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Research Paper

Mycobiota of compost, air, fruiting bodies of *Pleurotus ostreatus* (EM-1) and the mycotoxigenic potential of some resident *Aspergillus* species

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Abstract. The commercial and cottage industry entrepreneurs in Ghana have cultivated edible mushrooms such as *Pleurotus ostreatus* with appreciable success. However, the presence of fungal competitors and potential pathogens in the substrate obstructs yield. Selected farms in Greater Accra (6) and Central Region (1) were sampled for baseline data on the resident mycobiota of the compost (*Triplochiton scleroxylon* 'wawa' sawdust), fruiting body and aeromycoflora of the cropping rooms using conventional serial dilution technique and the Plate Exposure method respectively. Twenty (20) fungal species belonging to eleven (11) genera *Aspergillus, Cladosporium, Didymella, Fusarium, Mycelia, Penicillium, Rhizopus, Trichoderma, Verticillium* and *Saccharomyces* were encountered in the compost. The total fungal populations in compost varied significantly and with the highest recorded by E90 Mushroom Farm (4.0 to 5.24 log_{10}CFU/g) whereas 4E Farm (2.28 to 2.93 log_{10}CFU/g) obtained the lowest population. Eighteen (18) fungal species from ten (10) genera were isolated from the fruiting body of *P. ostreatus* from the farms. The most predominant species were *Aspergillus* species followed by *Cladosporium, Penicillium*, *Trichoderma, Rhizopus, Saccharomyces* and *Talaromyces*. The total mycoflora population resident in the fruiting body of the mushroom was lowest for Immaculate Gold Enterprise (2.59 to 3.22 log_{10}CFU/g) whereas PMC Mushroom Farm (4.71 to 5.34 log_{10}CFU/g) ranked highest. Aeromycoflora analysis showed nineteen (19) different fungal species isolated from the seven farms. Findings from the present data indicate fungi isolates from cropping rooms were also isolated from fruiting bodies and the compost. Potential toxin-producing *Aspergillus* species (*A. flavus, A. parasiticus, A. niger, A. alutaceus, A. fumigatus*) were present in the compost, the fruiting body of *P. ostreatus* and aeromycoflora.

Keywords: Mycobiota, *Trichoderma harzianum*, fruiting body, *Pleurotus ostreatus*, aeromycoflora of cropping rooms, toxin-producing *Aspergillus* species.

INTRODUCTION

Mushroom cultivation is currently a prominent biotechnological industry for the valorization of agro-industrial residue as bio-converters of lignocellulose waste in useful protein food with additional medicinal properties (Chang, 2007; Gregori *et al*., 2007). Hence, mushroom growing is a simple microbiological technique for commercial agro-waste recycling on a wide scale Stojchev, Asan, and Giicin (as cited in Altaf *et al.*, 2022). Cultivated mushrooms are an important food source for many people around the world, with a global production

estimated at over 10 million tons per year (FAO, 2014). In China alone, two-thirds of the production of edible mushrooms worldwide is harvested and form a more traditional role in food and medicine than in many Western or African countries. There are several important edible mushroom genera grown commercially. However, *Agaricus bisporus* (the button mushroom) especially white in colour, is the most extensively farmed edible mushroom, accounting for 31.8% of global mushroom output Choudhary, Agarwal, and Johri (as cited in Altaf *et al.*, 2022) while *Pleurotus* species follow (*P. ostreatus, P. eous, P. sajor-caju, P. florida, P. eryngii, P. pulmonarius, P. citrinopileteus*, etc.) are cultivated commercially. Others such as *Lentinula edodes* (shitake), *Auricularia* (*A. polytricha, A. auricularia-judae, A. auriculata*), *Flammulina velutipes* (enoki) and *Volvariella volvacea* (paddy straw mushroom) are also cultivated commercially (Royse, 2014; Chang and Miles 2004). In many African countries, large to moderate scale mushroom farms exist, cultivating mainly *P. ostreatus* (Mwai and Muchane, 2016). The mushroom industry is similar to the fruit and vegetable production industry, where it is often subjected to growing pressure to meet the growing demand of consumers. That notwithstanding consumers also demand healthier and safer goods. Mushrooms are usually cultivated under controlled indoor conditions; however, they are exposed to aeromicroflora of which some are potential mycotoxigenic species. Vijay and Sohi (as cited in Sarker *et al.*, 2021) reported that the mushroom substrates also harbour many weed fungi which act as competitor moulds either by competition for food material or through the production of toxic substances. Wiafe-Kwagyan *et al.* (2023) recently reported on the effect of mushroom pathogen *Trichoderma harzianum* Rafai on the quality, growth, and nutritional quality of *Pleurotus ostreatus* EM-1.

According to Raman *et al.* (2021), *Pleurotus* mushroom cultivation is most suitable and profitable in all three climatic conditions tropical, subtropical, and temperate regions. *P. ostreatus* are saprophytes which grow by degrading natural lignocellulose substrates, which are commonly available in large volumes as agricultural or industrial by-products. Many substrates which are used in the cultivation of *P. ostreatus* as well as other *Pleurotus* species have been listed by Cow (1991) and Howard *et al.* (2003). In Ghana, the primary and most common substrate for the production of *P. ostreatus* is 'wawa' sawdust (*Triplochiton scleroxcylon*) until when Frimpong-Manso *et al.* (2011) and Wiafe-Kwagyan *et al.* (2017) showed that rice waste which abounds in Ghana can be used as a substrate for the successful cultivation of *Pleurotus* to ameliorate environmental pollution by rice lignocellulose with prospective economic advantages.

The method for commercial cultivation of mushrooms on substrates can be divided into three (3) groups. Firstly, there is the cultivation of many wood-degrading mushrooms that are traditionally grown on wood or logs or harvested from trees e.g., *Lentinula* is grown on artificial logs of compacted, sterilized sawdust, while others e.g., *Flammulina* or *Auricularia* are cultivated on a partially composted mixture of sawdust and other components (bran, straw, corncob) which is sterilized at high temperatures (121°C for 15 to 20 min) before inoculation with the mycelium of the mushroom (Chang and Miles, 2004; Sanchez, 2010). During composting period varying up to 28 days, there is a change in pH, temperature, nutrient quality of substrate and changes in the microbial successive profile of the substrate which renders it suitable for the mushroom to compete favourably with the residual microorganisms in the substrate (Obodai and Odamtten, 2013).

The second group of cultivation methods either uses uncomposted substrate directly or partially composted substrates that have not been subjected to a rigorous sterilization process. This method is commonly used for *Pleurotus* and *Volvariella* species. However, *Pleurotus ostreatus* is grown on sterilized sawdust or rice straw substrate appropriately amended with lime to give favourable environmental conditions for the development of the inoculated mushroom spawn mycelium to have selective growth over other residual competitor organisms (Wiafe-Kwagyan *et al.*, 2017; Kortei *et al*., 2016; Chang and Miles, 2004).

The sterilized compost bags (70 in one oil drum barrel) are inoculated with a separately sterilized spawn supply nurtured on soaked grains (mainly sorghum) before incubation in the cropping room for fruiting body development. For *Pleurotus*, the growth of the mycelium and production of the commercial fruiting body is not dependent on the mushroom itself, but also on bacteria and other fungi in the substrate (Kertesz and Thai, 2018). These microorganisms (fungi and bacteria) play critical roles at several different stages of production including the conversion of lignocellulose feedstock into a selective nutrient-rich compost for mushroom growth; interaction with the fungal mycelium during hyphal elongation and proliferation through the substrate and induction of fruiting body formation during cropping (Kertez and Thai, 2018). Banfi *et al*. (2021) also confirmed the important role bacterial microbiome plays prior to *P. ostreatus* spawning (e.g., by the production of antimicrobial compounds, increasing the accessibility of celluloses). According to their findings, *P. ostreatus* can later use these microbes partially to build their biomass, but the microbes or their cell-free filtrate have a role also in inhibiting competing microbes without hindering *P. ostreatus* growth hence promoting fruiting body formation.

