



RESEARCH ARTICLE

Assessment of substrate blends and organic supplements on the yield performance of *Calocybe indica* strains CI-22-09 and CI-22-10

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Abstract

Milky mushroom (*Calocybe indica*) is an indigenous tropical fungus that can be grown on a variety of agricultural leftovers. An experiment was carried out using various substrate and additive combinations to assess the efficacy of organic additives and lignocellulosic substrates as sources of carbohydrates, which are essential energy sources for fungal growth and may also improve the overall nutritional profile of the mushroom crop. A variety of lignocellulosic substrates, such as wheat straw and paddy straw, were evaluated in conjunction with organic additives, including rice flour (5 % and 10 %), maize flour (5 % and 10 %), sorghum flour (5 % and 10 %) and pearl millet flour (5 % and 10 %). Wheat straw alone was used as the reference. With a biological efficiency (BE) of 97.10 %, maize flour at 5 % produced the most (971.16 g/kg dry substrate), closely followed by maize flour at 10 % (970.00 g/kg dry substrate; 97.00 % BE). The mixture of rice straw and wheat straw in a 1:4 ratio yielded the best yield (740.00 g/kg dry substrate; 74.00 % BE) when substrate combinations were evaluated. This was followed by the 1:3 ratio (720.00 g/kg dry substrate; 72.00 % BE). The findings showed that adding organic carbohydrate sources to substrates greatly increased the output of sporophores in *C. indica* growth.

Keywords: biological efficiency; *Calocybe*; cereal flour; mushroom; organic supplements; substrate; yield

Introduction

The fruiting body of macro-fungi, mushrooms, can be either epigeous or hypogeous and since they lack chlorophyll, they are unable to create their own sustenance. Instead, they eat complex organic components that are present in both live and dead plant and animal tissues. Of them, five to six species have been grown on an industrial scale, while 100 species are grown experimentally. More than 380 lakh tons of mushrooms are produced annually in around 100 nations, including India, up from 49.09 lakh tonnes in 1994. China is currently one of the top producers, accounting for almost 80 % of the world's mushroom production (1). In 2016, India produced 1.29 lakh tonnes of mushrooms; by 2018, that amount had risen to 1.55 lakh tons. After button and oyster mushrooms and other mushrooms (8 %), milky mushrooms are now the third most widely farmed commercially in India (2). The five types of mushrooms that make up less than 1 % of the world's total production are button mushrooms (*Agaricus* spp.), oyster mushrooms (*Pleurotus* spp.), straw mushrooms (*Volvariella volvacea*), milky mushrooms (*Calocybe indica*) and shiitake mushrooms (*Lentinula edodes*). The tropical mushroom *C. indica*

was initially discovered in West Bengal, India (3). Its milky, white tint has earned it the nickname "milky mushroom" or "white summer mushroom" and is a large-sized, delicious mushroom reported in India (4, 5). Its white colour is appealing and it keeps well. It thrives at temperatures between 25 and 35 °C (6). It has gained popularity as an edible mushroom in recent years due to its high conversion rate, ability to grow on a variety of agricultural wastes and most importantly, suitability for value addition.

The nutritional value of dried milky mushrooms is as follows: protein 21.4 %, ash 13.1 % and carbohydrate 48.5 % per 100 g, while fresh milky mushrooms have a moisture content of 87.4 %, protein 2.75 %, fat 0.65 %, fiber 1.63 %, ash 1.28 % and carbohydrates 6.8 % (7, 8). In addition to the mineral salts that the human body needs, the fruit body contains 12 amino acids, copper 0.44 mg/100 g, manganese 0.36 mg/100 g, zinc 0.05 mg/100 g, iron 0.13 mg/100 g, calcium 0.51 mg/100 g, phosphorous 0.38 mg/100 g, potassium 1.35 mg/100 g and sodium 0.21 mg/100 g (6, 9). Vitamin A (0.275 mg/g), vitamin C (1.03 mg/100 g), vitamin E (tocopherol) (2.8 mg/g) and glutathione (0.025 n mole/g) are all present in good amounts in milky mushrooms. Higher temperatures (30–35 °C) and humidity conditions (over 85 %) are ideal for its growth (10, 11).

