 2005). Collar rot is an important disease in warm and temperate groundnut growing areas. The fungus causes pre-emergence rotting of groundnut seed and the infected seed fails to germinate. In emerged young seedlings, *A. niger* infection results in sudden wilting (Middleton et al. 1994). The fungus is present in soils and is a common contaminant of groundnut seed. However, epidemics of the disease are sporadic and appear to be related to the prior occurrence of one or more stresses. Extreme heat or fluctuations in soil moisture during the seedling stage favours the disease development. Collar rot is prevalent in almost all groundnut growing states of India viz., Rajasthan, Gujarat, Punjab, Andhra Pradesh, Tamil Nadu, Uttar Pradesh, Maharashtra, Orissa, Karnataka and Madhya Pradesh particularly in sandy loam and medium black soils. In Gujarat the losses in terms of mortality of plants due to collar rot range from 28 to 50 per cent (Ghewande et al. 2002).

Disease symptoms: The major symptoms are pre-emergence rotting of seed, rotting of crown region tends to sudden wilting of young plants. The crown area of infected plants just below the soil surface may be swollen and eventually becomes covered with a black, sooty mass of fungus growth. As the fungus grows, the entire crown region becomes shredded and dark brown. The wilted and dead young plants are easily detached from crown region. Most affected plants die within 30 days from sowing. Mature plants are also attacked, symptoms include wilt of branches permanently and/or wilting of entire plant. The dead and dried branches are easily detached from the collar region. Infected pods reveals patches of black sooty spores (Gajera and Vakharia 2010). Tap root of affected plants reveals an internal discoloration of the vascular system that is dark grey in colour.

Disease epidemiology: The life cycle of *A. niger* begins with the production of conidia (asexual spores) that are easily dispersed into the air and causing disease. *A. niger* is predominantly

saprophytes grows aerobically on organic matter, therefore it can be found almost everywhere in environments that contain soil. Also, it is found in waste, decaying plant material and compost in outdoor environments (Deepake 2009). *Aspergillus niger* has been exhibited to sustain growth in freezing temperatures, which indicates it as a thermo-tolerant that can also survive at very high temperatures. Its thermo-tolerant abilities that enable growth in a wide temperature range from 6 to 47 °C with a preferred optimum temperature at 35-37 °C. The fungus is capable of growing over a very wide pH range, from 1.4 to 9.8 pH. The growth ability in various temperature ranges, pH ranges as well as the abundant amount of conidiospores allow species to be continuously widespread and Conidiospores are distributed by air (Schuster 2002).

Management practices: Cultural management practices like deep ploughing to bury crop residue, early sowing, avoid deep sowing, suitable crop rotation with wheat and gram and avoid any mechanical damage during intercultural operations are important in reducing collar rot incidence in groundnut (Ghewande et al. 2002). Biological control has been also proved to be a promising in disease management for collar rot. Seed treatment with *Trichoderma viride* or *T. harzianum* @ 10 g/kg seed and @ 4 kg/ha with 250 kg castor cake before sowing helps in management of collar rot in groundnut. The chemical seeds treatment with tebuconazole 2% DS @ 1.5 g/kg and spray tebuconazole 250 EC @ 1 ml/l water at 45 and 60 days after sowing. Seed treatment with propiconazole @ 2 ml/kg seed or vitavax @ 2 g/kg seed (Anonymous 2011). Currently there is no cultivars are known to be resistant to collar rot in groundnut except some genotypes have been found to show lower incidence than average susceptibility to collar rot under field conditions. Some tolerant groundnut varieties like OG-52-1, JCG 88 and J 11 found tolerant to collar rot under field conditions.

References

- Anonymous (2012). Agricultural Statistics at a glance-2012. Directorate of Economics and Statistics, Department of Agriculture and Cooperation, New Delhi. pp 97-98.
- Anonymous (2011). Annual Progress Report of AICRP on Groundnut, D.G.R., Junagadh.
- Deepake, U. (2009) Aero-microbiological studies of Moisture Affected Buildings in the Indoor Environment. *Journal of Young Investigators*. 19(11).
- Gajera, H.P. and Vakharia, D.N. (2010). Molecular and biochemical characterization of *Trichoderma* isolates inhibiting a phytopathogenic fungi *Aspergillus niger* Van Tieghem. *Physiol Mol Plant Pathol*. 74:274-282.
- Ghewande, M.P.; Desai, S. and Basu, M.S. (2002). Diagnosis and management of major diseases of groundnut. NRCG Bulletin. pp 8-9.
- Kishore, G.; Pande, S. and Podile, A.R. (2005). Biological control of collar rot disease with broad spectrum antifungal bacteria associated with

groundnut. *Can J Microbiol.* 51:123-132.

Middleton, K.J.; Pande, S.; Sharma, S.B. and Smith, D.H. (1994). Diseases. In The groundnut crop: a scientific basis for improvement. Edited by J. Smartt. Chapman and Hall, London, U.K. pp. 336-

394.

