Iranian Journal of Fisheries Sciences 19(2) 994-1005 2020
DOI: 10.22092/ijfs.2019.118664.0

Short communication:

Influence of encapsulated pomegranate peel extract on the chemical and microbial quality of silver carp (*Hypophthalmichthys molitrix* Val. 1844) fillet during refrigerating storage

Ganjian S.[1]; Javadian S.R.[2*]; Keshavarz M.[2]

Received: April 2015 Accepted: December 2018

1-Student of food science, Damghan Branch, Islamic Azad University, Damghan, Iran
2-Department of Fisheries, Qaemshahr Branch, Islamic Azad University, Qaemshahr, Iran
*Corresponding author Email: ro.javadian@gmail.com

Keywords: Silver carp, Pomegranate, Encapsulation, Extract, Fish preservation

Introduction

Fish meat plays an important role in supplement of human protein necessity (Rahimzade *et al.,* 2019)**.**

Fish products deteriorate rapidly as a result of high water activity, neutral pH, relatively large quantities of free amino acids, presence of autolytic enzymes and high percent of unsaturated fatty acids (Duan *et al.,* 2010). This problem and the increasing request for high quality fresh seafood has intensified the search for technologies that favor fresh fish preservation. Different methods have been used for extending fish products shelf life (Rostamzad *et al.,* 2010).

One of the most commonly used methods for fish preservation is cold storage. Nevertheless, it does not sufficiently prohibit the quality deterioration of fish (Jeon *et al.,* 2002) but it can be improved using antimicrobial and antioxidant compounds.

Moreover, there is an increasing demand for natural antimicrobial and antioxidant preservatives because of the concern about safety of synthetic materials due to possible carcinogenic effects (Ozogul *et al.,* 2010). During last decade, significant interest has been focused on the natural preservatives like plants extract and essential oil as an alternative to synthetic materials. Thus, the determination of the antioxidant and antimicrobial capacity of spices and their derivate in foods is being given greater importance by researchers and those involved in the agro-food industry (Viuda-Martos *et al.,* 2010). Among these, pomegranate has obtained popularity in recent years due to its multi-functionality and nutritional benefits in the human diet. Besides pomegranate fruit potential for reducing disease risk (Aviram *et al*., 2000), its peel which constitutes about 50% of the total fruit weight (Dahham *et al*., 2010), and it is often discarded as by-products contain higher amounts of polyphenol compounds than the fruit juice, and it shows stronger biological activities (Guleria and Kumar, 2006). In many studies it has been reported that pomegranate peel extracts possess a wide range of biological actions including anti-cancer properties (Kulkarni *et al*., 2005), antimicrobial properties (Al-Zoreky, 2009), apoptotic and anti-genotoxic activities (Kirilenko *et al*., 1978), anti-tyrosinase activity (De *et al*., 1999), anti-inflammatory and anti-diabetic properties (Dahham *et al*., 2010; Shuhua *et al.*, 2010). Biological activity of the fruit peel extracts is mainly related to their polyphenol compounds such as ellagic tannins, flavonols, anthocyanins, catechin, procyanidins, ellagic acid and gallic acid (Dahham *et al*., 2010; Fazeli *et al*. 2011). Several studies have also reported the efficacy of pomegranate juice, seed and peel extracts in meat-based products preservation (Naveena *et al*., 2008; Devatkal *et al*., 2010; Kanatt *et al*., 2010; Topuz *et al*., 2015; Vaithiyanathan *et al*., 2011; Zarei *et al*., 2015).

However, unfortunately, most natural active compounds are biologically instable, poorly soluble in water and they distribute poorly in the target sites. In recent years, some novel strategies have been introduced in order to improve their stability and their bioavailability, among which is the use of liposomal encapsulation (Shoji and Nakashima, 2004). Encapsulation decreases reactivity with the environment (water, oxygen, light), reduces the evaporation or the transfer rate of the active compounds to the outside environment. It also promotes their handling ability, the bioavailability and half-life of the compound (Fang and Bhandari, 2010), masks their unpleasant taste and increase dilution to achieve a uniform distribution in the food products when used in a very small amounts (Liolios *et al.*, 2009). Some studies (Gortzi *et al.*, 2007) have also showed that encapsulation can improve antimicrobial activity of compounds and maintain the stability of antimicrobials over prolonged periods of time.