In addition to paddy straw, it may be cultivated in several agricultural wastes, such as groundnut haulm, sugarcane bagasse, maize, wheat straw, Niger and so forth. However, the substrates affect how productive the mushrooms. In this regard, an experiment was carried out to assess the effects of various combinations of lignocellulosic substrates and organic additives on the formation of milky mushrooms. This mushroom has several advantages over other mushrooms, including ease of cultivation, low cost, a very appealing fruiting body, good storage quality, a pleasing milk white colour, the capacity to grow at temperatures above 30, high biological efficiency (up to 90–100 %) and ease of handling after harvest. But since its production and culture have not been very successful thus far, it was thought necessary to revalidate the production process using lignocellulosic substrate that was readily available in western Uttar Pradesh.

Materials and Methods

Collection and maintenance of fungus

The current study was conducted at Sardar Vallabhbhai Patel University of Agriculture and Technology Mushroom Research Training Centre, Meerut, Uttar Pradesh. The mushroom culture was collected from Directorate of Mushroom research and kept on potato dextrose agar (PDA). Throughout the course of the investigation, the pure culture of the fungus was subculturing every two months and used for a variety of experiments.

Preparation of spawn

The mother spawn was made from a pure milky mushroom culture. As substrate, spawn was made from wheat, rice, sorghum and maize grains. The spawn was prepared using wheat grains as a substrate. Wheat grains that were bold, healthy and clean were chosen, cleaned and then boiled in a container until they were tender. To remove extra water, boiled grains were spread out on a fresh polythene sheet and allowed to air dry in the shade. To absorb extra moisture while preserving the grains' ideal pH, calcium carbonate was completely mixed with 2 % dry weight of wheat grain. A 500 mL bottle was filled three-quarters of the way with the boiled and treated grains, sealed with non-absorbent cotton and autoclaved for 2 hr at 121 °C, 22 psi., to obtain mother spawn. Pure cultures of milky mushrooms were added to the sterile wheat bottles and cultured for 15 days at 28 ± 2 °C.

Substrate preparation

Rice flour (5 % and 10 %), maize flour (5 % and 10 %), sorghum flour (5 % and 10 %) and pearl millet flour (5 % and 10 %) were the different cereal flour mixtures that were used in the experiment to determine their effects utilised as cereal flour and a control group including only cereal flour was taken. Before being used, the additives were sun-dried, ground into a powder and autoclaved for 20 min at 121 °C (15 lbs of pressure). Three replications of each of the seven treatments were used in this experiment. Under cultivation technology, the crop was described in depth. Before spawning, these cereal flours were individually combined with substrate (wheat straw) based on dry weight. In three replications and seven treatments, the inoculated bags were set up to produce sporophores. Total spawn run days, pinhead formation days, initial harvest days, number of pinhead initiation days, number of fruiting

bodies (NOFB) and average weight of fruiting body (Avg WOFB (g/FB)) and sporophores yield (g/kg on dry substrate) were all recorded.

For these tests, bright, high-quality wheat straw was the most often utilised substrate. It was chemically treated with 7.5 g of carbendazim and 100 mL of formalin per 100 L of water for 18 hr to soften it out. A polythene sheet was placed over the tank to stop the formalin from evaporating. Straw was then taken out of the tank and spread out on a cement floor that had been coated with 2 % formalin for 2 hr to eliminate any remaining water. This is now prepared for mixing with spawn at a rate of 4 % of the substrates' dry weight.

