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Evaluation of Morphological Characteristics of *Pleurotus eryngii*

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ABSTRACT

Pleurotus eryngii, exhibits distinct morphological characteristics and commonly known as the king oyster mushroom. Morphological features of P. *eryngii* reveal its significance in commercial and ecological field. This research investigates the macroscopic morphology of *P. eryngii* under controlled environmental conditions. Key features of *P. eryngii* such as cap diameter, color, shape, stipe length, thickness, color, gill attachment, and its different developmental stages were evaluated thoroughly. Cultural characteristics were also evaluated by observing colony morphology, mycelial density, growth pattern, and mycelium color on malt extract agar medium. The results are useful in species identification, classification, strain selection for large scale cultivation and potential biotechnological applications of *P. eryngii*.

Keywords: *Pleurotus eryngii*, Morphology, Pileus, Stipe, Gills, Mycelium

INTRODUCTION

Macro-fungi (Mushrooms) are commonly widespread organisms on the earth that grow in a variety of natural habitats. It have been valued by humans as an important food source for centuries. Mushrooms are also widely used in the fields of pharmaceutical, cosmetic and industrial applications due to their highly valuable and beneficial bioactive compounds. Mushrooms, known as an amazing source of food, and are rich in terms of protein, fiber, vitamins, minerals and many different nutritional content, while they have low fat and carbohydrates content (Selem *et al.,* 2021).

The king oyster mushroom, scientifically known as *Pleurotus eryngii* belongs to division Basidiomycota, order Agaricales and family Pleurotaceae. (Zervakis *et al.,* 2014, Krüzselyi *et al.,* 2016). *Pleurotus eryngii* is the largest species in the *Pleurotus* genus, these *Pleurotus* genus also includes other species such as *Pleurotus ostreatus*, *Pleurotus sajor-caju*, and *Pleurotus florida* (Deora *et al.,* 2021). *Pleurotus* mushrooms, are globally recognized for their nutritional, medicinal, and ecological significance. Among them, *Pleurotus eryngii* is known for its large fruiting body, excellent shelf life, and rich umami flavor. *Pleurotus eryngii* has gained widespread attention in commercial mushroom cultivation due to its high market demand, delicious taste and adaptability to a range of substrates.

Pleurotus eryngii is regarded as one of the best superior species among the *Pleurotus* genus due to its firm and uniform texture of both pileus and stipe, outstanding culinary qualities, and comparatively extended shelf life relative to other species of *Pleurotus*. Its excellent dense consistency and desirable flavor profile enhance consumer preference and market value (Moonmoon *et al.,* 2010).

Pleurotus eryngii offers several advantages over traditional plant-based protein sources, such as a faster growth rate, a shorter developmental period, and the ability to be grown under controlled environmental conditions. As a result, *Pleurotus eryngii* holds strong potential to become high-quality protein source (Wu *et al.,* 2023).

In addition to its nutritional value, *P. eryngii* contains a wide range of metabolic content including polysaccharides, phenolics and ergothioneine, which help to its antioxidant, antimicrobial, and cholesterol-lowering properties. These qualities further increase its desirability in food, cosmetics and pharmaceutical industries. On the basis of agronomic perspective, *P. eryngii* offers sustainable production potential, as it can be cultivated on different lignocellulosic agricultural wastes. Its cultivation contributes in environmental conservation and effective recycling of agricultural waste.

Pleurotus eryngii could be suggested as a potential component for lowering cholesterol levels (Alam *et*

al., 2011) and improving insulin sensitivity (Krüzselyi *et al.,* 2016).

Pleurotus eryngii contains certain bioactive compounds that can contribute in improving human health. These compounds are triterpenoids, polysaccharides, antioxidants, low-molecular-weight proteins, immunomodulating compounds and glycoproteins. As a result of these bioactive compounds, king oyster mushroom has been seen with enhanced immune response, boost health condition, reduced cancer risk, inhibit tumor development, regulate blood sugar levels and help in the body's detoxification processes. *Pleurotus eryngii* has furnished therapeutic potential which are useful in various pharmaceutical, medicinal, and biotechnological applications (Couto and Herrera, 2006; Gregori *et al.,* 2007; Deora *et al.,* 2022).

