www.ThePharmaJournal.com

The Pharma Innovation



ISSN (E): 2277-7695 ISSN (P): 2349-8242 NAAS Rating: 5.23 TPI 2023; SP-12(9): 733-739 © 2023 TPI www.thepharmajournal.com Received: 16-07-2023

Accepted: 22-08-2023

R Kalpana Devi Department of Plant Pathology, TNAU, Coimbatore, Tamil Nadu, India

G Thiribhuvanamala

Department of Plant Pathology, TNAU, Coimbatore, Tamil Nadu, India

K Angappan

Department of Plant Pathology, TNAU, Coimbatore, Tamil Nadu, India

A Bharani

Department of Environmental Science, TNAU, Coimbatore, Tamil Nadu, India

M Thirunavukkarasu

Department of Agronomy Veterinary and Animal Sciences, TNAU, Coimbatore, Tamil Nadu, India

T Praveen

Department of Plant Pathology, TNAU, Coimbatore, Tamil Nadu, India

DVS Chakradhar Reddy

Agro-Climate Research Center, TNAU, Coimbatore, Tamil Nadu, India

K Ganesh Sarvanan Department of Plant Pathology, TNAU, Coimbatore, Tamil Nadu, India

Corresponding Author: G Thiribhuvanamala Department of Plant Pathology, TNAU, Coimbatore, Tamil Nadu, India

Exploitation and domestication of wild *Pleurotus* spp. from natural habitat of Coimbatore regions

R Kalpana Devi, G Thiribhuvanamala, K Angappan, A Bharani, M Thirunavukkarasu, T Praveen, DVS Chakradhar Reddy and K Ganesh Sarvanan

Abstract

Mushroom production has expanded globally due to its nutritional and functional properties. India habituate with diverse mushroom flora, albeit only half of these have been characterized yet. Wild mushrooms are natural source with nutritional rich food. In India due to its diverse climatic conditions, many types of mushrooms are found in the wild, a survey was conducted in the forest regions of Coimbatore District *viz.*, Siruvani, Sadivayal, Velliankadu and TNAU Botanical Garden and about six isolates of *Pleurotus* mushroom were collected. The isolates were identified morphologically on genus level and characterized molecularly through ITS 1 and ITS 4 sequencing *viz.*, *Pleurotus pulmonarius*, *Pleurotus djamor* 1, *Pleurotus djamor* 2, *Pleurotus djamor* 3, *Pleurotus djamor* 4, and *Pleurotus djamor* 5. Among the isolates, *P. djamor* 2 performed well in all aspects of yield parameters *viz.*, typical phenotypic character (a big white pileus with a wavy edge and a thin or rudimentary stipe), economic yield (482.7 g/ 500 g substrate) and bio-efficiency per cent (96.3 per cent). From the current study, the cultivation of *P. djamor* 2 could be suitable for commercial cultivation because of its distinct phenotypic character and bio-efficiency of 96.3 per cent.

Keywords: Mushrooms flora, Cultural characterization, Molecular characterization, Pleurotus sp.

Introduction

Mushrooms are edible macro-fungi, with unique fruiting body, can either be hypogenous or epigenous and have been regarded as a nutrient-dense food and a beneficial supplement to the diet. This nutritionally rich macrofungi are classified as a third food kingdom by the US Department of Agriculture and are usually pertained as "white vegetables and forgotten sources of nutrients" (Feeney *et al.*, 2014) ^[15]. Mushrooms are considered as the higher class flora among the fungal family and have been used as food and medicine since ancient times (Chang and Miles. 2004) ^[12]. The mushroom hunting embarked in India four decades ago. Around 1,40,000 species of mushroom is estimated to be found on earth, but only 10% (14,000 approximately) are known so far, of which around 2,200 species are identified as edible mushrooms. Out of which only 650 species have been widely exploited, cultivated, and consumed for health and medical applications. For instance, there are 850 native mushroom species in India alone, and researchers have looked at their nutritional and ethnomedical benefits as well as potential usage in food and pharmaceutical products (Thatoi & Singdevsachan, 2014)^[36]. So far, about 1,105 to 1,208 species of mushrooms belonging to 128-130 genera have been documented. Among the 300-315 species belonging to 75-80 genera are reported to be edible (Thiribhuvanamala et al., 2011)^[37]. India is haunt with manifold of mushroom flora accounting for one portion in three of world's fungal diversity, albeit only half of these have been characterized to yet (Manoharachary et al., 2005) ^[26]. Pleurotus spp. a Oyster mushroom very popular (Adejoye et al., 2006)^[17] and ranks second (19%) among mushrooms worldwide. There are over 70 species of Pleurotus has been discovered. A few Oyster mushrooms P. ostreatus, P. florida, P. eryngii, and P. sajor-caju have aroused the interest of fact finders in the quest for therapeutic metabolites, and have long been utilized as remedies in many regions of the world to cure a range of diseases such as coronary-artery disease, cancer, and rheumatoid arthritis (Jayakumar et al., 2009; Mohamed & Farghaly, 2014) ^[21, 28]. *Pleurotus* mushrooms are widely known for their ability to degrade lignocellulosic waste and produce palatable basidia with high nutraceutical and organoleptic properties, as well as vitamins, minerals, and some necessary amino acids. (Musieba et al., 2013) [30].

