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BIOLOGICAL AND MOLECULAR CHARACTERIZATION OF PLEUROTUS SP

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# Biological and Molecular Characterization of Pleurotus SP

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Abstract – Mushrooms have restorative just as nutritive worth and broadly utilized as human food from the time interminable. To decide the hereditary variety among Pleurotus types of mushroom utilizing morphological and arbitrary enhanced polymorphic DNA (RAPD) markers, around seven distinct species were gathered. Five species, naming Pleurotus platypus (P-6), Pleurotus flabelatus (P-7), Pleurotus florida (P-17), Pleurotus ostreatus (P-19) and Pleurotus sajor-caju (P-56) were from Canada and two Pleurotus warm-stram (P-9) and Pleurotus eryngii (P-16) from Philpines. Seven diverse morphological attributes that is, mycelial development (mm), cap measurement (cm), complete yield (kg), dampness substance (%), debris substance (%), nitrogen substance (%) and protein content (%) were recorded. The dendrogram dependent on morphological information isolated seven species in bunch 'A' and 'B' having four and three species, individually. The dendrogram dependent on RAPD examination created 3 groups 'A', 'B' and 'C'. Out of 14 irregular preliminaries, the greatest polymorphism was seen by groundworks OPL3 (72.70 %) and OPL11 (70%).

Keywords: Biological, Molecular, Pleurotus Sp

# INTRODUCTION

As a result of its nutritive and restorative properties, mushrooms have been widely used as food since antiquity. Mushrooms are produced in a regulated normal environment, and they have been widely utilised as food since antiquity (Manzi et al., 2011). With the passage of time, there has been a shift in the way people think about mushrooms and their nutritional and medicinal value (Cheung, 2012). Even while mushrooms have a low caloric and fat content, they are high in protein and fibre (Manzi et al., 2013). As a consequence of the high nutritional value of mushrooms, an amazing advancement has happened in the replication of its new strains, which has resulted in the discomfort of their unique verification being obsolete (Staniaszek et al., 2012). Furthermore, the confirmation of genotypic character is required in order to create appropriate natural assessments regarding individuals, structure, and exceptional methods inside and across species, among other things (Mahmood et al., 2009). In view of the fact that these features are substantially affected by the circumstances under which they are seen, there is an urgent need for an advanced method for identifying evidence that goes beyond morphological and physiological standards (Staniaszek et al., 2008; Iqbal et al., 2010).

The increase in explicit quality is a direct result of the environment and the natural beautifiers present in a specie or strain (Kumar, 2011; Astarini et al., 2014). Because biochemical markers are a rapid aftereffect of

traits, they may serve as a source of information on inherent capriciousness.... Additionally, DNA finger printing is one of the most effective technologies in plant biotechnology, and it is used to evaluate the genetic assortment of plants (Mehmood et al., 2008). DNA markers that are one of a kind and similar to morphological characteristics have been utilised for the confirmation of hereditary assortments at the molecular level for a very long time (Sajida et al., 2009). Molecular markers, particularly discretionary enhanced polymorphic DNA (RAPD), have been used to determine the inherent assortment of mushrooms. Because of the straightforwardness of its method, RAPD is utilised to evaluate inherited proximity and conduct phylogenetic investigation on a population. Moharram et al. (2012) worked on a representation of the oyster mushroom that was reliant on supplemental nutrition. Singh et al. (2016) used DNA fingerprinting and ribosomal RNA quality sequencing to characterise eighteen different strength mushroom increments, and they discovered the existence of genetic variety in all of them. As a result of their investigation, Hyeon et al. (2007) discovered that gathering subject to physiological limitations is inextricably linked to RAPD-based assembly in Pleurotus eryngii, and that this relationship is irreversible. Stajic et al. (2015) utilised a haphazardly improved polymorphic DNA method to evaluate the hereditary assortment among 37 pleurotus species of mushrooms in a study published in Nature Communications. According to the results of another study, RAPD-polymerase chain reaction

(PCR) upgrading was utilised to evaluate the innate assortment among 45 pleorous strains and it was discovered that this approach performed better than morphological evaluation. A current evaluation was required to examine the genetic assortment across various strains of produced mushroom (Pleurotus spp.) while taking into consideration the convenience of morphological and molecular markers).