In addition, several bacterial and fungal taxa act as pathogens of the mushroom crop causing either reduction in yield or severe loss in quality. Amin *et al*. (2021) reported improper pasteurization of compost and casing soil can be the major source of diseases. They further iterated that once the disease is introduced on the farm it can be carried out by different agencies like air, water, machines, and workers. Recently, Wiafe-Kwagyan *et al.* (2023) recorded

the effect of green mould disease fungus (*Trichoderma harzianum*) on the growth, yield and nutrient of *Pleurotus ostreatus* EM-1. Their finding supported previous findings by He *et al*. (as cited in Chen *et al*., 2021) also documented the white mould as a serious disease affecting the cultivation of *Morchella* spp. in China and it reduces the production and quality of morels during cultivation, storage, and transportation. Hassan *et al.*, (2022) isolated two mushroom pathogens *T. harzianum* and *Pseudomonas tolaasii* on *Agaricus bisporus*. However, their findings suggested that two desert weeds *Atriplex tatarica* and *Haloxylon salicornium* inhibited the growth of these two pathogens thereby increasing yield and improving the quality of fruiting bodies.

There is ample information on commercial mushroom farming in Ghana and where to buy mushroom spawns and compost. Small-scale mushroom farms have emerged in Southern Ghana because of the introduction of the National Mushroom Development Project aimed at promoting the economic welfare of rural communities Sawyer (as cited in Kortei *et al.*, 2018). There are 20 mushroom spawn and Compost Suppliers and Exporters in Ghana (https://ghana.tradeford.com). However, there is only one Centre which is internationally accredited namely, The Food Research Institute of the CSIR, Ghana.

Sharma *et al.* (2017) stated that in many instances, there is a complete crop failure of *P. ostreatus* and other cultivated mushrooms depending on the infection and quality of compost and spawns used not excepting the environmental conditions in the incubation and cropping houses which exacerbates the problem. Kortei (2015) stated that 64% of the Ghanaian mushroom farmers of *Pleurotus ostreatus* expressed dissatisfaction about the process of steam sterilization in oil drums of the spawn and compost. They attributed their low cropping and heavy financial losses to the inadequate sterilization of substrates and spawn grains leading to the proliferation of pathogens and the other competitors in the compost bags.

In recent years much emphasis has been placed on the provision of healthy and safe foods for human consumption by the publication of the International Standards Organization (ISO Standard EN22000) which spells out the specific requirements for a safe food management system. Manufacturers in the food chain need to demonstrate their ability to control food safety hazards to ensure their products are always safe for human consumption. ISO EN22000 is a combination of prerequisite programme, Good Manufacturing Practice (GMP) and Hazard Analysis and Critical Control Point (HACCP). While GMP applies to food processing that takes proactive steps to ensure that products are of good quality and safe to eat by eliminating the use of raw materials with poor quality. GMP addresses several issues such as premises, facilities, manufacturing, storage and distribution operations, sanitation, cleaning schedules, personal hygiene of workers, provision of protective materials, the procedure for a complaint of defective products as well as product recall and management responsibility.

HAACP is designed to prevent the occurrence of food safety hazards by ensuring that controls are applied at any point in food processing and cultivation where such critical situations could occur. It is therefore a preventive system to correct problems before they affect the safety of the food rather than inspection of finished products. Therefore, HACCP is based on the principles of a management system in which food safety is addressed through the analysis and control of biological, chemical, and physical hazards from raw material production, procurement and handling, to manufacturing, distribution and consumption of the finished product. In light of the report of Kortei (2015) that 64% of the farmers of *P. ostreatus* are dissatisfied with the sterilization of compost and spawns using the steam oil drum method we set out to critically look at the critical operational methods which result in contamination of substrate by resident fungi serving as competitors or pathogens. Results will provide baseline information for formulating future appropriate GMP and HACCP procedures for mushroom cultivation for increased production yield and quality mushrooms produced. This paper reports the sources and mycobiota profile and fungal species which may serve as pathogens or competitors in the substrate bags in the cultivation of oyster mushrooms. Cropping rooms of selected seven (7) mushroom farms in the Greater Accra Metropolitan Area and Kasoa in the Central Region of Ghana. Potential mycotoxin-producing species whose metabolites might adversely influence the nutrient and medicinal qualities of the mushroom were identified.

MATERIALS AND METHODS

Sample collection

The compost bags were collected from Greater Accra and the Central Region of Ghana using the list of dedicated Mushrooms Farmers provided by the Mycology Division, Food Research Institute of the Council for Scientific and Industrial Research, CSIR Ghana, Accra.

Greater Accra Region

E90 Mushroom Farm (Ogbojo); Immaculate Gold Enterprise (Nii Boi Town, Lapaz); Delabless Mushroom Farm (Adenta); 4E Farm (Ogbojo, Ashaley Botwe); Edeyef Mushroom Farm (Anyaa, Awoshie); PCM Mushroom Farm (Ashaley Botwe).

Central Region

Kwesi Babs Mushroom Farm (Kasoa)

The samples of mushroom and corresponding compost

bags were stored in sterile and sealed polyvinyl translucent bags and were transported to the Mycology Laboratory at the Department of Plant and Environmental Biology, University of Ghana, Legon.

Samples of the fruiting body with the green mould and the corresponding compost substrates were also placed in sterile bags as described above using a previously sterilized large forceps using 1% Sodium Hypochlorite mixed with 16.5% NaCl, and 70% Ethanol and then flame sterilized.

Mycobiota enumeration

The International Standards Organization, ISO methods described by Odamtten *et al.* (2018) and Dugan (2007) were followed. Exactly 50g of either the fruiting body or growth substrate (compost) were weighed into 100 ml 0.1% peptone water in 250 ml Erlenmeyer flasks using electronic weighing balance OHAUS®. The samples were shaken in a Gallenkamp Orbital Shaker (140 rev/min) for up to 30 min. From the stock suspension, serial dilution techniques up to $1:10^3$ were prepared. Exactly 1ml aliquots of each dilution level were dispensed into 20 ml media, either Potato Dextrose Agar (PDA), Oxytetracycline Glucose Yeast Extract Agar (OGYE) or Dichloran Rose Bengal Chloramphenicol Agar (DRBC) as previously outlined by Odamtten *et al.* (2018). There were triplicate samples for each and dilution level. The Petri plates were inoculated upside down at 28 to 30°C for up to 7 days. Colonies which appeared were counted and data was converted to $\log_{10}$CFU/g sample.

Identification of mycobiota in compost and on mushroom fruiting body

This was done using morphological, colour, mycelial and spore characteristics as described by the standard reference identification manuals (Barnett and Barry 1972; Samson, Hoekstra, Frisvad and Filtenborg, 1995; Dugan, 2007).

Aeromycoflora of cropping/incubation rooms of the seven farms

Petri dishes containing 20 ml media (PDA, OGYE or DRBC) were exposed in triplicates (at standing height) for five (5) minutes. The lids were eccentrically placed on the bottom of the media plate on the platform. The exposed plates were incubated at 28 to 30°C for 7 to 14 days for colonies to appear. The colonies were counted, and species were identified using morphological, colour, mycelial and spore characteristics.

Isolation and identification of the pathogenic species (*Trichoderma harzianum*)

The most frequently encountered green mould, *Trichoderma harzianum* causing the green mould disease was isolated from all the seven farms and sub-cultured on PDA in McCartney tubes on 9 mm Petri dishes in a refrigerator at 8°C until needed. The potential pathogen, *T. harzianum* was identified by cultural and morphological characteristics as well as by the molecular sequencing method described by Kredics *et al*. (2009) and Hatvani *et al*. (2007).

RESULTS

Mycofloral diversity from the growth compost (*Triplochiton scleroxylon* 'wawa' sawdust) used in the cultivation of oyster mushrooms in the seven (7) mushroom farms

Across all seven mushroom farms, twenty-one (21) fungal species belonging to 11 genera were isolated from the compost. The genera encountered were *Aspergillus, Cladosporium, Didymella, Fusarium, Penicillium, Rhizopus, Rhodotorula, Trichoderma, Sacharomyces, Verticillium* and *Mycelia sterilia. Aspergillus species* (*A. candidus, A. flavus, A. fumigatus, A. niger, A. oryzae, A. terreus, A*. *penicilloides*) predominated over the other species encountered, followed by *Fusarium (F. oxysporum, F. poae, F. solani), Penicillium (P. brevicompactum, P. camamberti), Cladosporium (C. herbarum, C. macrocarpum)* and *Yeast (Rhodotorula mucilaginosa, Saccharomyces)* and single species of *Didymella, Rhizopus oryzae, Trichoderma harzianum* and *Lecanicillium fungicola* (Table 1).

