

Effects of Grains Spawn and Substrates on Proximate Composition of Oyster Mushroom Under Different Cropping Shelters

Modeste Hakizimana^{1*}✉, Robert Morwani Gesimba²✉, Japhet Mburugu Muthamia³✉

¹Faculty of Agriculture, Department of Crops Horticulture and Soils, Egerton University, Kenya.

²Lecturer, Egerton University, Department of Crops Horticulture and Soils, P.O. Box 536-20115, Nakuru, Kenya.

³Lecturer, Egerton University, Department of Biological Sciences, P.O. Box 536-20115, Nakuru, Kenya.

*Corresponding author: Modeste Hakizimana, Faculty of Agriculture, Department of Crops, Horticulture, and Soils, Egerton University, Kenya. Email: modestehakim@gmail.com

Citation: Hakizimana M, Gesimba RM, Muthamia JM (2020) Effects of Grains Spawn and Substrates on Proximate Composition of Oyster Mushroom Under Different Cropping Shelters. GJ J Foo Sci Nutri: GJFSN:125.

Received Date: May 05th, 2022; **Accepted Date:** May 09th, 2022; **Published Date:** May 16th, 2022

Abstract

Oyster mushroom (*Pleurotus ostreatus*) can be considered as functional foods that can provide health benefits beyond the conventional supplements they contain. This study was conducted to evaluate the effects of different levels of cereal grain spawn and local substrates on the proximate composition of oyster mushrooms cultivated under different cropping shelters. The study was carried out as factorial experiment in a completely randomized design (CRD). Five proximate composition aspects i.e crude protein, fat, fibres, moisture content and ash analysis of oyster mushrooms cultivated in 16 different substrates and grown under 3 different cropping shelters (currently used dark-house shelter and shelter made in mikeka, 60% shade net shelter) were evaluated. Data was collected on crude protein, fat, fibres, moisture content and ash analysis. The data was analysed in SAS using PROC GLM in ANOVA and means separated using Tukeys HSD test ($P \leq 0.005$). Significant differences in proximate composition were observed among the samples taken from 16 different substrates under 3 different cropping shelters ($P \leq 0.005$). The highest and lowest mean value of moisture content was observed in sawdust substrate (90.0 %) cultivated under shade net cropping shelter and sawdust substrates (80.7 %) cultivated under dark house shelter, respectively. while the maximum and minimum mean value of ash content was obtained on a combination of wheat straws+ sawdust+ corn cobs substrate (13.4%) grown under shade net shelter and sawdust substrate (9.6%) cultivated under dark house shelter, respectively. The maximum crude fibre was found on combination sawdust+ corn cobs substrate (18.7.0%) under mikeka shelter, while the minimum was found in sawdust (14.7%) grown under dark house shelter. The highest crude fat was obtained in the combination of sawdust+ popcorn cobs + kikuyu grass (2.1%) cultivated under mikeka shelter and the lowest was found in kikuyu grass (0.85%) grown under shade net shelter. Crude protein found in wheat straws (39.1%) under dark house shelter was the best and crude protein observed in corn cobs (24.1%) under shade net was the least. The substrates and cropping shelters significantly affected the ash, crude fat, crude protein, crude fibre and moisture content of oyster mushrooms. The study revealed that the corn cob substrates cultivated under locally mikeka shelter highly influence growth and yield of oyster mushrooms.

Introduction

The nutritional deficiency levels remain high among a significant segment of the Kenyan population. Mushroom farming has so far been recognized as an important opportunity to enhance household food security in developing countries. The existing works of literature have been showing the evidence of the contribution of mushrooms to nutritional security through direct consumption and income stability among vulnerable groups through involvement in mushroom farming and value chain linkages [1, 2]. The problem of protein shortage in developing countries including Kenya is an existing reality and will continue for the foreseeable future. Protein

malnutrition will become even more acute since the supply of protein for the diet has not kept pace with population growth [3]. To meet the deficit, most developing countries tend to import essential protein sources of food from abroad, spending large sums of their meagre foreign exchange earnings. Such a situation has forced planners and nutritionists to think about unconventional alternative sources of protein such as mushrooms [4, 6].

Edible mushrooms are highly nutritious and are considered as functional foods because they are low in all of those substances that the body does not need much of fat, cholesterol, calories, and sodium [7]. Oyster mushroom (*Pleurotus ostreatus*) has high protein content as compared

to eggs, milk and meat [5, 8]. They are considered to be one of the most Efficiency producers of food protein, producing 30% of its dry weight [10] and it contains an adequate amount of phosphorous, iron, protein, lipid, riboflavin, and thiamine. The protein content of mushrooms varies from 4-44% according to the species [5]; whereas, other foods like beef and soybean contain protein of about 26 and 13%, respectively. Oyster mushroom fresh fruiting bodies indicates a high quantity of moisture (90.8%), whereas dry as well as fresh oyster mushrooms are rich in carbohydrate (57.6%), protein (30.4%), fibre (8.7%), fat (2.2%) and ash (9.8%) with 345 kilocalories energy value on 100 g dry weight [9]. Oyster Mushrooms are considered as substitute for meat in terms of nutritional value and are comparable to several vegetables [11].

Benjamin reported that mushrooms can improve metabolism (mushrooms are high in B vitamins like thiamine, riboflavin, niacin, pantothenic acid and folate and they also contain folic acid, which is blood building vitamin and counteracts anaemia [7]. Mushroom is the only food with vitamin D; boosts the immune system; Serves as antioxidants; contains Selenium for bladder heart; lowers the risk of cancer; anemia; diabetes; and certain heart diseases. The proteins of mushrooms are considered to be intermediate between that of vegetables and animals and the essential amino acids of the human body are found in the oyster mushroom [12]. The objective of this research is to evaluate different substrates and cereal grains spawn for the proximate composition of oyster mushrooms grown under different cropping shelters.

Materials and Methods

2.1. Experimental sites

The kikuyu grass, corn cobs, dry wheat straw and sawdust were air-dried and cleaned. The residuals were cut into small pieces and soaked in water for 3 days according to the suggestions made by [13]. Then were dried to reduce the moisture to 75%. All substrates were added to 5% by weight dry wheat bran to maximise the amount of nitrogen and some minerals and 1.5% by weight dry calcium carbonate to reduce the pH of organic residuals [14, 15]. The dried mixture of organic residuals was packed in polypropylene bags of (12×22) cm, then they were tied with a rubber band, and each bag was containing 1200g of a cultivation substrate. Pasteurization of substrates was performed using hot steam at 85°C for 5 hours by a metal barrel and firewood.

2.2. Spawning

Spawning was done under controlled conditions. The spawn (rice and popcorn) obtained from tissue culture were mixed in all substrates using 45g of the total weight of the packet. After spawning substrates were kept under dark environmental conditions and 8 holes were made on bags for aeration.

2.2. Spawn running

Humidity was maintained by irrigation two to three times a day; Room temperature and relative humidity were maintained during the spawn run. After full colonisation of the substrates, some of the bags were moved to mikeka shelter and shade net shelter for fruiting bodies production.

3. Data collection

In proximate composition determination, one sample from each treatment in all three cropping shelters was selected randomly to analyse for chemical compositions.

