



## Evaluation of different organic additives on spawn growth of shiitake mushroom (*Lentinula edodes* (Berk.) Pegler)

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### ABSTRACT

The present investigation was undertaken to examine the effect of organic additives on spawn growth of shiitake mushroom (*Lentinula edodes*). Different organic additives like cereal brans (wheat, rice and corncob powder), leguminous flour (pea, lentil and soybean flour) and oilseed cakes (castor, linseed and mustard) at three concentrations i.e. 2, 4 and 6% were mixed with wheat grains. A significantly maximum mean mycelial growth was recorded in wheat grain spawn supplemented with pea flour (82.22 mm) at 6% followed by lentil (79.11 mm) at 6% as compared to control (42.08 mm). Mycelial run rate (MRR) ranged between 0.88 to 3.43 mm/day. Maximum MRR was observed in wheat grain supplemented with pea flour (3.43 mm/day) up to 10 days of incubation. It was concluded that supplementation of pea flour at 6% concentration took lesser time of complete mycelial growth on wheat grain and spawn growth enhanced with supplementation of all additives.

**Keywords:** Spawn, Wheat grains, Organic additives, Mycelial growth

Mushrooms are fleshy fungi that develop spore-bearing reproductive structures on organic substrates. Their nutritional value and medicinal properties make them integral to human dietary habits and health, as highlighted by research (Nidhi *et al.* 2023; Thakur *et al.* 2023a). For thousand years, extracts from medicinal mushrooms have been esteemed as crucial remedies for preventing and treating numerous diseases (Israilides and Philippoussis 2003; Joshi and Sagar 2019). Mushrooms depend on substrates for nutrition which is normally a source of lignocellulosic material which supports growth, development and fruiting of mushroom (Tarushi *et al.* 2020). Shiitake mushrooms, scientifically known as *Lentinula edodes* (Berk.) Pegler, is a basidiomycetous fungus renowned as a Japanese specialty. The name “Shiitake” originates from two components: “Shii,” representing the oak tree species (*Castanopsis cuspidata*), and “take,” denoting mushroom (Sharma *et al.* 2024). Additionally, this fungus is referred to by alternative names including Sawtooth oak mushroom, Black forest mushroom, golden oak mushroom, and oakwood mushroom. The mushroom can be found naturally growing in Papua New Guinea, China,

Taiwan, the Himalayan Mountains, Borneo, and Japan (Annapu *et al.* 2019). Next to the button mushroom, shiitake is a significant culinary and medicinal fungus that ranks second in terms of global mushroom production (Chang and Miles 2004; Bisen *et al.* 2010). Its popularity continues to rise, owing in part to its exotic flavor, nutritional richness, and diverse medicinal benefits. Additionally, its unique advantage lies in the ability to be sold dried, offering an extended shelf life compared to most other mushrooms, which are typically marketed fresh (Chauhan and Jaswal 2015).

Mushroom cultivation begins with spawn production (Stanley and Awi-Waadu 2010; Thakur *et al.* 2023b). The production of spawn stands as a critical stage in the cultivation process of any mushroom. The quality of the spawn directly correlates with the robustness of basidiocarp yield. Acting as the inoculant, spawn is pivotal for fostering the vegetative growth of the mushroom. The production of shiitake spawn is contingent upon the selection of substrate type. Various materials, predominantly agricultural products, can be employed in the preparation of mushroom spawn. Commonly, wheat, rye, millet,

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sorghum, rice, and maize serve as primary substrates for spawn production (Stanley 2010). Healthy spawn leads to production of healthy fruit bodies and higher profits. Utilization of various agricultural and industrial byproducts or wastes as economic growth substrates has emerged as an attractive solution for mushroom production (Kuforiji and Fasidi 2008).

Keeping in view the importance and popularity of growing shiitake mushrooms in both rural and urban areas of the country, the experiment was conducted with the aim to study the effect of different organic additives (cereal brans, leguminous flours and oilseed cakes) on wheat grains for spawn growth of *Lentinula edodes*.

## Materials and Methods

### Pure culture of *Lentinula edodes*:

The pure culture of Shiitake mushroom (*Lentinula edodes*) was obtained from the Directorate of Mushroom Research Chambaghat, Solan. The culture was maintained on potato dextrose agar medium through successive sub-culturing after 15-20 days.