Figure 1 shows some of the fungal species isolated from the sterilized compost. The compost used in the different farms showed some similarities. However, there was some uniqueness in the diversity of the species from some farms. For example, *A. candidus* was isolated from compost used in Kwesi Babs Mushroom Farm (Kasoa, Central Region); *A. oryzae* was isolated from Delabless Mushroom Farm (Adenta, Greater Accra Region) only; *A. penicilloides* was isolated from PCM Mushroom Farm only (Ashaley Botwe, Greater Accra Region); *A. terreus* was detected in sawdust compost used in the same PCM Mushroom Farm while *Didymella* sp. was unique in the substrate used in PCM Mushroom Farm. *Fusarium oxysporum* contaminated substrate from PCM Mushroom Farm but was also isolated from substrate compost used in Delabless Mushroom Farm (Adenta, Greater Accra). Another *Fusarium* (*F. poae*) contaminated compost was used in Immaculate Gold Enterprise (Nii Boi Town, Lapaz, Greater Accra Region) as well as PCM Mushroom Farm (Ashaley Botwe, Greater Accra). However, *Fusarium solani* was unique to PCM Mushroom Farm only at Ashaley Botwe, Greater Accra (Table 1). *P. camemberti* was isolated from only 4E Mushroom Farm at Ogbojo, Greater Accra Region. These farms were all widely separated by distance and not juxtaposition. *T. harzianum* was the most frequently encountered and isolated fungal

Table 1. Fungal species resident in the growth substrate (*Triplochiton scleroxylon)* sampled from the seven (7) mushroom farms in Accra and Kasoa.

Fungal species	Farms sampled from (by code)	Media isolated on		
Aspergillus candidus Link	2	DRBC		
Aspergillus flavus Link	1, 4, 5, 6	DRBC	OGYE	PDA
Aspergillus fumigatus Fresineus	4 ,5, 7	DRBC	OGYE	PDA
Aspergillus niger van Tieghem	1, 2, 3, 4, 6	DRBC	OGYE	PDA
Aspergillus oryzae (Ahlburg) E. Cohn	4		OGYE	-
Aspergillus penicillioides Speg	7	DRBC	-	
Aspergillus terreus Thom	7			PDA
Cladosporium herbarum (Pers.)	4, 5, 6, 7	DRBC	OGYE	PDA
Cladosporium macrocarpum Preuss	4, 5, 6	DRBC	OGYE	PDA
Didymella sp. (Fuckel) Rehm	7	DRBC	-	
Fusarium oxysporium Schlecht. Emend. Snyder & Hansen	4, 7	DRBC	-	PDA
Fusarium poae (Peck) Wr.	3, 7	-	-	PDA
Fusarium solani (Mart.) Sacc.	7	DRBC	OGYE	-
Mycelia sterilia	1, 2		OGYE	-
Penicillium brevicompactum Dierckx	4, 5, 6, 7	DRBC	OGYE	PDA
Penicillium camemberti Thom	5	DRBC	-	
Rhizopus oryzae Went and *Prinsen Geerling*	1, 2, 4	DRBC	-	PDA
Rhodotorula mucilaginosa H. C Harrison	2, 3, 4, 5	DRBC	-	
Trichoderma harzianum Rifai	1, 2, 3, 4, 5, 6, 7	DRBC	OGYE	PDA
Verticillium fungicola (Preuss) Zare & W. Gams	4, 6, 7	DRBC	OGYE	PDA
Saccharomyces sp. Meyen	1, 2, 3, 4, 5, 6, 7	DRBC	OGYE	PDA

Key
1 – E90 Mushroom Farm, Ogbojo
2 – Kwesi Babs Farm, Kasoa
3 – Immaculate Gold Enterprise, Nii-Boi Town, Lapaz
4 – Delabless Mushroom Farm, Adenta
5 – 4E Mushroom Farm, Ogbojo/Ashaley Botwe
6 – Edeyef Mushroom Farm, Anyaa-Awoshie
7 - PCM Mushroom Farm, Ashaley Botwe
DRBC – Dichloran Rose Bengal Chloramphenicol Agar
OGYE – Oxytetracyclin Glucose Yeast Extract Agar
PDA – Potato Dexrose Agar

species (Table 1).

Total fungal population isolated from sawdust compost used in the cultivation of oyster mushrooms in the seven (7) mushrooms farms

Figure 2 summarises the results obtained. There was diversity in the mycobiota populations depending on the isolating media (PDA, OGYE, DRBC) (Figure 2) and from one farm to another. The population was highest in samples from E90 mushroom Farm (Ogbojo, Accra) recording 4.0 to 5.24 $\log_{10}$CFU/g to the lowest obtained in the compost from 4E Farm also at Ogbojo (2.28 to 2.30 $\log_{10}$CFU/g). The varying contamination of the composts used in the different farms can be ranked as follows in decreasing order:

E90 Mushroom Farm (4.0 to 5.24 $\log_{10}$CFU/g) > Delabless Mushroom Farm (4.5 to 4.72 $\log_{10}$CFU/g) > Edeyef Mushroom Farm (3.8 to 4.3 $\log_{10}$CFU/g) > Kwesi Babs Mushroom Farm (2.8 to 3.6 $\log_{10}$CFU/g) > PCM Mushroom Farm (2.9 to 3.5 $\log_{10}$CFU/g) > Immaculate Gold Enterprise (2.9 to 3.2 $\log_{10}$CFU/g) > 4E Farm (2.28 to 2.93 $\log_{10}$CFU/g).

There were differences in the mycological quality of the substrates used for the cultivation of the oyster mushroom on each farm. Potential mycotoxigenic species isolated from the sawdust were *A. flavus, A. fumigatus*, *A. niger* and *A. terreus* (Table 1).

Mycofloral diversity of the fruiting body sampled from the seven mushroom farms

Table 2 summarises the results obtained on three isolating media (PDA, OGYE, DRBC). The purpose was to encourage a wider spectrum of fungal species. Generally, eighteen (18) fungal species belonging to ten (10) genera (*Aspergillus, Cladosporium, Fusarium, Gliocladium, Penicillium, Rhizopus, Rhodotorula, Talaromyces,*

