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REVIEW ARTICLE **OPEN ACCESS**

Dry bubble disease *Lecanicillium fungicola* (Preuss) Zare & W. Gams] of white button mushroom (*Agaricus bisporus*)

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ABSTRACT
White button mushroom (Agaricus bisporus) is one of the premium quality exportable mushrooms. This mushroom contains a very high amount of protein, Vitamins and other important minerals. As far we talk about the taste about the mushroom it also gets infected by some serious pathogens. Dry bubble disease of button mushroom cause by Lecanicillium fungicola is a serious disease, which degrade the mushroom from export or marketable quality. This pathogen causes small narcotic spot like lesions on mushroom body which later disrupt the cap and stipe layer. This pathogen disperses by casing soil, wind, insects and some time by the workers of the farm. Researchers found that the pathogen can remain viable for more than a year under soil. The disease effects in the quality degradation which eventually means economic loss.
Keywords: *casing soil, disperses, dry bubble, economic loss, pathogen, viable*

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INTRODUCTION

Edible mushroom cultivation in large commercial stage, transform the various types of house-hold and agricultural wastes to a source of nutritional food which also solve the problems like healthy and quality food and environmental sustainability. The rapid growth of world population is craving for high quality food which can be fulfilled by mushrooms as it is a great source of protein and it can replace meat, vegetables and milk products for majority [44].

There are around 1.5 million of fungus and among them only 14,000 species are estimated to produce fruiting bodies that are considered as mushroom [23]. All over the world known edible mushroom species are around 7,000 and among them 200 species are successfully grown and only 10 species of edible mushrooms can be produced in industrial/ commercial basis [8].

Lion's shares of mushroom production have been occupied by America, East Asia, and Europe which provide almost 96% of world's mushroom production. India supply 3 per cent of total production which is around 4.60 lakh metric tons [2]. White button mushroom is the most popularly consumable mushroom in India as it is packed with essential amino acids, fibres, vitamins, valuable minerals (mainly potassium and iron), low calories and fat [29].

In India, mainly four types of edible mushrooms are, *Agaricus bisporus* (white button mushroom), *Volvariella* spp. (paddy straw/straw mushroom), *Calocybe indica* (milky mushroom), *Pleurotus* spp. (oyster mushroom) are grown in commercial basis. Mushrooms are attacked by numerous biotic and abiotic factors like bacteria, fungi, insects, nematodes and viruses which cause some serious damage in mushroom crop directly or indirectly [36].

Verticillium fungicola (Preuss) Ware causes dry bubble disease in white button mushroom which is one of the major fungal problems in the button mushroom production. The disease symptoms are small necrotic lesions on fruiting body and partial tissue disruption on cap and stipe. Thus the stipe blow out and undifferentiated mushroom masses that is known as dry bubble. These diseased mushrooms become unmarketable which ultimately cause significant yield loss. The pathogen *Verticillium fungicola* can be objectified by special erect structured conidiophores with unequal conidia in large, slimy globose heads, verticillate conidiophores with 2-3 whorls of 3-7 phiallides in number [51].

Verticillium fungicola species is consists of phialide Hyphomycetes, hyaline which is having the entomogenous and fungicolous characteristics. The pathogen *Verticillium fungicola* causes various diseases in basidiomycetes. White button mushroom covering almost 40% share of world mushroom

market and this mushroom is seriously affected by *Verticillium fungicola*. The compost used for production of *A. bisporus* is a mixture of manure and straw. After the compost is fully colonized by the fungal mycelium, the top portion of the compost is covered with casing layer which is mainly prepared by alkaline peat soil. We can protect the crop from *V. fungicola* by maintaining a hygienic place and by using some specific fungicides. But the excessive uses of fungicides provides some adverse effects on the mushroom, as this pathogen increasing resistance against the fungicides [7, 14, 20, 48].

Farmers use a fungicide named as prochlorazmagnanese for dry bubble diseases, but it is reported that this fungicide is becoming less effective against the pathogen [20, 22].

Taxonomy

Verticillium is a genus of hyaline, phialide Hyphomycetes, conidiophores generally raises from aerial, and submerged erect hyphae and sometimes it can be differentiated from the subtending hyphae. More or less globose shaped conidia can found stick on the phialide head transversely [50].

In some cases these conidia can be found in chain formation in short to ellipsoidal to fusiform or falcate arrangement with pointed ends. In this arrangement the very first formed conidia is known as macroconidia which is larger than the after formed small conidia or microconidia. These fungicolous or entomogenous fungus colonies are very rapid grower in agar medium. They can grow 15-30mm in 10 days on Potato Dextrose Agar (PDA) or in Malt Extract Agar (MEA) media.

At the beginning, *Lecanicillium* was being counted in paraphylatic genus from which other different genera like, *Beauveria*, *Microhilum*, *Isaria* have arisen(39). The fasciculate conidial arrangement is not to be specific in generic level, but if it is present it is a standard to level between *Lecanicillium* and *Simplicillium*.

