Studies on morphological and growth characters of new Pleurotus isolates

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Abstract
Oyster mushroom, Pleurotus sp. is suitable for commercial cultivation in subtropical regions and winter seasons in tropical regions of the world. The present study was carried out to isolate and characterize new Pleurotus isolates. Among a few collected isolates of Pleurotus sp., Pleurotus sp. isolate virutha1 produced well developed basidiocarp and it was compared with other cultivated Pleurotus spp. such as P. djamor isolate woody 1 and P. florida. These three Pleurotus spp. were analysed for different morphological traits viz., stipe length, diameter of the pileus, thickness of fruiting body, margin of fruiting body, colour of fruiting bodies and number of gills. Results indicated that all these three species of Pleurotus showed great diversity in their morphological characters. Typically, Pleurotus sp. isolate virutha and P. djamor isolate woody 1 appeared stipitate (no stipe). Whereas P. florida had well developed stipe. With regard to the colour, P. djamor isolate woody 1 appeared as white whereas Pleurotus sp. isolate virutha appeared light pink in colour at the initial growth phase and later growth phases, it appeared as white. P. florida appeared in creamly white in colour. All these Pleurotus spp. grown well in maize agar medium whereas potato dextrose agar medium supported slow growth.

Keywords: Oyster mushroom, phenotypic characters, basidiocarps

Introduction
The Indian subcontinent is blessed with numerous agroclimatic zones that harbour a wealth of fungal diversity. Though the occurrence of mushrooms is numerous in India, only some of them are cultivable and edible. Oyster mushroom (Pleurotus sp.) belongs to class Agaricomycetes and family Pleurotaceae. It grows naturally in the temperate and tropical region on dead and decaying wooden logs or generally on dying trunks of deciduous or coniferous woods. The oyster mushrooms have three distinct parts such as a fleshy shell or cap (pileus), a short or long lateral stalk known as stipe and long ridges and furrows beneath the pileus known as gills or lamellae. The gills stretch from the edge of the cap down to the stalk and bear the spores. The spores are smooth, cylindrical and germinate on any kind of mycological media at intervals of 48-96 hrs. The mycelium of Pleurotus is pure white in colour. The Pleurotus genus includes a various group of aromatic edible fungi that are praised for their culinary purpose and high price because they are wealthy in protein, fiber, vitamins and minerals [1, 2]. Additionally to their nutritional value, these fungi produce vital biomolecules [3]. Thus, in the present study, we focus on isolation and characterization of new Pleurotus isolates with good agronomic traits.

Materials and methods
Culture and media
Several fruiting bodies of Pleurotus spp. were collected from different places in southern districts of Tamil Nadu during 2017-2018 and preserved at the department of plant pathology, Agricultural college and research institute, Killikulam, Vandalanadu, Tamil Nadu. PDA medium was routinely used for culturing the mushrooms

Isolation and culturing of Pleurotus spp.
Well grown healthy sporocarps were collected from cultivated mushroom bed and from naturally grown on dead wood logs in different places and they were kept on a sterile tissue paper for 2-3 hours to evaporate the free moisture. The mushroom was surface sterilized with 70 % ethyl alcohol using absorbent cotton and it was split opened lengthwise into two halves employing a new sterilized blade. A little piece of plectenchymeous tissue was cut from the centre of the split mushroom at the junction point of the pileus and stipe and inoculated on the
PDA medium, amended with 100 ppm streptomycin sulphate, at equidistance in triangular position and incubated at 28°C. The plates were observed daily for the growth of mycelium. The pure culture of the Pleurotus sp. was maintained on PDA slants for further use during this study. Three Pleurotus spp. viz., P. djamor, isolate woody 1 and Pleurotus sp. isolate virutha and P. florida were used in this study.

**Morphological characterization of Pleurotus spp.**
Basidiocarp morphology for three Pleurotus spp. viz., P. djamor, isolate woody 1 and Pleurotus sp. isolate virutha and P. florida was assessed. Information on pileus phenotypic characters viz., diameter of pileus, its marginal shape, stip length, colour of the basidiocarp, thickness of the pileus, number of gills, and number of strip present per bunch was noted.

**Mycelial growth phenotype on different media**
To study the mycelial phenotypic characters of Pleurotus sp., five milli metre culture discs were cut with sterilized cork borer from advancing margins of the colonies and inoculated on the different media viz., PDA, oats agar medium, sorghum agar medium, maize agar medium, wheat agar medium supplemented with streptomycin sulphate (100 ppm). The plates were incubated at 28°C. Three replications were maintained for each medium. Radial growth of the mycelium was recorded when the mycelial growth covered in any one of the medium.

**Result and discussion**

**Isolation and culturing of the new Pleurotus sp.**
Isolation of mycelium from the new Pleurotus sp. isolates virutha, Pleurotus sp. isolate woody 1, and P. florida were taken from the healthy basidiocarp using PDA medium. The mycelial growth from the inoculated tissue pieces was attained on the second day of incubation. Pure culture was obtained from the corner of the colony. This pure culture was used for further studies.

**Phenotypic characterization of basidiocarp of Pleurotus spp.**
Morphological characters of fruiting body of three Pleurotus spp. were studied. Each Pleurotus sp. has typical differentiating phenotypic characters as described below (Table 1).

**Stipe length**
Among the three Pleurotus spp. commercially cultivated P. florida produced fruiting bodies with long stipe (stipe length was 52.93 mm). P. djamor isolate woody 1 produced basidiocarps with rudimentary stipe or no stipe at all. Pleurotus sp. isolate virutha is produced small sized stipe (stipe length was 26.11 mm). The result of the present study was similar to that reported by Mishra et al. (2015) [4].