Pleurotus ostreatus also exhibits antibiotic, antitumoral (Al-Saffar et al., 2020)^[5], antioxidant, anti-inflammatory and antimicrobial properties (Bains et al., 2021)^[8]. Many species of *Pleurotus* have been used in the field of food industry, pharmacology, environmental science, and nutrition (Chaurasia et al., 2020)^[39]. Pleurotus has potential to lessen cholesterol levels (Hossain et al., 2003) [20]. The whole fruiting body of mushroom is also an excellent fount of lignin and phenol degrading enzymes (Fountoulakis et al., 2002) ^[16]. Pleurotus is also employed as a biosorbent in industry (Tsioulpas et al., 2002; Barros et al., 2007) [38, 9]. Since thousands of years ago wild mushrooms have been collected and consumed by people as they are a natural resource with abundance of nutritious value (Khaund & Joshi, 2013)^[23]. Researchers have worked on wild mushrooms and have reported approximately 2,000 edible mushroom species from throughout the world, including 283 edible species from India (Adhikari et al., 2005; Purkayastha and Chandra, 1985) [1, 31] out of which some are cultivated.

As these wild mushrooms are not only exploited in terms of edibility but also in various other means, The Department of Plant Pathology, Centre for Plant Protection Studies, Tamil Nadu Agricultural University, Coimbatore, India has contributed to the development and domestication of six species of *Pleurotus* for commercial utilization. However, the understanding of the pharmacological and nutritional properties of mushrooms, more command and consumer desire for diverse species of mushrooms among people and farmers who are encouraged to harvest wild mushrooms for use. With the referred information, survey was conducted in forest region of Coimbatore to exploit the wild mushrooms for commercial utilization.

Materials and Methods

Collection, Isolation and morphological identification of wild mushrooms: The wild mushroom flora was collected from the forest regions of Coimbatore, Tamil Nadu (Siruvani, Sadivayal, Velliankadu and TNAU Botanical Garden). The collected specimens were brought to the lab and processed for detection. During the collection, the ecological and morphological characters of macro-fungi were observed and photographed for detection. (Kaya, 2005) [22]. The wild mushrooms were identified based on the various morphological characters such as mycelial growth, stipe length, stipe diameter and pileus diameter. The sporocarp were placed in sterilized tissue paper for evaporation of moisture for 2-3h. A small piece of inner tissue of the fresh basidiocarp was aseptically cut. The tissues were surface sterilized and placed on Potato Dextrose Agar medium (PDA) at equidistance in triangular position. The Petri plates were incubated at ambient temperature at 20-24 °C for 7-10 days for emergence of mycelial growth. The pure culture was maintained on PDA slants at 4±1 °C in a refrigerator for further studies.

Molecular characterization

The methodology used for extraction of total genomic DNA described by Muruke *et al.*, $(2002)^{[19]}$ with slight moderation. The genomic DNA was extracted directly from five days old fungal culture plate. The mycelium was macerated by adding 1-2ml of 2×CTAB buffer (hexadecyl trimethyl ammonium bromide) using sterilized pestle and mortar. 750µl of the macerated sample was taken in 1.5ml microfuge tubes and

incubated at 65 °C for 10 mins. After incubation, 750µl mixture of Phenol: Chloroform: Isoamyl alcohol (25:24:1) was added and centrifuged at 10,000 rpm for 10 mins. The supernatant was collected and equal amount of isopropanol was added to the macerate sample and incubated at -20 °C for 30-45 minutes or overnight incubation for DNA precipitation. The precipitated samples were centrifuged at 12,000 rpm for 10 minutes to pellet the nucleic acids and then washed the content using 70% ethanol and again centrifuged at 5,000 rpm for 5 mins. The pelleted DNA was dried at 37 °C and the DNA was re-suspended in 50µl of sterile water.

