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Cultivation of different strains of *P. pulmonarius* on wheat and paddy straw

Ananya Rao, Chetana Jangde and Raut Ravindra Sunderrao

Abstract
Ten strains of *Pleurotus pulmonarius* were evaluated on paddy straw and wheat straw substrate for their yield and biological efficiency. Variation in time duration was observed (for spawn run, time to express pin heads) and biological efficiency when strains of *Pleurotus pulmonarius* were evaluated on paddy and wheat substrates. Strain PP-21-03 and PP-21-04 and took 17.5 days and 13.2 days for spawn run on paddy and wheat straw respectively. On paddy straw pin heads were expressed early by PP-21-10 (5.5 days) where as two strains PP-21-05 and PP-21-09 expressed early pin heads in (4.1 days) on wheat straw. Strain PP-21-01 was identified as potential isolate as it expressed highest yield (1082.4 g) expressing 108.24% BE when evaluated on Paddy straw. On wheat straw strain PP-21-10 expressed highest yield (857.56 g) expressing 85.75% BE.

Keywords: *Pleurotus pulmonarius*, strain, paddy, wheat

Introduction
Mushrooms are non-timber forest products that are frequently seen growing as saprophytes on farmlands, open fields, wooded areas, and the sides of roads. It is possible to see the fruiting bodies with the naked eye since they are so huge. They are members of the order Agaricales and the class basidiomycetes. Mushrooms are gaining popularity as a delicious, high-protein vegetable. The common name "oyster mushroom" derives from the fruiting body's white shell-like appearance. Oyster mushroom includes number of species i.e., *Pleurotus ostreatus, Pleurotus sajor-caju, Pleurotus cystidiosus, Pleurotus cornucopiae, Pleurotus pulmonarius, Pleurotus tuber-regium, Pleurotus citrinopileatus, and Pleurotus flabellatus.* Among which, *Pleurotus pulmonarius*, is an edible white rot fungi (WRF) also called as Indian oyster, Italian oyster, phoenix mushroom, or lung oyster. *P. pulmonarius* has a tiny hat that is often cream or light brown in colour and measures 5 to 20 cm in size. It has a curved, occasionally ruffled edge and is formed like a shell and a paddle; in younger specimens, it has an oyster-like appearance. The gills are unremarkable; they are hat-colored, quite narrow, and converge profoundly to the stem.

Material and Methods
Experimental site
All the experiments were carried out in Mushroom Research Laboratory, Department of Plant Pathology, College of Agriculture, Indira Gandhi Krishi Vishwavidyalaya, Raipur (C.G.).

Source of material
All the strains of *Pleurotus pulmonarius* were obtained from All India Co-ordinate Mushroom Improvement Project (AICMIP), Department of Plant Pathology, Indira Gandhi Krishi Vishwavidyalaya, Raipur (C.G.) and all cultures were multiplied on PDA and kept at 4 °C for all different study. All the substrates used in the research were taken from instructional farm, College of Agriculture, IGKV, Raipur (C.G.). Empty glass bottles, polythene bags, cereal grains (wheat and sorghum) and other chemicals i.e., streptomycin sulphate, ethyl alcohol, calcium carbonate, calcium sulphate, Bavistin and formalin were obtained from department of plant pathology, Raipur (C.G.).

Statistical analysis
The experimental study was laid on Completely Randomized Design (CRD) with single factor analysis and CD was calculated.

Preparation of mother spawn

The mother spawn of different strains of *P. pulmonarius* namely, PP-21-01, PP-21-02, PP-21-03, PP-21-04, PP-21-05, PP-21-06, PP-21-07, PP-21-08, PP-21-09, PP-21-10 was developed on wheat and sorghum grains. Grains that were used for preparation of mother spawn should be clean, healthy and bold size. Grains were washed and soaked in water till night, on the next day grains were boiled for 10-15 minutes for softening of grains and the seed coat was remains intact. Muslin cloth was used for cooling on which grains were spreaded evenly, then the cooled grains were mixed with calcium carbonate (0.5%) and calcium sulphate (2%) on wet weight basis to avoid stickiness of grains and maintenance of pH. These processed grains were kept in empty glucose bottles and air tied with non-absorbent cotton and autoclaved at 20 lbs. PSI for 2 hours. After sterilization, bottles were cooled at room temperature and inoculated with small bit of pure culture and incubated for 15 days at 25±2 °C till the white mycelium covered all grains surfaces. The mother spawn of each strain was prepared in sufficient quantity and used when and as required.

