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Yield Performance and Economics of Grain Based Spawn on *Tricholoma giganteum*

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Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

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ABSTRACT

Earlier only *Pleurotus* spp., *Agaricus bisporus, Volvariella volvacea* and *Calocybe indica* were cultivated by the growers in India. The sequence of whole year mushroom production breaks due to unavailability of mushroom for production during summer season except *Calocybe indica* and *Volvariella volvacea,* but due to the pungent and bitter taste of *Calocybe indica* and bad shelf life of *Volvariella volvace*a are not accepted successfully by the farmers. *Tricholoma giganteum* was proved the best mushroom for summer season having a pleasant taste and good shelf life as compared to *C. indica* and *V. volvacea* and at par of Button mushroom (*Agaricus bisporus*).

Considering the above facts 5 local isolates of *T. giganteum* were tested for yield performance. The present studies include yield performance and economics of grains based spawn on *Tricholoma giganteum*. Altogether five isolates [CIP-12, CIP-17, CIP-18, CIP-19 and CIP-20] of *T. giganteum* and one isolate [CI-01] of *Calocybe indica* as a control were evaluated against the effect of two different grains [wheat and maize grains] for spawn production and their yield performance and economics of production was also studied.

Out of two grain substrate i.e. maize and wheat grain, wheat grain was observed most suitable substrate for spawn production with minimum days for complete colonization observed by CIP-19 and CIP-20 (13 days) followed by CIP-18 (14 days), CIP-12 (15 days), CIP-17 (16 days) and CI-01 (17 days), However complete colonization on maize grain substrate took too much time as compare to wheat grain substrate with minimum 23 days and maximum 27 days for complete colonisation.

Maximum yield was observed on wheat grain-based spawn followed by maize grain-based by all *T. giganteum* isolates. As regard wheat grain-based spawn CIP-20 gave maximum yield 76.4 kg /100 kg dry substrate, followed by CIP-19 and CIP-17, CIP-12, CIP-18 and minimum yield was obtained

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by CI-01 (58 kg /100 kg dry substrates), However, yield performance of maize grain-based spawn, CIP-12 gave maximum yield 57.2 kg /100 kg dry substrate followed by CIP-20, CIP-17, CIP-18, CIP-19, and minimum yield was recorded by CI-01 (50.2 kg /100 kg dry substrate).

Keywords: Maize grain; spawn; Tricholoma giganteum; wheat grain; yield performance.

1. INTRODUCTION

Mushroom have prized as the food of God on account of their special flavour, nutritive value and medicinal property. It can be grown on all types of plants and agricultural waste and thus constitute a highly nutritive source of food at low cost [1].

India is a traditional producer of native wild mushroom called 'Gucchi' (*Morchella spp*.) and 'Dhingri' (*Pleurotus spp*.).

1.1 Definition

Chang and Miles [2] defined, "The mushroom is a macrofungus with a distinctive fruiting body which can be either epigeous or hypogynous and large enough to be seen with the naked eye and to be picked by hand." It belongs to class basidiomycetes, and ascomycetes, saprophytic, parasitic, and symbiotic but mostly cultivated mushroom are saprophytic.

In India, commercial cultivation of mushrooms had been with the joint effort of scientists and farmers. Annual mushroom production has increased to 80,000 ton in 2006 from a mere 1,000 ton in 1981. Fifty percent of this is produced by marginal and small production units and the rest by industrial establishments. The major producers of mushrooms are Punjab (35,000 MT) Tamilnadu (15,000MT), and Andhra Pradesh (5000MT). Mushroom production of Uttarakhand alone increased from 2,640MT in 2000 to 5340MT in 2006, with Dehradun, Nainital, Haridwar and Udham Singh Nagar the major production centres. Button mushroom (*Agaricus bisporus*) constitutes about 90% of total production in India where that of other cultivated mushrooms viz. *Pleurotus, Lentinula, Auricularia* and *Calocybe* are very marginal [3].

India has achieved food security by producing over 257 million tonnes of food grain per annum. However, our struggle for nutritional security is still on. During the coming decades the increasing population depleting agricultural land, environmental issues. Water shortage and need for quality food are future challenges (Anonymous, 2013).

Tricholoma giganteum (Syn-*Macrocybe giganteum*), widely distributed in tropics of Asia and Africa. It is pure white resembling the morphology of *Calocybe indica.* It is larger than *Calocybe indica* and fleshier than *Agaricus bisporous*.

