Effect of different nutritional supplements on the productivity and quality of oyster mushroom (*Pleurotus ostreatus*)

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ABSTRACT

Species of mushroom are considered as a source of protein and possessing high medicinal and economical values. This study was carried out in growth chamber on *Pleurotus ostreatus* L. strain 66 to investigate the effect of different levels of supplements on yield quantity and quality during the two seasons of 2016/2017 and 2017/2018. Four supplements with three levels of each one was used in this investigation i.e. wheat and rice bran were added the levels of 5, 15 and 25%, while the urea and zinc sulfate were added the levels of 0.5, 1.5 and 2.5% were examined. The obtained results showed that, wheat bran with the second level gave the highest number of fruits/bag, total weight of fruits and biological efficiency/bag in the both seasons, respectively. While the lowest number of fruits/bag, total weight of fruits and biological efficiency/bag was noted from urea with the third level in the both seasons respectively. The highest stalk length was obtained from urea with the third level in the both seasons. While the largest stalk and fruit bodies diameters resulted from wheat bran with second level during both experimental seasons. The highest nutritional values of potassium and fat were obtained from rice bran with second level. The wheat bran with second level gave the highest value of protein and energy content. On contrast, the lowest results for protein and energy content were obtained from urea and the third level. The obtained results showed that the control gave the lowest value of potassium and fat content. These results recommended that the best yield and quality were obtained from wheat bran with second level of this supplement. The wheat and rice bran supplements increased the oyster mushroom productivity and quality.

Keywords: Biological efficiency; Oyster mushroom; Nitrogen; Supplements; Protein content; Wheat bran.

INTRODUCTION

Mushroom species are fruiting bodies of fungi. They are classified in Kingdom of Fungi, which is also called Kingdom of Mycetae (Ammirati and Seidl, 2007). The use of mushroom as food is an old age practice and there was cultivation record in 60 A.D. (Chang and Miles, 2004; Tripathi, 2005; Belachew *et al.,* 2011). Species of mushroom are considered as a source of protein and possessing high medicinal and economical values (Chang and Miles, 2004; Bhupinder and Ibitwar, 2007). Mushroom farming has two inseparable phases; spawn production and fruiting body cultivation. Spawn is the planting seed of mushroom and technically, it is an expanding mushroom mycelium colonizing a given substrate media (Oei, 2003). Currently, edible mushrooms are cultivated in more than 100 countries (Chang, 2006). The main constraints facing the majority of mushroom producers in developing countries including China is the lack of good quality spawn that meets consumers' preference (Belachew, 2011; Belachewl and Workie, 2013). The production of good quality of spawn requires a strict laboratory procedure in which maintaining sanitation and purity of the spawn are critical importance (Wach, 2012). The quality of spawn affects both the yield and quality of cultivated mushroom (Stanley, 2010; Mbogoh *et al.,* 2011). The nutritional supplementation of cultivation substrate is an important cultural practice of mushroom cultivation (Ayodele and Okhuoya, 2007). Most of the growth, yield and quality parameters varied when mushroom was cultivated with different levels of supplements (Mahbuba *et al.*, 2010). Therefore, the substrate supplementation with various additives including nitrogen sources has been reported to improve growth, yield and quality of mushrooms (Jadhav *et al.,* 1998; Khare *et al.,* 2010 and Onyango *et al.,* 2011). They usually change the decomposition rate of substrate components. In most cases the efficiency of agricultural waste acting as substrates is considerably enhanced when supplemented with protein-rich materials (Frimpong–Manso *et al.,* 2011). The various supplements (wheat bran, ammonium sulphate, gram flour, soybean meal, rice bran, mustard cake, cotton seed cake, and molasses) are recommended as substrate supplements prior to spawning to enhance oyster mushrooms (Naraian *et al.*, 2009). The substrate supplementation is a practice used in producing *Pleurotus spp.* in order to increase its productivity. Inclusion of additives to mushroom substrate is very important especially for substrates having low protein content to enhance the growth and yield of mushrooms (Assan and Mpofu, 2014). Generally, the type of base substrate and additives percentage could affect carbon to nitrogen ratio (C/N), pH, moisture content, compaction, O_2 and CO_2 concentrations and the temperature of the media (Dung *et al.*, 2012 and Randive, 2012).

MATERIALS AND METHODS

The experiment was conducted in Vegetables laboratory, at Horticulture Department, Faculty of Agriculture Al-Azhar University, Cairo and Central Laboratory for Agricultural Climate (CLAC) Minestry of Agriculture, Dokki, during the two successive seasons of 2016/2017 and 2017/2018 under the environmental control of growth chamber. In this study, cultivar of mushroom (*Pleurotus ostreatus L* starin 66) was used to evaluate their characterization under three levels of supplements by using JUNCAO technique. The spore of the cultivar was obtained from CLAC. Four supplements were prepared for this experiment as wheat bran, rice bran, urea, and zinc sulfate. Thirteen treatments of rice straw supplemented with the four nutritional supplements were established. The nutritional supplements were used in three concentrations. The addition of wheat and rice bran were in concentrations of 5, 15 and 25%, while, urea and $ZnSO_4$ were used in concentrations of 0.5, 1.5 and 2.5%. The nutritional supplements were mixed thoroughly with rice straw. The rice straw material was chopped into small pieces (2-3 cm) using a grinding machine at CLAC. Each treatment was constituted as following formula:

