



Effect of using desert weeds (Chenopodiaceae) as supplements in substrates of *Pleurotus ostreatus* (oyster mushroom) production

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Abstract

This study has been done to define the possibility of using two desert plant straws *Atriplex tatarica* and *Caroxylon cyclophyllum* for the first time as supplements with wheat straw substrate in producing mushroom (*Pleurotus osteratus*) to invest and build proper capacities for desert environments. This study included assessing the possibility of introducing 10% of these desert weeds straw as supplements in the formula of substrate to know their effects on the mycelial growth, mycelium completion period, primordial completion period, fruiting bodies number, yield and biological efficiency and protein content of *P. ostreatus* using three prepared spawns from barley, white corn and wheat seeds. Results showed that the produced spawn from the white corn exhibited the best results in the growth and yield of *P. ostreatus* compared to the spawn produced from wheat straw and barley. Also the *A. tatarica* straw-supplemented substrate showed a higher producing rate in fruiting bodies number, protein content, yield and biological efficiency. *A. tatarica* straw-supplemented substrate exhibited the shorter mycelial growth completion period (28.26 days). This substrate exhibited highest yield, and biological efficiency were 1489.2 g.kg⁻¹, and 40.01%, respectively. Also, results showed the best protein percent 24.14% and 24.00% when suing this substrate with barley seeds spawn during the first and second flushes, respectively. It exhibited the highest loss in the substrate weight was 44.3%. In conclusion, these results encourage the supplementation of agricultural substrates using desert weeds especially *A. tatarica* and apply in desert environments at the commercial level.

Key words – *Atriplex tatarica* – *Caroxylon cyclophyllum* – mushroom cultivation – *Salsola cyclophylla* – supplements

Introduction

The species *Pleurotus ostreatus* (oyster mushroom) grows naturally in nature and belongs to order Agaricales, Basidiomycetes class (Hibbett et al. 2007). *P. ostreatus* is characterized from other mushrooms having a simple and easy production technique, lower cost, higher growth rate

and biological efficiency and easy in preparing agricultural substrates (Çağlarirmak 2007). In addition, *P. ostreatus* has high protein content about 23%–25% (Owaid et al. 2017a), flavonoids (Akyüz et al. 2012), vitamins, fibers, carbohydrates and many bioactive compounds (Patel et al. 2012), and it comes next among edible mushrooms in regard to economic importance after *Agaricus bisporus*; and gives a good income to farmers with no need for a large agricultural area (Chang & Miles 2004). Besides, the spent mushroom substrate (SMS), after production, can be used as an bioorganic fertilizer to improve the productivity of soil which used for cultivating some edible mushrooms (Noonsong et al. 2016), vegetables and fruits in gardens and orchards (Mohd Hanafi et al. 2018, Owaid et al. 2017b, Polat et al. 2009). However, the cultivated oyster mushroom on various agro-wastes showed wide different antifungal activities, *in vitro* against same plant pathogenic fungi (Owaid et al. 2017c, 2017d).

The composition of substrates has an important effect in the mushroom growth (Barba et al. 2016) which can survive and grow in multi-cellulosic substrates (Mohd Hanafi et al. 2018). In order to get a higher rate in the mycelial growth, the used substrates must contain balanced quantities of nitrogen, carbohydrates and cellulose in addition to fibers (Owaid et al. 2015a, Jamil et al. 2019). The period of mycelial growth completion varied between three genera through 25–70 days, it was also found that the spawning period to produce fruiting bodies ranged between 8 to 15 days for different genera and substrates and the biological efficiency ranged between 26–38%. Besides, the cultivated oyster mushroom on the wheat straw substrate mixed with date-palm fibers and wood wastes showed productivity ranged 163–143g/ 500g substrate and biological efficiency between 45%–51% (Owaid et al. 2018).

