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Effect of Different Level of β -glucan extracted from Baker's Yeast *Saccharomyces cerevisiae* and Barley Bran in the Physicochemical Properties of Fish Patties at Cooling Storage Periods

Shaymaa A.J. Al-Jumaiee¹*, Khadeeja S.J. Al-Hussainy² & Alaa J.A. Al-Manhel²*

¹ Department of Marine Vertebrate, Marine Science Centre, University of Basrah, Iraq

² Department of Food Science, College of Agriculture, University of Basrah, Iraq

*Corresponding author e-mail: orchid.flowr@yahoo.com

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Abstract: β -glucan was extracted from Baker's Yeast of and barley bran using the conventional classical method and the hot water method respectively. The yield of β -glucan of Baker's Yeast and barley bran were (5.95 and 5.18) % respectively. with significant differences at the probability at the level of 0.05. β -glucan which extracted from two sources added were to fish patties at different levels (0.0, 0.1, 0.3, 0.5, and 1) g / 25g, and the patties were stored at 4±2 °C up to for 14 days. The results showed that the values of pH and water holding capacity of patties treated with β -glucan of yeast were lowest than those of patties treated with β -glucan of the patties treated with β -glucan of the patties treated with β -glucan of the patties treated with β -glucan extracted from barley bran.

Keywords: β -Glucan, Fish patties, pH, Water holding capacity, Acidity value, Peroxide value.

Introduction

Dietary fibers (DFs) are a part of edible foods that are indigestible by the human digestive system and that reach the colon area and improve the balance of natural flora in the body as well as its role in changing the nature of absorption of nutrients, in away helps to push food inside the digestive system with slow absorption of sugars. That's will be reflected positively on the stability of blood sugar for as long as possible as well as feeling and feeding full for a long time and thus maintaining weight (Bangari, 2011). β -glucan is a homopolysaccharide, composed of a linear chain (or a branch) of glucose units linked by the glycosidic bond between carbon and a hydroxyl group (Alves da Cunha *et al.*, 2017), β -glucan is proven to be indigestible and naturally found in the cell walls of cereals (such as barley, oats, and maize and wheat), fungi, algae and bacteria. The subunit of β glucan is glucose, and the most common source of β -glucan is fungi, especially yeast which is And its Structural unit was the glucose sugar and the most common source to extract from the wall of fungal cells because they are bio-effective in strengthening the immune system and act as inhibitor of cancer cells (Zhu *et al.*, 2016). There is a clear contrast between different types of the β glucan extracted from various sources. This is due to the type of the glycoside bond that binds the glucose units to each other, β (1-3) β (1-4) or β (1-6) or it may contain two types of bonds called mixed-linkage (Sofi *et al.*, 2017).

Zhang *et al.* (2019) noted that the addition of β -glucan at different levels have improved fish product surimi hardness and consistency with reduced viscosity, increased protein concentration and increased gel formation as well as reduced fish smell and taste delicious. The purpose of the present work was to extract β -glucan from cheap sources like bread yeast and barley bran.

Materials and Methods

Collection of samples

Samples of dried bakery yeast of *Saccharomyces cerevisiae* (Saf-instant-Turkish) and barley (Iraqi variety) were collected from the local markets of Basrah city. A process of grinding the barley was done to obtain the bran by using a grain mill then sieved and storage at 4°C.

Extraction of the β-glucan.

The classical method of the β -glucan was extracted from bread yeast according to was followed by the method of Asare (2015) by using 4 g of dry yeast and 150 ml of NaOH at 0.1 N. The mixture was mixed by magnetic stirrer at 100 °C for 15 minutes The mixture was left to cool then after centrifuge centrally at 5000 g for 5 minutes. The supernatant was removed and were then resuspended in another 50 ml of distilled water and the mixture then centrifuged under the same conditions. The pellets were washed with distilled water and the pH was adjusted to 6.9 - 7.1 using 0.1 M HCl or 0.1 M NaOH. Acid digestion was done by H3PO4 with 0.1 M to the precipitate and the mixture has to be was heated to the boiling point for 15 minutes. After cooling the mixture was centrifugation at 5000 g for 5 minutes subsequently, 50 ml of the ethanol was added to the pellets and heated at 80 °C for 15 minutes and centrifuged at 7000 g for 5 minutes.