Mushroom bed preparation

Wheat and paddy straw were used as substrate for mushroom bed preparation for oyster mushroom production. The required quantity of substrate was dipped in water for 12 hours in which 75 ppm carbendazim and 500 ppm formaldehyde were mixed. After dipping of substrate (chemically mixed water) mouth of the container was covered and sealed to avoid fumes of formaldehyde spread in environment. After sufficient time, Excess water was drained out and straw was spread on chemically sterilized cemented floor till the moisture content of straws comes to 60-70%. Formaldehyde was applied on the floor before spawning and after that spawn was mixed in substrate by layering method @ 4% on wet weight basis in all the experiment. A unit of 1 kg of dry substrate was used. The substrate mixed with spawn was filled in poly propylene bags (12"X 18"- 150 gauges). The mouth of the bag was tied with nylon string and then 8-10 holes were done in every single bag for air perforation. The spawned bags were kept in mushroom growing room, where appropriate temperature (20-25 °C) and relative humidity (75-85%) were maintained by frequently sprinkling of water on walls and floor. After complete colonization of mushroom mycelium (spawn run), the bags were teared and removed. The compact masses of aggregated straw were termed as 'bed' and were ready for hanging to the iron racks in cropping room.

Hanging and watering of the bed

The mushroom beds were hung on iron racks at a distance of 50 cm apart with the assistance of nylon string for fruitification and regularly watering was done with the help of a sprayer.

Harvesting of mushroom

Pin-head starts appearing after 3-4 days of the bag removal. First flush of mushrooms was obtained within 5-7 days after primordial initiation. Mature sphorophores were harvested by firmly holding the stipe and twisting it. Picking was done before the edges of pileus tend to fold or get curl upwards. The intervals between two consecutive flushes varied from 8-10 days for all the strains.

Weighing of sporophore

The harvested fruiting bodies were weighed with the help of a single pan electronic balance with a sensitivity of 1.0 g.

Yield of mushroom

The cumulative yield data of each replication was found out by summing the fresh weight of all the pickings of a particular replicon. This weight (g) was denoted per unit of the dry straw substrate.

Biological efficiency

The biological efficiency was calculated as per the formula given by Chang *et al.,* (1981) [21].

$$\text{Biological efficiency (\%)} = \frac{\text{Fresh weight of mushroom}}{\text{The dry weight of the substrate}} \times 100$$

Effect of wheat and paddy straw on different strains of *P. pulmonarius*

Wheat and paddy straw were taken to evaluate the yield of different strains. The substrate was dipped in water for 12 hours in which 75 ppm carbendazim and 500 ppm formaldehyde were mixed. Excess of water was drained out and straw was spread on cemented floor till the moisture content of straws comes to 60-70%. The substrate mixed with spawn was filled in poly propylene bags (12"X 18"- 150 gauges). The mouth of the bag was tied with nylon string and then 8-10 holes were done in every single bag for air perforation. The spawned bags were kept in mushroom growing room, where appropriate temperature (20-25^0 C) and relative humidity (75-85%) were maintained. After complete colonization of mushroom mycelium, the bags were teared and removed. The bags were monitored at regular intervals for pin head initiation, sporophore maturation, days for 1st. 2nd and 3rd harvesting, and yield.

Result and Discussions

Yield performance of different strains of *P. pulmonarius* on wheat straw substrate

To estimate production standards of all strains of *P. pulmonarius*, mainly two substrates were selected i.e., paddy straw and wheat straw. The data regarding growth and yield attributing characters were recorded and depicted in table 1. Considering this experiment, it can be inferred that PP-21-10 was the best strain for cultivation on wheat straw as substrate.

Different strains of *P. pulmonarius* did not show significant difference in period of spawn run. On wheat straw substrate, it varied from 13.2-19.5 days and considerably fastest (13.2 days) and slowest (19.5 days) spawn run was recorded in strain PP-21-04 and PP-21-08 respectively.

The appearance of pinhead initiation was simultaneously first observed in PP-21-05 and PP-21-09 (4.1 days), next was PP-21-01 (4.3 days) and at par with PP-21-04 (4.7 days). It was then followed by PP-21-10 (5.1 days), PP-21-07 (5.8 days), PP-21-08 (6.5 days), PP-21-03 (12.25 days). However, PP-21-06 (30.85 days) took maximum days for pin head initiation and next was PP-21-02 (25.00 days) which differ significantly with each other.