It is edible mushroom with 12-32 cm fruiting body diameter. It has good taste, nice aroma and crisp texture. Its fruiting body is rich in protein, polysaccharides, dietary fibre, mineral salt, vitamin and other healthful substances [4].

Modern pharmacological studies have shown that *Tricholoma giganteum* has antifungal [5], It has a novel angiotensin I converting inhibitory enzyme peptide [6] and can inhibit HIV-I reverse transcriptase [7].

The shelf life of *Tricholoma giganteum* is 3-4 days under room temperature and 5-6 days under refrigerated condition. It recorded a significantly higher yield with bio efficiency % of 164 to 174% [8].

The sequence of whole year mushroom production breaks due to unavailability of mushroom for production during summer because there is no any mushroom available for cultivation during summer season except *Calocybe indica* and *Volvariella volvacea,* but due to pungent and bitter taste of *Calocybe indica* and bad shelf life of *Volvariella volvacea* are not accepted successfully by the farmers. *T. giganteum* was proved the best mushroom for summer season having a pleasant taste and good shelf life as compared to *C. indica* and *V. volvacea* and at par of Button mushroom (*Agaricus bisporus*).

Keeping in view the nutritional and medicinal importance of this mushroom an experiment on "Yield Performance and Economics of Grain Based Spawn on *Tricholoma giganteum*" was conducted at RPCAU, Pusa, Samastipur, Bihar.

2. MATERIALS AND METHODS

2.1 Collection of Isolates

Five isolates of *Tricholoma giganteum* i.e. CIP-12, CIP-17, CIP-18 CIP-19, CIP-20 and one

isolate of *Calocybe indica* used as a control were obtained from Mushroom Centre, FBS&H Department of Microbiology, Dr. Rajendra Prasad Central Agricultural University, Pusa, Samastipur, Bihar, maintained on PDA (potato dextrose agar) medium at 30±2°C temperature.

2.2 Experimental Site

The research experiments were conducted in Mushroom Centre, FBS&H Department of Microbiology, Dr. Rajendra Prasad Central Agricultural University, Pusa, Samastipur, Bihar.

2.3 Washing and Sterilization

During the experimental work, BOROSIL glassware was used. The glass wares prior to use were washed with water and detergent powder followed by rinsing with tap water. The glass wares were dried and sterilised in an oven at 180°C for 2 hours. The inoculation needle, knife, scissors, and blades were sterilised by dipping them in 95% alcohol followed by heating over the flame.

2.4 Spawn Production

Preparation of starter spawn: Starter spawn was prepared from pure cultures of five isolates of *Tricholoma giganteum* [CIP-12, CIP-17, CIP-18 CIP-19 and CIP-20] and one strain of *Calocybe indica* [CI-01]. Clean healthy and bold wheat grain were taken then properly washed and dipped in water for 10-12 hours. The soaked grain were cooked for 10-15 minutes or till they become soft but not to be split or broken. Thereafter, the excess water was drained, and grains were spread on muslin cloth to eliminate excess moisture. In cooled grains, Calcium carbonate and Gypsum (1 and 2% w/w respectively) were mixed thoroughly. These grains were filled in the conical flask (250 ml) and plugged with non-absorbent cotton. Each flask contains 100 g grains and sterilised at 20 lbs psi for 2 hours. After cooling, take out the vessels from the autoclave and shake it properly to avoid clump formation. Inoculation was done with 7 days old culture growing mycelium of five strains of *T. giganteum* and one strain of *C. indica* then incubated at 30 ±2°C till the mycelium covered the entire grain surface.

Preparation of mother spawn: Processed and filled the wheat grains in flask or glucose bottle in similar to starter spawn. Thereafter these bags were sterilised. After sterilisation cooled bags were aseptically inoculated with 10-15 gm of starter spawn in laminar air flow and incubated at30 ±2°C till the mycelium covered the grain surface.

For planting spawn, preparation wheat and maize grains were used as the substrate. Wheat and maize grains were processed and filled in polypropylene bag (7" × 16" -150 gauges) bag in similar to starter spawn. Each bag contained half or one kg wheat grain substrates and thereafter these bags were sterilised. After cooling bags were aseptically inoculated with 25-30 gm of mother spawn in laminar air flow and incubated at 30 ± 2°C and frequently examined for any types of contamination. The observations were recorded for 100% mycelial growth on grains surface. There are 3 replications were kept in each treatment.