Pellets were resuspended three time with 50 ml distilled water using centrifuge to sediment the pellets to separate the pellets. The last precipitate was resuspended again with distilled water adjusted at PH 6.9-7 using 0.1 M of HCL or 0.1 M NaOH, and the centrifugation process was carried out again. The precipitate of β -glucan of bread yeast was dried in an air oven at a temperature of 55-60 °C for 24.

The barley bran β -glucan was obtained according to the method of hot water extraction applied by Ahmad et al. (2009). One hundred grams of barley bran were soaked in 300 ml of ethyl alcohol (80%) for 6 hours. Water was added to this mixture in the ratio of 1: 10 (w / v) and mixed by 55° C for 90 minutes and then centrifuged at 5000 rpm for 20 minutes at 40 °C. The pH was adjusted to 8.5 with sodium bicarbonate (Na₂CO₃) (20%) and the mixture was stirred by magnetic stirrer for 30 minutes at a temperature of 55 °C. Then, the mixture was centrifuged at a speed of 5000 g at 40 °C for 20 minutes and the precipitation was discarded and taking supernatant was adjusted to pH 4 using citric acid (2 M), followed by a centrifugation and offered to centrifugal at 5000 g for 25 minutes. The supernatant was suspended with ethyl alcohol (80%) in a ratio of 1:1 for 20 minutes, and the mixture was centrifuged again at 4000 g at 4°C for 25 minutes. B-glucan was transferred into petri dish and dried in an air oven at 55-60 °C for 24 hours the percentage of crude β -glucan yield (%) was calculated depending on the equation of Du et al. (2014) as follows:

 β -glucan yield (%) = Weight of crude β glucan (g) / weight of sample (g) ×100

Preparation of fish patties with β-glucan

Four kilograms of common carp fish (*Cyprinus carpio*) of Basrah markets and cleaned after removing the head and internal intestines. The skin and bones were isolated from muscle tissues. Fish meat was mixed

very well, and minced using electric grinder. The amount of fish meat was divided for in to five portions and β -glucan was added in quatity of (0, 0.1, 0.3, 0.5 and 1) g for each 25 g of the product. The patties were then formed by 25 g of each sample. Patties samples were placed in polyethylene bags and vacuum seale and stored at of 4 ± 2 °C for 14 days.

Physicochemical properties

1-pH, Water holding capacity and Acid degree value

pH, water holding capacity and acidity value of fish patties treated by β -glucan were tested during storage for (0, 3, 7, 10 and 14) days according to the method described by Al-Taii & Al-Mossawi (1992). The pH meter was used to measure estimate the pH. Water holding capacity was calculated according to the following equation:

WHC (ml) =total water content (ml) - amount of water included in the cylinder (ml) The pH value was calculated according to the following equation:

Acid degree value = number of milliliters of base \times 5.61 / sample weight (g)

Free fatty acids were calculated on the basis of Oleic acid according to the following equation

Free fatty acid ratio (FFA) = Acidity value / 2

2-Peroxide value

The Peroxide value of fish were, according to the method of Egan *et al.* (1981) and calculated as follow:

Peroxide value = sodium thiosulfate (ml) \times N \times 1000 \setminus Sample weight (g).

Statistical Analysis and Design

Complete Randomized Design (CRD) was used for two and three-factor, analyzed by the Stat Release 10.3 DE. Previously studied factors were tested by using the least significant difference (L.S.D.). at a probability level of ($p \le 0.05$) (Al-Rawi & Khalafallah, 2000).

Results and Discussion

Extraction of the β -glucan

The highest yield of β -glucan extracted from Baker's Yeast was 5.95 %, while it was for the β -glucan extracted from barley bran 5.18 % which was within the limits (2.9-10.3) % found by Asare (2015). However the yield of β -glucan extracted from bread yeast using the classical method with different concentrations of acid and base was within the limits (5.6-11.9) %. that found by Maheshwari *et al.* (2017). The yield of β -glucan with extacted from yeast bread was higher significantly (p≤0.05) as compared with that extracted from barely bran

Effect of adding different levels of β-glucan on the physiochemical properties of fish patties stored for 4 °C for different periods of time

Processed fish was produced without or with addition of β -glucan in aratio of (control sample, 0.1, 0.3, 0.5, 1) g for every 25 g of fish meat.