Many researches recorded a wide range of plant residues that can be used as agricultural substrates for the mushroom cultivation. *P. ostreatus* was grew on agricultural substrates and different plant residues in many formulae such as wheat straw, date-palm fibers, wood sawdust (Owaid et al. 2015a), corn cobs (León 2003, Kimenju et al. 2009), barley straw (Gaitan-Hernandez & Salmones 2008), cattails straw (*Typha latifolia*) (Vetayasuporn 2007), rice straw, bean straw (*Phaseolus vulgaris*), finger millet straw (*Seteria microcheata*), sawdust of *Eucalyptus* sp., water hyacinth (*Eichhornia crassipes*), coconut fibers, banana fibers (Kimenju et al. 2009), olive wastes (Atila 2017), sunflower (*Helianthus annuus*) husks (Myronycheva et al. 2017), asparagus old stalks (Zhai & Han 2018), and spent oyster substrate with wheat straw supplanted by nano-urea (Naim et al. 2020), or industrial wastes like cardboard (Owaid et al. 2015b) and waste paper (Tsfay et al. 2020). Each researcher needs to seek for other new substrates for entering in *P. ostreatus* production to increase the productivity and nutritional components. Many studies used other available wild plants and agricultural residues as alternative substrates for oyster mushroom cultivation due to their low costs and availability during all seasons (Sözbir et al. 2015).

However, *A. tatarica* (Löve 1970) and *Caroxylon cyclophyllum* (Gardens 1957), belong to Chenopodiaceae family, are classified as weeds available in most of the Iraqi lands particularly Anbar Desert. It was found that *A. tatarica* (Hanif et al. 2018) and *C. cyclophyllum* (Al-Waheeb & Lafta 2015) are considered desert weeds that could afford drought conditions, higher temperatures and saline soils. They have a rich nutritional value with big quantity of seeds (used for cattle). *A. tatarica* (Chenopodiaceae) distributes in Saudi Arabia, Iraq, Jordan, Palestine, Saudi, Syria and Turkey, (Le Houerou 1992), and the Gulf States (Al-Turki et al. 2000). *C. cyclophyllum* (= *Salsola cyclophylla* s.l.p.p.) is also distributes in Iraq deserts (Ghazanfar & McDaniel 2016) but only few studies on this plant (Al-Waheeb & Lafta 2015). *A. tatarica* is used by Bedouins as a local medicine as it contains 1.13% saponin components, volatile oils, in addition to 23% crude protein, 25.21% fibers, 15% acidic lignin and 5.1% nitrate (Toderich et al. 2012). This study is carried out at University Of Anbar in Center of Desert Studies to know the possibility of benefitting from straws of *A. tatarica* and *C. cyclophyllum* for the first time as supplements to encourage growth and production of the edible mushroom.

Materials & Methods

Agricultural substrates

Suitable quantities of *tatarica* and *C. cyclophyllum* (Chenopodiaceae family) straws were collected from the desert area western Ramadi (Anbar Desert), as in Fig. 1, and wheat straw was collected from some rural fields in Ramadi, Iraq.



Fig. 1 – The Iraqi desert weeds (Chenopodiaceae) used in this study.

Mother Spawn

Pleurotus ostreatus mother spawn was obtained from labs of Mushroom Cultivation, Center of Agricultural Research, University of Cairo, Egypt. In this center, spawns were produced.

Spawn Production

In order to multiply and prepare the spawn on certain local plant seeds, it was cultured on PDA plates and incubated at $25\pm 2^{\circ}\text{C}$ till growing mushroom mycelia during 7 days. The mycelial culture was partitioned by taking 5 mm discs from the culture. These mycelial discs were moved to flasks, which used to spawn preparation, containing wheat, barley, and white corn seeds separately, after being boiled in water (1w:2v). Flasks were left for 20 min to exhaust water and seeds were spread to avoid free water. About 12.5 g of CaSO_4 and 3.3 g of CaCO_3 were added to 1 kg of plant seeds depending based on wet weight. The seeds were divided each 50 g in a 1L bottle. The bottles were firmly closed, sterilized at 121°C and 15psi in the autoclave for one hour and left to the next day. The bottles were inoculated with mother spawn (one week old), mixed together and put in an incubator at $25\pm 2^{\circ}\text{C}$ then shake once a week to ensure for a better spread of mushroom mycelia and avoiding agglomeration of seeds. The period of spawn growth (spawn completion period) and length of every treatment were calculated (Oei 2005).

