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Quality Assessment of Salted Grass Carp (*Ctenopharyngodon idella*) Fillets Stored at Ambient Temperature.

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ABSTRACT

The present study was carried out to investigate the effect of salting (at 15% and 20% salt concentrations) and adding of some natural antioxidants (black seed, grape seed, jojoba extracts and chitosan) on the quality and shelf-life of grass carp stored at ambient temperature for 28 days. Chemical composition, pH value, total volatile basic nitrogen (TVB-N), thiobarbituric acid values (TBA); nutritional essential elements (Ca, K, P, and Na) and heavy metals (Cd, Cu, Fe, Mn, Ni, and Zn) and sensory scores were determined. Results indicated that, salting methods have a considerable effect on the nutritional value of fish, and in variations of moisture, protein, lipid, and ash contents. It caused significant decreases in moisture, protein, lipids, pH, Ca, K, P, Cd, Cu, Fe, Mn, Ni, and Zn, while it caused a significant increase in ash, carbohydrates, caloric values, TVBN, TBA and Na concentrations. On the other hand, moisture, protein, lipids were significantly decreased during storage, while ash, pH, TVBN and TBA were significantly increased but not exceed the maximum permissible levels. Such changes in chemical composition, physicochemical aspects and sensory scores during salting and storage of grass carp gave rise to recommend that salting of fish using natural antimicrobial and antioxidants compounds were better than control and they were in the following order black seed> chitosan>grape seed> jojoba.

Keywords: *Ctenopharyngodon idellus*, salting, black seed, grape seed, jojoba, chitosan.

INTRODUCTION

Natural antioxidants compounds which can be extracted from plant leaves, seeds, roots, and other plant parts or from seafood byproducts are preferred to synthetic antioxidants for preserving the quality and extending the shelf-life of fish products because they are safer than synthetics due to their antimicrobial and antioxidative properties (Tongnuanchan & Benjakul, 2014 and Huang, *et al.*, 2018).

Grass carp (*Ctenopharyngodon idellus*) is one of the most important commercial freshwater-cultured fish species. The global aquaculture production of grass carp was 5,537,794 tons in 2014, and it ranked first among principal aquaculture species (FAO, 2016). Fresh fish are highly perishable after death because of protein degradation, lipid oxidation or decomposition that is caused by microorganisms and endogenous enzymes under a high pH, and a high concentration of free amino acids conditions. Hence, taking some measures to delay the deterioration of fish quality and to extend the shelf-life of fish products are necessary (Cai, *et al.*, 2015 and Huang, *et al.*, 2018).

Several studies have shown that natural antioxidants compounds can maintain postmortem quality and extend shelf-life of fish and meat products. However, few studies have investigated the effect of natural antioxidants on the quality of fresh and salted grass carp fillets (Cai, *et al.*, 2015; Ghabraie, *et al.*, 2016 and Huang, *et al.*, 2018). Previous studies concluded that salting, apple polyphenols, and chitosan coating were effective in extending the shelf-life of grass carp (Talab, *et al.*, 2011; Sun, *et al.*, 2017; Wang, *et al.*, 2014a; Yu, *et al.*, 2017, and Huang, *et al.*, 2018).

Thus, the aim of this study was to evaluate the effect of (black seed extract, grape seed extract, jojoba and chitosan) on the quality of salted grass carp fillets stored at ambient temperature in terms of chemical composition, physicochemical, minerals, heavy metals and sensory score,.

MATERIALS AND METHODS

Fish samples

40 kg of fresh grass carp (mean weight of 1291.39 ± 12.23 and mean length of 41.37 ± 4.04 cm) were bought from El-Obour city fish market and they were carefully washed with potable water then packed in ice boxes and were transported to Fish Processing and Technology Laboratory, National Institute of Oceanography and Fisheries, El-Kanater El-Khiria City, El-Qaluobia Governorate, Egypt within 2 h. Then, fish samples were re-washed thoroughly with potable water scaled, beheaded, gutted, filleted and re-washed immediately and drained.

Fish salting

The obtained fish fillets were divided randomly into five groups. The control was not treated with natural antioxidants. The treatment groups were immersed in 1% (v/v) natural antioxidants solution (black seed extract, grape seed extract, jojoba and chitosan) for 1h at room temperature (4 kg grass carp fillets in 1000 ml of antioxidant solution). After that, each treated fillets were divided into two groups (15% and 20% salt concentration) and then packed individually in air tight plastic bottle with some air and stored at room temperature. Chemical composition, physical, chemical, sensory aspects, minor and major elements were carried out every week until they were denoted as completely spoiled by sensory panel.