### Spawn Production:

The mother spawn of shiitake mushroom on wheat grains was prepared by using standard procedure. Wheat grain substrate were assessed by adding different organic additives viz., Cereal brans (Wheat, Rice and Corncob powder), Leguminous flour (Pea, Lentil and Soyabean) and Oilseed cake powder (Castor, Linseed and Mustard) at three different concentrations (2, 4 and 6 %) for their capacity to support the mycelial growth of shiitake mushrooms spawn. For spawn production, wheat grains were boiled at 100°C for 15-20 minutes or till the grains become soft, allowed to cool down for 24 hours at room temperature. Next day, the grains were mixed with calcium carbonate @ 0.1% and organic additives at 2, 4 and 6 per cent concentration on the wet weight basis of grains. The grains were filled in the glass bottles and plugged with non-absorbent cotton. Bottles were autoclaved at 121°C for 2 hours to sterilize the grains. Bottles were inoculated with pure culture under aseptic conditions in laminar air flow and incubated at 22 + 2°C. The linear growth of mycelium on each substrate was measured at 10, 20, 30 and 40 days' interval until complete colonization.

The parameters for spawn production were recorded as Linear Mycelial growth (mm) and Mycelial run rate (MRR)

### Mycelial Run Rate (MRR)

Mycelial run was recorded with the help linear scale and growth was observed in mm. MRR was calculated by using the formula given as under:

$$\text{MRR} = L/N (\text{mm/day})$$

Where, L = Length of the mycelium in mm

N = Number of days

### Statistical analysis

The statistical analysis of the collected data was done by using OPSTAT software.

## Results and Discussions

### Effect of cereal bran as additive on linear mycelial growth of *Lentinula edodes* on wheat grains

The effect of cereal brans viz., wheat, rice and corncob powder at three concentrations (2, 4 and 6 %) on spawn production of *Lentinula edodes* (Table 1). Irrespective of days, the maximum mean linear mycelial growth (68.06 mm) was observed in the grains supplemented with wheat bran followed by corn cob powder (67.89 mm) at 6 per cent concentration which were statistically similar with each other. Among different concentrations of supplements tested maximum growth was observed at 6 per cent concentration followed by 4 per cent which were significantly different from each other. However, all mean values of linear mycelial growth were observed to be superior with respect to control (42.08 mm). Irrespective of the concentration of different organic additive, the maximum mean linear growth was observed after 60 days i.e. 104.43 mm and minimum after 10 days (20.55mm).

The maximum linear growth (119.67 mm) after 60 days of incubation was observed in the grains supplemented with wheat bran as additive at 6 per cent concentration followed by grains at 4 per cent concentration (116.33 mm) with wheat bran and rice bran (116.00 mm) at 6 per cent concentration which were statistically different from each other and control (Table 1). Minimum growth was recorded in control (74.33 mm) after 60 days of incubation. Observations were recorded up to 60 days of incubation but few treatments required more time to for completing linear growth of *L. edodes*. Additionally, the interaction

**Table 1. Supplementation of cereal bran additives on wheat grain spawn and its effect on linear growth of *Lentinula edodes***

Bran	Concentration (%)	Linear mycelial growth (mm/day)						Mean
		10	20	30	40	50	60	
Wheat	2	17.33	25.33	43.33	62.67	86.33	107.33	57.06
	4	19.33	32.67	49.67	73.67	96.00	116.33	64.61
	6	22.00	35.67	55.00	79.00	97.00	119.67	68.06
Rice	2	18.67	21.67	43.33	61.67	75.00	91.67	52.00
	4	21.33	23.67	49.67	65.33	88.33	112.00	60.06
	6	23.33	25.33	55.00	72.33	94.00	116.00	64.33
Corncob	2	23.33	35.67	47.33	64.33	81.33	93.33	57.56
	4	24.67	38.33	56.67	67.33	85.00	101.00	62.17
	6	27.00	39.00	63.33	74.33	91.00	112.67	67.89
Control		8.83	21.00	35.33	48.67	64.33	74.33	42.08
Mean		20.58	29.83	49.87	66.93	85.83	104.43	
Factors		C.D.	S.E.(d)					
Factor A		1.32	0.66					
Factor B		1.02	0.51					
Interaction(A×B)		3.22	1.62					

effect (A×B) of bran concentration and substrate type further influenced fungal growth and suggests that the combined effect of these factors differs from their individual effects. Results revealed that the supplementation of the wheat grains with organic brans additives help to increase the linear mycelial growth of the spawn as compared to grains without additives.