3.1. Determination of moisture content

The moisture content was calculated as per the reference of [16]. Three clean individually marked dishes were placed in an oven settled at 105°C for one hour after were cooled in a desiccator then were weighed accurately and recorded weight. Immediately about 2.0g were added of the samples with the dish and transferred to the dishes with the sample in the oven set at 105°C and leave them for 8 hours. After 8 hours the dishes were transferred to a desiccator and left to cool. They were removed from the desiccator one at the time and weigh again accurately.

The loss in weight is the moisture content of the sample and what is left is referred to as the dry matter (DM). The moisture is usually expressed as a percentage or g/kg.

3.2. Crude protein

The crude protein content of the oyster mushroom was determined using the Kjeldahl method as described by AOAC [18]. Three grams of each dry sample were weighed and put into a Kjeldahl digestion flask. Three grams of Selenium catalyst were added to each of the flasks moistened with distilled water and mixed with 10 ml of concentrated H₂SO₄. The mixture was heated with red hot temperature (440°C) under a fume cupboard for 2 hours to obtain a clear solution. The digest was transferred quantitatively to a 100 ml volume flask and diluted with 100 ml distilled water. 10 ml, of the digest, were mixed with 100 ml of 40% NaOH solution in a semi-micro Kjeldahl distillation apparatus. The mixture was distilled and the distillate was collected into 25 ml of 4% boric solution containing 3 drops of mixed indicator (methyl red and bromocresol green) and the solution were turned green. A total of 100 ml distillate were collected and titrated against 0.01N HCl solution until the solution turned pink in colour. A blank experiment was also set involving the digestion of all the materials except the sample. The distillation was also carried out on the blank. The titre value of the blank was subtracted from that of the sample and the difference obtained were used to calculate the crude protein. The percent nitrogen content was calculated using the following formula:

$$\text{Nitrogen content (\%)} = \frac{(\text{ml acid for sample} - \text{ml acid for blank}) \times \text{molarity} \times 14 \times 100}{\text{mg of sample}} =$$

- Crude protein (%) = % N x 6.25

3.3. Crude fibre determination

Crude fibre was determined by using the method described by Udo and [19]. One gram of each sample was digested with 100 ml of 1.25% H₂SO₄ solution under reflux for 30-minute boiling. The digest was allowed to cool and then filtered with a muslin cloth. The residue was washed thrice with hot water, took into a conical flask, and digested with 100 ml of 1.25% NaOH solution under reflux for 30 minute boiling. The digest was cooled, filtered, and washed thrice with distilled water. The residue was drained and took into a previously dried and weighed crucible and then put into the oven to dry at 105°C to a constant mass. The dish with its content were reweighed after drying and then placed in the

muffle furnace to ash at a temperature of 550°C for 3 hours. The ash was withdrawn at the end and put in a crucible and reweighed. The difference in mass of the sample were calculated as crude fibre and expressed as a percent of the initial mass.

3.4. Crude fat determination

For the determination of fat Soxhlet apparatus was used. The dried mushroom samples of weight 3g were taken in a thimble drops fell on the sample in the extraction tube petroleum ether (B.P. 40-60 °C) for was used for extraction hours. The solvent was evaporated under the fume hood and the samples were removed and dried in an oven at 105 °C for 30 minutes. After cooling in desiccators, the weight of the extract was recorded.

$Crude\ fat\ (\%) = (W2 - W1) \times 100 \text{ weight of sample}$

Where:

W1=weight of beaker

W2= weight of beaker+ oil

3.5. Determination of Total Ash

One gram of the sample was weighed accurately into a crucible. The crucible was placed on a clay pipe triangle and heated first over a low flame till all the material was completely charred, followed by heating in a muffle furnace for about 5-6 hours at 6000 C. It was then cooled in a dessicator and weighed. To ensure completion of ashing, the crucible was then heated in the muffle furnace for 1h, cooled and weighed. This was repeated till two consecutive weights were the same and the ash was almost white or grayish white in colour. Then total ash was calculated as following equation: Ash content (g/100g sample) = Wt of ash × 100 / Wt of sample taken [20].

4. Data analysis

The data were analysed using SAS statistical package 9.4 M6 [21]. Analysis of variance (ANOVA) was used to determine the significant differences in nutritional composition among

the samples that received the various treatments and conducted using proc glm. Tukey test was used to compare the means' significant differences.

Results

5.1. Effect of different substrates on moisture, ash content, crude fibre, fat and crude protein of oyster mushroom

The effects of different substrates on proximate composition are presented in Table 1 and Figure 1. The findings for analysis of variance showed that substrates did affect significantly (P≤0.005) the moisture, ash content, crude fibre, fat and crude protein of oyster mushrooms. The distributions of moisture content showed that the substrate levels at the combination of corn cobs+ sawdust+ wheat straw, sawdust, a combination of corn cobs+ sawdust+ kikuyu grass, a combination of sawdust+ kikuyu grass+ corn cobs and wheat straw highly influenced the moisture, ash content, crude fibre, fat and crude protein of oyster mushroom with 87.9, 12.9, 18.20, 1.97, 34.9% , respectively, while substrates levels at sawdust (82.1%), sawdust (16.5%), sawdust (10.3%), kikuyu grass (1.1%) and the combination of (kikuyu grass+ sawdust) with 27.4% were the least to influence the moisture, ash content, crude fibre, fat and crude protein of oyster mushroom, respectively. Tukey grouping for means showed that S7, S8, S5; S1, S2, S6; S4 and S2 were not significant different whereas S7, S6 and S3 were significantly different for ash content while Sawdust substrate was significantly different from all other substrates for crude fibre; The substrates S7, S6 and S3; S8, S6, S3 were significant while S8, S6,S2 and S4; S2,S4, S5 and S1 were not significant for crude fat and protein Tukey grouping for means of substrates (P≤0.005) showed that S1, S2 and S4; S2, S3 and S7 were significant while S1and S2; S6, S3 and S8; S4, S7 and S5 were not significant from each other.

Table 1: Effect of different substrates on moisture, ash content, crude fibre, fat and crude protein of oyster mushroom.

Substrates	Means %				
	Moisture	Ash	Fibre	Fat	Protein
S1	85.72	11.15	17.30	1.46	34.96
S2	82.18	10.35	16.58	1.67	34.18
S3	86.73	10.58	17.27	1.10	31.59
S4	85.10	10.37	17.98	1.67	28.69
S5	86.79	12.72	18.21	1.65	27.46
S6	85.91	11.68	17.93	1.70	32.53
S7	87.00	12.90	18.07	1.98	27.74
S8	87.82	12.88	17.29	1.89	31.11
St. Error	0.21	0.16	0.30	0.05	0.45

Whereas Sub: substrate, S1: wheat straw, S2: sawdust, S3: kikuyu grass, S4: popcorn cobs, S5: kikuyu grass+ sawdust, S6: sawdust+ popcorn cobs, S7: sawdust+ popcorn cobs+ kikuyu grass, S8: sawdust+ popcorn cobs+ wheat straw.