### **OBJECTIVE**

- 1. To explore oyster mushroom and its significance of collected mushroom cultures (Pleurotus florida and P. djamor).
- 2. To study the nutritional status of mushroom and biochemical parameters are also tested.

## **REVIEW OF LITERATURE**

The utilization of the Pleurotus species has a few beneficial outcomes on the overall human wellbeing due to various wellbeing advancing substances, they have (Kues and Liu, 2010). Numerous Pleurotus species have yielded potential, naturally dynamic mixtures that display anticancer action in vitro or in creature models (Borchers et al., 2012). These mixtures incorporate hemicelluloses, polysaccharides, lip polysaccharides, peptides, proteins, glycoproteins, nucleosides, triterpenoids, complex starches, lipid subordinates and different metabolites (Kalac and Svobod, 2015; Lindequist et al., 2016). Numerous basidiomycetes have the capacity to deliver all the while both the hydrolytic and oxidative chemicals which are expected to debase complex lignocellulosic substrates (Buswell et al., 2012; Kirk et al., 2013). Extraordinary variety inside Pleurotus recommends fluctuation as far as yield and natural Efficiency (BE) (Buswell et al., 2013). Pleurotus species can be utilized to productively deal with the horticultural waste materials left subsequent to collecting and simultaneously utilized as significant wellspring of food Environmental conditions are vital components to the mushroom's development and fruiting. The ecological factors such sas temperature, mugginess, gases, light, ventilation influence the shape and yield of mushroom The development of palatable mushrooms has advanced in a period and has become these days a movement of prudent significance, predominantly for the creation of types of the genera Agaricus, Pleurotus and Lentinula. Their reality creation increments, particularly Pleurotus sp. especially happened because of their capacity to fill in various deposits, like sawdust and agro mechanical waste, a trademark that made creation financially practical. Such attributes are relevants, respects the creation, however mushrooms are additionally significant in regards to their nourishing viewpoint. The sort of substrate, the natural conditions and the organism species utilized in development all have a huge impact in the compound sythesis of the Varieties happen fruitina bodies. principally comparable to minerals and protein substance (Crisan

and Sands, 1978). The end of water substance of the example to dry state will expand the centralization of supplement moderately. Accordingly, drying mushrooms is one of the strategies that would expand the time span of usability of mushrooms by lessening pointless biochemical response, for example, enzymatic searing and lipid oxidation that may prompt quality weakening.

The varieties of the protein substance among the palatable mushrooms are influenced by various elements, specifically the sort of mushrooms, the phase of advancement, level of nitrogen accessible. and the area (Longvah and Deosthale, 2016). Mushrooms are demonstrated to have great quality and higher protein content when contrasted with vegetables (Bozin et al., 2013).

#### MUSHROOM CULTIVATION

#### **Substrates**

Macrofungi from the class Pleurotus, generally known as clam mushrooms are liked by numerous individuals for their fragile taste, gentle yet chewy surface and their novel smell. The world exchange of these mushrooms shows an expanding example and offers promising chances for the dealers (Chang, 2011). This pattern happens not just due to popularity from the purchasers yet in addition the capacity to apply modest development methodologies. discovered becoming normally on certain spoiled woody material (Phillips, 2006). They have a wide of temperature versatility (Bano Rajarathnam, 1982) and substrate utilization. They have been developed in an enormous sums by utilizing lignocellulose materials like sawdust, paddy straw, wheat straw and cotton demonstrated to be effectively developed on banana pseudo stem, Bahia grass (Siqueira et al., 2011), bamboo leaves, yard grasses (Kumari and Achal, 2015), sweet potato peelings (Dioscorea sp.), cassava peelings (Manihot sp.), wild grass (Pennisetum sp.) corn stover (Zea mays) and oil palm (Elaeis guineensis) and natural product filaments (Okhuoya and Okogbo, 2014). The significance of the consumable mushrooms has expanded because of the advances the development innovation, which utilizes farming and mechanical buildups conceivable by reusing them as substrates for development, thus coming about in the lowcost creation and a constant market (Pandey et al., 2000).