#### **Effect of cereal bran additive on mycelial run rate of *Lentinula edodes* on wheat grain**

The mycelial run rate of *Lentinula edodes* starting from day 10 to 60 of incubation. It was observed that fortification with different cereal brans had varied effect on mycelial run rate of *L. edodes* (Table 2). Irrespective of days of incubation, mean maximum mycelial run rate was observed in the treatment

**Table 2. Supplementation of cereal bran additives on wheat grain spawn and its effect on mycelial run rate (MRR) of *Lentinula edodes***

Brans	Concentration (%)	MRR (mm/day)						Mean
		10	20	30	40	50	60	
Wheat	2	1.73	1.27	1.44	1.57	1.73	1.79	1.59
	4	1.93	1.63	1.66	1.84	1.92	1.94	1.82
	6	2.20	1.78	1.83	1.98	1.94	1.99	1.95
Rice	2	1.87	1.08	1.44	1.54	1.50	1.53	1.49
	4	2.13	1.18	1.66	1.64	1.77	1.87	1.71
	6	2.33	1.27	1.83	1.81	1.88	1.93	1.84
Corncob	2	2.33	1.78	1.58	1.61	1.63	1.56	1.75
	4	2.47	1.92	1.89	1.69	1.70	1.68	1.89
	6	2.70	1.95	2.11	1.86	1.82	1.88	2.05
Control		0.88	1.05	1.18	1.22	1.29	1.24	1.14
Mean		2.06	1.49	1.66	1.68	1.72	1.74	
Factors		C.D.	S.E.(d)					
Factor A		0.05	0.02					
Factor B		0.04	0.02					
Interaction (A×B)		0.12	0.06					

supplemented with corn cob powder (2.05 mm/day) at 6 per cent concentration followed by wheat bran (1.95 mm/day) at 6 per cent concentration which were statistically different from each other and lowest mean mycelial run rate was observed in control (1.14 mm/day). Irrespective of the concentrations of different organic additives, the mean mycelial growth was observed after 10 days of incubation (2.06 mm/day) followed by 60 days (1.74 mm/day) and minimum after 20 days of incubation (1.49 mm/day) which were significantly different from each other.

The maximum mycelial run rate was observed in corncob powder (2.70 mm/day) at 6 per cent concentration followed by 4 per cent corncob powder (2.47 mm/day) whereas, minimum mycelial run rate was observed in control (0.88 mm/day) after 10 days of incubation which was significantly different from each other (Table 2). It was concluded from the data that up to 10 days of incubation the mycelial run rate in all the treatments at all concentration started to decline, but after 20 days of incubation, run rate again started to increase. The increase in run rate after 60 days was statistically inferior to the values observed after 10 days of incubation. In case of control the mycelial run rate increased up to 50 days of incubation and then declined. Interaction of additives with wheat grains had a positive impact on mycelial run rate of

shiitake mushroom.

#### Effect of leguminous flour as additive on linear growth of *Lentinula edodes* on wheat grains

The effect of legume flour viz., pea, lentil and soybean flour at three concentrations (2, 4 and 6 %) on spawn production of *Lentinula edodes* (Table 3). Irrespective of days, the maximum mean linear mycelial growth (82.22 mm) was observed in the grains supplemented with pea flour followed by lentil flour (79.11 mm) at 6 per cent which were statistically different from each other. Among different concentration of supplements tested maximum growth was observed at 6 per cent concentration followed by 4 per cent which were significantly different from each other. However, all mean values of linear mycelial growth were observed to be superior with respect to control (42.08 mm). Irrespective of the concentration of different organic additive, the maximum mean linear growth was observed after 60 days i.e. 108.60 mm and minimum after 10 days (26.75mm).

