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EFFECT OF SOME PLANT EXTRACTS ON THE QUALITY OF SALTED FISH

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ABSTRACT

The main purpose of this work was to investigate the effect of some plant extracts such as cumin (*Carum carvi*), thyme (*Thymus vulgaris*) and licorice root (*Glycyrrhiza glabra*) at level 0.05% on the quality of salted Tubara (*Mugil capito*) fillets. Five batches i.e. control, cumin, thyme, licorice root and their combined were made. Each bag contained 200 g fillets and 100 ml filling media (20 % salt, 0.02% acetic acid and 0.05% plant extracts) were added. All batches were ripened under room temperature (16 ± 2°C) for 20 days and then stored in refrigerator at 4± 1°C for 8 weeks. Results showed that, both moisture and ash were clearly increased while both protein and lipid were decreased in all treatments at 0 time of salting. During ripening and cold storage periods, moisture content, pH value, total volatile bases nitrogen (TVB-N) and trimethylamine (TMA-N) content were increased while, thiobarbituric acid values were decreased. On the other hand, total bacterial count decreased whereas halophilic bacteria increased. The used plant extracts improved the quality properties comparing with control sample. It could be concluded that, plant extracts were more effective in controlling the biochemical changes and improved sensory attributes of salted fish fillets. Besides, safe salted product can be obtained.
Keywords: Fish – Salting – Plant extract – Quality criteria – Refrigerated storage.

INTRODUCTION

Spices and their essential oils are the most efficient natural antioxidant and antimicrobial agents have long been used to preserve food. The efficacy of these compounds can be enhanced, by combining their use with other preservatives (Harpaz *et al.*, 2003; Bagamboula *et al.*, 2004 and Burt, 2004). In addition, spice and its extracts are added to food primarily a flavoring agent. The functional properties (i.e., major flavor and aroma compounds and antimicrobial factors) of spice residue in its essential oil. Although spice and its extracts are antimicrobial effects, it must be noted that spices must be considered a potential source of high levels of microorganisms (Pivinick, 1980 and Pafumi, 1986). Moreover, many herbs and spices, usually used to flavor dishes, are an excellent source of phenolic compounds which have been reported to show good antioxidant activity. However, herbs and spices usually contain essential oil which antioxidant activity but also carry flavor (Rice-Evans *et al.*, 1996; Ruberto *et al.*, 2000; Teissedre and Waterhouse, 2000; Zheng and Wang, 2001). Thus, extracts are prepared by hydrodistillation to remove the intrinsic flavor from the plant material. Furthermore, use of an aqueous solvent may prevent solubility problems and this avoids harmful residues from organic solvents. The hydrodistilled extracts may also have use in the functionalization of foods and beverages as phenolic compounds have been ascribed health-promoting properties

especially synthetic antioxidants suffer from several drawbacks(Harborne and Williams, 2000 and Hinneburg *et al.*,2006). On the other hand, salting is one of the oldest techniques known not only to preserve the fish but also to improve its aroma and flavor. It is essentially intended to prolong the shelf-life of the product by reducing its water activity. Brines with low concentration of salt are known to promote better yield and water holding capacity than saturated brines (≥ 25 %) (Barate *et al.*, 2002).Therefore, the main purpose of our work was to investigate the effect of some spices extracts (0.05%) such as cumin *Carum carvi*, thyme *Thymus vulgaris,* licorice root *Glycyrrhiza glabra* and their combined on the quality attributes of salted Tubara (*Mugil capito*) fillets stored at refrigerated conditions.

MATERIALS AND METHODS

Materials

Fish: *Mugil capito* was obtained from El-Serw, National Institute of Oceanography and Fisheries (NIOF) during 2005. Fish were carefully washed with tap water. Heads, viscera, skin and fins were removed.

Salt: edible salt (produced by El-Nassr Co.) was obtained from a local market.

Dried plants: Cumin *(Carum carvi*), thyme *(Thymus vulgaris)* and Licorice root *(Glycyrrhiza glabra)* were purchased from a local market. Extracts of investigated plants were obtained as described by Hinneburg *et al.*,(2006) with some modifications. Fish fillets (200 g) were packed in polyethylene bags and 100 ml filling media (brine 20% salt, 0.02% acetic acid and 0.05% plant extracts) were added. Partially under vacuum for each bag was done.