An alluring component of this gathering of mushroom is because of the limit of emitting range of chemical. They can use an enormous assortment of farming byproducts containing (lignin, cellulose, starch, sugars and aged proteins) and change the lignocellulosic biomass into food sources superior grade, flavor and nutritive worth (Baysal and Peker, 2001). The vast majority of the parasites have solid catalyst and are equipped for using complex natural mixtures which happen as rural squanders and mechanical sideeffects. Mushroom growths likewise have a place with this gathering. Subsequently agrarian squanders can likewise be utilized as bedding material for mushroom development (Baysal and Peker, 2001). Lignocellulosic squanders, for example, neem frame, wheat grain, and sugarcane bagasse, accessible in plenitude, are brilliant substrates for the creation of ligninolytic compounds under strong state maturation by the white-decay parasites (Verma and Madamwar, 2002).

A few contagious animal types have the unmistakable and distinct capacity to debase lignin, cellulose and hemicellulose because of their intrinsic ability to emit a range of oxidizing and hydrolyzing proteins. An effective usage of lignocellulosic substrates by parasites is straightforwardly identified with their ability to use such substrates. The utilization of lignocellulose as carbon source relies upon the limit of growth to deliver lignocellulolytic catalysts and to discharge them to extracellular medium (Villas-Boas et al., 2012). Various sorts of straws are normally utilized for the Pleurotus sp. development. Straw can be treated the soil or sanitized and additional added substances can be utilized to build the BE. When utilizing rice and wheat straw for P. sajor-caju development, better returns were acquired on ground than on cleaved straw, and yields were 10 % higher on rice than on wheat straw. Higher generate levels upgraded mushroom yields (Zhang et al., 2013). Rice straw gave off an impression of being the best substrate for P. ostreatus mushroom development when contrasted with banana leaves, maize stover, corn husks, rice husks and elephant grass (Obodai et al., 2014).

# **Mushroom nutrition**

Mushroom contains apparent measure of potassium, phosphorous, copper, and iron however low degree of calcium. Mushroom development is profoundly work escalated, brief term yield and land saving, can be invited by the helpless formers (Anderson and Feller, 2013). The mushroom mycelia likewise contain amino acids like glycine, valine, threonine, serine, leucine, proline, methionine, asparagines, glutamine, lysine, arginine, histidine, cysteine and alanine (Quimio, 1976). For millennia, eastern culture has turned around mushrooms have medical advantages. Mushroom have for some time been commended as a wellspring of incredible supplements, however they can likewise help Americans meet the dietary proposals set out in the 2010 dietary rules and establishment of medication's dietary reference admissions for calcium and nutrients D. These positive advantages of mushroom can have likely effect (Lorenzo etal., 2014). Regularly gathered with vegetables, mushrooms give a significant number of the dietary credits of produce, just as trait all the more generally found in meat, beans or grains. Mushrooms are low in calories, without fat, sans cholesterol glutan free, and low in sodium, yet they give significant potassium, supplements, including selenium, riboflavin, niacin, nutrient D and the sky is the limit from there (Quimio, 2016). Shellfish mushrooms (Pleurotus species), the third biggest economically delivered mushroom on the planet (Van, 2009) have dietary and therapeutic properties (Garcha et al., 1993) Nutritionally, the mushroom has been found to contain nutrients B1 (thiamin), B2 (riboflavin), B5 (niacin), B6 (pyridoxine) and B7 (biotin) (Solomko and Eliseeva, 1988). Protein is perhaps the main supplements in food being especially significant for building body tissue. Mushroom with protein content going from 3-7% when new to 25-40% when dry it's anything but a significant part in improving human weight control plans (Ruiz etal., 2000). The family Pleurotus is a heterogeneous gathering of monetary significance. A few animal types are of nourishing as well as restorative significance (Guzman, 2010; Cohen et al., 2013). Pleurotus species can assimilate microelements from various development media and in this way they may introduce a brilliant dietary source (Stajic et al., 2002).