T0 = 100 % Rice straw substrate + 0 % supplement (Control).
T1= 95 % Rice straw + 5% wheat bran t (w/w).
T2= 85 %Rice straw +15% wheat bran (w/w).
T3= 75 % Rice straw + 25% wheat bran (w/w).
T4= 95 % Rice straw + 5% rice bran (w/w).
T5= 85 % Rice straw + 15 % rice bran (w/w).
T6= 75 % Rice straw + 2 5% rice bran (w/w).
T7= 99.5 % Rice straw +0.5% urea (w/w).
T8= 98.5 % Rice straw +1.5% urea (w/w).
T9= 97.5% Rice straw + 2.5% urea (w/w).
T10= 99.5 % Rice straw + 0.5% $ZnSO_4$ (w/w).
T11=98.5 % Rice straw + 1.5% $ZnSO_4$ (w/w).
T12= 97.5 % Rice straw + 2.5% $ZnSO_4$ (w/w).

The moisture content was adjusted to be about 60-70% and pH was adjusted to be 7 according to Zhanxi and Dongmei, 2008. The rice straw mixing s were filled in polypropylene bags (40 cm length ×18 cm in diameter). 12 bags were packaged for each treatment; each was containing 300g dry substrate. The bags were then marked by permanent marker. All bags were autoclaved at 121°C for two hours and allowed to cool overnight.

The spawn medium was filled in high-density polyethylene bags each was containing 1 kg. Bags were sterilized at 121°C for 2 hours according to Zhanxi and Dongmei, 2008**,** then inoculated with the spawn on the surface of substrate (about 5 %) under aseptic conditions. Each treatment consists of 12 bags divided into three groups each are four bags representing three replicates. Groups were distributed randomly on shelves in the growth chamber. Bags were allowed to complete mycelial growth in dark at temperature degrees 27°C (the favorable conditions of mycelial growth stage). After colonization (20 days later), the plastic bags were opened from cotton terminal plug, and climatic conditions then were changed; the temperature degree was reduced to be 20°C, the relative humidity was raised to be 90 % and light intensity was adjusted to be 350 lux (the favorable conditions of fruit bodies formation stage). In this experiment, the following measurements were recorded:

Yield characteristics: 1) Number of fruits/bag. 2) Total fruit weight /bag. 3) Biological efficiency/bag.

Characteristics of oyster mushroom

Physical characteristics: 1) Stalk length. 2) Stalk diameter. 3) Cap diameter.

Chemical characteristics of fruit bodies: 1) Potassium (g/100g.d.w.). 2) Nitrogen (g/100g.d.w.). 3) Fat (g/100g.d.w. 4) Protein (g/100g.d.w.). 5) Energy (k cal/100g. d. w.).

Determination procedures:

Yield

1) Number of fruit /bag: The harvested mature fruit bodies were counted per bag. 2) Total yield: the first flush, second flush, third flush and fourth flush were weighted in grams and

calculated. 3) Biological efficiency: was defined as the ratio of the fresh weight of harvested mushroom over dry weight of substrate (Pokhrel and Ohga, 2007).

Physical characteristics

1) Stalk length (cm): was measured by ruler from branching start point of junction. 2) Stalk diameter (cm) was measured by Vernier caliper. 3) Diameter of cap of fruit body (cm): was measured by Vernier caliper.

Chemical characteristics

Nitrogen content (g/100g. d. w.): The method for determining the nitrogen content was conducted according to Pella (1990). 2) Potassium content (g/100g. d. w.): was determined using an inductively coupled plasma atomic emission Spectrometer (ICP-AES0) according to Pella (1990). **3**) Fat percentage in oyster mushroom fruit body was determined by extracting certain weight of powdered sample with petroleum ether using the Soxhlet apparatus as described in the AOAC (1990). 4). Protein content of the samples was estimated by the macro Kjeldhal method employed to find the total nitrogen content. The contents of total nitrogen were multiplied by a factor of 6.25 to find the crude protein of the mushroom sample according to AOAC (1990). 5) Energy [Kcal/100g (d.w)] was determined by the equation of Sharma *et al.*, 2013 as follow = [(protein×4) + (Carbohydrate×4) + (fat×9)]**.**

Table 1. Chemical analysis of raw and spent substrates before and after oyster mushroom cultivation**.**