Analyses

Moisture, protein, lipids and ash were determined according to the methods described by AOAC, (2012). The pH value was done by the method of (Goulas, *et al.*, 2005) using pH meter (HANNA, pH213). Total

volatile bases nitrogen (TVBN) was done as described by **Mwansyemela (1973)**. Thiobarbituric acid (TBA) value was determined by the distillation method outlined by **Tarladgis, et al., (1960)**. Magnesium (Mg), calcium (Ca), sodium (Na), potassium (K), phosphor (P), copper (Cu), iron (Fe), manganese (Mn) and zinc (Zn) contents were analyzed using an inductively coupled plasma optical emission spectrophotometer (ICP-OES) (Model 4300 DV, Perkin Elmer, Shelton, CT, USA) according to the method of **AOAC (1999)**. Organoleptic evaluation was performed as described by **Twig, et al., (1976)**. Results were expressed as mean \pm SD. and analyzed by using Microsoft Excel 2010, windows seven and SPSS 20, Statistical Software. Data were subjected to analysis of variance (ANOVA).

RESULTS AND DISCUSSION

Chemical composition of raw and salted grass carp

Table (1) showed that, the mean (\pm SD) proximate composition of moisture, crude protein, crude fat, ash, carbohydrates and calories (g/100 g fish muscle) in the fresh grass carp was (78.11 \pm 0.69), (16.55 \pm 0.84), (2.31 \pm 0.01), (1.87 \pm 0.01), (1.16 \pm 0.01) and (91.63 \pm 1.49), while it recorded (74.56 \pm 0.09), (15.60 \pm 0.04), (1.99 \pm 0.04), (3.55 \pm 0.04),(4.29 \pm 0.04) and (97.49 \pm 0.26) in 15% salted grass carp and and (73.78 \pm 0.09), (15.57 \pm 0.04), (2.02 \pm 0.04), (3.63 \pm 0.04), (5.01 \pm 0.04), (100.46 \pm 0.44), for 20% salted grass carp, respectively.

The obtained results agree with those reported by **Yu, et al., 2017** who reported that, the mean (\pm SD) proximate composition of moisture, ash, crude protein and crude fat (g/100 g fish muscle) in the fresh grass carp were 78.98 \pm 0.61, 1.15 \pm 0.09, 18.03 \pm 0.35, and 2.12 \pm 0.21, respectively. **Ormanci and Colakoglu, (2015)** contributed the major changes in protein content in salted fish to the increased of NaCl concentration, which increases protein degradation. The decrease in moisture, protein and lipids content after salting process may be due to the mass transfer occurs basically between salt and water during salting process: the fish muscle takes up salt and loses water, while as the increase in nutritional components, such as ash, carbohydrates and calories may be due to the loss of water in fish muscle during salting process (**Clucas & Ward, 1996, Bras & Costa, 2010; Chaijan, 2011, Oliveira, et al., 2012, Bakhiet & Khogalie, 2012**).

Table (1): Chemical composition (on wet weight basis) of fresh and salted grass carp fish fillets

Constitutes	Raw	Salted grass carp	
	grass carp	15%	20%
Moisture (%)	78.11 \pm 0.69	74.56 \pm 0.09	73.78 \pm 0.09
Protein (%)	16.55 \pm 0.84	15.60 \pm 0.04	15.57 \pm 0.04
Lipids (%)	2.31 \pm 0.01	1.99 \pm 0.04	2.02 \pm 0.04
Ash (%)	1.87 \pm 0.01	3.55 \pm 0.04	3.63 \pm 0.04
Carbohydrates (%)	1.16 \pm 0.01	4.29 \pm 0.04	5.01 \pm 0.04
Caloric value (kcal/100 g)	91.63 \pm 1.49	97.49 \pm 0.26	100.46 \pm 0.44

Physicochemical quality aspects of raw and salted grass carp

Table (2) showed that, pH value, TVBN and TBA values of fresh grass carp, 15% and 20% salted grass carp were (6.55 \pm 0.01), (6.52 \pm 0.06), and (6.49 \pm 0.01); (8.43 \pm 0.05), (11.02 \pm 0.01) and (9.94 \pm 0.07) (mg/100g ww) and (0.43 \pm 0.01), (3.79 \pm 0.08) and (3.50 \pm 0.05) (mg MDA/Kg), respectively. It is clearly that, salt process led to higher increase in TVBN and TBA value compared with raw fish, while slightly increased in pH value were observed.

Table (2): Physicochemical quality aspects (on wet weight basis) of raw and salted grass carp fish fillets

Paramters	Raw	Salted grass carp	
	grass carp	15%	20%
pH value	6.55±0.01	6.52±0.06	6.49±0.01
TVBN (mg/100 g)	8.43±0.05	11.02±0.01	9.94±0.07
TBA (mg MDA/kg)	0.43±0.01	3.79±0.08	3.50±0.05

Mineral composition of raw and salted grass carp

Mineral composition (mg/100g on dry weight basis) of raw grass carp, 15% and 20% of salted grass carp were as follows: calcium (Ca) content (170±0.27), (31±0.08) and (35±3.54), respectively; potassium (K) content (897±0.26), (100±1.54) and (195±2.22), respectively; phosphorus (P) content (187±0.12), (78±1.11) and (125±2.03), respectively; sodium (Na) content (938±0.35), (1410±11.24) and (1504±33.45), respectively (Table 3).