1-Effect of adding of different quantities of β-glucan extracted from yeast and barley bran on the pH of fish patties stored at 4°C for different periods of time

Table (1) showed that the PH of fish patties was decreased proportionally with increasing the added percentage of β -glucan extracted from bread yeast and barley bran. The pH value of control sample and other treated patties was increased during cold storage. This release of nitrogen bases due to the protein degradation enzymatically., The pH value of fresh fish ranged from 6.2 to 6.8 (Al-Taii, 1987). It was observed that β -glucan extracted from barley bran was more effective in reducing the rate of increasing of pH values than with β-glucan extracted from yeast. Moreover, the variations pH values of were depending on the source and quantity of added β -glucan to the stored fish patties. This was due to the presence of the TMAO compound found in the fish, which acts as an buffer solution that resists the change in pH. It is either degraded enzymatically to DMA and formaldehyde, by bacteria into TMA. Both DMA and TMA are nitrogen bases which increased the pH value of patties . Nitrogen bases are also produced as result of protein degradation due to the bacterial activity that produced alkaline compound. On the other hand, the liberation of lipolytic enzymes, and the bacterial activity act of bacteria decompose produce fatty acids that play an opposite role leading to the reduction of pH (Deli, 1980), The results of the present study are in a good agreement with the findings of Al-Shawki (2018) regarding in the reduction of pH in patties with different added concentrations of β -glucan extracted from yeast.

Table (1): pH values of fish patties treated with different quantities of β -glucan extracted from yeast and barley bran stored at 4°C for 14 days.

Periods of cold storage (day)	Control	β-glucan ratios extracted from yeast bread (g)				β-glucan ratios extracted from barley bran(g)				
	_	0.1	0.3	0.5	1	0.1	0.3	0.5	1	
0	6.50	6.45	6.40	6.38	5.30	6.40	6.35	6.27	5.26	
3	6.52	6.48	6.45	6.40	5.36	6.46	6.38	6.30	5.28	
7	6.61	6.56	6.50	6.47	5.66	6.53	6.45	6.33	5.43	
10	6.70	6.65	6.59	6.53	5.75	6.67	6.54	6.41	5.50	
14	6.85	6.79	6.64	6.58	5.90	6.75	6.60	6.55	5.66	

2- Effect of adding different quantities of βglucan extracted from yeast and barley bran on water holding capacity of fish patties stored at 4 °C for different periods of time

Table (2) showed the values of water holding capacity (ml) in of fish patties samples cold stored for different periods of times. The results indicated an increase in water holding capacity, with increasing of the added β -glucan quantity from both sources and the storage periods at the probability level ((p ≤ 0.05). The water holding capacity values in the control sample increased from 14.75 to 16.50 during the storage period. This may be due to the increase in the pH which is associated with water retention.

The same pattern was observed in samples of fish patties treated with β -glucan extracted from the bread yeast and barley bran. An increase in water holding capacity with the added the β -glucan was observed added as it was (14.95) ml and (15.20) ml when the addition of 0.1 g and then increased to (15.20) and (15.35) ml when 0.3 g of β -glucan was added. These values increased when using 0.5 g and reached 15.55 ml and 15.85 ml. When using 1 g, WHC values were (15.75) ml and (16.70) ml respectivily.

The results showed a very clear increase in water holding capacity values when patties contained β-glucan from both sources stored for three days. When 0.1 g, was added WHC were increased from 16.00 and 16.10 ml to 16.20 and 16.40 ml when using 0.3 g respectivly. This increase was significant when the addition was $0.5 \text{ g} \beta$ -glucan. The water holding capacity remaining un changedbilized at an approximate value of 16.45 ml when using 1 g of β -glucan extracted from the yeast of bread while the water holding capacity value reached 17.10 ml when using the same percentage of β glucan extracted from barley bran. After seven days of storage, the water holding capacity values of treated fish patties of 0.1 g of β -glucan for both sources were 16.50 and 16.60 ml and increased significantly when using 0.3 g to 16.66 and 16.86 ml respectively. Additional of 0.5 g β -glucan resulted an increase up to 16.75 and 17.58 ml respectively. However, water holding capacity continued to rise to 16.90 and 17.85 ml when 1 g of β -glucan extracted from yeast and barley bran was added. The present study showed continuous increase in the values of water holding capacity with increasing the period of cold storage up to 14 days in samples treated with β -glucan extracted from the yeast of bread and barely bran.