Analytical methods:

The following analyses were carried out. Moisture (oven-drying at 105^{o}C to constant weight was obtained), ash (an electric furnace), ether extract (Soxhlet apparatus) and nitrogen content (Micro-Kjeldahl) x 6.25 to calculate protein content were determined using standard methods (AOAC,1995). Salt content was determined as described by FAO (1981). Total volatile bases nitrogen (TVB-N),Thiobarbituric acid (TBA), pH value and salt content were determined as described by Pearson, (1976).Trimethylamine nitrogen (TMA-N) was determined as mentioned by AOAC (1995). Microbial tests: 10g of sample were suspended in 90 ml sterile saline. Decimal dilutions were plotted to determine the following: total bacterial count yeast and moulds (FAO, 1979) and Halophilic bacteria (Del-Valle,1976). Sensory evaluation: investigated products were evaluated using a panel test of a point hedonic scale according to Fey and Regenstein (1982). All data were expressed as mean values (n = 3).

RESULTS AND DISCUSSION

Proximate analysis:

The Proximate analysis of raw and plant extract- fish fillets are shown in table (1). Raw fish flesh was composed 70.33% moisture, 18.34% protein,

9.66% lipid, 1.41% ash and 0.36% sod.chloride content (on weight wet basis). Similar results are reported by Yasin (1997). These parameters changed after fish fillets were soaked in brine (20 %) at 16 °C. In addition, both moisture and ash were clearly increased while both protein and lipid were decreased in all treatments. Similar trends were reported by El-Sharnouby (1993); Ahmed *et al.*, (1994); Ibrahim (1994); Awad (1999); El-Dessouky (1999).

Table (1): Proximate analysis (on wet weight basis) of raw and plant extract-fish fillets.

Constituent (%)	Raw	Control	Plant extract- salted fish fillets;			
			Cumin	Thyme	Licorice	Combined
Moisture	70.33	73.12	71.58	71.59	72.29	72.35
Protein	18.34	12.14	13.25	13.06	12.22	12.22
Lipid	9.66	6.08	6.11	6.12	6.27	6.01
Ash	1.41	8.59	8.95	9.08	9.10	9.20
Sod. chloride	0.36	8.12	8.87	8.92	8.29	8.77

All data are mean values (n = 3).

Effect of both ripening period and cold storage on:
Moisture

Results in table (2) showed the effect of both ripening period and cold storage on moisture. A little increase in moisture in all treatments was observed after 20 days of salting under room temp.((16 ± 2 °C). The same trend was noticed during cold storage (4 ± 1°C) up to 30 days, but it was slightly decreased till 45 days and increased at the end of storage period (60 days). This fluctuation occurred in moisture throughout storage time may be due to increase of salt content which consequently increase to osmotic pressure and then gained water content. These findings are in agreement with reported by Shlaby (1990); El- Sharnouby (1993); Ibrahim (1994); Awad (1999); El- Dessouky (1999); Kassem *et al.*, (2002) and Sarhan (2004).

Table(2): Effect of both ripening period and cold storage on moisture of plant extract-salted fish fillets .

Storage Period (day)	Control	Plant extract-Salted fish fillets;			
		Cumin	Thyme	Licorice	Combined
	Ripening period at room temp. (16 ± 2 °C)				
0	73.12	71.58	71.59	72.29	72.35
20	75.96	71.73	71.6 4	73.11	72.39
	Cold storage at (4 ± 1°C)				
15	76.16	72.39	71.75	73.43	72.55
30	76.60	72.66	73.63	74.01	73.19
45	76.01	71.88	71.82	73.58	73.01
60	77.67	74.22	73.88	74.93	74.17

All data are mean values (n = 3).

Quality criteria

pH value

The effect of both ripening period and cold storage on pH value of plant extract- salted fish fillets are illustrated in table (3). It could be observed that pH value in control sample was 6.26,slightly decreased in all fish fillets treated with plant extracts at zero time of salting. After ripening time (20 days) at room temp., the values of pH were slightly increased in all batches and the same trend was noticed in samples throughout different periods of cold storage. However, plant extracts used caused a little inhibition in pH value when compared with control sample. This increase in pH value may be due to the action of protease and lipase enzymes in the tissues to form pyridine bases. This data is in accordance with found by Rashad (1986); Ibrahim (1994);Kassem et al., (2002) and Sarhan (2004).

