ISSN 1110-0419

Annals Of Agric. Sc., Moshtohor,
Vol. 45(4): 1431-1444, (2007).

USING ROSEMARY AND SAGE FOR EXTENDING THE SHELF-LIFE OF COMMON CARP FISH FINGERS DURING COLD STORAGE

BY

Abd El-Razik, M. M.[*]; Elhariry, H. M.[*] and Abou-Taleb, M.[**]

[*] Department of Food Science, Fac. of Agri'c., Ain Shams Univ., Cairo, Egypt.
[**] Laboratory of Fish Processing and Technology, National Institute of Oceanography and Fisheries, Cairo, Egypt

ABSTRACT

Rosemary (*Rosemarinus officianalis* L.) or sage (*Salvia officinalis* L.) was incorporated at level of 1.5 or 3% in common carp (*Cyprinus carpio*) fish fingers. Quality and shelf life of the control and treated carp fingers during storage at 4 ±1 C were investigated periodically using organolyptic (appearance color, odor, taste, tenderness, juiciness, and overall acceptability), chemical (TVB-N, TMA, TBA, AV, and PV) and microbiological (mesophilic, psychrophilic, and *Enterobacteriacea* counts) analysis. All studied chemical parameters were significantly ($P<0.01$) correlated with storage time. Although, total volatile basic nitrogen (TVB-N) values were higher than 40 mg/100 g, the carp fish fingers were accepted based on the organolyptic evaluation. Trimethylamine-nitrogen (TMA-N) exhibits a great difference between treatments and seems to be a better index of spoilage for fish products processed from the freshwater such as common carp fish. The bacterial growth was significantly inhibited due to using rosemary and sage, where the enumerated mesophilic bacteria and psychrophilic counts did not excess than 4 log cfu/g and 2 log cfu/g, respectively during storage period at 4 ±1°C.

The obtained results indicated that, using of rosemary or sage at level of 3% led to delay the onset of organolyptic spoilage of carp fish fingers to the tenth day of cold storage, while to the fifth day in control sample.

Key words: Common carp – fish fingers- sage- rosemary- shelf-life - antioxidants – antimicrobial

INTRODUCTION

Common carp fish is considered as one of the main types of fish widely produced in fish farms, due to their faster growth rate and easy breeding. However, the fresh common carp fish did not find acceptance by consumers (Darweash 1996). Therefore, it has been an increasing interest in the consumption of restructured minced fish products in recent years (Montero *et al.*, 2005). Lipid oxidation, leading to rancidity, is one of the major reasons of fish products' quality deterioration, mainly because of their increased fat content, which characterized by the presence of polyunsaturated fatty acids (PUFA). In this sense, lipid oxidation causes degradation of PUFA and generation of residual products and lipid-derived volatiles (Bicchi *et al.*, 2000). The important biochemical parameters commonly used for the shelf-life estimation of seafoods are pH, trimethylamine nitrogen (TMA-N) and total volatile bases nitrogen (TVB-N). The rate of pH decreases due to the formation of lactic acid in the flesh just after death; but then the decomposition of nitrogenous compounds leads to an increase in pH. The TVB-N and TMA-N values increase during storage (Sikorski *et al.* 1990). Under refrigeration conditions, lipid oxidation compounds have shown to facilitate protein denaturation (Mackie, 1993; Sikorski and Kolakowska, 1994) and nutritional losses (Castrillon *et al.*, 1996). On the other hand, formation of carbonyl groups has been widely used as a measure of fat oxidation (Srinivasan and Hultin, 1995). These irreversible changes contribute to the development of unacceptable organoleptic characteristics and are more important in fish products preserved under cold or frozen conditions (Sikorski and Kolakowska, 1994).

To store and commercialize fish products in a safety and high quality state, deterioration occurring from lipid oxidation may be reduced by incorporation of natural or synthetic antioxidants (Bicchi *et al.*, 2000). The increasing consumer awareness and health consciousness, however, results in pressure to avoid the use of synthetic additives, which necessitates the use of natural additives or alternative methods to extend shelf life and/or improve safety (Bicchi *et al.*, 2000). Thus, many researches have been focused on the positive role of antioxidant molecules present in plants (Miyake and Shibamoto, 1997; Sebranek *et al.*, 2005; Estevez *et al.*, 2007).

The *Labiatae* family includes a large number of plants that are well known for their antioxidant properties. Among these, rosemary (*Rosemarinus officianalis* L.) and sage (*Salvia officinalis* L.) have been widely studied (Zheng and Wang 2001, Dorman, *et. al*, 2003). The antioxidant activity of sage and rosemary essential oils is mainly related to two phenolic diterpenes: carnosic acid and carnosol which are considered two effective free-radical scavengers (Dorman *et al.*, 2003; Ibanez *et al.*, 2003). The antioxidant activity of these molecules has been compared to that of other recognized synthetic antioxidant substances (Richheimer *et al.*, 1999; Ibanez *et al.*, 2003; Bicchi *et al.*, 2000; Sebranek, *et al.*, 2005; Estevez *et al.*, 2007). The antioxidant potential of the carnosic acid was higher than that of BHT and BHA (Richheimer *et al.*, 1999). Generally, the main phenolic compounds identified in rosemary and sage are rosmarinic, carnosic acids, carnosol, methyl carnosate, rosmanol, epirosmanol and rosmadial (Richheimer *et al.*, 1999; Zheng and Wang 2001). A number of researchers have reported the effectiveness of rosemary extracts for achieving higher sensory scores and retarding lipid oxidation in various foods (Shahidi and Wanasundara, 1992; Sebranek *et al.*, 2005). In addition to inhibition of lipid oxidation, colour changes during storage were inhibited by the addition of rosemary extract in irradiated ground beef (Formanek, *et al.*, 2003).