It was observed that after 60 days of incubation maximum linear growth (119.00 mm) was observed in the grains supplemented with pea flour as additive at 6 per cent concentration followed by grains at 6 per cent concentration of lentil flour (117.33 mm) which were significantly different from each other and statistically at par with 4 per cent concentration (117.00 mm) of pea

**Table 3. Supplementation of leguminous flour additives on wheat grain spawn and its effect on linear growth of *Lentinula edodes***

Flour	Concentration (%)	Linear mycelial growth (mm/day)						Mean
		10	20	30	40	50	60	
Pea	2	24.00	35.67	45.67	76.33	93.33	114.33	64.89
	4	33.67	60.67	70.00	81.33	96.33	117.00	76.50
	6	34.33	64.67	82.00	86.00	107.33	119.00	82.22
Lentil	2	28.67	46.00	62.33	77.00	93.67	110.67	69.72
	4	31.00	56.00	71.00	81.00	104.33	115.67	76.50
	6	31.33	59.00	74.00	86.33	106.67	117.33	79.11
Soybean	2	21.00	41.33	54.00	64.00	76.67	96.67	58.94
	4	24.67	56.00	72.33	84.33	97.33	105.67	73.39
	6	30.00	56.67	77.00	88.00	107.00	115.33	79.00
Control		8.83	21.00	35.33	48.67	64.33	74.33	42.08
Mean		26.75	49.70	64.37	77.30	94.70	108.60	
Factors		<b>C.D.</b>	<b>S.E.(d)</b>					
Factor A		1.69	0.85					
Factor B		1.31	0.66					
Interaction(A×B)		4.14	2.09					

flour. Minimum growth was recorded in control (74.33 mm) after 60 days of incubation. Observations were recorded up to 60 days of incubation but few treatments required more time for completing linear growth of *L. edodes*. Additionally, the interaction effect (A×B) of legume flour concentration and substrate type further influenced fungal growth and suggests that the combined effect of these factors differs from their individual effects. Results revealed that the supplementation of the wheat grains with organic legume flour as additives help to increase the linear mycelial growth of the spawn as compared to grains without additives.

#### Effect of leguminous flour as additive on mycelial run rate of *Lentinula edodes*

It was observed that fortification with different legume flour had varied effect on mycelial run rate of *L. edodes* (Table 4). Irrespective of days of incubation, mean maximum mycelial run rate was observed in pea flour (2.61 mm/day) followed by lentil flour (2.47 mm/day) at 6 per cent concentration which were statistically different from each other and lowest mean mycelial run rate was observed in control (1.14 mm/day). Irrespective of the concentrations of different organic additives, the maximum mean mycelial growth was observed after 10 days of incubation (2.68 mm/day) followed by 20 days (2.49

mm/day) and minimum after 60 days of incubation (1.81 mm/day) which were significantly different from each other.

Body of the table revealed that the maximum mycelial run rate was observed in pea flour (3.43 mm/day) at 6 per cent concentration followed by 4 per cent (3.37 mm/day) whereas, minimum mycelial run rate was observed in control (0.88 mm/day) after 10 days of incubation which was significantly less as compare to treatments. It was concluded from the data that up to 10 days of incubation the mycelial run rate in all the treatments at all concentration increases, but after 20 days of incubation, mycelial run rate started to decline. The decrease in run rate after 60 days was statistically inferior to the values observed after 10 days of incubation. In case of control the mycelial run rate increased up to 50 days of incubation and then declined. Interaction of legume flour additives with wheat grains had a positive impact on mycelial run rate of shiitake mushroom.

#### Effect of oilseed cakes as additive on linear growth of *Lentinula edodes*

Irrespective of days, the maximum mean linear mycelial growth (72.28 mm) was observed in the grains supplemented with castor oilseed cake followed by linseed oilseed cake (72.00 mm) at 6 per cent concentration which were statistically similar with

**Table 4. Supplementation of leguminous flour additives on wheat grain spawn and its effect on mycelial run rate of *Lentinula edodes***

Flour	Concentration (%)	MRR (mm/day)						Mean
		10	20	30	40	50	60	
Pea	2	2.40	1.78	1.52	1.91	1.87	1.91	1.90
	4	3.37	3.03	2.33	2.03	1.93	1.95	2.44
	6	3.43	3.23	2.73	2.15	2.15	1.98	2.61
Lentil	2	2.87	2.30	2.08	1.93	1.87	1.84	2.15
	4	3.10	2.80	2.36	2.03	2.09	1.93	2.38
	6	3.13	2.95	2.47	2.16	2.13	1.95	2.47
Soybean	2	2.10	2.07	1.80	1.60	1.53	1.61	1.79
	4	2.47	2.80	2.41	2.11	1.95	1.76	2.25
	6	3.00	2.83	2.57	2.20	2.14	1.92	2.44
Control		0.88	1.05	1.18	1.22	1.29	1.24	1.14
Mean		2.68	2.49	2.15	1.94	1.89	1.81	
Factors		C.D.	S.E.(d)					
Factor A		0.05	0.03					
Factor B		0.04	0.02					
Interaction(A×B)		0.12	0.06					