Characteristics		Nitrogen					Potassium				
Treatments	**Levels**	Raw	Spent 1	Fruit1	Spent 2	Fruit2	Raw	Spent 1	Fruit1	Spent 2	Fruit2
wheat bran	Frist	0.45	0.62	0.82	0.60	0.18	0.75	0.55	1.05	0.56	1.06
	Second	0.48	0.59	1.25	0.57	1.22	0.84	0.40	0.90	0.40	1.04
	Third	0.52	0.67	.087	0.64	0.87	0.88	0.59	0.97	0.57	0.99
Rice bran	Frist	0.36	0.60	0.65	0.58	0.65	0.62	0.50	0.97	0.51	0.92
	Second	0.38	0.58	0.91	0.60	0.93	0.66	0.35	1.71	0.34	1.35
	Third	0.41	0. 66	0.76	0.68	0.50	0.69	0.49	0.81	0.47	0.95
Urea	Frist	0.57	0.78	0.57	0.75	0.58	0.45	0.37	0.41	0.37	0.44
	Second	0.65	0.85	0.53	0.81	0.54	0.41	0.39	0.33	0.38	0.33
	Third	0.69	0.90	0.51	0.92	0.52	0.39	0.34	0.30	0.35	0.26
Zinc sulfate	Frist	0.55	0.69	0.59	0.66	0.61	0.47	0.40	0.53	0.41	0.52
	Second	0.58	0.65	0.62	0.64	0.62	0.44	0.37	0.47	0.38	0.46
	Third	0.61	0.70	0.67	0.75	0.68	0.38	0.33	0.42	0.33	0.43
Control		0.39	0.41	0.72	0.41	0.74	0.23	0.19	0.33	0.19	0.33
Characteristics		Protein					Fat				
Treatments	*Levels	Raw	Spent 1	Fruit1	Spent 2	Fruit2	Raw	Spent 1	Fruit1	Spent 2	Fruit2
Wheat bran	Frist	1.91	2.63	3.51	2.55	3.33	0.6.1	0.52	0.34	0.51	0.34
	Second	2.04	2.50	5.32	2.42	5.12	0.32	0.22	0.48	0.23	0.48
	Third	2.21	2.84	3.72	2.72	3.70	0.35	0.26	0.41	0.26	0.42
Rice bran	Frist	1.53	2.552	2.78	2.46	2.76	0.28	0.25	0.58	0.24	0.57
	Second	1.61	2.46	3.89	2.55	3.94	0.39	0.34	0.44	0.33	0.44
	Third	1.74	2.80.	3.24	2.89	3.13	0.25	0.22	0.51	0.22	0.50
Urea	Frist	2.42	3.31	2.44	3.18	2.48	0.44	0.32	0.40	0.31	0.39
	Second	2.76	3.61	2.26	3.44	2.30	0.2.3	0.20	0.47	0.22	0.46
	Third	2.93	3.82	2.16	3.91	2.22	0.58	0.45	0.55	0.43	0.52
Zinc sulfate	Frist	2.33	2.93	2.50	2.80	2.59	0.44	0.40	0.41	0.41	0.41
	Second	2.46	2.76	2.80	2.72	2.61	0.36	0.30	0.45	0.32	0.45
	Third	2.59	2.97	2.85	3.18	2.90	0.41	0.34	0.36	0.33	0.33
Control		1.65	1.82	3.08	1.82	3.17	0.39	0.28	0.35	0.39	0.36

*The levels of supplements were 5, 15 and 25 % in wheat and rice bran, while 0.5, 1.5 and 2.5 % in case of urea and zinc sulfate.

Statistical analysis

The experiment was statistically analyzed in a randomized complete block design one-way ANOVA with three replicates. The obtained data was subjected to the analysis of on way ANOVA and means were compared by L.S.D. method at 5% level of significance according to Snedecor and Cochran (1982).

RESULTS

Yield characteristics

Number of fruits/ bag

Data presented in Table (2) showed significant differences in fruit number /bag during the two experimental seasons. The highest fruit number/bag was obtained from wheat bran treatment (17.88 and 17.66), while the second level of supplement gave the best result (12.26 and 12.20) in the both seasons respectively. The interaction between treatments and additive levels exhibited highest fruit number/ bag 23.00 and 22.66) which obtained from wheat bran with the second level in the both seasons respectively.

Total weight of fruits /bag

The recorded data in Table (3) revealed significant differences in total weight of fruits/bag in the both seasons. The higher total weight of fruits /bag resulted from wheat bran treatment (248.66 and 244.77 g/bag), in contrast the second level of supplement which gave the heaviest weight (217.64 and 202.68 g/bag) during the two seasons respectively. The interaction between treatments and additive levels showed the highest total weight of fruits/bag (322.33 and 309.00 g/bag) which found in wheat bran with the second level of supplement in the both seasons respectively.

Table 2. Effect of mixing rice straw substrate with different levels of supplements on number of fruits/bag of *Pleurotus ostreatus* during the two seasons of 2016/ 2017 and 2017 /2018.