Preparation of substrates: The wheat straw was used as a substrate for cultivation of all isolates. It was dipped in water for overnight Thereafter, excess water was drained off the next morning and pasteurise the substrates by boiling it into the water for 30-35 minutes at 60°C temperature. Substrates were spread over cemented floor or polythene sheet or tabletop to drain off the excess water or till the moisture content of straw remained 65-70 per cent.

Spawning: Spawning was done by thoroughly mixing of spawn and substrates at the rate of 1.75 kg spawn/10 kg substrates (dry wt. basis). The spawned substrate was filled in polypropylene bags and mouth of the bag was covered with newspaper. The spawned bags were kept in a mushroom growing room, where appropriate temperature (30-35°C) and relative humidity (70-80 per cent) were maintained by a frequent sprinkling of water on walls and floor. After complete colonisation of the substrate by mushroom mycelium (spawn run), the casing was done to promote sporophore production. Casing layer was usually 2.0 to 5.0 cm deep and usually applied approximately after 2 weeks of spawning.

Meteorology of crop house: Crop house wall was covered with gunny bag. Temperature and moisture level of crop house was recorded on daily basis (6 AM and 2 PM) thereafter watering was done on gunny bag and spawned bag to maintain the temperature and moisture level of crop house.

Picking of the mushroom: Picking was done before margin twisting and colour change by slight twisting and pulling of sporophores. Three flushes were harvested from the same bed at an interval of 7-10 days.

2.5 Weighing of Sporophore

The freshly harvested sporophore was immediately weighed with the help of single pan balance with a sensitivity of 1 g.

2.6 Yield of Mushroom

The cumulative yield in each replication was recorded by summing up the fresh weight at the number of picking, which was collected as weight (kg) per 5 kg of the substrate, later on, change this value into weight (kg) per 100 kg of dry substrates.

2.7 Evaluation of Spawn Raised on Different Grains for Yield Performance

The effect of two grain-based spawn [wheat and maize grains] on different isolates were studied on the wheat straw substrate. The observations were recorded for spawn run, pinhead initiation, and another parameter i.e. pileus diameter, stipe length, avg. wt. of sporophore and yield (kg/100 kg substrates) of *T. giganteum.*

2.8 Statistical Analysis

All the experiments were evaluated with completely randomised design (CRD) and the data were statistical analysis using online software OPSTAT. The value of the standard error of the mean (SEm) and critical difference (C.D.) was calculated at 5% levels.

3. RESULTS AND DISCUSSION

3.1 Evaluation of Different Grain Substrate for Spawn Production of *Tricholoma giganteum*

Out of two-grain substrate [maize and wheat grains], wheat grain was observed the most suitable substrate for spawn production and yield potential of *Tricholoma giganteum* Massee presented in Table 1.

Complete colonization on wheat grain substrate was observed in ascending order i.e. CIP-19 and CIP-20 (13 days), followed by CIP-18 (14 days), CIP-12 (15 days), CIP-17 (16 days) and strain CI-01 (17 days), However complete colonization on maize grain substrate in ascending order CIP-19 and CIP-17 (23 days), CIP-18 (24 days), CIP-20 (24 days), CIP-12 (25 days) and CI-01 (27 days).

The present finding corroborates the finding of Prasuna [9] who studied different grain substrates for spawn production of *C. indica* and reported that early spawn development on wheat grain followed by jowar grains.

But contrary to Senthilnambi et al. [10] who reported that sorghum grains were the most suitable substrate for *C. indica* which took only 13.7 days for hundred percent mycelial growths. The maize grain substrate took 19 days for complete spawn run and recorded low yield when compared to other spawn substrates. Similarly Lakshmipathy et al. [11] and Krishnamoorthy, and Balan [12], where they have reported the best growth on sorghum grain.

3.2 Effect of Wheat and Maize Grain-Based Spawn on Yield Performance of Different Isolates of *Tricholoma giganteum*

The effect of two grain-based spawn viz: - wheat and maize grain on different strains was studied. It was observed that both grain-based spawn supported the yield parameters of all strains of *T. giganteum* Massee. Maximum yield was observed on wheat grain-based spawn by *T. giganteum* strains. The performance of each strain was as follows; - Maximum yield was obtained by CIP-20 (76.4 kg /100 kg substrate), followed by CIP-19 and CIP-17 which gave the same amount of yield (71.4 kg fruits/100kg substrates), then CIP-12 (70.8 kg /100 kg substrate), CIP-19 (67.2 kg /100 kg substrates) and minimum yield was obtained by CI-01 (58 kg /100 kg substrates) which was presented in Table 2.