It was found that there is a strong relationship correlation between water holding capacity and. This effect was reflected significantly in water holding capacity. Al-Taii (1987) pointed out that increased pH led to increase water holding capacity in muscles. It was observed that β -glucan extracted from barley bran was more effective in raising water-carrying values than with β -glucan extracted from yeast.

Kristinsson & Hultin (2003) noted that the increase in pH from 6.4 to 7.4 led to an increase water holding capacity in fish

proteins and otherwise when pH decreases. It has been observed that the added β -glucan from both source for patties has an effective role in increasing the water holding capacity of these patties. This is due to glucose it which represent as part of its structure and to the presence of water-hydrophilic hydroxyl groups. When β -glucan binds with muscle proteins of fish meat, hydrophilic proteins are more likely to increase water holding capacity.

A review of literature has shown that water holding capacity is at its maximum value when the pH is higher or lower than the Iso electric point (IP), which is 5.6 (Huff-Lonergan & Lonergan, 2005).

The statistical analysis results showed no significant differences at the probability level $(p \le 0.05)$ for the effect of the storage of cooled and the effect of the ratio of the addition of the β -glucan and its source as well as the binary interaction between the storage and the ratio of addition and the source of the β -glucan.

Table (2): Water Holding Capacity values (ml) of fish patties treated with different quantities
of β-glucan extracted from yeast and barley bran stored at $4^{\circ}C$ for 14 days.

Periods of storage (day)	Control _	β-gluca	n ratios (yeast br	extracted ead (g)	l from	β-glucan ratios extracted from barley bran(g)				
		0.1	0.3	0.5	1	0.1	0.3	0.5	1	
0	14.75	14.95	15.20	15.55	15.75	15.20	15.35	15.85	16.70	
3	15.00	16.00	16.20	16.40	16.45	16.10	16.40	16.75	17.10	
7	15.40	16.50	16.60	16.75	16.90	16.60	16.86	17.58	17.85	
10	16.00	17.15	16.80	16.90	18.85	17.25	17.40	17.75	18.00	
14	16.50	17.35	17.50	18.13	18.25	18.05	18.29	18.40	18.50	

3- Effect of adding different quantities of βglucan extracted from yeast of bread and barley bran on the peroxide value of fish patties stored at 4 °C for different periods of time

The results in table (3) showed the peroxide values (mEq / kg) in samples of fish at stored for different periods of times. The statistical analysis results showed that peroxide values varied with different quantities of added β -glucan for both of its source, and varied also with different storage periods of storage.

It was noted that the peroxide value of the control sample and fish patties showed clear increases due to cooling period of 14 days which indicated for the decomposition oxidation of fish fat, especially in control sample which is free β -glucan. However, there was a significant decrease in the peroxide values in samples contained β -glucan at different percentages in fish patties.

These results were in agreement with the findings of Al-Shawki (2018) peroxide values of sample were (0.71, 1.87, 2.02, 2.85 and 3.18) mEq /kg of fish initially and after different periods of storage 0, 3, 7, 10, for 14 days respectively.

The results of the statistical analysis showed significant differences at the (p ≤ 0.05) for the effect of the source of the β -glucan and the effect of the storage on the values of peroxide on the fish samples treated with different percentages of β -glucan. This is an indication that β -glucan extracted from yeast and barely bran have an antioxidant effect as it works to remove free radicals and capture the resulting hydrogen peroxide during the degradation of unsaturated fatty acids. This has been proven by some another studies on the possibility of using β -glucan as an antioxidant (Kayali *et al.*, 2005; Jaehrig *et al.*, 2007).