Table(3): Effect of both ripening period and cold storage on pH value of plant extract-salted fish fillets.

Storage Period (day)	Control	Plant extract-Salted fish fillets;			
		Cumin	Thyme	Licorice	Combined
	Ripening period at room temp.(16 ± 2 °C)				
0	6.26	5.81	5.72	5.85	5.78
20	6.78	5.93	5.91	5.99	5.92
	Cold storage at (4 ± 1°C)				
15	7.09	6.04	6.25	6.77	6.06
30	7.35	6.63	6.88	7.01	6.07
45	7.51	7.10	7.10	7.49	6.83
60	7.84	7.83	7.44	7.55	7.53

All data are mean values (n = 3).

Total volatile bases nitrogen (TVB-N):

The effect of both ripening period and cold storage on TVB-N content (on wet wt.) of plant extract-salted fish fillets are shown in table(4): The value of TVB-N in control sample was the highest (22.40 mg/100g flesh) than other treatments at 0 day of wet salting. In addition, it was clearly increased especially in control sample (37.80 mg /100 sample) after ripening period. Thyme extract and combined treatments were similar in TVB-N content (22.40 mg/100 sample) whereas licorice treatment was 29.40 mg /100g sample. On the other hand, its value was gradually increased in all treatments after cold storage for 15 days. Furthermore, the same trend was observed till the end of cold storage. This gradual increase may be due to protein decomposition as affected by microbial activity and protolytic enzymes. However, it could be observed that plant extracts controlled in rat of protein breakdown compared with control salted sample. This data is in agreement with reported by Abo-Raya (1975);Khallaf (1986);Shalaby (1990);Ahmed *et al.*, (1994); Ibrahim (1994);Kassem (2002) and Sarhan (2004).

Table(4): Effect of both ripening period and cold storage on TVB-N content (on wet wt.) salted fish fillets. of plant extract-

Storage Period (day)	Control	Plant extract-Salted fish fillets;			
		Cumin	Thyme	Licorice	Combined
	Ripening period at room temp. (16 ± 2 °C)				
0	22.40	19.20	18.20	18.20	19.60
20	37.80	26.60	22.40	29.40	22.40
	Cold storage at (4 ± 1°C)				
15	50.40	28.00	26.60	32.20	26.60
30	57.20	33.60	28.00	36.40	28.00
45	67.40	35.00	32.20	42.00	30.80
60	74.20	63.00	50.40	58.80	49.00

All data are mean values (n = 3).

Trimethylamine-Nitrogen

The effect of both ripening period and cold storage on TMA-N content (on wet wt.)of plant extract- salted fish fillets are shown in table (5). It could be observed that the value of TMA-N was ranged 1.19 to 1.88 mg /100 g flesh at 0 day, increased sharply after ripening period 20 days at room temperature. This increase may be due to the major changes occurred in bases nitrogenous compounds by microbial activity throughout fermentation time. However, its value was decreased in the investigated treatments after two weeks at cold storage. This decrease is probably caused by both the decreasing of enzyme activity and depression of enzyme synthesis. Vice versa was observed up to 30 and 45 days of storage. At the end of cold storage, its value was clearly decreased in all treatments. These results are in harmony with found by Nozawa *et al.*, (1979); Ibrahim (1994) and Sarhan (2004).

Table(5): Effect of both ripening period and cold storage on TMA-N content (on wet wt.) Salted fish fillets. of plant extract-

Storage Period (day)	Control	Plant extract-Salted fish fillets;			
		Cumin	Thyme	Licorice	Combined
	Ripening period at room temp. (16 ± 2 °C)				
0	1.88	1.25	1.82	1.19	1.39
20	3.91	3.85	3.22	3.51	3.12
	Cold storage at (4 ± 1°C)				
15	3.2 9	2.97	2.76	2.71	3.04
30	4.17	4.01	3.84	3.79	3.78
45	5.25	5.06	5.09	5.11	5.01
60	3.19	3.09	3.05	3.12	2.07

All data are mean values (n = 3).