Spawning and spawn run

Following spawning, the mixture was transferred into polythene bags at a rate of 1.2 kg per bag; the bags were folded and stapled at the top. To ensure enough aeration, 8–10 tiny holes were then drilled into polythene bags using a nail. For the 21-day spawn run, the bags were stored in a dark room (12). A temperature of 28 ± 2 °C and a relative humidity of 80–85 % were maintained. After about 3 weeks, the whole spawn in the wheat straw turns into a white, compact mass covered in mycelial growth that sticks to the polythene bag. The following some precautions should be taken like the floor should be sterilised before the substrate is spread, the substrate should be kept between 60 and 65 % moist during the filling bags, the spawn should be pure, spread and mixed properly in the substrate, the spawn dose should be applied as advised and the hands should be cleaned with Dettol or alcohol while the bags are being filled.

Casing

After spawning for 18 ± 2 days, casing was finished. Every two years, casing mixtures were made by combining vermicompost, garden soil and farm yard manure (1:1:1 w/w). They were then covered for 48 to 72 hr and treated with a formalin (2 %) solution. Calcium carbonate and calcium sulphate were added to the casing to maintain its pH at 8.5. The surface of the substrate was covered with a homogeneous layer that was roughly 1.5 cm thick. The crop rooms' relative humidity was maintained between 80 and 90 % by misting it with water using a sprayer. Pinhead initiation was started ten to twelve days after casing.

Harvesting

After maturing, *C. indica* sporophores were gathered. The yields of the strains were compared over a seven-week harvest period. When the fruiting body started to appear, water was sprayed on the blocks. The doors and windows were opened for a short time each night to provide sufficient ventilation. Although the fruiting bodies need to be protected from the sun, some light (2500–3000 Lux) was permitted to encourage the development of fruiting bodies. The walls and floor of the room were treated with 0.1 % carbaryl or cypermethrin to keep insects out. Two sprays of carbendazim 0.01 % were administered to avoid fungal infection.

The BE was calculated using the following formula (13):

Biological efficiency (%) =

$$\frac{\text{Fresh weight mushroom (g)}}{\text{Dry weight of substrate (g)}} \times 100 \quad (\text{Eq. 1})$$

Crop room experiment

Effect of different cereal flours on sporophore yield of *Calocybe indica*

The purpose of the experiment was to determine the effects of various cereal flours on the NOF, the number of days for spawn run (DFSR), the number of days for first harvesting (DFFH), the number of days for cropping period (DFCP), the stipe length (SL (mm)), the pileus width (PW (mm)), the Avg WOFB (g) and the biological efficiency (BE (in percentage)). The mixture of different cereal flours, namely rice flour (5 % and 10 %), maize flour (5 % and 10 %), sorghum flour (5 % and 10 %) and pearl millet flour (5 % and 10 %), was used as a cereal flour. The control group was taken without any additives; the additives were sun-dried, powdered and sterilised in an autoclave at 121 °C (15 lbs pressure) for 20 min before use. The crop was taken as described in detail under cultivation technology.

Before spawning, these cereal flours were individually combined with substrate (wheat straw) based on dry weight. In 3 replications and 7 treatments, the inoculated bags were set up in the crop room to produce sporophores. Total spawn run days, pinhead formation days, initial harvest days, number of pinhead initiation days, NOFB and Avg WOFB (g/fb) and sporophores yield (g/kg on dry substrate) were all recorded.

Effect of different straws and their combination on sporophore yield of *Calocybe indica*

Various combinations of rice straw and wheat were employed. The following combinations were used to observe their impact on *C. indica* yield: rice straw + wheat straw (1:1), rice straw + wheat (1:2), rice straw + wheat straw (1:3), rice straw + wheat straw (1:4), rice straw + wheat straw (2:1), rice straw + wheat straw (3:1) and rice straw + wheat straw (4:1). Control rice and wheat straw were also used. Nine treatments with 3 replications were used in this investigation. Total spawn run days, pinhead formation days, initial harvest days, number of pinhead initiation days, NOFB, Avg WOFB (g/fb) and sporophores yield (g/kg on dry substrate) were all recorded.