Thus, the present study was aimed to investigate the effects encapsulated and un-encapsulated pomegranate peel extract on the quality of silver carp fillets and the possible efficacy of liposomal encapsulation in the improvement of its antimicrobial and antioxidant activities during the preservation of the fillets at 4 ˚C.

Materials and methods

Materials

Pomegranate fruits were purchased from a local market. Fennel has also purchased from local market. All other chemicals were analytical grade and purchased from Merck Co., Germany.

Preparation of pomegranate peel extract and liposomes

The pomegranates were manually peeled and the peels immediately have been dried at 60 ˚C for 24 h. The dried peels were powdered and were extracted with 100 ml of methanol for 24 h according to Paari *et al*. (2011). The extracts having filtered, and were re-extracted with same solvent and concentrated under reduced pressure through rotator evaporator. Liposome has obtained from Sigma-Aldrich Chemical Co., USA.

Liposome, as a carrier for pomegranate peel extract, has been produced according to the method described by Gortzi *et al*. (2006) with some modifications. Liposome mixture has dissolved in chloroform/methanol (3/1) in a round bottom flask and the organic solvent has removed by a rotary evaporator until a thin film layer has formed on the walls. Pomegranate peel extract was also dissolved in dichloromethane/methanol (2/1) and mixed with liposome mixture (4/1 ratio, liposome/extract) and the solvents have evaporated under nitrogen steam. The produced lipid film was dissolved in 2 ml of phosphate buffer (10 mM, pH 7) and vortexed for 15 min at 35 ˚C. The obtained suspension was allowed to hydrate for 2 h in the dark at room temperature and then centrifuged at 6500 rpm at 4 ˚C. Finally, multi-lamellar lipid vesicles were obtained by freeze-drying. The freeze-drying process was as follows: (1) freezing at −50 °C for 8 h; (2) primary drying −50 ^{oC} for 48 h; and (3) secondary drying at 25 °C for 24 h.

Treatment of silver carp fillets by fennel extract

36 live silver carps with an average weight of 1000±100 g were purchased from a local aquaculture farm. They were transported to the laboratory within an hour in sealed foamed polystyrene boxes containing flaked ice. Then, the fish were gutted, skinned, filleted (100±10 g), and washed up by tap water in a laboratory. Furthermore, a ranking test previously carried out comparing fish samples with encapsulated and un-encapsulated pure pomegranate peel extract at different concentrations showed significantly lower acceptability of the samples incorporating 1.25 or 1.5% encapsulated and un-encapsulated pure pomegranate peel extract when compared to the rest (1% or lower) (data not shown). After these results, the encapsulated pure pomegranate peel extract concentration of 0.5% and 1% were chosen as optimal for the following study of fish preservation. 15 fillets from each treatment were randomly-selected-and divided to one of five treatments as presented in the following:

C: control, without treatment

PPE 0.3: treatment with 0.5% pure pomegranate peel extract

F 0.5: treatment with 1% pure pomegranate peel extract

FE 0.3: treatment with 0.5% encapsulated pomegranate peel extract

FE 0.5: treatment with 1% encapsulated pomegranate peel extract

Different concentrations of pomegranate peel extracts having been sprayed on the fillets by syringe. After packaging all samples in polyethylene dishes with cellophane blanket, they were stored at 4±1 ºC for subsequent quality assessment. Chemical and microbiological analyses were performed at 3-day intervals to determine the overall quality of the fish for 15 days.

Chemical analysis

The total volatile basic nitrogen (TVB-N)

TVB-N of the silver carp samples were measured by the micro-diffusion method as described by Goulas and Kontominas (2005). The values were reported in mgN $100g^{-1}$ of fish. Measurements were repeated three times for studying repeatability.

Evaluation of lipid oxidation

The colorimetric method described by Kirk and Sawyer (1991) has used to measure the thiobarbituric acid (TBA) value in fish fillets for secondary lipid oxidation products evaluation. All measurements were repeated three times as mentioned above.

Microbiological analysis

The pour plate method was used to determine total viable count (TVC) and total psychrotrophic count (TPC). 10 g of the fish minced sample was aseptically taken and homogenized in 90 ml of sterile 85% NaCl solution with a blender (HBM-400B, HBM Biomed, Tianjin, China) at room temperature. Appropriate dilutions were serially prepared and then 1 ml of each was spread onto plate count agar media (Merck, Darmstadt, Germany). The prepared plates were incubated at 37 ºC for 2 days for TVC and at 10 ºC for 7 days for TPC. All counts were expressed as log colony-forming units (CFU) g^{-1} and performed in triplicate.