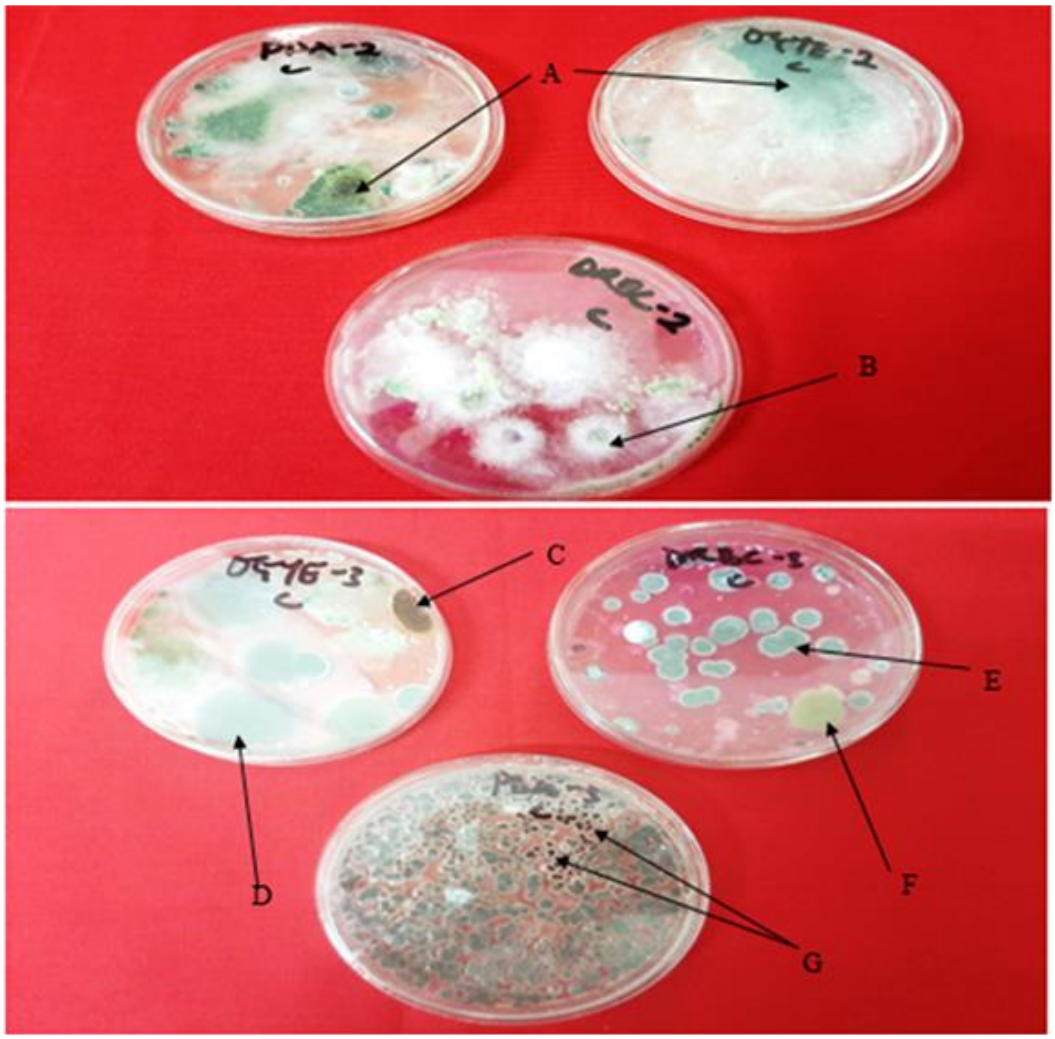

Figure 1. Representative mycoflora isolated from *Triplochiton scleroxylon* "wawa" growth substrate on PDA, OGYE and DRBC (Mag X 0.5). A - *Trichoderma harzianum*; B - *Fusarium poae*; C - *Aspergillus niger*; D - *Aspergillus fumigatus*; E - *Penicillium* sp.; F - *Aspergillus flavus*; G - *Cladosporium* sp.

Trichoderma and *Saccharomyces* were isolated. *Aspergillus* species (*A. ochraceus, A. candidus, A. flavus, A. fumigatus, A. niger* and *A. terreus*) predominated over the other species encountered. This was followed by *Cladosporium* isolates (*C. herbarum, C. macrocarpum*), *Fusarium* (*F. poae, F. oxysporum*), *Penicillium* (*P. brevicompactum, P. roqueforti*), *Rhizopus oryzae, Rhodotorula mucilaginosa, Trichoderma harzianum, Talaromyces flavus, Saccharomyces* spp. (Table 2). The mycobiota profile of the contaminants of the fruiting body from each of the seven mushroom farms was unique. For example, on the farm at Ogbojo, Greater Accra, *R. oryzae* was the least encountered whereas *A. niger* was the dominant species isolated from the fruiting body produced from Kwesi Babs at Kasoa, Central Region. On the other hand, at PCM Mushroom Farm (Ashaley Botwe, Greater Accra), *Aspergillus terreus* was the least predominant isolate whereas *T. harzianum* dominated. In general, *T. harzianum* was isolated from the fruiting body of *P. ostreatus* grown on all seven farms causing the green mould disease. Potential mycotoxigenic fungal species isolated were *A. flavus* (aflatoxin), *A. alutaceus* (ochratoxin A), *A. fumigatus* (fumigalin), *A. niger* (aflatoxin, nigerone), *A. terreus;* can produce tremorgenic mycotoxins, which are secondary fungal metabolites that elicit either intermittent or sustained tremors in vertebrate species (Noorabadi *et al*., 2020; Vassileva *et al*., 2020; Evans *et al*., 2018; Bartash et al., 2017; Burrows and Tyrl, 2001). (Figure 3).

Total fungal population isolated from the fruiting body grown in the storage rooms of the seven mushroom farms

Figure 4 shows the data obtained. The mycobiota populations varied depending on the isolating media (PDA, OGYE, DRBC) and from one farm to another. Microflora

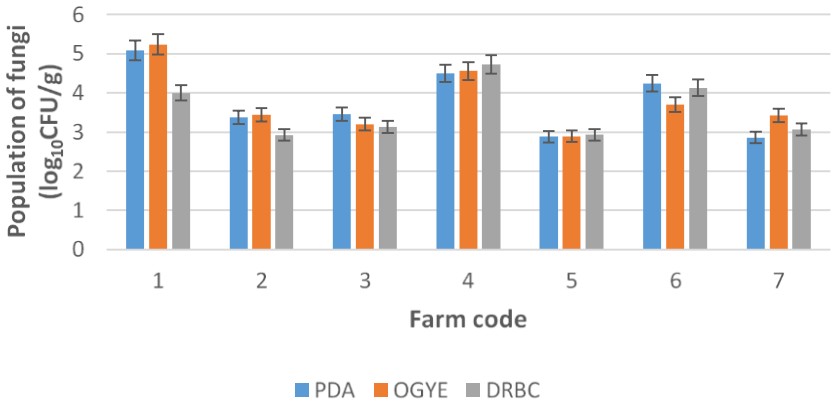

Figure 2. Comparison of the total fungal populations isolated from the growth substrate (*Triplochiton scleroxylon*) obtained from the seven mushroom farms. Key: Farm Code: 1 – E90 Mushroom Farm, Ogbojo; 2 – Kwesi Babs Farm, Kasoa; 3 – Immaculate Gold Enterprise, Nii-Boi Town, Lapaz; 4 – Delabless Mushroom Farm, Adenta; 5 – 4E Mushroom Farm, Ogbojo/Ashaley Botwe; 6 – Edeyef Mushroom Farm, Anyaa-Awoshie; 7 - PCM Mushroom Farm, Ashaley Botwe.

Table 2. Fungal species isolated from mushrooms sampled from seven (7) mushroom farms in Accra and Kasoa.

Fungal species	Code representing the name of mushroom farms sampled	Media for isolating fungal species		
Aspergillus alutaceus Wilhelm	7	-	-	PDA
Aspergillus candidus Link	3	DRBC	-	
Aspergillus flavus Link	4, 5, 6	DRBC	OGYE	PDA
Aspergillus fumigatus Fresineus	1, 2, 4	DRBC	OGYE	PDA
Aspergillus niger van Tieghem	1, 2, 6	DRBC	OGYE	PDA
Aspergillus terreus Thom	7	DRBC	-	PDA
Cladosporium herbarum (Pers.) Link	1, 3, 4, 5, 6, 7	DRBC	OGYE	PDA
Cladosporium macrocarpum Preuss	4, 6, 7	DRBC	OGYE	PDA
Fusarium oxysporium Schlecht. Emend. Snyder & Hansen	5, 7	DRBC	-	PDA
Fusarium poae (Peck) Wr.	7	DRBC	-	-
Gliocladium sp. (Link) Schroers	5	DRBC	-	-
Penicillium brevicompactum Dierckx	3, 5, 6		OGYE	PDA
Penicillium roqueforti Thom	6	DRBC	-	-
Rhizopus oryzae Went and Prinsen Geerling 1P, 2POD, 6D	1, 2, 6	DRBC	OGYE	PDA
Rhodotorula mucilaginosa H. C Harrison	4, 5, 6, 7	DRBC	-	-
Talaromyces flavus (P. A. Dang.) C. R. Benj.	1	DRBC	-	-
Trichoderma harzianum Rifai	1, 2, 3, 4, 5, 6, 7	DRBC	OGYE	PDA
Saccharomyces sp. Meyen	1, 2, 3, 4, 5, 6, 7	DRBC	OGYE	PDA