each other. Among different concentration of supplements tested maximum growth was observed at 6 per cent concentration followed by 4 per cent which was significantly different from each other. However, all mean values of linear mycelial growth were observed to be superior with respect to control (42.08 mm). But in case of mustard oilseed cake as additive, the maximum linear growth was observed at 2 per cent concentration (61.17 mm) followed by 6 per cent (50.78 mm) and minimum at 4 per cent (49.72 mm) which was significantly different from each other. Irrespective of the concentration of different oilseed cakes, the maximum mean linear growth was observed after 60 days i.e. 103.83 mm and minimum after 10 days (12.62 mm).

After 60 days of incubation maximum mycelial growth (119.33 mm) was observed in the grains supplemented with linseed as additive at 6 per cent concentration followed by grains at 6 per cent concentration of linseed cake (119.00 mm) which were similar with each other and different from mycelial growth on linseed cake at 4 per cent concentration (118.33 mm). Minimum growth was recorded in mustard at 4 per cent and control (74.33 mm) after 60 days of incubation which was statistically similar with each other. Observations were recorded up to 60 days of incubation but few treatments required more time to

for completing linear growth of *L. edodes*. Additionally, the interaction effect (A×B) of oilseed cakes concentration and substrate type further influenced fungal growth and suggests that the combined effect of these factors differs from their individual effects. Results revealed that the supplementation of the wheat grains with oilseed cakes as additives help to increase the linear mycelial growth of the spawn as compared to grains without additives.

#### **Effect of oilseed cakes as additive on mycelial run rate of *Lentinula edodes***

It was observed that fortification with different oilseed cakes had varied effect on mycelial run rate of *L. edodes* (Table 6). Irrespective of days of incubation, mean maximum mycelial run rate was observed in castor oilseed cake (1.98 mm/day) followed by linseed (1.96 mm/day) at 6 per cent concentration which were statistically similar with each other and lowest mean mycelial run rate was observed in control (1.14 mm/day). Irrespective of the concentrations of different organic additives, the maximum mean mycelial growth was observed after 40 days of incubation (1.88 mm/day) followed by 50 days (1.85 mm/day) and minimum after 10 days of incubation (1.26 mm/day) which were significantly different from each other.

The maximum mycelial run rate was observed in

**Table 5. Supplementation of oilseed cake additives on wheat grain spawn and its effect on linear growth of *Lentinula edodes***

Oilseed cake	Concentration (%)	Linear mycelial growth (mm/day)						Mean
		10	20	30	40	50	60	
Castor	2	12.67	31.67	54.67	71.00	91.00	111.33	62.06
	4	14.67	35.00	55.33	92.67	106.00	116.67	70.06
	6	15.00	36.67	61.33	93.33	108.33	119.00	72.28
Linseed	2	11.67	29.33	46.33	76.67	102.33	116.33	63.78
	4	13.00	34.67	56.00	86.67	105.33	118.33	69.00
	6	13.67	37.33	62.00	93.33	106.33	119.33	72.00
Mustard	2	10.67	28.00	46.00	70.00	99.00	113.33	61.17
	4	12.00	33.00	47.67	61.33	70.00	74.33	49.72
	6	14.00	36.67	49.67	58.00	71.00	75.33	50.78
Control		8.83	21.00	35.33	48.67	64.33	74.33	42.08
Mean		12.62	32.33	51.43	75.17	92.37	103.83	
Factors		C.D.	S.E.(d)					
Factor A		0.90	0.46					
Factor B		0.70	0.35					
Interaction(A×B)		2.21	1.12					

**Table 6. Supplementation of oilseed cakes additives on wheat grain spawn and its effect on mycelial run rate of *Lentinula edodes***