The genomic DNA extracted from the pure cultures were used for PCR analysis. The universal primers ITS 1 (5'-TCC GTA GGT GAA CCT GCG G-3') and ITS 4 (5'-TCC TCC GCT TAT TGA TAT GC-3') were used to amplify the ITS of ribosomal DNA. The PCR reaction was performed in a total reaction mixture of 40 µl containing 20 µl of master mix, 8 µl of distilled water, 4 µl each ITS 1 and ITS 4 primers and 4 µl of genomic DNA. The PCR conditions was setup with the following 35 cycles of 5 mins initial denaturation (95 °C), 1 min of denaturation (95 °C), 1 min of annealing (58 °C), 1 min of extension (72 °C) and 10 mins of final extension (72 °C) with the lid heating option at 110 °C. After amplification, 5 µl aliquot of each resultant PCR product was subjected to electrophoresis on 1.2% agarose gel. PCR products were resolved by running at 75 volts for 45 minutes. The electrophoresed gels were visualized under UV light and imaged using gel documentation system. PCR products of the DNA samples were sequenced using Sanger sequencing method (Sanger et al., 1977) [33] at Syngenome Private, Coimbatore. ITS sequences obtained were compared with other reported Pleurotus ITS nucleotide sequences in GenBank database using nucleotide BLAST (Basic Local Alignment Search Tool) search accessible through NCBI (National Centre for Biotechnology Information). Accession numbers were obtained by submitting the sequence at NCBI database.

Performance testing of wild isolates of *Pleurotus*

The yield performance of mushrooms was tested at the Mushroom Research Laboratory, Department of Plant Pathology, Tamil Nadu Agricultural University, Coimbatore. The ruling variety P. florida (PF) was used as standard isolate for comparison. The spawn was prepared using sorghum substrate and utilized for bed preparation. The cylindrical beds were prepared with paddy straw and sorghum-based spawn (500 g substrate/bed; 150 g spawn/bed). The beds were placed in cropping rooms made of thatched shed, where temperature of 23-30 °C and relative humidity of 80% was maintained throughout the cropping period. The beds were placed with a hanging rope system in thatched shed. The yield parameters including Days for Spawn Run (DFSR), Days for Pinhead emergence (DFPF), Days for First Harvest (DFFH), total yield (g per 500 g substrate) and the pest and diseases were recorded. The bio efficiency (BE) of mushroom was calculated by using the formula described by Chang and Miles (1989)^[13] as:

Biological Efficiency (%) = $\frac{\text{Fresh weight of mushroom}}{\text{Weight of Air-dried substrate}} \times 100$

The experiment was designed with three replications and four bags in each replication for each treatment.

Results and Discussion

Collection of wild mushroom flora: About 7 wild mushroom flora viz., Mushroom Isolate 1, Mushroom Isolate 2, Mushroom Isolate 3, Mushroom Isolate 4, Mushroom Isolate 5 and Mushroom Isolate 6 were collected from Siruvani, Sadivayal (Siruvani), Velliankadu (Karamadai), and TNAU Botanical Garden. The mushrooms are classified at the genus level based on the morphological characteristics of the basidiocarp produced. Rajarathnam et al. (1987) [32] characterized fruiting bodies of *Pleurotus* by an eccentric stalk, that might be short, long, or even missing; annulus and volva are absent; resemble flower petals may be clustered or separated. The pileus opens out like an oyster shell, far away from the stipe. The species of *Pleurotus* appropriately named as oyster mushrooms. Size varies across species and within species when grown under varying climatic and nutritional conditions. Depending on the species, the edge may be smooth, fractured, slightly serrated, or saw-toothed. Color is the most variable character in Pleurotus mushroom. According to the findings of Anderson et al., (1973)^[6] the fruit bodies of the same single P. ostreatus species might be white, cream colored, brown to black brown, gray to dark gray, or blue in color. On this basis, the novel wild mushrooms which is employed in this study was classified as Pleurotus at the genus level

