

Mycoparasitism of *Mycogone pernicioso* in edible fungi

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Introduction

Mycogone pernicioso causes wet bubble disease in mushroom fungi such as white button (*Agaricus bisporus*), shiitake (*Lentinula edodes*) and oyster mushroom (*Pleurotus spp*). It is among the major production lowering factors in these mushrooms. *Mycogone pernicioso* has a worldwide distribution and may cause complete crop failure under favourable environmental conditions. The perfect stage of *M. pernicioso* is hypomyces pernicioso. Mycelium of the pathogen is white, compact and fluffy mycelium. Hyphae are branched interwoven, septate and thin-walled. One-celled phialoconidia on Verticillium- like conidiophores and bicellular conidia which are commonly referred to as either aleuriospores or chlamydospores are produced by the *M. pernicioso*. In India, button mushroom contributes 73 percent in total mushroom production of the country.

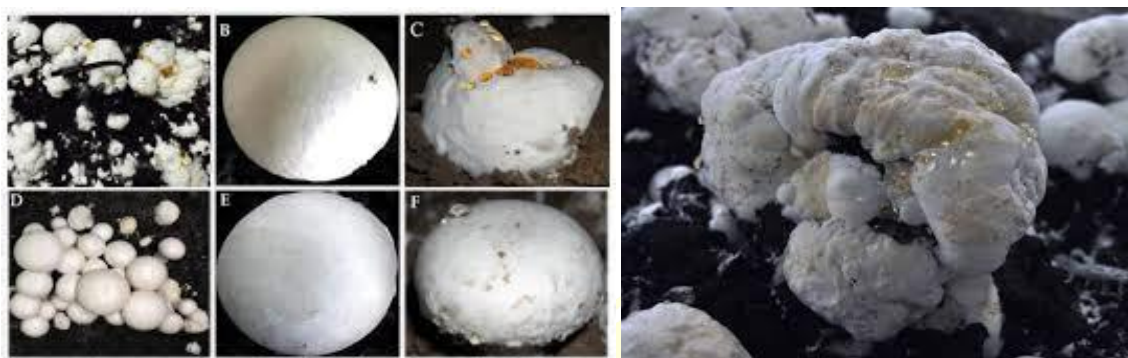
This disease was first reported for Peris in 1888. The disease has also been reported to assume serious proportions in other major mushroom growing countries of the world such as France, United Kingdom, Netherlands, USA, China, Taiwan, South Africa, Brazil, Hungary, Australia and Poland. In India, this disease reported for the first time in 1978 in Jammu and Kashmir. At present, it is found in almost all the button mushroom growing areas of the country. Major reasons for the widespread of the disease are lack of knowledge about hygiene required for mushroom crops, poor sterilization of casing soil, semi-controlled growing/sterilization facilities and a higher population of disease-transmitting vectors like mushroom flies Mycoparasitism is refers to the parasitism of one fungus by another. To protect the edible fungi from mycoparasitism, understanding of its mechanism is very important. For instance, during spawn run stage of the *Agaricus bisporus* (button mushroom), mycelium is not infected by *M. pernicioso*. But after the completion of spawn run and case

run, when the fresh air is introduced in the growing room and temperature is brought down to the range optimum for the primordial formation, the conidia of the *Mycogone perniciosa* are activated, and eventually, germ tubes are formed to invade the mushroom mycelium. At the onset of primordial formation, some openings are formed on the mushroom mycelium which allows the germ tube to enter into the mushroom hyphae to establish the parasitic relationship. The fact of the matter is that *A. bisporus* (button mushroom) genome is rich in the genes encoding hydrophobins. Hydrophobins form a uniform hydrophobic coat on the surface of mushroom hyphae and do not allow the *M. perniciosa* to penetrate the hyphae during spawn run. This is because the spores of *M. perniciosa* require sufficient moisture on the surface to be infected. Because of the hydrophobic nature of the surface of hyphae, germinated spores of *M. perniciosa* either get dried or stay as such near to the hyphae. During the process of invasion, many lytic enzymes like chitinases, laccases, cellulases etc are also secreted by the pathogen to degrade the host tissues. *M. perniciosa* is a strong parasite of *Agaricus bisporus*, and it grows deep into the mushroom fruit body and completely deformed it into sclera dermoid masses that means primordia is not differentiated into stipe and pileus. At the later stages of infection, droplets of dark brown colour are formed on the surface of infected fruit bodies. Because of these dark colour droplets, the disease is called a wet bubble