KEY
1 – E90 Mushroom Farm, Ogbojo
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6 – Edeyef Mushroom Farm, Anyaa-Awoshie
7 - PCM Mushroom Farm, Ashaley Botwe
DRBC – Dichloran Rose Bengal Chloramphenicol Agar
OGYE – Oxytetracycline Glucose Yeast Extract Agar
PDA – Potato Dextrose Agar

population on the media was highest in the fruiting bodies isolated from PCM Mushroom Farm (Ashaley Botwe), 4.71 to 5.34 $\log_{10}$CFU/g and the lowest recorded in 4E Farm (Ogbojo, Accra), 2.84 to 3.22 $\log_{10}$CFU/g. The varying

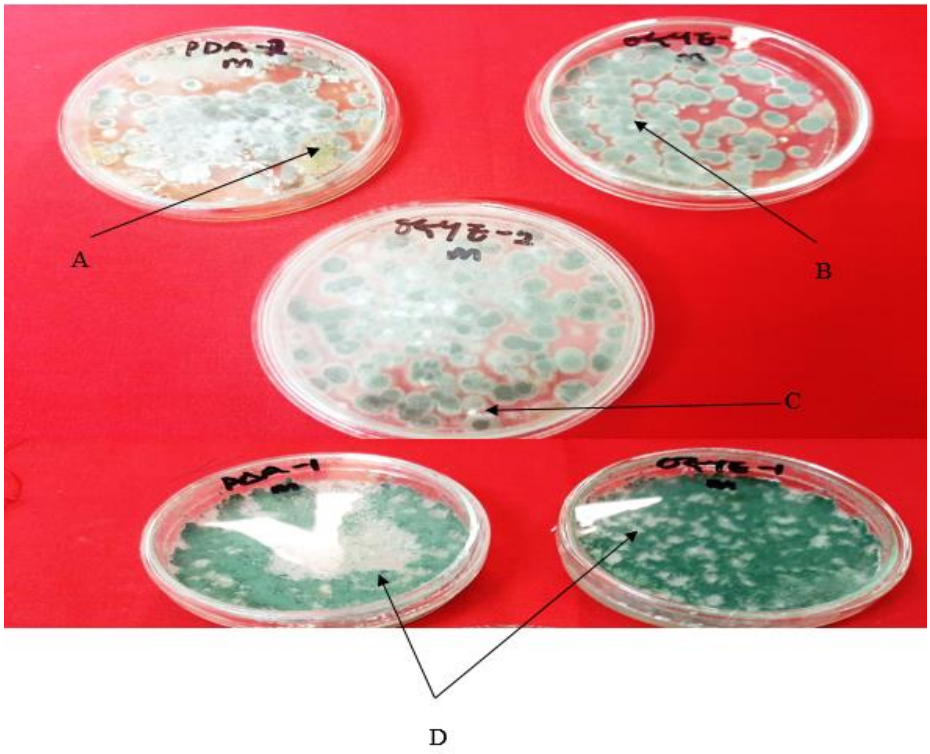

Figure 3. Representative mycoflora isolated from fruiting bodies of *Pleurotus ostreatus* sampled from the seven mushrooms on indicated media PDA and OGYE. **Key: A =** *Aspergillus flavus*; **B =** *Aspergillus fumigatus*; **C =** *Fusarium oxysporum*; **D =** *Trichoderma harzianum.*

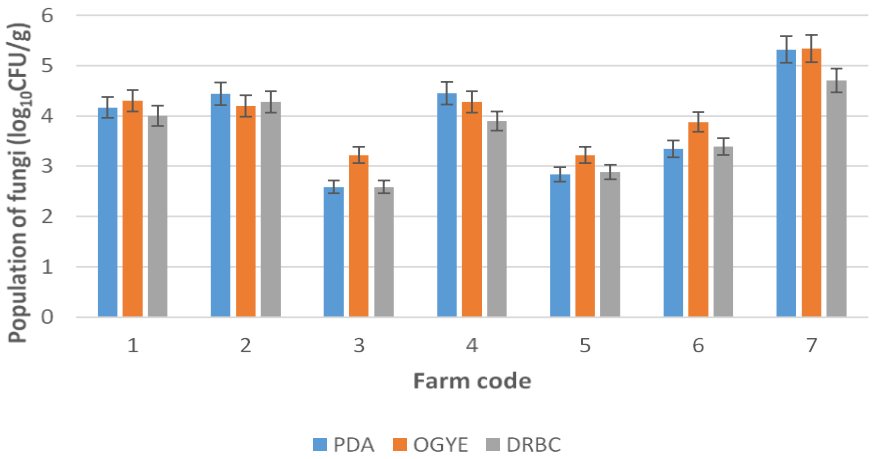

Figure 4. Comparison of the total fungal populations isolated from mushroom fruiting bodies obtained from the seven mushroom farms. **Key: Farm Code:** 1 – E90 Mushroom Farm, Ogbojo; 2 – Kwesi Babs Farm, Kasoa; 3 – Immaculate Gold Enterprise, Nii-Boi Town, Lapaz; 4 – Delabless Mushroom Farm, Adenta; 5 – 4E Mushroom Farm, Ogbojo/Ashaley Botwe; 6 – Edeyef Mushroom Farm, Anyaa-Awoshie; 7 - PCM Mushroom Farm, Ashaley Botwe.

quality of the fruiting bodies produced in the different farms can be ranked as follows (in decreasing order):
PCM Mushroom Farm (4.71 to 5.34 log_{10}CFU/g) > Kwesi Babs Mushroom Farm (4.20 to 4.44 log_{10}CFU/g) > E90 Mushroom Farm (4.0 to 4.30 log_{10}CFU/g) > Delables Mushroom Farm (3.90 to 4.45 log_{10}CFU/g) > Edeyef Mushroom Farm (3.35 to 3.88 log_{10}CFU/g) > Immaculate Gold Enterprise (2.59 to 3.22 log_{10}CFU/g) > 4E Farm (2.84

Table 3. Prevailing aeromicroflora in the cropping rooms of the seven mushroom farms isolated on three media (PDA, OGYE, DRBC) on Petri plates.

Fungal species	Code for mushroom farms sampled	Media isolated on		
Aspergillus candidus Link	4, 5	DRBC	-	PDA
Aspergillus flavus Link	3, 4, 5, 7	DRBC	OGYE	PDA
Aspergillus niger van Tieghem	1, 2, 4 , 6	DRBC	OGYE	PDA
Aspergillus oryzae (Ahlburg) E. Cohn	4	DRBC		
Aspergillus parasiticus Speare	2		OGYE	-
Cladosporium herbarum (Pers.) Link	3, 4, 5, 6, 7	DRBC	OGYE	PDA
Cladosporium macrocarpum Preuss	2, 3, 4, 5, 6	DRBC	OGYE	PDA
Epicoccum nigrum Link	3		-	PDA
Fusarium oxysporium Schecht Emend. Synder & Hansen	3, 5	DRBC	OGYE	-
Fusarium poae (Peck) Wr.	2, 4, 5	DRBC	OGYE	-
Gliocladium sp. (Link) Schroers	5	DRBC	-	-
Mycelia sterilia	5	DRBC	-	-
Penicillium brevicompactum Dierckx	3, 4, 5, 7	DRBC	OGYE	PDA
Penicillium citrinum Thom C.	2	DRBC		
Rhizopus oryzae Went and Prinsen Geerling	1, 2, 4	DRBC	OGYE	PDA
Rhodotorula mucilaginosa H. C Harrison	2, 3	DRBC		
Trichoderma harzianum Rafai	1, 2, 4, 5	DRBC	OGYE	PDA
Verticillium fungicola (Preuss) Zare & W. Gams	7	-	OGYE	-
Saccharomyces sp. Meyen	2, 3, 5, 7	DRBC	OGYE	PDA

Key
1 – E90 Mushroom Farm, Ogbojo
2 – Kwesi Babs Farm, Kasoa
3 – Immaculate Gold Enterprise, Nii-Boi Town, Lapaz
4 – Delabless Mushroom Farm, Adenta
5 – 4E Mushroom Farm, Ogbojo/Ashaley Botwe
6 – Edeyef Mushroom Farm, Anyaa-Awoshie
7 - PCM Mushroom Farm, Ashaley Botwe
DRBC – Dichloran Rose Bengal Chloramphenicol Agar
OGYE – Oxytetracycline Glucose Yeast Extract Agar
PDA – Potato Dextrose Agar

to 22 $\log_{10}$CFU/g). Furthermore, there were marked differences in the mycobiota population quality of the fruiting bodies of the oyster mushroom. However, 4E Farm had the lowest population followed by Immaculate Gold Enterprise. This is the same trend as obtained in the mycological quality of the compost.