Item	Season							
	2016/2017				2017 /2018			
Level / Treatment	First	Second	Third	Mean	First	Second	Third	Mean
Control	7.00	7.00	8.00	7.33	7.66	7.66	7.66	7.33
Wheat bran	14.33	23.00	16.33	17.88	14.00	22.33	16.66	17.66
Rice bran	11.00	15.00	10.00	12.00	9.00	14.33	10.33	11.22
Urea	8.66	8.00	4.33	7.00	8.66	8.66	5.00	7.44
Zinc sulfate	7.00	8.33	7.00	7.44	7.66	8.33	7.66	7.88
Mean	9.60	12.26	9.13		9.33	12.20	9.40	
L.S.D at 5%	Treatments	(A)		0.91	Treatments	(A)		0.78
	Levels	(B)		0.71	Levels	(B)		0.60
	Interaction	(AXB)		1.59	Interaction	(AXB)		1.36

*The levels of supplements were 5, 15 and 25 % in wheat and rice bran, while 0.5, 1.5 and 2.5 % in case of urea and zinc sulfate.

Table 3. Effect of mixing rice straw substrate with different level of supplements on the total weight of fruits/bag (g) of *Pleurotus ostreatus* during the two seasons of 2016/ 2017 and 2017 /2018.

Item	Season							
	2016/2017				2017 /2018			
Level / Treatment	First	Second	Third	Mean	First	Second	Third	Mean
Control	104.33	104.33	104.33	104.33	108.80	108.80	108.80	108.80
Wheat bran	235.66	322.33	188.00	248.66	236.00	309.00	189.33	244.77
Rice bran	241.66	267.13	203.66	237.48	236.00	269.00	170.00	225.11
Urea	77.92	160.40	59.16	99.16	78.60	90.23	59.16	76.00
Zinc sulfate	145.30	234.00	113.13	164.14	146.20	236.40	114.63	165.74
Mean	160.97	217.64	133.46		161.12	202.68	128.45	
L.S.D at 5%	Treatments	(A)		16.44	Treatments	(A)		8.74
	Levels	(B)		12.73	Levels	(B)		6.77
	Interaction	(AXB)		28.48	Interaction	(AXB)		15.1

*The levels of supplements were 5, 15 and 25 % in wheat and rice bran, while 0.5, 1.5 and 2.5 % in case of urea and zinc sulfate.

Biological efficiency/bag

The data tabulated in Table (4) showed significant differences in biological efficiency/bag in the first and second seasons. The highest biological efficiency/bag was obtained from wheat bran treatment (82.88 and 81.81%), while the second level of supplement gave the best result (76.54 and 71.25 %) during the two seasons. The interaction between treatments and additive levels showed the highest biological efficiency/bag (107.44 and 102.99 %) was noted from wheat bran with second level in both seasons respectively.

Characteristics of oyster mushroom

Physical characteristics

Stalk length.

The recorded values in Table (5) cleared significant differences in stalk length during the two seasons. The tallest stalk length resulted from wheat bran treatment (4.45 and 4.49 cm), in contrast the second level of supplement which gave the longest stalks (3.73 and 3.71 cm) in the first and second seasons. Interaction between treatments and additive levels showed the higher stalk length (5.06 and 4.92 cm) was found from urea with the third level of supplement during the two experimental seasons.

Stalk diameter

The recorded numbers in Table (6) showed significant differences in stalk diameter during the two seasons. The largest stalk diameter was noted from wheat bran treatment (2.23 and 2.22 cm), while the third level of supplement gave the best result (1.81 and 1.82 cm) in the first and the second seasons. The interaction between treatments and additive levels showed highest value of stalk diameter (2.50 and 2.45 cm) was found from wheat bran and the second level during the both experimental seasons

Table 4. Effect of mixing rice straw substrate with different levels of supplements on the biological efficiency/bag of *Pleurotus ostreatus* during the two seasons of 2016/ 2017 and 2017 /2018.

Item / Level / Treatment	2016/2017 First	Second	Third	Mean	2017 /2018 First	Second	Third	Mean
Control	54.77	54.77	54.77	54.77	56.26	56.26	56.26	56.26
Wheat bran	78.55	107.44	62.66	82.88	79.34	102.99	63.10	81.81
Rice bran	80.55	89.03	57.88	75.82	78.66	89.66	56.77	75.03
Urea	25.92	53.46	19.72	33.03	26.16	26.91	19.71	25.27
Zinc sulfate	48.43	77.99	37.70	54.71	48.06	78.77	38.20	55.01
Mean	57.64	76.54	46.55		57.70	71.25	46.81	
L.S.D at 5%	Treatments (A)			3.63	Treatments (A)			2.86
	Levels (B)			2.81	Levels (B)			2.21
	Interaction (AXB)			6.29	Interaction (AXB			4.95

*The levels of supplements were 5, 15 and 25 % in wheat and rice bran, while 0.5, 1.5 and 2.5 % in case of urea and zinc sulfate.

Table 5. Effect of mixing rice straw substrate with different levels of supplements on the stalk length cm of *Pleurotus ostreatus* during the two seasons of 2016/ 2017 and 2017 /2018.