A. *Pleurotus* isolate 1: was collected from TNAU botanical garden. The mushrooms blossomed in clusters on bottle palm tree stump and produced not anise like aroma. The pileus appeared broadly outcurved, becoming flat or shallowly depressed; kidney-shaped to fan-shaped in outline or nearly round when found on the top of the logs; somewhat greasy when young and fresh; bald; white or pale brown; fading to buff; sometimes fading slowly and becoming two-toned; the margin slightly curved inward when young 2.5 to 3.8 cm. The stipe was about 1.5cm length, small, thin, lateral, showed up dull white in color, hairy to velvety in texture. The gills laterally attached; close or nearly distant; short-gills; white to faintly vellowish, cylindrical to ellipsoid spores. The spore print retained gravish white spores. The characters obtained found to be similar to the fruiting bodies observed by Garibova and Sidorova (1997)^[40] in *P. pulmonarius*. The characters observed viz stipe 4-9 cm in diameter, tongueshaped, convex, white or sometimes greyish or pale yellow; descending gills; and a cylindrical, white, smooth eccentric or lateral column of 2-4 cm long and Hilber (1997) [19] also described white and gray colors for

https://www.thepharmajournal.com

basidiomata in P. pulmonarius.

- **B.** *Pleurotus* isolate 2: was grown singly on dead stump and produced mild smell. The pileus appeared light greyish white in color, 3.5cm in length, oyster shaped, broadly convex, flattened on maturity, textured smooth and glabrous. The stipe attached laterally to the pileus, Short or rudimentary 1.0 cm, short stipe base, Light greyish white. Gills were short, separable, close, white, free with smooth edges. The Spore print had Creamy with cylindrical spores
- C. *Pleurotus* isolate 3: was collected from Sadivayal, Siruvani. The mushrooms occurred as single/connate and had two sporophores. The mushrooms were found on decayed stump and produced mild smell. The pileus showed up light whitish cream and length of 5.2cm, Its shape appeared as cluster of flowers, soft to leathery, smooth surface, smooth margin. The stipe laterally attached, 1.5cm and creamy white in color. The gills appeared decurrent, separable, crowded, creamy white with smooth edges. The spore print had light creamy white spores
- **D.** *Pleurotus* **isolate 4:** occurred in groups on *Cassia fistula* tree trunk with bland smell (Velliankadu, Karamadai). The pileus measured about 4 to 5.2 cm, Fan shaped to oyster like, smooth surface, smooth margin becomes wavy at maturity. Surface textured soft to leathery. The stipe laterally attached to the cap, 1 to 1.5cm and showed up light creamy white. The gills appeared prominent, separable decurrent, creamy white and gill edges were smooth. It showed creamy white spores on spore print.
- **E.** *Pleurotus* isolate 5: The mushroom appeared single/connate with two sporophore and found on decayed stump with mild smell. The entire fruiting body was creamy white in color. The pileus was around 5.2 cm, Fan shaped to oyster like; smooth surface and leathery margin. The stipe found attached laterally to the cap, 1.5cm in length. The gills appeared prominent, decurrent with smooth gill edges. The spore print had light creamy white spores.
- **F.** *Pleurotus* **isolate 6:** The mushrooms were light pink in color, spatula shaped pileus, fleshly to leathery, soft, 6 to 7.5 cm, oyster to flower like curled shape, wavy margin scales absent. Stipe is pink and short, 2 to 3 cm base thin and fibrous. Gills are crowded, easily separable, running along stipe, gill pale pink, gill edges smooth, oval to cylindrical basidiospores.



Fig 1: Collection of mushrooms and their habitat (A) *Pleurotus* isolate 1, (B) *Pleurotus* isolate 2, (C) *Pleurotus* isolate 3, (D) *Pleurotus* isolate 4, (E) *Pleurotus* isolate 5 and (F) *Pleurotus* isolate 6

Isolation and cultural characteristics of wild *Pleurotus* isolates

Phenotypic characters on the mycelial growth of *Pleurotus* isolate 1, *Pleurotus* isolate 2 *Pleurotus* isolate 3, *Pleurotus* isolate 4, *Pleurotus* isolate 5, *Pleurotus* isolate 6 and *P. florida* were observed on PDA medium. The mycelia of the isolates appeared as follows *Pleurotus* isolate 1 produced cottony white, fluffy and dense strands of mycelium. *Pleurotus isolate 2* appeared filiform, flat, filamentous,

creamy white with concentric zonation, *Pleurotus isolate 3* appeared thick, irregular growth, raised, white with undulated margin and slow growing. *Pleurotus isolate 4* appeared thick, white, irregular growth with undulated margin. *Pleurotus isolate 5* appeared thin, filiform, white with concentric zonation, *Pleurotus* isolate 6 appeared thin, white, flat surface, filiform, filamentous with concentric zonation and *P. florida* produced thick strand of mycelium as they produce number of mycelial branches from one end.