Statistical analysis

Data that had been appropriately transformed were analysed using an analysis of variance for a complete randomised design (CRD) with 3 replications by OP Stat. The *F*-value was tested and the critical difference (CD) was calculated at a 5 % level of significance when comparing treatment means (14).

Results and Discussion

Effect of different cereal flours on sporophore yield

With a focus on the CI-22-09 strain, the experiment sought to determine how eight distinct grain flours affected the growth performance of two strains of milky mushrooms. The treatment that produced the shortest spawn run duration (17.33 days) was maize flour at 5 %. On the other hand, compared to all other treatments, sorghum flour at 10 % had the longest spawn run duration (22.16 days). The control treatment had the earliest time to first harvest (23.00 days). In contrast to all other treatments, maize flour at 5 % considerably postponed the first harvest (37.66 days). Maize flour at 5 % produced the greatest cropping period (85.33 days), while maize flour at 10 % produced the shortest cropping period (81.33 days), which was statistically less than the other treatments. In terms of yield characteristics, the control treatment generated the fewest fruiting bodies (15.33), whereas maize flour at 10 % produced the most no of fruiting bodies (22.66).

These findings show a definite trade-off between production time and yield potential. While 5 % maize flour increased spawn run efficiency and lengthened cropping time, it postponed the first harvest, which might not be the best option for growers looking for rapid profits. On the other hand, growers who prioritise quicker production cycles and early cash can benefit more from treatments that encourage earlier harvesting or larger fruiting bodies.

Comparing this to the other treatments, it was much lower. The wheat straw + rice flour at 5 % had the longest stipe (17.33 cm). However, compared to all other treatments, the smallest SL (13.00 cm) was found in wheat straw + pearl millet flour at 5 %. The wheat straw + pearl millet at 5 % showed the widest pileus (11.66 cm). However, wheat straw showed the lowest PW (7.00 cm), which was far smaller than any other treatment. Maize flour at 5 % produced the highest yield (971.16 g/kg of dry substrates with 97.10 % BE). However, the control showed the lowest yield (630 g/kg of dry substrates with 63 % BE). Comparing this to the other treatments, it was much lower (Table 1).

In contrast, the CI-22-10 strain required a minimum of 16.83 days for a spawn run in 5 % maize flour. The maximum spawn run days of 22.33 days were recorded in pearl millet flour at 10 %, which was noticeably longer than in any of the other treatments. Pearl millet at 5 % had the quickest initial harvesting time (26.16 days). However, maize flour at 10 % showed the largest number of DFFH (38.66 days), which was statistically greater than all other treatments. Sorghum flour at 5 % showed the longest cropping duration (85.16 days), whereas rice flour at 5 % showed the shortest DFPC (79.83 days), which was significantly less than all other treatments.

At 5 % maize flour, the greatest NOFB (22.66 g) were seen. Compared to other treatments, this was the highest. However, the control had the fewest NOFB (16.33 g), which was much fewer than any of the other treatments. The wheat straw + sorghum flour at 5 % showed the longest stipe (19.00 cm). However, compared to all other treatments, the smallest SL (14.00 cm) was found in wheat straw + pearl millet flour at 5 %. Wheat straw plus pearl millet flour at 5 % had the largest PW (10.66 cm), whereas wheat straw plus maize flour at 10 % had the smallest PW (7.83 cm), which was much less than all other treatments.

At 5 %, maize flour produced the highest yield (890.00 g/kg of dry substrates with 89 % BE). However, the control showed the lowest yield (622 g/kg of dry substrates with 62.20 % BE), which was less than all other treatments. Pearl millet flour at 5 % had the largest average weight of fruiting bodies (43.55 g). which, in comparison to the other treatments, was much higher. However, with 10 % rice flour, the lowest Avg WOFB (36.47 g) was recorded, which is much less than all other treatments (Table 2).