Oilseed cake	Concentration (%)	MRR (mm/day)					Mean	
		10	20	30	40	50	60	
Castor	2	1.27	1.58	1.82	1.78	1.82	1.85	1.69
	4	1.47	1.75	1.84	2.32	2.12	1.95	1.91
	6	1.50	1.83	2.05	<b>2.34</b>	2.17	1.98	1.98
Linseed	2	1.17	1.47	1.55	1.92	2.05	1.94	1.68
	4	1.30	1.73	1.87	2.17	2.11	1.97	1.86
	6	1.37	1.87	2.07	<b>2.34</b>	2.13	1.99	1.96
Mustard	2	1.07	1.40	1.53	1.75	1.98	1.89	1.60
	4	1.20	1.65	1.59	1.53	1.40	1.24	1.44
	6	1.40	1.83	1.66	1.45	1.42	1.26	1.50
Control		0.88	1.05	1.18	1.22	1.29	1.24	1.14
Mean		1.26	1.62	1.72	1.88	1.85	1.73	
Factors		<b>C.D.</b>	<b>S.E.(d)</b>					
Factor A		0.03	0.01					
Factor B		0.02	0.01					
Interaction(A×B)		0.07	0.03					

castor and linseed oilseed cake (2.34 mm/day) at 6 per cent concentration followed by castor oilseed cake at 4 per cent (2.32 mm/day) after 40 days of incubation whereas, minimum mycelial run rate was observed in control (0.88 mm/day) after 10 days of incubation which was significantly different from each other. It was concluded from the data that up to 40 days of incubation the mycelial run rate in all the treatments at all concentration increases, but after 50 days of incubation, run rate again started to decline. The decrease in run rate after 60 days was statistically inferior to the values observed after 10 days of incubation. But in mustard, at 2 per cent concentration mycelial run rate increased upto 50 days (1.98 mm) and then started to declines whereas, at 4 and 6 per cent the run rate (1.65 mm and 1.83 mm, respectively) increased upto 20 days and then started to decline. In case of control the mycelial run rate increased (1.29 mm) up to 50 days of incubation and then declined. Interaction of oilseed cakes as additives with wheat grains had a positive impact on mycelial run rate of shiitake mushroom except in mustard oilseed cake where increase in additive concentration led to decrease in mean mycelial run rate of spawn.

Our results were in accordance with Singh *et al.* (2017a) who reported that supplementation is best for the spawn production of *P. djamor*, the maximum

mycelial growth (100.00 mm) observed in pigeon pea flour @ 2 per cent followed by black gram flour (87.67 mm) @2 per cent concentration after 20 days and minimum growth in control (72.33 mm). Singh *et al.* (2017b) found that pigeon pea flour required lesser days (16 days) followed by black gram (18.33 days) for spawn run as compare to control (25.00 days) in *P. djamor*. Similarly, Kumar (2019) found maximum spawn growth in rice powder (90.00 mm) at 1 per cent concentration which was followed by pigeon pea (89.33 mm) and pea powder (85.33 mm) and minimum growth in the control (74.00 mm) after 20 days in CI-17-08 strain of *Calocybe indica*. Kumar *et al.* (2020) concluded that the maximum mycelial growth for spawn production in organic additive, pigeon pea (90.00 mm) and rice powder (90.00 mm) at 2 per cent concentration flour in He-02 strains of *Hericium* spp. after 20 days. Singh *et al.* (2021) also reported pigeon pea flour (89.67 mm) gave the maximum spawn growth followed by green gram (84.33 mm) and pea flour (82.00 mm) and in cereal flour, maximum mycelial growth was found in maize flour (89.33 mm) followed by barley flour (88.33 mm) as compare to control (75.00 mm) after 9 days in *Cordyceps militaris*. Thakur *et al.* (2023b) observed that supplementation helped to enhanced the spawn growth as well as minimize the days of spawn growth and concluded that

maximum mycelial growth in wheat grains with wheat flour additive at 6 per cent (120.40 mm) as compare to control (85.60 mm) after 13 days in *Pleurotus ostreatus*.

### Conclusion

The data indicates a notable impact of both additives and their respective concentrations (%) on the mycelial growth observed in the examined substrates. Among the various organic additives tested across different concentrations, the most effective additive was pea flour, particularly when used at the highest treatment concentration of 6 percent followed closely by lentil and soybean at same concentration.

The study concluded that the incorporation of leguminous flour as an additive significantly facilitated rapid mycelial growth. Additionally, supplementation was identified as the optimal approach for expediting spawn production.

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