Statistical analysis

The differences among all measurements were evaluated by one-way analysis of variance (ANOVA). Duncan's multiple range tests were used to compare the means to identify which groups were significantly different from other groups. Significance was defined at $p<0.05$. All data are presented as mean ±SD.

Results and discussion

Changes in total volatile basic nitrogen (TVB-N)

TVB-N is widely studied as an indicator of deterioration of fish muscles and measures the compounds composed of ammonia and primary, secondary and tertiary amines (Fan *et al.*, 2008; Abdollahi *et al*., 2014). Variations in TVB-N values for silver carp fillets are summarized in Figure 1. According to Leroi et al. (1998), fish flesh with a level of 30 mg TVB-N per 100 g is usually regarded as spoiled. The initial TVB-N value of the silver carp fillets was 10.09 mg $100g^{-1}$ which showed the good quality of the fresh samples in that, and freshwater fish muscle has 10–20 mg $100g^{-1}$ TVB-N after harvesting (Alçiçek, 2011). Figure 1 showed the value of TVB-N increased progressively with the time of storage for all fish samples. However, TVB-N content of the samples treated with PPE was significantly lower than the control during the storage period ($p<0.05$). TVB-N content of the control samples reached 31.47 mg $100g^{-1}$ while by day

12 while it was 28.20 in samples treated with 1% pomegranate encapsulated extract. The TVB-N values of the samples exceeded the maximum level by day 9 for control and by day 12 for samples treated with 0.5 and 1% PPE. Lower TVB-N content in the fillets treated with encapsulated pomegranate peel extract may be related to the antibacterial activity of the extract. Antibacterial compounds like plant extracts can reduce TVB-N production due to the decreased capacity of bacteria for oxidative deamination of non-protein nitrogen compounds or both (Banks *et al*., 1980). Dahham (2010) described the antibacterial activities of pomegranate peel extract (rind), seed extract, juice and whole fruit on the selected bacteria. According to their results, the peel extract showed the highest antimicrobial activity compared to other extracts. Opara *et al*. (2009) and Al-Zoreky *et al*. (2009) also reported appreciable antimicrobial activity for pomegranate peel extract against selected strains of bacteria and pathogenic fungi. Gokoglu *et al.* (2009) studied the effects of pomegranate sauce on the quality of marinated anchovy during refrigerated storage. Similarly, they have found lower TVB-N values for samples in pomegranate sauce samples compared to those in sunflower oil.

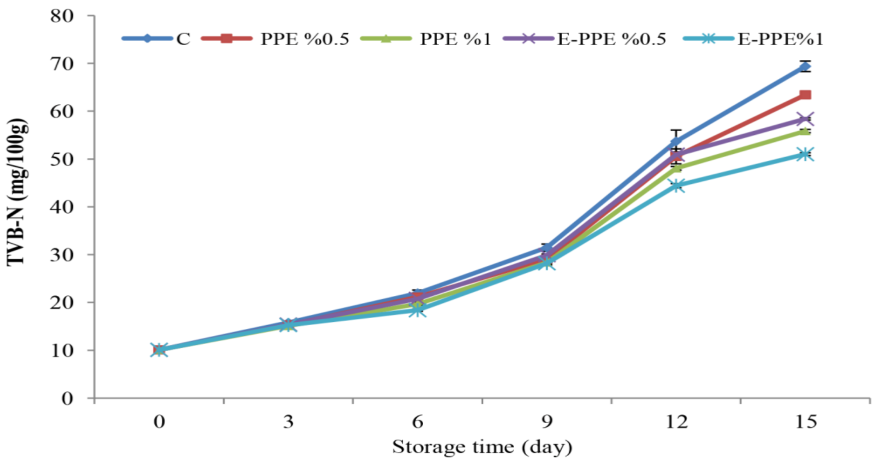

Figure 1: Changes in total volatile base nitrogen (mg N_2 $100g^{-1}$) value of silver carp fillets during storage.