Antimicrobial activities of spices and herbs and essential oils have been well known for long time. Many studies reported the activities of spices and herbs or essential oils to a variety of bacteria (Hirasa and Takemasa, 1998; Yutaka *et al.*, 2006).

Nevertheless, the protective activity of rosemary and sage against lipid oxidation on a product with the oxidative instability of common carp fish products remains unknown. The aims of the present work were to investigate the effect of the addition of sage and rosemary as natural sources of antioxidants on the lipid oxidative stability of carp fish finger during cold storage. The antimicrobial effects of these plants were also studied.

MATERIALS AND METHODS

Materials:

Fresh common carp fish of the species *Cyprinus carpio* were obtained from El-Sarwo farm, El-Dakahlia Governorate, National Institute of Oceanography and Fisheries, at July 2006 season. The common carp fish were approximately 2.5-3.0 kg weight each. The fish samples were put in ice box and transferred to Department of Food Science, Faculty of Agriculture, Ain Shams University for further technological treatments and analysis. Rosemary, sage and other materials used in spices mixture were obtained from the local market. The spices mixture was consisted of 42% black pepper, 23% cumin, 18% all spices, 5% ginger, 5% cardamom, 2% cubeb, 2% clove, 2% coriander and 1% red pepper.

Preparation of common carp fish fingers:

Fish samples were manually washed, deboned and cut to small pieces. These pieces were washed again to remove any blood traces and then drained for a few minutes. Fish meat was ground using a laboratory universal kitchen machine (Varimix- Spomoasy, Poland) and allowing passing through a 0.64 cm plate. Carp fish finger were prepared by mixing: 75% minced fish meat, with 9% vegetable oil, 8% broken ice meal, 2.3% sodium chloride, 2.5% onion, 0.5% garlic and 2% spices mixture (Chandrasekhar and Mohite, 1978).

To evaluate the effect of rosemary or sage as a source of antioxidant and antimicrobial agents, 1.5 or 3.0 % of each was added to the final mixture. Fish fingers (7.0 cm x 2.0 cm) were formed, packaged in polyethylene bags and stored at 4 ± 1°C for 10 days.

Proximate chemical analysis:

Moisture, crud protein, fat and ash contents of common carp fish meat before and after mixing with the ingredients (control sample) were determined as described by A.O.A.C. (2000).

Trimethylamine-nitrogen (TMA-N):

TMA-N was determined as described in A.O.A.C. (2000).

Peroxide value:

was determined as described in A.O.A.C. (2000), the peroxide values were expressed in units of meq/kg of oil.

Acid value (A.V.): was determined as described in A.O.A.C. (2000).

Total volatile basic nitrogen (TVB-N):

TVB-N associated with fish spoilage was determined according to the method of Conway (1950) which modified by Harold *et al.*, (1987).

Thiobarbituric acid (TBA):

TBA values of common carp fish finger were determined spectrophotometrically according to the procedure described by Harold *et al.*, (1987).

Total mesophilic aerobic bacterial counts:

Total mesophilic aerobic bacteria were determined as described in (ICMSF, 1986). Ten grams of common carp fish fingers were homogenized for 1 min at room temperature in 90 ml of sterilized 0.9% NaCl saline using a Stomacher 400 homogenizer (Seward, Basingstoke, England). Serial decimal dilutions were prepared in 0.9% NaCl saline solution. Spreading plate technique was applied to determine the total aerobic count on plate count agar (Difco). The cfu were enumerated after incubation at 37 °C for 48 h. The experiment was carried out three times in duplicate.

Psychrophilic counts:

Psychrophilic bacteria were determined in a similar method to that for total aerobic bacterial counts except that plates were incubated at 7 °C for 10 days (Baumgard, 1986.)

***Enterobacteriaceae* counts:**

Enterobacteriaceae counts were enumerated by the pour plate technique on Violet Red Bile Glucose Agar (VRBGA; Difco, Detroit, Michigan, USA). The plates were overlaid with a virgin layer of the same growth medium and then incubated at 37 °C for 48 h (ICMSF, 1986).

Sensory evaluation:

The organoleptic properties of fried common carp fish fingers were evaluated by a group of 10 trained panelists from the staff members of the Dept. of Food Sci., Fac. of Agric., Ain Shams Univ. Samples were fried in an electrical fryer pan using corn oil at 170 °C for 5 min just before sensory evaluation. The panelists were asked to evaluate the color, odor, texture, taste, tenderness, juiciness and overall acceptability according to 9-point hedonic scale (Larmond, 1974). Data from the ten independent panelists were pooled and statistically analyzed.

Statistical analysis:

The statistical analysis system (SAS, 1996) was used to carry out mean of three values, standard error in addition to an over all analysis of variance (ANOVA) and least significant differences (LSD) at 0.01.

RESULTS AND DISCUSSION

Proximate composition of raw common carp fish meat and fingers:

Main chemical composition of raw common carp fish meat and fingers were presented in Table (1). A reduction in moisture and protein contents was noticed after addition of spices mixture to carp fish meat. This result may be due to low moisture and protein content of the spices (Pradeep *et al.*, 2005). On the other hand addition of vegetable oil led to increase the fat content of the formulated fish finger. Also the ash content of carp fish finger was increased by addition of the spice mixture and other ingredients such as onion due to the high ash content of these additives (Nwinuka *et al.*, 2005; Pradeep *et al.*, 2005). This result indicates the high content of minerals in the formulated carp fish finger compared with carp fish meat.