## Molecular studies of Pleurotus sp.

A few highlights of rDNA make it suitable for efficient and phylogenetic examinations. To begin with, this area of the genome is very much described and monitored. Numerous preliminaries as of now are accessible to enhance districts of the rDNA rehash that would supply arrangement information for a wide scope of taxa (White et al., 2013). Second, considerable examination has been done on rDNA from numerous organisms, so plentiful datasets are accessible for reference. Furthermore, various districts of rDNA develop at variable rates, which can be utilized to research contagious connections at various ordered levels (Bruns et al., 2014). Generally ordered and phylogenetic investigations Basidiomycota have been founded examination of morphological characters. As of late, connections among species in a few genera of Basidiomycota have regularly been set up by enhancement of atomic arrangements by Polymerase Change Reaction (Bos, 2011; Pringle et al., 2012). Examinations have essentially centered around nucleotide arrangements of the inside interpreted spacer (ITS) situated between the atomic rDNA 18S and 28S subunit qualities, and made it conceivable to decide the connections between parasitic species from the family Pleurotus (Molcalvo et al., 2010). Points of view for fingerprinting the genomes of mushrooms have as of late emerged from molecular markers dependent on the polymerase chain response. These systems have given novel and extremely incredible reproducible and solid DNA fingerprinting techniques (Vos et al., 2010). Molecular markers like fast intensified polymorphic DNA (RAPD) markers, limitation part length polymorphic (RFLP) markers, microsatellite and mitochondrial genotypes have all been utilized to segregate mushroom species (Barroso et al., 2008).

There is a developing industry of palatable mushroom creation because of their nutritive worth and the

perceived reality that mushrooms are normal and good food varieties beginning from an eco-agreeable natural cultivating framework. The creation of eatable mushrooms is compromised by both abiotic and biotic elements, henceforth the need to improve rearing through hereditary devices. While trying to decide the morphological and hereditary variety among Pleurotus species, fourteen distinct strains of Pleurotus ostreatus and Pleurotus floridawild type and their freaks were morphological exposed to various attributes, ultrastuctural hyphae network studies and irregular intensified polymorphic **DNApolymerase** (RAPD-PCR) response marker. The mvcelia development yield on Petri-plates and in lowered maturation demonstrated that the strains PO90 and PF30 were essentially not quite the same as the other Pleurotus strains utilized in this investigation at P < 0.05. Additionally, the microscopy result showed checked contrasts among the Pleurotus strains. The dendrogram dependent on RAPD investigation produced two distinct groups. Out of 4 irregular preliminaries, particular polymorphism was seen by groundworks BG17, BG18, BG 23 and BG25. The rate closeness among the Pleurotus strains differs between 40-100% (Ola et al., 2013).

Mushooms have as of late stood out and are misused food and restorative purposes. Precise recognizable proof of mushrooms is key in using them to assist people. In any case, morphological distinguishing proof of mushrooms is tedious, drawnout and might be inclined to mistake. DNA markers are fast and solid instruments that are helpful in mushroom scientific categorization. To recognize of six Ghanaian mushrooms utilizing the inner interpreted spacer (ITS) arrangements. The ribosomal DNA-ITS pieces of genomic DNA of six wild mushrooms were intensified utilizing ITS1 and ITS4 preliminaries. The amplicons were sequenced and information gathered and broke down utilizing Bio Edit. Fundamental Local Alignment Search Tool (BLAST) search was completed utilizing the National Center for Biotechnology Information (NCBI) data set.

## RESEARCH METHODOLOGY

Out of seven species, five were obtained from the Vineland Agriculture Station in Ontario, Canada, and the remaining two were obtained from Dr. Qumio's Plant Pathology Department at the University of California, Davis. Pleurotus platypus (P-6), Pleurotus flabellatus (P-7), Pleurotus florida (P-17), Pleurotus Ostreatus (P-19), and Pleurotus sajorcaju (P-56) were obtained from Dr

Mushroom cultivation is a popular pastime.