On an average, highest sporophore weight was measured in PP-21-06 (8.98 gm), followed by PP-21-08 (6.25 gm) and PP-21-01 (5.04 gm). However, PP-21-10 gave least sporophore weight (2.92 gm).

The highest pileus diameter was found in PP-21-06 (5.3 cm), in other strains it was at par with each other. However, PP-21-02 had the smallest pileus diameter (2.5 cm). The strain PP-21-06 exhibit longest stipe length (2.4 cm) followed by 2 strains i.e., PP-21-02 and PP-21-08 (2.1 cm). However, smallest stipe length (0.5 cm) was found in PP-21-07.

Noticeably, PP-21-10 gave the highest yield (857.56 gm) with biological efficiency (85.75%). The other strains produced yields ranging from (433.99–691.65 gm) with biological efficiency ranging from (43.39–69.16%). However, Lowest yield (297.49 gm) with biological efficiency (29.74%) was found in strain PP-21-08.

The results are presented in above experiment are in accordance with the experiment performed by Gautam *et al.* (2020) [4]; Zhou *et al.* (2022) [20]; Getachew *et al.* (2019) [5]; Abid, (2020) [1] and have obtained higher yield of *Pleurotus ostreatus* on wheat straw substrate.

Table 1: Effect of wheat straw substrate on growth, yield and yield attributes of *P. pulmonarius*

S. No.	Strains	Spawn run* (days)	Pinhead initiation*(days)	Weight of sporophore**(gm)	Sporophore size**(cm)		Yield* (gm)	B.E. (%)
					Pileus diameter	Stipe length		
1	PP-21-01	16.8	4.3	5.04	4.5	1.6	624.1	62.41
2	PP-21-02	18.0	25.0	4.65	2.5	2.1	459.0	45.9
3	PP-21-03	18.0	12.2	4.91	4.5	1.7	503.12	50.31
4	PP-21-04	13.2	4.7	3.61	4.3	1.2	612.5	61.25
5	PP-21-05	17.8	4.1	3.48	4.2	1.3	691.65	69.16
6	PP-21-06	18.0	30.8	8.98	5.3	2.4	659.66	65.96
7	PP-21-07	17.6	5.8	3.76	4.2	0.5	433.99	43.39
8	PP-21-08	19.5	6.5	6.25	4.4	2.1	297.49	29.74
9	PP-21-09	17.3	4.1	4.30	4.7	0.6	442.87	44.28
10	PP-21-10	18.4	5.1	2.92	4.2	1.1	857.56	85.75
	SE(m)	2.45	2.04	0.85	0.55	0.23	22.56	
	CD	N.A.	5.75	2.41	1.55	0.65	67.03	

(*)- Average of 10 replications
(**)- Average of 10 sporophores of each replication