Genetic diversity

The dry bubble disease for the very first time and they described them as '*la môle*'. The scientists reported that the fungus *Hypomyces perniciosae* is causing the diseases, and it can express in two different forms: a big Verticillium-like conidia, a chlamydospores and second form is having only small Verticillium-like conidia(9).

Difference between dry bubble and wet bubble diseases, also named two different fungi as the causal organism of the diseases. As per Smith a fungus with small conidia named *Cephalosporium constantinii* is responsible for dry bubble diseases and it have similarities with Verticillium- like fungus(37).

Ware also worked on dry bubble diseases and suggested the name *Verticillium malthousei* as the causal organism of the disease as he thought the isolated species was not *Cephalosporium constantinii* [45].

Verticillium fungicola, Cephalosporium constaninii and *Verticillium malthousei* are from the same species(18). Short while ago, on the basis of small subunit rDNA sequences and ITS (Internal Transcribed Spacer) region it was concluded that *V. fungicola* is more close to the insect-pathogenic species of *Lecanicillium* genus than to plant-pathogenic species of *Verticillium* genus (51).

The different species of *Lacanicillium* is responsible for the diseases of dry bubble in different parts of the world as the climatic conditions are responsible for the diseases caused by different species.

Host range

Lecanicillium fungicola can infect *Agaricus bisporus*, *A. bitorquis* and *Pleurotus ostreatus*. The pathogen isolated from *Pleurotus ostreatus* can cause in healthy *P. ostreatus* which can be re-isolated, thus it certified the Koch's postulates (21).*Lecanicillium fungicola* has been isolated from various basidiomycetes e.g. *Hypoloma capnoides, Laccarialaccata,* and *Henningsomyces candidus* [17, 50]. Although the pathogenicity is not proven experimentally to all the mushrooms, still it makes a possibility that *L. fungicola*can infect a ranges of mushroom species.

L. fungicola is closely related to insect pathogen, and it can cause damage to insects [1, 49]. Apart from all these the evidence of pathogens entomo-pathogenicity is still very week to prove itself.

At the beginning, *Lecanicillium* was identified as a paraphyletic genus from which different genera like *Beauveria, Microhilum, Isaria* also have risen [39].

The most nearly matched genus is *Simplicillium* species that mainly have single phialides that is arises from prostate hyphae. Only a countable species of *Lecanicillium* have single conidia, risen subtending hyphae, phialides which are reduced at lateral ends.

Dispersal

Insects are associated with the dry bubble diseases [45]. A mix population of *Leptocera heteroneura* and *Megasalia halterata* was very useful for the spreading the spores in agar medium and the mix populations of these insects can spread the diseases from one effected farm to another farm [10].

Air also can be a effective path of dispersal but in their experiment there was no effective dispersal at wind speed up to 10.75 m/s, but dispersal by wind and equipment and employees was found very effective.

The initial stage of the mushroom production was affected by the insects, but in exponential spread of the diseases was due to the watering [47].

Primary source of infection

The spores of *Lecanicillium* can remain viable up to more than a year while it present in soil [10], and on mushroom farm the pathogen can remain viable for 7-8 months, even if the condition is dry. Due to the poor sanitation and hygiene or wind blow the spores of *Lecanicillium* become reservoir in the site.

The casing soil can be a source of the infection [45]. The pathogen cannot survive if the temperature is more than 40°C, and the composting process is done at a temperature of 70-80°C, so the casing material can be a source of the diseases [41]. Low pH and anaerobic conditions are not favourable for the growth of *L. fungicola*.

The surface peat was the main cause of *L. fungicola*attack as it may survive on basidiomycetes that mainly grow on the surface peat [43].

Presently, in commercial farms they used mainly black peat which is collected from the lower peat layer and it seem to be very less habitat for *L. fungicola*. Replacement of casing material of clay, humus, and loam with mixture of sphagnum peat, carbonate and sand is a resulted a considerable reduction of the pathogen in the mushroom industries.

Wild infected mushroom or an infected farm adjacent to a healthy farm could be an important primary source *Megaselia halterata* a kind of phorid flies attracted by the compost of *Agaricus bisporus* also can be a primary infection source. Rather than all these contaminated equipments also can be a great source of the infection to spread [51].

Ecology of*L. fungicola*inthe casing

The spores of *Lecanicillium fungicola* cannot infect *Agaricus bisporus* vegetative mycelium while in compost [6].

That spores do not germinate immediately, in natural soil and peat most of the spores do not germinate in 7days and the few germinated spores was having short germ tubes [10]. But in sterilized pear and soil the spores were readily germinate and was very extensive after 7 days of growth. This phenomenon of fungal propagules growth and germination is due to the active soil microorganisms, this is also known as soil fungistasis [28].

Germination of *L. fungicola* needs an external output of nutrition [10]. In casing the pathogens cannot germinate without the immediate help of Agaricus hyphae. After the germination of pathogen hyphae the pathogen grows along with the Agaricushyphae, as a result the pathogen soak the nutrient form the mushroom body.