**Diameter of the pileus**
Pileus diameter was maximum in P. djamor isolate woody1 (120.33mm) followed by Pleurotus isolate virutha (96.66mm) and P. florida (87.83mm). Similar results were found by Shubhra Shukla and Jaitly (2011) [5].

**Thickness of fruiting body**
Thickness of the pileus depends on the amount of plectenchymatous tissue present in the pileus. The thickness of the pileus was measured close to the intersection point of pileus and stipe. P. florida produced basidiocarp with maximum pileus thickness (13.43 mm diameter) followed by Pleurotus isolate virutha (7.76 mm diameter) and P. djamor isolate woody 1 had thickness of 5.33 mm diameter.

**Margin of fruiting body**
P. florida had pileus with smooth margin whereas P. djamor isolate woody 1 and Pleurotus isolate virutha had typical wavy margins.

**Colour of fruiting bodies**
P. florida produced creamy white colour basidiocarps whereas P. djamor isolate woody 1 produced white colour basidiocarps and Pleurotus sp. isolate virutha produced basidiocarps initially pink in colour later its turn white in colour.

**Number of gills**
P. djamor isolate woody 1 and Pleurotus isolate virutha had 23 and 21 gills/cm² respectively. Whereas, P. florida had less number of gills 19.56 gills/cm².

**Effect of the different media on the mycelial growth of Pleurotus spp.**
Effect of different media on the mycelial growth P. djamor isolate woody 1, Pleurotus sp. isolate virutha 1 and P. florida was studied on PDA, oats agar medium, wheat agar medium, sorghum agar medium, rice agar medium and maize agar medium. The maximum mycelial growth for all the three Pleurotus spp. was observed on maize and rice agar media. The maximum mycelial growth of P. florida was recorded on sorghum agar medium (90 cm), maize agar medium (90 cm), wheat agar medium (89.67 cm) followed by rice agar medium (87 cm) and minimal mycelial growth observed on PDA (57 mm). The maximum mycelial growth of P. djamor isolate woody was observed on maize agar medium (90.00) followed by rice agar medium (89.33), oats agar medium (87.33), sorghum agar medium (62.66), and PDA medium (57.00). The minimal mycelial growth observed on wheat agar medium (49.66). The maximum mycelial growth of Pleurotus sp. isolate virutha was observed on maize agar medium (69.00), rice agar medium (67.66), oats agar medium (47.66), PDA medium (42.00), sorghum agar medium (36.66) and minimal mycelial growth observed on wheat agar medium 21.00. Generally grain based media supports the mycelial growth of Pleurotus spp. (Figure 1, Table 2).
Fig 1: Effect of the different media on the mycelial growth of *Pleurotus* spp.

Table 1: Morphological characterization of basidiocarp of *Pleurotus* spp.

<table>
<thead>
<tr>
<th>Pleurotus spp.</th>
<th>Diameter of pileus (mm)</th>
<th>Length of stipe (mm)</th>
<th>Appearance of pileus margin</th>
<th>Thickness of pileus</th>
<th>Number of gills</th>
<th>Colour of fruiting bodies</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Pf</em></td>
<td>Primal Stage: 5.16&lt;sup&gt;a&lt;/sup&gt;</td>
<td>Mature Stage: 84.33&lt;sup&gt;c&lt;/sup&gt;</td>
<td>Harvesting Stage: 87.83&lt;sup&gt;c&lt;/sup&gt;</td>
<td>Primodial Stage: 11.00&lt;sup&gt;b&lt;/sup&gt;</td>
<td>Mature Stage: 49.83&lt;sup&gt;c&lt;/sup&gt;</td>
<td>Harvesting Stage: 52.93&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td><em>Woody</em></td>
<td>Primodial Stage: 15.33&lt;sup&gt;a&lt;/sup&gt;</td>
<td>Mature Stage: 80.30&lt;sup&gt;b&lt;/sup&gt;</td>
<td>Harvesting Stage: 120.33&lt;sup&gt;a&lt;/sup&gt;</td>
<td>Primodial Stage: 5.10&lt;sup&gt;b&lt;/sup&gt;</td>
<td>Mature Stage: 12.00&lt;sup&gt;b&lt;/sup&gt;</td>
<td>Harvesting Stage: 15.00&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td><em>Virutha</em></td>
<td>Primodial Stage: 11.33&lt;sup&gt;b&lt;/sup&gt;</td>
<td>Mature Stage: 54.00&lt;sup&gt;d&lt;/sup&gt;</td>
<td>Harvesting Stage: 96.66&lt;sup&gt;b&lt;/sup&gt;</td>
<td>Primodial Stage: 13.66&lt;sup&gt;a&lt;/sup&gt;</td>
<td>Mature Stage: 22.00&lt;sup&gt;b&lt;/sup&gt;</td>
<td>Harvesting Stage: 26.11&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
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Table 2: Effect of different media on the mycelial growth of *Pleurotus* spp.

<table>
<thead>
<tr>
<th>Pleurotus spp.</th>
<th>Mycelial growth</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Pf</em></td>
<td>PDA: 57.33&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td><em>Woody</em></td>
<td>Primodial Stage: 57.00&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td><em>Virutha</em></td>
<td>Primodial Stage: 42.00&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Reference
5. Shubhra Shukla, Jaitly AK. Microbiology Lab. Plant Science department M. J. P. Rohilkhand University, Bareilly, U.P., India.