In this study, all of the species were grown on potato dextrose agar (PDA) medium, which included the following components: potato starch (20 grammes), dextrose (20 grammes), agar (20 grammes), and distil water (to create a total volume of one litre). Antibiotic streptomycin was added at a concentration of 1 g/l.

The medium used for mushroom cultivation was wet sterilised at 121°C for 30 minutes at 15 psi and then cooled to 40°C (Khan et al., 2006) before being used for mushroom cultivation.

# Mushroom growing is a popular pastime

As a substrate for mushroom development, cotton waste material was utilised as a source of nutrition. It was necessary to prepare the substrate by soaking it in water for 72 hours, after which it was allowed to extrude excess moisture by spreading it out on an inclined surface. This substrate was put into polythene bags at a rate of one kilogramme per bag and then dried sterilised. A fully randomised design was used in the experiment, with three replications and 10 bags in each replication for each treatment. The results were analysed using a completely randomised approach 2006). Various morphological et al., characteristics, such as mycelial growth (mm) (after seven days on PDA), cap diameter (cm), total yield (kg) after four flushes, and biochemical characteristics, such as Mushrooms growing on cotton waste were harvested for their moisture content (percent), ash content (percent), nitrogen content (percent), and protein content (percent) of the fruiting body.

## **Analyzing morphological information**

Steel et al. (1997) conducted an analysis of variance for all morphological and biochemical characteristics in order to evaluate the significance of variation across all species, and a dendrogram was created using Minitab statistical software.

## **MOLECULAR ANALYSIS**

# DNA extraction

In organisms, the arrival of DNA is often poor due to the presence of cell dividers or instances that are not immediately powerless, among other factors. When it comes to filamentous organisms, they contain strong cell dividers that are notoriously difficult to break down using traditional DNA extraction methods (Van Burik et al., 1997). When it came to DNA disconnection from parasitic species, the protocol suggested by Sambrook et al. (2001) was followed to the letter.

After three days of duplication on potato dextrose medium at 25°C, the DNA of seven different species was extracted from mycelial string samples. Centrifuged for 5 minutes at 10000 rpm, followed by washing with 500 mL TE cradle, the mycelial mat was then dried. After being pounded in extraction cushion (200 mm Tris-HCl, pH-8.5, 250 mM NaCl, 25 mM EDTA, 0.5 percent SDS) using a cone-shaped processor, the range was expanded in 3M sodium acetic acid derivation for 15 minutes (pH-5.2). The lysates were centrifuged after being brooded at - 20°C for 10 minutes at a time. The extraction of DNA from the supernatant was aided by the addition of equal quantities of isopropanol, and the resulting bed was

washed with 70 percent ethanol. A 20-mL TE cushion was used to break down the DNA range after it had been air dried. A DNA measurement was carried out, and a weakening concentration of 15 ng/l was used in the downstream application.

# **RAPD** analysis

A total of 14 random decamer primers (Table 1) produced by Operon Technologies in the United States were used in individual PCR reactions for each isolate in 0.2 ml tubes. In 25-mL reactions comprising PCR buffer, 5mM each of the deoxyribonucleotide triphosphates, 10 pmol each of the relevant primers, DNA at a concentration of 15ng/mL, and 1unit of Taq polymerase, amplification was carried out to get the desired results. Following an initial denaturation phase at 94°C for 5 min, the amplification procedure consists of 40 cycles of amplification at 95°C for 1 min (denaturing), 36°C for 1 min (annealing), 72°C for 2 min (extension), and a final extension step at 72°C for 10 minutes.

