

**COMPARATIVE EVALUATION OF DIFFERENT SPECIES OF OYSTER
MUSHROOM SUITABLE TO KERALA**

**By
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(2019-11-233)**

THESIS

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**DEPARTMENT OF PLANT PATHOLOGY
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2021**


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DECLARATION

I, hereby declare that this thesis entitled “**Comparative evaluation of different species of oyster mushroom suitable to Kerala**” is a bonafide record of research work done by me during the course of research and the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

Place: Padannakkad

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LIST OF ABBREVIATIONS AND SYMBOLS USED

%	Per cent
µg	Microgram
µL	Microlitre
µm	Micrometer
0C	Degree Celsius
ANOVA	Analysis of variance
BE	Biological efficiency
CD	Critical difference
cm	Centimetre
CRD	Completely Randomised Design
<i>et al.</i>	And other co-workers
Fig.	Figure
GAE	Gallic Acid Equivalent
g	Gram
h	Hour
KAU	Kerala Agricultural University
kg	Kilo gram
L	Litre
MEA	Malt Extract Agar
mg	Milligram
min.	Minute
mL	Millilitre
mm	Millimeter
mM	Millimolar
N	Normality
ng	Nanogram
OMA	Oat Meal extract Agar
OD	Optical density
pH	Negative logarithm of hydrogen ions
PDA	Potato Dextrose Agar
PDPA	Potato Dextrose Peptone Agar
ppm	Parts per million

PYDA	Potato Yeast Dextrose Agar
rpm	Rotations per minute
SE (m) \pm	Standard error of mean
SD	Standard deviation
Sl.	Serial
sp. or spp.	Species (Singular and plural)
temp.	Temperature
Wt.	Weight
<i>viz.</i>	Namely
YEA	Yeast Extract Agar

1. INTRODUCTION

Mushrooms have long been used for food and medicinal purposes. They are regarded as vegetable meat and has been consumed by humans for their taste and pleasing flavour since time immemorial (Das, 2010). They have achieved significant importance because of their nutritional and medicinal attributes and also as an income generating enterprise in most of the countries. It is a good source of protein where people rely heavily on cereal diet (Sohi, 1986). They have high content of proteins, vitamins, minerals, fibres, trace elements and are low in calories, fat and cholesterol (Wani and Patil 2010).

More than 2000 species of fungi are reported to be edible throughout the world. Out of that, mushrooms belonging to the genus *Pleurotus* is commonly referred to as “Oyster mushroom” worldwide and as “Dhingri” in India. It is characterised by tongue shaped pileus with an eccentric lateral stipe. Oyster mushroom is the second largest cultivated mushroom around the world recording 27 per cent of the world production (Royse, 2014). Commonly cultivated species throughout the world are *Pleurotus florida*, *P. djamor*, *P. sajor-caju*, *P. flabellatus*, *P. ostreatus*, *P. eryngii*, *P. eous*, *P. cornucopiae*, *P. sapidus*, *P. membranaceous*, *P. citrinopileatus*, *P. cystidiosus*, *P. fossulatus*, *P. australis*, *P. opuntiae*, *P. platypus*, *P. columbinus*, *P. populinus* and *P. levis*. Some of the *Pleurotus* species are being used in the medical research since certain components have been known to have antibacterial, antifungal, antiviral, anticoagulant, anti-diabetic and antitumor activity (Lindequist and De, 2005).

Oyster mushrooms grow on varying types of substrates and does not require that much of specific conditions. Generally, they prefer a temperature of 20 to 30° C and humidity of 80 to 90 per cent. The cultivation of various species of oyster mushrooms indicated that all of them have the potential of thriving under a wide range of temperature and relative humidity (Zadrazil, 1982).

Among all the cultivated mushrooms, oyster mushrooms have the maximum number of commercially cultivated species which can be cultivated throughout the year. Kerala, having a tropical climate is found to be suitable for the cultivation of oyster

mushrooms. Some of the oyster mushrooms suitable for cultivation in Kerala are *P. florida*, *P. djamor*, *P. sajor-caju*, *P. citrinopileatus* and *Hypsizyguis ulmarius*. *P. florida*, the white oyster mushroom is pure white in colour from pinhead formation to maturity. The mushroom looks pure white with delicate flesh which is turgid in texture. *P. djamor* is widely known as the pink oyster mushroom due to the production of pinkish red sporocarp. *H. ulmarius* is commonly known as ‘blue oyster’ or ‘elm oyster’ and the sporophores are medium to large, greyish blue during pinhead stage, which later turns creamy colour at maturity (Sumi, 2016). *P. sajor-caju* is known as the Grey oyster mushroom. During pinhead stage, the pileus colour is grey to dark grey, which changes to light grey on maturity. *P. citrinopileatus* is also widely known as the Golden oyster mushroom. The sporocarps are medium to large in size which is yellowish white during pinhead stage and turns golden yellow colour on maturity.

Kerala is divided into five agro-ecological zones from coastal plains to high hills, with each zone having wider climatic conditions (KAU, 2016). But till now, no research has been conducted to study the suitability of oyster mushroom for specific agro-ecological zones. conducted in Kerala based on the suitability of different oyster mushroom for the specific agro-ecological zone. Success of oyster mushroom cultivation depends on the substrates which must be cheap and easily available as well as viable technology suitable for a particular agro-climatic region. Various agricultural by-products are being used as substrates for the cultivation of mushroom. C/N ratio of substrate formulas has close correlation with total colonization period, mushroom weight, yield, biological efficiency (BE) and protein content of mushrooms (Hoa *et al.*, 2015). Hence, it has become necessary to find cheap and suitable substrates for the cultivation of different oyster mushrooms.

Edible mushrooms have the potential to contribute immensely to the food value of our habitual diet as they may contribute enormously to the supply of both macro and micro-nutrients. Oyster mushroom is gaining more importance as health promoter and environmental restorer as compared to other medicinal mushrooms. The chemical nature of the bioactive compounds present in this mushroom includes polysaccharides, lipopolysaccharides, proteins, peptides, glycoproteins, nucleosides, triterpenoids,

lectins, lipids and their derivatives (Tolera and Abera, 2017). Variation in nutritional content with respect to inter and intra genus variation Inter and intra genus variation in nutrient content of oyster mushrooms has been reported by Zahid *et al.* (2010).

Oyster mushrooms are not only nutritive but also highly valued for their excellent taste and flavour. Eventhough mushrooms are produced by small scale farmers, major issue facing in the local market is spoilage due to its high perishability. The onset of deterioration starts immediately after the harvest, and get deteriorated leading to browning, wilting, liquefaction, loss of texture, aroma, flavour etc., making it unacceptable. Proper packaging, maintenance of the optimum moisture content and temperature levels are keys to keeping mushrooms fresh.

Hence, this study was undertaken with the objectives of cultivation of five species of oyster mushroom for five agro-ecological zones of Kerala and to detect their biochemical and organoleptic properties along with storage studies.

2. REVIEW OF LITERATURE

Mushrooms of the genus *Pleurotus* are commonly known as oyster mushrooms, which are edible and among the most popular mushrooms worldwide. They are consumed all over the world due to their taste, flavour, high nutritional values, and some medicinal properties. They are rich in proteins with essential amino acids, physiologically important polysaccharides and essential fatty acids, dietary fibers, important minerals, and some vitamins. (Khan and Tania, 2012). They are also low in calories, sodium, fat and cholesterol. These nutritional properties make them a very good dietary food (Manzi *et al.*, 1999). Oyster mushroom ranks second position in world in terms of production (19 %), the first being Shiitake mushroom (22 %) (Sharma *et al.*, 2017).

Oyster mushrooms have oyster like shape. The genus consists of more than 50 species, of which 25 have been reported from India and 12 are cultivated in different parts of the country. They have been recognized as a highly potential converter of cheap celluloses into valuable protein. They are promising as a medicinal mushroom due to its antiviral, antibacterial, antibiotic and immunomodulation properties (Narayanasamy *et al.*, 2008)

They prefer a temperature of 10⁰C to 35⁰C and grow on various lignocellulosic materials (Zadrazil, 1982). Kerala having a tropical climate is found to be a suitable place for the cultivation of oyster mushroom. Krishnapriya (2018) has reported that there are around 15 to 20 *Pleurotus* spp. available for cultivation in Kerala.

2.1 ISOLATION AND PURE CULTURING

Dundar *et al.* (2008) isolated the cultures of *P. sajor-caju* and *P. ostreatus* from Microbiology laboratory of Science and Arts Faculty of Dicle University. 2.0 per cent Malt-Extract Agar (MEA) was used for culturing. MEA plates (90-mm diameter) were inoculated with the mycelium (6 mm diameter) of a young, actively growing margin of the colony at the center of the plate and incubated at 25°C in the dark for seven days.

Jegadeesh *et al.* (2018) collected fresh basidiomata of *P. djamor* var. *roseus* from decomposed wood materials in the forests of Indian Institute of Technology (IIT) campus, Chennai, India. A small tissue of the fresh pileus was aseptically removed using a sterile forceps. Then, it was immediately placed on the surface of potato dextrose agar (PDA) plates and the plates were incubated at 20-24°C for 5-6 days. The pure culture obtained was maintained on PDA slants at 4 ± 1°C and subcultured at regular intervals for further studies.

Sardar (2015) conducted a series of experiments to investigate about the effect of various culture media for the growth and development of *Pleurotus* species. He concluded that potato dextrose agar (PDA) was superior to the other media studied *viz.*, malt extract agar (MEA) and wheat extract agar (WEA).

Ahmad *et al.* (2015) pure cultured the mycelium of *P. djamor* on PDA, MEA, potato yeast dextrose agar (PYDA) and yeast extract agar (YEA) and found out that yeast extract agar was superior compared to other culture media. Thakur (2019) evaluated seven solid media to study the best medium for the growth of *P. citrinopileatus* and reported that PDA medium is the best for culturing this mushroom as it took minimum number of days for complete colonization of mycelia in Petri plate. Out of nine spawn substrates used, the maximum mycelial growth rate was observed on sorghum grains and minimum on pigeon pea. The minimum time (27.33 days) for spawn run was observed on sorghum grains spawn.

2.2 Macroscopic observations

P. florida, the white oyster mushroom was pure white in colour from pinhead formation to maturity. The mushroom looks pure white with delicate flesh which was turgid in texture. It grows in bunches. Pileus have thin margins which was smooth and it has less thickness compared to *P. ostreatus* and *P. sajor-caju*. The mushroom resembles a white disc growing on a thick stipe. Gills were decurrent unlike *P. ostreatus* and *P. sajor-caju* (Dhar, 2011). Prasad (2008) observed that *P. florida* had an average pileal length and width of 7.12 cm and 7.40 cm respectively. The stipe length was 4.62 cm and stipe width was 1.27 cm. It had dentate margins. Krishnapriya *et al.* (2017)

observed that *P. florida* produced white coloured sporocarps with entire and enrolled pileus margins. Pileus length was 6.71 cm and width were 7.90 cm. Stipe was significantly longer (4.80 cm) compared to *P. djamor* (0.91 cm). Jose (2018) reported that the cap of *P. florida* had spatulate shape and the size being 4.5-7.5 cm x 2.5-3 cm. The stipe was attached laterally and stipe length was between 3 cm to 4.5 cm.

P. djamor (pink oyster) produces pinkish red sporocarp and it is also known as salmon oyster mushroom, the flamingo mushroom and the strawberry oyster. The mushroom prefers to grow in tropical and subtropical regions and was known for its earliness in fruiting and high tolerance to temperature. It exhibits a wide range of colour and morphology for fruit bodies. Boulware *et al.* (2014) reported that *P. djamor* is an extremely fast-growing mushroom which can grow on a variety of substrates such as cereal straws, wood byproducts, sugarcane bagasse, corn cobs and coffee residues. Corner (1981) described *P. djamor* as having a very short stipe, spatulate to flabelliform pileus, leathery sporocarp and dimitic hyphal system. The mushroom was having a pileus dimension of 70-100 mm x 55-85 mm. The pileal surface was found to be smooth. The stipe was usually absent or reduced to 5-10 mm x 4-8 mm in dimension. Lamella is decurrent. Spore print was initially pink and later turns to white or light yellowish when dry. Shukla and Jaitley (2011) recorded that the pileus length and width of *P. djamor* was 4.72 cm and 7.40 cm respectively. Periasamy and Natarajan (2003) reported that the pileus length and width of hybrids of *P. djamor* ranges from 5.2-8.4 cm and 6.0-12.1 cm, respectively.

H. ulmarius is commonly known as ‘blue oyster’ or ‘elm oyster’ (Sumi, 2016). The species was earlier named as *Pleurotus ulmarius* (Bull. Ex Fr.) Kummer, later *Lyophyllum ulmarium* (Bull.: Fr.) Kuhner and recently as *H. ulmarius*. “Blue oyster mushroom” are called so because of the blue-colored primordia. It is one of the important edible mushrooms and is popularly cultivated in Japan, China, Asian countries and in North America. Meera *et al.* (2011) reported that the mushroom is known for its anti-diabetic, cardiovascular and anti-tumour properties. The mushroom is widely accepted for its unique flavour, nutritive and medicinal properties. The sporophores of *H. ulmarius* were medium to large, greyish blue during pinhead stage, which later turns creamy colour at maturity. The fruiting bodies were heavier, larger

and bluish white in colour. The cap was 6-15 cm in diameter, convex with slightly inrolled margins initially, becoming almost flat later on. The stipe is off-centre to nearly central and the gills were attached to stipe and non decurrent (not running down). The stipe was 2-6 cm long, thick, often eccentric, fleshy, solid, stout and hard. It was sometimes enlarged at the base, having smooth to hairy texture. The spore print was pure white when fresh and turns pale cream when dry (Kushwaha *et al.*, 2011).

P. sajor-caju (grey oyster mushroom) is characterised by the production of grey coloured sporocarps. During pinhead stage, the pileus colour was grey to dark grey, which changes to light grey on maturity. The mushroom have fan shaped fruiting body and thick texture. The fruiting bodies are comparatively heavy when fully grown, and the pileus diameter may extend up to 4 inches (Dhar, 2011). The pileus diameter and stipe length of *P. sajor-caju* was 5.95- 7.38 cm and 1.30- 3.95 cm respectively (Vooticumpee, 1996)

P. citrinopileatus was also known as the golden oyster mushroom. The sporocarps are medium to large in size which was yellowish white during pinhead stage and turns golden yellow colour on maturity. Pileus diameter ranges from 3-9 cm, funnel shaped, convex and depressed towards the base. Stipe length varies from 2-4 cm long, thick, cylindrical, smooth, eccentric or lateral, whitish yellow coloured. Lamellae attached to the stem, decurrent, creamy white in colour, 0.3 to 1 cm in width. Spore print was off white in colour (Thakur, 2019).

2.3 Microscopic observations

The basidia of *P. florida* were four spored with a size of 30-38 μm x 6-10 μm . Basidiospores were oblong with 7-10 μm long. (Biswas *et al.*, 2011). Das *et al.* (2015) reported that the dimension of basidium of *P. florida* was 29.20 μm x 4.10 μm . The basidiospores formed had an area of 15.68 sq. μm .

Lechner *et al.* (2004) observed that the basidiospores of *P. djamor* are cylindrical to oblong, hyaline, thin walled and not amyloid. The basidiospore dimensions were 6.0-7.80 μm \times 2.60-3.12 μm . Club shaped basidia was produced (26-28 μm \times 4-5.2 μm). Cheilocystidia was mucronate and hymenophoral trama dimitic

with sharp-pointed skeletal hyphae. Junior *et al.* (2010) studied about the microscopic characters of *P. djamor*. The basidiospores were cylindrical, thin-walled, hyaline, smooth, inamyloid. The spore dimensions were 8.7- 11.2 $\mu\text{m} \times$ 3.7- 5.0 μm . Basidia measured 25- 26 $\mu\text{m} \times$ 5.0- 7.5 μm , clavate, four-spored, occasionally two or three spored. Basidioles were numerous. Pleurocystidia were not observed. Cheilocystidia measured 18.7- 26 $\mu\text{m} \times$ 6.2- 11.2 μm , subventricose to clavate. Mycelium composed of thick-walled hyphae, septate, with clamps ranging from 3.7- 6.2 μm in diameter.

The microscopic and cultural characters of *H. ulmarius* was studied by Kushwaha *et al.* (2011). They observed that the colony produced white to creamy buff, fluffy mycelium in PDA medium. The hyphae were septate, branched, subhyaline to creamy buff in colour. Hyphal width was measured to be 1-4 μm . Basidiospores were small in size, measuring 2.5-6.5 μm , broad-ellipsoid to ovoid, hyaline and smooth.

10.2 EVALUATION OF PADDY STRAW/RUBBER SAWDUST FOR MUSHROOM PRODUCTION

In India, among different agricultural wastes available, wheat straw and paddy straw were found to be most suitable for oyster mushroom cultivation (Bano and Srivastava, 1962). Singh *et al.* (1995) reported that *P. florida* when grown on paddy straw substrate gave maximum biological efficiency (87.5 per cent) and yield (700 g/kg substrate) compared to wheat and sugarcane thrash.

Ragunathan *et al.* (1996) cultivated three species of *Pleurotus* namely *P. sajor-caju*, *P. platypus* and *P. citrinopileatus*, on various agro-residues such as paddy straw, maize stover, sugarcane bagasse, coir pith and a mixture of these wastes. Maximum yield was obtained in *P. sajor-caju*, cultivated on paddy straw. Ragunathan *et al.* (1996) obtained highest yield of *P. sajor-caju* in paddy straw. The biological efficiency, nutrient composition, energy value and energy recovery of fruiting body varied with substrates used for cultivation. Geetha and Sivaprakasam (1998) observed that oyster mushrooms preferred substrates rich in cellulose. The substrates rich in lignin supported minimum growth of oyster mushrooms. There was a positive correlation between the

yield of oyster mushrooms to the cellulose, lignin and fibre contents of substrates (Obodai *et al.*, 2003).

Das *et al.* (2000) observed significantly faster rate of spawn run, earliest pin head initiation and maximum yield of *P. sajor-caju* and *P. florida* in paddy straw than when wild grass was used as substrates. Owseph *et al.* (2001) reported paddy straw as the best substrate for cultivation of *Pleurotus* spp. Kolsulkar *et al.* (2001) concluded that paddy straw is the best substrate for cultivation of oyster mushroom species. Sharma (2003) reported highest yield of *P. djamor* when cultivated on paddy straw.

Periasamy and Natarajan (2004) reported that paddy straw was the best substrate for *P. djamor* var. *roseus*. Ponmurugan *et al.* (2007) studied the effect of different biowastes such as paddy straw, saw dust, sorghum straw, sugarcane molasses, and paper wastes on the growth and proximate constituents of *P. florida*. The results showed that, mushroom growth and yield were better in paddy straw.

Mondal *et al.* (2010) cultivated *P. florida* in six substrates including rice straw, banana leaves, sawdust and a combination of substrates to evaluate the better performance of the mushroom in different substrate compositions and also to find out the better substrate for cultivation. They reported that the highest biological yield and economic yield of *P. florida* (164.4 g and 151.1 g) was obtained from rice straw which was significantly higher and was followed by sawdust.

Srivastava *et al.* (2012) evaluated locally available substrates *viz.*, paddy straw, wheat straw, sorghum straw, maize straw, sugarcane dry leaves and sugarcane bagasse for the production of *P. citrinopileatus* and reported that maximum yield was obtained from paddy straw (408.5 g kg⁻¹) and minimum in case of sorghum straw (213.20 g kg⁻¹). A study was conducted by Musieba *et al.* (2013) on indigenous Kenyan isolate of *P. citrinopileatus* to identify the suitable substrate. They concluded that bean straw was the best substrate for cultivation of the mushroom as its BE was 148.78 per cent when compared with 98.88 per cent on rice straw and 76.66 per cent on sugarcane bagasse.

Mago *et al.* (2014) cultivated two species of oyster mushrooms *viz.* *P. sajor-caju* and *P. florida* on three different substrates namely paddy straw, combination of

paddy straw and leaf litter, combination of paddy straw and sawdust. Paddy straw was found to be the most suitable substrate for commercial production of oyster mushroom. substrates used in combination with paddy straw in the study had also shown encouraging results. The highest yield of *P. sajor-caju* and *P. florida* (4142 g and 4785 g) respectively was achieved in paddy straw which was followed by the combination of paddy straw and leaf litter and the combination of paddy straw and sawdust. Also, the number of sporophores of *P. sajor-caju* and *P. florida* (665 and 614) respectively was recorded from paddy straw which was followed by the combination of paddy straw and leaf litter and combination of paddy straw and sawdust.

Paddy straw and sea grass was used for the cultivation of *P. sajor-caju* by Manimuthu and Rajendran (2015). They observed maximum biological efficiency on paddy straw (84.39 per cent), followed by sea grass (70.65 per cent). Mishra *et al.* (2015) reported that *P. citrinopileatus* had the biological efficiency of 68.52 per cent on wheat straw substrate.

Atila (2017) evaluated the suitability of various lignocellulosic wastes for the cultivation of *P. djamor*, *P. citrinopileatus* and *P. eryngii*. He recorded that *P. citrinopileatus* showed significantly higher biological efficiency of 73.9 per cent on oak sawdust substrate. On other substrates such as bean straw, safflower hay and sunflower head residue, the mushroom had biological efficiencies of 43 per cent, 42.5 per cent and 54.1 per cent respectively.

2.3. CULTIVATION OF MUSHROOM

2.3.1 Time taken for complete spawn run

Zadrazil (1982) studied about the spawn running of six species of oyster mushrooms. They observed that *P. sajor-caju* and *P. flabellatus* completed colonization after 10 days. *P. florida* and *P. sapidus* completed spawn running within 18 days after bed preparation. *P. ostreatus* and *P. eryngii* took 20 and 30 days respectively for complete spawn run.

Singh *et al.* (1995) noticed that *P. florida* inoculated in paddy straw completed spawn running within 15 to 20 days. The spawn running period of *P. florida* was 10-12 days and 11 days in case of *P. djamor* (Ram and Thakur, 2005). Prasad (2008) reported that *P. florida* completed spawn running in paddy straw in 13 days while *P. djamor* took 13.5 days to complete spawn run. Gaitan-Hernandez and Salmones (2008) reported that the duration for complete mycelial colonization of *Pleurotus* species ranged from 20 to 30 days. Mondal *et al.* (2010) observed an average of 24 days for completion of mycelial running of *P. florida* in rice straw.

Shukla and Jaitley (2011) reported that the *P. sajor-caju* took significantly less period (10.5 days) to complete spawn run while *P. florida* and *P. djamor* completed spawn run in a period of 13 and 13.5 days respectively.

The spawn running period of *P. djamor* was reported to be 18-19 days by Pandey *et al.* (2016) whereas, Suresh *et al.* (2017); Satpal *et al.* (2017) observed very short spawn running period of 12.33 days.

2.3.1 Time taken for pinhead formation

P. sajor-caju grown on wheat straw was reported to produce pinhead after 25 to 27 days (Sivaprakasham and Ramraj, 1991). Gaikwad (2004) reported that *P. sajor-caju* formed pinhead between 17 to 23 days after spawning.

Asmamaw *et al.* (2005) observed that light and temperature had significant impact on pinhead formation of oyster mushrooms. On different substrates, *P. ostreatus* completed spawn running in 17 to 20 days and the time taken for pinhead formation was 23 to 27 days. It was observed that *P. ostreatus* took 46 days for pinhead formation when the temperature was 3 to 19°C but took only 28 days when the temperature was 25°C.

Ram and Thakur (2005) observed that primordia were initiated after 16 days of mycelial colonization in *P. florida*. The pinheads were ready to harvest within the next four days. Mondal *et al.* (2008) reported that the pinheads of *P. florida* emerged 8 days after complete spawn run. Manimuthu and Rajendran (2015) reported the primordial

initiation of *P. florida* in 19 to 23 days. Iqbal (2016) observed 37 days for pinhead formation and 39 days for maturation of the oyster mushroom cultivated on paddy straw. Sumi (2016) reported that *H. ulmarius* took 22.4 days for complete spawn run and 38.1 days for pinhead formation on paddy straw. After 5 days of emergence of pinhead, the mushroom was ready to harvest.

2.4 EVALUATION OF PERFORMANCE OF OYSTER MUSHROOMS

Sohi (1986) reported that oyster mushrooms are better in consumer aspects than the generally grown button mushroom. According to Teweri and Pandey (1991), most part of South India is ideal for cultivation of oyster mushroom.

Sumi (2016) reported a yield of 1.096 kg kg⁻¹ dry weight of paddy straw (109.60 % BE) for *H. ulmarius* and 0.976 g with 97.5 per cent biological efficiency for *P. florida*.

Pani and Das (1999) recorded yield of 914 g from *P. sajor-caju* cultivated on mixed substrate of arecanut leaf waste and paddy straw. Sangitrao *et al.* (2000) reported that *P. sajor-caju* yielded 1060.67 g/ kg in wheat straw with a biological efficiency of 100 per cent, due to modified assembly method of cultivation.

Shukla and Jaitley (2011) compared the yield of *H. ulmarius* and *P. sajor-caju*. He observed that the maximum average yield of 855.52g per kg dry substrate was recorded by *H. ulmaris* followed by *P. sajor-caju* 742.98 g per kg dry weight substrate. Different species of oyster mushrooms such as *P. sajor-caju*, *P. flabellatus*, *P. florida*, *P. eous*, *P. ostreatus* and *H. ulmarius* were evaluated for cultural characters and yield by Biswas *et al.*, (2013). *H. ulmarius* was found to be most appropriate species in terms of biological efficiency (156 per cent), spawn run period (15 days) and average weight of sporophore (7.98 g), followed by *P. florida* (121.5 per cent), *P. sajor-caju* (115.5 per cent) and *P. ostreatus* (103.25 per cent) biological efficiency.

Bilal *et al.* (2014) studied about the yield of *P. ostreatus* on different grains and substrates. He recorded an average yield between 630 to 780 g from three flushes. The average yield from wheat straw alone was 630 g, from waste papers alone was 530 g

and the combination of wood chips and wheat straw was recorded to be 780 g. Joshi *et al.* (2018) cultivated four species of oyster mushrooms namely *P. florida*, *P. sajor-caju*, *P. citrinopileatus* and *H. ulmarius* for the assessment of biological efficiency. They reported that, among the four species cultivated on paddy straw, *H. ulmarius* recorded the highest BE (90.10 per cent) and it was followed by *P. citrinopileatus* (85.66 per cent), *P. sajor-caju* (77.68 per cent) and *P. florida* (57.63 per cent).

Rawte *et al.* (2019) selected five *Pleurotus spp.* viz., *P. florida*, *P. sajor-caju*, *P. eous*, *P. flabellatus*, *P. sp.* to estimate their potential biological efficiency during summer season on paddy straw. *P. sajor-caju* emerged out as the most potential isolate and exhibited maximum biological efficiency.

Four species of oyster mushroom including *P. djamor*, *P. eous*, *P. florida* and *P. citrinopileatus* were tested for their yield performance by Kavipriya *et al.* (2020). Out of them, the most promising yield was obtained in *P. eous* and *P. djamor* with minimum spawn running period. In terms of earliness in growth of spawn and total yield, sorghum grain was superior over other spawn substrates for quality spawn production followed by wheat grain. Additionally, Paddy chaffy grain spawn supplemented with bengal gram at 60 g kg⁻¹ increased the yield of *P. djamor* and *P. eous* with minimum spawn running time. Shah *et al.* (2004) reported that the average yield in three harvests of *P. ostreatus* on wheat leaves was 447.20 g while that of sawdust and leaves was 646.90 g and 210.60 g respectively.

2.5. ANALYSIS OF PROXIMATE CONSTITUENTS

Sadler (2003) reported that mushrooms are rich in protein, minerals and vitamins and contain abundance of essential amino acids. Oyster mushrooms are healthy foods, low in calories and in fat, rich in protein, chitin, vitamins minerals and amino acids (Chirinang and Intarapichet, 2009). Chattopadhyay *et al.* (2014) claimed that *Pleurotus spp.* was rich in proteins, carbohydrates, vitamins, minerals and crude fibres.

2.5.1. Estimation of Protein

Dunkwal *et al.* (2007) were of the opinion that the proteins of mushroom are of high quality and rich in various essential amino acids. Jiskani (2001) reported that the protein value of mushroom is twice as that of asparagus and potatoes, four times as that of tomatoes and carrots and six times as that of oranges.

Ghosh *et al.* (1991) reported that the protein content of *P. citrinopileatus* on dry weight basis was 17.1 per cent. Turner (1993) evaluated biological efficiency of *Pleurotus species* and reported to range from 12.5- 72.4 per cent with high protein. In *P. sajor-caju* the crude protein content range from 18.46- 27.78 per cent. (Gupta *et al.*, 2004). Randhawa and Ranote (2004) reported that *P. florida* has 3.02 per cent protein content.

Caglarirmak (2007) reported the mean protein value of *P. sajor-caju* to be 0.92 per cent on wet basis while Ashraf *et al.* (2013) reported that the crude protein content in *P. sajor-caju* is 25.24 per cent and *P. djamor* 24.83 per cent respectively. Alam *et al.* (2008) conducted a nutritional analysis of *P. sajor-caju* and *P. florida*. Hundred grams of fresh *P. sajor-caju* contained 3-3.6 g of proteins. In case of fresh *P. florida* these were observed to be 2.5- 2.75 g. The protein content in 100 g of dried *P. sajor-caju* was found to be 23- 26 g. 100 g of dried *P. florida* contained 19- 22 g of proteins.

Menaga *et al.* (2012) reported that *P. florida* contained 50.70 per cent protein content in dry basis. Gogavekar *et al.* (2014) calculated the concentration of protein in 100 g dry matter of *P. sajor-caju* to be 29.3 g. Khatun *et al.* (2015) estimated the protein content of three species of *Pleurotus* namely *P. florida*, *P. citrinopileatus* and *P. pulmonarius*. They found that protein content was highest in *P. florida* (23.80 per cent) which was followed by *P. citrinopileatus* (20.8 %) and *P. pulmonarius* (16.8 %). Gupta *et al.* (2004) reported that the crude protein content in *P. sajor-caju* ranges from 18.46- 27.78 per cent.

Sumi (2016) reported the protein content of *P. florida* and *H. ulmarius* to be 20.05 per cent and 32.00 per cent respectively. Jose (2018) recorded the protein content of *P. florida* and *P. djamor* to be 22.16 per cent and 21.98 per cent respectively. *H.*

ulmarius recorded the least protein content (20.76 %). Jyothi (2019) reported that the protein content of *P. florida* and *P. djamor* to be 21.61 per cent and 23.33 per cent respectively.

2.5.2. Estimation of Fat

According to Starton (1990) mushrooms have low fat content. Mushroom is a low-fat food containing 1.1- 8.3 per cent fat on dry weight basis. Mushrooms contain all the classes of lipids including free fatty acids, glycerides, sterols and phospholipids (Kumary and Murthy, 2002).

Ghosh *et al.* (1991) reported that the fat content of *P. citrinopileatus* on dry weight basis was 5.8 per cent. According to the nutritional analysis conducted by Alam *et al.* (2008), the lipid content in fresh weight of 100 g of *P. sajor- caju* and *P. florida* were found to be 0.52 - 0.62 g and 0.5- 0.6 g respectively. The lipid content in dry weight of 100 g of *P. sajor- caju* and *P. florida* were observed to be 4.2- 4.6 g and 4- 4.6 g respectively.

Ashraf *et al.* (2013) reported that the total fat content in *P. djamor* is 3.07 per cent as compared to *P. sajor-caju* (2.47 per cent). The crude fat content in *P. sajor-caju* was 0.91 g in 100 grams of dry matter (Gogavekar *et al.*, 2014).

Sumi (2016) reported the fat content of *P. florida* and *H. ulmarius* to be 1.50 per cent and 2.96 per cent respectively. Jose (2018) recorded the fat content of *P. florida*, *P. djamor* and *H. ulmarius* to be 2.28 per cent, 2.10 per cent and 1.72 per cent respectively.

2.5.3. Estimation of fibre

Alam *et al.* (2008) reported that the fibre content in fresh weight of one hundred grams of *P. sajor-caju* and *P. florida* were 2.8- 3.1 g and 2.9- 3.1 g respectively. The fibre content in dry weight of one hundred grams of *P. sajor- caju* and *P. florida* were observed to be 22- 23.6 g and 22- 24.6 g respectively.

Chirinang *et al.* (2009) reported that the dietary fibre content of *P. sajor-caju* was about 42 per cent dry matter. Ashraf *et al.* (2013) reported that the fiber content of *P. sajor-caju* was 26.28 per cent and *P. djamor* (22.03 per cent). Crude fiber content of *P. sajor-caju* was found to be 12.30 g/100 g on dry weight basis (Gogavekar *et al.*, 2014). Sumi (2016) reported the fibre content of *P. florida* and *H. ulmarius* to be 10.49 per cent and 17.69 per cent respectively. Jose (2018) reported that the crude fiber content in *P. djamor*, *P. florida* and *H. ulmarius* were 30.59 per cent, 24.38 per cent and 17.06 per cent respectively. Jyothi (2019) reported that the fibre content in *P. djamor* and *P. florida* were 9.73 per cent and 8.38 per cent respectively.

2.5.4. Estimation of carbohydrate

Starton (1990) estimated that mushroom has 3.8 per cent complex carbohydrate while starch was found to be 0.02- 0.3 per cent in mushrooms (Shanmugham and Jayarajan, 1990). Bano and Rajarathnam (1982) reported that Carbohydrate constituted the major component (46.6- 81.81 per cent) in dry matter of *Pleurotus* spp.

Ghosh *et al.* (1991) reported that the carbohydrate content of *P. citrinopileatus* on dry weight basis was 50.55 per cent. According to the nutritional analysis conducted by Alam *et al.* (2008), the fibre content in fresh weight of one hundred grams of *P. sajor-caju* and *P. florida* were found to be 2.8- 3.1 g and 2.9-3.1 g respectively.

Ashraf *et al.* (2013) reported that the carbohydrate content in *P. djamor* is 37.69 per cent followed by *P. sajor-caju* (37.22 per cent). Total carbohydrate content of *P. sajor-caju* was found to be 62.97 g/100 g on dry basis. (Gogavekar *et al.*, 2014).

Jose (2018) recorded 30.87 per cent carbohydrate from *H. ulmarius* while the carbohydrate content in *P. florida* and *P. djamor* were 26.68 per cent and 26.59 per cent respectively. Jyothi (2019) reported that the carbohydrate content in *P. djamor* and *P. florida* were 45.79 per cent and 52.42 per cent respectively.