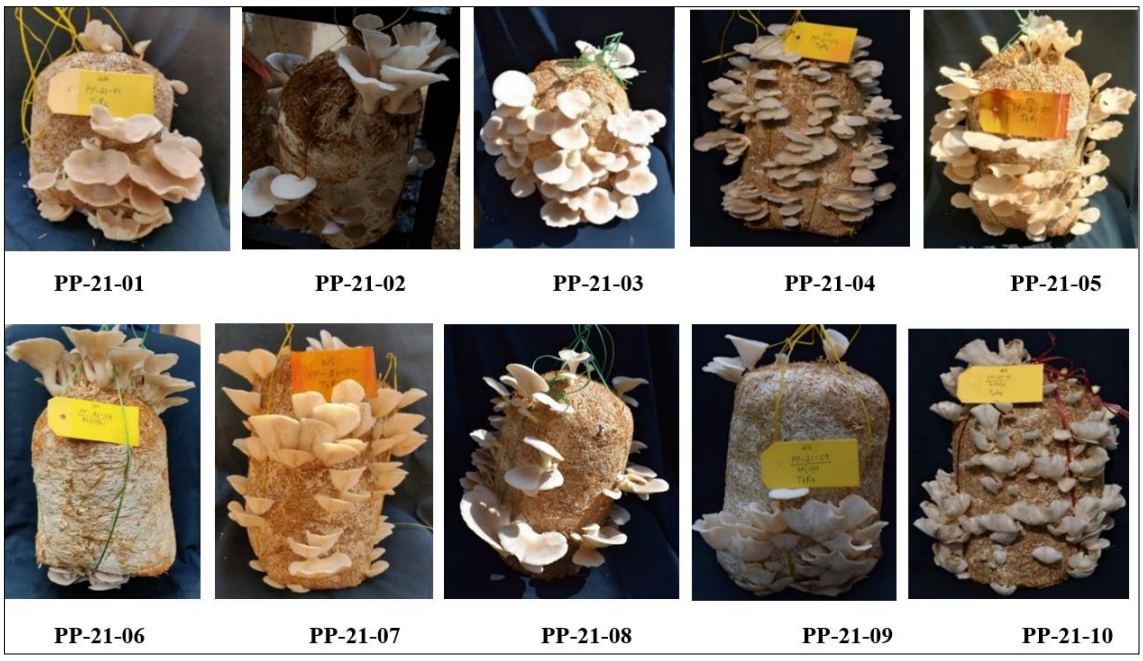

Fig 1: Performance of different strains of *P. pulmonarius* on wheat straw substrate

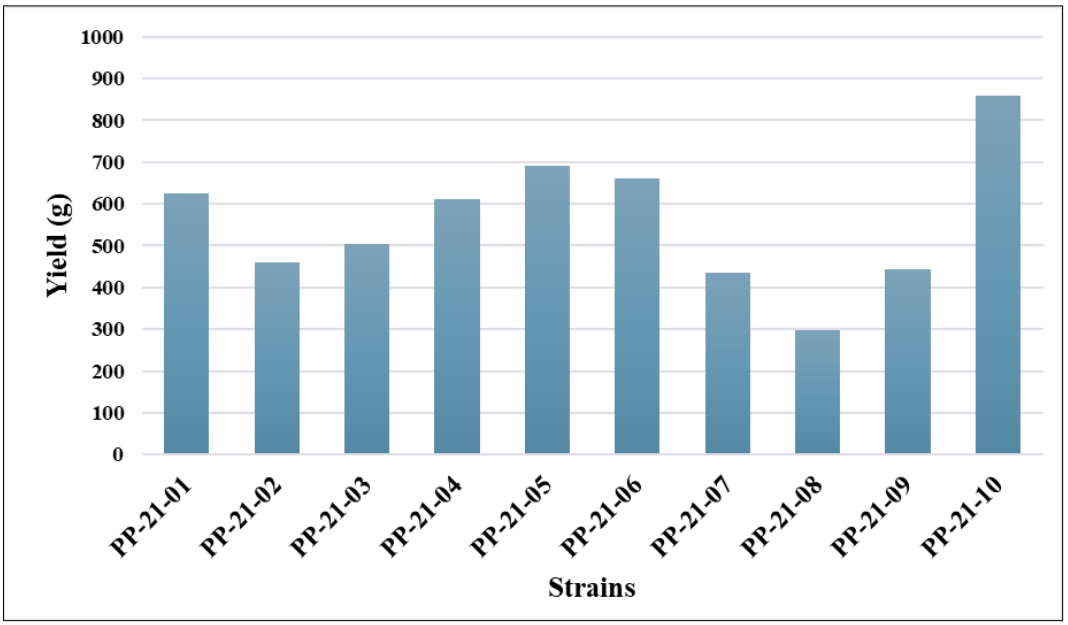

Fig 2: Yield performance of different strains of *P. pulmonarius* on wheat straw substrate

Yield performance of different strains of *P. pulmonarius* on paddy straw substrate

Paddy straw substrate was used for the cultivation of different strains of *P. pulmonarius* and the data regarding growth and yield attributing characters were recorded and depicted in table 2. Considering this experiment, it can be inferred that PP-21-01 was the best strain for cultivation on paddy straw substrate.

The colonization of different strains of *P. pulmonarius* on paddy straw substrate did not differ significantly with respect to spawn run period and it varied from 17.5 to 19.4 days.

Significantly, PP-21-10 gave earliest pinhead initiation (5.5 days) followed by PP-21-03 (5.6 days) and PP-21-09 (6.4 days). The pin head initiation period in other strains were at par with each other. However, strain PP-21-02 took significantly more period (22.0 days) and next was PP-21-06 (11.5 days) which were differed significantly with each other.

The largest sporophore weight was found in PP-21-01 (10.09 g) and next was PP-21-06 (9.33 g) and did not show significant difference in between. While other strains PP-21-09 (5.25 g), PP-21-06 (5.13 g), PP-21-08 (4.58 g), PP-21-04 (4.48 g), PP-21-03 (4.35 g), PP-21-07 (3.78 g) were at par with each other except PP-21-02 (6.57 g) and all were significantly lower with PP-21-01 and PP-21-06. However, PP-21-10 exhibit the minimum sporophore weight (3.43 g).

