

Review Article

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Microbial Diversity and their Role in *Agaricus bisporus* Production

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ABSTRACT

Mushrooms are a significant food crop for large population throughout the globe. The main edible mushroom is the button mushroom (*Agaricus bisporus*), a perfect example of economical food production which is manufactured on a specific manure delivered from farming residue materials. In mushroom cultivation successive microbial community consists of a variety of microorganisms including bacteria, actinomycetes and fungi at first breakdown the straw to form lignin humus complex and discharge the gases, and then metabolise the cellulose and hemicellulose into compost microbial biomass. This decayed straw along with microbial biomass turns into an organic and inorganic nutrient source for the mushroom mycelium and these microflora play a main role during the different stages of composting and resist the growth of other competitor in the crop production. In most farms, seasonal cultivation of this mushroom is being practiced, but they are vulnerable to a spread of viral, bacterial and fungal diseases. Standardization of compost composition and composting processes, disinfection of casing soil, cultural practices, and sanitation has significantly reduced the prevalence of those moulds in mushroom crops.

Keywords

Agaricus bisporus,
Compost, Casing,
Microbial ecology,
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Introduction

China, Malaysia, India, and Ireland are driving in worldwide mushroom production (Hanafi *et al.*, 2018). China is the world's biggest grower of eatable mushrooms, providing more than 30 million t, or 87 % of worldwide contribution (Royse and Beelman 2016). Multiple thousands mushroom species exist in nature, however just around 22 species are cultivated

(Raj and Thangaraj 2008). *Agaricus bisporus* is a heterotrophic edible basidiomycete which is the most famous consumable mushroom on the planet (Atila 2017). Mushrooms are devoured for their deliciousness, they are very rich in proteins with a significant substance of fundamental amino-acids and because of low starch and cholesterol, they suit diabetic and heart patients (Gupta *et al.*, 2019; Sharma *et al.*,

2017). The initial step of mushroom cultivation is the compost production and it is a complex microbial process in which microorganisms decompose and stabilize the organic substrates under controlled conditions (Johri and Rajni 1999) and many factors involved in the composting process like microbial succession, raw material used for compost preparation, pH, temperature, aeration and acidity or alkalinity (Antunes *et al.*, 2016).

Compost bacterial and fungal community

Microbial ecosystem of the compost changes drastically during different stages of fermentation in the mushroom crop production, bacterial community increases with every step of mushroom cultivation compare to fungal community in mushroom cropping process (Vieira and Pecchia, 2018); (Siyoun *et al.*, 2016); (McGee *et al.*, 2017). Phase I in mushroom crop production is a thermobiological process which involves the bioconversion of simple carbohydrates and proteins by mesophilic microorganisms such as *acinetobacter sphingomonas*, *Solibacillus*, *pseudomonas* and *comamonas* known as pioneer community (Kertesz and Thai 2018).

Mesophilic microorganisms present in Phase I digest easily degradable polysachride, which raise the temprature and cause the shifting of mesophilic microorganisms to thermophilic microbial community (Smith *et al.*, 1995). In phase I the dominant bacterial phylum are Firmicutes, Proteobacteria, Actinobacteria, Bacteroidetes and Thermi. During Phase II heat treatment is given to the compost which stimulates the growth of thermophilic microbial community and protects the crop from its parasites (Mouthier *et al.*, 2017); (Vieira and Pecchia 2018). Firmicutes and Proteobacteria phyla are the hydrogen producers from the wheat straw compost (Valdez *et al.*, 2017) whereas Actinobacteria

plays a main role in compost production responsible for cellulose decomposition, they are consistently exist and shows maximum growth in the later phase of composting (Wang *et al.*, 2011) ;(Zhang *et al.*,2014). In phase III during spawn run there is decrease in Actinobacteria and Firmicutes and increase in Proteobacteria (Kertesz *et al.*, 2016); (Carrasco *et al.*, 2019).

Fungi involve a significant role in biological biomass pretreatment because of their strong ligninolytic action (Stajić *et al.*, 2016). Thermophilic fungi promote the growth of *A.bisporus* by removing waste from compost and assimilate the free ammonia produced in Phase I and help to stimulate the growth of *Agaricus* mycelium (Ross and Harris 1983); (Straatsma *et al.*, 1994).