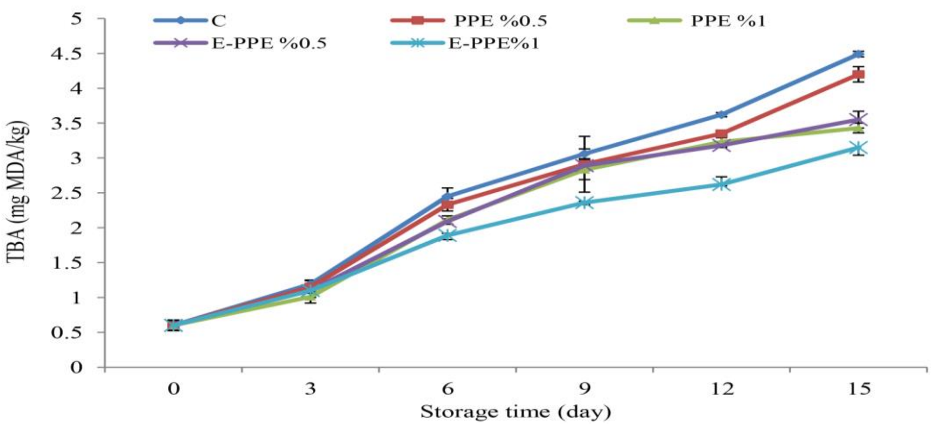

Figure 2: Changes in thiobarbituric acid (TBA) value of silver carp fillets during storage.

Table 1: Changes in total viable count (TVC) and total psychrotrophic count (TPC) of silver carp fillets during storage.

Attributes	Treatment	Storage period (days)					
		0	**3**	**6**	**9**	**12**	**15**
TVC (log_{10}cfu g^{-1})	C	4.28 ±0.07^{a}	5.27 ±0.04^{a}	7.47 ±0.03^{a}	9.67 ±0.05^{a}	10.53± 0.17^{a}	10.94 ± 0.02^{a}
	PPE 0.5%	4.28 ±0.07^{a}	5.03 ±0.06^{b}	5.87 ±0.06^{b}	6.66 ±0.09^{b}	7.83 ± 0.02^{b}	8.67 ± 0.03^{c}
	PPE 1%	4.28 ±0.07^{a}	4.91 ±0.03^{c}	5.79±0.06^{bd}	6.70 ±0.02^{b}	7.78 ± 0.04^{b}	8.77 ± 0.02^{b}
	EPPE 0.5%	4.28 ±0.07^{a}	4.94 ±0.02^{c}	5.78±0.01^{bd}	6.69 ±0.06^{b}	7.76 ± 0.03^{b}	8.68 ± 0.04^{c}
	EPPE 1%	4.28 ±0.07^{a}	4.77 ±0.05^{d}	5.68 ±0.06^{c}	6.61 ±0.02^{c}	7.66 ± 0.01^{b}	8.75 ± 0.01^{b}
TPC (log_{10}cfu g^{-1})	C	4.16 ±0.01^{a}	5.40 ±0.03^{a}	7.67 ±0.06^{a}	9.77 ±0.01^{a}	10.68 ±0.04^{b}	10.98 ± 0.0^{a}
	PPE 0.5%	4.16 ±0.01^{a}	5.27 ±0.11^{a}	5.88 ±0.01^{b}	6.64 ±0.10^{c}	7.77 ± 0.04^{b}	8.70 ± 0.04^{c}
	PPE 1%	4.16 ±0.01^{a}	5.03 ±0.06^{b}	5.87 ±0.04^{b}	6.79 ±0.01^{b}	7.80 ± 0.04^{b}	8.79 ± 0.02^{b}
	EPPE 0.5%	4.16 ±0.01^{a}	5.05 ±0.07^{b}	5.85 ±0.04^{b}	6.71 ±0.05^{b}	7.79 ± 0.02^{b}	8.71 ± 0.04^{c}
	EPPE 1%	4.16 ±0.01^{a}	5.87 ±0.04^{c}	5.84 ±0.01^{b}	6.73 ±0.02^{b}	7.69 ± 0.01^{c}	8.78 ± 0.01^{b}

^{a,b,c} Different small letters in the same column, represents significant difference ($p<0.05$).

In other hand, samples treated with liposomal encapsulated pomegranate peel extract showed significantly lower TVB-N content compared to the control and fillets treated with pure extract during the storage period ($p<0.05$). This observation may be explained by the enhanced antimicrobial activity of the extract after encapsulation or better protection of their functionality during the processing or storage period. Gortzi *et al*. (2007) also reported that after encapsulation in liposome, the antimicrobial activity of *Origanum dictamnus* extracts proved to be higher than those of the same extracts in pure form.