The use of *Pleurotus* mushrooms also have promising applications in dermatology and cosmetology. Their extracts are rich in antioxidants along with anti-wrinkle, anti-aging, skin-whitening, and moisturizing properties. These extracts can be used in the formulation of various cosmetic and cosmeceutical products (Golak-Siwulska *et al.,* 2018).

Morphological characterization of mushroom is a fundamental aspect of fungal taxonomy and for its cultivation studies. It provides important insights into strain variability, developmental physiology, and its adaptability to environmental conditions. The morphology of *P. eryngii*, including various features such as pileus shape, cap color, margin, diameter, stipe length, color, gill attachment, mycelial growth, and spore print. Its development influenced by several factors, including substrate type, humidity, temperature, light exposure and pH. Consistent and well-documented morphological features analysis is essential not only for understood species identification and taxonomical position but also for selecting mushroom different strains with desirable agronomic traits.

Spore print analysis was conducted to support species identification and document features of reproductive part. Mature fruiting bodies produced a white to creamy spore deposit, consistent with known traits of the *Pleurotus* genus. The spore print also provided important diagnostic information and was used to develop pure culture for further study.

Cultural characteristics were also evaluated by observing colony morphology, mycelial density, growth pattern, and its color on malt extract agar media. The mycelium exhibited rapid and uniform growth with cottony to fluffy texture, varying in appearance depending on the nutrient composition of the medium.

In recent years, morphological studies have gained revitalized attention due to increasing interest in cultivation enhancement, strain improvement and commercial mushroom breeding. Morphological characteristics play a crucial role in resolving the marketability of the fruiting bodies, as consumer preference is also affected by size, shape, color and uniformity.

A critical component of morphological studies involves the use of spore prints, which is a diagnostic tool to observe spore color, density and arrangement. These different salient features of mushrooms play an important role in authenticating species identity and differentiating them with closely related taxa. The establishment of pure culture of mushrooms from tissue culture is crucial for maintaining its genetic stability, ensuring reproducing ability in experiments and improving controlled cultivation for large scale production. Pure culture of mushroom can help in detailed study of its growth characteristics, colony morphology and mycelial behavior under *in-vitro* conditions, which are important for both research and commercial production.

This research aims to evaluate and document the basic biological and morphological characteristics of *Pleurotus eryngii*, which are essential for taxonomy, strain improvement and optimization of cultivation techniques. By analyzing key phenotypic characteristics, we can establish correlations between morphological outcomes and cultivation conditions. Findings of the experiment are expected to increase the knowledge about the growth behavior and qualities of *P. eryngii*, which can be useful for further studies and development of improved cultivation practices of *P. eryngii*. Morphological characteristics can also enhance the understanding of strain selection strategies for this delicious, economically and nutritionally valuable species.

MATERIALS AND METHODS

This research was carried out in research laboratory, Botany Department, Govt. Nagarjuna

P.G. College of Science, is on the Great Eastern Road, near the Pt. Ravi Shankar Shukla University Campus, Amanaka, Raipur, capital of Chhattisgarh, India. The climate of Chhattisgarh is tropical. Due to its location near Tropic of Cancer and its reliance on monsoon rainfall, it is characterized by high temperatures and humidity. The morphological features and cultural characteristics of *Pleurotus eryngii* were studied and described in detail.

A. Acquisition and Sub-culturing of culture

The pure culture of *Pleurotus eryngii* used in this study were procured from the Directorate of Mushroom Research, Chambaghat, Solan. The pure culture was maintained on Malt Extract Agar (MEA) medium and it was periodically sub-cultured in every three weeks to ensure the viability. These sub cultures were then used for further experimental process.