Item / Level / Treatment	2016/2017 First	Second	Third	Mean	2017 /2018 First	Second	Third	Mean
Control	2.11	2.11	2.11	2.11	2.29	2.29	2.29	2.29
Wheat bran	4.70	3.91	4.74	4.45	4.74	4.02	4.70	4. 49
Rice bran	3.38	3.91	2.83	3.38	3.23	3.87	2.84	3.31
Urea	3.57	4.53	5.06	4.39	3.50	4.44	4.92	4.29
Zinc sulfate	3.79	4.10	3.41	3.77	3.77	4.04	3.43	3.75
Mean	3.51	3.71	3.63		3.50	3.73	3.64	
L.S.D. at 5%	Treatments	(A)		0.12	Treatments	(A)		0.08
	Levels	(B)		0.09	Levels	(B)		0.06
	Interaction	(AXB)		0.12	Interaction	(AXB)		0.15

*The levels of supplements were 5, 15 and 25 % in wheat and rice bran, while 0.5, 1.5 and 2.5 % in case of urea and zinc sulfate.

Table 6. Effect of mixing rice straw substrate with different levels of supplements on the stalk diameter of *Pleurotus ostreatus* during the two seasons of 2016/ 2017 and 2017 /2018.

Item / Level / Treatment	2016/2017 First	Second	Third	Mean	2017 /2018 First	Second	Third	Mean
Control	0.90	0.90	0.90	0.90	1.03	1.03	1.03	1.03
Wheat bran	2.03	2.50	2.15	2.23	20.03	2.45	2.19	2.22
Rice bran	1.47	2.03	1.96	1.82	1.22	2.18	1.94	1.78
Urea	1.81	2.25	2.15	2.07	1.80	2.15	2.07	2.00
Zinc sulfate	1.41	1.22	1.92	1.51	1.44	1.24	1.85	1.51
Mean	1.52	1.78	1.81		1.50	1.81	1.82	
	Treatments	(A)		0.13	Treatments	(A)		0.09
L.S.D. at 5%	Levels	(B)		0.10	Levels	(B)		0.07
	Interaction	(AXB)		0.22	Interaction	(AXB)		0.17

*The levels of supplements were 5, 15 and 25 % in wheat and rice bran, while 0.5, 1.5 and 2.5 % in case of urea and zinc sulfate.

Table 7. Effect of mixing rice straw substrate with different levels of supplements on the cap diameter of *Pleurotus ostreatus* during the two seasons of 2016/ 2017 and 2017 /2018.

Item / Level / Treatment	2016/2017 First	Second	Third	Mean	2017 /2018 First	Second	Third	Mean
Control	7.10	7.10	7.10	7.10	7.13	7.13	7.13	7.13
Wheat bran	9.96	13.56	8.12	10.55	9.98	13.71	8.21	10.63
Rice bran	8.54	10.33	7.77	8.88	8.15	10.19	7.69	8.67
Urea	5.81	5.99	5.62	5.81	5.86	5.14	5.70	5.56
Zinc sulfate	8.06	8.79	7.67	8.17	8.10	8.75	7.62	8.15
Mean	7.89	9.15	7.26		7.84	8.98	7.27	
	Treatments	(A)		0.34	Treatments	(A)		0.30
L.S.D. at 5%	Levels	(B)		0.26	Levels	(B)		0.23
	Interaction	(AXB)		0.59	Interaction	(AXB)		0.53

*The levels of supplements were 5, 15 and 25 % with wheat and rice bran, while 0.5, 1.5 and 2.5 % in case of urea and zinc sulfate.

Cap diameter

The data in Table (7) showed significant differences in cap diameter during the both experimental seasons. Largest cap diameter resulted from wheat bran treatment (10.55 and 10.63 cm), while the second level of supplement gave the largest one (9.15 and 8.98 cm) during the two seasons. The interaction between treatments and additive levels show best value of cap diameter (13.56 and 13.71 cm) was obtained from wheat bran with the second level of additive during the two seasons, respectively.

***Chemical characteristics*:**

Nitrogen

The data presented in Table (8) showed significant differences in the nitrogen content during the two experimental seasons. The highest value of nitrogen content was noted from wheat bran treatment (0.98 and 0.97 g/100g,), in contrast the second levels of supplement gave the higher result (0.83 and 0.83g/100g) during the two seasons. The interaction between treatments and additive levels showed best value of nitrogen content (1.25 and 1.22g/100g) was found from wheat bran with second level in the two seasons respectively.

Potassium

The recorded value in Table (9) showed significant differences in potassium content of the fruits in the two experimental seasons. The higher value of potassium content (1.16 and 1.03 g/100g) was obtained from rice bran treatment, while the second levels of supplement gave the higher result (0.85 and 0.79g/100g) during the two seasons. The interaction between treatments and additive levels showed best value of potassium content (1.71 and 1.35g/100g), which was found from rice bran with second level in the two season.

Table 8. Effect of mixing rice straw substrate with different levels of supplements on the nitrogen content diameter fruits of *Pleurotus ostreatus* during the two seasons of 2016/ 2017 and 2017 /2018.