2.5.5. Estimation of moisture content

Mushrooms are composed of 90 per cent water and 10 per cent dry matter. The moisture percentage of *Pleurotus* spp. varied among different species and it depended

on factors such as stage of harvest, growing environment and storage conditions (Reis *et al.*, 2012). Agarwal *et al.* (2017) reported that the moisture content in oyster mushrooms ranged from 85 to 90 per cent which was responsible for the freshness in the mushroom.

Ghosh *et al.* (1991) reported that the moisture content of *P. citrinopileatus* was 90.22 per cent. The moisture content of *P. sajor-caju* was observed to be 94.04 per cent (Caglarimak, 2007). The moisture contents of *P. sajor-caju* and *P. florida* were found about 87 per cent and 87.5 per cent respectively according to Alam *et al.* (2008).

Gogavekar *et al.* (2014) calculated the moisture content of *P. sajor-caju* to be 90.02 ± 2.58 per cent. Ashraf *et al.* (2013) reported that the moisture content in *P. sajor-caju* was 87.37 per cent and *P. djamor* 82.77 per cent.

Sumi (2016) reported the moisture content of *P. florida* and *H. ulmarius* to be 93.95 per cent and 90.37 per cent respectively. Jose (2018) reported that the moisture content of *P. florida*, *P. djamor* and *H. ulmarius* were 84.92 per cent, 88.38 per cent and 89.28 per cent respectively. Jyothi (2019) reported that the moisture content of *P. florida* and *P. djamor* were 92.16 per cent and 85.97 per cent respectively.

2.5.6. Estimation of amino acid

The total amino acid content of *P. ostreatus* and *P. sajor-caju* was 21.11 mg/g and 20.12 mg/g respectively on fresh weight basis (Chirinang and Intarapichet, 2009). Sadler (2003) reported that mushrooms are rich in protein, minerals and vitamins and contain abundance of essential amino acids. Mushrooms form an excellent source of high-quality proteins comprising most of the essential amino acids in good proportion as well as vitamin and minerals (Richardson, 2010). Devi and Krishnakumari (2005) reported that the amino acid content in hot water extract of *P. sajor-caju* was 2.79 mg/g. Kim *et al.* (2009) reported that the average total free amino acid in edible and medicinal mushrooms are 120.79 and 61.47 mg per 100g respectively. Jose (2018) reported that the amino acid content in *P. djamor* and *P. florida* were 17.10 per cent and 14.75 per cent respectively. *H. ulmarius* recorded amino acid content of 10.62 per cent.

2.5.7. Estimation of mineral contents

Oyster mushrooms contain most of the mineral salts required by the human body (Stephanie, 2002). Marion (2006) reported that mushrooms contain several key minerals including copper, potassium, niacin and folate.

The nutritional analysis conducted by Alam *et al.* (2008) revealed that the total ash content in *P. sajor-caju* and *P. florida* were 1-1.2 g and 1.1-1.2 g respectively. In case of dry mushrooms these were 8- 8.6 g and 8.6- 9.5 g respectively. Ashraf *et al.* (2013) reported that the ash content in *P. sajor-caju* was 9.08 per cent while in *P. djamor* it was 8.35 per cent. Ash content of *P. sajor-caju* was 6.82 g per 100 g on a dry weight basis (Gogavekar *et al.*, 2014).

2.5.7.1 Sodium

Sodium is an important mineral which helps the body to keep fluids in a normal balance. Sodium plays a key role in normal nerve and muscle function. It retains the pH and water balance of body. Shelly *et al.* (2008) reported that the dried mushroom contains 3.90 per cent of sodium.

Vetter *et al.* (2005) reported that the sodium content of *P. pulmonarius* and *P. ostreatus* was 310 ppm and 191 ppm respectively. Caglarimak (2007) reported that the sodium content in *P. sajor-caju* was 750.77 mg kg⁻¹ on wet basis. The sodium content in *Hypsizygus sp.* was found to be 77.10 mg per 100g of oven dried sample (Chauhan *et al.* 2017). Salami *et al.* (2017) reported that the sodium content in *P. florida* was 277-359 mg per 100g. Jose (2018) reported that the sodium content of *P. florida*, *P. djamor* and *H. ulmarius* were 0.028 per cent, 0.096 per cent and 0.024 per cent respectively.

2.5.7.2 Phosphorous

Phosphorous is a mineral that the body uses to build bones and teeth and to make proteins that grow and repair cells and tissues.

Mallikarjuna *et al.* (2013) recorded the phosphorous content in *P. florida* and *P. djamor* as 640.2 mg per 100g and 743.2 mg per 100g respectively. Phosphorous

content of *P. ostreatus* cultivated on sawdust ranged from 0.77 to 0.91 per cent (Bhattacharjya *et al.* 2015).

Sumi (2016) reported that the phosphorous content in *H. ulmarius* and *P. florida* was 0.68 per cent and 0.67 per cent respectively. Jose (2018) reported that the phosphorous content of *P. florida*, *P. djamor* and *H. ulmarius* were 0.225 per cent, 0.217 per cent and 0.194 per cent respectively. Salami *et al.* (2017) reported that the phosphorous content in *P. florida* ranged between 1009-1133 mg per 100g.

Strmiskova (1992) observed that *P. ostreatus* contains 13387 mg kg⁻¹ of phosphorous. Caglarimak (2007) observed that the phosphorous content of *P. sajor-caju* was 716.31 mg kg⁻¹. Shelly *et al.* (2008) reported that the dried mushroom contains 1.94 per cent of phosphorous.

2.5.7.3 Potassium

Potassium helps our nerves to function and muscles to contract. It helps our heartbeat stay regular (Muthu and Shamnugasundaram, 2016). Bhattacharya *et al.* (2005) reported that the potassium content of *P. ostreatus* cultivated on different combination of substrates ranged from 1.16 to 1.28 per cent. Caglarimak (2007) reported that the potassium content of *P. sajor-caju* was 2687 mg kg⁻¹ on wet basis. Shelly *et al.* (2008) reported that the dried mushroom contains 3.70 mg per 100g potassium.

Sumi (2016) reported that the potassium content in *H. ulmarius* and *P. florida* was 1.98 per cent and 2.45 per cent respectively. Jose (2018) reported that the potassium content of *P. florida*, *P. djamor* and *H. ulmarius* were 0.39 per cent, 0.72 per cent and 0.70 per cent respectively.

2.5.7.4 Calcium

Calcium is vital for healthy teeth and bones. It is also important for the health and functioning of nerves and muscle tissue. Shelly *et al.* (2008) reported that dried mushroom contains 23.8 mg per 100g of calcium.

Caglarimak (2007) reported that the calcium content of *P. sajor-caju* was 23.66 mg kg⁻¹ on wet basis. The amount of calcium present in *P. florida* was estimated to be 2.7 mg per 100g (Masamba and Kazombo-Mwale, 2010). Bhattacharya (2015) reported that the calcium content in *P. ostreatus* was between 27.32 to 31.98 per cent. Jose (2018) reported that the calcium content of *P. florida*, *P. djamor* and *H. ulmarius* were 0.10 per cent, 0.08 per cent and 0.08 per cent respectively. Nagulwar *et al.* (2020) reported 24.27 mg per 100 g of calcium in oyster mushroom.

2.5.7.5 Magnesium

Magnesium is needed for more than 300 biochemical reactions in the body. It helps to maintain normal nerve and muscle function, supports a healthy immune system, keeps the heartbeat steady, and helps bones remain strong. Caglarimak (2007) reported that the magnesium content of *P. sajor-caju* was 157.67 mg per 100g on wet basis. Shelly *et al.* (2008) reported that dried mushroom contains 20.29 mg per 100g of magnesium. Masamba and Kazombo-Mwale (2010) reported that the magnesium content of *P. florida* was 9 mg per 100g. Bhattacharya (2015) reported that the magnesium content of *P. ostreatus* ranged from 13.3 to 19.85 mg per 100g. Jose (2018) reported that the calcium content of *P. florida*, *P. djamor* and *H. ulmarius* were 0.04 per cent, 0.05 per cent and 0.05 per cent respectively. Nagulwar *et al.* (2020) reported that magnesium content of oyster mushroom was 138.57 mg per 100g.

2.6 ANALYSIS OF MEDICINAL COMPONENTS

Medicinal value of mushrooms is known from time immemorial because of their ability to lower cholesterol and blood pressure, boost immune system and inhibit tumour growth (Ming *et al.*, 2010). Periyasamy and Natarajan (2002) had reported that *P. pulmonarius* act as an anti-tumour agent. Flat (2010) reported that due to their low sodium/ potassium ratio, fat, starch and calorific value, oyster mushrooms are suitable for persons with obesity, high blood pressure, and diabetes. He also reported that oyster mushrooms are natural sources of statin, which is a cholesterol lowering drug. Studies have shown that they typically contain 0.4-2.7 per cent statins. According to Dill (2007), oyster mushrooms help lowering cholesterol levels.

2.6.1 Beta-glucan

Sari *et al.* (2017) reported that *P. djamor* contains 20.70 per cent of β -Glucan followed by *P. pulmonarius* (17.46 per cent) and *P. citrinopileatus* (15.54 per cent). Toledo *et al.* (2013) analysed beta glucan content in edible mushroom samples by enzymatic method and HPLC. They reported that *P. ostreatus* and *P. sajor-caju* recorded 89.20 g/kg and 48.70 g per kg of beta-glucan respectively. Avni *et al.* (2017) reported that *P. eryngii* contains 43.47 per cent beta-glucan. Jose (2018) reported that the β -Glucan content of *P. florida*, *P. djamor* and *H. ulmarius* to be 20.80 per cent, 32.15 per cent and 26.62 per cent respectively.

2.6.2 Glycoprotein

Tanaka *et al.* (1989) confirmed the presence of glycoprotein in *G. luicudum*. Chen *et al.* (2009) isolated a functional glycoprotein (PCP- 3A) from the fruiting body of *P. citrinopileatus* and in vitro cell study showed that this glycoprotein at a concentration about 12.5 $\mu\text{g ml}^{-1}$ inhibits the proliferation of human tumor cell line in a time dependent manner.

2.6.3 Terpenoid

Menaga *et al.* (2012) reported that the aqueous extract of *P. florida* recorded high level of terpenoid on qualitative estimation. Sasidhara and Thirunalasundari (2014) studied on the antioxidant potentials of *P. djamor* and confirmed the presence of terpenoid in powdered sample. Hamzah *et al.* (2014) confirmed the presence of terpenoid in *P. pulmonarius*, *P. ostreatus* etc. The presence of terpenoid in *P. florida* and *P. ostreatus* was reported by Kinge *et al.* (2016). The terpenoid content in *P. florida*, *P. djamor* and *H. ulmarius* was reported to be 9.22 mg g^{-1} , 9.78 mg/g and 10.13 mg g^{-1} respectively (Jose, 2018).

2.6.4 Polyphenol

Chirinang and Intarapichet (2009) reported that the total phenolic content in water and ethanol extracts of *P. sajor-caju* was 37.98 and 29.30 gallic acid equivalent (GAE) respectively g^{-1} dry weight of sample. The total phenolic content of *P. florida*

was estimated as 62.72 mg catechol equivalent. Wandati *et al.* (2013) reported 675.56 mg GAE of polyphenol per 100 g of wild oyster mushroom sample. Hamzah *et al.* (2014) reported that the phenol content in *P. pulmonarius* and *P. ostreatus* were 223.11 mg g⁻¹ and 248.8 mg g⁻¹ respectively.

Sasidhara and Thirunalasundari (2014) estimated that the phenolic content of *P. djamor* was 32.55 mg/g of sample. Total phenolic content in *P. florida*, *P. citrinopileatus* and *P. pulmomrius* was estimated as 119 µg, 83 µg and 64 µg catechol equivalent per g of powdered sample (Khatun *et al.*, 2015). Methanolic extract of *P. ostreatus* contained maximum phenolic content of 24.01 mg GAE g⁻¹ of dry extract (Parihar *et al.*, 2015). Boonsong *et al.* (2016) observed 12.34 mg GAE g⁻¹ total phenolic content in the ethanolic extract of dried *P. sajor-caju* and 14.03 mg GAE in *P. eous*. The polyphenol content of *P. florida*, *P. djamor* and *H. ulmarius* to be 19.14 mg GAE g⁻¹, 14.56 mg GAE g⁻¹ and 20.14 mg GAE g⁻¹ respectively (Jose, 2018).

2.6.5 Beta-carotene

Beta-carotene content of *P. sajor-caju* (0.038 mg g⁻¹) was higher as compared to *P. florida* (0.018 mg g⁻¹). The β-carotene content of *P. florida*, *P. djamor* and *H. ulmarius* was reported to be 125.24 µg per 100g, 355.04 µg per 100g and 195.58 µg per 100g respectively (Jose, 2018).

2.7. SENSORY EVALUATION OF FIVE SPECIES OF OYSTER MUSHROOM

According to Adejumo and Awosanya (2005) mushrooms have been a food supplement in various cultures and they are cultivated and consumed for their edibility and delicacy. Mushrooms when used as ingredient in diet provide variety in taste, flavour, texture and nutrients (Kumar and Barmanray, 2007).

Mushrooms are low in calorific value but they rank very high for their vitamins, minerals and protein contents (Beetz and Greer, 1999). The consumption of oyster mushrooms is found to have an advantage of preventing as well as reducing diseases such as heart diseases, liver illness, kidney problems, diabetes, high blood cholesterol level, hypertension and microbial infection (Ooi, 2000). Apart from tasting great, they

are also a very nutritious addition to any cuisine (Mortimer *et al.*, 2012, Hilden *et al.*, 2013).

Sivaprakasham (1986) opined that the nutritive value of mushrooms as intermediate between vegetables, egg and meat protein. The nutritional profile of mushroom is far better than vegetarian foods. Mushrooms contain a high proportion of water (Wittingerova, 1991). Rai (2004) reported that *H. ulmarius* was a high yielding, highly palatable oyster mushroom with a good flavour.

Das (2010) opined that consumers preferred mushroom products based on different parameters such as appearance, colour, flavour, taste, texture and overall acceptability. Mehrotra *et al.* (2014) reported that sauteed mushroom is an ideal preparation to know the real taste of mushroom.

2.8 EVALUATION OF SHELF LIFE OF OYSTER MUSHROOM UNDER ROOM CONDITION AND REFRIGERATED CONDITION

Mushrooms have very short shelf-life and can remain as such only for a few hours under tropical conditions. In India, it is mostly sold as fresh and only negligible amount is used for processing. According to Sethi *et al.* (1991) mushrooms start deteriorating immediately after harvest due to enzymatic action resulting in the browning and softening of tissue, which is faster at higher temperatures. Bano *et al.* (1988) and Rai *et al.* (1988) also had similar opinion that mushrooms are predisposed to active desiccation due to high temperature which leads to reduction of moisture content of the fruiting bodies which in turn affect the texture, flavour and saleable weight of the produce. The storage temperature increased the rate of metabolic changes which resulted in quality deterioration (Rai and Saxena, 1989). (Ares *et al.*, 2007) opined that the short shelf life of mushrooms is mainly because of their increased respiration, loss of water and fast metabolic activity.

The shelf life of freshly harvested fruiting bodies of oyster mushrooms in non-perforated polythene bags was observed to be up to 72 h at room temperature and around 15 days at low temperature of 0-5⁰C (Mehta and Jandaik, 1989). Rai (2004) reported that *H. ulmarius* was an oyster mushroom with an attractive keeping quality.

According to Kim *et al.* (2006), one effective method to enhance shelf life of mushrooms during post harvest storage and commercialization is modified atmosphere packaging. Mota *et al.* (2006) packed mushrooms in plastic trays wrapped with perforated PVC films and stored under refrigerated condition.

3. MATERIALS AND METHODS

The experimental studies related to “Comparative evaluation of different species of Oyster mushroom suitable to Kerala” was carried out at Department of Plant Pathology, College of Agriculture, Padannakkad, during the period 2019-2021. Studies of five species of oyster mushrooms namely *P. florida*, *P. djamor*, *H. ulmarius*, *P. sajor-caju* and *P. citrinopileatus* have been undertaken with an objective to evaluate the performance of five species of oyster mushrooms in five agro-ecological zones of Kerala and to analyze the proximate and medicinal components present in five species. The methodologies used for the studies are described below.

3.1 ISOLATION AND PURE CULTURING

Five oyster mushrooms namely *Pleurotus florida* (Mont.), the white oyster mushroom, *Pleurotus djamor* (Fr.) Boedjn, the pink oyster mushroom, *Hypsizygos ulmarius* (Bull.: Fr.) Redhead, the blue oyster mushroom, *Pleurotus sajor-caju* (Fr.) Singer, the grey oyster mushroom and *Pleurotus citrinopileatus* Sing, the golden oyster mushroom were used for the study. Tissue culture method was used to isolate the mushrooms. Freshly harvested, medium aged, healthy mushrooms were collected and surface sterilized with ethyl alcohol. Using a sterile knife, the healthy stipe was split horizontally and a small portion of tissue from the junction of pileus and stipe was detached using a sterile inoculation needle. The detached tissue was placed in the Petri plates containing solidified Potato Dextrose Peptone Agar (PDPA) medium aseptically and incubated at room temperature. The mycelial growth observed after 72 hours was purified using hyphal tip method (Rangaswamy and Mahadevan, 2008).

The mushroom spawn of the five species were made as per the standard procedure given by Sinden, (1934). Clean and unbroken paddy grains were used as substrate for mushroom spawn production. The paddy grains were thoroughly washed in clean running water three to four times to remove soil debris, straw particles, chaff etc. Washed grains were cooked in boiling water until the seed coat just begun to split open. Full opening of the grain was avoided since it favoured the growth of microbial contaminants. Then the grains were spread evenly on a clean tarpaulin sheet for drying,

after draining the excess water. After sufficient drying, the boiled grains were mixed with calcium carbonate at the rate of 40 g kg⁻¹ of grains, to maintain the pH of the grains around 7 and to avoid sticking together. These were packed in polypropylene bags (12'' × 6'') at the rate of 300 g per bag and sterilized by autoclaving at a pressure of 1.02 kg cm⁻² at 121⁰C for 2 h. After cooling, the bags were inoculated aseptically with mycelial bits of size 1 cm x 1 cm from 10 days old cultures of species of oyster mushroom and incubated at room temperature (26±2⁰C) until the mycelium completely covered the grains.

Mushroom beds were prepared as per the procedure given by Bhaskaran *et al.* (1978). Paddy straw was sterilized chemically by soaking in 100 L of water containing 7.5 g carbendazim and 50 ml formalin for about 18 hours. Then excess water present in the straw was drained off and the straw were spread evenly over a clean tarpaulin sheet under sun to bring down the moisture content to 60 per cent. Polythene bags of 60 cm x 30 cm size were used for bed preparation. Paddy straw was twisted and placed in the polythene bag. Spawn was laid over the twisted straw towards the sides, over which paddy straw twists were again laid and spawning was done. Further, three to four layers were prepared and the top most layer was fully spread with spawn. Each bag was filled with 1 kg straw (dry weight) and 150 g of spawn. The bags were made compact, tied at the top and provided with around 15 pin holes for air circulation. The beds were then incubated in dark room with adequate aeration for the spawn run. After complete spawn run, 8-10 one-inch slits were put on the bed for the emergence of pinheads. The fruiting bodies produced were then used for taking the macroscopic and microscopic observations.

3.1.1 Macroscopic observations

Macroscopic characters of five species of oyster mushrooms *viz.*, sporocarp colour, texture, gill arrangement, stipe length, colour of pinhead was observed. The observed characters were recorded based on the data sheet provided by Nair (1990) as given in Appendix I.

Biometric observations such as sporocarp weight, number of sporocarp, pileus length, pileus breadth, stipe length and gills per cm were recorded.

3.1.2 Microscopic observations

Fresh, matured sporocarp of five species of oyster mushrooms were selected and as per the standard technique (Deepa, 2016), microscopic observations were taken. Thin sections of the gills were made using a new blade and stained with lactophenol cotton blue. The dimensions of basidia and cystidia were measured using compound microscope (Carl Zeiss Primo star, Germany) at 1000 X magnification. The microphotographs of basidia and cystidia were taken.

Microscopic studies of mycelium of five species of oyster mushrooms was conducted by following the standard slide culture technique (Riddell, 1950). Glass slides, coverslips, glass rods and blotter paper were placed inside petri plate and the whole unit was sterilized in autoclave at the temperature of 121⁰C and 1.05 kg cm⁻¹ pressure for 15 min. The unit was then taken inside laminar air flow chamber. Sterile water was added to the blotter paper and two glass rods were placed above the moist blotter paper. Glass slide was kept horizontally above the rods. About 10 ml. of 2 % plain agar medium was melted and poured into another sterile 9 cm petri dish. After the medium solidifies, a piece of 1 cm x 1 cm size plain agar was cut out using a flamed knife and placed at the centre of glass slide. Then the four sides were inoculated with the cultures of five species of oyster mushrooms. Then a flamed coverslip was placed over the agar piece and incubated at room temperature until sufficient growth has occurred. Then the cover slip was gently placed onto a small drop of lactophenol cotton blue (Appendix II) and observed under microscope.

Preparation of spore print was done using fresh sporocarps were harvested and selected for taking the spore print. The sporocarp were laid flat with the gill side facing the surface of a black chart paper kept overnight. A bell jar or Petri dish is placed over the sporocarp to avoid air current and also to provide a humid environment for the easy discharge of spores. The bell jar was removed after 24 hours and the pileus removed to obtain the spore print. The colour of spore dust was observed. To study the spore

characters, basidiospores obtained from the spore print were stained with lactophenol cotton blue and examined under 1000X magnification. Microphotographs of spores were also taken to examine the shape.

3.1.2. Cultural Characterization

The Pure cultures of five species of oyster mushroom were subcultured in 9 cm petri dishes containing solidified PDPA medium (20 mL). Mycelial discs were cut out from actively growing culture using a sterile cork borer and aseptically placed at the centre of petri plate containing the media. The cultures were then incubated under room temperature ($26\pm 2^{\circ}\text{C}$) and observed for colour of mycelia, growth pattern and density. To determine the rate and amount of mycelial growth of five species of oyster mushrooms, the diameter of the colony was measured every 24 h until the petri dish was completely colonized. The average mean growth was plotted against time (day) to obtain the growth rate in terms of mm day^{-1} (Guadarrama-Mendoza *et al.*, 2014). Four replications were maintained for each of the five species.

3.2 EVALUATION OF PRODUCTION OF MUSHROOMS ON SUITABLE SUBSTRATE

Two substrates namely paddy straw and rubber sawdust were selected for the cultivation of five species of oyster mushrooms namely *P. florida*, *P. djamor*, *H. ulmarius*, *P. sajor-caju* and *P. citrinopileatus*. The experiment was laid out in Factorial completely randomized design (FCRD) with five treatments and three replications were maintained for every treatment. Observations on the time taken for complete spawn run, pinhead formation, first harvest, total yield from three harvests and total crop period were recorded from both the substrates. The statistical analysis was carried out using GRAPES (KAU).

3.3 EVALUATION OF PERFORMANCE OF FIVE SPECIES OF OYSTER MUSHROOMS IN FIVE AGRO-ECOLOGICAL ZONES OF KERALA

Kerala is divided into five agro-ecological zones from coastal plains to high hills, with each zone having wider climatic conditions (KAU, 2016). To evaluate the

performance, five species were cultivated in farmer's field of five agro-ecological zones of Kerala. Five locations were selected with three replications for each of the five treatments. Pooled analysis was carried out using GRAPES (KAU).

The selected locations in five agro-ecological zones in Kerala were as following

1. Coastal plains - Mushroom production unit, Dept. of Plant Pathology, College of Agriculture, Padannakkad, Pin- 671328
2. Midland laterites - Suma Devi S, KRS bhavan, Moolayam, Aaliyad P.O, Venjaramoodu Thiruvananthapuram, Pin- 695607
3. Foothills - Lalu Thomas, Kalluvila Grace, Parankimammukal, Kunnicothu, Kollam, Pin- 691508
4. High hills - Bhagyaraj, Manjula bhavan, Punnamudi, Parakkottu, Vagamon, Pin- 685503
5. Palakkad plains - Abhilash R, House No 31, Green valley Colony, Chadayan kalai Kanjikode, Palakkad, Pin- 678623

3.4 ANALYSIS OF PROXIMATE CONSTITUENTS

Sample preparation

Healthy mature mushrooms were collected and cut into small pieces to facilitate drying. The pieces were kept inside a hot air oven at 55⁰C for 6 hours. After complete drying, the dried mushroom pieces were powdered with the help of a grinding machine.

3.4.1 Estimation of Protein

Estimation of protein content in the mushroom samples was done by Lowry's method (Lowry *et al.*,1951). One gram of powdered mushroom was mixed with 10 ml of 0.1 N NaOH and boiled for 30 minutes. Then the solution was cooled in room temperature and after cooling, the solution was centrifuged at 1000 rpm for 10 minutes to separate the supernatant.

Aliquot of 0.2, 0.4, 0.6, 0.8 and one millilitre of the working standard were taken into a series of test tubes. Then 0.1 and 0.2 ml of the sample extract were taken in two

test tubes and made upto one millilitre in all the test tubes. Five ml of alkaline copper solution (50 mL 2% Na₂CO₃ in 0.1 % NaOH) was added to each tube including the blank, mixed well and allowed to stand for 10 minutes. Then 0.5 ml of Folin-ciocalteau reagent was added and incubated at room temperature in dark for 25- 30 minutes. Absorbance of the blue colour developed was read at 660 nm against reagent blank. Standard graphs were prepared using bovine serum albumin. Using this graph, the concentration of protein content was estimated.

3.4.2 Estimation of Fat

Estimation of fat was carried out using Soxhlet extraction apparatus (Lees, 1975). Five grams of mushroom powder was taken in a thimble and placed inside the extractor. A piece of cotton wool was placed at the top of thimble for the proper distribution of solvent on the sample during extraction. Extraction of sample was then carried out with petroleum ether for 16 hours. The extract was transferred into a pre-weighed beaker (w₁), cooled in a desiccator and weighed (w₂). The percentage of fat was determined using the following equation.

$$\text{Per cent of fat content} = \frac{w_2 - w_1}{5} \times 100$$

3.4.3 Estimation of Fibre

Estimation of crude fibre content in mushroom was done by following the steps described by (De, 1965). Two grams of powdered mushroom sample was weighed out in a beaker. The sample was extracted using petroleum ether to remove the fat content. After adding 100 mL of 1.25 % sulphuric acid, it was boiled for 30 minutes with constant stirring. The digested sample was filtered through muslin cloth and washed with boiling water until washings are no longer acidic. The sample was again boiled with 200 ml of sodium hydroxide solution for 30 min. and filtered through muslin cloth and washed with 1.25% sulphuric acid, three 50 ml portions of water and 25 ml alcohol. The residue was removed and transferred to pre-weighed ash dish (w₁). Dried the residue for two hours at 130±2⁰C. It was then cooled in a desiccator and recorded the weight (w₂). The residue was further ignited for 30 min. at 600±15⁰C, cooled in a desiccator and reweighed.

$$\text{Per cent crude fibre in ground sample} = \frac{\text{loss in weight}}{\text{weight of the sample}} \times 100$$

3.4.4. Estimation of Carbohydrate

Estimation of total carbohydrate content of species of *Pleurotus* was done by following anthrone method (Aminoff *et al.*, 1970). 100 mg of mushroom powder was weighed into a boiling tube. It was hydrolysed by keeping the tube in a boiling water bath for three hours with 5 ml of 2.5 N hydrochloric acid. The hydrolysate was cooled to room temperature and neutralized with solid sodium carbonate until the effervescence ceased. The volume was made to 100 ml in a volumetric flask with the washings of the tube. It was thoroughly mixed and 10 ml of this solution was centrifuged at 5000 rpm for 10 min. The supernatant was collected and the aliquot was used for analysis. 0.5 ml aliquot was taken from the supernatant and made upto one ml by adding distilled water. Four ml anthrone reagent was added to the solution and heated for eight minutes in a boiling water bath. The solution was cooled rapidly and the absorbance was read at 630 nm in a spectrophotometer.

3.4.5. Estimation of Moisture content

Hundred grams (w1) of fresh mushroom samples were dried in an oven at 55°C until a constant weight was obtained (w2). The dry weight of the sample was noted and difference between fresh sample and dried sample weight gives the result which was converted into per cent (Geetha, 1993).

$$\text{Per cent of moisture content} = \frac{w1-w2}{w1} \times 100$$

3.4.6 Estimation of amino acid

Total amino acids were determined by following ninhydrin method which was given by Moore and Stein (1948). One ml of the sample was extracted using methanol and the extracted sample was mixed with 1 ml of ninhydrin in a test tube. The tubes were kept in a boiling water bath for 20 minutes and added 5 ml of diluent (equal

volume of water and n-propanol) incubate at room temperature for 15 minutes and the absorbance was read at 570 nm against a reagent blank. The results were expressed in mg/g sample.

3.4.7 Estimation of minerals

Digestion of sample

Digestion of the sample was carried out using kjeldhal's digestion assembly. 0.5 g of powdered sample was kept inside kjeldhal's distillation flask and 10 ml of concentrated sulphuric acid containing salicylic acid was added (1 g salicylic acid in 30 ml concentrated sulphuric acid). The mixture was allowed to stand for overnight. Initially the digestion was started on low flame for 10 to 15 minutes until the foaming stops. The digestion was continued at high flame for two to three hours until the liquid became clear which indicated the complete digestion. The contents were allowed to cool and transferred to a 50 ml volumetric flask through a Whatmann No.1 filter paper and made up the volume to 50 ml.

3.4.7.1 Estimation of Phosphorous

Estimation of phosphorous was carried out by the standard procedure given by Sadasivam and Manikam (1992). Working standards of 2, 4, 6, 8 and 10 ppm were prepared by pipetting out 2, 4, 6, 8 and 10 ml of 50 ppm stock solution into 50 ml volumetric flasks and the volume was made up. 10 ml of the digested sample was transferred to a 50 ml volumetric flask and 10 ml of Barton's reagent was added. Using distilled water, the volume was made up to 50 ml. The flask was allowed to stand for 30 minutes for the development of colour. The intensity of the yellow colour developed was read in a spectrophotometer at 470 nm. Concentration of phosphorous was found out with the help of standard curve. A blank was also prepared and read at 470 nm

Per cent of Phosphorous = $X \times 5010 \times 1000.5 \times 110,000$

X – Concentration of phosphorous from the graph

3.4.7.2 Estimation of sodium and potassium

From the stock solution of 100 ppm, working standards of 1, 2, 3, 4, 5, 6, 7, 8, 9 and 10 ppm potassium was prepared. From the digested sample, an aliquot of 5 ml was pipetted out and made up to the volume of 50 ml with distilled water. Prepared solution was fed into flame photometer and the readings were noted down. Standard graph was prepared as per the procedure given by Sadasivam and Manickam (1992).

$$\text{Per cent of potassium} = \frac{x * 50 * 50 * 100}{0.5 * 5}$$

X - Concentration of K from the graph

3.4.7.3 Estimation of calcium and magnesium

Wet ashing for determination of calcium and magnesium

2.5 g each of dried powdered samples of five species of oyster mushroom was weighed and collected in a conical flask. Few drops of distilled water were added to make the sample wet. 25 ml nitric acid was added and heated for 40 min. After cooling, 10 ml per chloric acid was added and heated again till the emission of white fumes appear. After cooling, 50 ml of distilled water was added to precipitate the remaining nitric acid. Then the contents were transferred to 250 ml volumetric flask and made up to the mark with distilled water.

Determination of Calcium and Magnesium

Five ml of the aliquot were pipetted in a titration flask and diluted to 100 ml with distilled water. Add 15 ml of buffer solution, ten drops of Eriochrome black T and two ml of triethanolamine. The mixture was titrated from red to blue. The minerals were calculated using the formula:

$$\text{Per cent of Ca} = \frac{a * N * 0.02 * 25 * 100}{V * W}$$

$$V * W$$

$$\text{Per cent of Mg} = \frac{a * N * 0.012 * 25 * 100}{V * W}$$

$$V * W$$

Weight of sample taken = W g

Volume of ammonium acetate made up = 25 ml

Volume taken for titration = V ml

Titrated volume of EDTA = a ml

Normality of EDTA =N

3.5 ANALYSIS OF MEDICINAL CONSTITUENTS

3.5.1 Estimation of β - Glucan

Beta-glucan in mushroom was estimated using acidic extraction method (Ibrahim *et al.*, 2017). To five grams of powdered mushroom sample, 80% of ethanol was mixed in 4:1 ratio using a magnetic stirrer for 2 hours at 600 rpm. One molar sodium hydroxide was added to in the ratio of 1:7 (Mushroom: NaOH) and allowed to mix using magnetic stirrer (45⁰C for 2 hours at 250 rpm). The solution was cooled until it became 20⁰C and centrifuged at 6000 rpm for 15 min. The supernatant was collected and centrifuged at 6000 rpm for 15 min. Then 15 per cent citric acid was added to the supernatant till the pH reaches 3.5 at 20⁰C. Then the supernatant was again centrifuged (6000 rpm for 15 min at 4⁰C). The supernatant separated and added 80% ethanol and left as such for 15 min at 4⁰C and again centrifuged to separate the sediment pellets. The pellets were transferred into a petri dish and weighed the extract. Then the pellets were dried in a hot air oven at 42⁰C for 16 hours until the drying was complete and the development of a dark colour and non-sticky nature was observed. It was weighed out and ground well to get a powdered form.

The powdered pellets were dissolved in distilled water using magnetic stirrer for two hours (at 25-30⁰C and 700 rpm). The reagents were prepared (cold 86 % sulphuric acid, every 1 ml contains 0.7 mg L-cysteine). Then two millilitres of reagent were added to each 400 μ l of dissolved extraction. Put them in boiling water bath directly for 3 min. Allowed to cool in room temperature and absorbance was read at 415 nm. The absorbance was compared with the standard curve of glucose to identify

the glucose concentration in the extract, and with the knowledge of molecular weight of beta- glucan and glucose, the percentage of beta- glucan in the extract was determined.

3.5.2 Estimation of Glycoprotein

Glycoprotein content in oyster mushroom was estimated qualitatively using orcinol (1,3- dihydroxy-5-methylbenzene) method (Koch *et al.*, 1991). Ready-made orcinol reagent was used. The purified mushroom extract was spotted on the thin layer chromatographic plate and air dried. Orcinol reagent was sprayed on the thin layer plate and placed it at 100⁰C for 20 minutes. Then it was allowed to cool and the presence of glycoprotein was confirmed qualitatively by the development of purple coloured spot.