Organolyptic evaluation of common carp fish fingers during cold storage:

Table 2 summarized the organolyptic properties of common carp fish fingers contained different concentrations (1.5 or 3 %) of either rosemary or sage in addition to the control samples which contained all other ingredients and spices mixture. No significant effect due to incorporation of rosemary or sage at any of the studied concentration was recorded except the odor and taste which were significantly enhanced. During the first three days of storage, the overall acceptability of all tested samples was significantly reduced ($P < 0.01$), except that contained 3 % rosemary or sage. The control samples were rejected according to the organolyptic evaluation (onset of spoilage) after on the third day of storage at 4 ±1°C. The onset of organolyptic spoilage was recorded on the seventh day of cold storage by carp fish fingers contained 1.5% of rosemary or sage. Whereas, incorporation of rosemary or sage at level of 3% led to extending the shelf-life of carp fish fingers to ten days under conditions of cold storage (Table 2).

Table (1): Proximate composition (± standard error) of raw common carp fish meat and fingers.

Fish sample	Moisture %	Protein* %	Fat* %	Ash* %	FNE %
Raw common carp fish meat	76.52 ±0.12	78.86 ±0.25	12.85 ±0.16	5.25 ±0.35	3.01 ±0.21
Raw common carp fish fingers	68.36 ±0.22	68.15 ±0.31	15.75 ±0.15	6.48 ±0.27	9.62 ±0.26

* On dry basis; FNE: Free nitrogen extract, calculated with difference.

Effect of rosemary and sage on chemical changes of common carp fish fingers during cold storage:

Total volatile basic nitrogen (TVB-N):

The determination of TVB-N is widely used to estimate the degree of fish decomposition. So the mean values of TVB-N of different carp fish fingers were determined and are summarized in Table (3). The initial values of TVB-N were ranged between 34.04-35.4 mg/100 g dry weight in the different samples. TVB-N values of common carp fish finger samples increased significantly on the third day of storage ($P < 0.01$). The highest value of TVB-N for control sample was 67.01 mg/100 g on the fifth day of storage. This value was significantly higher ($P < 0.01$) than the TVB-N value of the fresh common carp fish fingers. On the third day of cold storage, the control samples and carp fish fingers contained 1.5% rosemary showed TVB-N values higher than the level of 40 mg TVB-N/100g. Lakshmanan (2000) stated that this level is usually regarded as spoilage limit of fish muscle. On the fifth day of cold storage of carp fingers, all samples showed TVB-N values higher than the abovementioned limit, although only the control sample was organolyptically rejected by the panellists. This result indicated that the TVB-N test was not the bitter index of spoilage of the common carp fish fingers.

Trimethylamine-nitrogen (TMA-N):

TMA-N is also used as an index of quality for deciding the state of freshness of fish. At the beginning of storage TMA-N values were 3.22 -3.95 mg/100g based on dry weight (Table 4). Significant increases in the TMA-N values were noticed on the third day of cold storage of all tested samples. However, the development rate of TMA-N in the control sample was higher than those of the samples contained rosemary or sage. At the onset of organolyptical spoilage of control sample (on the fifth day), the TMA-N value was reached to 17.42 mg/100 g. On the seventh day of cold storage of carp fish finger contained 1.5 % of rosemary or sage, the TMA-N values were reached to 14.98 and 15.12 mg/100 g, respectively. Also, carp fish finger contained 3% of rosemary or sage were organolyptically rejected on the tenth day of cold storage when the TMA-N values were reached to 15.05 and 15.25 mg/100 g, respectively. The values of TMA-N determined in the present study at the onset of organolyptic spoilage were closed to the limit of acceptability for human consumption (15 mg/100 g) in chilled fish (Connell, 1975). The obtained results in the present study demonstrated that the TMA-N provide adequate quality index for common carp fish fingers stored at 4 ±1°C. This conclusion is in accordance with Manju *et al.* (2007); they stated that TMA-N could be used as a quality index for refrigerated fish fillets.

Thiobarbituric acid (as O.D.):

Data of TBA of different samples of common carp fish fingers during cold storage were tabulated in Table (5). There were incremental patterns in TBA values for different treatments. Both sage and rosemary were significantly reduced the increasing rate compared with those of control sample during storage period. These results suggest that the antioxidant content of both rosemary and sage retarded lipid oxidation during cold storage as mentioned by Giměnez *et al.*, (2005). The addition of either rosemary or sage at the level of 3% was significantly improved the storage stability of carp fish fingers at 4 ±1°C. The ability of rosemary and sage to inhibit lipid oxidation may be due to its high content of phenolic constituents which act as inhibitors for radical reactions on autoxidation as mentioned by Ibañez *et al.*, (2003).

Acid and peroxide values:

Acid and peroxide values are commonly used to determine quality in fish and meat muscle treated under different conditions of handling, processing and storage through the changes of proteins, water and lipids of muscle food (Ruiz-Capillas and Moral, 2001.). Data in Table 6 showed significant increasing in both acid and peroxide values of all tested common carp fish fingers during cold storage. At the onset of organolyptic spoilage, the measured acid and peroxide values were higher than 7 mg KOH/g oil and 20 meq/Kg oil, respectively, in all tested carp fish fingers (Table 6). These values showed a general trend to increase in lipid oxidation products with cold storage time in both control and treated carp fish fingers.

Effect of rosemary and sage on microbiological criteria of common carp fish fingers during cold storage:

Microorganisms are one of the important reasons for spoilage because they break down the food into a form that they can utilize. Therefore, food quality decreases and spoilage starts at this stage and estimation of the quality of food products relies on the quantification of total numbers of microorganisms (Turantas, 2000). In the present study, total mesophilic and psychrophilic aerobic counts were determined as microbiological criteria for fish spoilage. Data in Figure (1) illustrate the effect of rosemary and sag on total bacterial counts of common carp fish fingers during cold storage (4 ±1°C).

Table (2): Sensory properties of different common carp fish fingers stored at 4 ± 1 °C.