Lipid oxidation

Changes in thiobarbituric acid (TBA) values has been used to exhibit the degree of lipid oxidation as second stage auto-oxidation during chilled storage of silver carp fillets (Fig. 2). Presents the TBA values of different treatment groups during the storage period. As shown, the initial value of TBA was around 0.6 mg MDA kg^{-1}, close to the value reported for silver carp by Fan *et al*. (2009). The TBA value of the silver carp fillets increased through the whole storage period, especially in the control samples (reached to 4.29 mg MDA kg^{-1}) which shows secondary lipid oxidation in the samples. However, the TBA value of the samples treated with pomegranate peel extract (reached to 3.43 and 3.25 mg MDA kg^{-1} in PPE 1% and EPPE 1%, respectively) was significantly lower than the control during the storage, indicating the pomegranate peel extract could be effective in reducing lipid oxidation. Other authors have also been reported strong antioxidant properties for the ethanol extracts of pomegranate peel during in vitro studies (Negi and Jayaprakasha, 2003; Kanatt *et al*., 2010; Fazeli *et al*. 2011) which was explained by their high phenolic content. It has been well confirmed that phenolic compounds are able to donate a hydrogen atom to the free radicals, thus stopping the propagation chain reaction during lipid oxidation process (Singh *et al.* 2006). Kanatt *et al*. (2010) studied the effect of 0.1 and 0.5% pomegranate peel extract on the oxidative stability of chicken products and found significantly lower

TBA content in the samples treated with PPE. O¨zen *et al*. (2011) results showed that the formation of lipid hydro peroxides and thiobarbituric acid-reactive substances was significantly inhibited by pomegranate seed extract addition when compared with control in minced mackerel. Similarly, the addition of pomegranate peel extract to goat fish (Paari *et al*., 2011), halibut fillets (Ünalan and Korel, 2011), silver carp (Zarei *et al*., 2015) and anchovy fish oil (Topuz *et al*., 2015) could prohibit lipid oxidation and TBA formation during refrigerated storage.

Likely, samples treated with liposomal encapsulated pomegranate peel extract showed significantly lower TBA content compared to the control and fillets treated with pure extract during the storage period ($p<0.05$). This may show the potential of liposomal encapsulation to improve the antioxidant activity of the pomegranate peel extract during application on the fish fillet by prolonging its availability. As mentioned before, encapsulation decreases reactivity of bioactive compound with the environment (water, oxygen, light), reduces the evaporation or the transfer rate of the active compounds to the outside environment. It also promotes their handling ability, the bioavailability and half-life of the compound (Fang and Bhandari, 2010; Donsì *et al.,* 2011). Evidence of liposomes improving the bioactivity and bioavailability of polyphenols has been reported by a number of researchers (Fang and Bhandari, 2010). For example, Gortzi *et al*. (2007) reported higher antioxidant activity of *O. dictamnus* extracts after encapsulation in liposome.

Changes in total viable and psychrotrophic counts

The changes in total viable counts (TVC) with the storage period for the treated and untreated silver carp fillets are summarized in Table 1. The initial TVC of the samples was low (3.44 log_{10}cfu g^{-1}), indicating the high quality of fish fillets used in this study (ICMSF, 1986). TVC of all samples increased with storage time and the value of control increased faster and exceeded the maximum 10^6 log_{10}cfu g^{-1} after 6 days and reached to 10.94 Log CFU g^{-1} This acceptability limit of 10^6 CFU g^{-1} has been recommended for fresh fish (ICMSF, 1986). TVC of the fillets treated with pomegranate peel extract increased gradually and reached to 8.67, 8.77, 8.68, and 8.75 log_{10v}CFU g^{-1} for PPE 0.5, PPE 1, EPPE 0.5 and EPPE 1, respectively, at the end of storage period. As a result, all treatments significantly inhibited (about 2 Log) the growth of mesophilic bacteria in silver carp compared with the control samples during the storage period. A similar trend was also observed about psychrotrophic counts in all treatments (Table 1). The lower TVC and TPC observed in samples treated with pomegranate peel extract can be related to the antibacterial activity of the extract. Dahham (2010) described the antibacterial activities of pomegranate peel extract (rind), seed extract, juice and whole fruit on the selected bacteria. According to their results, the peel extract showed highest antimicrobial activity compared to other extracts. Opara *et al*. (2009) and Al-Zoreky *et al*. (2009) also have reported appreciable antimicrobial activity for pomegranate peel extract against selected strains of bacteria and pathogenic fungi. Our results coincide with those reported by Naveena *et al*. (2008) and Vaithiyanathan *et al*. (2011) which showed using pomegranate (*Punica granatum*) rind powder extract and fruit juice phenolic solution could inhibit the growth of microorganism in chicken patties and chicken meat, respectively, during refrigeration storage. Similar observations have been reported by Zarei *et al*. (2015) about silver carp filler treated with pomegranate peel extract combined with chitosan nanoparticles.