Fig 2: Fungal cultures - (A) *Pleurotus* isolate 1, (B) *Pleurotus* isolate 2, (C) *Pleurotus* isolate 3, (D) *Pleurotus* isolate 4, (E) *Pleurotus* isolate 5 and (F) *Pleurotus* isolate 6

Molecular characterization of wild *Pleurotus* isolates

The confirmation of different *Pleurotus* isolates were performed through molecular characterization. (Bruns *et al.*, 1991; Hibbett, 1992) ^[11]. A high molecular weight band was visualized in an agarose gel. Genomic DNA was used to amplify the ITS 1-5.8S-ITS 2 rDNA sequence region using the primer pair ITS 1 and ITS 4. PCR fragments of the 680 bp fragment were visualized as a single band on an agarose gel stained with ethidium bromide Shnyreva *et al.*, (2012) ^[35] proposed that the ITS primers generate 680 bp bands for *Pleurotus* isolates. No DNA template was added to the control reactions. The BLAST search analysis of ITS 1-5.8S-ITS2 region of *Pleurotus* isolate 1 matched with that of *Pleurotus*

pulmonarius at 92.95%, Pleurotus isolate 2 matched with that of Pleurotus djamor 1 at 83.97%, Pleurotus isolate 3 matched with that of Pleurotus djamor 2 at 86.83%, Pleurotus isolate 4 matched with that of Pleurotus djamor 3 at 99.54%, Pleurotus isolate 5 matched with that of Pleurotus djamor 4 at 99.54% and Pleurotus isolate 6 matched with that of Pleurotus djamor 5 at 99.54% identified in the database. The sequences were submitted to NCBI and given Gen bank Accession number viz., Pleurotus pulmonarius OR083329, Pleurotus djamor 1 OR086097, Pleurotus djamor 2 OR086099, Pleurotus djamor 3 OR086100, Pleurotus djamor 4 OR086098 and Pleurotus djamor 5 OR086095.



Fig 3: Amplified ITS products (using primer ITS-1 and ITS-4) of 6 *Pleurotus* isolate: L1- 1kb ladder, L2-Negative control, L3- *P. pulmonarius*, L4- *P. djamor* 1, L5- *P. djamor* 2, L6- *P. djamor* 3, L7- *P. djamor* 4 and L8- *P. djamor* 5.

Yield performance of wild isolates of *Pleurotus*

The results on number of days required by the wild isolates for Spawn Running, Pinhead Emergence and first blossom of the fruiting body illustrated in the Figure.5 Spawn Running is the expansion and colonization of fungal hyphae throughout the substrate. The days taken by the *Pleurotus* isolates to cover the entire substrate was around 17 to 20 days; *P. pulmonarius* resulted maximum number of days for running (20 days) similar results was reported by Akinfemi. A & Ogunwole. O.A (2012)^[3] that *P. pulmonarius* completes spawn running on paddy straw within 22 days. The faster mycelial run was seen in P. djamor 2 (17 days). According to Chang and Miles (2004) ^[12], The difference in the number of days taken by the spawn to colonize a given substrate depends on the fungal isolate, growth conditions, and substrate type. This might be affected by the chemical composition and Carbon to Nitrogen ratio (C:N) of the substrate used (Bhatti et al., 1987)^[10]. The period of pin-head formation varied among the isolates ranging from 20 to 24 days after spawn seeding. Pin-head formation occurred quickly in P. djamor 2 (20 days), followed by P. djamor 3 (21 days); while it took relatively longer time in P. pulmonarius (24.4 days). The invasion of mycelial growth on the substrate arose pinhead formation. The time required for the formation of pin-heads is comparable with other similar studies, Ahmed (1998)^[2] reported that pin-head formation of oyster mushrooms grown on various substrates took between 23 and 27 days from spawning, while Fan et al., (2000)^[14] found that it took 20-23 days. On the other hand, Shah et al., (2004) [34] found that pinheads appeared in about 6 days in oyster mushrooms. The time required for maturity of fruiting bodies varied from 23 days (for P. djamor 2) to 27.6 days (for P. pulmonarius). On comparison with ruling variety, P. djamor 2 showed lesser number of days for spawn running, pinhead emergence and

maturity that *P. florida*.