Table (3): Nutritional essential elements (mg/100 g on dry weight basis) of raw and salted grass carp fish fillets

Elements	Raw	Salted grass carp	
	grass carp	15%	20%
Ca	170±0.27	31±0.08	35±3.54
K	897±0.26	100±1.54	195±2.22
P	187±0.12	78±1.11	125±2.03
Na	938±0.35	1410±11.24	1504±33.45

The obtained results indicated that, salting process caused a significant decrease ($p \leq 0.05$) in (Ca, K and P) elements, while it caused a significant increase ($p \leq 0.05$) in Na content and this may be due to the sodium chloride added in processing procedure. This results agree with those reported by **Mohammed (2010)** who found that sodium content of raw and salted *Alestes dentex* fish were 142 and 612 mg/100g respectively.

Heavy metals concentrations of raw and salted grass carp

Heavy metals concentrations of fresh grass carp, 15% and 20% salted grass carp were as follows: Cadmium (Cd) content (0.07±0.001), (0.02±0.001) and (0.01±0.001); Copper (Cu) content (1.33±0.01), (0.97±0.001) and (1.19±0.09); iron (Fe) content (25.15±0.47), (15.48±1.03) and (16.68±1.01); manganese (Mn) content (0.98±0.01), (0.79±0.001) and (0.75±0.001); nickel (Ni) content (0.64±0.003), (0.49±0.01) and (0.47±0.001), and zinc (Zn) content (9.92±0.51), (9.16±1.01) and (7.92±0.08), respectively (Table 4).

Table (4): Heavy metals concentrations (mg/100 g on dry weight basis) of fresh and salted grass carp fish fillets

Metals	Raw	Salted grass carp	
	grass carp	15%	20%
Cd	0.07±0.001	0.02±0.001	0.01±0.001
Cu	1.33±0.01	0.97±0.001	1.19±0.09
Fe	25.15±0.47	15.48±1.03	16.68±1.01
Mn	0.98±0.01	0.79±0.001	0.75±0.001
Ni	0.64±0.003	0.49±0.01	0.47±0.001
Zn	9.92±0.51	9.16±1.01	7.92±0.08

The obtained results revealed that, salting process caused higher reduction in the concentrations of all tested metals. Mean values of Cd in the analyzed fish samples (mg/100g) were below the maximum

permissible value indicated by the European Community (EEC, 2001). The maximum Cd level permitted by the FAO (1983) is $0.5 \mu\text{g g}^{-1}$ and $0.2 \mu\text{g g}^{-1}$ by MAFF (1995) (FAO, 1984). Similar results were also reported by Sobhanardakani and Jafari (2014) who found that, cadmium concentration in the muscle of fresh grass carp was ranged from $(0.004\text{-}0.012 \mu\text{g g}^{-1})$. Pirestani, *et al.*, (2009) found that, the levels of Mn, Fe, Zn and Cu of fresh common carp were 0.17 ± 0.01 ; 12.55 ± 0.21 ; 7.44 ± 0.30 and $2.58 \pm 0.14 \text{ mg/kg}$ dry weight. While, Yi and Zhang (2012) found that, the mean concentrations of metals (copper zinc, cadmium) in the tissues of *Ctenopharyngodon idellus* fish caught from the Yangtze River were 0.834 ± 0.655 ; 2.8 ± 3.17 and 0.0457 ± 0.0449 (mg/kg wet weight), respectively.

Chemical composition changes of salted grass carp stored at ambient temperature

Moisture content

Figure (1) showed that, moisture content of 15% and 20% salted grass carp (control, chitosan, grape seed, black seed and jojoba) were (74.56, 75.46, 74.81, 75.35 and 74.78) and (73.78, 74.76, 73.58, 74.72 and 74.50) respectively at zero time, while it was recorded (68.99, 69.94, 69.91, 68.97 and 68.51) and (67.64, 69.04, 69.27, 67.94 and 66.20), respectively, at the end of storage period. Moisture content was significantly decreased in all treatments, where moisture content of salted grass carp with addition of chitosan, grape seed, black seed and jojoba were higher than control. The results are in accordance with those of Badawy (1979); Hernandez-Herrero, (1997) and Ahmed, *et al.*, (2010). The decrease in moisture content during storage may be due to high osmotic pressure which leads to diffuses salt into the fish tissues, but at the same time water moves by osmosis from the fish at a high speed into the surrounding brine and the fish declines in weight (Voskresensky, 1965 and Alsaban, *et al.*, 2014).