Preparation of substrate mixtures

The agricultural straws were cut into pieces (2–6 cm in length). Table 1 shows chemical properties of the used substrates including wheat straw enriched by 10% of *A. tatarica* straw (W1) or 10% *C. cyclophyllum* straw (W2), and wheat straw enriched with 2% urea as a control (W3). The prepared substrates were washed to clean the dust, then soaked for 48 hr and spread on a concrete ground. The mixtures were separately sterilized by using pasteurization unit at 65°C (Kimenju et al. 2009). To avoid free water, 2.5% gypsum and 3.0% of calcium carbonate were properly mixed (Oei 2005).

Cultivation of oyster mushroom

The inoculation technique using 5% of *P. ostreatus* spawn is done in plastic bags (40×50 cm) with 1 kg capacity of substrate on basis of the dry weight (Owaid et al. 2015b). This spawn was added in mutual layers with substrate (10 cm thickness). After inoculation, the bags were properly tightened and transferred to the incubation room at 25°C away from light for 3–4 weeks till the

growth of mycelia at the overall substrate inside bags. The first growth stage was completed as the substrate was full of mushroom mycelia. Then bags were moved to the production room well-conditioned environmentally where temperature was 25°C and humidity 80–90%, well ventilation using air discharger and white fluorescent lighting. The first yield was carried out after the fruiting bodies reached the proper size. The quantity of the yield was assessed for 1kg substrate (fresh weight) for each bag by counting the number of flushes, number of fruiting bodies and the yield weight of each flush. Percent of biological efficiency and substrate weight loss were counted according to the substrate weight (Owaid et al. 2015b). Also, the protein, carbon, carbohydrates (Banik & Nandi 2004), and total fibers content of substrates were assessed (Dávila et al. 2020). Biological efficiency = (fresh weight of oyster mushroom/ dry weight of substrate)×100 (Owaid et al. 2018).

Table 1 Chemical composition of the applied substrates used in this study

	Materials	C (%)	N (%)	Protein (%)	Carbohydrate (%)	Fibers (%)
W1	Wheat straw	62	0.01	-	38.54	26.54
	10% <i>Atriplex tatarica</i>	52	3.57	22.7	32.63	21.57
W2	Wheat straw	62	0.01	-	38.54	26.54
	10% <i>Caroxylon cyclophylla</i>	-	-	5.67	39.82	32.34
W3	Wheat straw	62	0.01	-	38.54	26.54
	2% of urea	19	48.6	-	-	-

Statistical analysis

The experiment was done according to Completely Randomized Design (CRD) in triplicates using factor experiments. The averages were compared by the Least Significance Difference (LSD) at $p \leq 0.05$ using General Statistical Package GENSTAT.

Results and discussion

Effect of plant seeds type on the spawn growth completion period

The superiority of white corn seeds and wheat seeds to record the least spawn growth completion period reached 20.63 and 22.14 days, respectively, whereas a delay in spawn development period of barely seeds was showed (25.75 days) (Table 2). White corn seeds obtained the biggest spawn length reached 854.6 mm, followed by wheat spawn length (822.4 mm), while barely spawn recorded the less length (750.4 mm). The superiority of white corn is due to its rich carbohydrates content, its absorbing high water quantities and seeds cover nature after boiling. These properties could form a good medium for growing big mycelia in length during a short period. However, this study is disagreed to Sagir & Yildiz (2004) that reported the barley spawn better than wheat spawn, also low crud protein of barely seeds. In addition, we also noted the solidness of barley seeds and their resistant seed cover nature led to slow mycelial growth and length. Generally, quality of mycelial growth depends on the genetic nature of *Pleurotus* species and the quality of spawn is affected by the applied plant seeds in this side (Owaid et al. 2017a).