Thiobarbituric acid (TBA)

Table (6) shows the effect of both ripening period and cold storage on TBA value (on wet wt.) of plant extract–salted fish fillets. TBA value in control sample was the highest (12.44 mg Malonaldhyde /kg sample) than treatments contained different plant extracts at 0 day of salting. After ripening period under room conditions its value was slightly decreased in all treatments. Similar observation was found in samples stored at 4 ± 1^{0}C for 15 days and but increased gradually up to 45 days. This increment may be due to salt concentration encourage the growth of halophilic lypolytic groups of bacteria which hydrolyze fish lipids (Quaglia *et al.*,1989). At the end of cold storage its value was decreased in all treatments. This decrement may be due to loss of formed malonaldhyde because of its reaction with protein–decomposed derivatives to produce tertiary products (Reddy and Setty, 1996). Therefore, plant extracts are responsible for the antioxidant activity (Rice-Evans *et al.*, 1996).

Table(6):Effect of both ripening period and cold storage on TBA value (on wet wt.) of plant extract-salted fish fillets.

Storage Period (day)	Control	Plant extract-Salted fish fillets;			
		Cumin	Thyme	Licorice	Combined
	Ripening period at room temp.(16 ± 2 ^{o}C)				
0	12.44	11.46	11.54	11.99	11.88
20	11.69	11.25	11.45	11.22	10.96
	Cold storage at (4 ± 1^{o}C)				
15	9.92	9.13	9.17	8.46	9.01
30	10.84	10.32	9.24	9.71	10.69
45	10.90	10.61	9.71	9.98	10.84
60	9.55	8.90	8.59	8.98	8.35

All data are mean values (n = 3).

Microbiological profiles:

Total bacterial count (TBC)

Effect of both ripening period and cold storage on TBC (log cfu/g) of plant extract- salted fish fillets are shown in table (7). It could be cleared that TBC in control sample was the highest (4.44 log cfu /g flesh) than that in all plant extract- treatments at first day of salting. This phenomenon may be due to the effect of plant extracts used as antimicrobial agents(Pafumi, 1986). After ripening time (20 days at (16 ± 2 ^{o}C), TBC was depressed in all treatments, this inhibition may be due to lactic acid formed during fermentation process. On the other hand, the count of total bacteria was increased in all treatments up to15 days of cold storage. In addition, cumin–treatment was higher in TBC than those in thyme-treatment. A gradual decrease was observed in TBC with extending cold storage up to 45 days and then increased at the end of storage. This fluctuation may be due to physico-chemical changes as affected by storage period extended. These results are in agreement with those found by Rashad (1986); Ibrahim (1994); Kassem *et al.*, (2002) and Sarhan (2004).

Table(7): Effect of both repining period and cold storage on TBC (log cfu/g) of plant extract-salted fish fillets.

Storage Period (day)	Control	Plant extract-Salted fish fillets;			
		Cumin	Thyme	Licorice	Combined
	Ripening period at room temp. (16 ± 2 °C)				
0	4.44	4.05	4.05	4.14	3.94
20	4.04	3.89	3.71	3.58	3.91
	Cold storage at (4 ± 1°C)				
15	4.89	4.97	3.85	4.29	4.09
30	4.07	4.09	3.71	3.94	4.06
45	3.69	4.04	3.69	3.87	3.86
60	4.46	4.07	3.90	3.91	4.09

All data are mean log (cfu/g flesh).

Halophilic bacterial count (HBC)

Effect of both ripening period and cold storage on halophilic bacterial count (log cfu/g) of plant extract-salted fish fillets are shown in table (8). It could be observed that HBC in control sample was the highest count (4.17 log cfu /g) than other treatments at 0 day and however vice versa was noticed after ripening time (20 days) at room temperature. A reduction trend in HBC in all treatments which stored for 15 days at 4 ± 1°C was observed. After that, HBC was fluctuated till the end of cold storage. This fluctuation may be attributed to the initial of TBC and type of used plant extracts. Similar results are observed by Ibrahim (1994) and Sarhan (2004).

Table(8): Effect of both repining period and cold storage on halophilic bacterial salted fish fillets. count (log cfu/g) of plant extract-

Storage Period (day)	Control	Plant extract-Salted fish fillets;			
		Cumin	Thyme	Licorice	Combined
	Ripening period at room temp. (16 ± 2 °C)				
0	4.17	3.65	3.97	3.25	3.79
20	4.37	4.04	4.09	4.09	3.96
	Cold storage at (4 ± 1°C)				
15	4.16	3.79	3.60	3.84	3.53
30	4.61	4.19	4.00	4.09	3.86
45	4.79	4.76	4.09	4.27	4.09
60	4.56	4.46	4.04	4.17	4.04

All data are mean log (cfu/g).