Symptomatology

Wet bubble disease causes extensive damage by rotting and causing deformities in the whole fruiting body. If not managed well in time, the pathogen causes economic losses by damaging the entire crop. Wet bubble is characterized by the development of whitish mouldy growth of the mycelium on the casing surface and on portions of fruit bodies, which eventually spreads covering the entire cap and causing distortion of the affected mushrooms. The infected pin heads have amorphic shapes, not resembling the typical mushrooms. Similar types of symptoms were produced on inoculation of pinheads and mature fruit bodies with mycelial bits from spore suspension sprays or inoculation with agar culture discs of the pathogen. However, the appearance of symptoms and the subsequent disease development was much quicker when fruit bodies of the mushroom (*A. bisporus*) were inoculated with mycelia. Characteristic symptoms in this case appeared within 1 to 2 days of inoculation. The reactions of undifferentiated primordia were vastly hyperplastic and of tumorous nature.

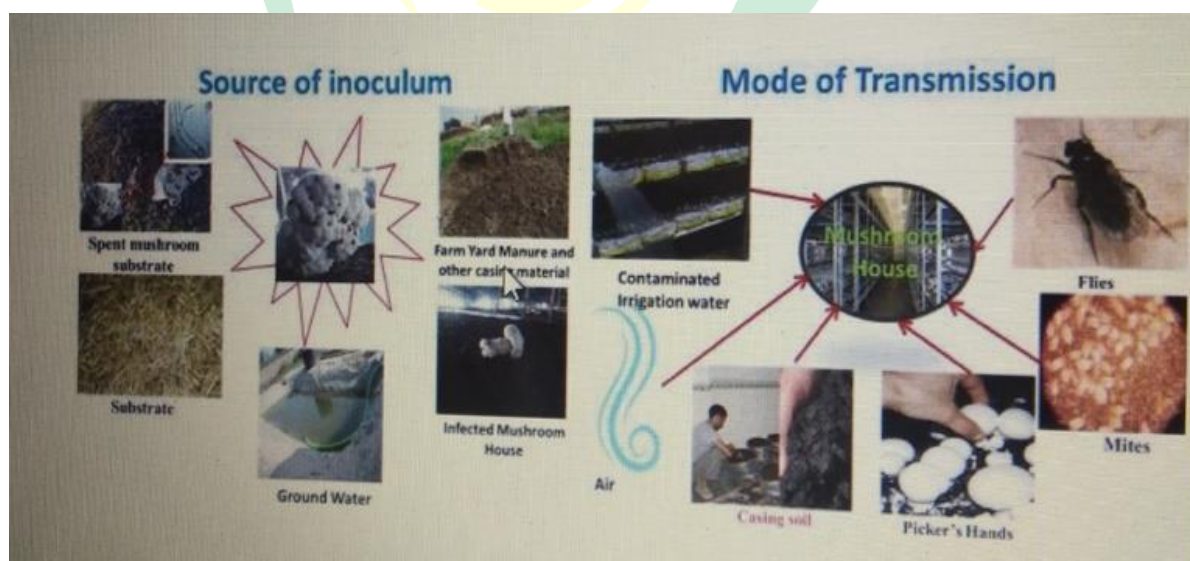
Finally, large irregular lumps of mushroom tissues were formed with no sign of differentiation or organogenesis. Dripping out of foul-smelling amber liquid as teardrops on the timorous bodies was consistently a typical sign of the wet bubble disease.



(Plate 1). Natural symptoms of wet bubble disease of white button mushroom.

Host range

Though *M. perniciosa* is a major pathogen of *Agaricus bisporus*, however, it is capable of infecting other mushroom species and crops like *A. campestris*, *Pleurotuseryngii* and *P. nebrodensis* and shiitake (*Lentinuse dodes*).



(Plate 2) : Source of inoculum and different modes of transmission

Spread

Spread of *M. pernicioso* occurs mainly through casing soil, air, water or maybe mechanically carried by mites, flies and human errors (Fig. 2). This pathogen can survive from many years in the form of chlamydospores in the spent mushroom substrate and in different cracks or unused areas of the growing rooms and are probably responsible for secondary infection.