Aeromycoflora of the cropping rooms used for the incubation of the mushroom during the growth and development of the fruiting bodies (basidiomata)

The results obtained are summarized in Table 3. Nineteen (19) different fungal isolates were sampled from the aeromycoflora. These nineteen (19) fungal species belong to twelve (12) genera (*Aspergillus, Cladosporium, Epicoccum, Fusarium, Gliocladium, Mycelia sterilia, Penicillium, Rhizopus, Rhodotorula, Trichoderma, Verticillium* and *Saccharomyces*). The aeromycoflora was predominated by *Aspergillus* species (*A. candidus, A. flavus, A. niger, A. oryzae, A. parasiticus*). This was followed by *Cladosporium* (*C. herbarum, C. macrocarpum*), *Fusarium* (*F. oxysporum, F. poae*), *Penicillium* (*P. brevicompactum, P. citrinum*). There were single species of *Trichoderma* (*T. harzianum*), *Epicoccum* (*E. nigrum*), *Gliocladium* sp., *Rhizopus* (*R. oryzae*), *Rhodotorula* (*R. mucilaginosa*), *Lecanicillium fungicola* (fr. *V. fungicola*), *Saccharomyces* sp. and *Mycelia sterilia* (Table 3).

The aeromycoflora profile in each Mushroom Farm was unique and the most predominant aeromycoflora was *Cladosporium herbarum* which was isolated from five (5) farms (Immaculate Gold Enterprise, Delabless Mushroom Farm, 4E Farm, Edeyef Mushroom Farm and PCM Mushroom Farm). *Cladosporium macrocarpum* was also isolated from the air in five (5) farms (Kwesi Babs Mushroom Farm, Immaculate Gold Enterprise, Delabless Mushroom Farm, 4E Farm, PCM Farm). The potential pathogenic *Trichoderma harzianum* was present in the aeromycoflora of four (4) farms (E90 Mushroom Farm, Kwesi Babs Mushroom Farm, Edeyef Mushroom Farm and 4E mushroom Farm). The potential toxin-producing *A. flavus* and *A. parasiticus* were among the aeromycoflora in Immaculate Gold Enterprise, Delabless Mushroom

Farm, 4E farm and PCM Mushroom Farm (Table 3). *Saccharomyce*s sp. was isolated from the air in four (4) farms (Kwesi Babs Mushroom Farm, Immaculate Gold Enterprise, 4E Farm and PCM Mushroom Farm).

On the contrary, *Aspergillus parasiticus* and *P. citrinum* were isolated only from Kwesi Babs Mushroom Farm; *Epicoccum nigrum* was in the air of Immaculate Gold Enterprise while *Gliocladium* sp. and *Mycelia sterilia* was only isolated from 4E Farm. *P. citrinum* was found in the aeromycoflora of the cropping house of Kwesi Babs Mushroom Farm only whereas *Lecanicillium fungicola* was encountered only from PCM Mushroom Farm. Presumably, the aeromycoflora in each farm cropping house may have contributed to the reduction in mycological quality of the product from that production facility.

The compost, fruiting body and air in the seven cropping rooms were laden with a miscellany of resident fungi predominated by *Aspergillus* species, *A. flavus*, *A. parasiticus, A. alutaceus* and *A. niger*, that has mycotoxigenic potential. However, *T. harzianum* was the pathogen proliferating in the compost, fruiting body, and air mycoflora causing the green mould disease recorded in this paper.

DISCUSSION

It is well-documented that mushrooms are an important food crop for many millions of people worldwide. Mushrooms are organic vegetables, and the production of mushrooms is an eco-friendly and profitable agribusiness but labour-intensive Banglapedia (as cited in Ferdousi *et al*., 2019). It does not require any cultivable land and can be grown in a room by racking vertically. Furthermore, mushroom cultivation can help reduce vulnerability to poverty and strengthens livelihoods through the generation of a fast-yielding and nutritious source of food and a reliable source of income; Marshall and Nair (as cited in Ferdousi *et al*., 2019). Mushroom cultivation is fast becoming one of the real solutions for utilizing the abundance of agrowaste specifically ligno-cellulolysic waste for a good harvest. There are approximately 14,000 described species of fungi that produce fruiting bodies and among them, 7000 are considered to possess a varying degree of edibility, and till now about 200 species have successfully been grown at a laboratory scale. Sixty species have the potential for commercial cultivation. Currently, only just about 10 species are farming at an industrial scale in many countries Chang; Chang and Miles; Kirk *et al.* (as cited in Raut, 2019); (Martins, 2017).

In Ghana, twenty-four (24) different species of mushrooms have been recorded; eighteen (18) are edible, and six or more are medicinal (Odamtten, 2018; Yafetto and Osei, 2017; Dhamodharan and Mirunalini, 2010; Obodai and Apetorgbor, 2001). The most common species of edible mushrooms cultivated in Ghana is the oyster mushroom, *Pleurotus ostreatus* (EM-1) imported into Ghana from Mauritius. It is also one of the most common edible mushrooms cultivated in the world (Hatvani, 2008). Ghana has over 12,000 mushroom farmers registered (https://mushroomsforwellbeing.org); however, the number of active mushroom farmers in Accra is 212. Although mushroom cultivation as an agribusiness for poverty alleviation has gained some modest success, there remains the problem of the inadequacy of sterilization of the spawn and substrate and the environmental quality of the cropping house (Kortei *et al*. 2018). The successful maintenance of the production of high-quality mushrooms is strict maintenance of substrate spawn microbiological quality and environmental cleanliness of the mushroom farm and their cropping houses. There are (20) mushroom and compost suppliers and exporters in Ghana (https://ghana.tradefood.com). However, there is one Centre officially accredited internationally, that is, the Food Research Institute (FRI) of the Council for Scientific and Industrial Research, CSIR-Ghana. One notable problem militating against the successful cultivation of *P. ostreatus* worldwide is the incidence of the green mould disease caused by the *Trichoderma* species (*T. harzianum, T. viride*); other species such as *Aspergillus* and *Penicillium* can also lead to decline in yield (Allaga *et al*. 2021; Wiafe-Kwagyan *et al.*, 2015; Chakraborty *et al*., 2013; Hatvani, 2008). Many mushroom farmers interviewed from several farms in Accra and the Central Region intimated their disappointment and heavy economic losses attributed to the prevailing incidence of green mould disease in their farms as well as a plethora of constraints. The present studies show that across all the seven mushroom farms selected for investigation, 20 fungal species belonging to 11 genera were isolated from the compost, *Triplochiton scleroxylon* ('wawa') sawdust (Table 1). *Aspergillus* species predominated the mycobiota followed by *Cladsosporium, Fusarium, Penicillium* and *Saccharomyces* among others. This implies that the composts used by the different farms shared some common contaminants. However, there was some uniqueness in the diversity of the species encountered from some farms (Table 1).