S/N	Primer code	Primer sequence	Nobands	bands of No of polymorphic	(%)polymorphic
1	OPQ1	GGGACGTATGG	H	5	55.60
2	OPQ6	GAGCGCCTTG	6	0	00.00
3	OP12	GGAGGAGAGG	7	4	57.10
4	OPL3	CCAGCAGCTT	11	8	72.70
5	OPL11	ACGATGAGCC	10	7	70.00
6	OPL15	AAGAGAGGGG	8	3	37.50
7	OPL13	ACCGCCTGCT	16	7	43.75
8	OPL2	TGGGCGTCAA	8	3	37.50
9	OP15	TGTTCCACGG	10	5	50.00
10	OPQ17	CCGTACGTAG	7	0	00.00
11	OPP11	AACGCGTCG	11	3	27.27
12	OPP17	TGACCGCCT	5	0	00.00
13	OPN10	ACAACTGGGG	6	2	33.33
14	OPLS	AGCAGGTGGA	9	4	44.44

Gel electrophoresis and RAPD data scoring The amplicons after PCR were analyzed by electrophoresis on 1.2% (W/V) agarose gels by running in 0.5X TBE buffer. After staining with ethidium bromide the gels were visualized under a UV transilluminator and photographed using Bio-Rad gel documentation system. The amplification product generated by each RAPD primer were scored as "1" or "0" for presence or absence of specific allele, respectively. To estimate the similarity and genetics distance among different Pleurotus species, cluster analysis based on Nei's unweighted pair-group with arithmetic average (UPGMA) was performed using the Popgen-32 software (Yeh et al., 2002) and a dendrogram was constructed.

# **RESULTS**

The examination of change (Table 2) demonstrated that, every one of the characteristics concentrated aside from protein substance were measurably critical (P 0.05). The dendrogram (Figure 1) gathered the seven mushroom species in two principle bunches. The group 'A' contains four animal types while bunch 'B' contain three species. In group 'A', species P-6 (P. platypus) and P-19 (P. ostreatus) were more comparative and fall in same subgroup and the examination of means for all out yield, debris

substance and protein substance showed that these species were measurably similar. Then again, in bunch 'B' the species P-9 (P. warm-starm) and P-16 (P. eryngii) were in same subgroup and correlation of means for complete yield, nitrogen substance and protein substance uncovered that these species were likewise genuinely comparative. The genomic DNA of seven Pleurotus species was dissected utilizing 14 ten mer arbitrary preliminaries. Every one of the preliminaries aside from OPQ6, OPQ17 and OPP17 were polymerphic. The quantity of groups and banding design were variable relying on the groundwork and sort of species tried and it went from 5 to 16 in checking. The greatest polymorphism was delivered by the groundwork OPL3 and OPL4 having polymorphism rate 72.70 and 70%, separately. The dendrogram dependent on comparability network separated the species into three unmistakable bunches A, B and C. Bunch An established animal varieties P-56 (P. sajorcaju), P-17 (P. florida) and P-6 (P. platypus), bunch B contained species P-19 (P. oysterus) and P-7 (P. flabellatus) and group C contained species P-9 (P. warm-starm) and P16 (P. eryngii) (Figure 2). The hereditary similitudes between species went from 75.5 and 86% (Table 3). The similitude among three types of group A was from 84 to 86%. The bunch 'B' and 'C' included two species in each gathering, which showed 80 and 81% similitude, separately. The closeness grid additionally portrayed that most firmly related confines (P-56 and P-17) were 86% comparable.

## **DISCUSSION**

The mean square and means for the thought qualities in group investigation uncovered that, there were critical contrasts among every one of the animal categories. The presence of species P-6 (P. platypus) and P19 (P. ostreatus) in same sub bunch of fundamental group 'A' and animal types P-9 (P. warmstarm) and P-16 (P. eryngii) in same subgroup of bunch 'B' uncovered that, the morphological conduct of these species was comparative or they may have same progenitors, yet in some cases, morphological based gathering didn't coordinate molecular/genomic relationship among the species (Stajic et al., 2005). In such manner, the morphological characteristics don't give a significant casing work to transformative arrangements. In light of the moderately basic fruiting designs and significant formative versatility of organisms, it is acknowledged that amassing contrasts in creating sub-populaces are not generally communicated as far as morphological uniqueness. Intently