Generally, *M. pernicioso* may resist the temperature up to 50 °C. However, the range may be exceeded beyond due to the genetic variability in the pathogen. Infected fruit bodies, spent mushroom substrate, farmyard manure, substrate material, groundwater etc. are the major sources of inoculum. *M. pernicioso* can also survive under high moisture conditions because it is also rich in genes encoding hydrophobins. Wet bubble disease spreads at a very fast rate and it therefore advisable to identify and manage the disease at the very early stage of infection i.e. when only 2-3 fruit bodies are infected in a single growing room.

Biology/physiology

M. pernicioso is very sensitive to comparatively high temperature i.e. >50°C. Potato Dextrose Agar (PDA) and Malt Extract Agar (MEA) are considered as the best medium to obtain maximum growth of the pathogen with optimum pH and temperature of 6.0 and 25°C, respectively. Mannose and asparagines are among the important source of carbon and nitrogen required for the growth and development of *M. pernicioso*.

Management

Quality compost and proper pasteurization or sterilization of casing soil is remained as effective means of management of *M. pernicioso* parasitism in edible fungi. Use standard crop management practices and be the earliest to jump to any disease control strategies. At the time of disease appearance, wear gloves in hands and sprinkles some pinches of common salt on the infected fruit body. Then take a piece of paper/newspaper and wrap up the infected fruit body, pluck it and bury it in a pit somewhere away from the mushroom house, after that sprinkle some common salt at the place from where the infected fruit bodies are removed. Don't add salt to the healthy area otherwise mushroom mycelium is also killed by the salt. Common salt is recommended only for spot treatment of wet bubble disease. For effective

management, one spray of Prabal/Derisom/Sheat guard(1-3ml/g/litre water) after casing is done (Prabal contains *Oryzussativus* + *Opuntiasp*, Derisom is the extract of *Pongamiaglabra/Pongamiapinnata* and whereas Sheath guard is a *Pseudomonas fluorescens* based biological fungicide followed by the second spray of Prabal/Derisom/Sheat guard 7 days after first spray (or before pinhead formation).

Spray of calcium carbonate (1g/litre water) is done on completion of each harvest followed by a spray of calcium chloride (1g/litre water) after 2 days. After each harvest of the crop pH level of the growing medium goes down to the acid level, which can be corrected back to the desirable level by adding calcium carbonate. Besides this, after first and second harvest, the incidence of bacterial diseases is also increased this is managed by the application of calcium chloride. Calcium chloride is also helpful in supporting the mushroom crop in drawing nutrition from the growing medium and resisting the disease pressure. Among chemical control, one spray of chlorothalonil (0.1%) immediately after casing and its second spray after 7 days or before pinhead formation. Besides chlorothalonil, drenching with any permissible fungicides of benzimidazole group (at 0.05% conc.) plus formalin (0.1%) is also effective against wet bubble disease.

After completion of crop harvesting, treat the used bags and room with formalin (2%) and keep the door closed for 3 days or adopt post-crop sterilization/cookout technique by keeping the room temperature at ≥ 70 °C temperature (for 3-4hr). Properly dispose of the spent mushroom substrate is most important to avoid the further spread of the disease. As quoted above in the text that casing soil is the most important source of inoculum for this disease. It is suggested to use 80-90% soil-less material (e.g. coir pith etc.) for the casing preparation and enhance its pH by adding lime (1%) into it and gypsum (2%) to stabilize the soil by reducing the dispersion of large soil aggregates into a smaller size. It is also helpful in maintaining the porosity of casing material and as results in creating continuous evaporation from the growing bags/beds. This whole process supports the fast growth and development of mushroom mycelium and eventually provides strength to resist the attack of a pathogen like *M. perniciosa*.

Conclusion

Among different parasites of edible fungi, management of mycoparasitism of *M. perniciosais* very difficult. It is tougher when the parasitism has to be managed curatively. Stage of manifestation of infection in the mushroom house and the time application of protection components (botanicals, microbial and synthetic pesticides) are very important. During the phase of colonization of substrate by the host fungi, application of any management strategy is of no use because of the presence of hydrophobins present on the surface of its hyphal surface. Hydrophobins prevent the attack of major parasite like *M. perniciososa*. However, care is required at the time of primordial formation. At this stage, natural openings are formed on hyphae, which are responsible for the invasion of parasitic fungi. Therefore, to prevent the attack of *M. perniciososa*, casing time and primordial formation stages are very important to apply any management strategy. Application of any antifungal compound can produce best results only when their protective applications are given to the crop and mushrooms are grown under good agricultural practices (GAP).

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