Storage period (day)	Common carp fish fingers contained				
	Control	1.5% Rosemary	3.0% Rosemary	1.5% Sage	3.0% Sage
Appearance					
Zero	8.^{55Aa}	8.75^{Aa}	8.67^{Aa}	8.88^{Aa}	8.78^{Aa}
3	6.67^{Bb}	7.67^{Ba}	7.55^{Ba}	7.43^{Ba}	8.00^{Aa}
5	5.00^{Bc*}	6.66^{Ca}	7.21^{Ba}	6.86^{Ba}	7.33^{Ba}
7		5.75^{Db*}	6.67^{Ca}	5.00^{Cc*}	6.32^{Ca}
10			5.55^{Ba*}		5.00^{Da*}
Color					
Zero	8.67^{Aa}	8.67^{Aa}	8.67^{Aa}	8.67^{Aa}	8.67^{Aa}
3	6.67^{Bb}	7.67^{Ba}	7.33^{Ba}	7.33^{Ba}	8.00^{Aa}
5	5.00^{Cb*}	6.66^{Ca}	7.00^{Ba}	6.67^{Ba}	7.33^{Ba}
7		5.55^{Db*}	6.55^{Cb}	5.12^{Cb*}	6.11^{Ca}
10		·	5.67^{Da*}		5.00^{Da*}
Odor					
Zero	8.00^{Ab}	8.55^{Aa}	8.67^{Aa}	9.00^{Aa}	9.55^{Aa}
3	7.85^{Bb}	7.75^{Bb}	8.00^{Aa}	8.55^{Aa}	9.00^{Aa}
5	6.55^{Cc*}	6.66^{Cc}	7.55^{Bb}	7.55^{Bb}	8.55^{Ba}
7		5.55^{Dc*}	6.67^{Ba}	6.00^{Cb*}	7.00^{Ba}
10			5.67^{Ca*}		5.50^{Ca*}
Taste					
Zero	7.85^{Ab}	8.67^{Aa}	8.67^{Aa}	8.67^{Aa}	9.67^{Aa}
3	6.67^{Bc}	7.00^{Bb}	8.33^{Aa}	7.55^{Bb}	9.00^{Aa}
5	5.00^{Cc*}	6.55^{Cb}	7.00^{Bb}	6.55^{Cb}	8.50^{Ba}
7		5.75^{Dc*}	6.55^{Bb}	6.00^{Cb}	7.55^{Ca}
10			5.12^{Ca*}		5.50^{Da*}
Tenderness					
Zero	8.00^{Aa}	8.55^{Aa}	8.62^{Aa}	8.65^{Aa}	8.65^{Aa}
3	7.55^{ABa}	7.67^{Aa}	8.12^{Aa}	7.55^{Ba}	8.12^{Aa}
5	5.00^{Bb*}	6.66^{Ba}	7.00^{Ba}	6.55^{Ca}	7.00^{Ba}
7		5.45^{Ca*}	6.67^{Ba}	6.00^{Ca*}	6.67^{Ba}
10			5.55^{Ca*}		5.66^{Ca*}
Juiciness					
Zero	8.00^{Aa}	8.55^{Aa}	8.67^{Aa}	8.67^{Aa}	8.67^{Aa}
3	7.55^{ABa}	7.67^{Aa}	8.00^{Aa}	7.55^{Ba}	8.00^{Aa}
5	5.00^{Bb*}	6.66^{Ba}	7.00^{Ba}	6.55^{Ca}	7.00^{Ba}
7		5.55^{Cb*}	6.67^{Ba}	6.00^{Ca*}	6.67^{Ba}
10			5.67^{Ca*}		5.67^{Ca*}
Overall acceptability					
Zero	8.67^{Aa}	8.67^{Aa}	9.67^{Aa}	8.67^{Aa}	9.67^{Aa}
3	6.67^{Bc}	7.67^{Ba}	8.33^{Aa}	7.55^{Bb}	9.00^{Aa}
5	5.00^{Cd*}	6.66^{Cc}	7.55^{Bb}	6.55^{Cb}	8.55^{Ba}
7		5.55^{Dc*}	6.55^{Cb}	5.50^{Dc*}	7.00^{Ba}
10			5.55^{Da*}		5.55^{Da*}

- Onset of the organolyptic spoilage.
- Different superscripted small letters mean significant differences between different treatments in the same storage periods ($p \le 0.01$).
- Different superscripted capital letters mean significant differences between different storage periods in the same treatment ($p \le 0.01$).

Table (3): Total volatile basic-nitrogen (mg/100 g dry weight) of common carp fish fingers stored at 4 ± 1°C

Storage Period (day)	Common carp fish fingers contained				
	Control	1.5% Rosemary	3.0% Rosemary	1.5% Sage	3.0% Sage
Zero	35.40^{Ca}	34.50^{Da}	34.97^{Ea}	35.08^{Da}	34.04^{Da}
3	53.41^{Ba}	43.43^{Cb}	37.13^{Dc}	37.82^{Cc}	39.53^{Cb}
5	67.01^{Aa}*	54.52^{Bb}	46.63Cc	48.50^{Bc}	42.78^{Cd}
7		64.07^{Aa}*	54.65^{abc}	58.26^{Ab}*	50.74^{Bc}
10			62.97^{Aa}*		59.59^{Aa}*

- Onset of the organolyptic spoilage.
- Different superscripted small letters mean significant differences between different treatments in the same storage periods ($p \leq 0.01$).
- Different superscripted capital letters mean significant differences between different storage periods in the same treatment ($p \leq 0.01$).

Table (4): Trimethylamine-nitrogen (mg/100 g dry weight) of common carp fish fingers stored at 4 ±1°C.