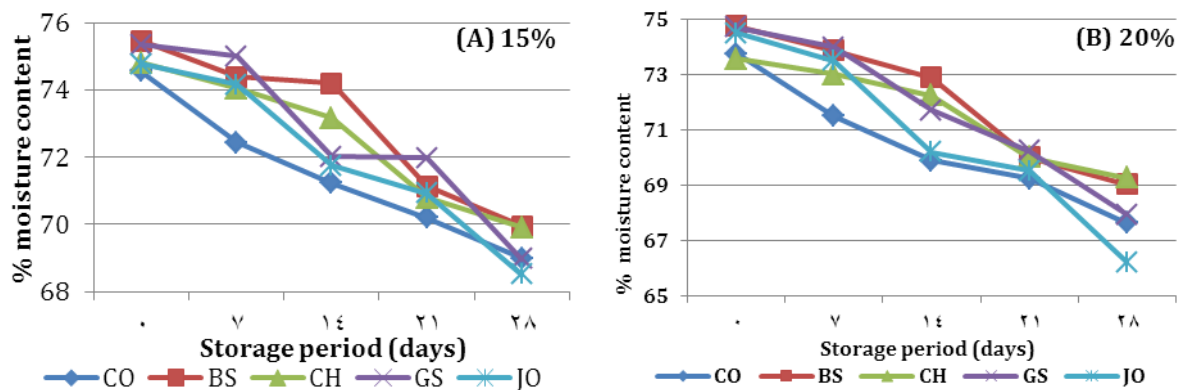


Fig. (1). Changes in moisture content of salted grass carp during storage at ambient temperature.

Protein content

Protein content of 15% and 20% salted grass carp (control, black seed, chitosan, grape seed, and jojoba) were (15.60, 15.67, 15.69, 15.64, 15.61) and (15.57, 15.65, 15.61, 15.67, 15.58) respectively at zero time, while it was recorded (12.49, 13.28, 13.64, 13.47, 12.99) and (12.33, 13.26, 13.18, 12.84, 12.90), respectively, at the end of storage period. Protein content was decreased in all treatments, where protein content of salted grass carp with addition of chitosan, grape seed, black seed and jojoba were higher than control (Figure 2).

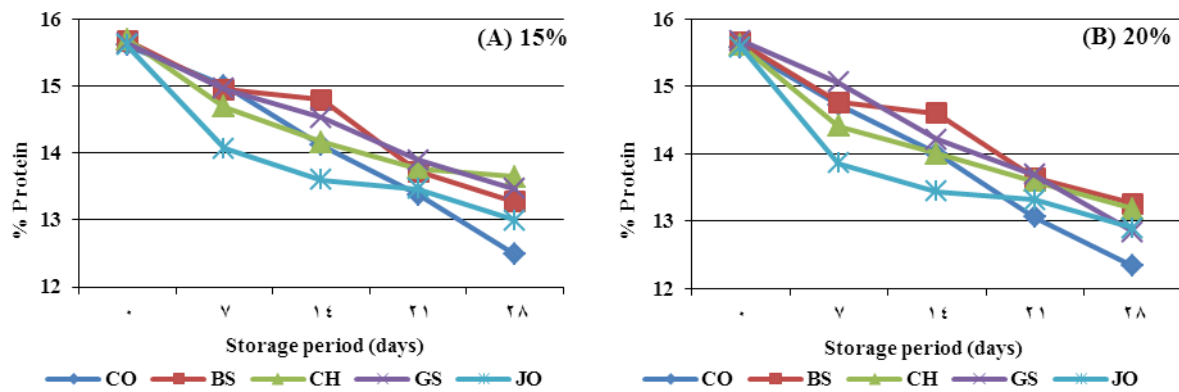


Fig. (2). Changes in protein content of salted grass carp during storage at ambient temperature.

As a result of decreasing in protein content salt penetrates the tissue, it alters the colloidal properties of the proteins and changes the nature of the water/protein relationship (Zaitzev, *et al.*, 1969). This finding can be explained as a result of the denaturation of both sarcoplasmic and myofibrillar proteins due to the effect heavy salting on fish muscle. These results are in accordance with (El-Sharnouby, 1989; Ahmed, *et al.*, 2010 and Alsaban, *et al.*, 2014)

Lipid content

Lipid contents of 15% and 20% salted grass carp (control, black seed, chitosan, grape seed, and jojoba) were (1.99, 1.76, 1.78, 1.78, 1.88) and (2.02, 1.91, 1.97, 1.95, 1.94) respectively at zero time, while it was recorded (1.22, 1.40, 1.28, 1.16, 1.26) and (1.45, 1.47, 1.39, 1.38, 1.54), respectively, at the end of storage period. Lipids content was decreased in 15 and 20% salt treatments, where lipids content of salted grass carp with addition of chitosan, grape seed, black seed and jojoba were higher than control (Figure 3).