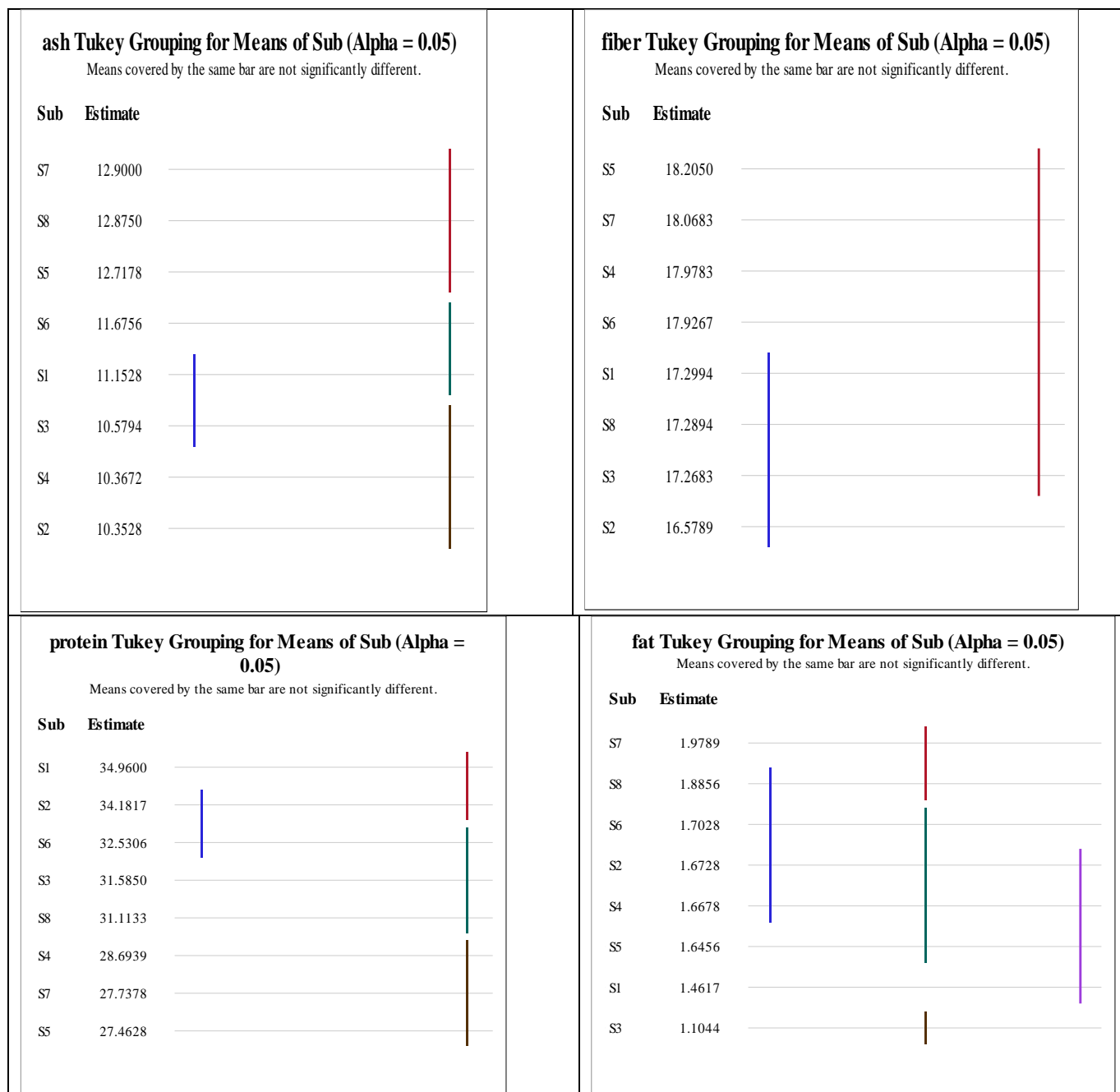


Figure 1: Moisture, ash, crude fibre, fat and protein Tukey grouping mean separation of substrates ($P \leq 0.005$). whereas Sub: substrate, S1: wheat straw, S2: sawdust, S3: kikuyu grass, S4: corn cobs, S5: kikuyu grass+ sawdust, S6: sawdust+ popcorn cobs, S7: sawdust+ popcorn cobs+ kikuyu grass, S8: sawdust+ popcorn cobs+ wheat straw.

5.2. Effect of different cropping shelters on proximate composition

The effects of different substrates on proximate composition are presented in Table 2, Figure 2 and Figure 3. The results indicated that the cropping shelters significantly influenced ($P \leq 0.005$) the moisture, ash content, fat and crude protein of oyster mushroom, though did not show any significant on crude fibre appendix 3. The maximum and minimum of moisture and ash content of 87.6 and 84.8 %; 11.8 and 11.3 % were observed in shade net and dark shelved house, respectively and ash mean separation revealed that shade net and mikeka shelter; dark house and mikeka shelter were not significant while shade net and mikeka cropping shelter

were significantly different from each other. None of the cropping shelters on crude fibre was statistically significant different from each other. Based on crude fat, dark shelved house was the best in influencing the crude fat followed mikeka cropping shelters and shade net shelter with 1.7, 1.6 and 1.5% of crude fat, respectively, and dark shelter and mikeka shelter; mikeka and shade net shelter were not significant while dark shelter and shade net were significantly different from each other. Dark house shelter was the best in influencing the crude proteins with 34.04% of crude proteins while the least was shade net with 27.7% Of crude protein and all the cropping shelters were significantly different from each other.

Table 2: Effect of different cropping shelters on moisture, ash content, crude fibre, fat and crude protein of oyster mushroom.

Shelters	Moisture	Means (%)			
		Ash	Fibre	Fat	Protein
Mikeka	85.42	11.54	17.54	1.64	31.33
Shade net	87.39	11.86	17.56	1.58	27.73
Dark house	84.90	11.33	17.63	1.71	34.05
St. Error	0.13	0.10	0.19	0.03	0.27