The pathogen can grow in sterile water and if we add nutrients in the water it can accelerate the growth of germ tube greatly. It was thought that Carbon was the key factor [40]. Fungistat is not the only cause of nutrient scarcity, but the inhibiting compound production was also the cause of spore inhibition. In the study it was shown that *L. fungicola* spores become dormant and germinate when *Agaricus* spores colonies on the casing, the pathogen awaits still the condition become favourable for the pathogen to grow.

Macroscopicsymptomsdevelopments

After *A. bisporus* gets affected by *L. fungicola* it shows 3 types of symptoms, as:

- Necrotic lesions: These are brown to light-brown or ash coloured spots develop on the cap or stipes. These spots later develop to warty outgrowths.
- Stipe blow-out: Mushroom sporophorebecome partially deformed.
- Dry bubble: white coloured mass can be seen on mushroom bags.

The severity of infection mainly depends on the time inthis study harvested mushrooms from *Lecanicillium* effected bags at vary points of time after apply casing layer [24]. Diseases severity was minimum when we applied pathogen at the time of casing of the bags which later geared up its pathogenicity after 14 days of casing.

L. fungicola introduced in the mushroom bags before the colonization of *Agaricus* it lost its germination capability [24]. He also suggested the inoculation period of the pathogen at 14 days, 21 days and 28 days after applying of casing layer. Thus it results in smaller numbers of mushrooms with adequate symptoms.

If mushrooms are found without any symptoms it's not necessary that it is uninfected as hyphae and conidia of *L. fungicola*can found on the cap surface before it develops any kind of discoloration [30].

Disease Resistance

In many studies it has been observed that mushrooms are investing in their defence apart from their ability to produce fruiting bodies. But it still yet to be confirmed that the poisoning of the mushroom is their adaptive advantage or it is just like any other by-products. Mushrooms also use their defence against microbial effects. Dry bubble affects *Agaricus bisporus* to form its fruiting body which eventually damages

its spore generating capability. *Agaricus bisporus* stains uses for commercial purposes are majorly selected by their ability of reproduce not for defence.

Narcotic lesions on the *Agaricus bisporus* cap of brown partial resistance cultivar they found in this type of cultivar there are less sporulation and hyphae of the pathogen *L.fungicola* [11]. This enlightens the hypersensitive response in which the damaged on infected cells die and capsule the infection. 17 stains of *Agaricus bisporus* to find any susceptibility correlation between the mushroom and pathogen, but in all cases they found negative correlation [33].

The various biochemical mechanisms that is responsible for the diseases in *A. bisporus.* Reveal that resistance that is partial is not requisite outcome of active defence system [26]. Other characteristics that are not related to defence can also act against infection. Time of pathogen affect is a major impact of disease.The stains that fruits earlier causes less in diseases [27].

Induced Resistance

Many bio control agents elicits induce some kinds of Induced Systemic Resistance (ISR) in plants [42]. It is a type of resistance in which plant's resistance boosted against a variety of adverse condition. Induced resistance can be triggered by some beneficial micro-organisms, but few times this also helps the pathogen [12]. When pathogen is observed in a certain plant part, the resistance introduced in other parts of the parts, as a result the plant become capable of systemic acquired resistance (SAR). In case of mushrooms mainly *Agaricusbisporus* no such kinds of resistance (both ISR & SAR) still have been found [26].

Biological Resistance

For effective fungal pathogen control of plants soils must have use microorganisms which have antagonistic characteristics [16]. The screening of *Pseudomonas* spp. as a bio control agent could prove to be an effective mechanism against the dry bubble disease of *Agaricusbisporus*. When*A. bisporus* starts its colonization the relative number density of the bio control agent (*Pseudomonas*) expand [13, 31].

The super effective traits of *Pseudomonas* for which it become a very effective bio agent against the pathogens of plants(46). The ability to grow on a vast substrate, high broadening rate, quality to grow at a very low temperature, is the mechanisms which alienate other microbes. Some resistance methods for bio control methods and breeding, i.e. use of *Lippiacitriodora* (Lemon verbena), *Thymus vulgaris* (Thyme oils), *Origanum vulgare* (Oregano) [38, 32]. Spent mushroom substrate (SMS) treated with teas eventually reduce the effects of *L. fungicola* in *A. bisporus* [19].

CONCLUSION

Mushroom is one of the most nutritious foods. Button mushroom is in high demand throughout the world for its deliciousness. In mushroom farm growers facing this dry bubble disease caused by the pathogen *Verticilliumfungicola* (*Lecanicilliumfungicola*) which 1st leaves a brown spot on the cap of mushroom which later disrupt the cap and the stipe of the mushroom. The pathogen spread by the infected soil used for the casing soil. It remains viable under soil for more than a year. Scientists have found that the pathogen can also be spread by the water uses for spraying, and by the insects of mushrooms beds. When a bed infected by the pathogen it releases some spores as this is a fungal diseases, later insects sat on the infected beds it carries the spores to the healthy beds. The pathogen needs external nutrition for its growth, which it gets from the mushroom mycelium. When the mycelium of the mushroom grows the pathogen soaks nutrition from the sporophore. The pathogen can be control only by using proper sanitation of mushroom growing room.

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