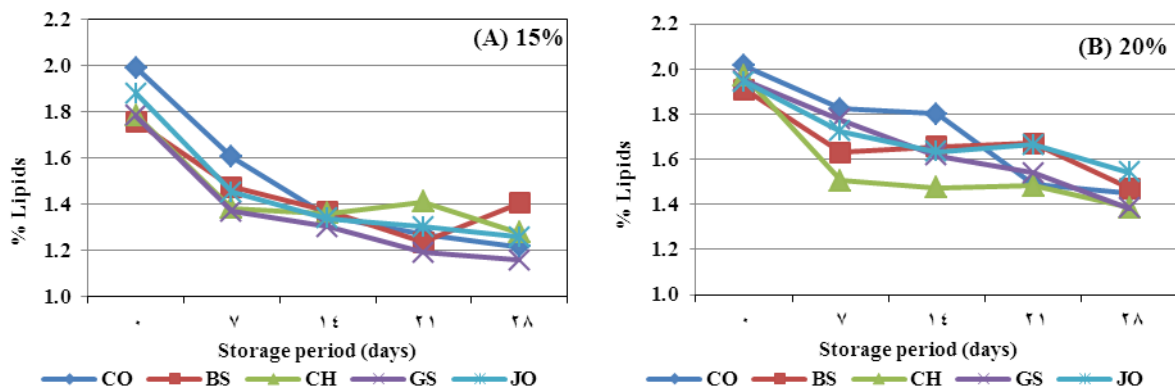


Fig. (3). Changes in lipid content of salted grass carp during storage at ambient temperature.

The decreasing in lipid content, may be attributed to their insolubility in water, which diffusion throughout the cell walls to the brine solution and the hydrolysis of triglycerides and phospholipids, which is catalyzed by lipases and phospholipases and release of free fatty acids that is soluble in water, then leaching into the drip (Voskresensky, 1966; Aman and Shehata, 1978; Al-Habib & Al-aswad, 1985 and Alsaban, *et al.*, 2014).

Ash content

Ash content of 15% and 20% salted grass carp (control, black seed, chitosan, grape seed, and jojoba) were (3.55, 3.97, 3.52, 3.59, 3.98) and (3.63, 4.05, 4.18, 4.08, 4.23) respectively at zero time, while it was

recorded (6.22, 6.35, 6.23, 6.35, 6.40) and (6.43, 6.53, 6.75, 6.54, 6.84), respectively, at the end of storage period.

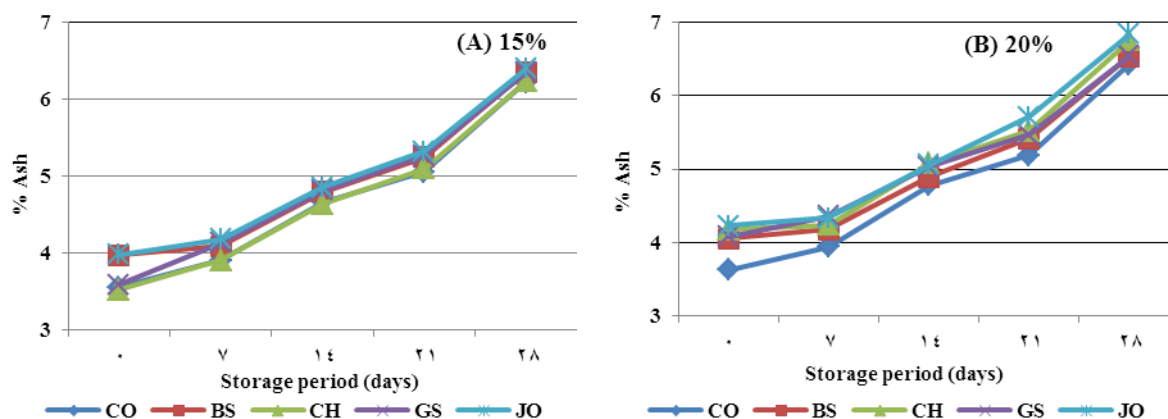


Fig. (4). Changes in ash content of salted grass carp during storage at ambient temperature.

From the above findings, it can be concluded that, ash content was decreased in all treatments, where ash content of salted grass carp with addition of chitosan, grape seed, black seed and jojoba were higher than control (Figure 4).