Microbial ecology of casing layer

Casing is the top covering placed on the substrate, colonized by the host mycelium and it helps to stimulate the fructification in the crop. Quality, yield, and uniformity of the mushroom crop production depend upon the casing layer (Noble and Gaze 1996).

Bacterial population present in casing influence the *A.bisporus* production by releasing growth stimulating substances (Hume and Hayes 1972).

Most of bacterial species present in casing is mainly related to *Pseudomonas*, *Pedobacter* and *Caulobacter*, act as growth promoting strains, promote mycelia growth or fruiting body formation (Schisler 1982). *A.bisporus* produces 1-Octen -3-ol and ethylene which has inhibitory role in the process of fructification and some bacteria has the property to lower the level of this compound which promote the growth of mycelium (Zhang *et al.*, 2016); (Kües *et al.*, 2018); (Fermor *et al.*, 1991).

Role of bacteria and fungi as disease causal agents

The white button mushroom is vulnerable to numerous diseases that unfavourably influence the crop productivity. There are different types of *Trichoderma* strains which are responsible for the serious diseases in the mushroom crop like green mould (Seaby 1996). Cobweb disease of white button mushroom caused by the *Cladobotryum dendroides* responsible for the major loss in the crop (Grogan 2008). Bacterial pathogens involved in mushroom diseases mostly present in casing material and cause the Bacterial blotch in the crop. *Pseudomonas tolaasii* causes Agaricus brown blotch and develop light yellow injuries which result in tissue damage and *Pseudomonas gingeri* causes Agaricus ginger blotch (Wells *et al.*, 1996). *A.bisporus* is seriously affected by a disease, for example, *Mycogone pernicioso*, the causal agent of Wet Bubble Disease, Wet Bubble Disease causes economic loss in button mushroom overall (Sharma and Kumar 2000).

Beneficial role of bacteria and fungi in mushroom crop production

Mushroom growth promoting bacteria (MGPB) are potential agent to increase the growth of mushroom, MGP microbes promote the mycorrhizal growths, shortening the soil composting procedure, improving nature of the substrate by secretion of secondary metabolites and help in mushroom fructification (Pratiksha *et al.*, 2017). *A. bisporus* produces 1-aminocyclopropane-1-carboxylic corrosive (ACC) act as self inhibitory compound degenerate by the 1-aminocyclopropane-1-carboxylic corrosive (ACC) deaminase producing bacteria present in the casing layer and reduce the ethylene level which obstruct the fructification (Chen *et al.*, 2013). *Pseudomonas*, *Azotobacter*, *Bacillus*, *Paenibacillus*, *Bradyrhizobium* has a essential and stimulatory role for the growth

of mushroom while indicating threat against competitive molds, have been accounted as biofertilizers (Zarenejad *et al.*, 2012); (Jadhav *et al.*, 2014); (Pratiksha *et al.*, 2017). *Mycothermus thermophilus* (*Scytalidium thermophilum*) the thermophilic fungus, has been depicted as significant for development, improvement and yield of *A. bisporus* (Natvig *et al.*, 2015). Disease management with the help of biological methods is the best alternate over the other methods, *Bacillus velezensis* QST 713 and *Bacillus amyloliquefeciens* MBI 600 are use to control the green mould disease and affect the growth of *T. aggressivum* (Milijaševi *et al.*, 2015); (Pandin *et al.*, 2018).

Mushroom cropping comprises of a number of events and microbial population dynamics varies from compost, casing to fruit body formation and it present in large amount in mushroom compost compared to casing and fresh mushroom samples.

There are many significant factors that impact the different varieties of microorganisms in the substrate such as type of cultivation, substrate material, fermentation time and type of wheat straw material. The dominating bacterial community present in the mushroom compost are members of the phyla Actinobacteria, Bacterioidetes, Firmicutes and Proteobacteria and except the basidiomycetous fungi *A. bisporus*, most fungal species found within the mushroom growing medium tend to belong to the phylum Ascomycotina.

Pseudomonadales contain the genera *Pseudomonas* are the dominant bacterial population in compost and casing layer is associate with the promotion of mushroom fructification and metabolise the volatile compounds which act as inhibitory component in the *A.bisporus* primordial formation and acts as bioinoculant and Mushroom Growth Promoting Bacterial (MGPB) to increase the yield in mushroom crop production.