Periods of C storage (day)	Control	β-gluo	can ratios yeast b	extracted read (g)	d from	β-glucan ratios extracted from barley bran(g)				
	-	0.1	0.3	0.5	1	0.1	0.3	0.5	1	
0	0.77	0.75	0.72	0.69	0.67	0.74	0.71	0.68	0.64	
3	1.78	1.62	1.56	1.51	1.41	1.49	1.39	1.35	1.33	
7	2.02	1.97	1.93	1.91	1.86	1.90	1.87	1.83	1.79	
10	2.85	2.55	2.34	2.19	2.13	2.27	2.21	2.09	2.03	
14	3.18	2.87	2.78	2.73	2.66	2.79	2.75	2.62	2.53	

Table (3): Peroxide values (mEq / kg) of fish patties treated with different quantities of β -glucan extracted from yeast and barley bran stored at 4°C for 14 days.

4- Effect of adding different quantities of β glucan extracted from yeast and bran on the value of acidity and free fatty acid content of fish patties stored at 4 °C at different periods of time

The results in table (4) and table (5) focused on the acidity and free fatty acid content in fish samples treated with different quantities of β -glucan, which showed gradual increases in their values (1.23, 1.84, 2.18, 2.69 and 3.25) mg KOH/g (0.61, 0.92, 1.09, 1.34, and 1.62)% respectively in the control sample and during the 14 day storage period, samples with added with β -glucan extracted from the Baker's Yeast and barley bran. gave significant variations in acidity and FFA (3.01, 2.80, 2.69 and 2.58) mg KOH/g and (2.91, 2.69, 2.58 and 2.50) mg KOH/g as well as (1.50, 1.40, 1.34 and 1.29) % and (1.45, 1.34, 1.29 and 1.25)% respectively after 14

days of cold storage Increases in FFA is an important test to indicate hydrolysis of fish by lipase and phosphlipase. This study indicated that acidity values depression with the addition of higher quantities of beta-glucan for the fish patties could reduce fat hydrolysis. It has an active role in controlling the activities of psychrophilic bacteria and reducing its secretion of lipid analysis enzymes that produce free fatty acids. These results are consistent with Ozcan & Ertan (2018) who studied the effect of antioxidant and antimicrobial β -glucan extracted from some types of mushrooms. Also it was found that proportional of increases were found with increasing added β -glucan concentration.

Table (4): Acidity values (mg KOH/g) of fish patties treated with different quantities of β -glucan extracted from yeast and barley bran stored at 4C for 14 days.

Periods of storage (day)	Control	β-glu	can ratio yeast l	s extracte oread (g)	β-glucan ratios extracted from barley bran(g)				
		0.1	0.3	0.5	1	0.1	0.3	0.5	1
0	1.23	1.12	1.10	0.94	0.75	1.12	1.06	0.80	0.70
3	1.84	1.73	1.58	1.54	1.44	1.62	1.54	1.44	1.34
7	2.18	2.07	1.79	1.59	1.51	1.82	1.61	1.48	1.43
10	2.69	2.63	2.01	1.90	1.68	2.55	1.87	1.79	1.58
14	3.25	3.01	2.80	2.69	2.58	2.91	2.75	2.58	2.50

Table (5): Values of free fatty acids (F.F.A.) % of fish patties treated with different quantities of β-glucan extracted from yeast and barley bran stored at 4°C for up to 14 days.

Periods of storage (day)	Control	β-gluc	β-glucan ratios extracted from barley bran(g)						
		0.1	0.3	0.5	1	0.1	0.3	0.5	1
0	0.61	0.56	0.55	0.47	0.37	0.56	0.53	0.40	0.35
3	0.92	086	0.79	0.77	0.72	0.81	0.77	0.72	0.67
7	1.09	1.03	0.89	0.79	0.75	0.91	0.80	0.74	0.71
10	1.34	1.31	1.00	0.95	0.84	1.27	0.93	0.89	0.79
14	1.62	1.50	1.40	1.34	1.29	1.45	1.34	1.29	1.25

Conclusions

Extraction and source methods and β -glucan source play an important role in determining the amount of extracted β -glucan. The ratio of β -glucan content of yeast was higher than that of barley bran, β -glucan has the ability to improve the specific qualities of some products of fish patties, and prolonging the life of the fish patties. The results showed that the ratio of free fatty acids and values peroxide, pH, water holding and pH values were depending on the quantities of added β glucan in the preparation of fish patties.

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