Sample preparation

Mushroom powder was dissolved in methanol: water (1:1, v/v), and then vortexing and incubating at 70⁰C for 2 h. The mixture was then neutralized by adding 20 µl of 0.3 M hydrochloric acid solution. Phase separation was enhanced by brief centrifugation. For analysis of glycoprotein content, 100 mg mL⁻¹ of extracts were prepared by dissolving 1 g of mushroom powder in 10 mL of solvent. Mushroom extracts were centrifuged at 4000 rpm for 10 min. The supernatants were used as such.

Glycoprotein content was determined by using High-Performance Liquid Chromatography (HPLC) method. Supernatants of mushroom extracts were filtered through 0.45 µm PTFE membranes. Then, 20 µl was injected into HPLC system. The mobile phase was an isocratic mixture of ammonium acetate and 25 % acetonitrile (65:35), flow rate of 200 µL per min at room temperature. The detection wavelength (λ) was 245 nm using a UV-Vis detector. A gradient of 45 to 100 % acetonitrile was used for separation.

3.5.3 Estimation of Terpenoid

One gram of the powdered mushroom sample was taken in a test tube and 10 ml of methanol was added to it and shaken well and filtered to take five millilitre extract of the sample. The selected extracts were mixed with two millilitres of chloroform and

three millilitres of sulphuric acid. Then the prepared samples were transferred from the tube to colorimetric cuvette [95 % (v/v) Methanol will be used as blank] to read the absorbance at 538 nm. For the standard curve 200 µl of previously prepared linalool solution in methanol was added to 1.5 ml chloroform and serial dilution must be done. In case of serial dilution total volume of 200 µl will be made up by addition of 95 % (v/v) Methanol, the total terpenoid content was determined by using standard curve.

3.5.4 Estimation of Polyphenol

Estimation of polyphenol was done by following the Folin- Ciocalteu method as per the method given by Ondo *et al.* (2013). Aliquots of 0.25 ml of extracts (1mg ml⁻¹) were mixed with 1.25 ml of Folin- Ciocalteu reagent (0.2 N diluted in methanol). A reagent blank was prepared by using methanol instead of the sample. It is incubated at room temperature for five minutes. After that, one millilitres sodium carbonate solution (75g l⁻¹) was added. Samples were incubated at room temperature for two hours and the absorbance was measured at 765 nm.

3.5.5 Estimation of beta-carotene

Five grams of dried mushroom was grinded in 10-15 ml acetone and a few crystals of anhydrous sodium sulphate were added with the help of pestle and mortar. Supernatant was decanted into a beaker. The process was repeated twice and the combined supernatant was transferred to a separating funnel. 10-15 ml petroleum ether was added into it and mixed thoroughly. Two layers were separated out on standing. The lower layer was discarded and the upper layer was collected in a 100 ml volumetric flask, volume was made to 100 ml with petroleum ether and the optical density was recorded at 452 nm using petroleum ether as blank (Srivastava and Kumar, 2002).

$$\text{Beta-carotene } (\mu\text{g/ g}) = \frac{\text{O.D.} * 13.9 * 10^4}{\text{Wt. of sample} * 560 * 1000}$$

3.5.6 Estimation of Lovastatin

Sample preparation

Mushroom powder was dissolved in methanol: water (1:1, v/v), and then shaken at room temperature for 5 h. For analysis of lovastatin content, 100 mg mL⁻¹ of extracts were prepared by dissolving 1 g of mushroom powder in 10 mL of solvent. Mushroom extracts were centrifuged at 4000 rpm for 10 min. The supernatants were used as such.

Determination of Lovastatin content

Lovastatin was determined by using High-Performance Liquid Chromatography (HPLC) and following the method adapted from (Silva *et al.*, 2012). Supernatants of mushroom extracts were filtered through 0.45 µm PTFE membranes. Then, 20 µL was injected into HPLC system. The mobile phase was an isocratic mixture of acetonitrile and 0.1% phosphoric acid (65:35), flow rate of 1.5 ml per min at room temperature. The detection wavelength (λ) was 237 nm using a UV-Vis detector. Lovastatin stock solutions were prepared by dissolving lovastatin lactone in methanol: water (1:1, v/v) at a concentration of 100 µg/ ml.

3.6 ORGANOLEPTIC STUDIES

100g fresh oyster mushrooms was weighed and was cooked by sauting method by following the standard procedure as described in Appendix VI. Sensory evaluation was performed based on parameters such as appearance, colour, texture, flavour, taste, and overall acceptability. The overall acceptability was calculated by taking the average of all the five parameters. 12 were selected as the judging panel. The prepared sauted mushroom were analysed for organoleptic evaluation by 9-point hedonic rating scale for each of the quality aspects as per the reference given by Jellinick (1985). The scores were tabulated and statistically evaluated based on Kruskal Wallis test.

Score card is provided in Appendix IV. 9-point Hedonic rating scale is provided in Appendix V.

3.7 SHELF LIFE STUDIES

For the storage studies, half matured bunches of five species of oyster mushrooms were harvested, cleaned and used. They were packed in polypropylene covers with ten to fifteen holes of 5 mm diameter. One set was stored at room temperature ($26\pm 2^{\circ}\text{C}$) and another set was stored under refrigerated conditions (15°C). Visual observations in terms of days for change in colour, texture, smell and physical appearance was recorded at an interval of 24 hours.

3.8 STATISTICAL ANALYSIS

The data obtained from the experiments were subjected to analysis of variance (ANOVA). Critical difference (CD) was calculated at 5 per cent level of significance and used for comparison of difference between the treatment means. Standard error mean and standard deviation of observations were also determined. R based web application GRAPES was used for statistical data analysis (Gopinath *et al.*, 2020).

4. RESULTS

Oyster mushroom ranks second in cultivation among all the mushrooms both in the world as well as in India. They are rich source of proteins, vitamins, minerals and are also a reservoir of medicinal components such as β -Glucan, β -carotene and Lovastatin. *Pleurotus* has the maximum number of commercially cultivated species for round the year cultivation. Kerala having a tropical climate is found to be a suitable place for the cultivation of oyster mushroom. Kerala is divided into five agro-ecological zones from coastal plains to high hills, with each zone having wider climatic conditions (KAU, 2016). But till now research on suitability of different oyster mushroom for the specific agro-ecological zone of Kerala has not been conducted. Five species of oyster mushrooms viz., *P. florida*, *P. djamor*, *Hypsizyguis ulmarius*, *P. sajor-caju* and *P. citrinopileatus* were cultivated under five agro-ecological zones of Kerala viz., Coastal plains, Midland laterites, Foothills, High hills and Palakkad plains. Hence, this study was undertaken at Department of Plant Pathology, College of Agriculture Padannakkad to evaluate the suitability of oyster mushroom species for cultivation in different agro-ecological zones of Kerala and also to determine the biochemical, medicinal and storage properties of each species.

4.1 ISOLATION, PURE CULTURING AND MAINTENANCE OF NATIVE ISOLATES OF MUSHROOMS

Three days old healthy, pest and disease free sporocarps of the five species of oyster mushrooms were cultured on Potato Dextrose Peptone Agar (PDPA) medium by following tissue culture technique. Then actively growing mycelium of 5 mm diameter was cut using a sterile forceps and inoculated into the centre of 9 cm Petri plates. Observations on nature of mycelial growth, colour of mycelia, rate of mycelial growth (mm day^{-1}) and number of days to complete growth in 9 cm petri dish were recorded (Plate 1, plate 2).

P. florida and *H. ulmarius* produced white coloured thick, fluffy mycelium with even margins. *P. djamor* produced cream-coloured mycelia with concentric pattern and thick cottony growth. Off white coloured mycelia with cottony growth

were observed in *P. sajor-caju*. *P. citrinopileatus* produced off white coloured mycelia with thin cottony growth. The rate of mycelial growth was significantly higher in all the species and took 7-9 days to complete mycelial growth in 9 cm Petri dish except in *P. djamor* (7.50 mm day⁻¹) which took 12 days to complete mycelial growth. *H. ulmarius* and *P. sajor-caju* attained complete mycelial growth in minimum days (seven days). The cultural characters and mycelial growth of five species of oyster mushroom in PDPA medium is described in Table 1.

4.1.1 Macroscopic observations

The sporocarps from the five species of oyster mushrooms were studied based on the sporocarp colour, gill arrangement, stipe length, colour of pinhead and texture of sporocarp (Plate 3, plate 4). Macroscopic characteristics of five species of oyster mushrooms cultivated in paddy straw are described in Table 2.

P. florida produced white coloured and delicate fleshy sporocarp and pinhead and had leathery texture. Average weight of sporocarp was 11.71 g and comparatively large pileus with 6.62 cm x 7.52 cm were produced. Long and stout stipe (3.22 cm) was recorded from the sporocarp of *P. florida*. It had 12 gills per cm of the pileus and were attached decurrently to the stipe and the pinheads were white in colour. The sporocarp was produced in bunches and attained harvesting maturity in three days from pinhead emergence.

P. djamor produced pinkish white leathery sporocarps with smaller pileus. The pinheads were initially pink in colour; turned to pinkish white on maturity. The sporocarp was having a delicate texture with average pileus dimensions of 4.90 cm x 6.50 cm. The sporocarp was usually devoid of stipe and if present, was very short with an average length of 0.8 cm. The average sporocarp weight was 8.75 g and the number of gills from the margin was 17.50 gills/cm which were significantly higher than other species studied. The gills were attached decurrently to the stipe. The pinkish pinheads attained harvesting maturity within three days of primordial initiation.

H. ulmarius produced creamy white fleshy sporocarps with significantly large pileus. The pinheads were initially greyish blue, later turns to creamy white.

Table 1. Cultural characters and mycelial growth of five species of oyster mushroom in PDPA medium

Oyster mushroom	Mycelial growth pattern	Colour of mycelia	Rate of mycelial growth (mm day ⁻¹)	Days taken for complete growth in 9 cm Petridish
<i>P. florida</i>	Fluffy growth with even margins	White	11.20±1.55 ^a	8
<i>P. djamor</i>	Thick cottony growth with concentric pattern	Cream	7.50±0.14 ^b	12
<i>H. ulmarius</i>	Radial, thick and fluffy	White	12.75±1.70 ^a	7
<i>P. sajor-caju</i>	Cottony growth	Off white	12.50±1.91 ^a	7
<i>P. citrinopileatus</i>	Cottony growth	Off white	10.62±1.58 ^a	9
CD (0.05)			2.291	-
SE (m)			0.760	-

Values are mean ± SD of five replications

Values followed by similar superscripts are not significantly different at 5 % level

Table 2. Macroscopic observations of five species of oyster mushrooms cultivated in paddy straw substrate

Characters	Sporocarp colour	Gill arrangement	Stipe length	Colour of pinhead	Texture of sporocarp
<i>P. florida</i>	Pure white, fleshy	Decurrent	Comparatively longer	Pure white	Leathery
<i>P. djamor</i>	Pinkish white	Decurrent	Comparatively short stipe	Pinkish, faded on maturity	Delicate and watery
<i>H. ulmarius</i>	Creamy white	Non decurrent	Comparatively longer	Greyish blue, later turns to creamy white	Fleshy
<i>P. sajor-caju</i>	Greyish white	Decurrent	Longer	Grey colour later turns to greyish white	Leathery
<i>P. citrinopileatus</i>	Golden yellow	Decurrent	Comparatively short stipe	Bright yellow colour, later turns to golden yellow	Delicate and watery

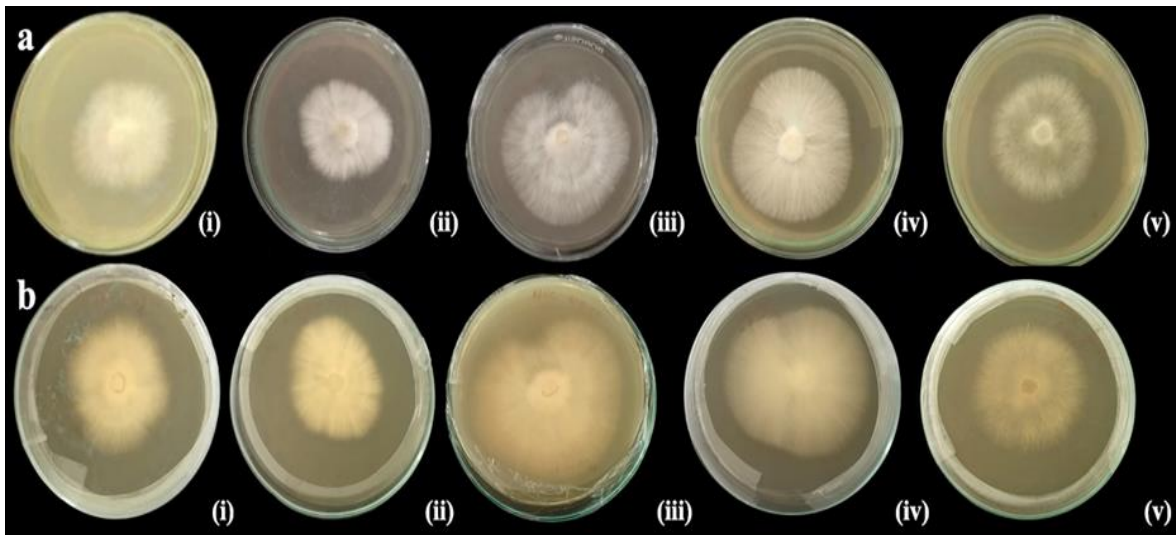


Plate 1. Radial growth of five species of Oyster mushroom in PDPA medium on 6th day of inoculation; a. Upper side; b. Rear side

i. *P. florida* ii. *P. djamor* iii. *H. ulmarius* iv. *P. sajor-caju* v. *P. citrinopileatus*

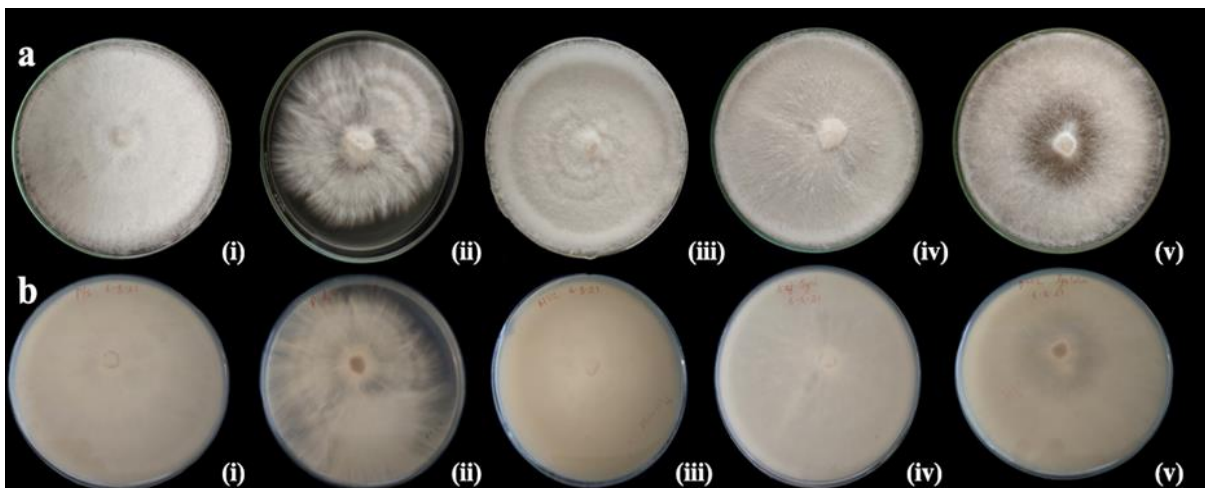
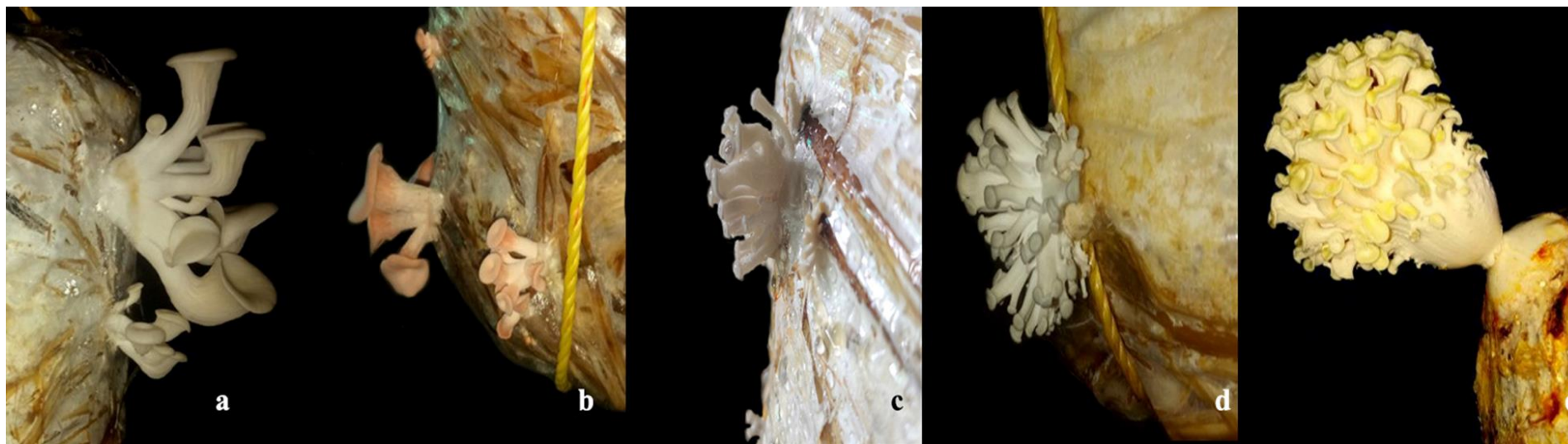
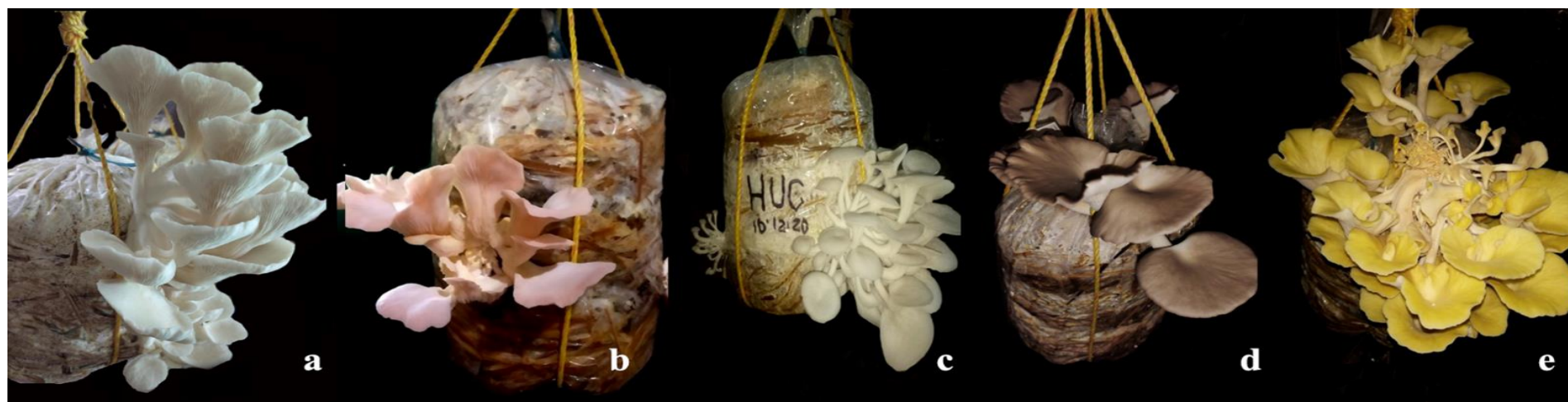


Plate 2. Radial growth of five species of Oyster mushroom in PDPA medium on 9th day of Inoculation; a. Upper side; b. Rear side

i. *P. florida* ii. *P. djamor* iii. *H. ulmarius* iv. *P. sajor-caju* v. *P. citrinopileatus*



**Plate 3. Pin-head formation of five species of oyster mushrooms in paddy straw;
a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus***



**Plate 4. Matured sporocarps of five species of oyster mushrooms
a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus***

The average pileus size was 7.0 cm x 7.52 cm. The average length of stipe was 3.35 cm which was comparatively higher. The average sporocarp weight was 9.33 g. Gills were attached to the stipe in a non-decurrent fashion and the average number of gills from the margin was 13.75 gills/cm. The creamy white pinheads attained harvesting maturity within three to four days from pinhead emergence.

P. sajor-caju produced greyish white leathery sporocarp. The pinheads were grey in colour later turned greyish white. The average length and breadth of pileus was 7.10 cm and 5.32 cm respectively. Significantly long and stout stipe with an average length of 5.32 cm was one of the characteristic features of *P. sajor-caju*. The sporocarps were produced singly and weighed 11.07 g. Gills were attached to the stipe in a non-decurrent fashion and the average number of gills was 17.50 gills per cm. The pinheads attained harvesting maturity within four days.

P. citrinopileatus produced golden yellow delicate sporocarp with comparatively short stipe. The pinheads were bright yellow in colour initially and turned to light yellow. The average size of pileus was 5.45 cm x 6.17 cm and gills were attached decurrently to the stipe. The average stipe length recorded was 3.30 cm and the average sporocarp weight was 5.05 g. The average number of gills from the margin was 11.75 gills per cm. The bright yellow pinheads attained harvesting maturity within three to four days from pinhead emergence (Plate 5, plate 6). The biometric observations of five species of oyster mushrooms are given in Table 3.

Spore print of five species of oyster mushroom were taken. Spore print of *P. florida* and *H. ulmarius* was pure white. *P. djamor* produced light pink coloured spore print which changed to creamish white. *P. sajor-caju* and *P. citrinopileatus* produced pale white spore print (Plate 7).

Table 3. Biometric characters of sporocarp of five species of oyster mushroom cultivated in paddy straw substrate

Species of oyster mushroom	Sporocarp weight (g)	No. of sporocarp	Pileus length (cm)	Pileus breadth (cm)	Stipe length (cm)	Gills (No. cm ⁻¹)
<i>P. florida</i>	11.71±3.32 ^a	106.50±6.55 ^{cd}	6.62±0.35 ^a	7.52±0.59 ^a	3.22±0.22 ^b	12.00±0.81 ^c
<i>P. djamor</i>	8.75±4.00 ^{ab}	98.25±5.25 ^d	4.90±0.18 ^b	6.50±0.14 ^{bc}	0.80±0.07 ^c	17.50±0.57 ^a
<i>H. ulmarius</i>	9.33±1.90 ^a	115.50±5.50 ^{bc}	7.0±0.54 ^a	7.52±0.37 ^a	3.35±0.25 ^b	13.75±0.95 ^b
<i>P. sajor-caju</i>	11.07±2.34 ^a	125.25±12.03 ^b	6.67±0.97 ^a	7.10±0.76 ^{ab}	5.32±0.65 ^a	17.50±1.00 ^a
<i>P. citrinopileatus</i>	5.05±1.79 ^b	169.50±8.96 ^a	5.45±0.70 ^b	6.17±0.53 ^c	3.30±0.21 ^b	11.75±0.50 ^c
CD (0.05)	4.228	10.802	0.929	0.792	0.519	1.199
SE (m)	1.403	3.584	0.308	0.263	0.172	0.398

Values were recorded on 3rd day after primordial initiation

Values are mean ± SD of four replications

Values followed by similar superscripts are not significantly different at 5 % level



Plate 5. Front view of sporocarp of five species of oyster mushrooms
a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

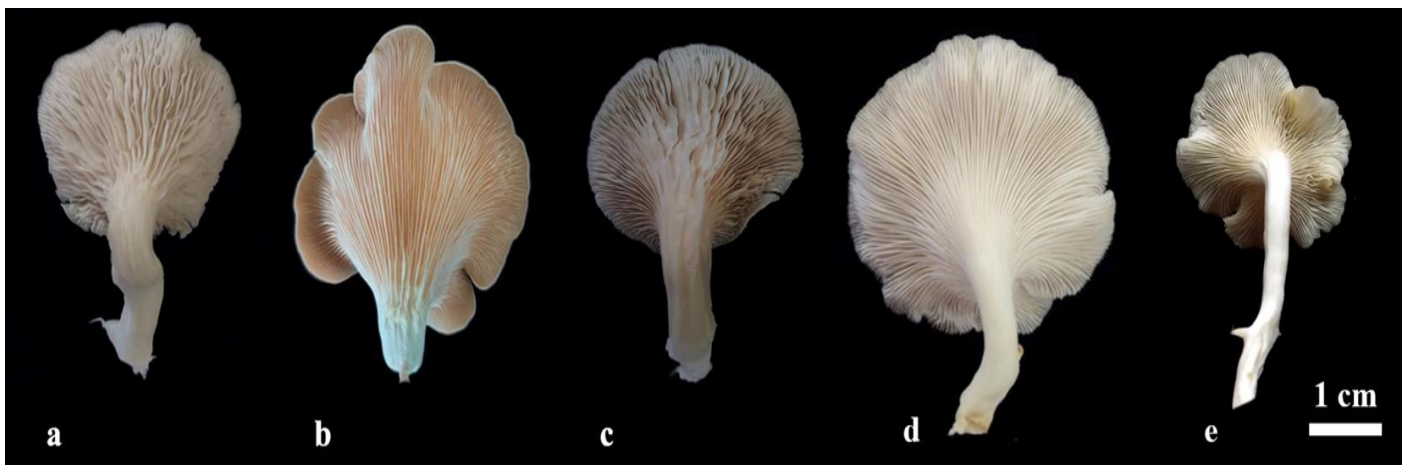


Plate 6. Sporocarp showing gills
a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

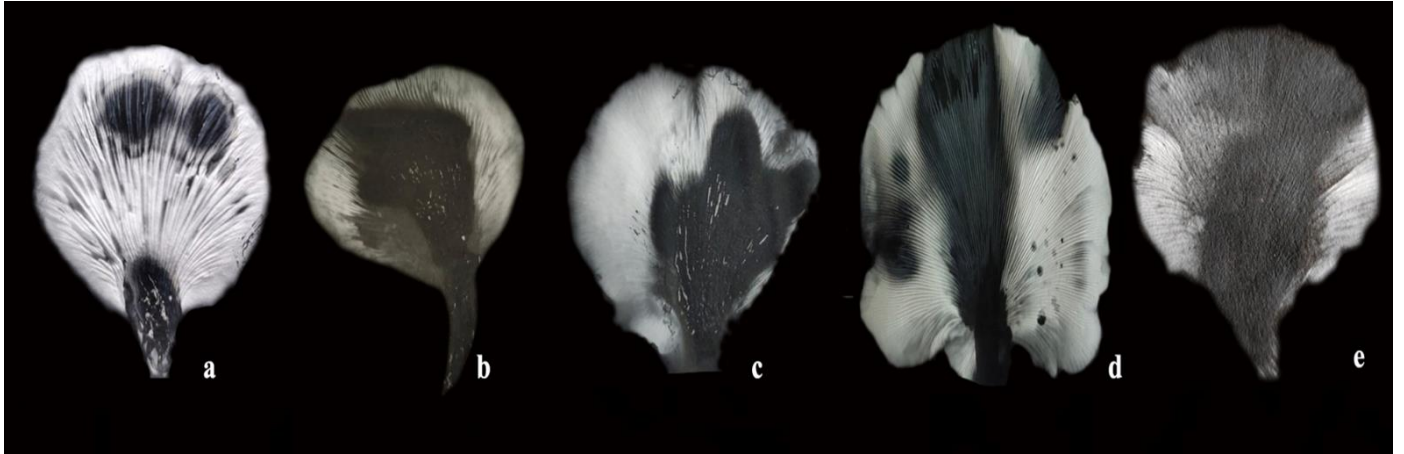


Plate 7. Spore print of five species of oyster mushrooms indicating their gill arrangements
a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

4.1.2 Microscopic observations

Microscopic observations of hyphae and spores of five species of oyster mushrooms were recorded (Table 4). Hyphae of all the five species were septate, branched and hyaline with clamp connections (Plate 9). The width of the hyphae did not vary significantly among each other and it ranged from 1.5-4.5 μm . Basidiospores of *P. florida* and *P. djamor* recorded 7-12 μm in length and 2-5 μm in width whereas, *H. ulmarius* and *P. sajor-caju* produced spores with dimension of 8-12 $\mu\text{m} \times 3-6 \mu\text{m}$. *P. citrinopileatus* produced spores with dimension of 7-10 $\mu\text{m} \times 2-3 \mu\text{m}$ (Plate 8).

4.2 EVALUATION OF PRODUCTION OF MUSHROOMS ON SUITABLE SUBSTRATE

The two substrates namely paddy straw and rubber sawdust were evaluated for the cultivation of five species of oyster mushrooms. Observations on time taken for complete spawn run, time taken for pin-head formation, time taken for first harvest, total crop period and total yield from three harvests were recorded from five species of oyster mushrooms on both the substrates (Plate 10, 11, 12, 13). The comparative performance of five species of oyster mushrooms cultivated in paddy straw and rubber sawdust substrates are given in Table 5 and Table 6.

The yield recorded was highest for *H. ulmarius* cultivated on both paddy straw and rubber sawdust (1233 g kg^{-1} and 1611 g kg^{-1} respectively) which was followed by *P. florida* (1148.30 g kg^{-1} and 1582.66 g kg^{-1} respectively) whereas significantly lower yield was obtained from *P. citrinopileatus* (610 g kg^{-1} and 979 g kg^{-1}). Comparatively less crop period was observed in the case of *P. djamor* cultivated on both the substrates (54.00 days and 90.33 days). Significantly higher crop period was recorded for *P. sajor-caju* cultivated on both the substrates (67.33 and 111.66 days respectively). *P. florida* and *P. citrinopileatus* had a total crop period of 59.00 and 60.66 days respectively from paddy straw substrate.

Table 4: Microscopic characters of five species of oyster mushroom

Oyster mushroom	Basidiospores (μm) (l \times b)	Mycelial width (μm)
<i>P. florida</i>	7-12 x 2-5	1.9 - 4.2
<i>P. djamor</i>	7-11 x 2-4	1.5 - 4.5
<i>H. ulmarius</i>	8-12 x 3-5	2.0 - 4.5
<i>P. sajor-caju</i>	8-12 x 3-6	2.0 - 4.5
<i>P. citrinopileatus</i>	7-10 x 2-3	1.9 - 4.2

Values are range of 30 observations

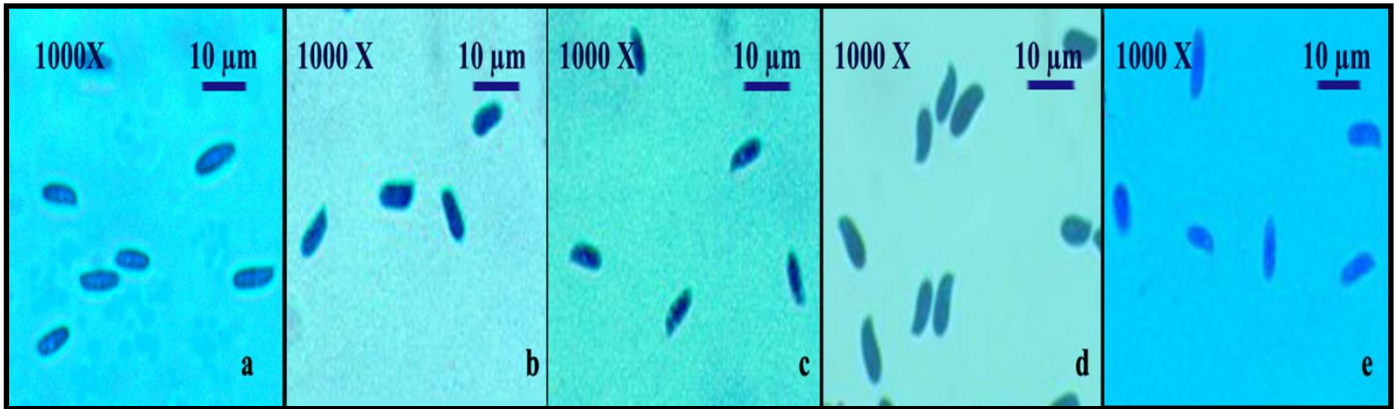


Plate 8. Cylindrical shaped basidiospores of five species of oyster mushrooms
 a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

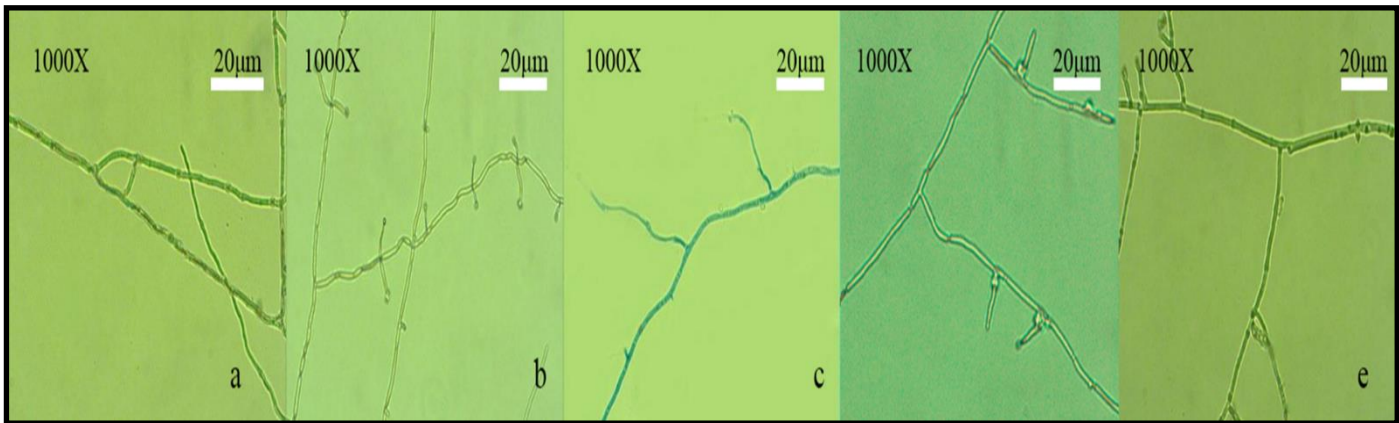


Plate 9. Mycelium of five species of oyster mushrooms
 a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

Table 5. Comparative performance of five species of oyster mushrooms cultivated in paddy straw substrate

Sl. No	Species of Oyster mushroom	Time taken for complete spawn run (days)	Time taken for pin-head formation (days)	Time taken for first harvest (days)	Total crop period (days)	Total yield from three harvests (g)	BE (%)
1	<i>P. florida</i>	15.66± 1.15 ^c	19.00± 1.00 ^c	23.00± 1.00 ^b	59.00± 1.73 ^c	1148.30± 39.06 ^b	114.83
2	<i>P. djamor</i>	11.66± 0.57 ^d	15.00± 0.00 ^d	17.66± 0.57 ^c	54.00± 1.00 ^d	1050.30± 10.78 ^c	105.03
3	<i>H. ulmarius</i>	16.33± 0.57 ^{bc}	20.33± 0.57 ^b	23.66± 1.15 ^b	63.66± 1.15 ^b	1233± 19.46 ^a	123.30
4	<i>P. sajor-caju</i>	19.33± 0.57 ^a	23.00± 0.00 ^a	26.33± 0.57 ^a	67.33± 2.08 ^a	731.60± 24.00 ^d	73.16
5	<i>P. citrinopileatus</i>	17.66± 0.57 ^b	21.33± 0.57 ^b	24.33± 0.57 ^b	60.66± 0.57 ^c	610.00± 68.00 ^e	61.00
	CD (0.05)	1.329	1.050	1.485	2.573	69.142	
	SE (m)	0.422	0.333	0.471	0.816	21.942	

Values are mean ± SD of three replications

Values followed by similar superscripts are not significantly different at 5 % level

Table 6. Comparative performance of five species of oyster mushrooms cultivated in rubber sawdust substrate

Sl. No	Species of Oyster mushroom	Time taken for complete spawn run (days)	Time taken for pin-head formation (days)	Time taken for first harvest (days)	Total crop period (days)	Total yield from three harvests (g)	BE (%)
1	<i>P. florida</i>	25.66± 0.57 ^{ab}	30.00± 1.15 ^b	34.66± 0.57 ^a	98.33± 5.03 ^{bc}	1582.66± 59.53 ^a	158.26
2	<i>P. djamor</i>	21.66± 1.52 ^c	25.33± 1.52 ^c	29.00± 1.00 ^c	90.33± 1.52 ^c	1292.33± 11.24 ^b	129.23
3	<i>H. ulmarius</i>	25.00± 1.00 ^b	29.66± 0.57 ^b	32.66± 0.57 ^b	101.00± 7.93 ^b	1611.00± 22.51 ^a	161.10
4	<i>P. sajor-caju</i>	27.33± 1.15 ^a	32.00± 1.00 ^a	35.00± 1.00 ^a	111.66± 3.05 ^a	1067.00± 73.89 ^c	106.70
5	<i>P. citrinopileatus</i>	24.66± 0.57 ^b	29.33± 1.15 ^b	32.00± 1.00 ^b	102.66± 2.51 ^b	979.00± 31.79 ^d	97.90
	CD (0.05)	1.879	1.993	1.558	8.390	83.961	
	SE (m)	0.596	0.632	0.494	2.662	26.645	

Values are mean ± SD of three replications

Values followed by similar superscripts are not significantly different at 5 % level

The effect of paddy straw and rubber sawdust on time taken for complete spawn run, time taken for pinhead formation, total crop period and total yield from three harvests are produced in Table 7.