Effect of treatments on mycelial growth and primordial completion periods

The superiority of *Atriplex tatarica* straw-supplemented substrate (W1) which recorded the shorter rate for mycelial growth completion period (28.26 days), followed 31.76 and 30.2 days for the *C. cyclophyllum* straw-supplemented substrate (W2) and urea-enriched wheat straw substrate as a control (W3), respectively (Table 3). The spawn of white corn seeds recorded the least period of 28.33 days to complete covering the substrate by mycelia followed by the usage of barley spawn, and wheat spawn 30.00 and 31.86 days, respectively. It was clear that there was a significant ($p < 0.05$) effect for the mycelial growth completion period between treatments of substrate and spawn types. W1 substrate with white corn spawn recorded 26.2 days which considered the least

significant effect for the mycelial growth completion period, whereas the longest period was 33.6 days by the treatment of control (W3) with the spawn loaded on wheat seeds. These results confirmed the importance of supplements of *A. tatarica* straw in W1 substrate to obtain good protein content according to Table 1 in speeding up the mycelial growth completion period for the whole substrate area. It also showed the importance of white corn seeds in forming an active thick spawn by mycelia which reduced the mycelial growth completion period for the whole substrate, significantly, and more effective than other spawns used in this study. It was clearer in the interaction between treatments which reduced the period to 21.2% than other treatments.

Table 2 Completion period rate of spawn growth and length

Spawn type	Spawn completion period (days)	Length of completed spawn (mm)
Barley seeds	25.75	750.4
White corn seeds	20.63	854.6
Wheat seeds	22.14	822.4
LSD ($p < 0.05$)	2.15	34.32

Table 3 Effect of treatments on the mycelial growth completion period (days)

Treatments Spawn type (S)	Substrate type (W)			Average of substrates
	W1	W2	W3 (control)	
S1	28.2	30.4	31.4	30.00
S2	26.2	28.5	30.4	28.33
S3	30.4	31.6	33.6	31.86
Average of S	28.26	30.20	31.76	-
LSD ($p < 0.05$)	W = 1.61, S = 1.76, W*S = 2.63			

Note: W1 = 10% *Atriplex tartaric* straw-supplemented wheat straw substrate, W2 = 10% *Caroxylon cyclophyllum* straw-supplemented wheat straw substrate, W3 = 2% urea-supplemented wheat straw substrate (control), S1 = spawn of barley seeds, S2 = spawn of white corn seeds, S3 = spawn of wheat seeds, F1 = 1st flush, F2 = 2nd flush, F3 = 3rd flush, F4 = 4th flush

Table 4 showed the superiority of the substrate supplemented by *C. cyclophyllum* (W2) and *A. tatarica* (W1) which recorded the least period for spawning (11.8 and 11.4 days), compared with wheat straw substrate supplemented with urea (control, W3) which recorded 10.8 days. The spawn loaded on white corn seeds recorded the least time 9.67 days for the primordial completion period. But, spawns of barley and white corn seeds recorded a period of 11.4 and 12.2 days, respectively. It was clear there was a significant ($p < 0.05$) effect for the interaction between the substrate type and spawn type. The treatments of W1 and W2 with spawn of white corn seeds recorded the least average of primordial completion period reached 7.6 and 8.8 days significantly, whereas the longest period was 13.2 and 13.4 days in the case of using the treatment of control (W3) with spawns of barley and wheat seeds, respectively.

The mycelial completion period of *Pleurotus ostreatus* on the substrate and the primordial completion period are very important for its cultivation and very essential to give a short time to accomplish the growth stages in the case of mushroom production (Owaid et al. 2018). The difference between growths of mycelia is attributed to the disparity of substrate contents and the type and activity of the applied spawn. The different completion periods of mycelia growth and primordia depend on the nature and contents of substrate (Owaid et al. 2015a, Mkhize et al. 2016) and genetic diversity of oyster mushroom (Adebayo et al. 2014). These results agreed with finding of Adebayo et al. (2014) who recorded primordia formation days (11–19). Besides, the mycelial completion period in bags and time of forming primary primordia depend on substrate content of cellulose, lignin, proteins and substrate capability to provide necessary nutrients for different growth stages of *P. ostreatus* (Oei 2005, Vetayasuporn 2007, Mohd Hanafi et al. 2018).

Table 4 Effect of treatments on the primordial completion period (days).