Results of the yield attributes of wild oyster mushroom grown in paddy straw substrate are presented in Table 1. Accordingly, it was found that the length of the pileus of fruiting bodies produced showed maximum length *P. djamor* 4 (12.4cm)followed by *P. djamor* 2 (6.7cm) and minimum length was recorded in P. djamor 3 (4.1cm). The width of the pileus ranged from 7.5 cm (P. *djamor* 2) to 2.7 cm (*Pleurotus pulmonarius*). The length of the stipe showed maximum length in *P. djamor* 4 (2.4cm) and minimum length in *P. pulmonarius* (0.8 cm). Rajarathnam *et al.*, (1987) ^[32] reported that the width of the fruiting bodies varies between 2 to 3 cm and 15 to 20 cm.

The largest yield among the cultivated wild isolates was recorded in *P. djamor* 2 (482.7g/500g substrate) followed by *P. djamor* 3 (424.0 g/500g substrate) and minimum was seen in *P. pulmonarius* (153.5g/500g substrate). Liang et al., (2009)^[25] reported that varying ranges of BE was seen when different lignocellulosic materials were used as substrates for cultivation of *Pleurotus* mushrooms. The bio-efficiency percentage range from 30.6% (*P. pulmonarius*) to 96.4% in *P. djamor* 2. In comparison with the cultivated variety *P. florida*, all the wild isolates cultivated showed lower yield attributes *viz.*, length and width of pileus, length of the stipe, economic yield and bio-efficiency %.



Fig 4: Graph illustrates the Days of Spawn Run, Pinhead Emergence and First Harvest of the collected wild isolates of mushrooms and *Pleurotus florida* (PF)

Isolate	Pileus		Stipe	Total Yield	Die officiency (0/)
	Length(cm)	Width(cm)	Length(cm)	(g/500 g substrate)	BIO efficiency (%)
Pleurotus pulmonarius	4.2 ^{de}	2.7 ^g	0.8 ^e	153.5 ^g	30.7 ^g
Pleurotus djamor 1	4.5°	5.6 ^d	1.0 ^d	410.0 ^d	82.0 ^d
Pleurotus djamor 2	6.7 ^b	7.5ª	1.5°	482.7 ^b	96.3 ^b
Pleurotus djamor 3	4.1 ^e	5.0 ^e	0.9 ^{de}	424.0°	84.8 ^c
Pleurotus djamor 4	12.4 ^a	6.3°	2.4 ^b	371.0 ^f	74.2 ^f
Pleurotus djamor 5	4.4 ^{cd}	4.1 ^f	1.0 ^d	390.0 ^e	78.2 ^e
Pleurotus florida (PF)	4.4 ^{cd}	7.2 ^b	2.7 ^a	530.3ª	106.0 ^a
CD	0.289	0.278	0.229	1.468	0.294
SEd	0.1341	0.1290	0.1064	0.6845	0.1341

Table 1: Phenotypic characterization of basidiocarp of Pleurotus isolates

SEd \pm : Standard error of the difference. CD (p = 0.05): Critical difference, where p = level of significance. All the observations are average of three replications.



Fig 5: Bed Cultivation of mushrooms- (A) Pleurotus florida (PF), (B) Pleurotus pulmonarius, (C) Pleurotus djamor 1, (D) Pleurotus djamor 2, (E) Pleurotus djamor 3, (F) Pleurotus djamor 4 and (G) Pleurotus djamor 5

Conclusion

The review of the literatures shows that India's unique climatic conditions have made it a natural habitat for numerous mushrooms. In recent times, several mushroom species are threatened as a result of deforestation activities, making it critical to maintain the mushroom flora in order to increase a country's food security. In this approach, the current study vividly displays the richness of mushroom flora existing in some forest regions of Coimbatore and their salvation in order to identify plausible isolate for domestication. Based on yield parameters including phenotypic character, economic yield and bio-efficiency percent P. djamor 2 showed values slightly higher variation from P. djamor 3. Many Pleurotus species produce smooth pileus with a long stipe. However, P. djamor 2 has a big white pileus with a wavy edge and a small stipe in comparison with ruling variety P. florida (PF). As a result, it is concluded that this new Pleurotus djamor 2 has typical phenotypic features that extricate from other reported Pleurotus spp., and thus, P. djamor 2 could be suitable for commercial cultivation and count on mushroom germplasm collection.

Acknowledgements

The author would like to thank Mushroom Research Laboratory, Department of Plant Pathology, Tamil Nadu Agricultural University, Coimbatore for providing facilities to conduct the research work.