Table 2. Means, mean square and LSD value of morphological traits

Danning	Morelel greeds (rest)	Cop Remote (and	Total state (Ital)	Marabas contents (%)	Ash content (Na	Minger coldent (%)	Project receiped (%)
F-b	34.00 to .	JH REV.	\$94.1 bp.	Thi00 a	6.000 to	5.790a	41 dithe
60	28.500	200.004	255.0 4	R1309c	1.000 1:	8 600 8	30 MHz
P-D	21.00 w	10.00.4	diff on	79.00-36	1.00%	9.000 Y	00.01 lis
P-10:	31.0504	2020 4	diff de	BE186/90	0 0 M to	4-147.9	thittle .
P.17:	36.00A:	26.00 %	30.76	71.00 st	1.000 %	8.200 H	36-860a
P.18	40366	40.004	981 1700	84:50 S.	0.000 h	0.600.8	AG-000la
P.M.	AT Disc.	48.00k	886 Tan	86.7%s	1.000 11	9.607 8	40 10 lin
150	2000	2.09	54.05	2400	E710	1.09	3.661
Most squares	THEFT.	**************************************	1102*	TERRY:	1000	B 1800**	4.35 <sup>30</sup>

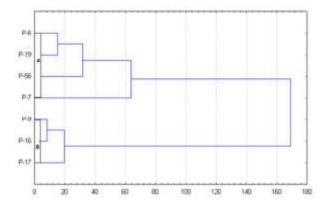


Figure 1. Dendrogram based on morphological traits.

Table 3. Similarity matrix for Nei's and Li's coefficient of seven fungal species

Specie	P-8	P-58	P-17	P-19	P-16	P-6	p.7
P-9	1.000	- Sheles	-91005-	- 1000	-0.00	- 3322	777.
P-56	0.723	1,000					
P-17	0.784	0.867	1.000				
P-19	5.000	0.754	0.755	1.000			
P-16	0.852	0.767	0.755	0.755	1.000		
P-6	0.790	0.043	0.838	0.791	0.811	1,000	
P-7	0.793	5.805	0.804	0.857	0.760	0.785	1.00

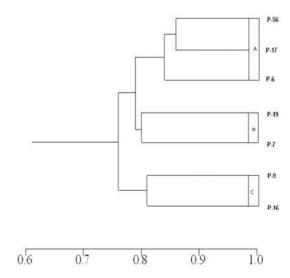


Figure 2. Dendrogram of seven fungal strains developed from RAPD data using the unweighted pair group method of arithmetic means (UPGMA). The scale is based on Nei's and Li's coefficients of similarity.

Related or kin species may along these lines, need systematically valuable morphological contrasts long speciation after underlying occasion. Morphological highlights that had been utilized to recognize Pleurotus spp. in the past unmistakably recognize the distinctive phylogenetic types of which it is involved. The essential DNA arrangement of a living being can be dared to be harsh toward transient natural change and subsequently ought to give a more steady choice to strain/species recognizable proof. Accordingly, the arbitrary and genomic wide nature of the RAPD procedure is ideal to show over all hereditary relatedness/disparity than the morphological investigation (Ravash et al., 2009). The various preliminaries delivered distinctive number of groups in PCR. This variety in the quantity of groups might be because of the arrangement of preliminary. accessibility of tempering destinations in the genome and format quality.