Treatments Spawn type (S)	Substrate type (W)			Average of substrates
	W1	W2	W3 (control)	
S1	10.2	10.8	13.2	11.40
S2	7.6	8.8	10.8	9.67
S3	11.4	11.8	13.4	12.20
Average of S	9.63	10.20	12.46	-
LSD ($p < 0.05$)	W = 1.05, S = 1.12, W*S = 2.23			

Note: W1 = 10% *Atriplex tartaric* straw-supplemented wheat straw substrate, W2 = 10% *Caroxylon cyclophyllum* straw-supplemented wheat straw substrate, W3 = 2% urea-supplemented wheat straw substrate (control), S1 = spawn of barley seeds, S2 = spawn of white corn seeds, S3 = spawn of wheat seeds

Effect of treatments on the flushes number and mushroom production

The results of Table 5 showed that the optimal substrate was W1 (10% *A. tartaric* straw-supplemented wheat straw substrate) which recorded the best production average from four flushes, significantly ($p < 0.05$). A single flush of W1 substrate recorded 479.1 g.kg⁻¹. While W2 (10% *C. cyclophyllum* straw-supplemented wheat straw substrate) recorded a decrease to 308.5 g.kg⁻¹, whereas the production average of single flush of wheat straw substrate supplemented by urea (control) was 190.5 g.kg⁻¹. Moreover, it was found that the highest flush average recorded 372.4 g.kg⁻¹ using barley seeds spawn, the next average recorded 337.4 g.kg⁻¹ for spawn of white corn seeds. The lowest flush average was by wheat seeds spawn which recorded 268 g.kg⁻¹. Generally, it was clear that the highest flush average got 356.1 g.kg⁻¹ in the first flush, but the flush average decreased between 304.6 and 326.4 g.kg⁻¹ for the other flushes (2nd, 3rd, and 4th). The results proved the mutual interactions between the substrate type and applied spawn from side, and substrate or spawn type and number of flushes from another side. The best average of flushes recorded 536.3 and 524.8 g.kg⁻¹ with W1 substrate using barley seed and white corn seed spawns, respectively. The triple interaction showed the best single flush was 547 g.kg⁻¹ using W1 substrate with the white corn spawn, whereas the lowest one was 136 g.kg⁻¹ using W3 (control) with wheat seeds spawn. These results agree with (Owaid et al. 2015a) who referred to the positive correlation between flushes number from side and total yield, fruiting bodies number and biological efficacy from another side, also the suitable properties to encourage mycelial growth led to best flush number achieved.

Table 6 exhibited that the highest average of the total production was 1489.2 g.kg⁻¹ recorded by W1 substrate, while the lowest average was 1072.13 g.kg⁻¹ by the control (W3) substrate. Using the wheat seed spawn got the highest total production (1489.2 g.kg⁻¹), whereas the interaction between substrate type and spawn type showed total production 2145.6 and 2123.03 g.kg⁻¹ by W1 substrate with spawns of barley and white corn seeds, respectively. The total production of this mushroom depends on the straw type as agro-substrate (Owaid et al. 2015b).

The results of Table 7 showed significantly ($p < 0.05$) that the best applied substrates recorded the highest fruiting bodies number average are W2 and W1 (4.33 and 4.06 fruiting bodies per a single flush, respectively) compared with 3.97 fruiting bodies in the control (W3). The highest averages of fruiting bodies were 4.31 and 4.17 fruit/flush recorded by spawns of wheat and barley seeds, respectively, followed by spawn of white corn seeds (3.89 fruit/flush). It was clear that the highest number of fruiting bodies is in this sequence: the 3rd flush, 1st flush and 2nd flush 4.26, 4.22 and 4.04 fruit/flush, respectively. The mutual interactions between the types of substrate and spawn or substrate and the flushes number or the spawn type and the flushes number proved that the best average of the fruiting bodies recorded was 5.65 fruit/flush by spawn of white corn seeds in flush 4. Besides, many treatments recorded higher values of the fruiting bodies number reached 5.0 fruit/flush, such as W1 substrate with barley seeds spawn in the first flush, and W2 substrate with barely seeds and wheat seeds spawns in the fourth flush, and W3 substrate (control) with wheat seeds spawn in the third flush. It was noted that the lowest average of fruiting bodies recorded 3.33

fruit/flush by different treatments as in Table 7 which agreed with findings by Oei (2005). The superiority of W1 substrate in the number of fruiting bodies and their weights maybe referred to the organic protein content of this substrate (Table 1) and its nutritional content characteristics which encouraged the mycelia growth. These results agree with the some previous studies (Vetayasuporn 2007).