B. Preparation of Basal Medium

Malt Extract Agar (MEA) medium was used for sub culturing *P. eryngii* pure culture. The medium was prepared by adding 20 g of malt extract powder, 20 g of dextrose, and 20 g of agar-agar in distilled water and make a final volume of one liter, and the pH was adjusted to 6.5. The MEA medium was sterilized in an autoclave at 121°C for 40 minutes at 15 psi pressure. After sterilization, the medium was poured into culture tubes and culture plates and allowed to solidify. The following day, these prepared tubes and plates were used for sub-culturing *P. eryngii*.

C. Establishment of *P. eryngii* Pure Culture

Fresh, healthy, medium-sized fruiting bodies of *P. eryngii* were harvested and surface-sterilized using 70% ethanol. The fruiting bodies were cut longitudinally using a sterilized scalpel. Tissues were taken from the upper central portion of stipe and placed aseptically on the surface of MEA medium with the help of sterilized inoculation needle. These culture tubes and culture plates were sealed in sterilized condition and incubated at 25 ± 2°C for 7 days of incubation period (Singh *et al.*, 2011).

D. Preparation of mother and sister Spawn

Wheat grain spawn was prepared by boiling the grains until they were softened but not overcooked. After boiling, the excess water was drained off, and the grains were spread out to dry for 1-2 hours. When partially dried, 0.5% calcium carbonate and 2% calcium sulfate were mixed with grains thoroughly. These grains were packed into autoclavable polypropylene (pp) bags and bottles. The openings of pp bags and bottles were closed with cotton plug, and the grains were sterilized at 15 psi for 2 hours. After sterilization, they were allowed to cool to room temperature. In a laminar airflow under aseptic conditions, the mycelium of *P. eryngii* was inoculated in these grains packets and bottles with the help of sterilized inoculation needle. The inoculated pp bags and bottles were incubated at 25 ± 2°C for 10–15 days. The appearance of white mycelial cottony growth, which completely colonized the grains indicated that the spawn was ready for use (Singh *et al.*, 2011).

E. Cultivation of *Pleurotus eryngii*

Sawdust was used as the substrate for king oyster mushroom cultivation. It was supplemented with wheat bran 10-20%, calcium sulfate 1.5%, calcium carbonate 0.5% and soaked in hot water (65-70%) for 2 hours. The substrate was filled into pp bags and sterilized in an autoclave at 15 psi for 2 hours. When bags cooled to room temperature, they were inoculated with spawns of *Pleurotus eryngii* on laminar air flow under aseptic conditions. The inoculated bags then transferred to a cropping room and incubated at a temperature range of 25 ± 2°C to allow mycelial colonization. When the substrate was fully colonized and turned completely white, the top of the bags was carefully opened. A heat shock was applied by reducing the temperature to approximately 14–16°C to trigger fruiting body formation, and fresh water was sprayed regularly to maintain 85–90% relative humidity in the cropping room (Singh *et al.*, 2011).

F. Morphological Characterization of the Mushroom

The freshly harvested mushrooms were carefully observed, recorded and documented based on macroscopic characteristics (visible to naked eyes). Spore print was collected by following the method of Senthilarasu and Kumaresan, (2018). Documented characteristics included the appearance, texture, color of fruiting bodies, spore print color, pileus color, diameter, lamellae, stipe color, length and position of the stipe. The size of pileus and stipe were measured in centimeters.

G. Study of developmental stages of mushroom

Each stage of development of *Pleurotus eryngii* was carefully observed and recorded for detailed study.

RESULTS AND DISCUSSION

Based on its unique morphological characteristics *Pleurotus eryngii* was distinguished successfully. These morphological and cultural characteristics of *Pleurotus eryngii* are presented in the next part:

1. Cultural Characteristics

The pure culture of *P. eryngii* was observed after seven days of incubation. Mycelium was milky white colored, showed radial and cottony growth, forming a smooth matted texture that fully covered the plate in 11-12 days. Mature mycelium exhibited thick cottony soothing appearance (**Figure 1**).