Furthermore, in the present study, the lowest TVC and TPC have been observed in the samples treated with encapsulated pomegranate peel extract (Table 1). The improvement of the antimicrobial activity of natural plant extracts and essential oils when encapsulated into liposomal delivery systems has also reported by others (Gortzi *et al.*, 2006; Gortzi *et al.*, 2007; Liolios *et al.*, 2009; Donsì *et al*., 2011). The encapsulation of eugenol and carvacrol into nanometric surfactant micelles also resulted in improved antimicrobial activity (Gaysinsky *et al.*, 2005).

The effects of encapsulated and un-encapsulated pomegranate peel extract on the quality of refrigerated silver carp fillet has studied. Results have showed that the extract could reduce chemical deterioration and lipid oxidation in the fillets compared to the control, as reflected with lower TVBN and TBA values. Also, pomegranate peel extract reduced TVC of the fillets about 2 Log10CFU g^{-1} compared with control. Moreover, the efficacy of the extract was improved with liposomal encapsulation.

References

Abdollahi, M., Rezaei, M. and Farzi, G., 2014. Influence of chitosan/clay functional bionanocomposite activated with rosemary essential oil on the shelf life of fresh silver carp. *International Journal of Food Science and Technology*, 49, 811–818.

Alçiçek, Z., 2011. The effects of thyme (*Thymus vulgaris* L.) oil concentration on liquid-smoked vacuum-packed rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) fillets during chilled storage. *Food Chemistry*, 128, 683-688.

Al-Zoreky, N.S., 2009. Antimicrobial activity of pomegranate (*Punica granatum* L.) fruit peels. *International Journal of Food Microbiology*, 134(**3**), 244–8.

Aviram, M., Dornfeld, L, Rosenblat M, Volkova N, Kanegative M, Coleman R, Hayek T, Presser D, Fuhrman B. 2000. Pomegranate juice consumption reduces oxidative stress, atherogenic modification to LDL and platelet aggregation: studies in human and in atherosclerotic apolipoprotein deficient mice. *American Journal of Clinical Nutrition*, 71, 1062–1076.

Banks, H., Nickelson, R. and Finne, G., 1980. Shelf life studies on carbon dioxide packaged finfish from Gulf of Mexico. *Journal of Food Science,* 45, 157–162.

Dahham, S.S., Ali, M.N., Tabassum, H. and Khan, M., 2010. Studies on Antibacterial and Antifungal Activity of Pomegranate (*Punica granatum* L.). *American-Eurasian Journal of Agricultural and Environmental Science,* 9(**3**), 273–281.

De, M., Krishna De, A. and Banerjee, A.B., 1999. Antimicrobial screening of some Indian spices. *Phytotherapy Research,* 13(**7**), 616–618.

Devatkal, S.K., Narsaiah, K. and Borah, A., 2010. Anti-oxidant effect of extracts of kinnow rind, pomegranate rind and seed powders in cooked goat meat patties. *Meat Science,* 85, 155–9.

Donsì, F., Annunziata, M., Sessa, M. and Ferrari, G., 2011. Nanoencapsulation of essential oils to enhance their antimicrobial activity in foods. *LWT-Food Science and Technology,* 44, 1908–1914.

Duan, J., Cherian, G. and Zhao, Y., 2010. Quality enhancement in fresh and frozen lingcod (*Ophiodon elongates*) fillets by employment of fish oil incorporated chitosan coatings. *Food Chemistry,* 119, 524–532.