Commercial substrate preparation aims to produce a substrate that is optimal and selective for vegetative mycelial growth. Biologically, the substrate must have a population of suitable microorganisms and during the growth of these microorganisms on the compost, there is a production of secondary metabolites by these microorganisms. Imperfect fungi are among microorganisms which grow in the compost and may compete for nutrients and space and therefore there will be antagonism between fungal contaminants which may be undesired in mushroom cultivation since these species affect the growth and development as well as the quality of the mushroom crop. Many researchers (Chakraborty *et al.*, 2013; Singh *et al*., 2010; Reyes *et al.*, 2009; Sharma *et al.*, 2007) described these as competitor weeds. Contaminants are primarily moulds, bacteria, viruses, and insects and can be divided into two well-defined groups; those attacking the mushrooms are called pathogens while

those competing for the substrate are known as indicators and competitors (Rajarathnam *et al.*, 1997; Ragunathan *et al.*, 1996; Sandhu and Sidhu, 1980; Cailleux and Diop, 1978). Although, in this paper we concentrated on the fungi there is an excellent review on compost bacteria and fungi that influence the growth and deve lopment of commercial mushrooms (Kertesz and Thai, 2018). The seven farms were all widely separated and not juxtapositioned. It is instructive to note the variation in resident mycobiota in the bags after sterilization for mushroom production. Mycofloral population (log_{10}CFU/g) of the bags could be ranked as follows (in decreasing order) E90 Mushroom Farm > Delabless Mushroom Farm > Edeyef Mushroom Farm > Kwesi Babs Mushroom Farms > Immaculate Gold Enterprise > 4E Farm.

It was higher in samples from E90 Farm (4.0 to 5.24 log_{10}CFU/g) to the lowest in 4E Farm (2.29 to 2.93 log_{10}CFU/g). It was evident that the differences recorded could be attributed to the varying sources of the compost used accentuated by the insufficient heating time of compost during sterilization. However, in all instances, *T. harzianum* was the most frequently encountered and isolated fungal isolate in the compost, followed by *Saccharomyces* (Table 1).

The mycofloral diversity of the fruiting body of *P. ostreatus* harvested from the seven mushroom farms followed a similar trend as compost mycobiota. Generally, eighteen (18) fungal species belonging to ten (10) genera were isolated and were predominated by *Aspergillus* species followed by *Cladosporium, Fusarium* and *Penicillium* (Table 2). The mycological quality of the fruiting body produced in each farm differed considerably and could be ranked as follows (in decreasing order): PCM Mushroom Farm > Kwesi Babs Farm > E90 Mushroom Farm > Delabless Mushroom Farm > Edeyef Mushroom Farm > Immaculate Gold Enterprise > 4E Farm.

It was highest in fruiting bodies sampled from PCM Mushroom Farm (4.71 to 5.34 log_{10}CFU/g) whereas the less mycofloral population was isolated on fruiting bodies sampled from 4E Farm (2.84 to 3.22 log_{10}CFU/g). Interestingly, 4E Mushroom Farm had the highest contamination population of fungi in the compost followed by Immaculate Gold Enterprise. It has been reported by Woo *et al*. (2007) that *T. harzianum* as well as other contaminants can be transmitted to the fruiting bodies from the growth compost as was seen in these instances. During the composting, the substrate is continually changing both physically and chemically such that its suitability for colonization by different organisms also changes (Obodai and Odamtten, 2013; Obodai *et al.* 2010). Several workers (Obodai and Odamtten, 2013; Obodai *et al.*, 2010; Obodai, 1992; Sandhu and Sidhu, 1980) have reported that certain fungi are associated with the composting process such as *Trichoderma, Aspergillus fumigatus, A. flavus, A. terreus, Monila, Fusarium, Penicillium, Sclerotium rolfsii, Coprinus, Cinereus, Mucor pusillus, Rhizopus microsporous* and *Chaetomium thermophile*.

In general, mushroom pathogens are not as numerous as the competitor or contaminant fungi, though they can be much more devastating as well as fungi and bacteria are damaging to the mushroom crop. On the other hand, several contaminants are beneficial and can promote the growth of mushrooms. Examples of yield-enhancing microorganisms are several thermophilic fungi and bacteria such as *Humicola, Torula, Actinomyces, Streptomyces, Pseudomonas* and *Bacillus* species (Kertesz and Thai, 2018).

Trichoderma species are known to be pathogenic to *P. ostreatus* cultivation, for example, *T. viride* metabolites are antagonistic to *P. ostreatus* and *P. sajor-caju*. Some members of the genus *Trichoderma* (*T. kongii, T. hamatum, T. crissum, T. spirale* and *T. harzianum* have been isolated from mushroom compost (Ospina – Geraldo *et al.*, 1999; Jandaik and Guleria, 1999; Castle *et al.*, 1998). *Trichoderma aggressivum* caused extensive crop loses in both America and Europe in the 1990s and is still problematic worldwide caused of two slightly different strains in America and Europe (*T. aggressivium* and *T. aggressivum f. europaeum*) (Samuels *et al.*, 2002; Hatvani *et al*., 2017). *Pleurotus* is also affected by green mould disease, but the disease is caused by a related but phylogenetically different species called *T. harzianum* (Kredics *et al.*, 2009). Previous aggressive colonization of mushroom compost causing an epidemic outbreak of green mould in *P. ostreatus* was attributed to *T. harzianum* (Morris *et al.*, 1995; Doyle, 1991; Seaby, 1996, 1989, 1987). Jayal and Adikaram (2007), however, isolated*T. harzianum* from mushroom compost causing green mould in *P. ostreatus* resulted in considerable inhibition of the growth of mycelium and formation of fruiting bodies thus lowering substantially the yield. Wiafe-Kwagyan *et al.* (2015) showed that the culture metabolites of *Aspergillus flavus, Penicillium citrinum* and *Trichoderma harzianum* were antagonistic to dry matter accumulation by mycelium of *P. ostreatus* and *P. eous. T. harzianum* metabolite was the most potent in the antibiosis effect on the *Pleurotus* species.

Antibiosis is the inhibition of one microorganism by the metabolic product of another. Although it is usually an inhibition of growth and sporulation, it may be lethal. The metabolite penetrates the cells and inhibits chemical toxicity. Lysis is the final destruction and decomposition of biological materials by the enzymes of parasites (Obodai and Odamtten, 2013; Goltapeh *et al.*, 2000; Mumpuni *et al*., 1998). The sawdust compost for the cultivation of the mushroom contained contaminants or competitors as shown in this present paper.

Aeromycoflora refers to airborne fungal contributors to the environment. Fungal spores are ubiquitous and are found in every environment including cities (Ianovici and Tudorica, 2009), traffic and residential areas (Raj and Joshi, 2016) indoors (Ankush and Bhajbhuye, 2014), milk dairy (Barui and Chande, 2000) banana fields (Galenda *et al.*, 2020) to mention a few. These surveys were undertaken to understand the qualitative and quantitative

incidence of fungal spores and their contribution to plant aetiology, human diseases and crop production. Very little is reported on the impact of airborne fungi on mushroom cultivation. The number and type of fungi vary with the time of day, weather and seasonal fluctuation, conditions of the surrounding area, climatic conditions and local sources of spores (Pepelnjak and Kleric, 2003). Further, Gea *et al*. (2021) also reported some mycoparasites such as dry bubble (*Lecanicillium fungicola*), cobweb (*Cladobotryum* spp.), wet bubble (*Mycogone perniciosa*), and green mould (*Trichoderma* spp.) and their negative impact on mushroom yield and quality and at the same time reducing the cropping surface or damaging basidiomes. Fungal spores are normal and major component of indoor as well as outdoor air hence, all normal indoor conditions provide a conducive environment for the growth and proliferation of a wide range of fungal spores (Fashola *et al*., 2023; Odebode *et al.*, 2020; Shelton *et al.*, 2002).

This paper provides novel information on the diversity of fungi constituting the aeromycoflora of seven mushroom cultivation facilities in Ghana under tropical environmental conditions. Results show that nineteen (19) different fungi belonging to twelve (12) genera were encountered in the aeromycoflora (Table 3). The aeromycoflora was predominated by *Aspergillus* species followed by *Cladosporium. Cladosporium* and *Aspergillus* can grow both indoors and outdoors and can cause respiratory diseases and are the most abundant in tropical and subtropical regions (CABI, 2021; Mousavi *et al.*, 2016; Lacey and Venette, 1996; Ingold, 1971).