Storage Period (day)	Common carp fish fingers contained				
	Control	1.5% Rosemary	3.0% Rosemary	1.5% Sage	3.0% Sage
Zero	3.95^{Ca}	3.61^{Dab}	3.33^{Eb}	3.48^{Db}	3.22^{Db}
3	9.07^{Ba}	5.95^{Cb}	4.22^{Dc}	5.34^{Cb}	3.54^{CDd}
5	17.42^{Aa}*	8.72^{Bb}	5.32^{Cc}	8.51^{Bb}	4.28^{Cd}
7		14.98^{Aa}*	8.41^{Bb}	15.18^{Aa}*	7.23^{Bc}
10			15.05^{Aa}*		15.25^{Aa}*

- Onset of the organolyptic spoilage.
- Different superscripted small letters mean significant differences between different treatments in the same storage periods ($p \leq 0.01$).
- Different superscripted capital letters mean significant differences between different storage periods in the same treatment ($p \leq 0.01$).

Table (5): Thiobarbituric acid (presented as OD) of common carp fish fingers stored at 4 ±1°C.

Storage Period (day)	Common carp fish fingers contained				
	Control	1.5% Rosemary	3.0% Rosemary	1.5% Sage	3.0% Sage
Zero	0.238^{Ca}	0.198^{Cb}	0.156^{Dd}	0.179^{Cc}	0.149^{Dd}
3	0.944^{Ba}	0.435^{BCb}	0.346^{CDc}	0.320^{Cc}	0.292^{Dc}
5	1.475^{Aa}*	0.785^{Bb}	0.525^{Cc}	0.736^{Bb}	0.554^{Cc}
7		1.056^{Aa}*	0.727^{Bb}	1.078^{Aa}*	0.662^{Bb}
10			1.177^{Aa}*		1.051^{Aa*}

- Onset of the organolyptic spoilage.
- Different superscripted small letters mean significant differences between different treatments in the same storage periods ($p \leq 0.01$).
- Different superscripted capital letters mean significant differences between different storage periods in the same treatment ($p \leq 0.01$).

Table (6): Acid (mg KOH/g oil) and peroxide (meq/Kg oil) values of common carp fish fingers stored at 4 ±1°C.

Storage period (day)	Common carp fish fingers contained									
	Control		1.5% Rosemary		3.0% Rosemary		1.5% Sage		3.0% Sage	
	AV	PV	AV	PV	AV	PV	AV	PV	AV	PV
Zero	2.10^{Ca}	5.75^{Ca}	2.10^{Da}	5.15^{Dab}	1.98^{Dab}	4.98^{Eb}	2.10^{Da}	5.02^{Db}	1.85^{Dab}	4.75^{Eb}
3	4.55^{Ba}	10.12^{Ba}	3.15^{Cb}	8.18^{Cb}	2.15^{Dc}	7.75^{Dbc}	2.98^{Cb}	7.92^{Cc}	2.12^{Dc}	7.25^{Dc}
5	7.18^{*Aa}	25.31^{*Aa}	4.15^{Bb}	15.12^{Bb}	3.55^{Cc}	12.82^{Cc}	4.20^{Bb}	14.75^{Bb}	3.25^{Cc}	11.95^{Cc}
7			7.12^{*Aa}	22.15^{*Aa}	5.25^{Bb}	18.72^{Bb}	7.09^{*Aa}	21.89^{*Aa}	5.12^{Bb}	18.52^{Bb}
10					7.15^{*Aa}	21.15^{*Aa}			6.95^{*Aa}	20.75^{*Aa}

- Onset of the organolyptic spoilage.
- AC, acid value; PV, peroxide value;
- Different superscripted small letters mean significant differences between different treatments in the same storage periods ($p \leq 0.01$).
- Different superscripted capital letters mean significant differences between different storage periods in the same treatment ($p \leq 0.01$).

Total mesophilic aerobic bacteria counts were low for all samples (4 log cfu/g) at the beginning of storage and this value increased slowly during cold storage of both control and treated samples. However, the increasing rate of mesophilic counts in control sample was higher than those of the carp fish fingers which containe1 either rosemary or sage. The unacceptable level of mesophilicbacteria in seafood is higher than 5 log cfu/g, (CDPH, 2000), or over 6–7 log cfu/g in freshwater fish (Turantas, 2000). Interestingly, in the present study, the number of total mesophilic aerobic bacteria remained lower than 5 log cfu/g during the cold storage of carp fish fingers (control, and treated samples) even if they organolyptically spoiled. This result could be explained by the antimicrobial effect of most ingredients specially, rosemary and sage, used in the preparation of fish fingers, where the shelf-life was extended by increasing the concentration (up to 3%) of rosemary and sage (Figure 1).

Same behaviour was recorded when pschrophilic bacteria were determined in this study (Figure 2), however, their counts were at the level of 2 log cfu/g at the beginning of storage.

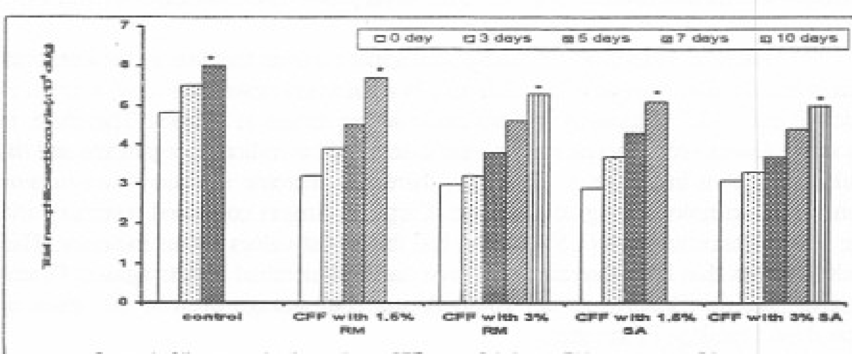

Fig. (1): Total mesophilic aerobic bacterial counts of common carp fish fingers contained rosemary or sage during cold storage (4 ± 1°C)