These results are consistent with the previous findings (13). The optimal nutrients and doses for commercial milky white mushroom growing were investigated. The yield and yield-contributing characteristics of *C. indica* were evaluated using supplements of rice bran, maize powder and 68 wheat bran at various doses (10, 20, 30, 40 and 50 %). Primordia were shown to begin between 13.5 and 19.3 days. According to the findings, the 30 % maize powder supplement was successful in generating fruiting bodies. With 30 % maize powder, the pileus and stalk diameters reached their maximum. The maximum biological and economic yield, together with the highest BE, was obtained by supplementing with 30 % maize powder. Research investigated how

Table 1. Effect of different cereal flours on sporophore yield of *Calocybe indica* (CI-22-09)

Treatment	DFSR	DFFH	DFCP	NOFB	Yield (kg/kg)	SL (cm)	PW (cm)	Avg WOFB (g)	BE (%)
WS + RF at 5 %	20.00	25.00	80.33	20.66	843.33	17.33	10.16	40.81	84.33
WS + RF at 10 %	18.00	31.00	83.33	19.66	823.33	16.00	8.00	41.87	82.33
WS + MF at 5 %	17.33	37.33	85.33	22.00	971.66	16.00	7.33	44.16	97.16
WS + MF at 10 %	17.66	37.66	81.33	22.66	970.00	14.33	8.00	42.80	97.00
WS + SF at 5 %	20.33	27.00	84.00	18.66	760.00	17.00	10.00	40.72	76.00
WS + SF at 10 %	22.16	29.66	84.33	18.00	773.33	14.00	9.00	42.96	77.33
WS + PMF at 5 %	22.00	23.16	82.16	18.00	766.66	13.00	11.66	42.59	76.66
WS + PMF at 10 %	21.33	25.00	83.00	16.66	740.00	16.00	8.00	44.41	74.00
WS (control)	18.00	23.00	82.00	15.33	630.00	14.00	7.00	41.09	63.00
CD at 5 %	1.59	1.91	2.61	1.63	35.40	1.70	1.68	-	-
SE (m)	0.54	0.65	0.89	0.56	12.10	0.58	0.57	-	-

WS: Wheat straw; RF: Rice flour; MF: Maize flour; SF: Sorghum flour; PMF: Pearl millet flour; DFSR: Days for spawn run; DFFH: Days for first harvesting; DFCP: Days for cropping period; NOFB: Number of fruiting bodies; SL: Stipe length; PW: Pileus width; Avg WOFB: Average weight of fruiting body; BE: Biological efficiency; CD: Critical difference; SE: Standard error.

Table 2. Effect of different cereal flour on sporophore yield of *Calocybe indica* (CI-22-10)

Treatment	DFSR	DFFH	DFCP	NOFB	Yield (kg/kg)	SL (cm)	PW (cm)	Avg WOFB (g)	BE (%)
WS + RF at 5 %	19.66	28.33	79.83	21.66	793.33	18.33	9.16	36.62	84.33
WS + RF at 10 %	18.33	30.50	81.33	19.83	723.33	17.3	9.16	36.47	82.33
WS + MF at 5 %	16.83	36.33	82.00	22.66	871.66	18.00	8.33	38.46	97.16
WS + MF at 10 %	17.33	38.66	84.00	21.66	890.00	16.33	7.83	41.08	97.00
WS + SF at 5 %	19.33	28.33	85.16	19.66	750.00	19.00	9.00	38.14	76.00
WS + SF at 10 %	21.00	27.66	84.66	18.33	783.33	17.83	8.66	42.73	77.33
WS + PMF at 5 %	20.16	26.16	83.33	17.83	776.66	14.00	10.66	43.55	76.66
WS + PMF at 10 %	22.33	28.66	82.33	17.66	750.00	15.00	9.00	42.46	74.00
WS (control)	17.00	26.83	85.00	16.33	622.00	15.33	8.33	38.08	63.00
CD at 5 %	1.50	2.64	3.36	1.61	64.33	1.34	0.71	-	-
SE (m)	0.51	0.90	1.15	0.55	21.99	0.46	0.24	-	-