Table 5 Effect of the used substrate and spawn type on the flushes number (g.kg⁻¹)

Treatments		No. of flushes (F)				Average W*S	Average W
Substrate type (W)	Spawn type (S)	F1	F2	F3	F4		
W1	S1	548.0	520.0	541.0	536.0	536.3	479.1
	S2	547.0	510.3	541.0	511.3	524.8	
	S3	531.0	330.7	334.7	297.7	376.0	
W2	S1	348.0	387.0	374.3	336.0	361.3	308.5
	S2	347.0	343.7	264.0	244.7	299.8	
	S3	231.7	308.7	247.1	277.9	264.4	
W3 (Control)	S1	248.0	187.0	241.0	202.7	219.7	190.5
	S2	196.7	199.7	172.3	181.7	187.6	
	S3	211.3	155.7	136.0	154.3	164.0	
Average of F		356.1	326.3	316.8	304.6	-	-
Average of S*F		F1	F2	F3	F4	Average of S	-
S1		381.3	364.8	385.4	458.2	372.4	-
S2		363.6	351.2	322.3	297.7	337.4	-
S3		32.4	263.0	242.6	218.0	268.3	-
LSD ($p < 0.05$)		W = 19.38, S = 19.40, F = 22.38, W*S = 33.57, W*F = 38.76, S*F = 38.80, W*S*F = 67.4					

Note: W1 = 10% *Atriplex tartaric* straw-supplemented wheat straw substrate, W2 = 10% *Caroxylon cyclophyllum* straw-supplemented wheat straw substrate, W3 = 2% urea-supplemented wheat straw substrate (control), S1 = spawn of barley seeds, S2 = spawn of white corn seeds, S3 = spawn of wheat seeds, F1 = 1st flush, F2 = 2nd flush, F3 = 3rd flush, F4 = 4th flush

Table 6 Effect of the used substrate and spawn type on mushroom total production (g.kg⁻¹)

Treatments		Substrate type (W)			Average of W
Spawn type (S)	W1	W2	W3 (control)		
S1	2145.60	1445.3	876.7	1489.20	
S2	2123.03	1199.5	750.4	1357.64	
S3	1494.1	1065.0	657.3	1072.13	
Average of S		1920.68	1131.7	761.5	-
LSD ($p < 0.05$)		W = 58.12, S = 60.03, W*S = 100.71			

Note: W1 = 10% *Atriplex tartaric* straw-supplemented wheat straw substrate, W2 = 10% *Caroxylon cyclophyllum* straw-supplemented wheat straw substrate, W3 = 2% urea-supplemented wheat straw substrate (control), S1 = spawn of barley seeds, S2 = spawn of white corn seeds, S3 = spawn of wheat seeds

Table 7 Effect of the applied substrate and spawn type on the fruiting bodies number per flush

Treatments		No. of flushes (F)				Average W*S	Average W
Substrate type (W)	Spawn type (S)	F1	F2	F3	F4		
W1	S1	5.00	4.33	4.67	3.67	4.42	4.06
	S2	4.33	3.33	4.00	3.67	3.83	
	S3	4.33	3.67	4.33	3.33	3.92	
W2	S1	4.00	3.67	4.33	5.00	4.25	4.33
	S2	4.33	4.67	4.00	3.67	4.17	

Table 7 Continued.

Treatments		No. of flushes (F)				Average W*S	Average W	
Substrate type (W)	Spawn type (S)	F1	F2	F3	F4			
W3 (Control)	S3	4.33	5.00	4.33	4.67	4.58		
	S1	3.67	4.33	3.33	4.00	3.83		
	S2	3.67	3.33	4.33	3.33	3.67	3.97	
	S3	4.33	4.33	5.00	4.00	4.42		
Average of F		4.22	4.04	4.26	3.97	-	-	
Average of S*F		F1	F2	F3	F4	Average of S	-	
S1		4.22	4.11	4.11	4.22	4.17	-	
S2		4.11	3.78	4.11	5.65	3.89	-	
S3		4.33	4.33	4.65	4.00	4.31	-	
LSD ($p < 0.05$)		W = 0.55, S = 0.56, F = 0.63, W*S = 0.95, W*F = 1.10, S*F = 1.101, W*S*F = 1.91						