Fan, W.J., Chi, Y.L. and Zhang, S., 2008. The use of a tea polyphenol dip to extend the shelf life of silver carp (*Hypophthalmicthys molitrix*) during storage in ice. *Food Chemistry,* 108, 148-153.

Fang, Z. and Bhandari, B., 2010. Encapsulation of polyphenols–a review. *Trends in Food Science and Technology,* 21(**10**), 510–523.

Fazeli, M.R., Bahmani, S., Jamalifar, H. and Samadi, N., 2011. Effect of probiotication on antioxidant and antibacterial activities of pomegranate juices from sour and sweet cultivars. *Natural Product Research,* 25(**3**), 288–97.

Gaysinsky, S., Davidson, P.M., Bruce, B.D. and Weiss, J., 2005. Growth inhibition of Escherichia coli O157: H7 and *Listeria monocytogenes* by carvacrol and eugenol encapsulated in surfactant micelles. *Journal of Food Protection,* 68(**12**), 2559-2566.

Gokoglu, N., Topuz, O.K. and Yerlikaya, P., 2009. Effects of pomegranate sauce on quality of marinated anchovy during refrigerated storage. *LWT - Food Science and Technology,* 42(**1**), 113-118.

Gortzi, O., Lalas, S., Tsaknis, J. and Chinou, I., 2006. Reevaluation of antimicrobial and antioxidant activity of *Thymus* spp. extracts before and after encapsulation in liposomes. *Journal of Food Protection,* 69(**12**), 2998-3005.

Gortzi, O., Lalas, S., Tsaknis, J. and Chinou, I., 2007. Enhanced bioactivity of *Citrus limon* (Lemon Greek cultivar) extracts, essential oil and isolated compounds before and after encapsulation in liposomes. *Planta Medica,* 73(**9**), 184-189.

Goulas, A.E. and Kontominas, M.G., 2005. Effect of salting and smoking-method on the keeping quality of chub mackerel (*Scomber japonicus*): Biochemical and sensory attributes. *Food Chemistry,* 93, 511–520.

Guleria, S. and Kumar, A., 2006. Antifungal activity of some Himalayan medicinal plants using direct bioautography. *Journal of Cell and Molecullar Biology,* 5, 95-98

ICMSF. 1986. Microorganisms in foods. The International Commission on Microbiological Specifications for Foods of the International Union of Biological Societies. Oxford: Blackwell Scientific Publications. pp. 181–196.

Jeon, Y.J., Kamil, J.Y.V.A. and Shahidi, F., 2002. Chitosan as an edible invisible film for quality preservation of herring and atlantic cod. *Journal of Agriculture and Food Chemistry,* 50, 5167–5178.

Kanatt, S.R., Chander, R. and Sharma, A., 2010. Antioxidant and antimicrobial activity of pomegranate peel extract improves the shelf life of chicken products. *International Journal of Food Science and Technology*, 45(**2**), 216–222.

Kirilenko, O.A., Linkevich, O.A., Suryaninova E.I. and Lysogor, T.A., 1978. Antibacterial properties of juice of various types of pomegranate. *Konservnaya I Ovoshchesushilnaya Promyschlennost,* 12, 12-18.

Kirk, R.S. and Sawyer, R., 1991. Pearson's composition and analysis of foods, 9th ed. London: Longman Scientific and Technical.page?

Kulkarni, A.P. and Aradhya, S.M., 2005. Chemical changes and antioxidant activity in pomegranate arils during fruit development. *Food Chemistry,* 93, 319-324.

Leroi, F., Joffraud, J.J., Chevalier, F. and Cardinal, M., 1998. Study of the microbial ecology of cold-smoked salmon during storage at 8 ºC. *International Journal of Food Microbiology*, 39, 1-2.

Liolios, C.C., Gortzi, O., Lalas, S., Tsaknis, J. and Chinou, I., 2009. Liposomal incorporation of carvacrol and thymol isolated from the essential oil of *Origanum dictamnus* L. and in vitro antimicrobial activity. *Food Chemistry*, 112(**1**), 77–83

Naveena, B.M., Sen, R., Vaithiyanathan, S., Babji, Y. and Kondaiah, N., 2008. Comparative efficacy of pomegranate juice, pomegranate rind powder extract and BHT as antioxidants in cooked chicken patties. *Meat Science,* 80, 1304–8.