Pileus diameter was notably highest recorded in PP-21-01 (6.0 cm) and it was significantly with other strains and they were at par with each other. The lowest pileus diameter was observed in PP-21-07 (3.97 cm). The PP-21-06 strain had the longest stipe length (2.9 cm), followed by the PP-21-02 (2.2 cm) and the PP-21-08 (2.0 cm) and other strains were at par with each other. However, PP-21-09 had the shortest stipe length (0.83 cm).

The highest fresh yield (1082.4 gm) with B.E. (108.24%) was obtained from PP-21-01, followed by PP-21-10 yield (979.7 gm) with B.E. (97.97%). The other strains produced yield ranging from (410.75–807.5 gm). However, PP-21-08 produced lowest yield (350.6 gm) with B.E. (35.06%).

The superiority of paddy straw to obtain higher yield of oyster mushroom have been shown by Menon *et al.* (2021) [9]; Zárate *et al.* (2020) [19]; Mubasshira *et al.* (2020) [10]; Shifa *et al.* (2020) [17] for *Pleurotus ostreatus*; Sen *et al.* (2022) [14] for *Hypsizygus ulmarius*; Paswal *et al.* (2021) [18] for *Pleurotus djamor*; Sayed *et al.* (2019) [22] for *Pleurotus florida* and Deora *et al.* (2020) [3] for *Pleurotus eryngii*.

Table 2: Effect of Paddy straw substrate on yield and yield attributing characters of *P. pulmonarius*

S. No.	Strains	Spawn run*(days)	Pinhead initiation*(days)	Weight of sporophore**(gm)	Sporophore size**(cm)		Yield*(gm)	B.E. (%)
					Pileus diameter	Stipe length		
1	PP-21-01	18.1	7.1	10.09	6.0	1.7	1082.4	108.24
2	PP-21-02	18.5	22.0	6.57	5.1	2.2	410.75	41.07
3	PP-21-03	17.5	5.6	4.35	4.5	1.8	505.12	50.51
4	PP-21-04	18.0	6.6	4.48	4.9	1.3	591.6	59.16
5	PP-21-05	18.2	7.1	5.13	4.2	1.1	807.5	80.75
6	PP-21-06	18.6	11.5	9.33	5.4	2.9	709.99	70.99
7	PP-21-07	18.0	8.4	3.78	3.9	0.9	516.0	51.6
8	PP-21-08	19.4	8.6	4.58	4.8	2.0	350.6	35.06
9	PP-21-09	19.1	6.4	5.25	4.0	0.8	669.1	66.91
10	PP-21-10	18.1	5.5	3.43	4.1	1.1	979.7	97.97
	SE(m)	2.16	1.42	0.72	0.58	0.22	43.60	
	CD	N.A.	4.01	2.04	1.64	0.64	129.52	