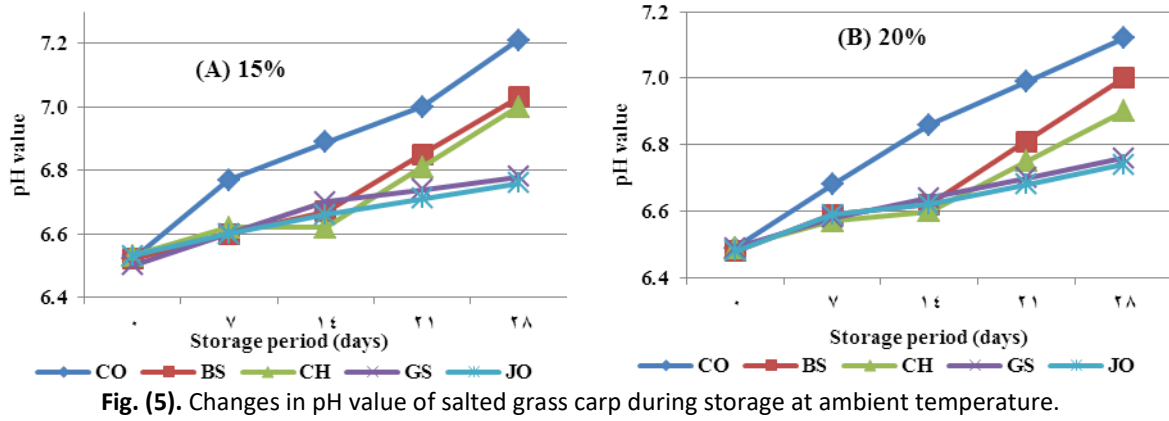
The increasing in ash content during salting period may be due to effect of extracted lipid which helps to create a crusted surface on each dried fish (Mohammed, 2007) and effect of ground bones and scales in dried meat, also the presence of residues from salt during preparation of samples for analysis and this consequently lead to increase the ash content. These results were agreement with (Hernandez-Herrero, 1997; Ahmed, et al., 2010 and Alsaban, et al., 2014). It is known that inorganic matter increases in salted fish and it depends on preservation period and salt rate. Inorganic matter and salt rates increase along the storage period in anchovy stored that had been salt cured and were in storage for 29 weeks, and that this condition affects other components (Kolsarici & Candoğan, 1997 and (Binici &Kaya, 2017).

Physicochemical quality changes of salted grass carp stored at ambient temperature

pH value

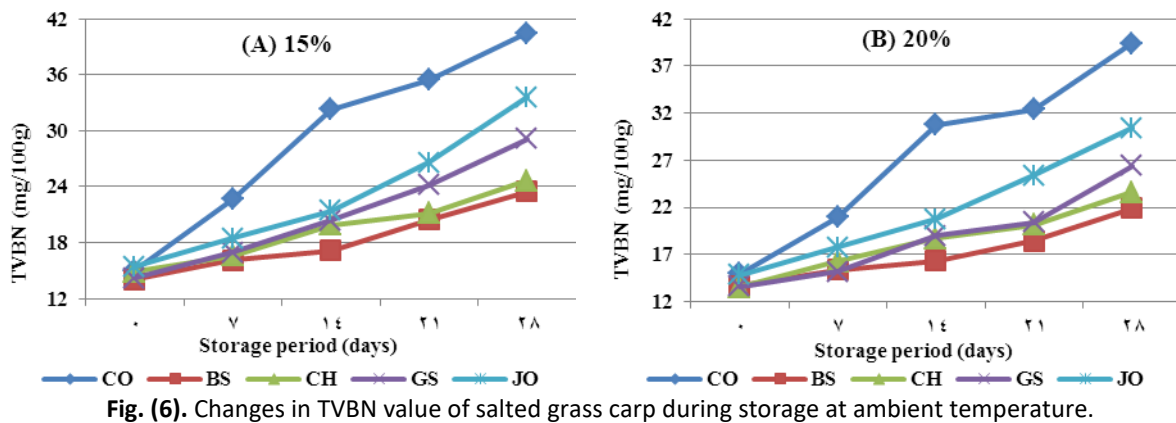
pH value of 15% and 20% salted grass carp (control, chitosan, grape seed, black seed and jojoba) were (6.52, 6.52, 6.53, 6.50, 6.53) and (6.49, 6.48, 6.49, 6.49, 6.48) respectively at zero time, while it was recorded (7.21, 7.03, 7.00, 6.78, 6.76) and (7.12, 7.00, 6.90, 6.76, 6.74), respectively, at the end of storage period. The pH values were decreased in 15 and 20% salt treatments, where pH value content of salted grass carp with addition of chitosan, grape seed, black seed and jojoba were higher than control (Figure 5).

The decrease in pH value from 6.55 to 6.52 and 6.49 of 15% and 20% of salted grass carp may be due to the ionic strength of the solution inside of the cells. This results agree with those reported by (Goulas & Kontominas, 2005; Leroi & Joffraud, 2000).