As regard yield performance of maize grain based spawn on different strains of *T. giganteum* Massee it was observed as follows: - Maximum yield was obtained by CIP-12 (57.2 kg /100 kg substrate, followed by CIP-20 (52.6 kg /100 kg substrate), CIP-17 (52.2 kg /100 kg substrate), CIP-18 (51.6 kg /100 kg substrate), CIP-19 (51.4 kg /100 kg substrate) and minimum yield was obtained by CI-01 (50.2 kg /100 kg substrate) which was presented in Table 3.

Table 1. Evaluation of different grain substrate for spawn production of *T. giganteum*

Grains	Complete spawn development by strains (days)*						Mean
	CI-01	CIP-12	CIP-17	CIP-18	CIP-19	CIP-20	
Wheat	17.00	15.00	16.00	14.00	13.00	13.00	14.66
Maize	27.00	25.00	23.00	24.00	23.00	24.00	24.33
Mean	22.00	20.00	19.50	19.00	18.00	18.50	
	Factors		C.D (5%)	SE(d)	SEm±		
	Grains		0.531	0.255	0.180		
	Strains		0.920	0.441	0.312		
	Grains × Strains		1.302	0.624	0.441		

(*) - Average of three replications

Fig. 1. Spawn production of *T. giganteum* isolates on wheat grain

Fig. 2. Spawn production of *T. giganteum* isolates on maize grain

Table 2. Effect of wheat grain-based spawn on yield performance of different isolates of *Tricholoma giganteum*

Strains	Yield kg/100 kg dry wheat straw*					
	SRD	PHID	Pileus diameter (cm)	Stipe length (cm)	Avg. wt. of sporophore (g)	Yield (kg)/ 100 kg substrate
CI-01	15	35	8.46	9.73	91.66	58.0
CIP-12	13	32	9.10	10.70	118.33	70.8
CIP-17	14	33	9.73	11.53	100.23	71.4
CIP-18	13	32	10.36	12.53	96.96	67.2
CIP-19	12	34	11.00	12.23	104.53	71.4
CIP-20	14	31	11.63	11.76	103.10	76.4
C.D (5%)	1.387	1.903	0.527	1.098	NS	0.399
SEm±	0.435	0.596	0.165	0.344	8.601	0.125
SE(d)	0.615	0.843	0.233	0.487	12.163	0.177

(*) : - Average of three replications.
SRD: - Spawn Run Days, PHID: - Pin head Initiation Days, NS: - Non significant

CI-01

CIP-12

CIP-17

CIP-18

CIP-19

CIP-20

Fig. 3. Yield performance of wheat grain based spawn on different isolates of *T. giganteum*

CI-01

CIP-12

CIP-17

CIP-18

CIP-19

CIP-20

Fig. 4. Yield performance of Maize grain based spawn on different isolates of *T. giganteum*

Table 3. Effect of maize grain-based spawn on yield performance of different isolates of *T. giganteum*

Strains	Yield kg/ 100 kg dry wheat straw*					
	SRD	PHID	Pileus diameter (cm)	Stipe length (cm)	Avg. wt. of sporophore (g)	Yield (kg)/ 100 kg substrate
CI-01	21	45	8.43	8.13	73.67	50.2
CIP-12	20	44	9.53	9.76	79.06	57.2
CIP-17	18	42	10.13	9.86	76.76	52.2
CIP-18	20	43	9.70	10.27	80.16	51.6
CIP-19	19	40	9.48	10.54	80.46	51.4
CIP-20	18	41	10.06	10.30	78.46	52.6
C.D (5%)	1.812	1.387	0.989	1.313	NS	0.180
SEm±	0.568	0.435	0.310	0.411	2.660	0.057
SE(d)	0.803	0.615	0.438	0.582	3.762	0.080

(*): - Average of three replications.
SRD: - Spawn Run Days, PHID: - Pin head Initiation Days, NS: - Non significant