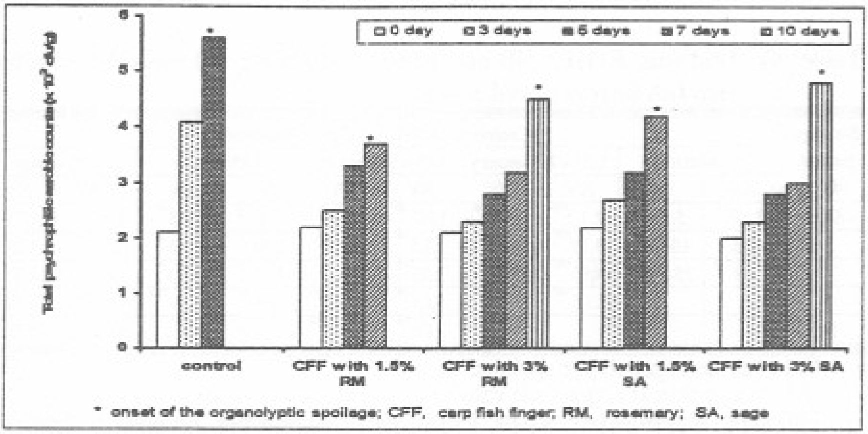

Fig. (2): Total psychrophilic aerobic bacterial counts of common carp fish fingers contained rosemary or sage during cold storage (4 ± 1°C).

The obtained results demonstrated that the number of total mesophilic and psychrophilic aerobic bacteria is not adequate to reflect the quality of chilled fish products which contained spices, especially in the early days of storage. Scott *et al.*,(1992) studied the quality changes of ice-stored smooth Oreo dory (*Pseudocyttus maculatus*) and did not estimate a significant growth of mesophilic aerobic microorganisms during the first 4 days of storage. Köse and Erdem (2004) stored anchovies at ambient and refrigerated temperatures. They estimated the counts of total mesophilic and psychrophilic aerobic bacteria counts in the samples stored at ambient temperatures. However, they preferred to analyze only psychrophilic aerobic bacteria when studying fresh fish samples stored at refrigerated temperatures and did not estimate the others. From the data obtained in the present study, it was seen that total mesophilic and psychrophilic aerobic bacteria counts did not indicate the spoilage of fish products that contained spices, where their counts remaining lower than the limit of spoilage. This conclusion is due to the inhibition effect of rosemary or sage against both mesophilic and psychrophilic bacteria. The inhibition effect could be explained by the high phenol content of both rosemary and sage and by the highly electronegative nature of aldehyde and carbonyl moieties afford bacteriostatic characteristics as described by Dorman and Deans (2000) and Dorman *et al.*, (2003).

Enterobacteriaceae is a family of microorganisms that live in the intestines of man and animals. Included in this family is a bacteria known as *E .coli*, a strain of which, *E.coli 0157*, is harmful and can cause severe illness in humans. Therefore, in this study, *Enterobacteriaceae* counts were determined as indicator organisms and the results are shown in Figure 3. Generally, there was increase in *Enterobacteriaceae* counts of all samples during cold storage. Carp fish fingers contained rosemary and sage at both concentrations (1.5% or 3%) had the lowest values of this indicator. This result indicates that both rosemary and sage had antimicrobial effects against Gram-negative bacteria (Santoyo *et al.*, 2005) during cold storage even at the onset of organolyptic spoilage of samples.

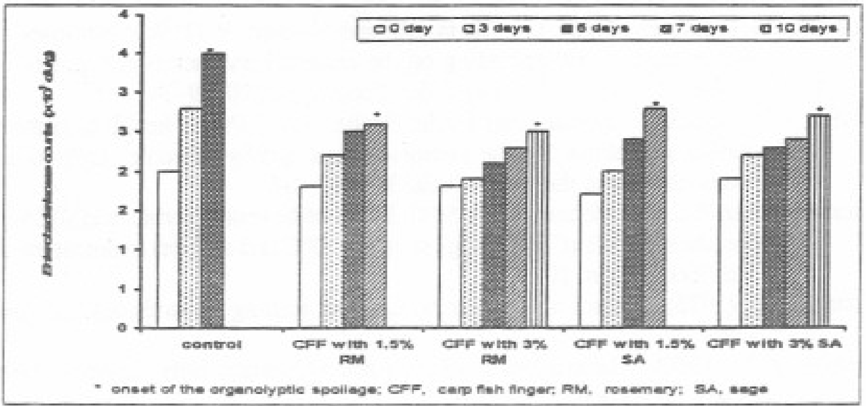

Fig. (3): *Enterobacteriaceae* counts of common carp fish fingers contained rosemary or sage during cold storage (4 ± 1°C).

CONCLUSION

From the results obtained in the present study it can be concluded that rosemary or sage can be incorporated in common carp fish fingers at level of 3% to improve its shelf-life at 4 ±1°C. The traditional methods that measure primary changes such as acid value and peroxide values and those that measure secondary changes such as TMA-N and TBA tests, could be applied for determination of the quality of common carp fish fingers supplemented with rosemary or sage. However, the relationship between the organolyptic spoilage, on one hand, of carp fish fingers, and TVB-N and the microbiological criteria, on the other hand, was not demonstrated in this study. This may be due to the antimicrobial effect of the ingredients added to the carp fish finger mixture, which inhibit the microbial growth during storage. At the same time, other deterioration factors such autolysis enzymes may be responsible to the organolyptic spoilage. Also, TMA-N exhibits a great difference between treatments and seems to be a better index of spoilage for fish products processed from the freshwater such as common carp fish. This conclusion is in accordance with those stated by Scherer *et al.,* (2006).

REFERENCES

A.O.A.C., (2000): *Official Methods of Analysis of AOAC International*, (17th ed.). Gaithersburg, Maryland, U.S.A: AOAC International.