Table.1 Diversity of bacterial communities in compost and casing material

| Substrate | Stage | Microbes (bacterial community) | Characteristics | Reference |
|-----------|---------------------------|---|--|---|
| Compost | Phase I | <i>Solibacillus, Comamonas, Acinetobacter, Pseudomonas, Sphingomonas</i> | Mesophilic pioneer microorganisms | (Basotra <i>et al.</i> ,2016) |
| | | <i>Thermophilic Bacillus Paenibacillus, Actinobacteria (Corynebacterium Streptomyces)</i> | Theomorphilic microbial community | (Zhang <i>et al.</i> , 2014); (Kertesz <i>et al.</i> , 2016); (Vieira and Pecchia 2018) |
| | Phase II of composting | <i>Proteus, Micrococcus, Aerobacter</i> | Consume the ammonia produced in phase I and promote the growth of mycelium of button mushroom. | (Mouthier <i>et al.</i> , 2017); (Chang and Miles 2004). |
| | | <i>Actinobacteria</i> | Cellulose decomposition during compost decomposition. | (Wang <i>et al.</i> , 2011); (Zhang <i>et al.</i> , 2014) |
| | | <i>Bacillus</i> | Degradation of organic component in the form of nutrition for the growth of <i>Agaricus bisporus</i> mycelium. | (Vieira and Pecchia 2018) |
| | Compost conditioning | <i>Pseudomonas taiwanesis</i> | Work as a hetrotrophic nitrifier. | (Kertesz <i>et al.</i> , 2016); (Székely <i>et al.</i> , 2009); (Vieira and Pecchia 2018) |
| | | <i>Actinobacteria</i> | Nitrogen fixing bacteria. Major bacterial phylum present in casing layer. | (Choudhary 2011) |
| | <i>Pseudomonas putida</i> | Mushroom fructification and metabolise the volatile compounds which has an inhibitory role in the formation of <i>Agaricus bisporus</i> primordial. | (Riahi <i>et al.</i> , 2011) | |

Table.2 Diversity of fungal communities in compost and casing material

| Substrate | Stage | Microbes(fungal community) | Characteristics | Reference |
|-----------|------------------------|--|---|--|
| Compost | Phase I | <i>Lewia, Rhizomucor, Aspergillus</i> | Mesophilic fungi | (Kertesz <i>et al.</i> , 2016) |
| | | <i>Aspergillus spp., Rhizopus oryzae, Trichoderma viride, Chaetomium spp., Penicillium spp., Alternaria spp, Talaromyces, thermomyces</i> | Dominant mycoflora in the initial phases of composting, thermophilic fungi has positive influence for the growth of <i>A.bisporus</i> by decreasing the ammonia concentration and it immobilize the nutrients so that it is easily available to the mycelium of mushroom. | (McGee <i>et al.</i> , 2017) |
| | Conditioning process | Thermophilic fungus <i>Scytalidium thermophilum</i> | Dominating cellulytic ascomycete, help in degradation of polymeric carbohydrates | (Vajna <i>et al.</i> , 2012); (Kertesz <i>et al.</i> , 2016); (Basotra <i>et al.</i> , 2016) |
| | Phase II of composting | <i>Thermomyces ibadanensis, Thermomyces lanuginosus and Scytalidium thermophilum</i> | Most abundant thermophilic fungal species | (Zhang <i>et al.</i> , 2014) |
| Casing | | <i>Paecilomyces niveus, Thermomyces lanuginosus, Aspergillus spp., Myceliophthora spp., Sordaria spp., Candida subhashii, Lecanicillium fungicola, and Cercophora spp.</i> | Dominant fungal community in casing | (Kertesz <i>et al.</i> , 2016) |

Thermophilic fungi have positive influence for the growth of *A.bisporus* by decreasing the ammonia concentration and it immobilizes the nutrients so that it is easily available to the mycelium of mushroom.

The main constraints with the good productivity of this crop are microbial diseases, caused by different types of pathogens that result the partial or total failure of the crop. Disease control with the help of chemical fungicides stimulates the production of harmful component which effect the environment adversely however, evolution of resistance to fungicides and host sensitivity to fungicides are serious issues.

Throughout this review, it's emphasize that understanding the structure, dynamics and usefulness of the mushroom microbiota present in at different stages of crop cycle provides a foundation to change and improve current cultivation ways. More detailed and advanced studies are needed to explore the biocontrol agents and to develop consortia of bacteria and fungi that may be utilized in bioaugmentation and may be a potential tool and a chance to modify the mushroom crop production.

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