Selection of substrate significantly affected the time taken for complete spawn run. Time taken for complete spawn was significantly less (16.20 days) in paddy straw (S₁) and superior to rubber sawdust (S₂) which took 24.86 days to complete spawn run. In case of mushroom species, *P. djamor* completed spawn run within 16.66 days and was significantly superior to other species and it was followed by *P. florida* (20.66 days), *H. ulmarius* (20.83 days) and *P. citrinopileatus* (21.16 days) which were on par and significantly superior to *P. sajor-caju* (23.33 days).

Interaction effect showed significant differences in time taken for complete spawn run. The treatment combination S₁M₂ (*P. djamor* on paddy straw) took less time to complete spawn run and was found to be significantly superior to other treatment combinations.

Selection of substrate significantly affected the total yield. Total yield from first three harvests were significantly less in paddy straw, S₁ (954.66) compared to rubber sawdust (S₂) which was superior and given a yield of 1306.40 g. In the case of mushroom species, *H. ulmarius* recorded the highest yield (1422.00 g) and found to be superior. It was followed by *P. florida* and *P. djamor* (1365.50 g and 1171.33 g) respectively which did vary significantly. The lowest yield was recorded from *P. citrinopileatus* (794.50 g).

Interaction effect showed significant differences in total yield from three harvests. The treatment combination S₂M₁ (*P. florida* in rubber sawdust) produced the highest yield (1582.66 g) and was found to be significantly superior to other treatment combinations. It was followed by S₁M₃ (*H. ulmarius* in paddy straw), S₂M₂ (*P. djamor* in rubber sawdust) and S₂M₃ (*H. ulmarius* in rubber sawdust) which were statistically on par with each other. Significantly lower yield was recorded from S₁M₅ (*P. citrinopileatus* in paddy straw) and S₂M₅ (*P. citrinopileatus* in rubber sawdust) which were on par with each other.

Table 7. Effect of paddy straw and rubber saw dust on time taken for complete spawn run, pinhead formation, total crop period and total yield from three harvests

Treatments	Time taken for complete spawn run (days)	Time taken for pin-head formation (days)	Time taken for first harvest (days)	Total crop period (days)	Total yield from three harvests (g)
SUBSTRATE					
Paddy straw (S ₁)	16.20± 2.73 ^b	19.73± 2.84 ^b	23.00± 3.07 ^b	60.93± 4.78 ^b	954.66± 252.47 ^b
Rubber sawdust (S ₂)	24.86± 2.10 ^a	29.26± 2.43 ^a	32.66± 2.35 ^a	100.80± 8.12 ^a	1306.40± 270.28 ^a
SE (m)	0.231	0.226	0.216	0.881	10.915
CD (0.05)	0.681	0.667	0.637	2.598	32.20
MUSHROOM SPECIES					
<i>P. florida</i> (M ₁)	20.66± 5.53 ^b	24.50± 6.09 ^b	28.83± 6.43 ^b	78.66± 21.80	1365.50± 242.12 ^b
<i>P. djamor</i> (M ₂)	16.66± 5.57 ^c	20.16± 5.74 ^c	23.33± 6.25 ^c	72.16± 19.93	1171.33± 132.91 ^c
<i>H. ulmarius</i> (M ₃)	20.83± 4.62 ^b	25.00± 5.13 ^b	28.16± 4.99 ^b	82.33± 21.068	1422.00± 207.89 ^a
<i>P. sajor-caju</i> (M ₄)	23.33± 4.45 ^a	27.50± 4.97 ^a	30.66± 4.80 ^a	89.50± 24.39	899.33± 190.13 ^d
<i>P. citrinopileatus</i> (M ₅)	21.16± 3.86 ^b	25.33± 4.45 ^b	28.16± 4.26 ^b	81.66± 23.06	794.50± 207.61 ^e
SE (m)	0.365	0.357	0.342	1.392	10.915
CD (0.05)	1.077	1.055	1.008	4.108	32.200
INTERACTION EFFECTS					
S ₁ M ₁	15.66± 1.15 ^f	19.00± 1.00	23.00± 1.00 ^e	59.00± 1.73	1148.33± 39.06 ^c
S ₁ M ₂	11.66± 0.57 ^g	15.00± 0.00	17.66± 0.57 ^f	54.00± 1.00	1050.33± 10.78 ^{de}
S ₁ M ₃	16.66± 0.57 ^{ef}	20.33± 0.57	23.66± 1.15 ^e	63.66± 1.15	1233.00± 19.46 ^b
S ₁ M ₄	19.33± 0.57 ^d	23.00± 0.00	26.33± 0.57 ^d	67.33± 2.08	731.66± 24.00 ^f
S ₁ M ₅	17.66± 0.57 ^e	21.33± 0.57	24.33± 0.57 ^e	60.66± 0.57	610.00± 68.00 ^e
S ₂ M ₁	25.66± 0.57 ^b	30.00± 1.00	34.66± 0.57 ^a	98.33± 5.03	1582.66± 59.53 ^a
S ₂ M ₂	21.66± 1.52 ^c	25.33± 1.52	29.00± 1.00 ^c	90.33± 1.52	1292.33± 11.24 ^b
S ₂ M ₃	25.00± 1.00 ^b	29.66± 0.57	32.66± 0.57 ^b	101.00± 7.93	1611.00± 22.51 ^b
S ₂ M ₄	27.33± 1.15 ^a	32.00± 1.00	35.00± 1.00 ^a	111.66± 3.05	1067.00± 73.89 ^d
S ₂ M ₅	24.66± 0.57 ^b	29.33± 1.15	32.00± 1.00 ^b	102.66± 2.51	979.00± 31.79 ^e
SE (m)	0.516	0.506	0.483	1.969	24.407
CD (0.05)	1.523	NS	1.425	NS	72.002



Plate 12. Spawn run of different species of oyster mushrooms in rubber sawdust substrate after 10 days
a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*



Plate 13. Spawn run of different species of oyster mushrooms rubber sawdust substrate after 20 days
a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*



Plate 10. Spawn run of different species of oyster mushrooms in paddy straw substrate after 6 days
a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

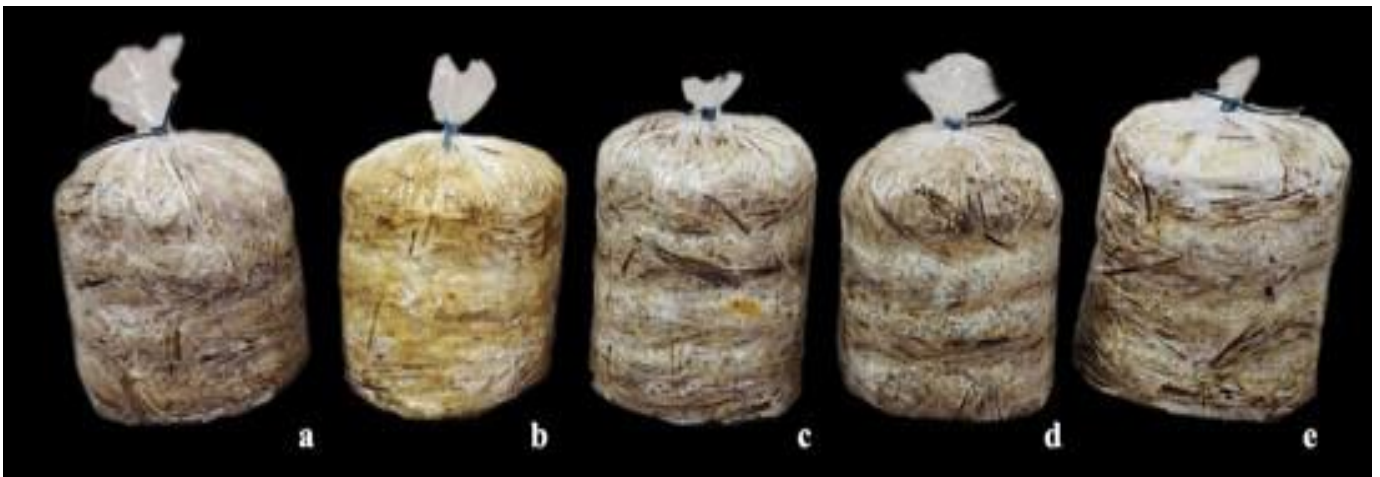


Plate 11. Spawn run of different species of oyster mushrooms in paddy straw substrate after 13 days
a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

4.3 EVALUATION OF PRODUCTION OF MUSHROOMS UNDER DIFFERENT AGRO-ECOLOGICAL CONDITION

Kerala is divided into five agro-ecological zones from coastal plains to high hills with each zone having wider climatic conditions. Locations and mushroom farmers were selected in each of the five agro-ecological zones and the five species of oyster mushrooms were cultivated in farmer's field (Plate 14). In all the five locations, observations on time taken for complete spawn run, time taken for pin-head formation, time taken for first harvest, total crop period and total yield from three harvests were recorded from five species of oyster mushrooms. The location and address of mushroom farmers are given in Table 8.

Five species of oyster mushrooms were cultivated in coastal plains (Plate 15, 16). In coastal plains, the highest yield of 1029.96 g with BE of 102.99 per cent was recorded from *H. ulmarius* which was followed by *P. florida* (886.20 g) and *P. djamor* (723.00 g). Lower yield of 550.10 g was obtained from *P. sajor-caju* and *P. citrinopileatus* (630.00 g) which vary significantly. In terms of time taken for complete spawn run, *P. djamor* was significantly superior which took the least number of days (11.66) followed by *H. ulmarius* (15.33). The time taken for complete spawn run was significantly higher in *P. sajor-caju* (18.33 days) followed by *P. citrinopileatus* (16.66). First harvest was obtained from *P. djamor* in 18.66 days which was followed by *H. ulmarius* and *P. florida* (22.33 and 23.33 days) respectively. *P. djamor* completed the crop period within 53.66 days followed by *H. ulmarius* and *P. citrinopileatus* (58.66 and 60.66 days respectively). Total yield from first harvest was significantly higher in *H. ulmarius* (520.00 g) followed by *P. florida* (432.20 g). Total yield from third harvest was significantly lower in *P. sajor-caju* (50.03 g) and *P. citrinopileatus* (62.00g). *H. ulmarius* was found to be the best suited oyster mushroom species for cultivation in coastal plains. Comparative performance of five species of oyster mushrooms cultivated in paddy straw substrate in coastal plains is given in Table 9.

Table 8. Selected locations in five agro-ecological zones in Kerala

Sl. No	Agro-ecological zone	Location	Latitude and longitude
1	Coastal plains	Mushroom production unit, Dept. of Plant Pathology, College of Agriculture, Padannakkad, Pin- 671328	12° 15' 13.7" N 75° 07' 03.4" E
2	Midland laterites	Suma Devi S, KRS bhavan, Moolayam, Aaliyad P.O, Venjaramoodu Thiruvananthapuram, Pin- 695607	8° 40' 18.8" N 76° 53' 37.0" E
3	Foothills	Lalu Thomas, Kalluvila Grace, Parankimammukal, Kunnicothu, Kottarakkara, Kollam, Pin- 691508	9° 03' 34.1" N 76° 50' 20.0" E
4	High hills	Bhagyaraj, Manjula bhavan, Punnamudi, Parakkottu, Vagamon, Pin- 685503	9° 41' 10.3" N 76° 54' 18.7" E
5	Palakkad plains	Abhilash R, House No 31, Green valley Colony, Chadayan kalai Kanjikode, Palakkad, Pin- 678623	10° 47' 38.8" N 76° 43' 51.2" E

Table 9. Comparative performance of five species of oyster mushrooms cultivated in paddy straw substrate in coastal plains

Sl. No	Species of oyster mushroom	Time taken for complete spawn run (days)	Time taken for pin-head formation (days)	Time taken for first harvest (days)	Total crop period (days)	Total yield from first harvest (g)	Total yield from second harvest (g)	Total yield from third harvest (g)	Total yield from three harvests (g)	BE (%)
1	<i>P. florida</i>	16.00± 1.00 ^{ab}	19.33± 1.15 ^b	23.33± 1.55 ^b	61.66± 3.21 ^b	432.20± 1.00 ^b	274.00±4.00 ^b	180.00± 15.00 ^b	886.20± 10.41 ^b	88.62
2	<i>P. djamor</i>	11.66± 1.15 ^c	14.66± 0.57 ^c	18.66± 1.15 ^c	53.66± 1.52 ^c	373.00±3.00 ^c	210.00±10.00 ^c	140.00± 5.00 ^c	723.00± 10.14 ^c	72.30
3	<i>H. ulmarius</i>	15.33± 1.15 ^b	19.66± 0.57 ^b	22.33± 0.57 ^b	58.66± 3.21 ^b	520.00±5.00 ^a	309.09±0.00 ^a	200.87± 0.00 ^a	1029.96± 22.96 ^a	102.99
4	<i>P. sajor caju</i>	18.33± 1.52 ^a	22.66± 2.08 ^a	26.00± 2.00 ^a	67.00± 1.73 ^a	350.00±3.00 ^d	150.10±2.00 ^e	50.00± 3.00 ^d	550.10± 18.67 ^e	55.01
5	<i>P. citrinopileatus</i>	16.66± 1.52 ^{ab}	18.66± 1.52 ^b	24.33± 1.52 ^{ab}	60.66± 2.08 ^b	370.00±7.00 ^c	198.00± 2.00 ^d	62.00± 2.00 ^d	630.00± 13.22 ^d	63.00
	CD (0.05)	2.349	2.395	2.486	4.481	7.846	9.060	13.194	28.908	
	SE (m)	0.745	0.760	0.789	1.422	2.490	2.875	4.187	9.174	

Values are mean ± SD of three replications

Values followed by similar superscripts are not significantly different at 5 % level

In midland laterites, the five species of oyster mushrooms were cultivated (Plate 17). Highest yield of 827.00 g was obtained from *P. djamor*. The yield recorded from *H. ulmarius* and *P. djamor* were on par with each other (806.00 g and 773.03 g respectively). Significantly lower yield of 658.83 g was recorded from *P. sajor-caju* in midland laterites. Total yield from first harvest was significantly higher in *P. djamor* (425.00 g) followed by *H. ulmarius* (389.00 g). Total yield from third harvest was significantly low in *P. citrinopileatus* (75.00 g). *P. djamor* took significantly less time to complete the spawn run (12.66 days). *H. ulmarius*, *P. florida* and *P. citrinopileatus* did not vary significantly in the time taken for completing spawn run and took 17.33 days, 17.66 days and 18.33 days respectively. *P. sajor-caju* took the highest number of days to complete spawn run (20.33 days). Earliness in primordial initiation was observed from *P. djamor* (16.00 days) which also was harvested in 19.33 days. *P. djamor* completed the total crop period within 57.00 days followed by *P. florida* (63.00 days). *P. djamor* was found to be the best suited oyster mushroom species for cultivation in midland laterites. Comparative performance of five species of oyster mushrooms cultivated in paddy straw substrate in midland laterites is given in Table 10.

Five species of oyster mushrooms were cultivated in foothills (Plate 18). In foothills, the highest yield of 927.33 g was recorded from *H. ulmarius*, which was followed by *P. florida* (832.33 g). The yield obtained from *P. sajor-caju* (811.00 g) and *P. citrinopileatus* (800.00 g) did not vary significantly. Significantly lower yield was recorded from *P. djamor*. Total yield from first harvest was significantly higher in *H. ulmarius* (455.33 g) followed by *P. florida* (428.33 g). Of all the five species of oyster mushrooms, *P. djamor* took the least number of days to complete spawn run (15.33 days) which was significantly less and followed by *H. ulmarius* (17.66 days). *P. sajor-caju* took the maximum number of days to complete spawn run (19.66 days). Earliness in primordial initiation was recorded from *P. djamor* (17.00 days) and time taken for first harvest was found to be significantly less in *P. djamor*, which yielded in 20.00 days. *P. florida*, *H. ulmarius* and *P. citrinopileatus* took 25.33 days, 24.00 days and 25.33 days respectively for first harvest. *P. djamor* completed the total crop period within 53.66 days followed by *P. florida* (59.33 days). *H. ulmarius* was found



Plate 14. Mushroom farmers in five agro-ecological zones of Kerala

a. Coastal plains; b. Foot hills; c. Midland laterites; d. High hills; e. Palakkad plains



Plate 15. Pin-head formation in five species of oyster mushrooms in Coastal plains
a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

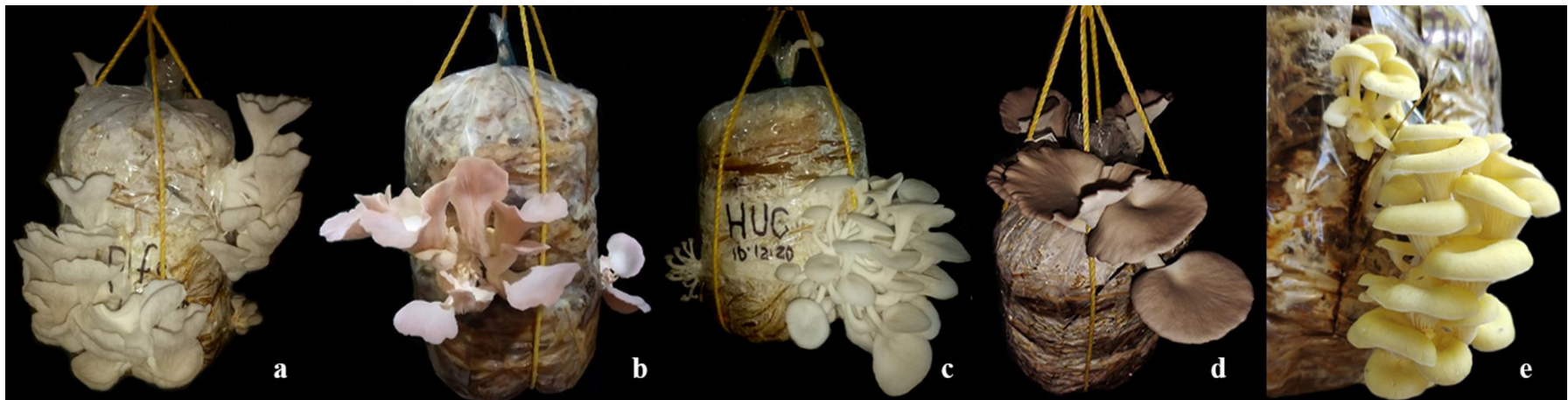


Plate 16. Mature fruiting bodies of oyster mushrooms cultivated in Coastal plains
a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

Table 10. Comparative performance of five species of Oyster mushrooms cultivated in paddy straw substrate in midland laterites

Sl. No	Species of oyster mushroom	Time taken for complete spawn run (days)	Time taken for pin-head formation (days)	Time taken for first harvest (days)	Total crop period (days)	Total yield from first harvest (g)	Total yield from second harvest (g)	Total yield from third harvest (g)	Total yield from three harvests (g)	BE (%)
1	<i>P. florida</i>	17.66± 1.15 ^b	21.00± 1.00 ^b	23.33± 0.57 ^c	63.00± 2.64 ^c	73.00±1.00 ^c	250.00±4.00 ^b	150.03±1.00 ^a	773.03± 50.26 ^{ab}	77.30
2	<i>P. djamor</i>	12.66±0.57 ^c	16.00± 1.00 ^c	19.33± 0.57 ^d	57.00± 2.64 ^d	425.00±2.00 ^a	252.00±2.00 ^b	150.00±6.00 ^a	827.00± 36.51 ^a	82.70
3	<i>H. ulmarius</i>	17.33± 1.15 ^b	21.66± 0.57 ^b	24.33± 0.57 ^{bc}	67.66±2.51 ^b	389.00±11.00 ^b	277.00±3.00 ^a	140.00±2.00 ^b	806.00± 19.51 ^{ab}	80.60
4	<i>P. sajor caju</i>	20.33± 1.15 ^a	23.33± 0.57 ^a	27.33± 0.57 ^a	74.33± 2.08 ^a	345.83±1.00 ^d	223.00±3.00 ^c	90.00±3.00 ^c	658.83± 30.52 ^c	65.88
5	<i>P. citrinopileatus</i>	18.33± 1.15 ^b	21.00± 1.00 ^b	25.33± 0.57 ^b	69.33± 0.57 ^b	390.00±5.00 ^b	85.00±10.00 ^a	75.00±5.00 ^d	750.00± 15.00 ^b	75.00
	CD (0.05)	1.937	1.329	1.050	4.068	10.031	9.558	7.046	59.774	
	SE (m)	0.615	0.422	0.333	1.291	3.183	3.033	2.236	18.969	

Values are mean ± SD of three replications

Values followed by similar superscripts are not significantly different at 5 % level

Table 11. Comparative performance of five species of Oyster mushrooms cultivated in paddy straw substrate in foothills

Sl. No	Species of oyster mushroom	Time taken for complete spawn run (days)	Time taken for pin-head formation (days)	Time taken for first harvest (days)	Total crop period (days)	Total yield from first harvest (g)	Total yield from second harvest (g)	Total yield from third harvest (g)	Total yield from three harvests (g)	BE (%)
1	<i>P. florida</i>	18.33± 1.52 ^{ab}	22.00± 1.00 ^{bc}	25.33± 0.57 ^b	59.33± 2.30 ^c	428.33±2.00 ^b	234.00±2.00 ^c	170.00±5.00 ^b	832.33±40.07 ^b	83.23
2	<i>P. djamor</i>	15.33± 0.57 ^c	17.00± 1.00 ^d	20.00± 1.00 ^c	53.66± 2.51 ^d	385.50±2.00 ^d	295.10±3.00 ^a	90.00±3.00 ^e	768.60±23.15 ^c	76.86
3	<i>H. ulmarius</i>	17.66± 1.15 ^b	20.66± 0.57 ^c	24.00± 1.00 ^b	60.33± 0.57 ^{bc}	5.33±10.00 ^a	252.00±4.00 ^b	220.00±0.00 ^a	927.33±42.02 ^a	92.73
4	<i>P. sajor caju</i>	19.66± 1.15 ^a	23.66± 1.00 ^a	27.00± 1.00 ^a	64.00± 1.73 ^a	423.00±3.00 ^b	58.00±10.00 ^b	130.00±2.00 ^c	811.00±34.59 ^{bc}	81.10
5	<i>P. citrinopileatus</i>	18.66± 0.57 ^{ab}	22.33± 0.57 ^{ab}	25.33± 0.57 ^b	63.66± 1.52 ^{ab}	395.00±5.00 ^c	290.00±5.00 ^a	15.00±10.00 ^d	800.00±28.05 ^{bc}	80.00
	CD (0.05)	1.937	1.627	1.558	3.387	9.695	10.097	9.558	62.457	
	SE (m)	0.615	0.516	0.494	1.075	3.077	3.204	3.033	19.821	

Values are mean ± SD of three replications

Values followed by similar superscripts are not significantly different at 5 % level

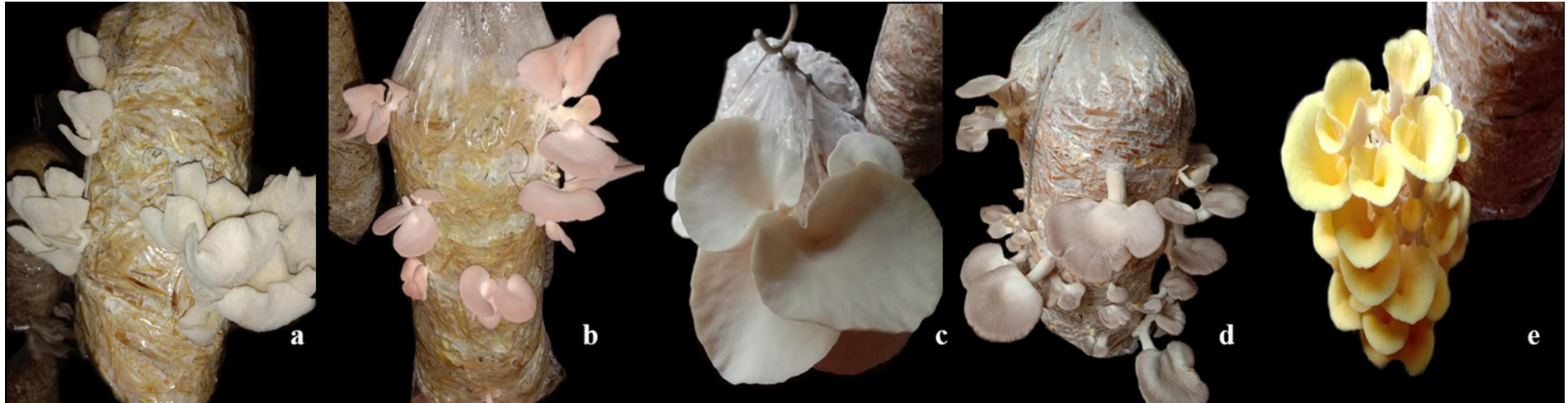


Plate 17. Matured fruiting bodies of oyster mushrooms cultivated in midland laterites
 a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

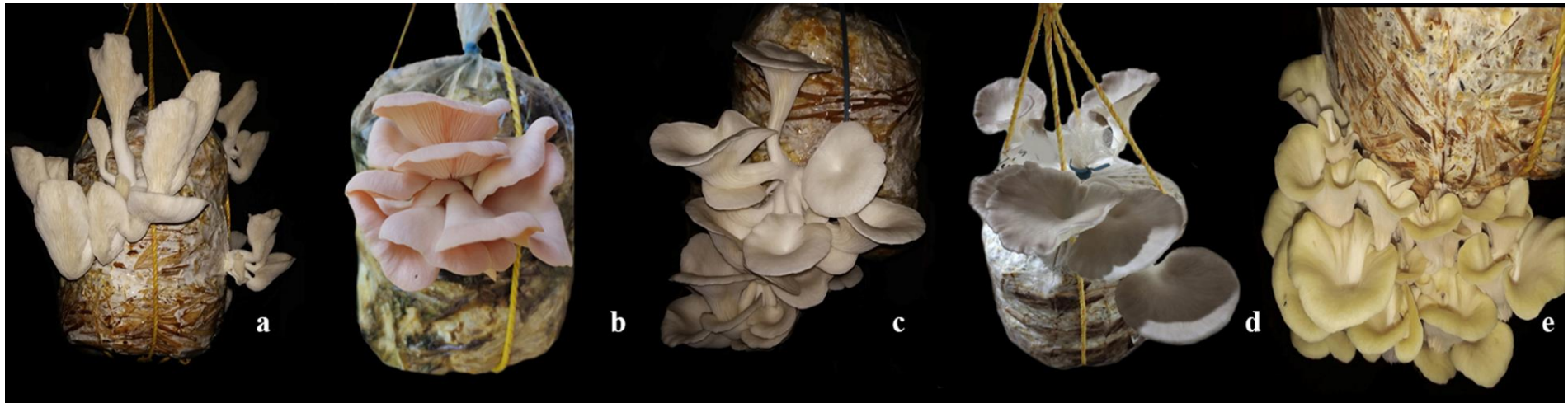


Plate 18. Matured fruiting bodies of oyster mushrooms cultivated in foothills
 a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

to be the best suited oyster mushroom species for cultivation in foothills. Comparative performance of five species of oyster mushrooms cultivated in paddy straw substrate in foothills in Table 11.

Five species of Oyster mushrooms were cultivated in high hills (Plate 19). In high hills, the highest yield of 1233.00 g was obtained from *H. ulmarius*, followed by *P. florida* (1148.30 g) and *P. djamor* (1050.30 g). Significantly lower yield was obtained from *P. citrinopileatus* (610.00 g). Total yield from first harvest was significantly higher in *H. ulmarius* (658.30 g) followed by *P. djamor* (530.30 g). *P. djamor* took the least number of days to complete spawn run in high hills (11.66 days) followed by *P. florida* (15.66 days). First harvest was obtained from *P. djamor* within 17.66 days after bed preparation. *P. florida*, *H. ulmarius* and *P. citrinopileatus* took 23.00 days, 23.66 days and 24.33 days respectively for first harvest which were on par with each other. *P. djamor* completed total crop period in 54.00 days while *P. sajor-caju* took 67.33 days to complete the crop period. *H. ulmarius*, *P. florida* and *P. djamor* were found to be best suitable for cultivation in high hills based on comparatively low total cropping period and high yield. Comparative performance of five species of oyster mushrooms cultivated in paddy straw substrate in high hills in Table 12.

Five species of Oyster mushrooms were cultivated in Palakkad plains (Plate 20). In Palakkad plains, *P. djamor* produced significantly higher yield of 1038.00 g compared to other species of oyster mushrooms. It was followed by *H. ulmarius* (824.60 g) and *P. florida* (749.00 g). Comparatively lower yield of 560.00 g was obtained from *P. citrinopileatus* in Palakkad plains. Total yield from first, second and third harvest was obtained from *P. djamor* which yielded 502.00 g, 335.00g and 201.00 g respectively. Significantly low yield was recorded from *P. citrinopileatus* from third harvest (28.00 g). *P. djamor* took 15.00 days to complete spawn run and was followed by *P. florida* and *H. ulmarius* (17.66 days and 18.00 days respectively) which were on par with each other. *P. sajor-caju* took the higher number of days to complete spawn run (21.33 days). *P. djamor* was harvested within 22.33 days after bed preparation, followed by *H. ulmarius* (24.33 days). *P. djamor* and *P. florida* completed total crop period in 60.66 days and 62.33 days respectively and did not vary significantly. *P. sajor-caju* took 72.66 days to complete the crop period which was significantly higher. *P. djamor* was found to be the best suited oyster mushroom species for cultivation in Palakkad plains. Comparative performance of five species of oyster mushrooms cultivated in paddy straw substrate in Palakkad plains is given in Table 13.