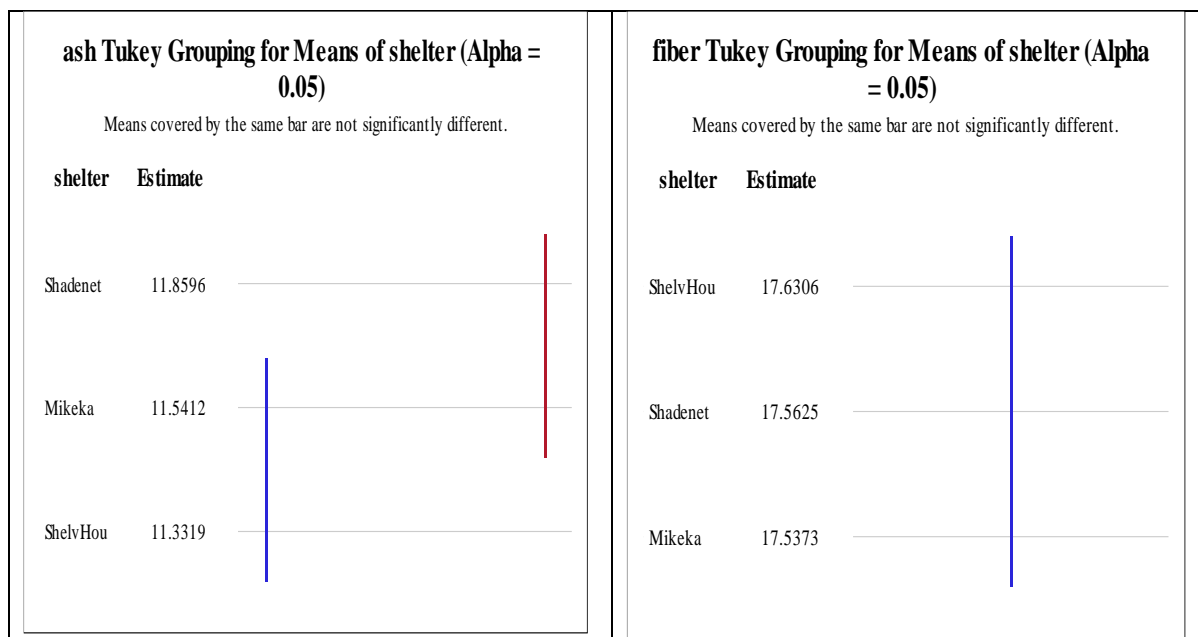


Figure 2: Moisture and crude fibre, fat and protein Tukey grouping mean separation of cropping shelters ($P \leq 0.005$). Means covered by the same bar are not significantly different.

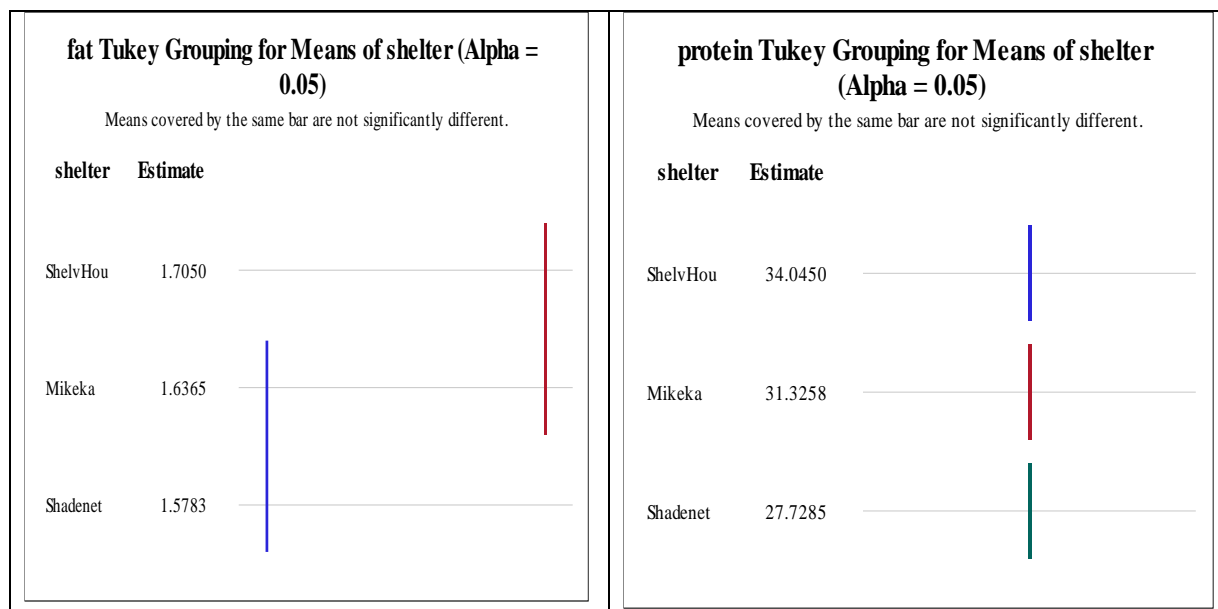


Figure 3: Crude fat and crude protein Tukey grouping mean separation of cropping shelters ($P \leq 0.005$). Means covered by the same bar are not significantly different.

5.3. Effect of different cereal grain spawn on moisture, ash content, crude fibre, fat and crude protein of oyster mushroom

The effects of different substrates on proximate composition are presented in Table 5.3, Figure 4 and figure 5. Results

showed a significant difference in ash content crude fibre and crude protein from cereal grain spawn, though there were not significantly different ($P \leq 0.005$) in moisture content and crude fat. Both popcorn and rice spawn were not significant from each other on moisture content and

crude fat whereas rice grain spawn showed high ash content (11.7%) than popcorn spawn (11.4%) and both grain spawn was significantly different from each other on ash content and showed high crude fibre content of 17.8% while rice

spawn was 17.2% and were significantly different from each other. Rice grain spawn showed high crude protein with 31.5% which was significantly different from popcorn spawn with 30.5% of proteins.

Table 3: Effect of different cereal grain spawn on moisture, ash content, crude fibre, fat and crude protein of oyster mushroom.

Spawn	Means (%)				
	Moisture content	Ash content	Crude Fibre	Crude Fat	Crude Protein
Rice	86.00	11.70	17.26	1.63	31.51
Popcorn	85.81	11.45	17.90	1.65	30.56
St.errors	0.08	0.08	0.08	0.08	0.08

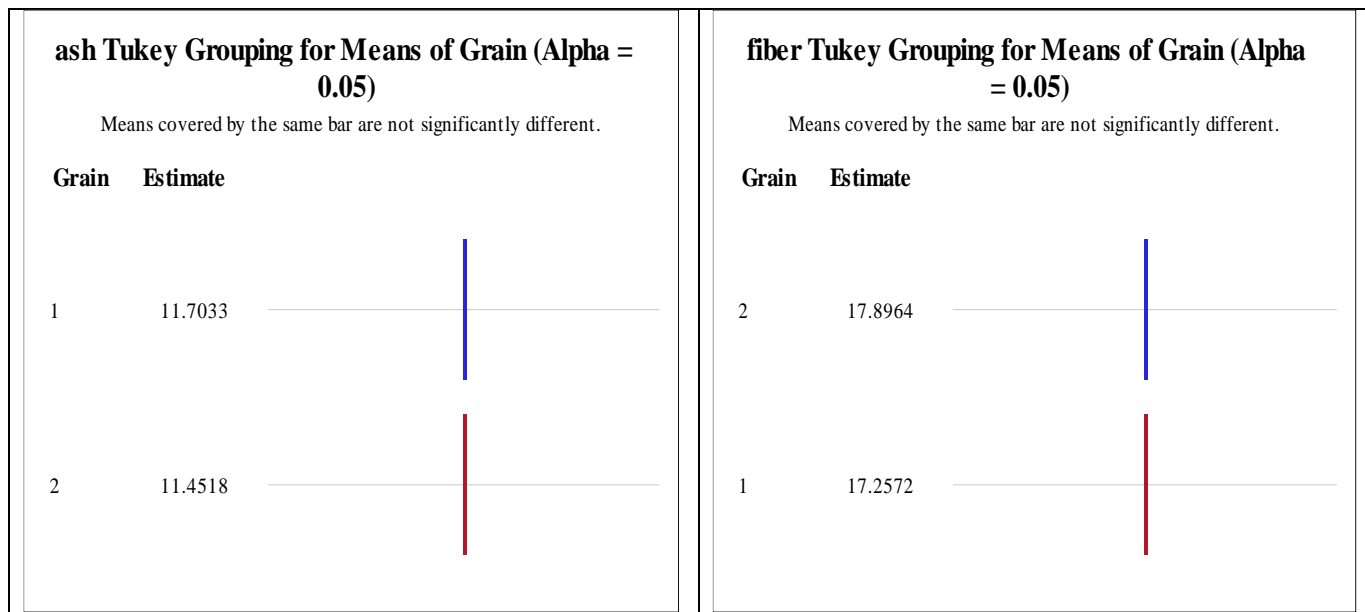


Figure 5.4: Ash content and Crude fibre Tukey grouping mean separation of grain spawn ($P \leq 0.005$). Whereas Grain 1: Rice spawn, Grain 2: Popcorn spawn.