WS: Wheat straw; RF: Rice flour; MF: Maize flour; SF: Sorghum flour; PMF: Pearl millet flour; DFSR: Days for spawn run; DFFH: Days for first harvesting; DFCP: Days for cropping period; NOFB: Number of fruiting bodies; SL: Stipe length; PW: Pileus width; Avg WOFB: Average weight of fruiting body; BE: Biological efficiency; CD: Critical difference; SE: Standard error

two strains (CI-16-02 and CI-16-03) of *C. indica* were produced in response to various grain additions, including sorghum flour, rice flour and pearl millet flour at 1 and 2 % (15). They found that when rice flour at 2 % was added, strains CI-16-02 and CI-16-03 took the fewest days (16.50 and 17.50 days respectively) to spawn and that their yields (640 g/kg dry substrate and 650 g/kg dry substrate respectively) were the highest of all treatments followed by 2 % sorghum flour (620 and 625 g/kg dry substrate).

Effect of different straws and their combination on sporophore yield

The purpose of the experiment was to determine how various substrates and their combinations affected the milky mushroom sporophore yield. Under the CI-22-09 strain in control, the spawn run minimum of 14.00 days was noted. However, the rice straw + wheat straw (4:1) treatment had the longest spawn run (22.16 days), longer than any other treatment. The shortest first harvesting duration (29.00 days) was for wheat straw. However, rice straw + wheat straw (4:1) had the highest number of days for initial harvesting (39.00 days), which was statistically higher than all other treatments. The rice straw + wheat straw (3:1) combination had the longest cropping duration (86.00 days). Although rice straw + wheat straw (1:1) had the shortest cropping period (82.00 days), it was still far shorter than any other treatment. The most fruiting bodies (18.00) were formed by the rice straw and wheat straw (1:4) combination. In comparison to the other treatments, rice straw had the fewest fruiting bodies (12.00) (Fig. 1). The combination of rice straw and wheat straw (1:2) produced the longest stipe (18.00 cm). However, rice straw + wheat straw (3:1) had the smallest SL (14.66 cm), which was less than all other treatments. Rice straw + wheat straw (1:4) had the largest PW (9.33 cm), whereas rice straw + wheat straw (3:1) had

the smallest (6.00 cm), which was much smaller than all other treatments. The rice straw + wheat straw (1:4) combination produced the highest yield (740.00 g/kg of dry substrates with

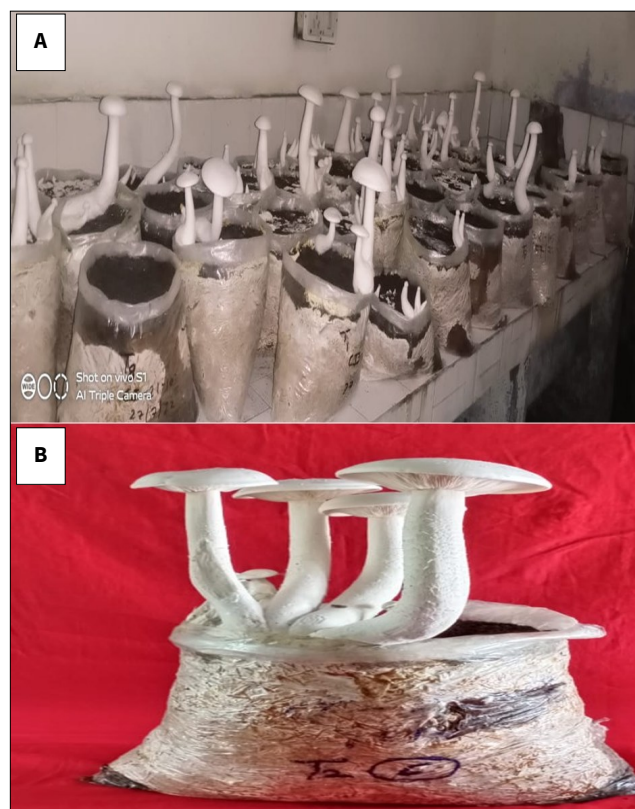


Fig. 1. A: Effect of different substrate combinations on the sporophore yield of milky mushroom; B: Effect of different cereal flour on the sporophore yield of milky mushroom.