Conflict of interest: The author declares no conflict of interest

References

- 1. Adhikari MK, Devkota S, Tiwari RD. Ethnomycological knowledge on uses of wild mushrooms in western and central Nepal. Our nature. 2005;3(1):13-9.
- 2. Ahmed S. Development of mushroom varieties suitable for rural level in Bangladesh. Report presented in BARC Annual Review Programme; c1998. p. 72-73.
- 3. Akinfemi A, Ogunwole OA. Chemical composition and

in vitro digestibility of rice straw treated with Pleurotus ostreatus, Pleurotus pulmonarius and Pleurotus tuber-Regium. Slovak Journal of Animal Science. 2012 Mar 31;45(1):14-20.

- Alam N, Khan A, Hossain M, Amin SM, Khan LA. Nutritional analysis of dietary mushroom *Pleurotus florida* Eger and Pleurotus sajor-caju (Fr.) Singer. Bangladesh Journal of Mushroom. 2007;1(2):1-7.
- 5. Al-Saffar AZ, Hadi NA, Khalaf HM. Antitumor activity of β -glucan extracted from Pleurotus eryngii. Indian Journal of Forensic Medicine & Toxicology. 2020 Jul 1;14(3):2493.
- Anderson NA, Wang SS, Schwandt JW. The Pleurotus ostreatus-sapidus species complex. Mycologia. 1973 Jan 1;65(1):28-35.
- Atri NS, Sharma SK, Gulati A. Study on Mycelial growth pattern of five wild Pleurotus species from North West India. American-Eurasian Journal of Scientific Research. 2012;7(1):12-5.
- Bains A, Chawla P, Tripathi A, Sadh PK. A comparative study of antimicrobial and anti-inflammatory efficiency of modified solvent evaporated and vacuum oven dried bioactive components of Pleurotus floridanus. Journal of Food Science and Technology. 2021 Sep;58:3328-37.
- Barros L, Ferreira MJ, Queiros B, Ferreira IC, Baptista P. Total phenols, ascorbic acid, β-carotene and lycopene in Portuguese wild edible mushrooms and their antioxidant activities. Food chemistry. 2007 Jan 1;103(2):413-9.
- Bhatti MA, Mir FA, Siddiq M. Effect of different bedding materials on relative yield of oyster mushroom in the successive flushes. Pak J Agri. Res. 1987;8:256– 259.
- 11. Bruns TD, White TJ, Taylor JW. Fungal molecular systematics. Annual Review of Ecology and systematics. 1991 Nov;22(1):525-64.
- Chang ST, Miles PG. Mushrooms: cultivation, nutritional value, medicinal effect, and environmental impact, 2nd Edn. CRC Press, Boca Raton; c2004.
- 13. Chang ST, Miles PG. Edible Mushrooms and their

Cultivation, 1st edition. ERC Press, Boca Raton, Florida USA; c1989. p. 345.