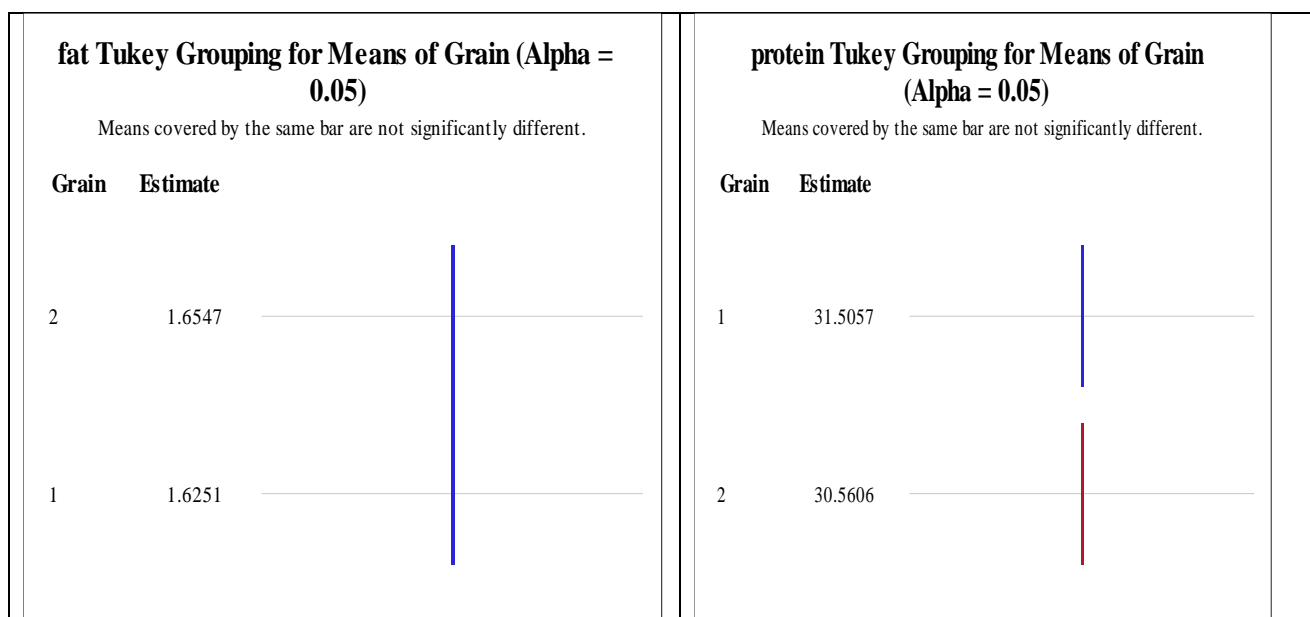


Figure 5: Crude fat and Crude protein Tukey grouping mean separation of grain spawn ($P \leq 0.005$). Whereas Grain 1: Rice spawn, Grain 2: Popcorn spawn.

5.4. Effect of interaction between substrates and shelters on moisture, ash content, crude fibre, fat and crude protein of oyster mushroom

The effects of different substrates on proximate composition are presented in table 4. The findings for analysis of variance showed that the interaction between substrate and cropping shelters did affect significantly ($P \leq 0.005$) the moisture content and crude protein whereas did not affect ash content, crude fibre and crude fat of oyster mushroom. The highest and lowest moisture content were observed on the interaction between shade net and combination of (wheat straw+ sawdust+ popcorn cobs) substrate and interaction between dark house shelter and popcorn cobs with 89.4 and 80.8 %, respectively, The maximum and minimum ash content were found on the interaction between shade net

and combination of (kikuyu grass+ sawdust+ popcorn cobs) substrate and On the interaction between dark house shelter and popcorn cobs substrate with 13.11 and 9.76%; The interaction between dark shelter and combination (popcorn cobs+ sawdust+ kikuyu grass) had the highest crude fibres of 18.4% while the least was on the interaction between mikeka and sawdust substrates with 15.7% while the interaction between mikeka shelter and (corn cobs+ sawdust+ kikuyu grass) were the highest in influencing crude fat with 2.02% and the interaction between shade net and kikuyu grass was the lowest with 0.89 % of crude fat. The maximum and minimum crude protein were observed on the interaction between dark shelter and wheat straw with 39.1% and interaction of shade net and corn cobs with 25.4% crude protein.

Table 4: Effect of interaction between substrates and shelters on moisture, ash content, crude fibre, fat and crude protein of oyster mushroom.

Shelters	Substrates	Moisture	Ash	Fibre	Fat	Protein
Mikeka	S1	85.18	11.08	17.23	1.46	34.87
Mikeka	S2	81.52	10.25	16.62	1.69	35.04
Mikeka	S3	86.93	10.47	17.31	1.15	32.65
Mikeka	S4	84.35	10.32	18.04	1.62	28.49
Mikeka	S5	86.52	12.74	18.07	1.64	27.65
Mikeka	S6	85.48	11.71	17.88	1.71	32.08
Mikeka	S7	86.17	12.88	17.83	2.02	27.44
Mikeka	S8	87.23	12.89	17.33	1.80	32.38
Shade net	S1	87.42	11.31	17.23	1.38	30.91
Shade net	S2	84.15	10.66	16.62	1.60	29.53
Shade net	S3	85.88	10.75	17.31	0.89	26.57
Shade net	S4	87.50	11.02	18.04	1.67	25.43
Shade net	S5	88.32	12.91	18.01	1.57	25.76
Shade net	S6	87.47	12.09	17.99	1.63	30.28
Shade net	S7	89.00	13.11	17.96	1.98	25.61
Shade net	S8	89.40	13.03	17.34	1.91	27.75
Dark house	S1	84.57	11.08	17.44	1.55	39.10
Dark house	S2	80.87	10.15	16.50	1.73	37.98
Dark house	S3	87.36	10.52	17.20	1.28	35.53
Dark house	S4	83.45	9.76	17.86	1.71	32.17
Dark house	S5	85.55	12.51	18.54	1.72	28.98
Dark house	S6	84.77	11.23	17.91	1.78	35.24
Dark house	S7	85.83	12.71	18.41	1.93	30.16
Dark house	S8	86.82	12.71	17.20	1.94	33.21
St. Error		0.37	0.28	0.52	0.09	0.77

Whereas substrate S1: wheat straw, S2: sawdust, S3: kikuyu grass, S4: popcorn cobs, S5: kikuyu grass+ sawdust, S6: sawdust+ popcorn cobs, S7: sawdust+ popcorn cobs+ kikuyu grass, S8: sawdust+ popcorn cobs+ wheat straw.