Table 3. Effect of different straws and their combination on sporophore yield of *Calocybe indica* (CI-22-09)

Treatment	DFSR	DFFH	DFCP	NOFB	Yield (g/Kg)	SL (cm)	PW (cm)	Avg WOFB (g)	BE (%)
RS + WS (1:1)	17.00	36.00	82.00	16.00	640.00	16.00	7.33	40.00	64.00
RS + WS (1:2)	15.00	30.00	83.33	16.00	650.00	18.00	8.33	40.62	65.00
RS + WS (1:3)	16.00	33.00	82.00	17.00	720.00	17.33	8.66	42.35	72.00
RS + WS (1:4)	17.33	35.00	84.33	18.00	740.00	16.66	9.33	41.11	74.00
RS + WS (2:1)	18.00	37.00	82.33	14.00	560.00	15.33	6.00	40.00	56.00
RS + WS (3:1)	19.83	38.00	86.00	13.00	550.00	14.66	9.00	42.30	55.00
RS + WS (4:1)	22.16	39.00	83.00	14.00	510.00	16.00	6.33	36.42	51.00
Rice straw	21.00	37.00	82.33	12.00	500.00	16.33	7.33	41.66	50.00
Wheat straw	14.00	29.00	84.66	16.00	620.00	17.00	8.00	38.75	62.00
CD at 5 %	1.51	2.16	2.63	1.43	39.57	1.90	1.96	-	-
SE (m)	0.52	0.74	0.90	0.49	13.53	0.65	0.67	-	-

RS: Rice straw; WS: Wheat straw; DFSR: Days for spawn run; DFFH: Days for first harvesting; DFCP: Days for cropping period; NOFB: Number of fruiting bodies; PW: Pileus width; Avg WOFB: Average weight of fruiting body; BE: Biological efficiency; CD: Critical difference; SE: Standard error

74.00 % BE). However, rice straw showed the lowest yield (500.00 g/kg of dry substrates with 50.00 % BE), which is statistically less than all other treatments. Rice straw + wheat straw (1:3) had the highest Avg WOFB (42.35 g), whereas rice straw + wheat straw (4:1) had the lowest Avg WOFB (36.42 g), which was less than all other treatments (Table 3).

While in strain CI-22-10 the spawn run minimum of 14.00 days was noted. However, rice had the longest spawn run (21.00 days), longer than any other treatment. The minimal number of days required for the initial harvest (30.00 days) was preserved in wheat straw. However, the rice straw + wheat straw (4:1) combination showed the highest number of DFFH (38.00 days), which was statistically greater than all other treatments. The combination of rice straw and wheat straw (3:1) resulted in the longest cropping time (85.33 days). However, rice straw + wheat straw (1:4) had the shortest cropping period (81.00 days), which was significantly less than all other treatments. The most fruiting bodies (18.33) were formed by the rice straw and wheat straw (1:4) combination. In comparison to the other treatments, rice straw had the fewest fruiting bodies (13.00). The rice straw + wheat straw (1:3) and (1:4) combinations showed the longest SL (17.66 cm). However, rice straw + wheat straw (3:1) had the smallest SL (15.66 cm), which was less than all other treatments. Rice straw + wheat straw (1:4) and (1:3) had the largest PW (8.33 cm), while rice straw + wheat straw (2:1) had the smallest PW (7.00 cm), which was much smaller than all other treatments. The rice straw + wheat straw (1:4) produced the highest yield (730.00 g/kg of dry substrates with 73.00 % BE), while the control rice straw produced the lowest yield (520.66 g/kg of dry

substrates with 52.66 % BE), which was statistically less than that of any other treatment.