In this current study, *C. herbarum* and *C. macrocarpum* were isolated from the aeromycoflora of five (5) farms (Table 3), although *Aspergillus* predominated on the aeromycoflora of all seven farms. The presence of *Aspergillus* species could pose a danger of respiratory problems for the farm workers especially if any among them is immunocompromised. *A. flavus, A. parasiticus, A. alutaceus, A. niger* and *P. citrinum* were part of the aeromycoflora encountered with mycotoxigenic potential. The human health hazard posed by mycotoxin exposure at home, offices, schools, and hospitals depends on mycotoxin concentration, size, duration of exposure, immunogenicity, and individual sensitivities (Saad-Hussein and Ibrahim, 2021). Odamtten (2005; 2018) reiterated that the danger of mycotoxins, especially, aflatoxins still exists in Africa and cannot be discounted in our health delivery system. There was a preponderance of *T. harzianum* in the air of four (4) mushroom farms (E90, Kwesi Babs, Edeyef, 4E mushroom farms). *T. harzianum* was the same pathogen proliferating the compost, although the aeromycoflora profile in each mushroom farm was unique (Table 3). They shared the species found in the compost and fruiting body of the mushroom indicative of the possibility of being a source of contaminants especially the emerging fruiting bodies from the incubated compost bags.

Mushroom production in Ghana is faced with a plethora of constraints, some of which are mentioned in this paper. However, a more fundamental health problem posed by the infection of mushrooms by toxigenic species of fungi in the genera *Aspergillus, Fusarium* and *Penicillium* cannot be overlooked. In this present study, *Aspergillus flavus, A. parasiticus, A. niger* and *A. alutaceus* (*A. ochraceus*) not excepting *Fusarium oxysporum* were isolated from air and the fruiting bodies of *P. ostreatus*. There is a positive correlation between substrate moisture content and mycotoxin production in foods (Kanya *et al.*, 2005). Moisture content is a measure of the amount of free humidity (moisture) in a product and water activity (aw) is defined as the water vapour pressure of the substance divided by the vapour pressure of pure water at the same temperature. Water activity beyond 0.85_{aw} (85% ERH) at 25°C provides a conducive environment for fungal growth and spore germination (Hassane *et al.*, 2017; Lasram *et al*., 2010). The optimum water activity (aw) for mycotoxin production in fungi is 0.996_{aw} and the minimum is 0.80 to 0.82_{aw} (Grorni *et al.*, 2012; Northolt *et al.*, 1977). The moisture content or (aw) of *P. ostreatus* was high and falls within the stated range for growth and toxin formation. *Aspergillus* and *Penicillium* are linked to agricultural commodities during post-harvest storage (Agriopoulou *et al.,* 2020; Balandres *et al*., 2019). These mycotoxigenic species in the genera *Aspergillus, Fusarium* and *Penicillium* produce toxic compounds (Fashola *et al*., 2023; Mohammed *et al*., 2013).

A. niger is known to produce aflatoxins, but it possesses the ability to produce other toxins such as ochratoxin A, malformin and nigerone (Siddiquee, 2018; Wagacha *et al.*, 2013; Wang *et al.*, 2016). *A. alutaceus* which is known to produce Ochratoxin A was also isolated in this study. Ochratoxin A is lipid soluble and is not excreted efficiently and thus accumulates in meat and other products which exposes humans to health risks after consuming contaminated products (Denil and Perez, 2010). *A. flavus* and *A. parasiticus* infection and aflatoxins production in food have been extensively documented (Upadhyaya *et al.*, 2002; Wallyar *et al.*, 2016; Upadhyaya, 2005) and together with *A. parasiticus* produce aflatoxin (B1, B2, G1 and G2) in agricultural crops prior to harvest, or during storage (Yu *et al*., 2004). Prolonged consumption of aflatoxins have been associated with impaired immune function (immunosuppressive effect), malnutrition and stunted growth in children, disabilities, and death (Achagtinkame *et al.*, 2017; Bbosa *et al.*, 2013). Other adverse health effects of the intake of aflatoxins include liver cirrhosis, hepatitis B and C infection and liver cancer (Bbosa *et al.*, 2020; Raffe and Keller, 2019). The activity of fumagillin on its target, the methionine aminopeptidase type 2 (MetAP2) enzyme and the effects of blocking this enzyme in the host was also described by Gruceaga *et al*. (2020) and Fallon *et al*. (2010). The same fungus also produces several mycotoxins such as gliotoxin and pseurotin A. Fumagillin can inhibit the function of neutrophils in the blood inducing cell death in erythrocytes;

and play a role in the damage of epithelial cells which opens the way for fungal invasion (Gayathri *et al.*, 2020; Gruceaga *et al.,* 2020). Therefore, the presence of *A. fumigatus* in the fruiting body, spawn or compost cannot be discounted as it has serious health implications and toxic effects on human functions such as metabolism (Gruceaga *et al*., 2018, 2020; Raffa and Keller, 2019). These findings open a new direction of studies to look for the presence of fumagillin in samples of stored fresh and dry oyster mushrooms in Ghana because of the adverse effect on health. About 23 strains of *A. terreus* produced sterigmatocystin (STG) while 13 of them produced patulin (PAT) (Noorabadi *et al*., 2020). In this present study, *A. terreus* was isolated from the fruiting bodies of PCM Mushroom Farms.

There was another interesting observation of pathological importance i.e., the presence of *Fusarium oxysporum*, which is a well-known plant pathogen causing severe damage in many agricultural crops, both in the field and during post-harvest storage (Mondani *et al.*, 2021; de Lamo and Takka, 2020). Interaction between plant and root-colonizing *F. oxysporum* can be neutral, beneficial, or detrimental to the host. *F. oxysporum* is famous for its ability to cause wilt, root and fruit rot in many plant species including agriculturally important crops (Dean *et al.*, 2012). This fungus ranks among the ten (10) most devastating fungal plant pathogens worldwide (Dean *et al.*, 2012) and also causes wilts (pre- and post-emergence) which are a major threat to agricultural productivity (Fisher *et al.*, 2012). The association of *F. oxysporum* with the spent compost and the waste from the mushroom farms leaves much to be desired because of the indiscriminate way of disposal of the spent compost after harvesting the fruiting bodies into the ecosystem in and around farms.

CONCLUSION

Seven mushroom farms in Greater Accra (6) and Central Region (1) were sampled for resident mycobiota of the compost (*Triplochiton scleroxylon* 'wawa' sawdust), fruiting body and aeromycoflora of the cropping rooms to ascertain mycological quality of mushrooms, the compost bags, and the cropping rooms. Though mycoflora varied from one farm to another there were some common species found in all seven farms e.g., *Aspergillus*, *Cladosporium* and *Trichoderma* species. The presence of some species of *Aspergillus* and *Penicillium* possess health risks to humans upon consumption due to the potential mycotoxin production by these species.

RECOMMENDATIONS

To sustain profitable entrepreneurial production of oyster mushrooms producers should make a concerted effort to demonstrate control of food hazards through the application of Good Manufacturing Practices and Hazard Analysis of Critical Control Point, HACCP which is based on seven principles (Addo, 2008). Quality manuals for the production and processing of cocoa have been developed for cocoa (Amoa-Awua *et al*., 2007) and cassava (Dziedzoave *et al*., 2006). The present findings from this paper have demonstrated that the quality of mushrooms leaves much to be desired and GMP and HACCP should be formulated for the mushroom production industry in Ghana. Due to the predominance of fungi, especially *T, harzianum* causing green mould pathogen and potential mycotoxigenic species such as *A. niger, A. parasiticus*, *A. fumigatus* and *A. alutaceus* (fr. *A. ochraceus*). Poor environmental hygiene in farms that exacerbated the prevalence of mycobiota must be curtailed by regular fumigation and use of certified composts and spawns enhanced by proper sterilization protocols and timing sued in the microbiological laboratories. There should be more Certified Centres in each of the Regions where mushroom farming is practised which will produce quality and disease-free substrates and spawns for mushroom cultivation. There ought to be regular refresher courses for farmers on the best practices to sustain the economic production of mushrooms in the cottage industry as shown in this paper. If these recommendations are implemented, they will serve as a catalyst and springboard to increase the local supply and export of good quality oyster mushrooms from Ghana. This will also offer employment opportunities and will be a source of extra income in our effort to alleviate the poverty of the rural dwellers and also contribute to food security. Studies on the effect and authentication of the pathogenic *T. harzianum* on the yield and other growth parameters of *P. ostreatus* in the mushroom houses are being pursued and will be reported in a subsequent paper.

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