5.5. Effect of interaction between substrates and grain spawn on moisture, ash content, crude fibre, fat and crude protein of oyster mushroom

The effects of different substrates on proximate composition are presented in table 5. There were significantly different on moisture content and crude fibre from cereal grain spawn, though there were not significantly different in ash content and crude fat and crude protein from interaction between substrates and grain spawn. The interaction between the combination of (wheat straw+ sawdust+ corn cobs) and popcorn grain spawn showed the highest

influence in moisture content (88.0) and interaction between sawdust substrate and popcorn spawn showed the lowest with 81.9% moisture content, respectively. The maximum and minimum ash content observed in the interaction between (kikuyu grass+ sawdust) substrates and rice grain spawn and interaction between corn cobs and corn spawn with 13.1 and 10.2%, respectively. The highest and lowest crude fibre found on the interaction between (sawdust+ wheat straw+ corn cobs) and popcorn spawn (18.67 %) and on the interaction of sawdust and popcorn cobs with 15.4% of crude fibre while also the interaction

between (Sawdust+ corn cobs+ kikuyu grass) substrates and popcorn spawn was the highest (2.01%) and the lowest (1.03%) crude fat found in the interaction between kikuyu grass substrate and popcorn spawn. The maximum and

minimum crude protein observed on the interaction between wheat straw and rice spawn and interaction between (kikuyu grass+ sawdust) substrate and popcorn spawn with 35.4 and 26.7% of crude protein, respectively.

Table 5: Effects of interaction between substrates and cereal grain spawn on proximate composition of oyster mushroom.

Substrates	Grains	Means				
		Moisture	Ash	Fibre	Fat	Protein
S1	1	85.96cdefg	11.32	17.17abc	1.42	35.42
S1	2	85.49efg	10.98	17.43abc	1.50	34.50
S2	1	82.42h	10.47	15.43c	1.62	34.50
S2	2	81.93h	10.23	17.73ab	1.72	33.86
S3	1	87.34abc	10.64	16.90abc	1.18	32.09
S3	2	86.11cdefg	10.52	17.64ab	1.03	31.08
S4	1	84.81g	10.41	17.53abc	1.64	28.53
S4	2	85.39fg	10.33	18.43a	1.69	28.86
S5	1	87.14abc	13.11	18.31a	1.66	28.20
S5	2	86.44bcdef	12.32	18.10a	1.63	26.73
S6	1	85.63defg	11.80	18.60a	1.68	33.03
S6	2	86.18bcdefg	11.55	17.25abc	1.73	32.03
S7	1	87.08abcd	13.04	18.21a	1.95	27.85
S7	2	86.92abcde	12.76	17.92ab	2.01	27.63
S8	1	87.62ab	12.83	15.91bc	1.85	32.43
S8	2	88.01a	12.92	18.67a	1.92	29.80
Stand.Error		0.30	0.23	0.43	0.08	0.63

The means followed by the same letters in the same (columns of moisture and fibre) are not significantly different using Tukeys' honest significant difference (HSD) test. Whereas substrate S1: wheat straw, S2: sawdust, S3: kikuyu grass, S4: popcorn cobs, S5: kikuyu grass+ sawdust, S6: sawdust+ popcorn cobs, S7: sawdust+ popcorn cobs+ kikuyu grass, S8: sawdust+ popcorn cobs+ wheat straw. Grain 1: Rice spawn; Grain 2: Popcorn spawn.

5.6. Effect of interaction between grain spawn and cropping shelters on moisture, ash content, crude fibre, fat and crude protein of oyster mushroom

The effects of different substrates on proximate composition are presented in table 6. The findings for analysis of variance showed that the interaction between grain spawn and cropping shelters statistically did not influence the moisture content, crude fibre, crude fat, ash content and crude protein ($P \leq 0.005$). The maximum and minimum moisture observed on the interaction between shade net shelter and rice spawn and on the interaction between dark house shelter and popcorn spawn with 87.4 and 84%, respectively. The interaction between shade net and rice spawn highly influenced the ash content (11.9%) of oyster mushrooms

while the interaction between dark house shelter and popcorn spawn showed the least (11.1%) to influence ash content. The highest and lowest crude fibre found on the interaction between shade net shelter and popcorn spawn (17.9) and interaction between mikeka shelter and rice spawn (17.2%), respectively. Based on crude fat, the interaction between dark house shelter and popcorn spawn was the highest (1.7%) and interaction of the shade net and rice spawn was the lowest (1.5%) and the crude protein results showed that the interaction between dark house shelter and rice spawn was higher (34.3%) while the interaction between shade net shelter and popcorn spawn with 27.1% crude protein showed the least amount.

Table 6: Effects of interaction between cropping shelters and cereal grains on proximate composition of oyster mushroom.

Shelters	Grains	Means				
		Moisture	Ash	Fibre	Fat	Protein
Mikeka	Rice	85.50	11.68	17.20	1.64	31.84
Mikeka	Popcorn	85.35	11.40	17.87	1.63	30.81
Shade net	Rice	87.40	11.92	17.20	1.55	28.35
Shade net	Popcorn	87.39	11.80	17.92	1.61	27.11
Dark house	Rice	85.11	11.51	17.37	1.68	34.33
Dark house	Popcorn	84.70	11.16	17.89	1.73	33.76
Stand.Errors		0.19	0.14	0.26	0.05	0.39

5.7. Effects of interaction among the substrates, cropping shelters and cereal grain spawn on moisture, ash content, crude fibre, fat and crude protein of oyster mushroom

The effects of different substrates on proximate composition are presented in table 7. The results indicated that interaction among the substrates, cropping shelters and cereal grain spawn statistically did not affect ($P \leq 0.005$) the moisture content, crude fibre, crude fat, ash content and crude protein. The maximum and minimum of moisture content indicated that the interaction among shade net shelter, (sawdust + popcorn cobs+ wheat straw) and popcorn spawn and on the interaction among dark house shelter, sawdust substrates and rice spawn with 90.0 and 80.8%, respectively. While the interaction among shade net, (sawdust + corn cobs+ wheat straw) and corn spawn and the interaction among dark house shelter, popcorn cobs and popcorn spawn showed the highest and lowest ash content of 13.4 and 9.7%, respectively. The higher value of crude fibre obtained on the interaction among dark house shelter, (Sawdust+ corn cobs) substrate with and rice spawn showed the lower value (19.07%) while the lower value found on the interaction between dark house, sawdust and rice spawn with 14.7% of crude fibre. The interaction among Mikeka shelter, (Sawdust+ corn cobs+ kikuyu grass) substrate and rice spawn was the highest with 2.1% crude fat whereas the interaction among shade net shelter, kikuyu grass and rice spawn was the lowest with 0.85% crude fat of oyster mushroom. Finally, the results obtained on crude protein showed that the maximum and minimum found on the interaction among dark house shelter, wheat straw substrate and rice grain spawn with 39.2% and on the interaction among shade net, popcorn cobs, and rice spawn with 24.1% crude protein of oyster, respectively.