(*)- Average of 10 replications

(**)- Average of 10 sporophores of each replication

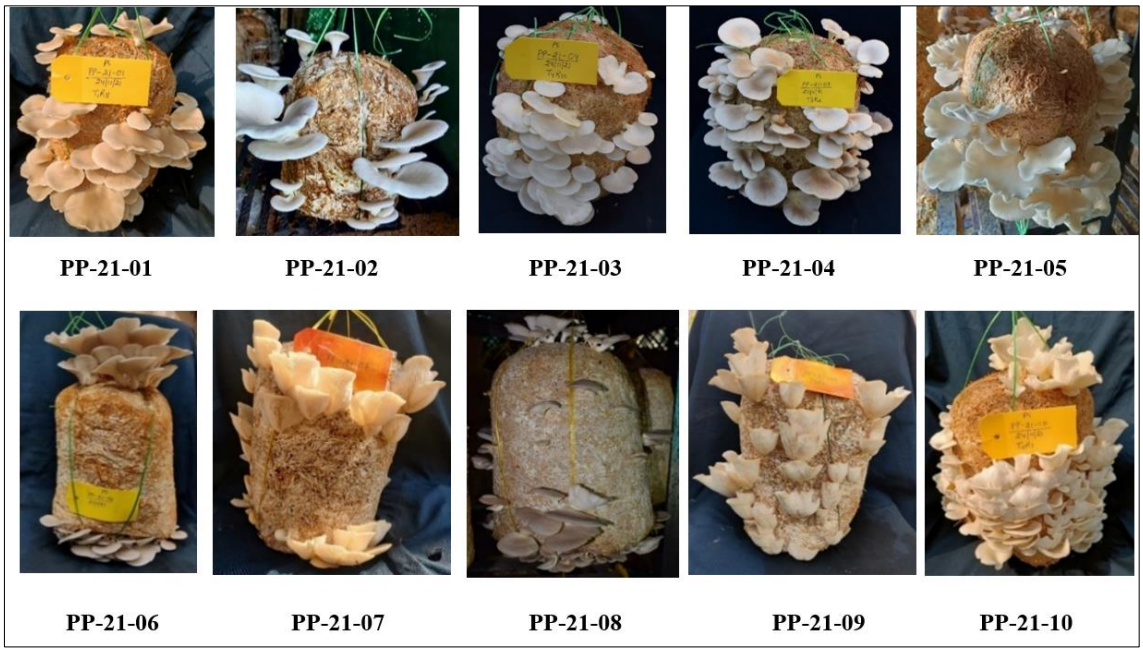

Fig 3: Performance of different strains of *P. pulmonarius* on paddy straw substrate

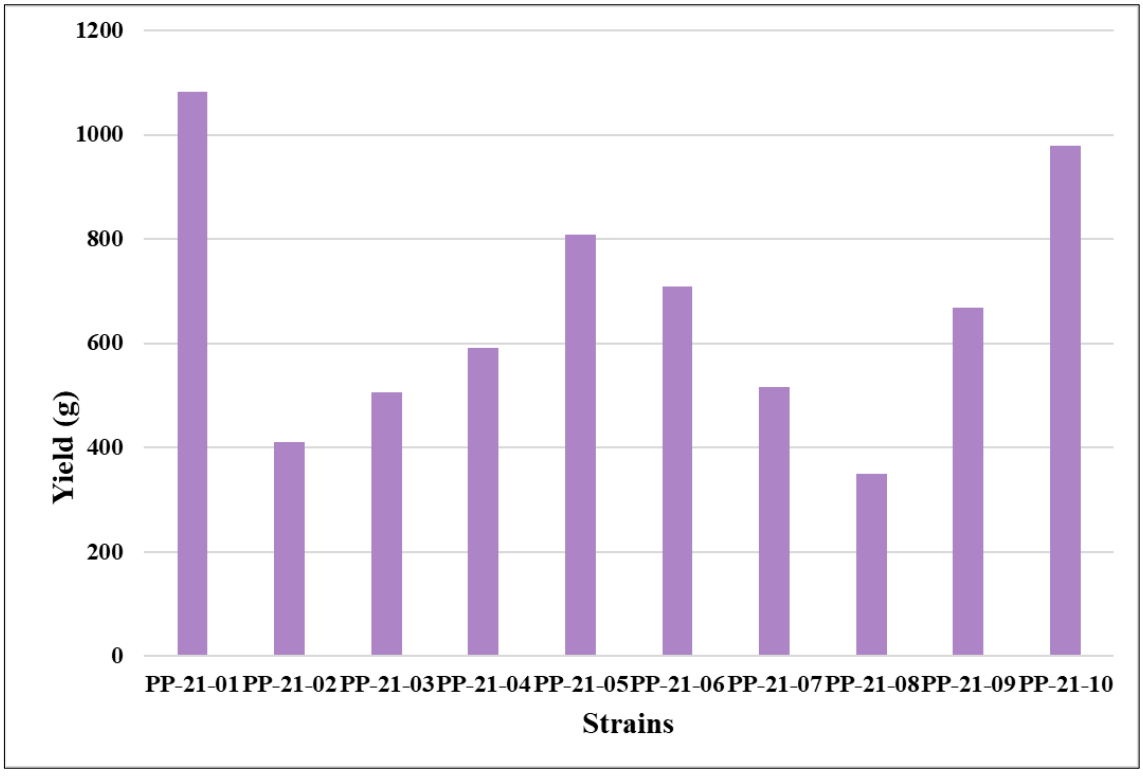

Fig 4: Yield performance of different strains of *P. pulmonarius* on paddy straw substrate

Conclusion

All the strains of *Pleurotus pulmonarius* were cultivated on wheat and paddy straw. PP-21-10 gave maximum yield (857.56 g) with B.E. (85.75%) while PP-21-08 achieved minimum yield (297.49 g) with B.E. (29.74%) on wheat straw. On paddy straw, PP-21-01 produced maximum yield (1082.4 g) with B.E. (108.24%) while PP-21-08 shows minimum yield (350.6 g) with B.E. (35.06%).

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