Item / Level / Treatment	Season 2016/2017 First	Second	Third	Mean	Season 2017 /2018 First	Second	Third	Mean
Control	0.72	0.72	0.72	0.72	0.74	0.74	0.74	0.74
Wheat bran	0.82	1.25	0.87	0.98	0.81	1.22	0.87	0.97
Rice bran	0.65	0.91	0.76	0.77	0.65	0.93	0.50	0.69
Urea	0.57	0.53	0.51	0.54	0.58	0.54	0.52	0.55
Zinc sulfate	0.59	0.62	0.67	0.62	0.61	0.62	0.68	0.63
Mean	0.66	0.83	0.70		0.66	0.83	0.64	
L.S.D. at 5%	Treatments	(A)		0.02	Treatments	(A)		0.18
	Levels	(B)		0.01	Levels	(B)		0.17
	Interaction	(AXB)		0.03	Interaction	(AXB)		0.18

*The levels of supplements were 5, 15 and 25 % in wheat and rice bran, while 0.5, 1.5 and 2.5 % in case of urea and zinc sulfate.

Table 9. Effect of mixing rice straw substrate with different levels of supplements on the potassium content of *Pleurotus ostreatus* during the two seasons of 2016/ 2017 and 2017 /2018.

Item / Level / Treatment	Season 2016/2017 First	Second	Third	Mean	Season 2017 /2018 First	Second	Third	Mean
Control	0.33	0.33	0.33	0.33	0.33	0.33	0.33	0.33
Wheat bran	1.05	0.90	0.97	0.97	1.06	1.04	0.99	1.07
Rice bran	0.97	1.71	0.81	1.16	0.92	1.35	0.95	1.03
Urea	0.41	0.33	0.30	0.35	0.44	0.33	0.26	0.34
Zinc sulfate	0.53	0.47	0.42	0.47	0.52	0.46	0.43	0.47
Mean	0.74	0.85	0.62		0.73	0.79	0.65	
L.S.D. at 5%	Treatments	(A)		0.09	Treatments	(A)		0.08
	Levels	(B)		0.08	Levels	(B)		0.09
	Interaction	(AXB)		0.09	Interaction	(AXB)		0.08

*The levels of supplements were 5, 15 and 25 % in wheat and rice bran, while 0.5, 1.5 and 2.5 % in case of urea and zinc sulfate.

Fat content

The results data in Table (10) showed significant differences in fat content of the fruits in the first and the second seasons. The higher value of fat content (0.51 and 0.50 g/100g) was obtained from rice bran treatment, while the second levels of supplement gave the higher result (0.44 and 0.44g/100g) during the two seasons respectively. The interaction between the treatments and additive levels showed the best value of fat content (0.58 and 0.57g/100g) was found from rice bran with first level during the two seasons.

Protein

The presented data in Table (11) exhibited significant differences in the protein content of the fruits during the two seasons of 2016/2017 and 2017/2018. The highest value of protein content was obtained from wheat bran treatment (4.18 and 4.08g /100g,), while the second levels of supplement gave the highest result (3.57 and 3.51g/100g) in the two seasons. The interaction between the treatments and the additive levels showed best value of protein content (5.32 and 5.21g/100g) was found from wheat bran with the second level during the both seasons respectively.

Energy (kcal/100g. d. w.)

The presented data in Table (12) showed significant differences in the energy content of fruit bodies during the two experimental seasons. The highest value of energy content was found from wheat bran treatment (175.66 kcal/100g) in the first season and with rice bran (153.33 kcal/100g) in the second season. The first level of supplement gave the higher result (149.57 kcal/100g in the first season and 148.04 kcal/100g) during the second season. The interaction between treatments and additive levels showed the best value of energy content (162.81 and 158.12 kcal/100g) was found from wheat bran with second level during both seasons respectively.

Table 10. Effect of mixing rice straw substrate with different levels of supplements on the fat content of *Pleurotus ostreatus* during the two seasons of 2016/ 2017 and 2017 /2018.

Item / Level / Treatment	Season 2016/2017				Season 2017 /2018			
	First	Second	Third	Mean	First	Second	Third	Mean
Control	0.35	0.35	0.35	0.28	0.36	0.36	0.36	0.36
Wheat bran	0.34	0.48	0.41	0.41.	0.34	0.48	0.42	0.41
Rice bran	0.58	0.44	0.51	0.51	0.57	0.44	0.50	0.50
Urea	0.40	0.47	0.55	0.47	0.39	0.46	0.52	0.46
Zinc sulfate	0.41	0.45	0.36	0.40	0.41	0.45	0.33	0.40
Mean	0.37	0.44	0.44		0.41	0.44	0.43	
L.S.D. at 5%	Treatments	(A)		0.06	Treatments	(A)		0.02
	Levels	(B)		0.04	Levels	(B)		0.01
	Interaction	(AXB)		0.10	Interaction	(AXB)		0.03

*The levels of supplements were 5, 15 and 25 % in wheat and rice bran, while 0.5, 1.5 and 2.5 % in case of urea and zinc sulfate.

Table 11. Effect of mixing rice straw substrate with different levels of supplements on the nitrogen content of *Pleurotus ostreatus* during the two seasons of 2016/ 2017 and 2017 /2018.