Note: W1 = 10% *Atriplex tartaric* straw-supplemented wheat straw substrate, W2 = 10% *Caroxylon cyclophyllum* straw-supplemented wheat straw substrate, W3: 2% urea-supplemented wheat straw substrate (control), S1 = spawn of barley seeds, S2 = spawn of white corn seeds, S3 = spawn of wheat seeds, F1 = 1st flush, F2 = 2nd flush, F3 = 3rd flush, F4 = 4th flush

Effect of treatments on the protein content of fruiting bodies

In Table 8, this study proved that the protein content in fruiting bodies has been affected by the type of the used substrate and spawn. The highest protein content average was 22.54% by W1 substrate. The higher percent was 22.35% by barley seeds spawn. It was also found that the highest percent in the first and second flushes recorded 22.79% and 22.20%, respectively. Also, the collaborative interactions led to record the highest protein percent 23.41% when using W1 substrate with barley seeds spawn. Also the triple interaction showed the best protein percent 24.14% and 24.00% when using W1 substrate with barley seeds spawn during the first and second flushes, respectively. Whereas it was found that the lowest protein percent was 18.56% when using wheat straw (W3) and wheat seeds spawn in the fourth flush. The reason of decreasing in the protein content of the produced fruiting bodies resulted in a substrate compared to another one (Owaid et al. 2017a) because of difference of substrate components made some of them more suitable for fungal growth due to the availability of a wet content and nutritional needs which reflected on the mushroom ability in utilizing them to speed up protein composition in cells of *Pleurotus* spp. (ranged 14.06-25.50)% cultured in different agricultural substrates (Dundar et al. 2009). This study agreed with some studies which recorded 23.30-32.21% protein content in fruiting bodies when growing 4 species of *Pleurotus* in enriched wheat straw using date-palm fibers and phosphate rocks (Owaid et al. 2017a). Generally, *Pleurotus* spp. is capable to myco-degrade lignocelluloses to protein-rich food (Owaid et al. 2018).

Effect of treatments on substrate weight loss and the biological efficiency

Table 9 exhibited the highest loss in substrate weight was 44.3% for W1 substrate whereas W2 and W3 substrates recorded lower weight loss reached 38.13% and 34.2%, respectively. It was also found that using the two spawns of barley and white corn seeds got the highest loss in substrate weights after the production 39.33% and 40.4%, respectively. The interaction between substrate and spawn type exhibited that the highest rate in weight loss was 46.3% by W1 substrate with barley seeds spawn, whereas the lowest one was 33.5% by the control (W3) substrate with wheat seeds spawn. These results agree with results of the total production (Table 6) and fruiting bodies number (Table 7), which referred that W1 substrate showed the best growth that led to exhaust nutritional compositions of this substrate and decrease its weight. The biological efficiency links with the total yield in a positive correlation (Owaid et al. 2015b), thus it depends on the size productivity of oyster mushroom especially in the case of using supplements (Tesfay et al. 2020).

Table 8 Effect of substrate and spawn type in protein content (%) in fruiting bodies

Treatments		No. of flushes (F)				Average W*S	Average W
Substrate type (W)	Spawn type (S)	F1	F2	F3	F4		
W1	S1	24.14	24.00	22.63	21.61	23.10	22.54
	S2	23.96	23.10	22.40	20.51	22.39	
	S3	23.65	22.81	21.86	20.22	22.13	
W2	S1	22.89	22.10	21.20	18.60	21.21	21.19
	S2	23.14	22.81	21.10	19.15	21.55	
	S3	22.63	21.71	20.03	18.94	20.83	
W3 (Control)	S1	22.65	21.60	20.40	18.60	20.81	20.64
	S2	21.95	21.20	20.50	19.31	20.74	
	S3	21.63	21.31	20.00	18.56	20.37	
Average of F		22.79	22.20	21.11	19.61	-	-
Average of S*F		F1	F2	F3	F4	Average of S	-
S1		23.41	22.96	22.26	20.78	22.35	-
S2		22.88	22.21	20.77	19.24	21.28	-
S3		22.07	21.36	20.30	18.82	20.64	-
LSD ($p < 0.05$)		W=0.65, S=0.73, F=0.86, W*S=0.95, W*F=1.12, S*F=v, W*S*F=1.53					