Table 12. Comparative performance of five species of oyster mushrooms cultivated in paddy straw in high hills

Sl. No	Species of oyster mushroom	Time taken for complete spawn run (days)	Time taken for pin-head formation (days)	Time taken for first harvest (days)	Total crop period (days)	Total yield from first harvest (g)	Total yield from second harvest (g)	Total yield from third harvest (g)	Total yield from three harvests (g)	BE (%)
1	<i>P. florida</i>	15.66± 1.15 ^c	19.00± 1.00 ^c	23.00± 1.00 ^b	59.00± 1.73 ^c	495.30±5.00 ^c	393.00±0.00 ^a	60.00±10.00 ^a	1148.30± 39.06 ^b	114.83
2	<i>P. djamor</i>	11.66± 0.57 ^d	15.00± 0.00 ^d	17.66± 0.57 ^c	54.00± 1.00 ^d	530.30±10.00 ^b	25.00±10.00 ^c	195.00±5.00 ^c	1050.30± 10.78 ^c	105.03
3	<i>H. ulmarius</i>	16.33± 0.57 ^{bc}	20.33± 0.57 ^b	23.66± 1.15 ^b	63.66± 1.15 ^b	658.30±2.00 ^a	354.70±4.00 ^b	220.00±0.00 ^b	1233± 19.46 ^a	123.30
4	<i>P. sajor caju</i>	19.33± 0.57 ^a	23.00± 0.00 ^a	26.33± 0.57 ^a	67.33± 2.08 ^a	382.50±2.00 ^d	269.10±0.00 ^d	80.00±20.00 ^d	731.60± 24.00 ^d	73.16
5	<i>P. citrinopileatus</i>	17.66± 0.57 ^b	21.33± 0.57 ^b	24.33± 0.57 ^b	60.66± 0.57 ^c	320.00±10.00 ^e	225.00±5.00 ^e	65.00±5.00 ^d	610.00± 68.00 ^e	61.00
	CD (0.05)	1.329	1.050	1.485	2.573	12.419	9.661	19.081	69.142	
	SE (m)	0.422	0.333	0.471	0.816	5.574	4.336	6.055	21.942	

Values are mean ± SD of three replications

Values followed by similar superscripts are not significantly different at 5 % level

Table 13. Comparative performance of five species of Oyster mushrooms cultivated in paddy straw substrate in Palakkad plains

Sl. No	Species of oyster mushroom	Time taken for complete spawn run (days)	Time taken for pin-head formation (days)	Time taken for first harvest (days)	Total crop period (days)	Total yield from first harvest (g)	Total yield from second harvest (g)	Total yield from third harvest (g)	Total yield from three harvests (g)	BE (%)
1	<i>P. florida</i>	17.66± 0.57 ^c	21.33± 0.57 ^c	25.00± 1.00 ^{bc}	62.33± 2.51 ^c	450.00±10.00 ^b	185.00±5.00 ^e	114.00±6.00 ^b	749± 13.52 ^c	74.90
2	<i>P. djamor</i>	15± 1.00 ^d	19.33± 0.57 ^d	22.33± 0.57 ^d	60.66± 2.51 ^c	502.00±8.00 ^a	335.00±1.00 ^a	201.00±10.00 ^a	1038± 27.62 ^a	103.80
3	<i>H. ulmarius</i>	18± 0.00 ^c	21.33± 0.57 ^c	24.33± 0.57 ^c	66.66± 1.52 ^b	386.50±10.00 ^c	325.10±1.00 ^b	113.00±3.00 ^b	824.6± 13.50 ^b	82.46
4	<i>P. sajor caju</i>	21.33± 0.57 ^a	24.33± 0.57 ^a	27.66± 0.57 ^a	72.66± 2.08 ^a	325.50±5.00 ^d	202.30±2.00 ^d	86.50±1.00 ^c	614.3± 12.01 ^d	61.43
5	<i>P. citrinopileatus</i>	19.33± 0.57 ^b	23.00± 1.00 ^b	25.66± 0.57 ^b	68.66± 1.52 ^b	320.00±2.00 ^d	212.00±2.00 ^c	28.00±0.00 ^d	560± 27.18 ^e	56.02
	CD (0.05)	1.151	1.243	1.243	3.787	13.927	4.813	9.831	36.491	
	SE (m)	0.365	0.394	0.394	1.202	4.420	1.528	3.120	11.581	

Values are mean ± SD of three replications

Values followed by similar superscripts are not significantly different at 5 % level

Comparative yield performance of five species of oyster mushrooms in five agro-ecological zones revealed that *H. ulmarius* was the best suited oyster mushroom species for cultivation in coastal plains (102.99 % BE), foothills (92.73 % BE) and high hills (123.30 % BE) (Table 14). *P. djamor* was found to be the best yielder (82.70 % BE) in midland laterites and Palakkad plains (103.8 % BE).

4.4 ANALYSIS OF PROXIMATE CONSTITUENTS

Proximate constituents in the five species of oyster mushroom were evaluated. The results on protein, fat, fibre, carbohydrate, moisture content and amino acid contents are presented in Table (15)

4.4.1 Estimation of protein

Protein content recorded was significantly higher in *P. sajor-caju* (26.02 %). This was followed by *P. citrinopileatus* (23.07 %) and *P. florida* (22.42 %) which were on par with each other. Significantly low protein content was found to be present in *P. djamor* (19.55 %).

4.4.2 Estimation of fat

Minimum fat content was estimated on *H. ulmarius* (1.70 %) which is superior to other species studied. Fat content of *P. florida* (2.82 %) and *P. sajor-caju* (2.50 %) did not vary significantly. *P. djamor* recorded fat content of 1.97 per cent and significantly higher fat content was recorded on *P. citrinopileatus* (3.60 %) which differed significantly from other species of oyster mushrooms.

4.4.3 Estimation of fibre

Maximum crude fibre content was estimated in *P. djamor* (26.20 %) followed by *P. florida* (24.21%). *P. sajor-caju* and *P. citrinopileatus* had fibre content of 23.77 and 21.15 per cent respectively. The least crude fibre content was recorded in *H. ulmarius* (18.47 %) which differed significantly from other species of oyster mushrooms.

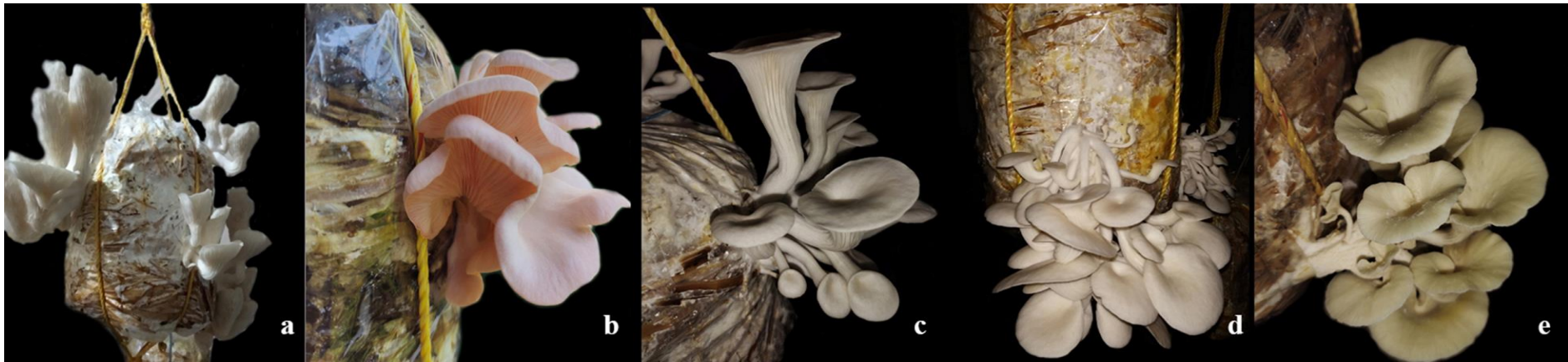


Plate 19. Matured fruiting bodies of oyster mushrooms cultivated in High hills
 a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

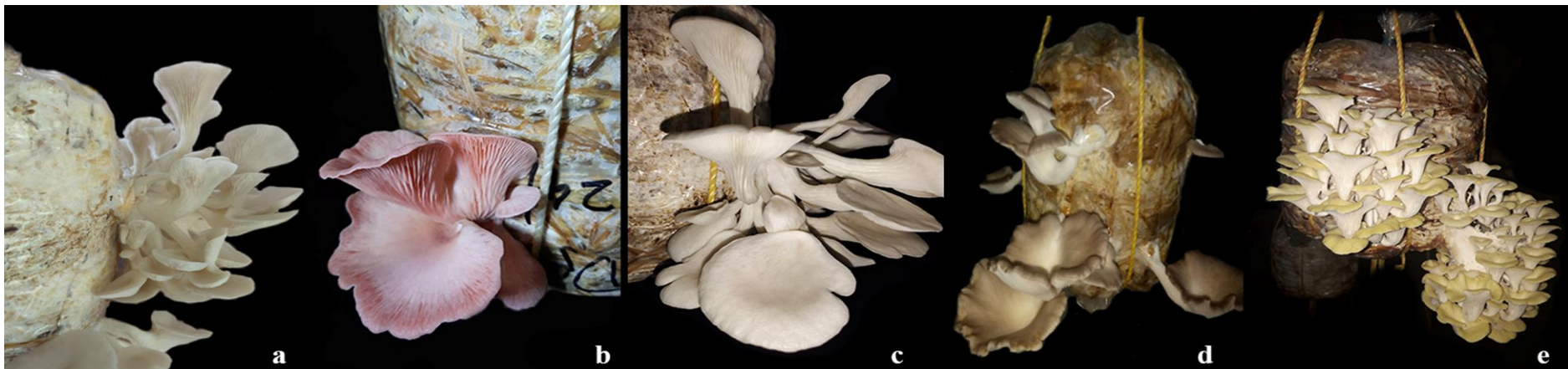


Plate 20. Matured fruiting bodies of oyster mushrooms cultivated in Palakkad plains
 a. *P. florida*; b. *P. djamor*; c. *H. ulmarius*; d. *P. sajor-caju*; e. *P. citrinopileatus*

Table 14. Yield of five species of oyster mushrooms in five agro-ecological zones of Kerala

	Yield of Oyster mushroom (g/kg of substrate)				
	<i>P. florida</i>	<i>P. djamor</i>	<i>H. ulmarius</i>	<i>P. sajor-caju</i>	<i>P. citrinopileatus</i>
Coastal plains	886.20	723.00	1029.96	550.10	630.00
Midland laterites	773.03	827.00	806.00	658.83	750.00
Foothills	823.33	768.60	927.33	811.00	800.00
High hills	1148.30	1050.30	1233.00	731.60	610.00
Palakkad plains	749.00	1038.00	824.60	614.30	560.00

Table 15. Proximate constituents in five species of oyster mushroom

Mushroom	Protein (per cent content)	Fat (per cent content)	Fibre (per cent content)	Carbohydrate (per cent content)	Moisture content (per cent content)	Amino acid (per cent content)
<i>P. florida</i>	22.42± 2.26 ^b	2.82± 0.17 ^b	24.21± 1.14 ^{ab}	29.17± 0.94 ^{ab}	90.97± 1.00 ^a	13.57± 0.51 ^c
<i>P. djamor</i>	19.55± 0.55 ^c	1.97±0.42 ^c	26.20± 2.89 ^a	27.57± 1.16 ^{bc}	84.32± 4.49 ^c	17.47± 0.99 ^b
<i>H. ulmarius</i>	21.07± 2.02 ^{bc}	1.70± 0.28 ^c	18.47± 0.67 ^d	31.27± 2.05 ^a	86.85± 2.60 ^{bc}	10.62± 1.48 ^d
<i>P. sajor caju</i>	26.02± 1.269 ^a	2.50± 0.29 ^b	23.77± 0.78 ^b	25.55± 1.04 ^{cd}	88.22± 0.27 ^{ab}	19.40± 1.48 ^a
<i>P. citrinopileatus</i>	23.07± 2.029 ^b	3.60± 0.31 ^a	21.15± 0.26 ^c	24.27± 1.51 ^d	85.95± 0.40 ^{bc}	15.92± 0.71 ^b
CD (0.05)	2.524	0.466	2.216	2.116	3.581	1.674
SE (m)	0.837	0.155	0.735	0.702	1.188	0.555

Values are mean ± SD of four replications

Values followed by similar superscripts are not significantly different at 5 % level

4.4.4 Estimation of carbohydrate

Carbohydrate content was found to be significantly higher in *H. ulmarius* (31.27%) followed by *P. florida* (29.17 %) and *P. djamor* (27.57 %) which were significantly different from each other. Carbohydrate content was minimum in *P. citrinopileatus* (24.27 %) followed by *P. sajor-caju* (25.55 %).

4.4.5 Estimation of moisture content

Moisture content of *Pleurotus* spp. was determined based on fresh weight. *P. florida* recorded the maximum moisture content (90.97 %). It was followed by *P. sajor-caju* (88.22%) and *H. ulmarius* (86.85 %), which were significantly different from each other. *P. citrinopileatus* recorded moisture content of 85.95 per cent. Minimum moisture content was observed in *P. djamor* (84.32 %) which differed significantly from other species.

4.4.6 Estimation of amino acid

Amino acid content was maximum in *P. sajor-caju* (19.40 %) which was significantly higher than other species. *P. djamor* (17.47 %) and *P. citrinopileatus* (15.92 %) were on par with each other. Amino acid content was less in *H. ulmarius* (10.62 %) followed by *P. florida* (13.57 %).

4.4.7 Estimation of Minerals

Minerals present in the five species of oyster mushroom were evaluated. The results on sodium, phosphorous, potassium, calcium and magnesium contents are presented in Table (16)

4.4.7.1 Estimation of sodium

Sodium content in oyster mushrooms was relatively less in quantity compared to other minerals. Among the five species of oyster mushrooms, sodium content was found significantly more in *P. djamor* (965 ppm), followed by *P. citrinopileatus* (605

Table 16. Mineral constituents in five species of oyster mushroom

Mushroom	Sodium (ppm)	Phosphorous (ppm)	Potassium (ppm)	Calcium (ppm)	Magnesium (ppm)
<i>P. florida</i>	290.00± 28.55 ^d	86.75± 2.36 ^{bc}	3725.00± 125.83 ^e	1000.00± 70.71 ^a	4025.00± 50.00 ^a
<i>P. djamor</i>	965.00± 23.80 ^a	82.75± 2.06 ^c	7175.00± 150 ^a	795.00± 10.00 ^{bc}	577.50± 26.30 ^d
<i>H. ulmarius</i>	255.00± 17.32 ^d	90.25± 3.40 ^b	7000.00± 81.65 ^b	810.00± 11.54 ^b	530.00± 35.59 ^d
<i>P. sajor caju</i>	470.00± 24.49 ^c	90.50± 3.41 ^b	6550.00± 57.73 ^c	550.00± 00 ^d	1200.00± 00 ^c
<i>P. citrinopileatus</i>	605.00± 23.80 ^b	96.50± 3.41 ^a	4575.00± 50.00 ^d	752.50± 20.61 ^c	2160.00± 58.87 ^b
CD (0.05)	35.983	4.509	151.967	50.701	60.003
SE (m)	11.937	1.496	50.415	16.820	19.906

Values are mean ± SD of four replications

Values followed by similar superscripts are not significantly different at 5 % level

ppm) whereas the content in other mushrooms such as *P. florida* (290 ppm) and *H. ulmarius* (255 ppm) were significantly less and were statistically on par.

4.4.7.4 Estimation of calcium

The calcium content recorded was significantly higher in *P. florida* (1000 ppm) followed by *H. ulmarius* (810 ppm). Significantly lower calcium content (550 ppm) was recorded from *P. sajor-caju*.

4.4.7.5 Estimation of magnesium

Magnesium content was significantly higher in *P. florida* (4025 ppm) followed by *P. citrinopileatus* (2160 ppm). Significantly lower magnesium content was recorded from *P. djamor* (577.50 ppm) and *H. ulmarius* (530.00 ppm) which were statistically on par.

4.5 ANALYSIS OF MEDICINAL COMPONENTS

Medicinal components in the five species of oyster mushroom were evaluated. The results on β -Glucan, glycoprotein, terpenoid, polyphenol, β -carotene and lovastatin contents are presented in Table (17)

4.5.1 Estimation of β -glucan

β -Glucan content was significantly higher in *P. djamor* (30.25 g per 100g) followed by *H. ulmarius* (28.00 g/100g) which vary significantly. *P. citrinopileatus* (16.75 g per 100g) recorded the least β -Glucan content followed by *P. florida* (20.22 per 100g).

4.5.2 Estimation of glycoprotein

Glycoprotein content recorded was significantly higher in *P. sajor-caju* (435 μ g/100g), followed by *P. citrinopileatus* (337.50 μ g per 100g). Glycoprotein content was significantly less in *P. djamor* (117.50 μ g per 100g).

Table 17. Medicinal components in five species of oyster mushroom

Mushroom	β -Glucan (g/100g)	Glycoprotein (μ g/100g)	Terpenoid (per cent)	Polyphenol (mg/100g)	β -carotene (μ g/100g)	Lovastatin (μ g/100g)
<i>P. florida</i>	20.22 \pm 0.82 ^d	210.00 \pm 14.14 ^d	0.94 \pm 0.08 ^c	1.86 \pm 0.06 ^c	1.78 \pm 0.13 ^c	129.00 \pm 7.25 ^d
<i>P. djamor</i>	30.25 \pm 1.09 ^a	117.50 \pm 22.17 ^e	0.99 \pm 0.02 ^c	1.50 \pm 0.08 ^d	3.70 \pm 0.16 ^a	370.75 \pm 36.61 ^a
<i>H. ulmarius</i>	28.00 \pm 0.46 ^b	277.50 \pm 20.61 ^c	1.01 \pm 0.06 ^c	2.05 \pm 0.15 ^c	2.12 \pm 0.28 ^c	197.50 \pm 11.21 ^c
<i>P. sajor caju</i>	25.72 \pm 0.84 ^c	435.00 \pm 23.80 ^a	1.26 \pm 0.04 ^a	3.5 \pm 0.34 ^b	2.60 \pm 0.35 ^b	257.25 \pm 29.71 ^b
<i>P. citrinopileatus</i>	16.75 \pm 0.66 ^e	337.50 \pm 15.00 ^b	1.13 \pm 0.06 ^b	4.17 \pm 0.34 ^a	0.85 \pm 0.34 ^d	85 \pm 34.15 ^e
CD (0.05)	1.214	29.44	0.089	0.349	0.411	40.266
SE (m)	0.403	9.768	0.030	0.116	0.136	13.35

Values are mean \pm SD of four replications

Values followed by similar superscripts are not significantly different at 5 % level

4.5.3 Estimation of terpenoid

Terpenoid content was significantly higher in *P. sajor-caju* (1.26 %) followed by *P. citrinopileatus* (1.13 %) which were significantly different from each other. Terpenoid content was significantly less in *P. florida* (0.94 %), *P. djamor* (0.99 %) and *H. ulmarius* (1.01 %) which were on par with each other.

4.5.4 Estimation of polyphenol

P. citrinopileatus (4.17 mg per 100g) recorded higher polyphenol content which differed significantly from other species of oyster mushrooms. *P. florida* (1.86 mg per 100g) and *H. ulmarius* (2.05 mg per 100g) did not vary significantly. *P. djamor* (1.50 mg per 100g) recorded the least polyphenol content among the five species of oyster mushrooms.

4.5.5 Estimation of β -carotene

P. djamor (3.70 μg per 100g) recorded significantly higher β -carotene content compared to other species of oyster mushrooms which was followed by *P. sajor-caju* (2.60 μg per 100g). *P. florida* (1.78 μg per 100g) and *H. ulmarius* (2.12 μg per 100g) were on par with each other. β -carotene content was significantly less in *P. citrinopileatus* (0.85 μg per 100g).

4.5.6 Estimation of lovastatin

Lovastatin content was estimated to be higher in *P. djamor* (370.75 μg per 100g) which was significantly different, which was followed by *P. sajor-caju* (257.25 μg per 100g). The least content of lovastatin was recorded from *P. citrinopileatus* (85.00 μg per 100g) followed by *P. florida* (129.00 μg per 100g).

4.6 SENSORY EVALUATION OF FIVE SPECIES OF OYSTER MUSHROOMS

Sensory evaluation of five species of oyster mushrooms was done using sauteed mushroom recipe to evaluate the cooking quality for quality attributes like appearance, colour, texture, flavour, taste, cooking quality and overall acceptability

using nine-point score card (Plate 21). The sensory scores of sauteed mushroom developed from five species of oyster mushrooms are given in Table (18) The mean score of *H. ulmarius* for appearance parameter, was 8.83 on a maximum score of 9 which was significantly higher than other species. It was followed by *P. florida* (8.00), *P. sajor-caju* (7.58) and *P. djamor* (7.66) which was on par with each other. In case of colour also, the mean score of *H. ulmarius* (8.75) was much preferred than the other samples evaluated, which was followed by *P. djamor* (8.33), *P. florida* (8.16) and *P. citrinopileatus* (8.08) which did not vary significantly. The mean score of the texture attribute of saute of *H. ulmarius* (8.66) was the highest followed by *P. florida* (7.91) and *P. sajor-caju* (7.50). The flavour attribute was significantly highest for *H. ulmarius* (8.58) on a maximum score of 9. This was followed by *P. djamor* (7.75). *H. ulmarius* scored 8.83 for the taste attribute, followed by *P. florida* and *P. djamor* both of which scored 7.41.

It was observed that among the five species of oyster mushrooms evaluated, *H. ulmarius* exceeded in all the sensory parameters.

4.7 EVALUATION OF SHELF LIFE OF FIVE SPECIES OF OYSTER MUSHROOM UNDER ROOM CONDITION AND REFRIGERATED CONDITION

Shelf life of five species of oyster mushrooms were undertaken both in room temperature ($26\pm 2^{\circ}\text{C}$) and in refrigerated conditions (15°C). Number of days for change in colour, texture and physical appearance were recorded based on visual observation. Observations were taken on 24 h intervals.

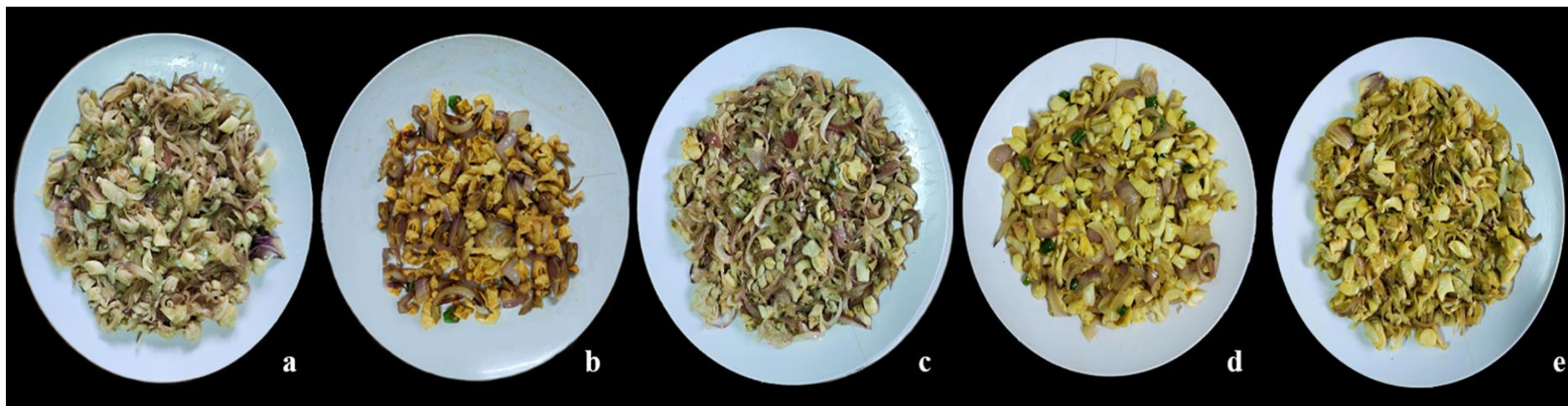


Plate 21. Saute developed from Oyster mushrooms
a. *P. florida* b. *P. djamor* c. *H. ulmarius* d. *P. sajor-caju* e. *P. citrinopileatus*

Table 18. Sensory scores (mean values) of saute developed from five species of oyster mushrooms - Kruskal value test statistics

Sample	Appearance		Colour		Texture		Flavour		Taste	
	Mean Rank	Mean Score	Mean Rank	Mean Score	Mean Rank	Mean Score	Mean Rank	Mean Score	Mean Rank	Mean Score
<i>P. florida</i>	35.54	8.00 ^b	29.41	8.16 ^{ab}	35.08	7.91 ^{ab}	35.29	7.75 ^{ab}	30.50	7.41 ^b
<i>P. djamor</i>	29.50	7.66 ^b	32.50	8.33 ^{ab}	26.58	7.41 ^{bc}	31.33	7.50 ^b	32.33	7.41 ^b
<i>H. ulmarius</i>	50.91	8.83 ^a	43.12	8.75 ^a	48.33	8.66 ^a	48.04	8.58 ^a	52.00	8.83 ^a
<i>P. sajor-caju</i>	28.75	7.58 ^b	20.16	7.66 ^b	30.41	7.50 ^b	30.33	7.50 ^b	29.16	7.33 ^b
<i>P. citrinopileatus</i>	7.79	5.66 ^c	27.29	8.08 ^{ab}	12.08	6.00 ^c	7.50	4.83 ^c	8.50	4.58 ^c
K value	41.33		13.26		29.53		35.95		39.40	

Freshly harvested bunch of *P. florida* was pure white in colour and remained fresh for one day after harvest in room condition and on the second day the colour began to fade and the bunch started to shrink and decay (Plate 22). *P. florida* had lost the marketability on day 2. On the third day, the bunch kept in room temperature began to emit slightly foul smell, the colour changed to yellow and rotted appearance was observed. On the fourth day, colour turned to yellowish brown, and it became watery with foul smell (Table 19). The bunch kept under refrigerated condition remained fresh for two days after harvest (Plate 23). The colour turned to pale yellow and starts to crinkle three days after harvesting and lost its marketability. It became pale yellow in colour and became watery after 5 days (Table 20).

Freshly harvested bunch of *P. djamor* was pink in colour and remained fresh for two days after harvest in room condition and three days after harvest, the colour changed to pale pink and slight crinkling and rotting was observed in the bunch (Plate 24). Four days after harvest, the colour of bunch turned to yellowish pink, and it became watery with foul smell (Table 21). The bunch kept under refrigerated condition (15⁰C) remained fresh for three days after which the colour began to fade (Plate 25). Four days after harvest, the colour turned to pale white and became unfit for consumption. After five days, the colour turned to slight yellow with a foul smell (Table 22).

Freshly harvested bunch of *H. ulmarius* was white in colour and remained fresh for only one day after harvest in room condition and on the second day, colour changed to pale white with slight smell and crinkled appearance (Plate 26). It had lost its marketability due to loss of weight and shrivelling. Three days after harvesting, the bunch kept in room temperature began to emit slightly foul smell, the colour changed to slight yellow and the bunch became rotten. On the fourth day, colour turned to yellowish brown, and it became watery with foul smell (Table 23). The bunch kept under refrigerated condition remained fresh for two days after harvest (Plate 27). It became pale yellow in colour and became watery after five days and lost its marketability (Table 24).

Table 19. Keeping quality of *P. florida* in room condition

Sl. No	Time after harvest	Observation on room condition
1	Fresh mushroom (0 day after harvest)	Pure white colour, Fresh appearance
2	1 day after harvest	Pale white colour, Fresh appearance
3	2 days after harvest	Pale white colour, starts to crinkle and rot
4	3 days after harvest	Pale yellowish colour, start to emit slightly foul smell
5	4 days after harvest	Colour turns to yellow, emits foul smell, rotted and watery appearance

Table 20. Keeping quality of *P. florida* in refrigerated condition

Sl. No	Time after harvest	Observation on refrigerated condition
1	Fresh mushroom (0 day after harvest)	Pure white colour, Fresh appearance
2	2 days after harvest	Pure white colour, Fresh appearance
3	3 days after harvest	Pale white colour, starts to crinkle
4	4 days after harvest	Pale yellowish colour, slightly foul smell, shredded
5	5 days after harvest	Colour turns to yellow, foul smell, watery appearance

Table 21. Keeping quality of *P. djamor* in room condition

Sl. No	Time after harvest	Observation on room condition
1	Fresh mushroom (0 day after harvest)	Pink colour, Fresh appearance
2	1 day after harvest	Pink colour, Fresh appearance
3	2 days after harvest	Pink colour starts to fade, fresh appearance
4	3 days after harvest	Pale pink colour, slight smell, bunch begin to crinkle and rot
5	4 days after harvest	Colour turns to yellowish pink, foul smell, rotted appearance

Table 22. Keeping quality of *P. djamor* in refrigerated condition

Sl. No	Time after harvest	Observation on refrigerated condition
1	Fresh mushroom (0 day after harvest)	Pink colour, Fresh appearance
2	2 days after harvest	Pink colour, Fresh appearance
3	3 days after harvest	Pink colour, Fresh appearance
4	4 days after harvest	Pale white colour, no change in appearance of bunch
5	5 days after harvest	Colour turns to slight yellow, bunch starts to crinkle with slight smell



Plate 22. Shelf life of *P. florida* under room condition; a. Fresh mushroom (0 day after harvest); b. 1 day after harvest; c. 2 days after harvest; d. 3 days after harvest; e. 4 days after harvest

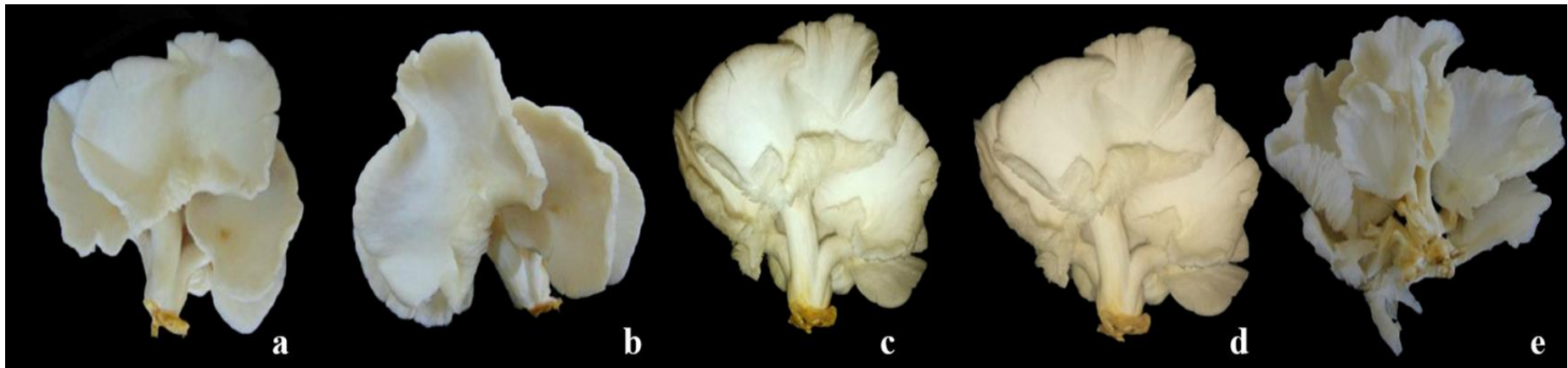


Plate 23. Shelf life of *P. florida* under refrigerated condition; a. Fresh mushroom (0 day after harvest); b. 2 days after harvest; c. 3 days after harvest; d. 4 days after harvest; e. 5 days after harvest

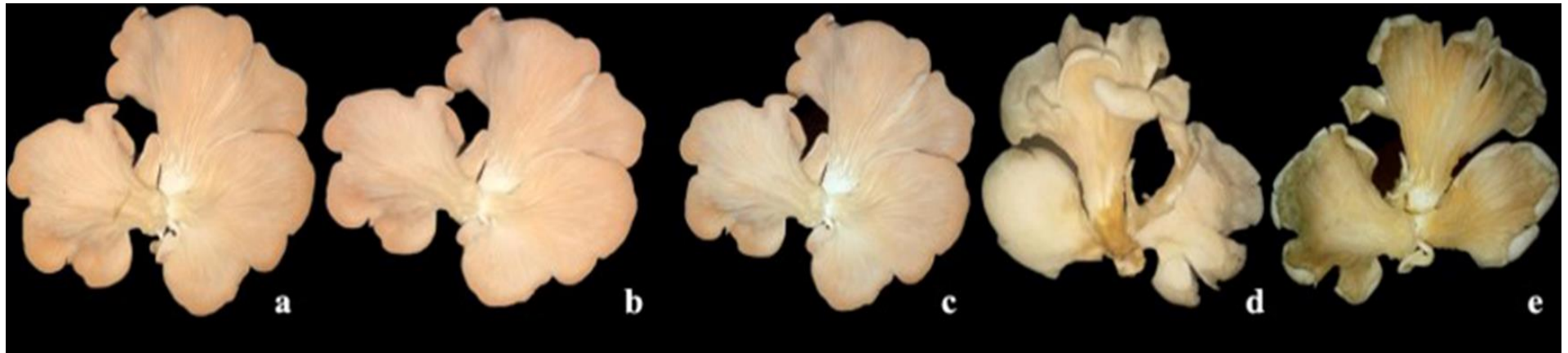


Plate 24. Shelf life of *P. djamor* under room condition; a. Fresh mushroom (0 day after harvest) b. 1 day after harvest c. 2 days after harvest d. 3 days after harvest e. 4 days after harvest



Plate 25. Shelf life of *P. djamor* under refrigerated condition; a. Fresh mushroom (0 day after harvest) b. 2 days after harvest c. 3 days after harvest d. 4 days after harvest e. 5 days after harvest



Plate 26. Shelf life of *H. ulmarius* under room condition; a. Fresh mushroom (0 day after harvest) b. 1 day after harvest
c. 2 days after harvest d. 3 days after harvest e. 4 days after harvest

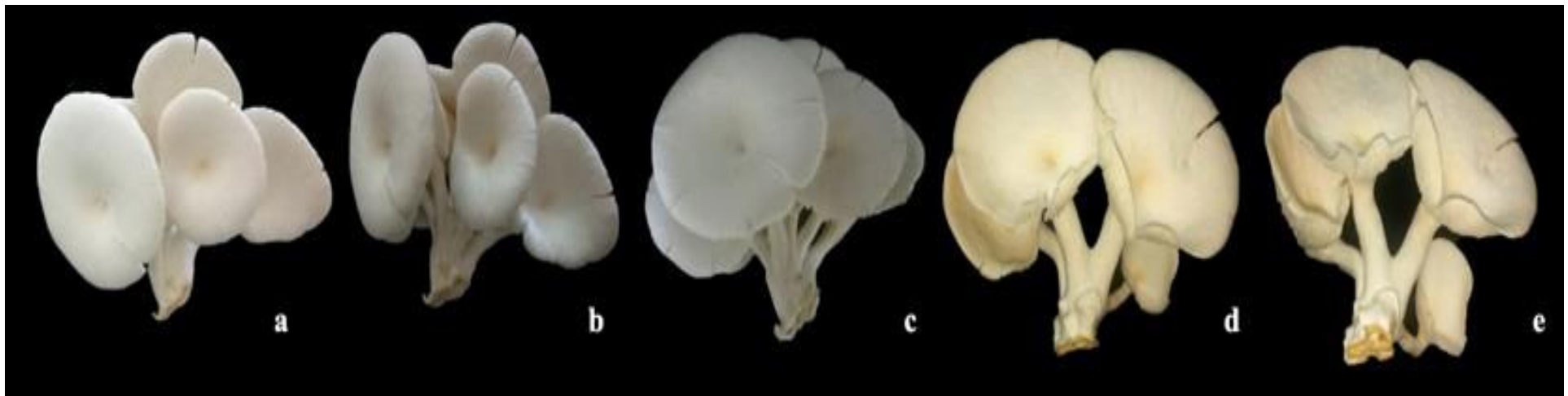


Plate 27. Shelf life of *H. ulmarius* under refrigerated condition; a. Fresh mushroom (0 day after harvest) b. 2 days after
harvest c. 3 days after harvest d. 4 days after harvest e. 5 days after harvest

Table 23. Keeping quality of *H. ulmarius* in room condition

Sl. No	Time after harvest	Observation on room condition
1	Fresh mushroom (0 day after harvest)	Pure white colour, Fresh appearance
2	1 day after harvest	White colour, bunch starts shredding
3	2 days after harvest	Colour fades to pale white, slight smell and crinkled appearance
4	3 days after harvest	Slightly yellow colour, foul smell, rotted appearance
5	4 days after harvest	Colour turns to yellow-brown, foul smell, watery appearance

Table 24. Keeping quality of *H. ulmarius* in refrigerated condition

Sl. No	Time after harvest	Observation on refrigerated condition
1	Fresh mushroom (0 day after harvest)	Pure white colour, Fresh appearance
2	2 days after harvest	White colour, Fresh appearance
3	3 days after harvest	White colour starts to fade, no change in appearance
4	4 days after harvest	Pale white colour, bunch starts to crinkle and emits slightly foul smell
5	5 days after harvest	Colour turns to slight yellow with foul smell and rotted appearance

Freshly harvested bunch of *P. sajor-caju* was greyish white in colour and remained fresh for two days after harvest in room condition and three days after harvest, the bunch became unfit for consumption (Plate 28). Three days after harvest, colour of the bunch turned to slight yellow with rotted appearance. Four days after harvest, the bunch began to emit smell, the colour changed to yellow and loss of weight and watery appearance was observed (Table 25). The bunch kept under refrigerated condition remained fresh for four days (Plate 29). It became pale cream in colour and became watery after seven days (Table 26).