Item / Level / Treatment	Season 2016/2017				Season 2017 /2018			
	First	Second	Third	Mean	First	Second	Third	Mean
Control	3.08	3.08	3.08	3.08	3.17	3.17	3.17	3.17
Wheat bran	3.51	5.32	3.72	4.18	3.33	5.21	3.70	4.08
Rice bran	2.75	3.89	3.24	3.29	2.76	3.94	3.13	3.27
Urea	2.44	2.26	2.16	2.29	2.48	2.30	2.22	2.33
Zinc sulfate	2.50	2.80	2.85	2.72	2.59	2.61	2.90	2.70
Mean	2.80	3.57	2.99		2.79	3.51	2.99	
L.S.D. at 5%	Treatments	(A)		0.10	Treatments	(A)		0.18
	Levels	(B)		0.11	Levels	(B)		0.16
	Interaction	(AXB)		0.12	Interaction	(AXB)		0.17

*The levels of supplements were 5, 15 and 25 % in wheat and rice bran, while 0.5, 1.5 and 2.5 % in case of urea and zinc sulfate.

Table 12. Effect of mixing rice straw substrate with different levels of supplements on the energy content of *Pleurotus ostreatus* during the two seasons of 2016/ 2017 and 2017 /2018.

Item / Level / Treatment	Season 2016/2017				Season 2017 /2018			
	First	Second	Third	Mean	First	Second	Third	Mean
Control	134.55	134.55	134.55	134.55	136.74	136.74	136.74	136.74
Wheat bran	153.93	162.81	156.24	175.66	141.92	158.12	147.49	149.18
Rice bran	144.02	160.38	115.21	139.87	152.88	1558.20	148.91	153.33
Urea	151.41	87.96	128.30	122.56	142.25	85.70	152.82	126.92
Zinc sulfate	148.93	143.38	153.52	148.61	149.20	152.96	142.93	148.39
Mean	149.57	138.63	138.32		146.58	138.74	148.04	
L.S.D. at 5%	Treatments	(A)		4.03	Treatments	(A)		8.03
	Levels	(B)		4.31	Levels	(B)		8.02
	Interaction	(AXB)		4.30	Interaction	(AXB)		8.01

*The levels of supplements were 5, 15 and 25 % in wheat and rice bran, while 0.5, 1.5 and 2.5 % in case of urea and zinc sulfate.

DISCUSSIONS

Oyster mushroom was successfully produced on agricultural wastes (substrates) supplemented with various levels of additives such as (wheat bran, rice bran, zinc sulfate and urea). The obtained results showed that the highest number of fruits/ bag, total weight, biological efficiency, stalk length and diameter, cap diameter, contents of nitrogen and protein were found from wheat bran supplement with the second level. Whereas the highest value of potassium and fat content resulted from rice bran with the second level. This result may be due to the different supplement combination in both physical and nutritional composition as well as microclimates (Amin *et al.*, 2008). In general, the number of fruit bodies per flush decreased from flush to flush indicating that the nature and amount of nitrogen available in a substrate after each flush influence the degree of cellulose degradation which in turn affects the yield (Frimpong–Manso, *et al.,* 2011). The highest, total weight and biological efficiency were found with wheat bran in the second level of these supplements. These results can be attributed to the nitrogen supplementation enhanced mushroom weight of fruit/ bag and biological efficiency especially when organic sources were used as wheat and rice bran. The supplementation of the substrate with various sources of organic nitrogen, such as wheat bran, rice bran, increased the weight of fruit/ bag and biological efficiency of oyster mushroom (Loss *et al.* 2009). Therefore, the organic sources of nitrogen for wheat and rice bran can be easily used by mushroom because the absorption of these molecules is more energetically efficient than synthesizing the molecules, which allow the mushroom to obtain more energy for mycelia growth and fruit formation. The increased total weight of fruits and biological efficiency can be due to the high availability of water in the substrate with wheat bran, since addition of rice bran decreases the granulometry of substrate, which improve the moisture retention (Özçelik and Peksen, 2007). The wheat bran with second level of this supplement contain middle concentration of nitrogen and protein content before cultivation of oyster mushroom in comparison to the other levels as shown Table (1).

For another point of view, considerable attention has been paid in these experiments towards the chemical contents which have been affected by using mixing rice straw substrate with three levels of four supplements (wheat bran, bran rice bran, urea and zinc sulfate). Therefore, the highest nitrogen, protein and energy content were found from wheat bran with second level of this supplement. The potassium content was found from rice bran with second level while, the fat content was found from rice bran with the first level of this supplement during the two seasons of 2016/2017 and 2017/2018. While, the lowest values of these previous characteristics was resulted from rice straw without any supplements except potassium and fat content. These results may be due to, the difference of nutritional composition of different supplements (Khan *et al.*, 2008). In turn the nutritional composition of mushroom depends to large extent on the status of the nutritional sources such as C/N ratio, vitamins, phytohormones, macro and microelements (Adenipekun and Gbolagade, 2015) and the biological differences of the substrates and supplements (Sangwan and Saini, 1999). The changes in nitrogen and protein contents in the fruit bodies depended on the C/N ratio in the cultivation substrate with the supplements (Yehia, 2012). Also, the protein content of mushrooms depends on several factors, such as the supplements chemical composition especially C/N ratio (Mane *et al.*, 2007). The excess of nitrogen may have affected the degradation of lignin, which may prevent the mycelium from developing (Zanetti and Ranal, 1997). Wheat bran was observed better supplement, which is rich in protein and fats, and is supposed to increase mushroom yields by promising growth of mycelium by certain amino acids present in wheat bran (Naraian *et al.*, 2009).