Table 1: Growth and cultural characters of *Pleurotus eryngii*

Sl.no.	Name of Mushroom	Radial growth in the day (cm)			Time taken for completion of growth (days)	Nature of growth
		3	5	7		
1.	*Pleurotus eryngii*	0.3	2.1	4.5	11-12	Pure milky white, thick mycelium, fluffy cottony growth, smooth margin



Figure 1: Pure culture of *Pleurotus eryngii*

2. Fruiting Body

Fruiting bodies were grown individually or also joined at their base to form small group (**Figure 2**). Fresh mushroom had a larger size soft texture, mild odor and fleshy consistency.



Figure 2: Fruiting Bag of *Pleurotus eryngii*

3. Pileus

The cap ranged from 3.4 to 8.5 cm in diameter and exhibited colors from brownish white to brown. Its shape was initially convex, but when the cap matured gradually expanding with age into a slightly depressed center, flat structure, with a soft upper surface (**Figure 3**). The cap margins were entire at young stages, enrolled at mature stage.



Figure 3: Fruiting Body of *Pleurotus eryngii*

4. Stipe

The stipe measured 2.3–5.0 cm in length and 0.8–3.2 cm in diameter, with a brownish white-colored, smooth surface. Stipe was eccentric, clavate shaped, and solid. When fresh, the stipe was long, thick, and fleshy fibrous.

Table 2: Sporophore characters of *Pleurotus eryngii*

Sl. No.	Pileus size (cm)	Stipe length (cm)	Stipe thickness (cm)	Color	Margin
1.	3.4	2.3	0.8	Brownish white	Entire
2.	3.9	2.5	1.4	Brownish white	Entire
3.	4.5	2.6	0.9	Brown	Entire
4.	4.8	4.8	3.2	Brown	Entire
5.	5.3	3.1	1.3	Brown	Entire
6.	5.6	3.2	2.2	Brown	Entire
7.	5.8	3.8	2.5	Brown	Entire
8.	6.1	4.8	3.2	Brown	Entire
9.	7.4	4.9	2.3	Brown	Entire
10.	8.5	5.0	2.3	Brown	Entire

5. Hymenium

The hymenium was fertile layer containing basidia, spore-bearing part of a mushroom and found on gills, underside region of the cap.

6. Gills

The gills were smooth, sub-decurrent, and at young stage brownish white and at mature stage light brown in color with a smooth, entire surface. The gills were crowded and closed. In mature fruiting bodies, the gills played a critical role in spore dispersal.

7. Spore Print

The spore print was milky white colored (**Figure 4**).



Figure 4: Spore print of *Pleurotus eryngii*

Table 3 (i, ii, iii): Morphological characterization of *Pleurotus eryngii*

Pileus	
Color	Brownish white to Brown
Shape	Broadly convex
Depression	Slightly depressed
Margin	Margins were entire at young stages, inrolled at mature stage
Surface of the margin	Striate
Texture of pileal surface	Smooth
Odor	Mild odor
Fruiting body attachment	Stipitate
Gills	
Color	At the young stage white and at mature stage creamish white
Spacing	Crowded
Margin	Smooth
Lamellulae	Absent
Latex	Absent
Arrangement	Free

Texture	Fleshy
Consistency	Smooth
Stipe	
Shape	Clavate
Color	brownish white-colored
Surface	Smooth
Attachment to the pileus	Eccentric
Consistency	Solid
Annulus	Absent
Volva	Absent

8. Developmental stages of *Pleurotus eryngii*

The **Developmental stages** of *Pleurotus eryngii* consists of different phases:

A. Growth of Mycelium

Pure culture of *P. eryngii* was prepared by using the central stipe portion of the fruiting body which was introduced in the culture plates containing MEA medium. The culture plate was covered with thread-like milky white colored hyphae and completely colonized the plate in 10-12 days.