Freshly harvested bunch of *P. citrinopileatus* was golden yellow in colour and remained fresh for one day after harvest in room condition and on the second day after harvest, the colour began to fade and slight smell started to emerge and lost the marketability (Plate 30). On the third day after harvest, the bunch kept in room temperature began to emit foul smell, the colour changed to yellowish white and the texture changed to watery. On the fourth day, the colour turned to yellowish brown, and it became watery with foul smell (Table 27). The bunch kept under refrigerated condition remained fresh for two days after which the colour began to fade and the bunch started to crinkle (Plate 31). The bunch became pale white in colour and emitted foul smell after four days of harvesting. After five days, the bunch became pale cream and watery (Table 28).

Table 25. Keeping quality of *P. sajor-caju* in room condition

Sl. No	Time after harvest	Observation on room condition
1	Fresh mushroom (0 day after harvest)	Greyish white colour, Fresh appearance
2	1 day after harvest	Greyish white colour, Fresh appearance
3	2 days after harvest	Greyish white, the bunch start to crinkle
4	3 days after harvest	Slightly yellow colour, foul smell, rotted appearance
5	4 days after harvest	Colour turns to yellow-brown, foul smell, watery appearance

Table 26. Keeping quality of *P. sajor-caju* in refrigerated condition

Sl. No	Time after harvest	Observation on refrigerated condition
1	Fresh mushroom (0 day after harvest)	Greyish white colour, Fresh appearance
2	2 days after harvest	Greyish white colour, Fresh appearance
3	3 days after harvest	Greyish white colour, Fresh appearance
4	4 days after harvest	Pale white colour, no smell, slight crinkling of the bunch
5	5 days after harvest	Pale white colour, no smell, slight crinkling of the bunch

Table 27. Keeping quality of *P. citrinopileatus* in room condition

Sl. No	Time after harvest	Observation on room condition
1	Fresh mushroom (0 day after harvest)	Golden yellow colour, Fresh appearance
2	1 day after harvest	Pale yellow colour, No change in smell
3	2 days after harvest	Colour fades to pale white, slight smell and the bunch starts to lose water
4	3 days after harvest	Pale white colour, foul smell, rotted appearance
5	4 days after harvest	Colour turns to yellowish white, foul smell, watery appearance

Table 28. Keeping quality of *P. citrinopileatus* in refrigerated condition

Sl. No	Time after harvest	Observation on refrigerated condition
1	Fresh mushroom (0 day after harvest)	Golden yellow colour, Fresh appearance
2	2 days after harvest	Light yellow colour, Fresh appearance
3	3 days after harvest	Yellow colour starts to fade, bunch starts to crinkle
4	4 days after harvest	Pale white colour, emits foul smell
5	5 days after harvest	Pale yellow colour, foul smell, and watery appearance



Plate 28. Shelf life of *P. sajor-caju* under room condition; a. Fresh mushroom (0 day after harvest) b. 1 day after harvest
c. 2 days after harvest d. 3 days after harvest; e. 4 days after harvest



Plate 29. Shelf life of *P. sajor-caju* under refrigerated condition; a. Fresh mushroom (0 day after harvest) b. 2 days after
harvest c. 3 days after harvest d. 4 days after harvest e. 5 days after harvest



Plate 30. Shelf life of *P. citrinopileatus* under room condition ; a. Fresh mushroom (0 day after harvest) b. 1 day after harvest
c. 2 days after harvest d. 3 days after harvest e. 4 days after harvest



Plate 31. Shelf life of *P. citrinopileatus* under refrigerated condition; a. Fresh mushroom (0 day after harvest) b. 2 days after
harvest c. 3 days after harvest d. 4 days after harvest e. 5 days after harvest

5. DISCUSSION

Oyster mushroom is commonly called as 'Dhingri' in India and stands second among the total mushroom production in the world. Mushrooms have a long history of use among humans both as a food and medicine. The demand and production of mushroom is increasing day by day due to the awareness about its nutraceutical and therapeutic properties. The present research project is aimed to evaluate the suitability of five species of oyster mushrooms viz, *P. florida*, *P. djamor*, *H. ulmarius*, *P. sajor-caju* and *P. citrinopileatus* under five agro-ecological zones of Kerala. The proximate and medicinal constituents present in the five species of oyster mushrooms are evaluated. Sensory evaluation and shelf-life studies were also conducted.

5.1 Isolation and Pure culturing

Healthy mushrooms collected from pest and disease-free high yielding beds were taken for tissue culturing. A small portion of tissue from the junction of pileus and stipe was detached aseptically and placed in the Petri plates containing solidified Potato Dextrose Peptone Agar (PDPA) medium and incubated at room temperature. After 72 h, the mycelial growth observed was purified by hyphal tip method in which the growing mycelial tip is transferred to petri plates containing PDPA medium and pure cultures of the five species of oyster mushrooms were maintained.

Krishnapriya (2018) isolated three isolates of *Pleurotus* spp. viz. *P. cystidiosus coremial*, *P. cystidiosus non-coremial* and *P. opuntia* in different types of media such as PDA, MEA, PDPA, OMA and CEA and concluded that PDPA was the best medium.

5.1.1 Macroscopic observations

The sporocarps of five species of oyster mushrooms were studied for their morphological characters. Observations on the sporocarp colour, sporocarp weight, stipe length, colour of pinhead, texture of sporocarp, pileus length, pileus breadth, gill arrangement and number of gills per cm were recorded.

In the present study, *P. florida* produced white, delicate and fleshy sporocarp with entire, enrolled type pileus margin with leathery texture. The stipe was long and stout (3.22 cm). Average weight of sporocarp was 11.71g and comparatively large pileus with 6.62 cm x 7.52 cm were produced. Krishnapriya *et al.* (2017) reported that the pileus dimensions of *P. florida* were 6.71 cm x 7.90 cm and similar finding was recorded by Jose (2018) who reported that the cap of *P. florida* had a spatulate shape and the size being 4.5-7.5 cm x 2.5-3 cm. The stipe was attached laterally and stipe length was between 3 cm to 4.5 cm. The sporocarp was produced in bunches and attained harvesting maturity in three days from pinhead emergence. Stipe was significantly longer (4.80 cm) compared to *P. djamor* (0.91 cm). Jyothi (2019) also reported that *P. florida* produced larger pileus with dimension of 6.46 cm x 7.58 cm. Long and stout stipe (3.33 cm) was recorded and the average weight of individual sporocarp was reported to be 11.47 g.

P. djamor produced pinkish white leathery sporocarps with smaller pileus. Average pileus dimensions of 4.90 cm x 6.50 cm were recorded. The sporocarp was usually devoid of stipe and if present, was very short with an average length of 0.80 cm. The average sporocarp weight was 8.75g and the number of gills from the margin was 17.50 gills/cm which were significantly higher than other species studied. The pinkish pinheads attained harvesting maturity within three days. Shukla and Jaitly (2011); Krishnapriya *et al.* (2017) reported that the pileus dimensions of *P. djamor* were 4.80 cm x 6.57 cm which is smaller than that of *P. florida*. Jyothi (2019) reported that the pileus dimension of *P. djamor* were 4.80 cm x 6.57 cm. The average length of stipe is 0.92 cm and the average weight of individual sporocarp was 6.03 g.

H. ulmarius produced creamy white fleshy sporocarps with significantly large pileus. The average pileus size was 7.0 cm x 7.52 cm. The average length of stipe was 3.35 cm which was comparatively higher. The average sporocarp weight was 9.33 g. Gills were attached to the stipe in a non- decurrent fashion and the average number of gills from the margin was 13.75 gills/cm. The greyish white pinheads attained harvesting maturity within three to four days from pinhead emergence. Sumi (2016) reported that the pileus dimension of *H. ulmarius* was 5-15 cm and the stipe was 1.5-

10 cm long, thick and cylindrical. Gills were attached to the stem but not running down, non decurrent. The results are in concurrence with the report of Meyers (2004).

P. sajor-caju produced greyish white leathery sporocarp. The average length and breadth of pileus was 7.10 cm and 5.32 cm respectively. Significantly long and stout stipe with an average length of 5.32 cm was one of the characteristic features of *P. sajor-caju*. The pinheads attained harvesting maturity within four days. Vooticumpee (1996) reported that the pileus diameter and stipe length of *P. sajor-caju* was 5.95 cm -7.38 cm and 1.30 cm -3.95 cm respectively. Dhar (2011) also reported that the fruiting bodies of *P. sajor-caju* was comparatively heavy when fully grown.

P. citrinopileatus produced golden yellow delicate sporocarp with comparatively short stipe. The average size of pileus was 5.45 cm x 6.17 cm and gills were attached decurrently to the stipe. The average stipe length recorded was 3.30 cm and the average sporocarp weight was 5.05 g. The average number of gills from the margin was 11.75 gills/cm. The bright yellow pinheads attained harvesting maturity within three to four days from pinhead emergence. (Thakur, 2019) reported that the pileus diameter of *P. citrinopileatus* ranged from 3-9 cm and stipe length varied from 2-4 cm.

5.1.1 Microscopic observations

Basidiospores of *P. florida* and *P. djamor* recorded 7-12 μm in length and 2-5 μm in width whereas, *H. ulmarius* and *P. sajor-caju* produced spores with dimension of 8-12 μm x 3-6 μm . *P. citrinopileatus* produced spores with dimension of 7-10 μm x 2-3 μm . Hyphae of all the five species were septate, branched and hyaline with clamp connections. The width of the hyphae did not vary significantly among each other and it ranged from 1.5-4.5 μm .

In confirmation with the above observations, Biswas *et al.* (2011) observed that the basidiospores of *P. florida* were oblong with 7-10 μm long while Das *et al.* (2015) reported that the basidiospores of *P. florida* had an area of 15.68 sq. μm . Lechner *et al.* (2004) observed that the spore dimensions of *P. djamor* were 6.0-7.80 μm x 2.60-3.12 μm . Junior *et al.* (2010) also studied about the microscopic characters of *P. djamor* and

reported that the basidiospores were cylindrical, thin-walled, hyaline, smooth and inamyloid. The spore dimensions were $8.7\text{--}11.2\ \mu\text{m} \times 3.7\text{--}5.0\ \mu\text{m}$. Kushwaha *et al.* (2011) studied about the microscopic characters of *H. ulmarius* and reported that the hyphae were septate, branched, subhyaline to creamy buff in colour. Hyphal width was measured to be $1\text{--}4\ \mu\text{m}$. Basidiospores were small in size, measuring $2.5\text{--}6.5\ \mu\text{m}$, broad-ellipsoid to ovoid, hyaline and smooth. The microscopic characters of *P. citrinopileatus* was studied by Thakur (2019). The hyphae of *P. citrinopileatus* was $1.5\text{--}3\ \mu\text{m}$ width, monomitic and clamp connections were present. Basidiospores were thin-walled, hyaline, smooth and cylindrical to sub-cylindrical in shape with an average size of $2.30\text{--}3.94\ \mu\text{m} \times 0.77\text{--}1.54\ \mu\text{m}$.

5.2 EVALUATION OF PADDY STRAW/RUBBER SAWDUST FOR MUSHROOM PRODUCTION

Two substrates namely paddy straw and rubber sawdust were evaluated for the cultivation of five species of oyster mushrooms. In the present study, highest yield was obtained from *H. ulmarius* cultivated on both paddy straw and rubber sawdust ($1233\ \text{g kg}^{-1}$ and $1611\ \text{g kg}^{-1}$ respectively) which was followed by *P. florida* ($1148.30\ \text{g kg}^{-1}$ and $1582.66\ \text{g kg}^{-1}$ respectively). Crop period was comparatively less for *P. djamor* cultivated on both the substrates (54.00 days and 90.33 days). Significantly higher crop period was recorded for *P. sajor-caju* cultivated on both the substrates (67.33 and 111.66 days respectively).

Paddy straw is a rich source of cellulose and it is composed of 37 per cent cellulose, 24 per cent hemicellulose and 14 per cent lignin abundant biomass. The yield of mushroom is proportional to the amount of cellulose (Nguyen, 2004). The superiority of paddy straw over the other substrates was also reported earlier by Pal and Thapa (1979) and Bano *et al.* (1988). This is in agreement with the findings of Sivaprakasam and Kandaswamy (1981) who reported a positive correlation of sporophore yield to the cellulose content and cellulose – lignin ratio. Desai (1982) and Bisaria *et al.* (1987) also reported the superiority of paddy straw over other substrates for the cultivation of *P. sajor-caju*.

The present study revealed that both the substrates *viz.*, paddy straw and rubber sawdust were suitable for the cultivation of oyster mushrooms. It has been evident that the selection of substrate significantly affected the time taken for complete spawn run and total yield. Comparatively higher yield was obtained from rubber sawdust but the total number of days needed for cultivation was longer. The superiority of paddy straw over rubber sawdust may be due to the higher content of cellulose and hemicellulose which can be easily degraded by the oyster mushrooms while that from rubber sawdust is used up comparatively slowly by the mushroom and hence the crop period will be higher. So, both the substrates can be utilized for cultivation based on the ease of availability and cost effectiveness.

5.3 EVALUATION OF PRODUCTION OF MUSHROOMS UNDER DIFFERENT AGRO-ECOLOGICAL CONDITION

Oyster mushroom has the maximum number of commercially cultivated species for year-round cultivation. Kerala having a tropical climate is suitable for the cultivation of oyster mushroom. Sumi (2016) conducted multilocational trials in Idukki, Wayanad and Vellayani and revealed that *H. ulmarius* can be cultivated throughout the year in all the three regions except April-May in cool climate of Idukki and Wayanad. An average yield of 1.096 kg kg⁻¹ dry weight of paddy straw (109.60 % BE) was reported for *H. ulmarius* and 0.976 kg with 97.5 (%) BE for *P. florida*.

The coastal plains are characterised by tropical monsoon climate having rainfall ranging from 3133 to 3254 mm and mean annual temperature of 27.5⁰C. The weather data from RARS Pilicode revealed an average temperature of 27.57⁰C and a relative humidity of 93.87. In coastal plains, the highest yield of 1029.96 g was recorded from *H. ulmarius* which was followed by *P. florida* (886.20 g) and *P. djamor* (723.00 g). Significantly lower yield of 550.10 g was obtained from *P. sajor-caju* and *P. citrinopileatus* (630.00 g). The total yield from first harvest was significantly higher from *H. ulmarius* (520.00 g) followed by *P. florida* (432.20 g). Significantly low yield was obtained from *P. sajor-caju* (50.00 g) and *P. citrinopileatus* (62.00 g) from third harvest which indicates the possibility of limiting the economic cropping period of

these mushroom upto second harvest. *H. ulmarius* was found to be the best suited oyster mushroom species for cultivation in coastal plains.

Midland laterites are characterised by tropical moist subhumid monsoon climate with mean annual temperature of 27.1⁰C and rainfall 1884 mm. The weather data from Trivandrum city revealed an average temperature of 27.57⁰C and an average relative humidity of 85.81. Total yield from first harvest was significantly higher in *P. djamor* (425.00 g) followed by *H. ulmarius* (389.00 g). It has been observed that the total yield from third harvest was significantly low in *P. citrinopileatus* (75.00 g) followed by *P. sajor-caju* (90.03 g). In midland laterites, the highest yield of 827 g was obtained from *P. djamor* which was on par with *H. ulmarius* and *P. florida* (806.00 g and 773.03 g respectively).

The climate of foothills is tropical humid monsoon type with mean annual temperature of 27.5⁰C and rainfall 3462 mm. The weather data from Trivandrum Airport revealed an average temperature of 27.51⁰C and an average relative humidity of 87.8. In foothills, the highest yield of 927.33 g was recorded from *H. ulmarius*, which was followed by *P. florida* (823.33 g). The yield obtained from *P. sajor-caju* (811.00 g) and *P. citrinopileatus* (800.00 g) did not vary significantly. Significantly lower yield was recorded from *P. djamor*. Total yield from first harvest was significantly higher in *H. ulmarius* (455.33 g) followed by *P. florida* (428.33 g). Earliness in primordial initiation was recorded from *P. djamor* (17.00 days) and time taken for first harvest was found to be significantly less in *P. djamor*, which yielded in 20.00 days. *H. ulmarius* was found to be the best suited oyster mushroom species for cultivation in foothills.

In High hills, the climate is tropical monsoon type with mean annual temperature of 21.6⁰C and rainfall of 3602 mm. The weather data from CRS, Pampadumpara revealed an average temperature of 15.23⁰C and an average relative humidity of 94.93 per cent. In high hills, the highest yield of 1233.00 g was obtained from *H. ulmarius*, followed by *P. florida* (1148.30 g) and *P. djamor* (1050.30 g). Significantly lower yield was obtained from *P. citrinopileatus* (610.00 g). Total yield from first harvest was significantly higher in *H. ulmarius* (658.30 g) followed by *P. djamor* (530.30 g). Total yield from third harvest was significantly low for *P.*

citrinopileatus (65.00 g) followed by *P. sajor-caju* (80.00 g). *P. djamor* took the least number of days to complete spawn run in high hills (11.66 days) followed by *P. florida* (15.66 days). First harvest was obtained from *P. djamor* within 17.66 days after bed preparation. *H. ulmarius*, *P. florida* and *P. djamor* were found to be best suitable for cultivation in high hills based on comparatively low total cropping period and high yield.

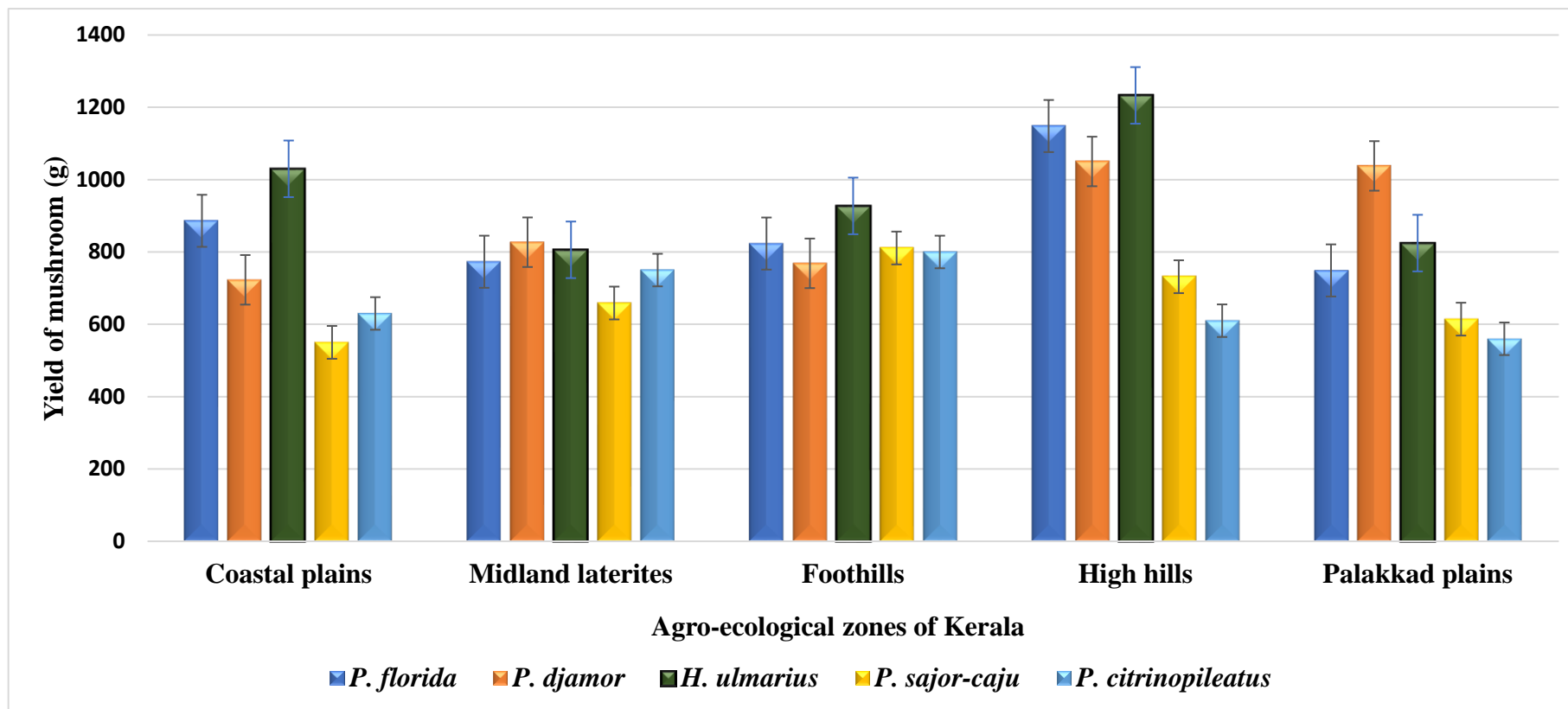
The climate in Palakkad plains is dry subhumid monsoon type with mean annual temperature of 27.6⁰C and rainfall 1340 mm. The weather data from RARS Pattambi revealed an average temperature of 27.69⁰C and an average relative humidity of 84.87 per cent. *P. djamor* produced significantly higher yield of 1038.00 g compared to other species of oyster mushrooms. It was followed by *H. ulmarius* (824.60 g) and *P. florida* (749.00 g). Comparatively lower yield of 560.00 g was obtained from *P. citrinopileatus* in Palakkad plains. Total yield from first, second and third harvest was obtained from *P. djamor* which yielded 502.00 g, 335.00g and 201.00 g respectively. Significantly low yield was recorded from *P. citrinopileatus* from third harvest (28.00 g). *P. djamor* was found to be the best suited oyster mushroom species for cultivation in Palakkad plains.

Shukla and Jaitley (2011) compared the yield of *H. ulmarius* and *P. sajor-caju*. They observed that the maximum average yield of 855.52g per kg dry substrate was recorded by *H. ulmarius* followed by *P. sajor-caju* 742.98 g per kg dry weight substrate. Biswas *et al.* (2013) evaluated the cultural characters and yield of *P. sajor-caju*, *P. flabellatus*, *P. florida*, *P. eous*, *P. ostreatus* and *H. ulmarius*. *H. ulmarius* was found to be most appropriate species in terms of biological efficiency (156 per cent), spawn run period (15 days) and average weight of sporophore (7.98 g), followed by *P. florida* (121.5 % BE), *P. sajor-caju* (115.5 % BE) and *P. ostreatus* (103.25 % BE). From the present study it was evident that *H. ulmarius* was adapted to the climatic conditions prevailing throughout Kerala. High yield of *H. ulmarius* and *P. florida* may be due to the large fruiting clusters and large caps, higher moisture content of pileus and also the adaptability of both mushrooms to different temperature and humidity ranges.

Ahmed *et al.* (2009) reported that the optimum temperature range for spawn running and basidiocarp production of *P. florida* was 21-25⁰C and 20-28⁰C respectively while Dhar *et al.* (2011) observed an optimum temperature range of 20-25⁰C and 20-28⁰C for the spawn running and sporocarp formation of *P. sajor-caju*. Studies indicated that *P. djamor* required higher temperature of 30-35⁰C for spawn running and 21-35⁰C for basidiocarp production (Jatwa *et al.*, 2016). *P. djamor*, although reported to be performing better under warm conditions also yielded well under the different agro-ecological zones of Kerala. It has been evident that *P. djamor* performed well in areas having higher temperature, lesser rainfall and lesser humidity. Both earliness in primordial initiation and comparatively less time taken for first harvest was observed from *P. djamor*, which took an average of 15.00-19.33 days for primordial initiation and 17.66-22.33 days for first harvest respectively.

The performance of *P. sajor-caju* was poor in all the agro-ecological zones. The cropping period was also higher for *P. sajor-caju*. Comparatively low yield of *P. citrinopileatus* and *P. sajor-caju* in Kerala conditions is due to the fact that they prefer warm conditions and are suitable for cultivation in summer months. Compared to other oyster mushroom the average weight of fruiting body was less for *P. citrinopileatus* due to lower moisture content. Since the cultivation of golden oyster mushroom, *P. citrinopileatus* was done for the first time in Kerala its cultivation technology has to be finalised. Also, typically small and numerous caps were produced by *P. citrinopileatus* which also may be responsible for the lower crop yield.

The temperature required for primordial formation of selected *Pleurotus* species was 10-25⁰C (Kong 2004). Results from the current study was also similar to Viziteu (2000), who reported that the temperature required for primordial formation of oyster mushroom was 18-25⁰C. According to Das *et al.* (1987, 1991), variations in season seriously affected the number, weight and crop production period of mushroom. They reported that the production of fruiting bodies of mushroom can be enhanced by favorable temperature and moisture condition. The highest number of effective fruiting body was produced in December to February for selected *Pleurotus* species. Tripathi (2005) reported that for fruiting body development, temperature of 16-22⁰C for *P. ostreatus* and 10-26⁰C for *P. florida* and *P. sajor-caju* were suitable.



Error bar represents standard error of means of observed values

Figure 1. Yield of five species of oyster mushrooms under five agro-ecological zones of Kerala

5.4 ANALYSIS OF PROXIMATE CONSTITUENTS

Mushrooms are good source of nutrients; some have medicinal values and some have both of these properties. Mushroom production has increased rapidly in the recent decades with the realization that they are not only delicious, but also are good sources of nutrients and having medicinal values. Carbohydrates and fats are the source of energy food and proteins are the sources of body building materials. In addition to carbohydrates, fats and proteins, vitamins and minerals are crucial to good health. Determination of nutritional value involves the analysis of composition of nutrients present in the mushroom. In the present study, the proximate constituents such as protein, fat, fibre, carbohydrate, moisture content, amino acid and mineral contents in the five species of oyster mushroom were evaluated.

5.4.1 Estimation of protein

The quality of protein present in oyster mushrooms are reported to be as good as animal proteins. In the present study, the protein content recorded was significantly higher in *P. sajor-caju* (26.02 %). This was followed by *P. citrinopileatus* (23.07 %) and *P. florida* (22.42 %) which were on par with each other.

In confirmation with the above results, Gupta *et al.* (2004) reported that the crude protein content in *P. sajor-caju* ranges from 18.46- 27.78 per cent. Ashraf *et al.* (2013) also observed similar results and reported that the crude protein content in *P. sajor-caju* is 25.24 per cent. Alam *et al.* (2008) reported that the protein content in 100 g of dried *P. sajor-caju* were 23- 26 g and that of *P. florida* were 19- 22 g. Gogavekar *et al.* (2014) calculated the concentration of protein in 100 g dry matter of *P. sajor-caju* to be 29.3 g. Khatun *et al.* (2015) estimated the protein content of three species of *Pleurotus* namely *P. florida*, *P. citrinopileatus* and *P. pulmonarius* and found that protein content was highest in *P. florida* (23.80 per cent) which was followed by *P. citrinopileatus* (20.8 %) and *P. pulmonarius* (16.8 %).

5.4.2 Estimation of fat

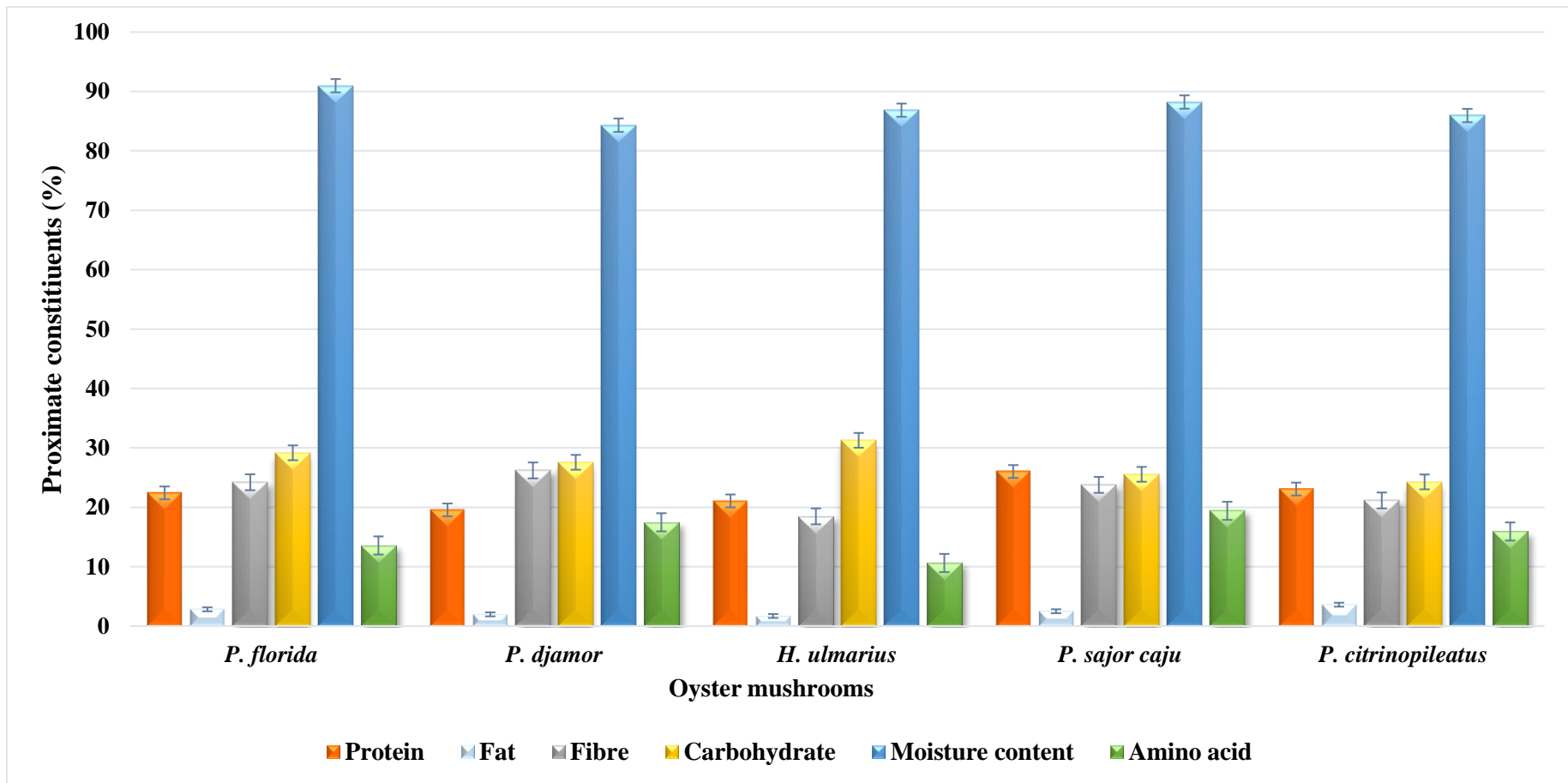
Mushrooms are acclaimed for their low-fat contents. In the present study, minimum fat content was estimated on *H. ulmarius* which is superior to other species studied. Fat content of *P. florida* (2.82 %) and *P. sajor-caju* (2.50 %) did not vary significantly. *P. djamor* recorded fat content of 1.97 per cent and significantly higher fat content was recorded on *P. citrinopileatus* (3.60 %) which differed significantly from other species of oyster mushrooms.

In agreement with the above results, Ashraf *et al.* (2013) reported that the total fat content in *P. djamor* is 3.07 per cent as compared to *P. sajor-caju* (2.47 per cent). According to the nutritional analysis conducted by Alam *et al.* (2008), the lipid content in dry weight of one hundred grams of *P. sajor-caju* and *P. florida* were observed to be 4.2- 4.6 g and 4- 4.6 g respectively. Sumi (2016) reported that the fat content in *H. ulmarius* and *P. florida* were 2.96 and 1.55 per cent respectively on paddy straw substrate. Jose (2018) recorded the fat content of *P. florida*, *P. djamor* and *H. ulmarius* as 2.28 per cent, 2.10 per cent and 1.72 per cent respectively.

5.4.3 Estimation of fibre

The fiber content in oyster mushroom ranges from 7.4 to 27.6 per cent. Fibre is considered as an important ingredient in a balanced and healthy diet. In the present study, the maximum crude fibre content was estimated in *P. djamor* (26.20 %) followed by *P. florida* (24.21 %). *P. sajor-caju* and *P. citrinopileatus* had fibre content of 23.77 and 21.15 per cent respectively. The least crude fibre content was recorded in *H. ulmarius* (18.47 %) which differed significantly from other species of oyster mushrooms.

This is in accordance with the results of Alam *et al.* (2008) who reported that the fibre content in dry weight of 100 g of *P. sajor-caju* and *P. florida* were 22- 23.6 g and 22- 24.6 g respectively. Chirinang *et al.* (2009) reported that the dietary fibre content of *P. sajor-caju* was about 42 per cent dry matter. Ashraf *et al.* (2013) reported that the fiber content of *P. sajor-caju* is 26.28 per cent and *P. djamor* (22.03 per cent).



Error bar represents standard error of means of observed values

Figure 2. Proximate constituents of five species of oyster mushrooms

Jose (2018) reported that the crude fiber content in *P. djamor*, *P. florida* and *H. ulmarius* were 30.59 per cent, 24.38 per cent and 17.06 per cent respectively

5.4.4 Estimation of carbohydrate

Carbohydrate content in the present study was found to be significantly higher in *H. ulmarius* (31.27 %) followed by *P. florida* (29.17 %) and *P. djamor* (27.57 %). Carbohydrate content was minimum in *P. citrinopileatus* (24.27 %) followed by *P. sajor-caju* (25.55 %).