The Avg WOFB was lower than all other treatments, with rice straw + wheat straw (1:3) having the highest average weight (43.49 g) and the lowest average weight (38.22 g) (Table 4). Research has noticed a similar finding from their observation (16). Research discovered that the lowest BE was in paddy straw (36.27 %), while the highest was in wheat straw + sugarcane bagasse (61.13 %) and wheat straw (45.00 %). To determine the ideal substrate for *C. indica* cultivation, research indicates the five distinct substrates, i.e. wheat straw, paddy straw, sugarcane bagasse and paddy straw alone and in combination (17). The largest yield (1052.50 g), the greatest number of fruiting bodies (40.75), the earliest spawn run (21.50 days) and the earliest first harvest (33.25 days) were all produced by the wheat straw substrate. The yield from the combination of wheat straw and paddy straw came in second (932.50 g).

Conclusion

Based on the results of the experiment, the efficiency of milky mushroom (*Calocybe indica*) production is significantly impacted by various lignocellulosic substrates and grain additions. Maize flour at 5 % concentration produced the best yield (971.16 g/kg of dry substrate) with a BE of 97.10 % among the tested grain additions. It was closely followed by maize flour at 10 % concentration (970.00 g/kg, 97.00 % efficiency). Rice straw + wheat straw (1:4) showed the highest sporophore output (740.00 g/kg with 74.00 % BE) among substrate combinations,

Table 4. Effect of different straws and their combination on sporophore yield of *Calocybe indica* (CI-22-10)

Treatment	DFSR	DFFH	DFCP	NOFB	Yield (kg/kg)	SL (cm)	PW (cm)	Avg WOFB (g)	BE (%)
RS + WS (1:1)	17.00	33.33	83.00	17.33	643.33	16	7.66	38.22	64.00
RS + WS (1:2)	14.00	31.66	81.33	17.00	660.00	17.33	7.33	38.82	65.00
RS + WS (1:3)	16.33	32.66	83.33	16.33	710.33	17.66	8.33	43.49	72.00
RS + WS (1:4)	16.66	34.00	81.00	18.33	730.66	17.66	8.33	40.24	74.00
RS + WS (2:1)	17.66	36.33	82.00	14.66	570.00	16.33	7.00	38.43	56.00
RS + WS (3:1)	17.33	37.33	85.33	14.33	564.00	15.66	8.00	39.35	55.00
RS + WS (4:1)	20.66	38.00	83.66	14.00	530.66	15.33	7.33	38.33	51.00
Rice straw	21.00	36.00	82.00	13.00	520.66	16.66	8.33	40.05	50.00
Wheat straw	14.00	30.00	83.66	15.33	633.00	17.33	8.33	41.29	62.00
CD at 5 %	1.48	2.24	2.03	1.32	64.56	1.37	1.40	-	-
SE (m)	0.51	0.77	0.70	0.45	22.07	0.47	0.48	-	-

RS: Rice straw; WS: Wheat straw; DFSR: Days for spawn run; DFFH: Days for first harvesting; DFCP: Days for cropping period; NOFB: Number of fruiting bodies; PW: Pileus width; Avg WOFB: Average weight of fruiting body; BE: Biological efficiency; CD: Critical difference; SE: Standard error.

suggesting that it is a suitable substrate blend. The lowest output, however, was 500.00 g/kg of rice straw alone (50 % efficiency), followed by rice straw + wheat straw (4:1) (510.00 g/kg, 51 % efficiency). All things considered, the results indicate that adding maize flour to cereals greatly increases sporophore yield and that using a substrate mixture of rice straw and wheat straw (1:4) works better than using only rice straw. These findings may help maximise the production of milky mushrooms in tropical areas.

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Authors' contributions

AY conducted the trial. GS designed all the technical programs and also provided the necessary equipment and chemicals. VS helped in manuscript preparation and financial support. SS, HY, UR, JK and SK provided data analysis and interpretation of the results. All authors read and approved the final manuscript.

Compliance with ethical standards

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