Note: W1 = 10% *Atriplex tartaric* straw-supplemented wheat straw substrate, W2 = 10% *Caroxylon cyclophyllum* straw-supplemented wheat straw substrate, W3 = 2% urea-supplemented wheat straw substrate (control), S1 = spawn of barley seeds, S2 = spawn of white corn seeds, S3 = spawn of wheat seeds, F1 = 1st flush, F2 = 2nd flush, F3 = 3rd flush, F4 = 4th flush

Table 9 Effect of the applied substrate and spawn type on the weight loss of substrate (%)

Treatments Spawn type (S)	Substrate type (W)			Average of W
	W1	W2	W3 (control)	
S1	2145.60	1445.3	876.7	1489.20
S2	2123.03	1199.5	750.4	1357.64
S3	1494.1	1065.0	657.3	1072.13
Average of S	1920.68	1131.7	761.5	-
LSD ($p < 0.05$)	W = 3.61, S = 3.74, W*S = 4.54			

Note: W1 = 10% *Atriplex tartaric* straw-supplemented wheat straw substrate, W2 = 10% *Caroxylon cyclophyllum* straw-supplemented wheat straw substrate, W3 = 2% urea-supplemented wheat straw substrate (control), S1 = spawn of barley seeds, S2 = spawn of white corn seeds, S3 = spawn of wheat seeds

Finally, Table 10 showed that the highest rate of the biological efficiency was 40.01% using W1 substrate, while W2 and W3 substrates recoded 36.51% and 33.33%, respectively. No significant differences in the biological efficiency were seen when using the spawns of barley seeds, white corn seeds and wheat seeds. The interaction between substrate and spawn type showed the highest rate of biological efficiency 41.3% and 40.9% with W1 substrate with spawn of white corn seeds and barley seeds, respectively, whereas the lowest rate was 31.6% when using W3 substrate with wheat seeds spawn. The quality of spawn is very important for oyster mushroom cultivation (Sagir & Yildiz 2004, Liu et al. 2018). These results agree with finding by Al-Qarawi et al. (2013) who recorded biological efficiency 20–30% and Owaid et al. (2018) who recorded about 37–45% in the mixture substrates.

Table 10 Effect of the substrate and spawn type on the biological efficiency (%) of mushroom

Treatments Spawn type (S)	Substrate type (W)			Average of W
	W1	W2	W3 (control)	
S1	40.9	36.4	34.2	36.13
S2	41.3	37.8	34.3	37.13

Table 10 Continued.

Treatments Spawn type (S)	Substrate type (W)			Average of W
	W1	W2	W3 (control)	
S3	38.1	35.4	31.6	36.33
Average of S	40.01	36.51	33.33	-
LSD ($p < 0.05$)	W = 2.12, S = NS, W*S = 3.12			

Note: W1 = 10% *Atriplex tartaric* straw-supplemented wheat straw substrate, W2 = 10% *Caroxylon cyclophyllum* straw-supplemented wheat straw substrate, W3 = 2% urea-supplemented wheat straw substrate (control), S1 = spawn of barley seeds, S2 = spawn of white corn seeds, S3 = spawn of wheat seeds, NS = Non-significant

Conclusion

In conclusion, these results encourage the supplementation of agricultural substrates using desert weeds especially *A. tatarica* and apply in desert environments at the commercial level. Results of this study showed that the produced spawn from the white corn exhibited the best results in the growth and yield of *P. ostreatus* compared to the spawn produced from wheat straw and barley. Also the *A. tatarica* straw-supplemented substrate showed a higher producing rate in fruiting bodies number, protein content, yield and biological efficiency. *A. tatarica* straw-supplemented substrate exhibited the shorter mycelial growth completion period (28.26 days). This substrate exhibited highest yield, and biological efficiency were 1489.2 g.kg⁻¹, and 40.01%, respectively. Also, results showed the best protein percent 24.14% and 24.00% when using this substrate with barley seeds spawn during the first and second flushes, respectively.

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