Discussion

The study indicated that *P. ostreatus* can be successfully grown on almost all agricultural wastes but wheat straw under dark house shelter gave better results in terms of proximate composition mainly crude protein content. Statistically, significant variation was recorded in terms of crude protein, crude fibre, crude fat, moisture content and ash content of *Pleurotus ostreatus*. The proximate composition of mature fruiting bodies of oyster mushrooms cultivated on different substrates (wheat straw, corn cobs, kikuyu grass and sawdust) under different cropping shelters (Mikeka, shade net and dark house shelter) are shown in the Tables 1,2,3,4,5,6 and Table 7.

The moisture content of the present study ranged from 82.10 % to 90.06 % when the interaction of shade net shelter, (corn cobs+ sawdust+ wheat straw) substrates and popcorn spawn indicated high moisture content of 90.06 %. The result of the present study found more similar to the study of previous researchers like [22, 1, 24] who found the moisture content ranged from 88.8 to 90.33%, 88.2 to 93.4% and 88.15 to 91.6%, respectively. Whereas the results obtained in the study are lower than that reported by Kumar and Tripathi [26] who indicated the moisture ranged from 85.84 to 87.7%. The high moisture content found in mushrooms cultivated under shade net shelter might be due to the high quantity of water from irrigation and rainfalls

hold by casing soils used to moisture the substrates and it might depend on the maturity of fruiting bodies, species and storage conditions during packaging or processing as well [25, 27].

The present study revealed that the ash content of studied mushroom ranged between 10.2% - 12.9%. The amount of ash was higher in the interaction of shade net× (kikuyu grass+ sawdust) substrates× rice grain spawn substrates with 13.4% of ash content, than other interactions. The result of the present study matches with the findings of the previous one reported by Kumar & Tripathi [26] who reported the ash content ranged 12.8-13.35%, [22] who reported 8.5-13% of ash content and [29] who reported 8.89-13.9% of ash content. The findings of the present study were differed by the findings of [1] who found 0.33-1.06% of ash content, [28] reported 0.5-0.6% of ash in dried *P. sajor-caju*; whereas [31] recorded 1.1 and 8.28 g/100g in fresh and dried *P. sajor-caju*, respectively and [34] who found that ash content was ranged in 6.58 to 8.41%. The amount of ash depends on salt content in substrates [9]. This might be the reason why kikuyu grass and sawdust substrates showed the highest amount of ash content. The study observed that the crude fibres ranged between 14.6 and 19.6%. The same as ash content, the quantity was high in the interaction of dark shelter× (Sawdust+ kikuyu grass) substrate× rice spawn with 19.06% of fibres. The findings were in agreement with [22, 29], who indicated the crude fibre ranged between 17.3 and 20.5%, 14.7-19.15%, respectively; whereas [26, 1, 33] reported lower crude protein of 13.3-14.03 %, 2.2-3.5% and 1.6-3.9% than the present findings. The high dietary fibre content of fruiting bodies from sawdust substrates might have resulted from the high lignin and cellulose content of sawdust.

Crude fat content data of the dried mushroom are presented in the range of 0.85 and 2.10 % and were significantly different among them. The results obtained in this study were close to those obtained by [29] who reported the crude fat of 1.9 and 2.4% and [34] who reported 2.66% crude fat for dried oyster mushroom. The findings were less similar to the results ranged between from 4 to 10% of crude fat obtained by [1]. The variation of crude fat and fibre may be probably due to different substrates, biological, chemical differences and C/N ratio of the substrates which are also indicated by several authors [30].

The analysis of mushroom composition indicated that the crude protein ranged from 24.19% to 39.1% and wheat straws spawned by rice grain spawn cultivated under dark house cropping shelters showed higher crude protein (39.19%) than other substrates. Dark house shelter showed higher dry matter and crude protein than other shelters, this may be associated with a lower moisture content of substrates hanging under dark house from less substrates water uptake. The findings collaborated with [35] who reported that extending the irrigation interval and limiting irrigation volume for the second part of the tomato crop cycle, appeared to be the best management strategy to optimize the yield and nutritional quality of processing tomato. These results were confirmed with the findings of [22, 29] who reported the findings close to the present study of 26.2% and 27.3% of crude protein. The nitrogen present in the

substrate after spawn running enhances the mushroom yield and quality [15]. The carbon and nitrogen influence the proximate composition of oyster mushrooms in the mycelium [36]. The results corroborate the findings of [37] which reported that *Pleurotus ostreatus* are considered to be one of the most efficiency producers of the food protein. Regardless of the crude protein of oyster mushrooms grown on different substrates, the overall crude proteins were relatively superior to several other research findings.

Conclusion

The findings from the present study are the first to find out the nutritional content of *Pleurotus ostreatus* fruiting bodies grown under semi-controlled environmental conditions. The study revealed that the highest moisture content was obtained on sawdust substrates under shade net, ash content on a combination of sawdust+ wheat straw+ corn cobs substrates under shade net, crude fibre on the combination of sawdust + corn cobs substrates under mikeka, crude fat on a combination of corn cobs+ sawdust+ kikuyu grass substrates under mikeka and crude protein was observed on wheat straw grown under dark house shelter. From the result, it is recommended that the farmers harvest the fruiting body before they become over mature to maintain a high amount of crude protein found in mature oyster mushrooms mainly grown in dark house shelter for better crude protein, crude fibre and low fat.

Acknowledgments

This study was funded by my parents.