Total Volatile Basic Nitrogen (TVBN)

TVBN values of 15% and 20% salted grass carp (control, black seed, chitosan, grape seed, and jojoba) were (15.02, 14.04, 14.90, 14.22 and 15.42) and (14.94, 13.64, 13.54, 13.62, 14.80) respectively at zero time, while it were recorded (40.45, 23.45, 24.62, 29.14 and 33.59) and (39.40, 21.90, 23.60, 26.40 and 30.40), mg/100g on wet weight basis, respectively, at the end of storage period. Data showed that, TVBN values were increased in 15 and 20% salt treatments, where TVBN content of salted grass carp with addition of chitosan, grape seed, black seed and jojoba were lower than control (**Figure 6**).



Thiobarbituric acid value (TBA)

TBA values of 15% and 20% salted grass carp (control, black seed, chitosan, grape seed, and jojoba) were (3.79, 3.36, 3.76, 3.44, 3.63) and (3.50, 3.24, 3.39, 3.38, 3.35) respectively at zero time, while it was recorded (11.51, 10.37, 6.54, 7.57, 8.00) and (11.06, 9.94, 5.83, 6.55, 7.08), mg MDA/kg respectively, at the end of storage period.

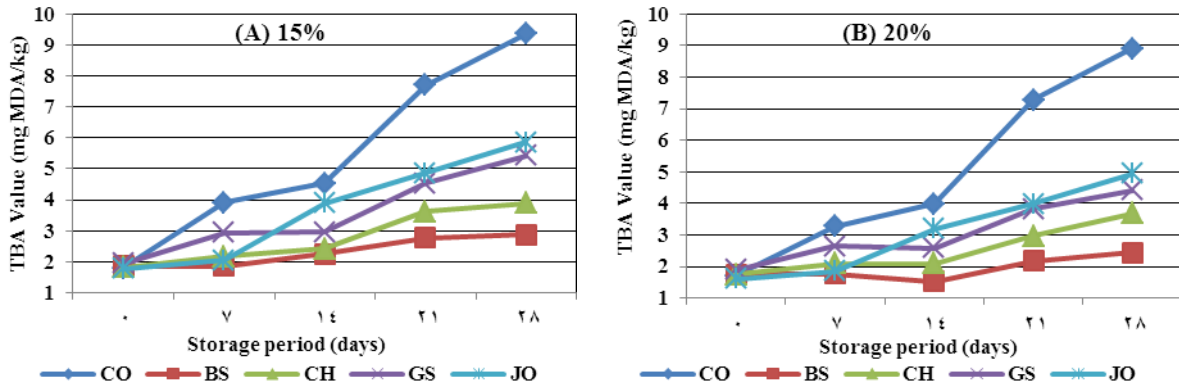


Fig. (7). Changes in TBA value of salted grass carp during storage at ambient temperature.

TBA values were increased in 15 and 20% salt treatments, where TBA content of salted grass carp with addition of chitosan, grape seed, black seed and jojoba were higher than control (Figure 7). The increase in TBA indicated the formation of secondary oxidation products such as aldehydes and other volatile compounds responsible for rancid flavour and off odors as well as colour and texture deterioration. The oxidative breakdown of lipids is also evidenced by the incidence of high TBA values in the respective samples (Kolakowska, 2002 and Jeyasanta, et al., 2016).

Sensory evaluation of salted grass carp stored at ambient temperature

Sensory analysis of 15% and 20% salted grass carp stored at ambient temperature are illustrated at figures (8) and (9). The overall acceptability scores decreased with increase in storage period. Samples with 20% salt received higher scores than 15% salt concentration due to increasing salt concentration had a progressive effect on increasing the sensory quality at ambient temperature.