Menaga *et al.* (2012) reported that *P. florida* contains 26.60 per cent carbohydrate on dry weight basis which was in accordance with the current study. Ashraf *et al.* (2013) reported that the carbohydrate content in *P. djamor* is 37.69 per cent followed by *P. sajor-caju* (37.22 per cent). The present results were also in accordance with the findings of Jose (2018) who reported 30.87 per cent carbohydrate from *H. ulmarius* while the carbohydrate content in *P. florida* and *P. djamor* were 26.68 per cent and 26.59 per cent respectively. Jyothi (2019) reported almost similar results in the carbohydrate content of *P. djamor* and *P. florida*. Similar results were given by Sumi (2016) who reported that the carbohydrate content of *H. ulmarius* was 29 per cent.

5.4.5 Estimation of moisture content

Moisture content is one of the important quality parameters which determine the stability and quality of foods. The moisture contents in mushroom are generally very high and hence mushrooms are highly perishable and susceptible to microbial growth. In the present study, *P. florida* recorded the maximum moisture content (90.97 %). It was followed by *P. sajor-caju* (88.22 %) and *H. ulmarius* (86.85 %). *P. citrinopileatus* recorded moisture content of 85.95 per cent. Minimum moisture content was observed in *P. djamor* (84.32 %).

Similar findings were reported by Ghosh *et al.* (1991) that the moisture content of *P. citrinopileatus* was 90.22 per cent whereas the moisture content of *P. sajor-caju* was observed to be 94.04 per cent (Caglarimak 2007). The moisture contents of *P. sajor-caju* and *P. florida* were found to be 87 per cent and 87.5 per cent respectively

according to Alam *et al.* (2008). In accordance with the above reports, Jose (2018) reported that the moisture content of *P. florida*, *P. djamor* and *H. ulmarius* were 84.92 per cent, 88.38 per cent and 89.28 per cent respectively. Jyothi (2019) reported that the moisture content of *P. florida* and *P. djamor* were 92.16 per cent and 85.97 per cent respectively. Menaga *et al.* (2012) recorded moisture content of *P. florida* as 87.30 per cent which were also in agreement with the current results.

5.4.6 Estimation of amino acid

Amino acid content in the present study was maximum in *P. sajor-caju* (19.40 %). *P. djamor* (17.47 %) and *P. citrinopileatus* (15.92 %) were on par with each other. Amino acid content was minimum in *H. ulmarius* (10.62 %) followed by *P. florida* (13.57 %). Chirinang and Intarapichet (2009) recorded 21.10 per cent and 20.12 per cent total amino acid content in dried samples of *P. ostreatus* and *P. sajor-caju* respectively. Similar results were reported by Jose (2018) that the amino acid content in *P. djamor* and *P. florida* were 17.10 per cent and 14.75 per cent respectively. *H. ulmarius* recorded amino acid content of 10.62 per cent.

4.4.7 Estimation of Minerals

Mushroom is an important source of minerals such as sodium, potassium, calcium, magnesium and phosphorous. The mineral composition varies with different mushroom species. Mushrooms contain higher amounts of potassium followed by phosphorous like in higher plants.

4.4.7.1 Estimation of sodium

Mushrooms are generally low in sodium and high in potassium concentration. This low Na to high K ratio (<0.6) implies that mushrooms are suitable for healthy diet. Among the five species of oyster mushrooms, sodium content was found significantly more in *P. djamor* (965 ppm), followed by *P. citrinopileatus* (605 ppm) whereas the content in other mushrooms such as *P. florida* and *H. ulmarius* were 290 ppm and 255 ppm respectively.

The findings were in accordance with Caglarimak (2007) observed that the sodium content in *P. sajor-caju* was 750.77 mg kg⁻¹ on wet basis. The sodium content in *Hypsizygus sp.* was found to be 77.10 mg per 100g of oven dried sample (Chauhan *et al.* 2017). Salami *et al.* (2017) reported that the sodium content in *P. florida* was 277-359 mg per 100g. Jose (2018) reported that the sodium content of *P. florida*, *P. djamor* and *H. ulmarius* were 0.028 per cent, 0.096 per cent and 0.024 per cent respectively.

5.4.7.2 Estimation of phosphorous

Phosphorous is crucial for the generation of ATP in our body and maintenance of acid-base homeostasis. Phosphorous also have important roles in cell structure such as maintenance of cell membrane integrity and nucleic acid synthesis. Phosphorous also plays an important role in the mineralization of bones and teeth. In the present study, *P. citrinopileatus* recorded significantly higher content of phosphorous (96.50 ppm) which differed from other species. *P. sajor-caju* (90.50 ppm) and *H. ulmarius* (90.25 ppm) were on par with each other. *P. djamor* recorded the least phosphorous content (82.75 ppm). Jose (2018) reported that the phosphorous content of *P. florida*, *P. djamor* and *H. ulmarius* were 0.225 per cent, 0.217 per cent and 0.194 per cent respectively which were similar to the present study. Sumi (2016) reported that *H. ulmarius* and *P. florida* contains 0.69 per cent and 0.68 per cent respectively which shows deviation from the present study which may be due to the variation in mineral uptake by the mushroom.

4.4.7.3 Estimation of potassium

Potassium is the most abundant mineral element in various species of mushrooms. It is an essential element which helps to maintain normal heart rhythm, fluid balance and nerve functions (Muthu and Shanmugasundaram, 2016). Sodium and Potassium are crucial in the maintenance of cell osmotic and interstitial fluid balance in animal systems. The potassium content is high in oyster mushroom species.

In the present study, *P. djamor* recorded significantly higher content of Potassium (7175 ppm) followed by *H. ulmarius* (7000 ppm). Potassium content was significantly lower in *P. florida* (3725 ppm). In agreement with the above results, Jose

(2018) reported that the potassium content of *P. florida*, *P. djamor* and *H. ulmarius* were 0.39 per cent, 0.72 per cent and 0.70 per cent respectively. Caglarimak (2007) reported that the potassium content of *P. sajor-caju* was 2687 mg kg⁻¹ on wet basis. Bano and Rajarathnam (1982) reported 3.26 per cent potassium in *P. sajor caju*, 4.57 per cent in *P. eous* and 4.66 per cent in *P. florida*.

4.4.7.4 Estimation of calcium

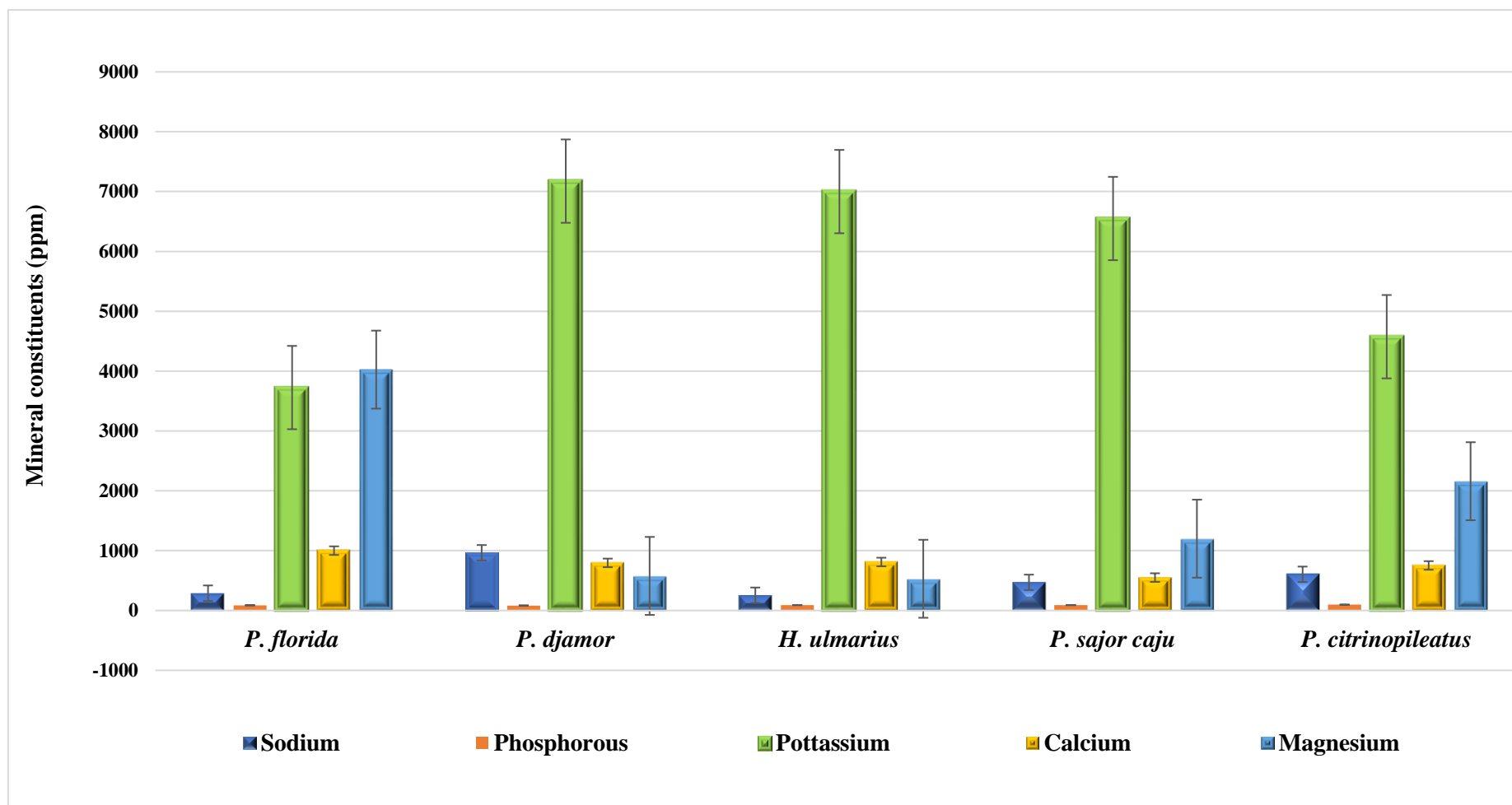
Calcium plays an important role in the circulatory system for mediating vascular contraction and dilation. Calcium regulates muscle function, nerve transmission, intracellular signalling and hormonal secretion in the tissue. Bone tissue serve as a reservoir for and of calcium.

In the present study, significantly higher content of calcium was estimated in *P. florida* (1000 ppm) followed by *H. ulmarius* (810 ppm). Significantly lower calcium content (550 ppm) was recorded from *P. sajor-caju*. Jose (2018) reported that the calcium content of *P. florida*, *P. djamor* and *H. ulmarius* were 0.10 per cent, 0.08 per cent and 0.08 per cent respectively which was in confirmation with the above results.

4.4.7.5 Estimation of magnesium

Magnesium acts as an important co-factor for certain enzymes in many biochemical pathways and help to maintain the functions of nerves and muscles. Also, magnesium supports healthy immune system and keep the bones strong.

In the present study, magnesium content was significantly higher in *P. florida* (4025 ppm) followed by *P. citrinopileatus* (2160 ppm). Significantly lower magnesium content was recorded from *P. djamor* (577.50 ppm) and *H. ulmarius* (530.00 ppm). Similar results were obtained by Jose (2018) who reported that the calcium content of *P. florida*, *P. djamor* and *H. ulmarius* were 0.04 per cent, 0.05 per cent and 0.05 per cent respectively. Kathiravan and Krishnakumari (2017) reported magnesium content of *H. ulmarius* and *P. eous* as 0.019 and 0.064 per cent respectively which is almost in agreement with the present study.



Error bar represents standard error of means of observed values

Figure 3. Mineral constituents of five species of oyster mushrooms

From the present study, it has been confirmed that oyster mushrooms are rich in mineral components. Low sodium and high potassium ratio (1:3) present in oyster mushroom have been reported to lower blood pressure and reduce the risk of heart diseases. Calcium and magnesium content present in oyster mushrooms has been reported to support our immune system and keep the bones strong.

5.5 ANALYSIS OF MEDICINAL COMPONENTS

Mushrooms are the storehouse of medicinal components and many bioactive immunomodulation components have been isolated from mushrooms. These bioactive components such as polysaccharides, glycoproteins, terpenoid, polyphenols and lovastatin are found to be retarding the progress of cancer and other diseases such as hyperglycemia and hypercholesterolemia through immune modulation.

5.5.1 Estimation of β -glucan

β -Glucans are polysaccharides which are the building blocks in fungi since their cell walls are composed of chitin and beta-glucan. It has antitumor activities and the mechanisms include stimulation of hematopoietic stem cells and activation of immune cells such as lymphocytes, macrophages, T cells and B cells (Popovic *et al.* 2013).

In the present study, β -Glucan content was significantly higher in *P. djamor* (30.25 g per 100g) followed by *H. ulmarius* (28.00 g per 100g) which vary significantly. *P. citrinopileatus* (16.75 g per 100g) recorded the least β -Glucan content followed by *P. florida* (20.22 g per 100g).

In confirmation with the above results Sari *et al.* (2017) reported that the β -Glucan content of *P. florida*, *P. djamor* and *P. citrinopileatus* were 20.70 per cent, 17.46 per cent and 15.54 per cent respectively. This was also in agreement with the findings of Jose (2018) who reported that the β -Glucan content of *P. florida*, *P. djamor* and *H. ulmarius* were 20.80 per cent, 32.15 per cent and 26.62 per cent respectively. Avni *et al.* (2017) reported that the beta-glucan content of various *Pleurotus* sp. Ranged between 17.09 per cent and 48.90 per cent.

5.5.2 Estimation of glycoprotein

Certain mushroom glycoproteins have been reported to be inhibitory against cancer cells including that from *Flammulina velutipes* (Ko *et al.*, 1995). Only limited studies are available regarding the analysis and medicinal properties of glycoprotein.

The glycoprotein content recorded was significantly higher in *P. sajor-caju* (435 µg per 100g), followed by *P. citrinopileatus* (337.50 µg per 100g). Glycoprotein content was significantly less in *P. djamor* (117.50 µg per 100g).

5.5.3 Estimation of terpenoid

Triterpenes act as an important intermediate in the biosynthetic pathway of steroids. These have cytotoxic, hepatoprotective and hypolipidemic properties (Chang and Miles, 2004). The terpenoid content recorded from the present study was significantly higher in *P. sajor-caju* (1.26 %) followed by *P. citrinopileatus* (1.13 %). Terpenoid content was significantly less in *P. florida* (0.94 %), *P. djamor* (0.99 %) and *H. ulmarius* (1.01 %).

Menaga *et al.* (2012) reported that the aqueous extract of *P. florida* recorded high levels of terpenoid on qualitative analysis. Sasidhara and Thirunalasundari (2014) studied on the antioxidant potentials of *P. djamor* and confirmed the presence of terpenoid in powdered sample. The terpenoid content in *P. florida*, *P. djamor* and *H. ulmarius* was reported to be 9.22 mg g⁻¹, 9.78 mg g⁻¹ and 10.13 mg g⁻¹ respectively (Jose, 2018), which was in accordance with the present study.

4.5.4 Estimation of Polyphenol

Polyphenols are secondary metabolites with a wide range of medicinal properties. Among the anti-oxidant compounds, polyphenols are of much importance because of the free radical scavenging, metal chelation and enzyme modulation activities. Polyphenol content in mushrooms is responsible for its anti-bacterial and anti-inflammatory activities. (Li *et al.*, 2012)

P. citrinopileatus (4.17 mg per 100g) recorded higher polyphenol content which differed significantly from other species of oyster mushrooms. *P. florida* (1.86 mg per 100g) and *H. ulmarius* (2.05 mg per 100g) did not vary significantly. *P. djamor* (1.50 mg per 100g) recorded the least polyphenol content among the five species of oyster mushrooms. Chirinang and Intarapichet (2009) reported that the total phenolic content in water and ethanol extracts of *P. sajor-caju* was 37.98 and 29.30 gallic acid equivalent (GAE) respectively. The total phenolic content of *P. florida* was estimated as 62.72 mg catechol equivalent. The polyphenol content of *P. florida*, *P. djamor* and *H. ulmarius* to be 19.14 mg GAE g⁻¹, 14.56 mg GAE g⁻¹ and 20.14 mg GAE g⁻¹ respectively (Jose, 2018) which was in agreement with the present study.

4.5.5 Estimation of β -carotene

Beta-carotene, the precursor of vitamin A is one of the major compounds contributing anti-oxidant properties of mushroom. These act as principal compound in biosynthesis of several molecules (Ullah *et al.*, 2011).

P. djamor (3.70 μ g per 100g) recorded significantly higher β -carotene content which was followed by *P. sajor-caju* (2.60 μ g per 100g). *P. florida* (1.78 μ g per 100g) and *H. ulmarius* (2.12 μ g per 100g) were on par with each other. β -carotene content was significantly less in *P. citrinopileatus* (0.85 μ g per 100g). Rajoriya *et al.* (2014) reported that the beta- carotene content in *P. florida* was 18 μ g g⁻¹. which was in contrary to the current results. Beta-carotene content of *P. sajor-caju* (0.038 mg g⁻¹) was higher as compared to *P. florida* (0.018 mg g⁻¹). The β -carotene content of *P. florida*, *P. djamor* and *H. ulmarius* was reported to be 125.24 μ g per 100g, 355.04 μ g per 100g and 195.58 μ g per 100g respectively (Jose, 2018), which was similar to the present study.

4.5.6 Estimation of lovastatin

Lovastatin is a statin medication, to treat high blood cholesterol and reduce the risk of cardiovascular disease. Lovastatin is reported to be naturally produced by *Pleurotus ostreatus* and closely related *Pleurotus* spp. Lovastatin content was estimated to be higher in *P. djamor* (370.75 μ g per 100g), which was followed by *P.*

sajor-caju (257.25 µg per 100g). The least content of lovastatin was recorded from *P. citrinopileatus* (85.00 µg per 100g) followed by *P. florida* (129.00 µg per 100g).

Medicinal analysis of five species of oyster mushrooms revealed that even though *P. djamor*, *P. citrinopileatus* and *P. sajour-caju* were poor yielders compared to *H. ulmarius* and *P. florida*, they were rich in protein, fibre and aminoacids. *P. djamor* was rich in β-Glucan which is having antitumour and immunomodulation activities, β-carotene having antioxidant properties and lovastatin which reduce blood cholesterol and the risk of cardiovascular diseases. *P. citrinopileatus* and *P. sajour-caju* were rich in glycoprotein, polyphenols and terpenoids. Glycoproteins have been reported to be inhibitory against cancer cells. Terpenoids have hepatoprotective and hypolipidemic properties. Even though *P. citrinopileatus* and *P. djamor* are not widely preferred for consumption, it has been understood from the present study that both of them have high medicinal value which can make a strong case for their addition in our diets.

5.6 SENSORY EVALUATION OF FIVE SPECIES OF OYSTER MUSHROOMS

Mushrooms can be used as an ingredient in many dishes since it enhances taste, flavour, texture and nutrient content (Kumar and Barmanray, 2007). Jaziya (2011) reported that the addition of oyster mushrooms in diet was acceptable to the consumers. Prabhu (1991) reported that the appreciation per cent of *P. djamor* and *P. sajour-caju* was 80 per cent and 56.66 per cent.

Sensory evaluation was done on sauteed mushroom recipe to evaluate the quality of mushroom for attributes like appearance, colour, flavour, texture, taste and overall acceptability. The evaluation was done using nine-point score card and Hedonic rating scale. A panel of twelve judges evaluated the recipe from five species of oyster mushrooms for the various attributes. Comparison between all the five species of oyster mushroom were carried out based on the sensory parameters. It was revealed that *H. ulmarius* excelled in all the sensory parameters such as appearance (8.83), colour (8.75), texture (8.66), flavour (8.58) and taste (8.83) on a maximum score of 9. *H. ulmarius* scored 8.80 in terms of overall acceptability followed by *P. florida* (7.60).

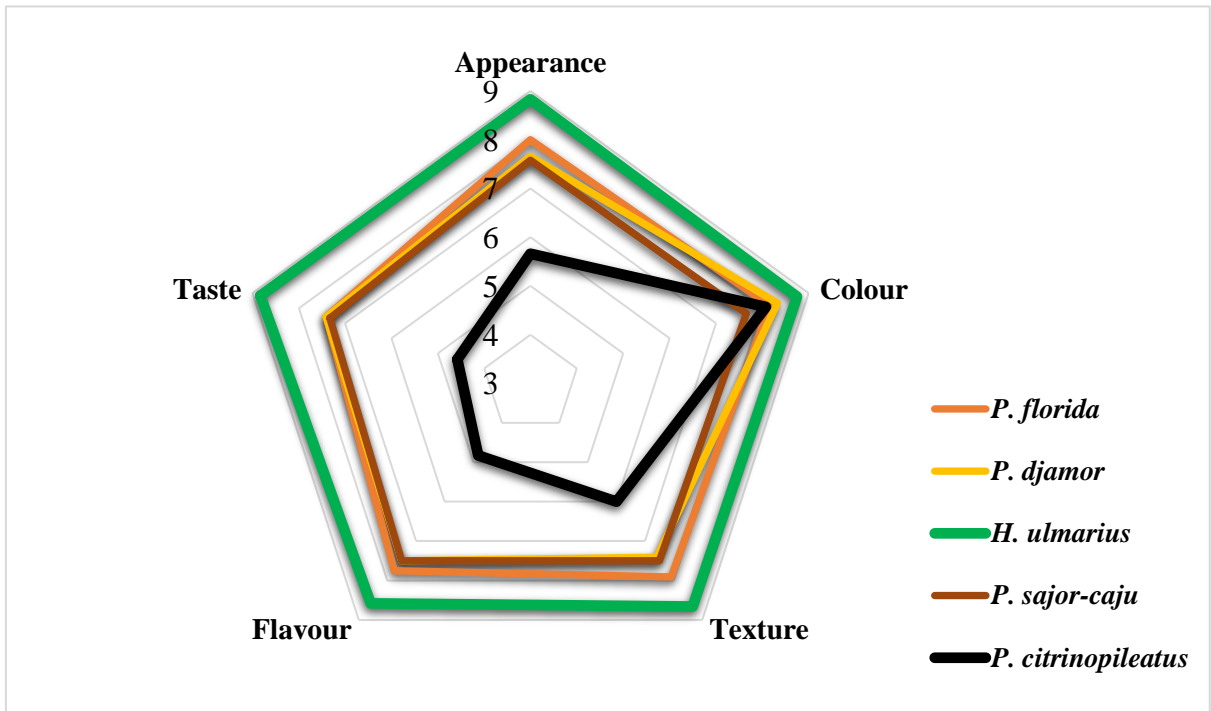


Figure 4. Sensory scores (mean values) of saute developed from oyster mushroom

From this study it was found that oyster mushrooms, especially *H. ulmarius* and *P. florida* were highly favoured for consumption.

5.7 EVALUATION OF SHELF LIFE OF FIVE SPECIES OF OYSTER MUSHROOM UNDER ROOM CONDITION AND REFRIGERATED CONDITION

The moisture contents in mushroom are generally very high and hence mushrooms are highly perishable and susceptible to microbial growth. Since mushroom is a highly perishable commodity, preservation is having much importance

In the present study, comparatively higher shelf life was observed on *P. djamor* and *P. sajor-caju* which could be stored for two days after harvesting in room temperature ($26\pm 2^{\circ}\text{C}$) and three days in refrigerated conditions (15°C). This may be due to the higher fiber content and lower moisture content in their fruiting bodies. *P. florida*, *H. ulmarius* and *P. citrinopileatus* can be stored under room conditions upto only one day after harvest. Higher shelf life was observed on *P. djamor* followed by *P. sajor-caju*.

Lal and Sharma (1995) reported that the higher moisture content of the species may be attributed to the fast deterioration. Even after harvesting, mushrooms continue to grow and respire which results in weight loss and microbial spoilage. Hammond and Nichols (1975) reported that the very high respiration rate of 28.2 – 43.6 mg CO₂ per kg of fresh weight per hour is responsible for the shorter shelf life of mushrooms. Storage under low temperature is a very good method for restricting deterioration of harvested mushrooms for a limited period of time (Rai and Arumuganathan, 2008).

6. SUMMARY

The present research project was aimed to identify the best suited oyster mushroom in five agro-ecological zones of Kerala and to detect the biochemical, medicinal and storage properties of each species. Five species of oyster mushrooms viz, *Pleurotus florida*, *Pleurotus djamor*, *Hypsizygus ulmarius*, *Pleurotus sajor-caju* and *Pleurotus citrinopileatus* were cultivated in the farmers field of five agro-ecological zones of Kerala namely coastal plains, midland laterites, foothills, High hills and Palakkad plains.

Isolation of five species has been done by tissue culture technique and pure culturing has been done by hyphal tip method. The mushrooms were grown on PDPA medium and growth rate assessed. The rate of mycelial growth was significantly higher in all the species and took 7-9 days to complete mycelial growth except *P. djamor* which took 12 days to complete mycelial growth in 9 cm Petri dish. *H. ulmarius* and *P. sajor-caju* attained complete mycelial growth in minimum days (seven days).

P. florida produced white, fleshy sporocarp having comparatively large pileus with 6.62 cm x 7.52 cm. Long and stout stipe (3.22 cm) was recorded from the sporocarp and it had 12 gills per cm of the pileus. *P. djamor* produced pinkish white leathery sporocarps with smaller pileus. Average pileus dimension of 4.90 cm x 6.50 cm with very short stipe (0.8 cm) was recorded from *P. djamor*. Number of gills from the margin was 17.50 gills/cm which were significantly higher than other species studied. *H. ulmarius* produced creamy white fleshy sporocarps with significantly large pileus. The average pileus size was 7.0 cm x 7.52 cm. The average length of stipe was 3.35 cm which was comparatively higher. The average sporocarp weight was 9.33 g. Gills were attached to the stipe in a non- decurrent fashion and the average number of gills from the margin was 13.75 gills per cm. *P. sajor-caju* produced greyish white leathery sporocarp. The average length and breadth of pileus was 7.10 cm and 5.32 cm respectively. Significantly long and stout stipe with an average length of 5.32 cm was one of the characteristic features of *P. sajor-caju*. The sporocarps were produced singly and the average number of gills was 17.50 gills per cm. *P. citrinopileatus*

produced golden yellow delicate sporocarp with comparatively short stipe. The average size of pileus was 5.45 cm x 6.17 cm and the average number of gills from the margin was 11.75 gills per cm. The average stipe length recorded was 3.30 cm and the average sporocarp weight was 5.05 g.

Spore print of *P. florida* and *H. ulmarius* was pure white. *P. djamor* produced light pink coloured spore print which changed to creamish white. *P. sajor-caju* and *P. citrinopileatus* produced pale white spore print.

Microscopic observations of hyphae and spores of five species of oyster mushrooms shows that hyphae of all the five species were septate, branched and hyaline with clamp connections. The width of the hyphae of all the five species ranged from 1.5-4.5 μm . Basidiospores of *P. florida* and *P. djamor* recorded 7-12 μm in length and 2-5 μm in width whereas, *H. ulmarius* and *P. sajor-caju* produced spores with dimension of 8-12 $\mu\text{m} \times 3-6 \mu\text{m}$. *P. citrinopileatus* produced spores with dimension of 7-10 $\mu\text{m} \times 2-3 \mu\text{m}$.

Two substrates namely paddy straw and rubber sawdust were evaluated for the cultivation of five species of oyster mushrooms. Observations on time taken for complete spawn run, time taken for pin-head formation, time taken for first harvest, total crop period and total yield from three harvests were recorded from five species of oyster mushrooms on both the substrates. Selection of substrate significantly affected the total yield. Total yield from first three harvests were significantly less in paddy straw, (954.66 g) compared to rubber sawdust which was superior and given a yield of 1306.40 g. The yield recorded was highest for *H. ulmarius* cultivated on both paddy straw and rubber sawdust (1233 g kg⁻¹ and 1611 g kg⁻¹ respectively) which was followed by *P. florida* (1148.30 g kg⁻¹ and 1582.66 g kg⁻¹ respectively). Selection of substrate significantly affected the time taken for complete spawn run. Time taken for complete spawn was significantly less (16.20 days) in paddy straw and superior to rubber sawdust substrate which took 24.86 days to complete spawn run. Less crop period was observed on *P. djamor* cultivated on both the substrates (54.00 days and 90.33 days). Significantly higher crop period was recorded for *P. sajor-caju* cultivated on both the substrates (67.33 and 111.66 days respectively).

Among all the cultivated mushrooms, oyster mushrooms have the maximum number of commercially cultivated species which can be cultivated throughout the year. Kerala having a tropical climate is found to be suitable for the cultivation of oyster mushrooms. Kerala is divided into five agro-ecological zones from coastal plains to high hills, with each zone having wider climatic conditions (KAU, 2016). But studies have not been yet conducted in Kerala based on the suitability of different oyster mushroom for the specific agro-ecological zone. In the present study, the suitability of five species of oyster mushrooms were analysed under five agro-ecological zones of Kerala

In coastal plains, the highest yield of 1029.96 g was recorded from *H. ulmarius* which was followed by *P. florida* (886.20 g) and *P. djamor* (723.00 g). Significantly lower yield of 550.10 g was obtained from *P. sajor-caju* and *P. citrinopileatus* (630.00 g). The time taken for first harvest was found to be significantly less in *P. djamor*, which yielded in 18.66 days. and completed the crop period within 53.66 days. *H. ulmarius* was found to be the best suited oyster mushroom species for cultivation in coastal plains in terms of yield.

In midland laterites, the highest yield of 827.00 g was obtained from *P. djamor*. The yield recorded from *H. ulmarius* and *P. djamor* were on par with each other (806.00 g and 773.03 g respectively). Significantly lower yield of 658.83 g was recorded from *P. sajor-caju* in midland laterites. The time taken for first harvest was found to be significantly less in *P. djamor*, which yielded in 19.33 days and completed the total crop period within 57.00 days followed by *P. florida* (63.00 days). *P. djamor* was found to be the best suited oyster mushroom species for cultivation in midland laterites in terms of yield and lesser crop period.

In foothills, the highest yield of 927.33 g was recorded from *H. ulmarius*, which was followed by *P. florida* (823.33 g). The time taken for first harvest was found to be significantly less in *P. djamor*, which yielded in 20.00 days. *P. florida*, *H. ulmarius* and *P. citrinopileatus* took 25.33 days, 24.00 days and 25.33 days respectively for first harvest. *H. ulmarius* was found to be the best suited oyster mushroom species for cultivation in foothills.

In high hills, the highest yield of 1233.00 g was obtained from *H. ulmarius*, followed by *P. florida* (1148.30 g) and *P. djamor* (1050.30 g). Significantly lower yield was obtained from *P. citrinopileatus* (610.00 g). *P. djamor* took the least number of days to complete spawn run in high hills (11.66 days) followed by *P. florida* (15.66 days). *H. ulmarius*, *P. florida* and *P. djamor* were found to be best suitable for cultivation in high hills based on comparatively low total cropping period and high yield.

In Palakkad plains, *P. djamor* produced significantly higher yield of 1038.00 g compared to other species of oyster mushrooms. It was followed by *H. ulmarius* (824.60 g) and *P. florida* (749.00 g). *P. djamor* took lesser number of days to yield (22.33 days) followed by *H. ulmarius* (24.33 days). *P. djamor* was found to be the best suited oyster mushroom species for cultivation in Palakkad plains.

In all the five locations, *P. djamor* took the minimum number of days to complete both spawn running, pin-head formation, first harvest and total crop period while *P. sajor-caju* took the maximum number of days to complete both spawn running, pin-head formation, first harvest and total crop period.

The proximate analysis of five species of oyster mushrooms was carried out and it revealed that mushrooms are rich source of proteins, carbohydrates, fibres, amino acids, vitamins and minerals with less fat content. The protein content in *P. sajor-caju* was estimated to be 26.02 per cent which was the highest among the five mushrooms followed by *P. citrinopileatus* (23.07 per cent) and *P. florida* (22.42 per cent). *P. djamor* was recorded with the least protein content (19.55 %). Fibre is considered as an important ingredient in a healthy diet. The crude fibre content of *P. djamor* was estimated as 26.20 per cent which was highest among five mushrooms followed by *P. florida* (24.21%) while *P. sajor-caju* and *P. citrinopileatus* had fibre content of 23.77 and 21.15 per cent respectively. *H. ulmarius* recorded 18.47 per cent fibre content on dry weight basis which was the least among the five mushrooms studied.

Mushrooms contain very low fat. Fat content was the least in *H. ulmarius* with 1.70 per cent on dry weight basis. *P. djamor*, *P. florida* and *P. sajor-caju* contains fat content of 1.97 per cent, 2.82 per cent and 2.50 per cent respectively. Highest fat content was recorded from *P. citrinopileatus* (3.60 %). *H. ulmarius* (31.27 %) recorded the highest carbohydrate content among the five species of oyster mushrooms and was followed by *P. florida* (29.17%) and *P. djamor* (27.57 %). Carbohydrate content was minimum in *P. citrinopileatus* (24.27%) followed by *P. sajor-caju* (25.55%). The moisture content of five species were determined on fresh weight basis. *P. florida* recorded the maximum moisture content (90.97 %) followed by *P. sajor-caju* (88.22 %) and *H. ulmarius* (86.85 %). Low moisture content was estimated from *P. djamor* (84.32 %) followed by *P. citrinopileatus* (85.95 %). Highest amino acid content of 19.40 per cent was recorded from *P. sajor-caju* followed by *P. djamor* (17.47 %) and *P. citrinopileatus* (15.92 %). Amino acid content was less in *H. ulmarius* (10.62 %) followed by *P. florida* (13.57 %).

Mushrooms are an important source of minerals such as sodium, potassium, phosphorous, calcium and magnesium. Studies conducted on the mineral composition of mushrooms revealed that sodium content in oyster mushrooms was relatively less in quantity compared to other minerals. *P. djamor* (965 ppm) recorded the highest sodium content followed by *P. citrinopileatus* (605 ppm). *P. florida* (290 ppm) and *H. ulmarius* (255 ppm) were significantly less. *P. citrinopileatus* recorded highest phosphorous content of 96.50 ppm followed by *P. sajor-caju* (90.50 ppm) and *H. ulmarius* (90.25 ppm). *P. djamor* recorded the least phosphorous content (82.75 ppm). Among the five species of oyster mushrooms, *P. djamor* recorded higher content of Potassium (7175 ppm) followed by *H. ulmarius* (7000 ppm). Potassium content was significantly lower in *P. florida* (3725 ppm). The calcium content recorded was higher in *P. florida* (1000 ppm) followed by *H. ulmarius* (810 ppm). Significantly lower calcium content (550 ppm) was recorded from *P. sajor-caju*. Magnesium content recorded was highest in *P. florida* (4025 ppm) followed by *P. citrinopileatus* (2160 ppm). Lower magnesium content was recorded from *P. djamor* (577.50 ppm) and *H. ulmarius* (530.00 ppm).