Yeasts and moulds count

The effect of both ripening period and cold storage on yeasts and moulds count of plant extract-salted fish fillets are shown in table (9). It was found that their counts ranged from 2.00 to 2.48 log cfu /g in plant extract-treatments compared with 2.78 log cfu/g in control sample at 0 day of salting. In addition, thyme extract was more effective on growth of yeasts and moulds

than other extracts. However, yeasts and moulds were decreased in all samples after ripening period (20 days). This inhibition may be due to effect of lactic acid bacteria during fermentation and lactic acid formed. On the other hand, their counts were similar in samples treated with plant extracts while in control sample was a high count after 15 days storage at 4 ± 1°C. After that, their counts were slightly fluctuated in all treatments up to 60 days under the same conditions. These findings are similarly to those of Ibrahim (1994) and Sarahan (2004).

Table(9): Effect of both ripening period and cold storage on yeasts and molds count salted fish fillets. (log cfu/g) of plant extract-

Storage Period (day)	Control	Plant extract-Salted fish fillets;			
		Cumin	Thyme	Licorice	Combined
	Ripening period at room temp. (16 ± 2 °C)				
0	2.78	2.30	2.00	2.40	2.48
20	1.22	1.05	0.85	0.98	0.93
	Cold storage at (4 ± 1°C)				
15	3.70	3.00	3.00	3.00	3.00
30	3.48	2.00	3.00	2.00	2.18
45	3.86	3.30	3.48	3.00	3.48
60	3.48	3.00	2.48	3.00	3.00

All data are mean log (cfu)/g.

Sensory evaluation

Mean of sensory tests of salted fish fillets as affected by different plant extracts and cold storage are illustrated in fig. (1). Appearance, texture, odor, taste and overall acceptability are the most important quality attributes investigated in this study. The obtained data showed that cumin extract improved all tests studied followed by thyme, licorice and their combined respectively compared with control sample during different cold storage periods. However, thyme extract was more effective in controlling of protein and lipid decomposition and microbiological profiles comparing with other extracts. Furthermore, fishy odor was observed in control sample comparing with treatments contained plant extracts. Concern texture, it could be observed that combined treatment was a high tender compared others.