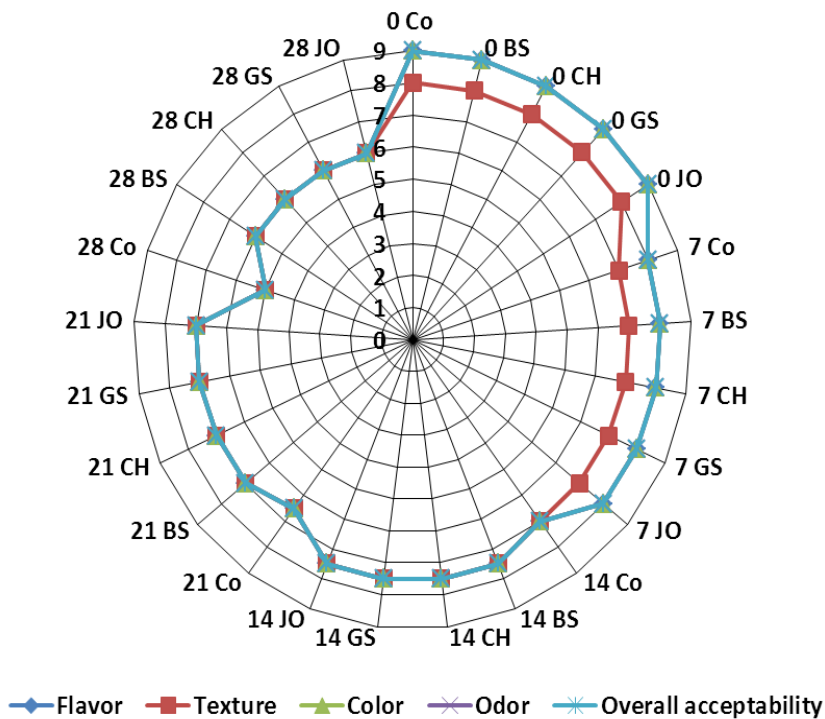


Fig.8. Sensory evaluation of 15% salted grass carp stored at ambient temperature

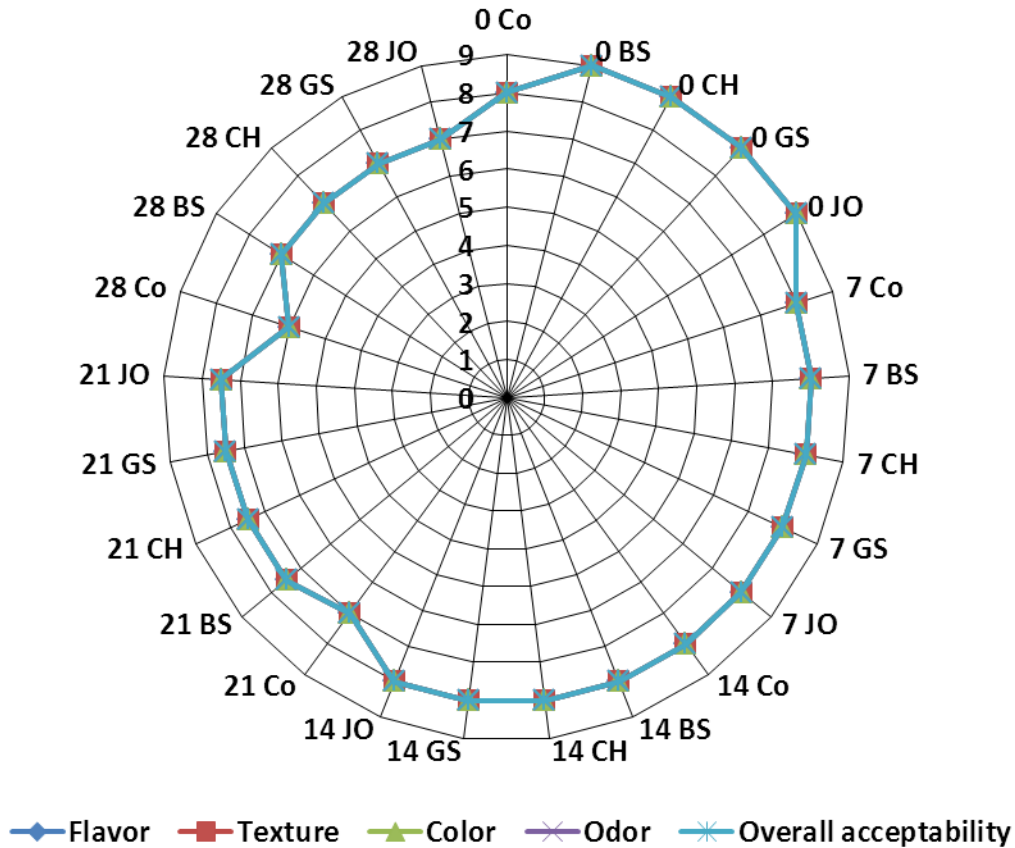


Fig.9. Sensory evaluation of 20% salted grass carp stored at ambient temperature

CONCLUSION

Salting methods have considerable effect on the nutritional value of fish, and in variations of moisture, protein, lipid, and ash contents. It caused significant decreases in moisture, protein, lipids, pH, Ca, K, P, Cd, Cu, Fe, Mn, Ni, and Zn, while it caused a significant increase in ash, carbohydrates, caloric values, TVBN, TBA and Na concentrations. On the other hand, moisture, protein, lipids were significantly decreased during storage, while ash, pH, TVBN and TBA were significantly increased but not exceed the maximum permissible levels. Such changes in chemical composition and physicochemical aspects during salting and storage of grass carp gave rise to recommend that salting of fish using natural antimicrobial and antioxidants compounds were better than control and they were in the following order black seed> chitosan>grape seed> jojoba.

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