Analysis of medicinal constituents confirmed that mushrooms were rich in bioactive components such as β -Glucan, glycoprotein, terpenoid, polyphenol, β -carotene and lovastatin. β -Glucan content recorded was highest in *P. djamor* (30.25 g per 100g) followed by *H. ulmarius* (28.00 g per 100g). *P. citrinopileatus* (16.75 g per 100g) recorded the least β -Glucan content followed by *P. florida* (20.22 g per 100g). Terpenoid content was significantly higher in *P. sajor-caju* (1.26 %) followed by *P. citrinopileatus* (1.13 %). Terpenoid content recorded was less in *P. florida* (0.94 %), *P. djamor* (0.99 %) and *H. ulmarius* (1.01 %). *P. djamor* (3.70 μ g per 100g) recorded highest β -carotene content compared to other species of oyster mushrooms which was followed by *P. sajor-caju* (2.60 μ g per 100g). *P. florida* (1.78 μ g per 100g) and *H. ulmarius* (2.12 μ g per 100g) were on par with each other. β -carotene content was less in *P. citrinopileatus* (0.85 μ g per 100g). Lovastatin content was estimated to be higher in *P. djamor* (370.75 μ g per 100g), which was followed by *P. sajor-caju* (257.25 μ g per 100g). The least content of lovastatin was recorded from *P. citrinopileatus* (85.00 μ g per 100g) followed by *P. florida* (129.00 μ g per 100g).

Sensory evaluation of five species of oyster mushrooms was done using sauteed mushroom recipe to evaluate the quality attributes and consumability of the species. Comparison between all the five species of oyster mushroom were carried out based on the sensory parameters. *H. ulmarius* recorded highest scores for appearance (8.83), colour (8.75), texture (8.66), flavour (8.58) and taste (8.83) on a maximum score of 9. It was observed that *H. ulmarius* excelled in all the sensory parameters. Overall, from this study it was found that oyster mushrooms, especially *H. ulmarius* and *P. florida* are highly favoured for consumption.

Shelf life studies of five species of oyster mushrooms were undertaken both in room temperature ($26\pm 2^{\circ}\text{C}$) and in refrigerated conditions (15°C). Observations were recorded in terms of number of days for change in colour, texture and physical appearance based on visual observation.

In the present study, comparatively higher shelf life was observed on *P. djamor* and *P. sajor-caju* which could be stored for two days after harvesting in room temperature ($26\pm 2^{\circ}\text{C}$) and three days in refrigerated conditions (15°C). *P. florida*, *H.*

ulmarius and *P. citrinopileatus* can be stored under room conditions upto only one day after harvest.

The present study demonstrated the exploitability of five species of oyster mushrooms in Kerala conditions. All the five species can be cultivated under the five agro-ecological zones of Kerala but *H. ulmarius*, *P. florida* and *P. djamor* yielded well compared to *P. citrinopileatus* and *P. sajor-caju*. The study revealed that both the substrates *viz.*, paddy straw and rubber sawdust were suitable for the cultivation of oyster mushrooms and can be selected for cultivation based on the ease of availability and cost. Analysis of proximate and medicinal components present in five species of oyster mushrooms revealed that they are a storehouse of nutritive and medicinal value. Sensory evaluation suggests that apart from their nutritive and medicinal value, oyster mushrooms are very tasty and can be included in our diets. From the present study, it has been understood that *H. ulmarius* and *P. florida* are highly favoured for consumption. Shelf-life studies revealed that oyster mushrooms are highly perishable. *P. djamor* has higher storage life among the five species studied. It has been proven that storage under low temperature is a very good method for restricting deterioration of harvested mushrooms for a limited period of time.

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Petaloid/ Flabelliform/ Mucronate / Depressed /
Dimidate / Resupinate

When young :

Size :

At maturity :

Colour :

Texture : Soft/ Brittle/ Fleshy/ Coriaceous/ Hyphanous/
Fragile/ Cartilaginous/ Membraneous

Surface : Smooth/ Scaly/ Rugose/ Rugulose/ Viscid/ Striate/ Dry/
Squamulose/ Velutinous/ Pubescent/ Strigose/ Sulcate/
Tomentose/ Aveolate/ Farinose/ Floccose/ Punctate/ Rivose/
Rivulose

Margin : Serrate/ Serrulate/ smooth/ Undulate/ Reflexed/ Involute/
Fimbriate/ Incised/ Lobed/ Revolute

Context colour:

Before cutting :

After cutting :

Colour changes with :-

Melzer's reagent : Amyloid/ Psuedoamyloid/ Inamyloid

Green Vitrial :

Phenol :

Sulphovanilin :

GILLS

Arrangement : Remote/ Free/ Decurrent/ Adanate/ Adnexed/
Sinuate

Shape : Rounded anteriorly or posteriorly/
Lanceolate/ Ventricose/ Reticulate

Texture : Soft/ Brittle/ Ceraceous/ Waxy/ Thick/
Papery/ Opaque

Margin : Smooth/ Wavy/ Serrate/ Fimbriate/ Dentate

Length

Size :

Shape : Clavate/ Obclavate/ Cylindrical/ Solid/ Hollow/ Slender/ Short

Attachment to pileus : Lateral/ Eccentric/Central/ Resupinate/ Glabrous/
Scaly/ Pubescent/ velutinous/ Squamose/ Tomentose

Colour

Before cutting :

After cutting :

Reaction with Melzer's reagent : Amyloid/ Psuedoamyloid/ Inamyloid

Basal part : Globular/ Annular stipes/ Fusoid/ Bulbous/
Sheathing bulbous/ marginately depressed
bulb/Pseudorhizoid/Rhizines/Rhizemorphoid

VOLVA

Present/ Absent Persistent/ Evanescent

Shape : Free/ Lobed/ Irregular/ Cup like

Colour :

Texture : Soft/ Fleshy/ Tough/ Papery

Odour

Before cutting :

After cutting :

Taste : Acrid/ Mealy/ Acidulous/ Blunt

SPORE PRINT

Colour :

Other details :

BASIDIA

Size :

Shape :

Sterigmata :

SPORES

Colour :

Reaction with Melzer's reagent : Amyloid/ Psuedoamyloid/ Inamyloid

Shape : Ovate/ Elliptical/ Globose/
subglobose/ Apiculate/ Cylindrical/
Fusiform/ Angular/ Echinulate/
Verrucose/ Reticulate/ Tuberculate/
Ovoid/ Abtuselyfusiform/ Allantoid/
Guttulate/ Pip shaped/ Piriform/
Pedicilate/ Muriform/ Filiform

Other characters of spores :

ANY OTHER DETAILS

APPENDIX- II

Composition of stain used

1. Lactophenol Cotton blue

Phenol crystals	-	20.0 g
Cotton blue	-	0.05 g
Lactic acid	-	20.0 mL
Glycerol	-	20.0 mL
Distilled water	-	20.0 mL

APPENDIX III

Composition of media

1. Potato Dextrose Peptone Agar

Potatoes (Sliced)	-	200.00 g
Agar-agar	-	20.00 g
Dextrose	-	20.00 g
Peptone	-	10 g
Distilled H ₂ O	-	1000 ml

APPENDIX IV

Score Card

Appearance	
Like Extremely	9
Like Very Much	8
Like Moderately	7
Like Slightly	6
Neither Like nor Dislike	5
Dislike Slightly	4
Dislike moderately	3
Dislike Very Much	2
Dislike Extremely	1
Colour	
Like Extremely	9
Like Very Much	8
Like Moderately	7
Like Slightly	6
Neither Like nor Dislike	5
Dislike Slightly	4
Dislike moderately	3
Dislike Very Much	2
Dislike Extremely	1
Flavour	
Like Extremely	9
Like Very Much	8
Like Moderately	7
Like Slightly	6
Neither Like nor Dislike	5
Dislike Slightly	4
Dislike moderately	3
Dislike Very Much	2

Dislike Extremely	1
Texture	
Extremely soft and fleshy	9
Very soft and fleshy	8
Moderately soft and fleshy	7
Slightly soft and fleshy	6
Neither soft nor fibrous	5
Slightly fibrous	4
Moderately fibrous	3
Very Fibrous	2
Extremely fibrous	1
Taste	
Like Extremely	9
Like Very Much	8
Like Moderately	7
Like Slightly	6
Neither Like nor Dislike	5
Dislike Slightly	4
Dislike moderately	3
Dislike Very Much	2
Dislike Extremely	1
Overall acceptability	
Like Extremely	9
Like Very Much	8
Like Moderately	7
Like Slightly	6
Neither Like nor Dislike	5
Dislike Slightly	4
Dislike moderately	3
Dislike Very Much	2
Dislike Extremely	1

APPENDIX V
Hedonic rating scale

Particulars	Score
Like Extremely	9
Like Very Much	8
Like Moderately	7
Like Slightly	6
Neither Like nor Dislike	5
Dislike Slightly	4
Dislike moderately	3
Dislike Very Much	2
Dislike Extremely	1

APPENDIX VI

Recipe and method of preparation

Sauted mushroom

Mushroom	-	100g
Big onion	-	10g
Green chilli	-	5g
Tomato	-	20g
Coconut oil	-	1 teaspoon
Pepper powder	-	1/8 teaspoon
Turmeric powder	-	a pinch
Red chilli powder	-	a pinch
Salt	-	sufficiently
Curry leaves	-	one sprig

Heated the oil, sauted big onion, green chilli and tomato. Added mushroom pieces, salt and other ingredients. Then sauted and cooked by covering the vessel. Served hot.

APPENDIX VII

Weather data at RARS, Pilicode, Kerala Agricultural University

Date	Outdoor				Indoor (Mushroom House)		
	Temp (°C)		RH (%)	Total rainfall (mm)	Temp. (°C) (9 am)	Temp. (°C) of mushroom bed	RH (%)
	Max	Min					
10-01-2021	32	23	96	0	28	29	90
11-01-2021	32	23	91	0	27	27.5	85
12-01-2021	32	22.5	96	0	26.5	27	95
13-01-2021	33	22	91	0	27	28	85
14-01-2021	32	22	96	0	27.5	28	90
15-01-2021	31	24	88	0	27	28	90
16-01-2021	32	23	96	0	26.5	27	90
17-01-2021	32	23	96	0	28	27	85
18-01-2021	32	23	91	0	27	27	85
19-01-2021	31	22.5	96	0	27	27	85
20-01-2021	31	21	93	0	27.5	28	90
21-01-2021	34	21	96	0	28	29	85
22-01-2021	34	22	96	0	28	29	90
23-01-2021	34	24	88	0	28.5	29	90
24-01-2021	34	24	96	0	29	28	90
25-01-2021	34	22	96	0	28	28	90

APPENDIX VIII

Weather data from Thiruvananthapuram City

Date	Outdoor			
	Temp (° C)		RH (%)	Total rainfall (mm)
	Max.	Min		
10-01-2021	32.1	24.2	93	2.3
11-01-2021	29.5	23.8	93	0.6
12-01-2021	31.5	23.3	89	1.8
13-01-2021	29	23.3	96	4.9
14-01-2021	27.2	23.3	96	17.1
15-01-2021	32.2	24.7	92	0
16-01-2021	32	25.2	92	0
17-01-2021	32.1	24	90	0
18-01-2021	32.5	21.8	88	0
19-01-2021	33.5	21.6	84	0
20-01-2021	32.5	23.4	88	0
21-01-2021	32.1	24.8	92	0
22-01-2021	32.3	24.9	92	0
23-01-2021	32.8	24.2	93	0
24-01-2021	32.7	23.7	93	0
25-01-2021	32.3	21.8	76	0

APPENDIX IX

Weather data from Thiruvananthapuram Airport

Date	Outdoor			
	Temp (° C)		RH (%)	Total rainfall (mm)
	Max.	Min		
10-01-2021	31.8	24.2	89	2.5
11-01-2021	29.9	23.8	84	0.8
12-01-2021	30.7	23.5	82	1
13-01-2021	27.8	23.1	95	2.1
14-01-2021	28.4	23.2	87	5.1
15-01-2021	31.6	24.7	84	0
16-01-2021	31.7	25.1	86	0
17-01-2021	31.5	23.7	85	0
18-01-2021	31.8	22.6	78	0
19-01-2021	33.2	22.4	69	0
20-01-2021	32.6	23.6	70	0
21-01-2021	31.5	24.8	83	0
22-01-2021	31.8	24.8	90	0
23-01-2021	31.8	24.3	80	0
24-01-2021	32.1	23.5	77	0
25-01-2021	32	23.1	60	0

APPENDIX X

Weather data at CRS, Pampadumpara, Kerala Agricultural University

Date	Outdoor			
	Temp (° C)		RH (%)	Total rainfall (mm)
	Max.	Min		
10-01-2021	20	10	95	6.0
11-01-2021	20	10	95	0.0
12-01-2021	18.5	9	95	1.0
13-01-2021	18.5	9.5	95	34.0
14-01-2021	19	9.5	95	51.2
15-01-2021	18.5	9	94	5.0
16-01-2021	20	10	95	3.0
17-01-2021	20.5	10.5	95	3.0
18-01-2021	20.5	10.5	95	0.0
19-01-2021	18.5	9.5	95	0.0
20-01-2021	20.5	11	95	0.0
21-01-2021	22.5	12.5	95	0.0
22-01-2021	22.5	13	95	0.0
23-01-2021	20.5	11.5	95	0.0
24-01-2021	21	11.5	95	0.0
25-01-2021	19.5	10	95	0.0

APPENDIX XI

Weather data at RARS Pattambi, Kerala Agricultural University

Date	Outdoor			
	Temp (° C)		RH (%)	Total rainfall (mm)
	Max.	Min		
10-01-2021	32	24	84	0.0
11-01-2021	30.8	24.4	87	0.0
12-01-2021	30.4	23.6	84	0.0
13-01-2021	29.8	23.8	91	0.0
14-01-2021	31	24.2	84	0.0
15-01-2021	31.8	24.2	76	0.0
16-01-2021	33.4	23.1	80	0.0
17-01-2021	33.2	23.8	74	0.0
18-01-2021	30.4	22.4	81	0.0
19-01-2021	32.2	21.4	83	0.0
20-01-2021	33.8	20.6	96	0.0
21-01-2021	34.4	22.6	91	0.0
22-01-2021	33.8	23.2	90	0.0
23-01-2021	33.8	22.4	85	0.0
24-01-2021	33	22	80	0.0
25-01-2021	32.8	23.9	92	0.0

**COMPARATIVE EVALUATION OF DIFFERENT SPECIES OF OYSTER
MUSHROOM SUITABLE TO KERALA**

By
AKHIL G. L.
(2019-11-233)

Abstract of the Thesis
Submitted in partial fulfilment of the requirement
For the degree of

MASTER OF SCIENCE IN AGRICULTURE
Faculty of Agriculture
Kerala Agricultural University



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ABSTRACT

The present study entitled “Comparative evaluation of different species of Oyster mushroom suitable to Kerala” was carried out in the College of Agriculture, Padannakkad during 2019-2021 with the objective to identify the oyster mushroom species best suitable for cultivation under five agro-ecological zones of Kerala.

Five species of Oyster mushrooms were selected for the study viz., *Pleurotus florida*, *P. djamor*, *Hypsizygus ulmarius*, *P. sajor-caju* and *P. citrinopileatus*. To evaluate the yield performance, five species were cultivated in farmer’s field of five agro-ecological zones of Kerala. The study also aims to evaluate the proximate constituents and medicinal components present in oyster mushrooms. Sensory evaluation and shelf life studies were also carried out.

The sporocarps of five species of oyster mushrooms were studied for their morphological characters. *P. florida* produced white, delicate and fleshy sporocarp with an average weight of 11.71 g. *P. djamor* produced pinkish white leathery sporocarps with smaller pileus having very short stipe (0.8 cm). *H. ulmarius* produced creamy white fleshy sporocarps with significantly large pileus having average size of 7.0 cm x 7.52 cm. *P. sajor-caju* produced greyish white leathery sporocarp. *P. citrinopileatus* produced golden yellow delicate sporocarp with comparatively short stipe. The average size of pileus was 5.45 cm x 6.17 cm.

Microscopic observations of hyphae and spores of five species of oyster mushrooms shows that hyphae of all the five species were septate, branched and hyaline with clamp connections. The width of the hyphae of all the five species ranged from 1.5-4.5 μm . Basidiospores of *P. florida* and *P. djamor* recorded 7-12 μm in length and 2-5 μm in width whereas, *H. ulmarius* and *P. sajor-caju* produced spores with dimension of 8-12 μm x 3-6 μm . *P. citrinopileatus* produced spores with dimension of 7-10 μm x 2-3 μm .

Two substrates namely paddy straw and rubber sawdust were evaluated for the cultivation of five species of oyster mushrooms. Total yield from three harvests were significantly less in paddy straw, (954.66 g) compared to rubber sawdust which has given a yield of 1306.40 g. The yield recorded was highest for *H. ulmarius* cultivated on both paddy straw and rubber sawdust (1233 g kg⁻¹ and 1611 g kg⁻¹ respectively) which was followed by *P. florida* (1148.30 g kg⁻¹ and 1582.66 g kg⁻¹ respectively). It has been evident that selection of substrate significantly affected time taken for complete spawn run, time taken for pin-head formation, time taken for first harvest, total crop period and total yield from three harvests. The present study revealed that both the substrates *viz.*, paddy straw and rubber sawdust were suitable for the cultivation of oyster mushrooms. So, both the substrates can be selected for cultivation based on the ease of availability and cost.

In the present study, the suitability of five species of oyster mushrooms in five agro-ecological zones of Kerala were evaluated by cultivating the mushroom species at coastal plains, midland laterites, foothills, high hills and Palakkad plains. *H. ulmarius* was found to be the best suited oyster mushroom species for cultivation in coastal plains (102.99 % BE), foot hills (92.73 % BE) and high hills (123.30 % BE). In midland laterites and Palakkad plains, highest BE of 82.70 (%) and 103.80 (%) respectively were obtained from *P. djamor*. *P. djamor*, although reported to be performing well under warm conditions, yielded well under midland laterites and Palakkad plains. This may be due to low relative humidity and comparatively higher temperature in these zones. Earliness in primordial initiation (14-19 days) and extremely fast growth also make this species promising for cultivation in Kerala especially during dry months.

The proximate analysis of five species of oyster mushrooms was carried out and it revealed that mushrooms are rich source of proteins, carbohydrates, fibres, amino acids, vitamins and minerals with less fat content. The protein content was highest in *P. sajor-caju* (26.02 %) and the fibre content was highest in *P. florida* (26.20 %). Fat content was the least in *H. ulmarius* with 1.70 per cent on dry weight basis. *H. ulmarius* (31.27 %) recorded the highest carbohydrate content among the five species of oyster mushrooms.

P. florida recorded the maximum moisture content (90.97 %). Highest amino acid content of 19.40 per cent was recorded from *P. sajor-caju*

Analysis of mineral components revealed that highest content of sodium (965 ppm) and potassium (7175 ppm) was present in *P. djamor*. *P. florida* recorded highest content of calcium (1000 ppm) and magnesium (4025 ppm) which supports healthy immune system and keep bones strong.

Medicinal analysis confirmed that these mushrooms were rich in bioactive components. *P. djamor* recorded highest content of β -glucan (30.25 g per 100g) having antitumour and immunomodulation activities, β -carotene (3.70 μ g per 100g) having antioxidant properties and lovastatin (370.75 μ g per 100g) which reduce blood cholesterol. *P. sajor-caju* recorded highest glycoprotein (435.00 μ g per 100g) and terpenoid (1.26 %) content.

Sensory evaluation of five species of oyster mushrooms was done using sauteed mushroom recipe to evaluate the quality attributes and consumability of the species. *H. ulmarius* recorded highest scores for appearance (8.83), colour (8.75), texture (8.66), flavour (8.58) and taste (8.83) on a maximum score of 9. *H. ulmarius* scored 8.80 in terms of overall acceptability followed by *P. florida* (7.60). Overall, from this study it was found that oyster mushrooms, especially *H. ulmarius* and *P. florida* are highly favoured for consumption.

Shelf life of five species of oyster mushrooms were undertaken both in room temperature ($26\pm 2^{\circ}\text{C}$) and in refrigerated conditions (15°C). In the present study, comparatively higher shelf life was observed on *P. djamor* and *P. sajor-caju* which could be stored for two days after harvesting in room temperature and three days in refrigerated conditions.

The present study demonstrated the exploitability of five species of oyster mushrooms in Kerala conditions. All the five species can be cultivated under the five agro-ecological zones of Kerala but *H. ulmarius*, *P. florida* and *P. djamor* yielded well compared to *P. citrinopileatus* and *P. sajor-caju*. *H. ulmarius* was found to be the best

suited oyster mushroom species for cultivation in coastal plains, foot hills and high hills. In midland laterites and Palakkad plains, highest yields were obtained from *P. djamor* can be economically cultivated regarding the yield and earliness in fruiting.

സംഗ്രഹം

ഹരിത രഹിതമായ സസ്യങ്ങളിൽ ഉൾപ്പെടുന്ന ഒരു വിഭാഗമാണ് കൂണുകൾ. ചരിത്രാതീത കാലം മുതൽ അവ ഭക്ഷണത്തിനും ഔഷധത്തിനുമായി ഉപയോഗിച്ചുവരുന്നു. സാധാരണക്കാർ കിടയിലും ശാസ്ത്രസമൂഹത്തിനിടയിലും കൂണിന്റെ പ്രാധാന്യം വളരെ വലുതാണ്. വാണിജ്യാടിസ്ഥാനത്തിൽ കൃഷിചെയ്യപ്പെടുന്ന ഏറ്റവും പ്രധാനമായ ഒരു കൂൺ വിഭാഗമാണ് ചിപ്പിക്കൂൺ. ശാസ്ത്രീയമായി പ്ലൂറോട്ടസ് എന്ന ജനുസ്സിൽ ആണ് ചിപ്പിക്കൂൺ ഉൾപ്പെടുന്നത്. പൊതുവെ 18 മുതൽ 30°C വരെയുള്ള താപനിലയാണ് ചിപ്പിക്കൂൺ കൃഷിക്ക് അഭികാമ്യം, എന്നിരുന്നാലും പ്ലൂറോട്ടസ് ജനുസ്സുകളിൽ തന്നെ ഓരോ ഇനം കൂണുകളുടെയും വളർച്ചയ്ക്കായി നിർദ്ദിഷ്ടമായ താപനിലയും ഈർപ്പവും അത്യന്താപേക്ഷിതമാണെന്ന് പഠനങ്ങൾ വ്യക്തമാക്കുന്നു. കേരളത്തിന്റെ ഉഷ്ണമേഖലാ കാലാവസ്ഥ ചിപ്പിക്കൂൺ കൃഷിക്ക് അനുയോജ്യമാണ്. കേരള കാർഷിക സർവകലാശാലയുടെ വിള പരിപാലന മൂറുകൾ (2016) പ്രകാരം കേരളത്തെ സമുദ്രതീരപ്രദേശം മുതൽ പാലക്കാട് സമതലപ്രദേശങ്ങൾ വരെയുള്ള അഞ്ച് കാർഷിക പരിസ്ഥിതിമേഖലകളായി തരംതിരിച്ചിരിക്കുന്നു. ഈ അഞ്ച് മേഖലകളിലും തിരഞ്ഞെടുത്ത അഞ്ച് ചിപ്പിക്കൂണുകൾ കൃഷിചെയ്ത് ഓരോ മേഖലയ്ക്കും അനുയോജ്യമായ കൂൺ ഇനത്തെ കണ്ടെത്തുക എന്നതാണ് പഠനോദ്ദേശ്യം. അതോടൊപ്പം അവയുടെ ജൈവരാസഘടന, ഔഷധഘടന, ഉപഭോക്തൃസ്വീകാര്യത കൂടാതെ സംഭരണ കാലയളവും പഠനവിധേയമാക്കി.

പ്ലൂറോട്ടസ് ഫ്ലോറിഡ, പ്ലൂറോട്ടസ് ജാമൊർ, ഹിപ്സിസൈഗസ് അൾമാരിയസ്, പ്ലൂറോട്ടസ് സാജോർ-കാജു, പ്ലൂറോട്ടസ് സിട്രിനോപിലിയേറ്റസ് എന്നിവയാണ് പഠനവിധേയമാക്കിയ അഞ്ചിനം ചിപ്പിക്കൂണുകൾ.

അഞ്ചിനം ചിപ്പിക്കൂണുകളുടെയും സ്റ്റോറഫലങ്ങളെ കുറിച്ച് വിശദമായ പഠനം നടത്തുകയുണ്ടായി. തുവെള്ള നിറത്തിലുള്ള സ്റ്റോറഫലങ്ങൾ പ്ലൂറോട്ടസ് ഫ്ലോറിഡ ഉല്പാദിപ്പിച്ചപ്പോൾ

ഹിപ്പിസൈഗസ് അശ്മാരിയസ് വെള്ള നിറത്തിലുള്ള, താരതമ്യേന വലിപ്പം കുടിയ സ്റ്റോറഫലങ്ങൾ ഉല്പാദിപ്പിച്ചു. പ്ലൂറോട്ടസ് ജാമൊനിന്റെ പിങ്ക് നിറത്തിലുള്ള സ്റ്റോറഫലങ്ങളുടെ തണ്ടുകൾ വളരെ നീളം കുറഞ്ഞവയായിരുന്നു. ചാരനിറത്തിലുള്ള സ്റ്റോറഫലങ്ങൾ ഉല്പാദിപ്പിച്ച പ്ലൂറോട്ടസ് സാജോർ-കാജുവിന്റെ തണ്ടുകൾ താരതമ്യേന നീളം കൂടുതൽ ഉള്ളവയായി കാണപ്പെട്ടു. സ്വർണനിറത്തിലുള്ള, താരതമ്യേന വലിപ്പം കുറഞ്ഞ നിരവധി സ്റ്റോറഫലങ്ങളായിരുന്നു പ്ലൂറോട്ടസ് സിട്രിനോപിലിയേറ്റസിന്റെ പ്രത്യേകത.

വയേക്കാൽ, റബർ അറക്കപ്പൊടി എന്നീ രണ്ട് മാധ്യമങ്ങളിൽ ചിപ്പിക്കുൺ കൃഷി ചെയ്ത് ഓരോ മാധ്യമത്തിലും അഞ്ചിനം ചിപ്പിക്കുണുകളുടെ വളർച്ചയും വിളവും നിരീക്ഷിച്ചു. മൂന്നു വിളവെടുപ്പിൽ നിന്നും താരതമ്യേന കുറഞ്ഞ വിളവായ ശരാശരി 954.66 ഗ്രാം വയേക്കാലിൽ നിന്നു ലഭ്യമായപ്പോൾ റബർ അറക്കപ്പൊടിയിൽ നിന്നും ശരാശരി 1306.40 ഗ്രാം വിളവ് ലഭിച്ചു. റബർ അറക്കപ്പൊടി മാധ്യമത്തിൽ കുണിന്റെ കാലാവധി ശരാശരി 100.80 ദിവസങ്ങളായിരുന്നു, മറുവശത്ത് വയേക്കാൽ മാധ്യമത്തിൽ ശരാശരി 60.93 ദിവസത്തിൽ മുഴുവൻ വിളവും ലഭിക്കുകയുണ്ടായി. മേല്പറഞ്ഞ രണ്ട് മാധ്യമങ്ങളും ചിപ്പിക്കുൺ കൃഷിയ്ക്ക് ഉത്തമമാണെന്ന് ഈ പഠനം തെളിയിക്കുന്നു. അതിനാൽ ലഭ്യതയുടെയും ലാഭത്തിന്റെയും അടിസ്ഥാനത്തിൽ രണ്ട് മാധ്യമങ്ങളും തെരഞ്ഞെടുക്കാം.

കേരളത്തിലെ അഞ്ച് കാർഷിക പരിസ്ഥിതി മേഖലകളിലും കർഷകരുടെ കുൺപുരയിൽ അഞ്ചിനം ചിപ്പിക്കുണുകളും കൃഷി ചെയ്യുകയുണ്ടായി.

ഹിപ്പിസൈഗസ് അശ്മാരിയസ് ആണ് സമുദ്രതീര പ്രദേശങ്ങളിലും (1029.96 ഗ്രാം), മലയടിവാരങ്ങളിലും (927.33 ഗ്രാം), ഉയർന്ന മലകളിലും (1233.00 ഗ്രാം) താരതമ്യേന കൂടുതൽ വിളവ് നല്കിയത്. ഹിപ്പിസൈഗസ് അശ്മാരിയസിന്റെയും പ്ലൂറോട്ടസ് സ്ലോറിഡയുടെയും മികച്ച വിളവിന്റെ കാരണം താരതമ്യേന വലിപ്പം കുടിയ സ്റ്റോറഫലങ്ങളുടെ ഉല്പാദനം, സ്റ്റോറഫലങ്ങളിലെ ഉയർന്ന

ഈർപ്പം, എന്നിവയോടൊപ്പം വിശാലമായ ശ്രേണിയിലുള്ള താപനിലയിലും ഈർപ്പത്തിലും പൊരുത്തപ്പെട്ട് വളരാൻ കഴിയുന്നതുമാണ്. മലയടിവാരങ്ങളിൽനിന്നും (823.33 ഗ്രാം), പാലക്കാട് സമതലപ്രദേശങ്ങളിൽ നിന്നും (1038 ഗ്രാം) ഉയർന്ന വിളവ് പ്ലൂറോട്ടസ് ജാമൊറിൽ നിന്ന് രേഖപ്പെടുത്തി. സാധാരണ വേനൽക്കാലത്തെ കൃഷിയ്ക്ക് അനുയോജ്യമായ പ്ലൂറോട്ടസ് ജാമൊറിൽ നിന്ന് താരതമ്യേന അധികം വിളവ് ലഭിക്കുവാൻ കാരണം ഈ രണ്ട് കാർഷിക പരിസ്ഥിതി മേഖലകളിലെ ഉയർന്ന താപനിലയും കുറഞ്ഞ ഈർപ്പ വുമായാണ് മനസ്സിലാക്കുന്നു.

ജൈവരാസഘടന പഠനത്തിൽ നിന്നും ചിപ്പിക്കുണുക്കൾ വളരെയധികം പോഷകമൂല്യമുള്ള ഒരു ഭക്ഷണ വിഭാവമാണെന്ന് മനസ്സിലാക്കുന്നു. കൂടുതൽ മാംസ്യം, ജലാംശം, നാരുകൾ, അമിനോ അമ്ലം എന്നിവയോടൊപ്പം കുറഞ്ഞ കൊഴുപ്പ്, ധാന്യകം എന്നിവ ചിപ്പിക്കുണുക്കളെ മറ്റു ഭക്ഷണപദാർഥങ്ങളിൽ നിന്ന് വേറിട്ട് നിർത്തുന്നു. മറ്റു പച്ചക്കറികളിലും, പഴവർഗങ്ങളിലും അടങ്ങിയിട്ടുള്ളതിനെക്കാൾ കൂടുതലായ അളവിൽ ധാതു ലവണങ്ങളും ചിപ്പിക്കുണിയിൽ ഉണ്ട്. സോഡിയം : പൊട്ടാസിയം കാനപ്പെട്ടത് 1:3 എന്ന അനുപാതത്തിലാണ്. ഇങ്ങനെയുള്ള ആഹാര പദാർഥങ്ങൾ രക്ത സമ്മർദ്ദം കുറയ്ക്കുന്നതിനും ഹൃദ്രോഗങ്ങൾക്കെതിരെയും ശുപാർശ ചെയ്യപ്പെടുന്നു. ഔഷധ ഘടനാവിശകലനത്തിൽ നിന്നും ചിപ്പിക്കുണുക്കളിൽ വിവിധങ്ങളായ സംയുക്തങ്ങൾ അടങ്ങിയിട്ടുണ്ടെന്ന് നിരീക്ഷിച്ചു. ശരീരത്തിൽ മുഴുകൽ ഉണ്ടാകാതിരിക്കാൻ സഹായിക്കുന്ന ബീറ്റ- ഗ്ലൂക്കോസ്, ആന്റി ഓക്സിഡന്റായ ബീറ്റ കരോട്ടീൻ, കോളസ്ട്രോൾ കുറയ്ക്കാൻ സഹായിക്കുന്ന ലോവസ്റ്റാറ്റിൻ എന്നിവ പ്ലൂറോട്ടസ് ജാമൊറിൽ താരതമ്യേന കൂടുതലായി കാണപ്പെട്ടു.

ചിപ്പിക്കുണുക്കളുടെ ഉപഭോക്തൃസ്വീകാര്യത പഠിക്കു വാനായി അവയിൽ നിന്നും വിഭവം തയ്യാറാക്കി നിറം, മണം, രുചി, പാചക നിലവാരം, സ്വീകാര്യത തുടങ്ങിയ മാനദണ്ഡങ്ങളുടെ

അടിസ്ഥാനത്തിൽ തരംതിരിച്ചു. മറ്റു നാല് ഇനങ്ങളെയും അപേക്ഷിച്ച് ഹിപ്ലിസൈഗസ് അൾമാരിയസ് ഉപഭോക്തൃസ്വീകാര്യതയിൽ മുന്നിട്ട് നിൽക്കുന്നതായി കണ്ടെത്തി.

അഞ്ചിനം ചിപ്പിക്കുണുക്കളും എത്രനാൾ കേടുകൂടാതെ ശേഖരിക്കാം എന്നു നിരീക്ഷിച്ചു. പ്ലാസ്റ്റിക് കവറുകളിൽ മുദ്ര ചെയ്ത് മുറിയിലെ താപനിലയിലും ($26 \pm 2^\circ\text{C}$), ശീതീകരിച്ച നിലയിലും (15°C) വച്ചു, ദൈനംദിനം കാഴ്ചയിലും, നിറത്തിലും, മണത്തിലും ഉണ്ടാകുന്ന മാറ്റം നിരീക്ഷിച്ചു. പ്ലൂറോട്ടസ് ജാമൊർ, പ്ലൂറോട്ടസ് സാജോർ-കാജു എന്ന ഇനങ്ങൾ വിളവെടുപ്പിനു ശേഷം രണ്ടുനാൾവരെ മുറിയിലെ താപനിലയിലും, മൂന്നുനാൾവരെ ശീതീകരിച്ച നിലയിലും കേടുകൂടാതെ സൂക്ഷിക്കാമെന്ന് കണ്ടെത്തി.