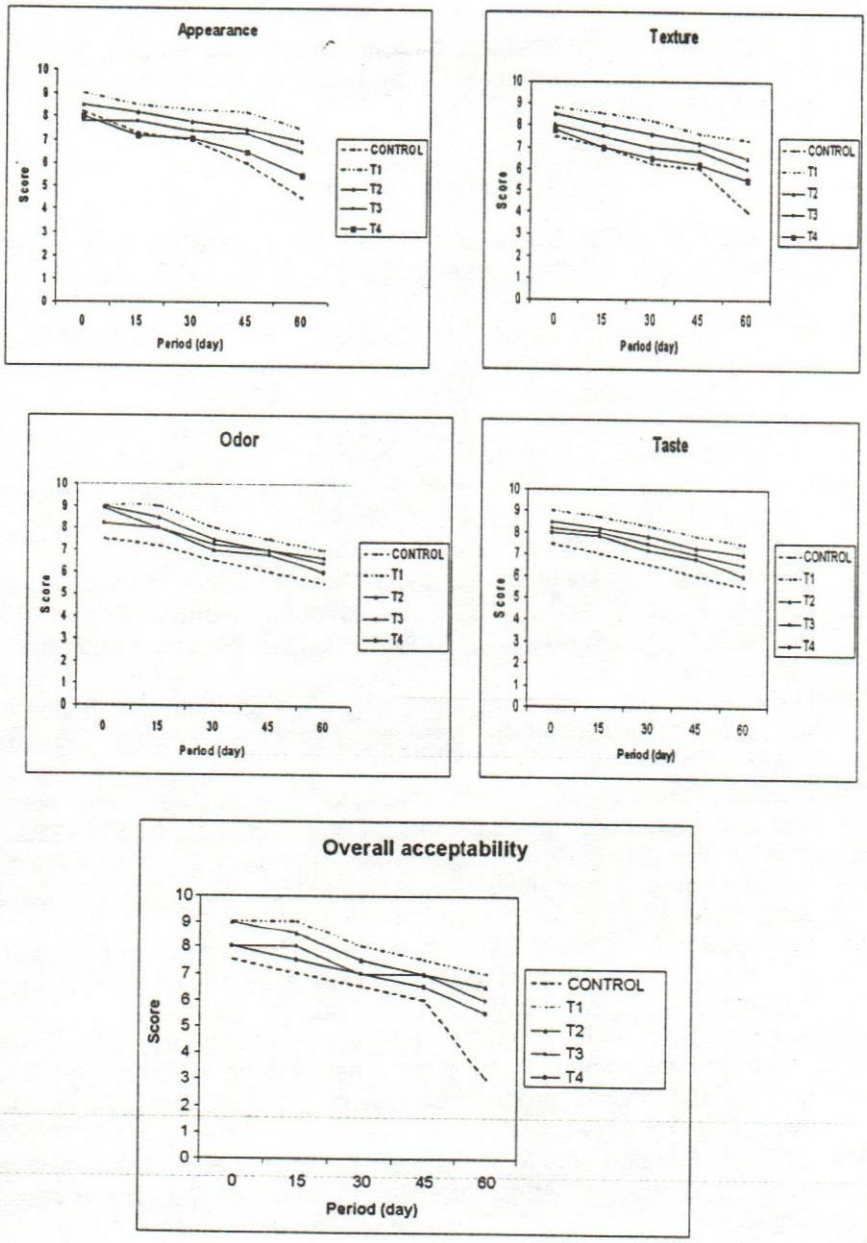

Fig. (1): sensory tests of salted fish fillets as affected by different plant extracts and cold storage. Control (no additives),T1:Cumin-treatment;T2:Thyme-treatment;T3:Licorice-treatment and T4: Combined- treatment).

Conclusion

According to the obtained data, it could be concluded that using of plant extracts (0.05%) improved both biochemical indices and sensory parameters of brined fish fillets.

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تأثير بعض المستخلصات النباتية على جودة شرائح سمك الطوبارة المملح

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يهدف هذا البحث الى دراسة تاثير بعض المستخلصات النباتية مثل الكمون والزعتر وعرق السوس ومخلوط منها (بتركيز ٠,٠٥%) على جودة شرائح سمك الطوبارة المملحة. حيث تم تعبئة حوالى ٢٠٠ جرام شرائح سمك فى اكياس بولى ايثلين محتوية على ١٠٠ملى محلول ملحى (تركيز ٢٠% ملح ، ٠,٠٢ % حمض الخليك ، ٠,٠٥ % مستخلص نباتى) ومفرغة الهواء جزئيا . ولقد استغرقت فترة التسوية ٢٠ يوما على درجة حرارة الغرفة (١٦ ± ٢° م) ، ثم حفظت فى الثلاجة (٤ ± ١° م) لمدة ٨ أسابيع. و لقد أجريت بعض الاختبارات الطبيعية والكيميائية والميكروبيولوجية والحسية لهذه المعاملات. وأوضحت النتائج المتحصل عليها مايلى:

زيادة كلا من المحتوى الرطوبى والرماد بينما حدث انخفاض فى كلا من البروتين والدهن فى المعاملات موضع الدراسة وذلك فى اليوم الأول من التمليح. وخلال فترات التسوية والتخزين فقد حدث زيادة فى المحتوى الرطوبى وكذلك قيم الأس الهيدروجينى والقواعد النيتروجية وثلاثى ميثايل الأمين فى حين انخفضت قيم حمض الثيوباربتيوريك . وميكروبيولوجيا فقد حدث انخفاض فى الأعداد الكلية للبكتيريا بينما زادت أعداد البكتيريا المحبة للملوحة. وحسيا فقد حسنت المستخلصات النباتية المستخدمة من خصائص الجودة مقارنة بالعينة الضابطة.

وبناء على ماسبق فان المستخلصات النباتية ذات تأثير واضح فى التحكم فى التغيرات البيوكيميائية وتحسين الخواص الحسية لشرائح السمك المملحة ، علاوة على امكانية الحصول على منتجات مملحة امنة.