Notwithstanding this, every one of the animal groups were developed on comparative culture yet the varieties in the banding design were reflected. Chandra et al. (2010) utilized the RAPD markers to separate the eight Pleurotus types of mushrooms and furthermore discovered varieties in banding design. The polymorphism delivered by 11 RAPD preliminaries with the exception of OPQ6, OPR17 and OPP17 might be because of the base replacement, inclusion and erasure or assortment of hereditary material from various sources (Chopra, 2005; Jusuf, 2010). The most extreme similitude (86%) was seen between P-17 (Pleurotus florida) and P-56 (Pleurotus sajor-caju) having a place with group 'A'. Chandra et al. (2010) discovered smaller than usual mum comparability level (27 %) between Pleurotus florida and Pleurotus sajorcaju. This logical inconsistency in the finding might be because of various development conditions. The base (72%) likeness was seen between P-56 (Pleurotus sajor-caju) and P-9 (Pleurotus warm-starm) which had a place with two unique bunches 'A' and 'C', separately. During development examines, both of these, P-56 (P. sajor-caju) and P-9 (Pleurotus warmstarm), end up being quick and moderate developing separately and showed both have a place with two diverse living space that is the reason these species were seen to join various bunches, An and C, individually. These discoveries uncovered that, the hereditary cosmetics is related with natural 1998). The examination heterogeneity (Nevo, portrayed that social varieties was likewise held at molecular level. P-19 (Pleurotus ostreatus) is a low temperature received animal varieties while P7 (Pleurotus flabelatus) is tropical in nature yet in spite of this differentiation, they framed a different group 'B'. P-9 (Pleurotus warm-starm) and P-16 (Pleurotus eryngii) framed a particular sub-bunch in group. At the molecular level, both of these specie additionally held gathering dependent on morphological information. The outcomes portrayed that, there is solid connection among's molecular and morphological measures (Zervakis et al., 2004).

#### CONCLUSIONS

The types of organisms having a place with natural nearness or distinctive geological beginnings can be characterized through morphological and molecular markers. The current investigation showed that, the RAPD examination and morphological assessment both are valuable for portrayal, hereditary variety and distinguishing connections among Pleurotus types of mushrooms. Study likewise uncovered that, RAPD examination can be exceptionally valuable apparatus for mushroom cultivator for grouping and upkeep of good quality generates.

- 1. Astarini AI, Plummer AJ, Lancaster AR, Yan G (2014). Fingerprinting of cauliflower cultivars using RAPD markers. Aust. J. Agri. Res. 55: pp. 112- 124.
- 2. Chandra S, Ghosh K, Acharya K (2010). Comparative study on the Indian cultivated Pleurotus species by RAPD fingerprinting. Nature & Sci. 8: pp. 90-93
- 3. Cheung ST (2012). World production of cultivated edible and medicinal mushrooms in (1997). with emphasis on lentinus edodes (berk). Sing in China. J. Med. Mushrooms. 1: pp. 291-300.
- 4. Chopra VL (2013). Mutagenesis: Investigating the process and processing the outcome for crop improvement. Curr. Sci. 89: pp. 353-359.
- 5. Gepts P (2011). The use of molecular and biochemical markers in crop Evolution studies. In: Hecht MK (ed.). Evol. Biol. 27: pp. 51-94.
- 6. Hyeon-Su RO, Sung SK, Jae SR, Che-Ok J, Tae SL, Hyun-Sook L (2013). Comparative studies on the diversity of the edible mushroom Pleurotus eryngii: ITS sequence analysis, RAPD fingerprinting, and physiological characteristics. Mycological Res. 111: pp. 710-715
- Jusuf M (2010). Amplified fragment length polymorphism diversity of cultivated white Oyster mushroom Pleurotus ostreatus. HAYATI J. Biosci. 17: pp. 21-26
- 8. Mehmood S, Bashir A, Ahmad A, Akram Z (2014). Molecular characterization of regional sorghum bicolor varieties from Pakistan. Pak. J. Bot. 40: pp. 2015-2021.
- 9. Stajic M, Sikorski J, Wasser SP, Nevo E (2016). Genetic similarity and taxonomic relationships within the genus Pleurotus (higher Basidiomycetes) determined by RAPD analysis. Mycotoxon. 93: pp. 247- 255
- Zervakis GI, Moncalvo JM, Vilgalys R (2015). Molecular phylogeny, biogeography and speciation of mushroom species Pleurotus cystidiosus and allied taxa. Microbiology, 150: pp. 715-726.
- 11. Yeh FC, Yang R, Boyle TJ, Ye Z. (2013)
  Popgene 32, Microsoft ware windows based
  freeware for population genetic analysis.
  Molecular Biology and Biotechnology Centre,
  Edmonton.

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