- 14. Fan L, Pandey A, Mohan R, Soccol CR. Use of various coffee industry residues for the cultivation of Pleurotus ostreatus in solid state fermentation. Acta Bio technological. 2000;20(1):41-52.
- Feeney MJ, Miller AM, Roupas P. Mushrooms-Biologically distinct and nutritionally unique: Exploring a "third food kingdom". Nutrition today. 2014 Nov;49(6):301.
- 16. Fountoulakis MS, Dokianakis SN, Kornaros ME, Aggelis GG, Lyberatos G. Removal of phenolics in olive mill wastewaters using the white-rot fungus Pleurotus ostreatus. Water research. 2002 Nov 1;36(19):4735-44.
- 17. Gbolagade J, Sobowale A, Adejoye D. Optimization of sub-merged culture conditions for biomass production in *Pleurotus florida* (Mont.) Singer, a Nigerian edible fungus. African Journal of Biotechnology. 2006;5(16).
- Hibbett D. Ribosomal RNA and fungal systematics. Transaction of Mycological society of Japan. 1992;33:533-556
- 19. Hilber O. The genus' Pleurotus' (Fr.) Kummer (2). O. Hilber; c1997.
- Hossain S, Hashimoto M, Choudhury EK, Alam N, Hussain S, Hasan M, Choudhury SK, Mahmud I. Dietary mushroom (*Pleurotus ostreatus*) ameliorates atherogenic lipid in hypercholesterolemia rats. Clinical and Experimental Pharmacology and Physiology. 2003 Jul;30(7):470-5.
- Jayakumar T, Thomas PA, Geraldine P. *In-vitro* antioxidant activities of an ethanolic extract of the oyster mushroom, Pleurotus ostreatus. Innovative Food Science & Emerging Technologies. 2009 Apr 1;10(2):228-34.
- 22. Kaya A. Macrofungi determined in Gölbaşı (Adıyaman) district. Turkish Journal of Botany. 2005;29(1):45-50.
- 23. Khaund P, Joshi SR. Wild edible macrofungal species consumed by the Khasi tribe of Meghalaya, India; c2013.
- 24. Khlood A, Ahmad A. Production of oyster mushroom (*Pleurotus ostreatus*) on olive cake agro-waste. Dissertation of Agricultural Science. 2005;32(1):64-70
- 25. Liang Z, Wu C, Shieh Z, Cheng S. Utilization of grass plants for cultivation of *Pleurotus citrinopileatus*. Int. Biodeterior Biodegrad. 2009;63(4):509-514.
- 26. Manoharachary C, Sridhar K, Singh R, Adholeya A, Suryanarayanan TS, Rawat S, *et al.* Fungal biodiversity: distribution, conservation and prospecting of fungi from India. Current Science. 2005 Jul 10:58-71.
- 27. Miles PG, Chang ST. Mushrooms: cultivation, nutritional value, medicinal effect, and environmental impact. CRC press; 2004 Mar 29.
- 28. Mohamed EM, Farghaly FA. Bioactive compounds of fresh and dried Pleurotus ostreatus mushroom. International journal of biotechnology for wellness industries. 2014 Mar 1;3(1):4.
- 29. Muruke MH, Kivaisi AK, Magingo FS, Danell E. Identification of mushroom mycelia using DNA techniques. Tanzania Journal of Science. 2002;28(1):115-28.
- Musieba F, Okoth S, Mibey RK, Wanjiku S, Moraa K. Proximate composition, amino acids and vitamins profile of Pleurotus citrinopileatus singer: an indigenous mushroom in Kenya. American Journal of Food Technology. 2013;8(3):200-6.
- 31. Purkayastha RP, Chandra A. Manual of Indian edible

mushrooms. New Delhi: Today and Tomorrow's Printers and Publishers; c1985. p. 41-48.

- 32. Rajarathnam S, Bano Z, Miles PG. Pleurotus mushrooms. Part I A. Morphology, life cycle, taxonomy, breeding, and cultivation. Critical Reviews in Food Science & Nutrition. 1987 Jan 1;26(2):157-223. https://doi.org/10.1080/10408398709527465
- 33. Sanger F, Nicklen S, Coulson AR. DNA sequencing with chain-terminating inhibitors. Proceedings of the national academy of sciences. 1977 Dec;74(12):5463-7.
- 34. Shah ZA, Ashraf M, Ishtiaq M. Comparative study on cultivation and yield performance of oyster mushroom (*Pleurotus ostreatus*) on different substrates (wheat straw, leaves, saw dust). Pakistan Journal of Nutrition. 2004;3(3):158-60.
- 35. Shnyreva AA, Sivolapova AB, Shnyreva AV. The commercially cultivated edible oyster mushrooms Pleurotus sajor-caju and P. pulmonarius are two separate species, similar in morphology but reproductively isolated. Russian Journal of Genetics. 2012 Nov;48:1080-8.
- Thatoi H, Singdevsachan SK. Diversity, nutritional composition and medicinal potential of Indian mushrooms: A review. African journal of biotechnology. 2014, 13(4).
- 37. Thiribhuvanamala GURUDEVAN, Prakasam V, Chandrasekar G, Sakthivel K, Veeralakshmi S, Velazhahan R, Kalaiselvi G. Biodiversity, conservation and utilization of mushroom flora from the Western Ghats region of India. In Proceedings of the 7th International Conference on Mushroom Biology and Mushroom Products (ICMBMP7), 2011, October, 155-164.
- 38. Tsioulpas A, Dimou D, Iconomou D, Aggelis G. Phenolic removal in olive oil mill wastewater by strains of Pleurotus spp. in respect to their phenol oxidase (laccase) activity. Bioresource Technology. 2002 Sep 1;84(3):251-7.
- Chaurasia V, Pal S. Application of machine learning time series analysis for prediction COVID-19 pandemic. Research on Biomedical Engineering. 2020 Oct 24:1-3.
- 40. Garibova LV, Sidorova II. Griby. Entsiklopediya prirody Rossii; c1997.