Baumgard, J. (1986): Lebensmittel tierischer Herkunft, Feinkosterzeugnisse, gefrorene, tiefgefrorene und etrocknete lebensmittel, Fertiggerichte, hitzekonservierte Lebensmittel, Speiseeis, Zucker, Kakao, Zuckerwaren, Rohmassen. In *Mikrobiologische Untersuchung von Lebensmitteln*, (J. Baumgart, M.J. Firnhaber and G. Spicher, eds.) Behr's Verlag, Hamburg, Germany.

Bicchi, C.; Binello A. and Rubinolo, P. (2000): Determination of phenolic diterpene antioxidants in rosemary (Rosmarinus officinalis L.) with different methods of extraction and analysis. Phytochem. Anal. 11: 236 - 242.

Castrillon, A., Alvarez-Pontes, E., Garcıa, M., and Navarro, P. (1996): Influence of frozen storage and defrosting on the chemical and nutritional quality of sardine (*Clupea pilchardus*). J. Sci. Food Agric., 70, 29–34.

CDPH; Communicable Diseases and Public Health (2000) Public health laboratory service guidelines for the microbiological quality of some ready-to-eat foods sampled at the point of sale. 3: 163 – 167.

Chandrasekhar, T.C. and Mohite, R.R. (1978): Effect of fat coated sorbic acid (FCSA) and the shelf-life of fish susage stored at 10°C and ambient temperature. J. Sea Food Export, 10: 19-23.

Connell, J. J. (1975): Control of fish quality. London: Fishing News Books Ltd., pp. 137.

Conway, E.J., (1950): Microdiffusion analysis and volumetric error. London, UK: Cosby Lochwood.

Daraweash, B.M. (1996): Chemical composition of kobebah carp and bolti fish, Egypt J. Food Sci. 24, 345-358.

Dorman H. J. and Deans S. G (2000): Antimicrobial agents from plants: Antibacterial activity of plant volatile oils. J. Appl. Microbiol. 88: 308-316.

Dorman, H.J.D., Peltoketo, A., Hiltunen, R., and Tikkanen, M.J. (2003): Characterisation of the antioxidant properties of deodorised aqueous extracts from selected *Lamiaceae* herbs. Food Chemistry, 83, 255–262.

Estevez, M., Ramırez, R, Ventanas, S., and Cava, R. (2007): Sage and rosemary essential oils versus BHT for the inhibition of lipid oxidative reactions in liver pate. LWT 40 58–65

Formanek, Z., Lynch, A., Galvin, K., Farkas, J., and Kerry, J. P. (2003): Combined effects of irradiation and the use of natural antioxidants on the shelf life stability of over wrapped minced beef. Meat Sci., 63(4), 433–440.

Giménez, B.; Roncalés P. and Beltrán, J.A. (2005):The effects of natural antioxidants and lighting conditions on the quality of salmon (*Salmo salar*) fillets packaged in modified atmosphere. J. Sci. Food Agric. 85: 1033 - 1040.

Harold, E.; Ronald S.K. and Roland S. (1987): Pearson s chemical analysis of foods. 8th Ed. Longman House, Burnt, M., Harbow, Essex CM 202 JE, England.

Hirasa, K. and Takemasa, M. (1998): Antimicrobial and antioxidant properties of spices. In: Hirasa, K., Takemasa, M. (Eds.), Spice Science and Technology. Marcel Dekker Inc., New York, pp. 163–200.

Ibanez, E., Kubatova, A., Senorans, F. J., Cavero, S., Reglero, G., and Hawthorne, S. B. (2003): Subcritical water extraction of antioxidant compounds from rosemary plants. J. of Agric. Food Chem., 51, 375–382.

ICMSF "International Commission on Microbiological Specification for Foods" (1986): Microorganisms in foods. 1. Their significance and methods of enumeration (2nd ed.). Elliot, R.P. Buffalo, N.Y., University of Toronto Press.

Köse, S. and Erdem, M.E. (2004): An investigation of quality changes in anchovy (*Engraulis encrasicolus* L, 1758) stored at different temperatures. Turk. J. Vet. Anim. Sci. *28*, 575–582.

Lakshmanan, P. T. (2000): Fish spoilage and quality assessment. In T. S. G. Iyer, M. K. Kandoran, Mary Thomas, & P. T. Mathew (Eds.), Quality assurance in seafood processing (pp. 26–40). Cochin: Society of Fisheries Technologists (India).

Larmond, E., (1974): Method of sensory evaluation of foods. Canada Dept. of Agric. Ottawa KiAOC, 7.

Mackie, I. (1993): The effects of freezing on flesh proteins. Food Reviews International, 9, 575–610.

Manju, S., Jose, L., Srinivasa Gopal T.K., Ravishankar, C.N. and Lalitha, K.V. (2007) Effects of sodium acetate dip treatment and vacuum-packaging on chemical, microbiological, textural and sensory changes of Pearlspot (*Etroplus suratensis*) during chill storage. Food Chem. 102: 27–35

Miyake, T., and Shibamoto, T. (1997): Antioxidative activities of natural compounds found in plants. J. Agric. Food Chem., 45, 1819–1822.

Montero, P. Gimenez, B. Perez-Mateos, M. Go´mez-Guillen, M.C. (2005) Oxidation stability of muscle with quercetin and rosemary during thermal and high-pressure gelation. Food Chem. 93 17–23

Nwinuka, N. M., Ibeh, G. O. and Ekeke, G. I. (2005) Proximate Composition And Levels Of Some Toxicants In Four Commonly Consumed Spices. J. Appl. Sci. Environ. Manag. 9: 150-155.