Table 4. Meteorological data of crop house during cultivation of *T. giganteum*

Month- May 2017				
Date	Temperature (°C)		Relative humidity (%)	
	Minimum	Maximum	Minimum	Maximum
23-05-2017	29	33.1	50	55
24-05-2017	29.2	33.2	51	56
25-05-2017	28.7	32.2	53	56
26-05-2017	28.4	33.5	52	57
27-05-2017	27.2	33.2	52	56
28-05-2017	28	32.4	51	55
29-05-2017	27	32.2	55	58
30-05-2017	28.5	33.3	52	57
31-05-2017	29.2	33.6	54	58

Month- June 2017				
Date	Temperature (°C)		Relative humidity (%)	
	Minimum	Maximum	Minimum	Maximum
1-06-17	30.1	34.1	64	70
2-06-17	31.1	34.3	65	71
3-06-17	32.1	34.3	65	72
4-06-17	32.3	34.5	66	70
5-06-17	33.0	34.6	64	72
6-06-17	32	35.3	65	72
7-06-17	31	35.1	65	73
8-06-17	33	37.8	64	71
9-06-17	30	34.1	64	68

Month- June 2017				
Date	Temperature (°C)		Relative humidity (%)	
	Minimum	Maximum	Minimum	Maximum
10-06-17	30.2	34.5	65	69
11-06-17	30.3	34.6	65	71
12-06-17	30.2	34.7	62	72
13-06-17	30.4	37.5	64	73
14-06-17	30.5	35.6	65	73
15-06-17	32.2	35.4	64	73
16-06-17	32.3	35.4	65	69
17-06-17	32.5	35.6	63	70
18-06-17	32.3	36.2	64	71
19-06-17	32.7	36.1	65	72
20-06-17	33	36.2	67	72
21-06-17	33.1	36.2	65	72
22-06-17	33.2	36.3	66	74
23-06-17	33.5	36.4	64	71
24-06-17	33.4	36.4	65	71
25-06-17	33.3	36	66	71
26-06-17	33.2	36.1	64	72
27-06-17	31.2	36.3	64	72
28-06-17	32.3	36.3	65	73
29-06-17	32.4	36.2	65	74
30-06-17	32.6	36.4	66	74

Month- July 2017				
Date	Temperature (°C)		Relative humidity (%)	
	Minimum	Maximum	Minimum	Maximum
1-07-17	33	36.1	71	78
2-07-17	32.3	36.2	70	79
3-07-17	33.4	36.2	72	78
4-07-17	34	36.3	71	80
5-07-17	33.4	36.4	72	81
6-07-17	33.6	36.4	70	79
7-07-17	33.5	36.4	71	78
8-07-17	33.4	36.5	69	79
9-07-17	32.4	37.4	72	82
10-07-17	34.2	37.2	72	85
11-07-17	34.1	37.1	73	85
12-07-17	34.2	37.2	71	83
13-07-17	34.1	37.1	71	84
14-07-17	34.2	37.2	72	82
15-07-17	34.2	37.4	72	83
16-07-17	34.1	37.4	70	82
17-07-17	34.2	37.2	71	83
18-07-17	32.2	36.4	71	79
19-07-17	32.5	38.4	72	78
20-07-17	32.6	38.2	72	81
21-07-17	32.5	38.1	73	82
22-07-17	32.3	38.1	72	84
23-07-17	32.3	38.2	73	83
24-07-17	32.7	38.2	72	84
25-07-17	32.7	38.3	71	85

3.3 Economics of Production of *T. giganteum*

Benefit-Cost Ratio of Production of *Tricholoma giganteum:* In the present study the benefit-cost ratio is estimated at one quintal dry substrates and two and a half months (~75 days) of crop duration.

Table 5. Total expenditure

Item	Quantity	Rate (in Rs.)	Cost (in Rs.)
Wheat Straw	1q	500/q	500
Spawn	18kg	100/kg	1800
Gas	10 cylinders	886/cylinder	8860
Man Power	10	300/ man power	3000
Electric Supply Charge	-	-	7000
Marketing Expenditure	-	-	5000
Miscellaneous	-	-	5000
		Total	31,160

Total production - 60 kg (avg. production)
Sale price - 150 Rs./kg
Total income - 150 x 60 = Rs. 90,000
***Net profit** = Total income – Total expenditure*
= 90,000 – 31,160
*= **58,840.00***
*So, the **B:C** ratio = approximately **3:1** ratio.*

4. CONCLUSION

In the present study the benefit-cost ratio is estimated at one quintal dry substrates and two and a half months (~75 days) of crop duration, and the **B:C** ratio is approximately **3:1** ratio.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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