The highest nitrogen and protein content of the fruit bodies were obtained from applying the wheat bran with second level, while the highest potassium and fat content of the fruit bodies resulted from rice bran with the second level. These results may be attributed to the wheat bran with second level contain of middle nitrogen and protein content before cultivation of oyster mushroom in comparison to the other levels as shown Table (1). As a notice, the nature of the growing mushroom on wheat bran with second level consists of 1.25 and 1.04 g/100g nitrogen and 5.32and 5.12g/100 protein. The rice bran with the second level gave the best result of the potassium. The highest fat content resulted from rice bran with the first level, these results may be due to the rice bran with the second level contain of middle potassium and fat content before cultivation of oyster mushroom in comparison to the other levels as shown in Table (1). As a notice, the nature of the growing mushroom on rice bran with second level consists of 1.71 and1.35/100g potassium and level first 0.58 and 0.57g/100 fat. These findings may be due to, the application of second level of supplement proved it to be a viable option for oyster mushroom and recommended for commercial use while any supplementation above this level would reduce the growth and yield of oyster mushroom (Oseni *et al.*, 2012). Therefore, the best growth, yield and comical composition of oyster mushroom appeared with medium concentration of the additives, while the higher concentrations reduced the growth, yield comical composition of mushroom (Naraian *et al.*, 2009). These results may be due to the second level of supplement give rise to provide aeration of the substrates, which results from sufficient utilization of nutrients (Jafarpour *et al.*, 2010).

CONCLUSION

For seeking to follow the results in this experiment, the second point can be put in mind as recommendations for practical work. The first point recommends the highest number of fruits, total weight of fruits and biological efficiency were obtained from rice straw plus wheat bran with second level of supplement. The second point, recommend the best quality and nutritional value as (cap diameter and contents of nitrogen protein, and energy were resulted from the mixing rice straw with the wheat bran and the second level of supplement. The exploitation of spent mushroom substrate for the management of environment, agriculture and production of recyclable energy requires strict watch on its physical, chemical and microbiological properties.

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تاثير المكملات الغذائية المختلفة على انتاجية وجودة عيش الغراب المحاري

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الملخص العربي

تعتبر انواع عيش الغراب مصدرا للبروتين وتمتلك قيما طبية واقتصادية عالية. اجريت هذه الدراسة في غرفة النمو على سلالة *Pleurotus ostreatus* L. 66 لدراسة تاثير مستويات مختلفة من المكملات الغذائية على كمية ونوعية المحصول خلال موسمي ٢٠١٧/٢٠١٦ و٢٠١٨/٢٠١٧. تم استخدام اربعة مكملات تحتوي كل منها على ثلاثة مستويات في هذا البحث، اي تمت اضافة نخالة القمح والارز بمستويات ٥ و١٥ و٢٥٪، في حين تمت اضافة اليوريا وكبريتات الزنك بمستويات ٠,٥ و١,٥ و٢,٥٪. اظهرت النتائج التي تم الحصول عليها ان نخالة القمح بالمستوى الثاني اعطت اكبر عدد من الفاكهة / الكيس والوزن الاجمالي للفاكهة والكفاءة البيولوجية / الكيس في كلا الموسمين على التوالي. في حين لوحظ ان اقل عدد من الثمار/العبوة، الوزن الاجمالي للفاكهة والكفاءة البيولوجية /للعبوة من اليوريا مع المستوى الثالث في كلا الموسمين على التوالي. تم الحصول على اعلي طول ساق من اليوريا مع المستوى الثالث في الموسمين. بينما اكبر اقطار الساق والفاكهة ناتجة عن نخالة القمح بالمستوى الثاني خلال موسمي التجربة. تم الحصول على اعلي القيم الغذائية من البوتاسيوم والدهون من نخالة الارز مع المستوى الثاني. اعطى نخالة القمح مع المستوى الثاني اعلي قيمة للبروتين ومحتوى الطاقة. في المقابل، تم الحصول على ادنى النتائج لمحتوى البروتين والطاقة من اليوريا والمستوى الثالث. اوضحت النتائج ان عنصر التحكم اعطى اقل قيمة من محتوى البوتاسيوم والدهون. اوصت هذه النتائج بالحصول على افضل محصول وجودة من نخالة القمح مع المستوى الثاني من هذا المكمل. زادت مكملات نخالة القمح والارز من انتاجية وجودة الفطر المدروس.

الكلمات المفتاحية: الفعالية البيولوجية، عيش الغراب المحاري، نيتروجين، المكملات، محتوى البروتين، نخالة القمح.