Richheimer, S. L., Bailey, D. T., Bernart, M. W., Kent, M., Viniski, J. V., and Andersen, L. D. (1999): Antioxidant activity and oxidative degradation of phenolic compounds isolated from rosemary. Recent Research Development in Oil Chemistry. 3: 45–48.

Ruiz-Capillas, C., & Moral, A. (2001): Correlation between biochemical and sensory quality indices in hake stored in ice. Food Research International. 34: 441–447.

Santoyo, S., Cavero,S., Jaime, L., Ibanez, E., Senorans, F.J. and Reglero, G. (2005): Chemical composition and antimicrobial activity of *rosmarinus officinalis* L. essential oil obtained via supercritical fluid extraction. J. Food Prot. 68: 790-795.

SAS program (1996): SAS/STAT user's guide relase 6.12 ed. Cary, NC, USA: SAS Inst. Inc.

Scott, D.N., Fletcher, G.C., Charles, J. C., Wong, R. J. (1992): Spoilage changes in the deep water fish, smooth oreo dory during storage in ice Int. J. Food Sci. Technol. 27: 577–587.

Sebranek, J. G., Sewalt, V. J. H., Robbins, K . L., and Houser, T. A. (2005): Comparison of a natural rosemary extract and BHA/BHT for relative antioxidant effectiveness in pork sausage. Meat Sci. 69: 289–296.

Shahidi, F., and Wanasundara, P. K. J. P. D. (1992): Phenolic antioxidants. Crit. Rev. Food Sci. Nutr., 32: 67–103.

Sikorski, Z., and Kolakowska, A. (1994): Changes in protein in frozen stored fish. In Sikorski, Z. Sun Pan B., and Shahidi F. (Eds.), Seafood proteins (pp. 99–112). New York, USA: Chapman & Hall.

Sikorski, Z.E., Kolakowska, A. and Burt, J.R. (1990): Postharvest biochemical and microbiological changes. In *Seafood: Resources Nutritional Composition and Preservation* (Z.E. Sikorski, ed.) pp. 55–76, CRC Press Inc., Boca Raton, FL.

Srinivasan, S., and Hultin, H. (1995): Hydroxil radical modification of fish mucle protein. J. Food Biochem. 18: 405–425.

Turantas, F. (2000): Mikrobiyolojik kriterler. In: Gida Mikrobiyolojisi (Ünlütürk A. and Turantas, F. eds.) pp. 517–549, Mengi Tan Basimevi, Izmir, Turkey. (Abs.).

Yutaka, Y., Masataka S.,and Hiroshi O. (2006): Antimicrobial effect of spices and herbs on Vibrio parahaemolyticus. Int. J. Food Microbiol. 111, 6–11.

Zheng, W. and Wang, S.Y. (2001): antioxidant activity and phenolic compounds in selected herbs. J. Agric. Food Chem. 49:5165-70.

استخدام حصي البان والمريمية لإطالة فترة صلاحية أصابع سمك المبروك خلال التخزين بالتبريد

محمد مصطفي عبد الرازق* ، هشام محسن الحريري* ، محمد أبو طالب**

* قسم علوم الأغذية – كلية الزراعة – جامعة عين شمس – شبرا الخيمة – مصر

** معمل تكنولوجيا تصنيع الأسماك– المعهد القومي لعلوم البحار والمصايد– مصر

تم إنتاج أصابع سمك المبروك المحتوي علي ١,٥ أو ٣,٠ % من نباتات حصاالبان والمريمية. قدرت خصائص الجودة المختلفة وفترة الصلاحية لأصابع سمك المبروك المنتجة وذلك خلال التخزين علي درجة حرارة ٤ ± ١ °م وذلك من خلال تتبع الخصائص الحسية مثل (المظهر العام – اللون – الرائحة – الطعم – الطراوة – العصيرية – القبول العام)، بعض التقديرات الكيميائية مثل (المواد النيتروجينية المتطايرة الكلية – ثلاثي مثيل الأمين – حمض الثيوباربتيوريك – رقم الحامض – رقم البيروكسيد)، بعض التقديرات الميكروبيولوجية مثل (العد الكلي للبكتيريا المحبة لدرجة الحرارة المتوسطة – العد الكلي للبكتيريا المحبة لدرجة الحرارة المنخفضة – العد الكلي للبكتيريا المعوية). اظهرت النتائج أن جميع المقاييس التي تم تتبعها للمنتج ذات علاقة معنوية مع فترة التخزين. وبالرغم من أن قيمة المواد النيتروجينية المتطايرة الكلية كانت أعلي من ٤٠ مللجرام /١٠٠ جرام الا أن أصابع السمك لاقت قبولا حسيا. أظهرت نتائج قياس ثلاثي مثيل الأمين إختلافات كبيرة بين المعاملات المختلفة واتضح أنة يمكن الاعتماد علية في تقييم مدي فساد منتجات الأسماك المنتجة من أسماك المياه العذبة مثل سمك المبروك. أوضحت نتائج التقديرات الميكروبيولوجية حدوث تثبيط بصورة معنوية نتيجة استعمال النباتات سابقة الذكر، حيث أن كل من العد الكلي للبكتيريا المحبة لدرجة الحرارة المتوسطة و البكتيريا المحبة لدرجة الحرارة المنخفضة لم يزد عن لوغاريتم العدد ٤ وحده مكونة للمستعمرات /جرام و لوغاريتم العدد ٢ وحده مكونة للمستعمرات /جرام علي الترتيب أثناء فترة التخزين علي درجة حرارة ٤ ± ١ °م .

أوضحت النتائج المتحصل عليها إمكانية إستعمال كل من حصاالبان والمريمية بنسبة ٣,٠% لتأخير ظهور الفساد الحسى لأصابع سمك المبروك الى اليوم العاشر مقارنتا بالعينه الكنترول والتي فسدت فى اليوم الخامس من التخزين على درجة حرارة ٤ ± ١ °م.