Schuster, E.; Dunn-Coleman, N.; Frisvad, J. and Van Dijk, P. (2002). On the safety of *Aspergillus niger* - a review. *Applied Microbiology and Biotechnology.* 59(4-5): 426-435.

47. PLANT PATHOLOGY

Siderophore Mediated Fe³⁺ Uptake in Fungi

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Iron is an essential nutrient for all eukaryotes and nearly all prokaryotes. As a transition element, iron can adopt either of two ionic forms, reduced ferrous (Fe²⁺) or oxidized ferric (Fe³⁺). The capacity to gain or lose electrons makes iron the major redox mediator in biology. Either alone or incorporated into iron sulfur clusters or in heme, iron is indispensable for a variety of cellular processes including respiration, the tricarboxylic acid cycle, oxidative stress detoxification, as well as synthesis of amino acids, desoxyribonucleotides, lipids and sterols. Iron is one of the most abundant elements on Earth. In an aerobic environment, however, iron is largely present in the oxidized form, as oxyhydroxide colloid particles that have a solubility below 10⁻⁹ M at neutral pH. As this is well below the need, iron-dependent microorganisms have evolved different strategies to solve the bioavailability problem.

Mechanisms of iron acquisition in fungi: In fungi, four different mechanisms for iron uptake have been characterized at the molecular level: (1) Siderophore-mediated Fe³⁺ uptake (high affinity iron uptake systems). (2) Reductive iron assimilation (RIA) RIA is a two-step process that begins with the extracellular reduction of Fe³⁺ to Fe²⁺ at the plasma membrane followed by high-affinity uptake of Fe²⁺ (high affinity iron uptake systems). (3) Heme uptake: represents the utilization of a special iron source typically found within hosts. (4) Direct Fe²⁺ uptake (low affinity iron acquisition).

Siderophores: Siderophores are low molecular mass, Fe³⁺ specific chelators which form tight complexes with Fe³⁺ to overcome the problem of low bioavailability by solubilization.

Fungal Siderophores: All fungal siderophores identified so far are hydroxamates. Fungal hydroxamates are derived from the nonproteinogenic amino acid ornithine and different acyl groups and can be grouped into five structural families:

1. **Rhodotorulic acid:** Produced by *Rhodotorula*, *Sporobolomyces*
2. **Fusarinines:** Produced by *Fusarium*, *Gliocladium*, *Penicillium*
3. **Coprogens:** Produced by strains of *Penicillium*, *Neurospora crassa*
4. **Ferrichromes:** Produced by *Ustilago*

sphaerogena, *Aspergillus*, *Trichoderma*, *Penicillium*

5. **Ferritins:** Produced by Zygomycetes (*Rhizopus spp.*, *Mucor spp.*)

Most fungal species employ more than one of these systems in parallel but rarely are all four strategies present in the same species. High-affinity systems are important under iron-limited conditions, whereas low affinity systems are of use only when iron is abundant.

Fungal siderophore biosynthesis: A schematic view of the siderophore biosynthetic pathway in general given in Figure 1. The first committed step in the biosynthesis of fungal hydroxamate siderophores is N⁵-hydroxylation of ornithine catalyzed by ornithine-N⁵-monooxygenase. The first fungal gene shown to encode this enzyme activity was *sidi1* of *Ustilago maydis*, followed by characterization of orthologs from *Aspergillus nidulans* (*sida*), *Aspergillus oryzae* (*dfaA*), *Aspergillus fumigatus* (*sida*), *Fusarium graminearum* (*Gibberella zeae*) (*SID1*) and *Cochliobolus heterostrophus* (*SIDA*).

Deletion of the gene encoding ornithine-N⁵-monooxygenase blocks synthesis of all siderophores in all *Aspergillus*, *Ustilago* and *Fusarium* species investigated. Notably, the *A. nidulans sida*-deficient mutant is barely able to grow unless supplemented with siderophores or high amounts of Fe²⁺ owing to the lack of another high-affinity iron acquisition system. In contrast, lack of siderophore biosynthesis affected axenic growth of *U. maydis*, *A. oryzae*, *A. fumigatus*, *C. heterostrophus* and *F. graminearum* only to some degree when mutant strains were grown under iron-depleted conditions.

Analysis of the pathogenicity of mutants carrying a null allele of *sidi1* suggests that the biosynthetic pathway of siderophores does not play an essential role in the infection of maize by *U. maydis*. Hence, until recently there was a long thought that siderophores are not involved in virulence of pathogen.

Recent studies showed that, in the smut fungus *Ustilago maydis*, a tightly regulated cAMP signaling cascade is necessary for pathogenic development. Transcriptome analysis using whole genome microarrays set up to identify putative target genes of the protein kinase A catalytic subunit *Adr1*