B. Spawn

Mycelium of *P. eryngii* introduced into sterilized wheat grains, covered completely these grains with milky white colored mycelium. White mycelium without contamination was used as spawn for further cultivation.

C. Spawn run

Prepared spawn was introduced in sterilized substrate and rest it in cropping room at 25°C for spawn run. In spawn running process, the mycelium spread throughout the substrate filled in bag by utilizing available nutrients. When mycelium covered substrate completely and bag turned white without contamination indicated that bag was completely prepared for fruiting.

D. Primordia Formation

Temperature and humidity was triggered the formation of pin head like structures known as fruiting bodies precursors or primordial. For primordial formation mouth of bags were opened and 14-16°C temperature was maintained. Through spraying of water humidity was maintained. In 5-7 days pin head structures appeared. Primordial are tiny ball-like structures.

E. Fruiting Body Development

Pin head structures developed in mature fruiting body into 4-8 days. Fully developed fruiting body structure was made up of cap, lamellae, and stipe. Volva and annulus were absent. In 1-2 Days this fruiting body formed basidiospores for its reproduction.

F. Spore Release and Dispersal

Gills within the Basidium produced basidiospores that were spread into air, for its germination and regeneration.

DISCUSSION

The present research gives detailed description on the morphological and cultural characteristics of *Pleurotus eryngii*, a nutritionally and economically important edible oyster mushroom species. The findings revealed significant observations in both macroscopic and cultural characteristics, which are important for species identification, different strain selection, and optimization of its large-scale cultivation protocols.

The morphological characteristics analysis showed noticeable variation in pileus diameter, stipe length, and color patterns of *Pleurotus eryngii*. The pileus was initially convex, becoming funnel-shaped with maturity, and displayed inrolled margins that extended with age. The gills were creamish white and decurrent, a characteristic of typical *Pleurotus* species, supporting accurate species identification.

Cultural characteristics, including colony morphology, mycelial texture, and growth timing on MEA media, provided important insights into the adaptability and vigor of the *P. eryngii*. The mycelium was observed cottony to fluffy in texture, and milky white in color. These characteristics indicate a healthy and viable culture suitable for commercial spawn production. The successful development of spore print and pure

culture further validated the identity and reproductive capability of the *P. eryngii*.

Although morphological and cultural characteristics of *Pleurotus eryngii* have been previously studied but most reports provide only general descriptions. Some reports like Bisko *et al.*, (2022) worked on biology, morphology, and phylogeny of some strains of *Pleurotus eryngii* species. Bandura *et al.,* (2022) evaluated fruiting body yield and morphological characteristics of *Pleurotus eryngii* different strains. Dharmasena *et al.*, (2025) highlighted the morphology of *Pleurotus eryngii* cultivated on invasive grass species.

The present study offers a detailed and systematic characterization of *P. eryngii* macroscopic features along with cultural characteristics. Such detailed documentation contributes to improved strain identification, comparative taxonomy and the selection of better isolates for large scale cultivation to improve yield.

CONCLUSION

For all that *Pleurotus eryngii* is widely cultivated at the global level, its large-scale organized cultivation has not been well established in Chhattisgarh, and scientific reports of *Pleurotus eryngii* strains remains limited. The present study provides the first detailed morphological and cultural characterization of a *Pleurotus eryngii*. Comprehensive evaluation of macroscopic features, along with growth behaviour on culture media, establishes standardized attributes under controlled laboratory conditions. The generation of region-specific baseline data strengthens strain authentication and supports the selection of *Pleurotus eryngii* strains that can locally adapted for sustainable mushroom cultivation in Chhattisgarh.

Overall, this study contributes to the foundational understanding of *P. eryngii*'s biological and cultural traits and serves as a reference for mushroom growers, researchers, and mycologists involved in mushroom taxonomy, breeding, and production.

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