Negi, P.S. and Jayaprakasha, G.K., 2003. Antioxidant and antibacterial activities of *Punica granatum* peel extracts. *Journal of Food Sciences,* 68, 1473–1477

Opara, L.U., Al-Ani, M.R. and Al-Shuaibi, Y.S., 2009. Physico-chemical properties, vitamin C content, and antimicrobial properties of pomegranate fruit (*Punica granatum* L.). *Food and Bioprocess Technology,* 2(**3**), 315–321.

Özen, B., Eren, M., Pala, A., Özmen, İ. and Soyer, A., 2011. Effect of plant extracts on lipid oxidation during frozen storage of minced fish muscle. *International Journal of*

Food Science and Technology, 46(**4**), 724-731.

Ozogul, Y., Ayas, D., Yazgan, H., Ozogul, F., Boga, E.K. and Ozyurt, G., 2010. The capability of rosemary extract in preventing oxidation of fish lipid. *International Journal of Food Science and Technology*, 45(**8**), 1717-1723.

Paari, A., Naidu, H.K., Kanmani, P., Satishkumar, R., Yuvaraj, N., Pattukumar, V. and Arul, V., 2011. Evaluation of irradiation and heat treatment on antioxidant properties of fruit peel extracts and its potential application during preservation of goat fish *Parupenaeus indicus*. *Food and Bioprocess Technology*, 5(**5**), 1860–1870.

Rahimzade, E., Bahri, A.H., Moini, S. and Nokhbe Zare, D., 2019. Influence of vacuum packaging and frozen storage time on fatty acids, amino acids and ω -3/ ω-6 ratio of rainbow trout (*Oncorhynchus mykiss*). *Iranian Journal of Fisheries Sciences*, 18(**4**), 1083-1092.

Rostamzad H., Shabanpour B., Kashaninejad M. and Shabani A., 2010. Inhibitory impacts of natural antioxidants (ascorbic and citric acid) and vacuum packaging on lipid oxidation in frozen Persian sturgeon fillets. *Iranian Journal of Fisheries Sciences*, 9(**2**), 279-292

Shoji, Y. and Nakashima, H., 2004. Nutraceutics and delivery systems. *Journal of Drug Targeting*, 12(**6**), 385–391.

Shuhua, Q., Hongyun, J., Yanning, Z. and Weizhi, H., 2010. Inhibitory effects of *Punica granatum* peel extracts on Botrytis cinerea. *Journal of Plant Diseases and Protection*, 36, 148-150.

Singh, G., Maurya, S., De Lampasona, M.P. and Catalan, C., 2006. Chemical constituents, antifungal and antioxidative potential of *Foeniculum vulgare* volatile oil and its acetone extract. *Food Control*, 17(**9**), 745–752.

Topuz, O.K., Yerlikaya, P., Uçak, İ., Gümüş, B., Büyükbenli, H. and Gökoğlum. 2015. Influence of pomegranate peel (*Punica granatum*) extract on lipid oxidation in anchovy fish oil under heat accelerated conditions. *Journal of Food Science and Technology*, 52(**1**), 625-632.

Ünalan, U., Dalgaard, P. and Korel, F., 2011. Effect of pomegranate (*Punica granutum*) and rosemary (*Rosmarinus officinalis* L.) extracts on shelf-life for chilled Greenland halibut (*Reinhardtius hippoglossoides*) fillets in modified atmosphere packaging at 2 ^{o}C. *International Food Congress-Novel Approaches in Food Industry*, 1, 189-196.

Vaithiyanathan, S., Naveena, B.M., Muthukumar, M., Girish, P.S. and Kondaiah, N., 2011. Effect of dipping in pomegranate (*Punica granatum*) fruit juice phenolic solution on the shelf life of chicken meat under refrigerated storage (4°C). *Meat Science*, 88, 409–14.

Viuda-Martos, M., Ruiz-Navajas, Y. and Ferna, J., 2010. Effect of adding citrus fibre washing water and rosemary essential oil on the quality characteristics of a bologna sausage. *LWT - Food Science and Technology,* 43, 958-963.

Zarei, M., Ramezani, Z., EinTavasoly, S. and Chadorbaf, M., 2015. Coating effects of orange and pomegranate peel extracts combined with chitosan nanoparticles on the quality of refrigerated silver carp fillets. *Journal of Food Processing and Preservation,* 39(**6**), 2180-2187.