References

- Ahmed N, Saifullah S, Shah B, Iqbal B., Khan H, Khan I, Naeem A, Ullah W, Khan N, Adnan M, Rizwan AS, Junaid K, Iqbal M (2016) Substrates evaluation for the quality, production and growth of oyster mushroom (*Pleurotus florida* Cetto). Article in Journal of Entomology and Zoology Studies. 4, 98-107.
- Marshall E, Nair NG (2009). Make money by growing mushrooms (Vol. 7). Rome: FAO. Retrieved from <http://www.fao.org/3/a-i0522e.pdf>
- Müller O, Krawinkel M (2005) Malnutrition and health in developing countries. *Cmaj*. 173, 279-286.
- Chang ST (2007) Training manual on mushroom cultivation technology, APCAEM, from <http://www.un-csam.org/publication/TMMushroom.pdf>
- Oei p (2003) Mushroom Cultivation: Appropriate Technology for Mushroom Growers. The Netherlands BackhuysPublishing: Leiden.
- Raman J, Jang KY, Oh YL, Oh M, Im JH, Lakshmanan H, Sabaratnam V (2021). Cultivation and nutritional value of prominent *Pleurotus* spp.: An overview. *Mycobiology*. 49, 1-14.
- Benjamin (2015) Mushrooms A Beginners Guide To Home Cultivation(2 nd edition).CC public Domain, pp 24-27 .www.pixabay.com.
- Hoa HT, Wang CL (2015) The effects of temperature and nutritional conditions on mycelium growth of two oyster mushrooms (*Pleurotus ostreatus* and *Pleurotus cystidiosus*). *Mycobiology*. 4, 14-23.
- Iqbal B, Khan H, Saifullah IK, Shah B, Naeem A, Ullah W, Ahmed N (2016) Substrates evaluation for the quality, production and growth of oyster mushroom (*Pleurotus florida* Cetto). *Journal of Entomology and Zoology Studies*. 4, 98-107.
- Salami AO, Bankole FA, Olawole OI (2016) Effect of different substrates on the growth and protein content of oyster mushroom (*Pleurotus florida*). *International Journal of Biological and Chemical Sciences*. 10, 475.
- Thakur MP (2020) Advances in mushroom production: key to food, nutritional and employment security: A review. *Indian Phytopathology*. 73, 377-395.
- Singh A, Research PD, Singh IS, Pradesh U, Singh S (2021) Nutritional and Health Importance of Fresh and Dehydrated Oyster Mushroom (*Pleurotus florida*). *Journal of Current Research in Food Science*. 2, 10-14.
- Kimenju JW, Odero GOM, Mutitu EW, Wachira PM, Narla RD, Muiro WM (2009). Suitability of locally available substrates for oyster mushroom (*Pleurotus ostreatus*) cultivation in Kenya. *Asian Journal of Plant Sciences*. 8, 510 – 554.
- Zakil FA, Xuan LH, Zaman N, Alan NI, Salahutheen NAA, Sueb MS, Isha R (2022) Growth performance and mineral analysis of *Pleurotus ostreatus* from various agricultural wastes mixed with rubber tree sawdust in Malaysia. *Bioresource Technology Reports*. 17, 100873.
- Carrasco J, Zied DC, Pardo JE, Preston GM, Pardo-Giménez A (2018) Supplementation in mushroom crops and its impact on yield and quality, *AMB Express*. 8, 1-9.
- Yirankinyuki FF, Lamayi DW, Sadiq BA, Usman YM (2013) Proximate and some minerals analysis of *Colocasia esculenta* (Taro) leaves, *Journal of Medical and Biological Sciences*. 3, 9-10.
- Udo EJ, Ogunwele DA (1986). *Laboratory Manual for Analysis in Soil, Plants and Water Analysis 3rd Edition*, Ilorin, University of Ilorin, Kwara State Nigeria. 131-152.
- Official methods of the Association of Official Analytical Chemists (16th ed.) AOAC (1995) Arlington, VA, Association of Official Analytical Chemists.
- Udo EJ, Ogunwele DA (1986). *Laboratory Manual for Analysis in Soil, Plants and Water Analysis 3rd Edition*, Ilorin, University of Ilorin, Kwara State Nigeria. 131-152.
- Raghuramulu N, Madhavan NK, Kalyanasundaram S (2003) A Manual of Laboratory Techniques. National Institute of Nutrition. Indian Council of Medical Research, Hyderabad. 500, 56-58
- SAS Institute Inc (2017) SAS® 9.4 System Options: Reference, Fifth Edition. Cary, NC: SAS Institute Inc. AS®. 27513-2414.
- Roy Supta T, Nazmul Hossain M, Nuruddin M M, Uddin AK (2017) Effect of Different Sawdust substrates on the Growth, Yield, and Proximate composition of Oyster Mushroom (*Pleurotus florida*). *Bioresearch Communications*. 10, 47-58.
- Saifullah S, Shah B, Iqbal B., Khan H, Khan I, Naeem A, Ullah W, Khan N, Adnan M, Rizwan AS, Junaid K, Ahmed N, Iqbal M (2016) Substrates evaluation for the quality, production and growth of oyster mushroom (*Pleurotus florida* Cetto). Article in Journal of Entomology and Zoology Studies. 4, 98-107.

24. Asneti T (2013). Effect of different substrate supplements on oyster mushroom (*Pleurotus* spp.) production. *Food Science and Technology*. 44-51.
25. Tirkey VJ, Simon S, Lal, AA (2017) Efficacy of different substrates on the growth, yield and nutritional composition of oyster mushroom-*Pleurotus florida* (Mont.) Singer. *Journal of Pharmacognosy and Phytochemistry*. 6, 1097-1100.
26. Kumar M. and Tripathi, A. (2017). Study of the Different Types of Sugar Cane Planter in India. *IOSR Journal of Agriculture and Veterinary Science*, 10(7): 1-7. <https://doi.org/10.9790/2380-1007030107>.
27. Ares G, Lareo C, Lema P (2007) Modified atmosphere packaging for postharvest storage of mushrooms. A review. *Fresh Produce* 1, 32-40.
28. Bonatti M, Karnopp P, Soares HM, Furlan SA (2004) Evaluation of *Pleurotus ostreatus* and *Pleurotus sajorcaju* nutritional characteristics when cultivated in different lignocellulosic waste. *Food Chemistry*. 7: 425-428.
29. Tolera KD, Abera S (2017) Nutritional quality of Oyster Mushroom (*Pleurotus ostreatus*) as affected by osmotic pretreatments and drying methods. *Food Science and Nutrition*. 5, 989-996.
30. Li, H, Zhang Z, Li M, Li X, Sun Z (2017) Yield, size, nutritional value, and antioxidant activity of oyster mushrooms grown on perilla stalks. *Saudi Journal of Biological Sciences*, 24, 347-354.
31. Alam N, Amin R, Khan A, Ara I, Shim MJ, Lee MW (2001) Nutritional analysis of cultivated mushrooms in Bangladesh *Pleurotus ostreatus*, *Pleurotus sajor-caju*, *Pleurotus florida* and *Calocybe indica*, *Mycobiology*. 5, 228-232.
32. Kulsum U, Hoque S, Ahmed KU (2009) Effect of different levels of cow dung with sawdust on yield and proximate composition of oyster mushroom (*Pleurotus ostreatus*). *Bangladesh J. Mushroom*. 3, 25-31.
33. Onyeka EU, Okechie MA (2018) Effect of substrate media on growth, yield and nutritional composition of domestically grown oyster mushroom (*Pleurotus ostreatus*). *African Journal of Plant Science*, 12, 141-147.
34. Favati F, Lovelli S, Galgano F, Miccolis V, Di T, Candido V (2009) Processing tomato quality as affected by irrigation scheduling. *Scientia Horticulturae*. 122, 562-571.
35. Reguła J, Siwulski M (2007) Dried shiitake (*Lentinula edodes*) and oyster (*Pleurotus ostreatus*) mushrooms as a good source of nutrient. *Acta Scientiarum Polonorum Technologia Alimentaria*, 6, 135-142.
36. Prasad S, Rathore H, Sharma S, Tiwari G (2018). Yield and proximate composition of *Pleurotus florida* cultivated on wheat straw supplemented with perennial grasses. *Indian J. Agric. Sci*. 88, 91-94.
37. Dril AA, Mayurnikova LA, Rozhdestvenskaya LN (2021) The influence of ionizing radiation on free protein content and microorganisms' growth in oyster mushrooms. In *IOP Conference Series: Earth and Environmental Science*. IOP Publishing, 640, 032-060.