



Original research

## Comparison antioxidant effect of fennel extract and BHT on the quality of minced silver carp during refrigerated storage

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### ABSTRACT

This study evaluated the ability of fennel extract to improve the quality of refrigerated minced silver carp over a period of 16 days. Minced silver carp were treated with fennel extract (0.5, 1 and 1.5% w/w), and beta-hydroxy toluene (BHT 200 ppm). The control and the treated fish samples were analyzed periodically (every 4 days) by chemical analysis (Peroxide value (PV), thiobarbituric acid (TBA) and free fatty acid (FFA)). Results showed that the fennel extract and BHT could reduce lipid oxidation in the samples compared to the control. Moreover, minced silver carp treated with 1.5% fennel extract showed the lowest amount of lipid oxidation. The results indicated that fennel extract was improving the quality characteristics of treated fresh minced fish.

Keywords: Silver carp, Fennel extract, Synthetic antioxidant, Lipid oxidation

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## 1. Introduction

An Silver carp (*Hypophthalmichthys molitrix*) is a prevalent freshwater fish and one of the most widely cultured species all over the world due to its fast growth rate, easy cultivation, high feed efficiency ratio and high nutritional value (Fan et al., 2009). However, this fresh water fish with extremely muddy flavour and many fish bones is a low market-value resource. Consequently to increase the amount of consumption, it is proposed to have various fish products of it. Today, production of minced fish which are vastly consumed could be one way of increasing fish consumption (Asgharzadeh et al., 2006; Bagheri et al., 2015).

Fish are very susceptible to both microbiological and chemical spoilage and lipid oxidation, due to large amount of polyunsaturated fatty acid and free amino acids, volatile nitrogen bases, and higher final pH (Razavi Shirazi, 2001; Ibrahim Sallam, 2007). Effective measures for extending shelf-life and improving nutritional quality of fresh fish are necessary.

Synthetic antioxidants, such as butylated hydroxyl anisole (BHA), butylated hydroxyl toluene (BHT) and tert-butyl hydroquinone (TBHQ) have been widely used as food additives to provide protection against oxidative degradation and to prolong the storage stability of foods. According to some reports, these compounds have possible toxic properties to human health and environment and can exhibit carcinogenic effects in living organisms (Ames, 1983; Baardseth, 1989). Hence, there is a

tendency towards the use of essential oil and extract from natural source replace these synthetic antioxidants (Mohledy et al., 2011).

Fennel (*Foeniculum vulgare*) is a small green brown seed belonging to the *Umbelliferae* (*Apiaceae*) family, known and used by humans since antiquity. It grows in the Mediterranean region and western Asia. Traditionally, fennel seeds are used as anti-inflammatory, analgesic, carminative, diuretic and antispasmodic agents (Muckensturm et al., 1997; Anwar et al., 2008). It has shown antimicrobial (Gulfranz et al., 2008)

Antioxidant activity in various in vivo and in vitro experiments (Barros et al., 2009; El Ouariachi et al., 2014), and in the some studies its ability in fish preservation has been reported (Bagheri et al., 2015; Alipour et al., 2015).

Thus, this study was aimed to investigate the effect of fennel extract on the quality of minced silver carp and to compare them with commercially available antioxidants (BHT) during storage at refrigeration condition ( $4 \pm 1^\circ\text{C}$ ).

## 2. Material and Methods

### 2.1. Materials

The plant fennel (*Foeniculum vulgare*) was purchased from a local market and authenticated by the Medical Plant Incubator Center (Islamic Azad University, Ayatollah Amoli Branch, Amol, Iran). They were washed and air dried in the shade and change to

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the powder using an electric device and stored at refrigerator (4 °C) until use.

BHT prepared from Sigma-Aldrich (St. Louis, MO, USA). All other chemicals were analytical grade and purchased from Merck Co., Frankfurt, Germany.

## 2.2. Preparation of fennel extract

Ethanol was added to powdered fennel in ratio of 1: 10 and the resulting mixture was shaken overnight to extract fennel's phenol compound. After 24 h, the extract was filtered through Whatman No. 42 filter paper to separate fennel particles. Ethanol was completely evaporated in an oven at 40°C. Finally, they were placed in a refrigerator (Bagheri et al. 2015).

## 2.3. Fish sample preparation and storage condition

Thirty-six live silver carp with an average weight of 1000 ± 100 g were purchased from a local aquaculture farm. In 1 h, they were transported to the laboratory in sealed foamed polystyrene boxes containing flaked ice. Then, the fishes were washed by hand and minced twice, and then minced fish were randomly assigned into five batches (100±10 g minced fish in each group) as presented in the following:

C: control, without treatment

FE 0.5%: treatment with 0.5% fennel extract

FE 1%: treatment with 1% fennel extract

FE 1.5%: treatment with 1.5% fennel extract

BHT 200: treatment with 200 ppm BHT (at its legal limit)

After packaging all samples in polyethylene dishes with cellophane blanket, they were stored at 4 ± 1 °C for subsequent quality assessment. Chemical analyses were performed at 4 day intervals to determine the overall quality of the minces.

## 2.4. Chemical analysis

The peroxide value (PV) was determined in the total lipid extracts according to the method of Pearson (Egan et al., 1997). Results were expressed in meq oxygen/lipid. The thiobarbituric acid (TBA) value was determined colorimetrically by the method as described by Kirk and Sawyer (1991) and expressed as mg malondialdehyde/kg sample. Free fatty acid (FFA) was estimated by the procedure explained by AOAC (2005) and its content was expressed as percentage of oleic acid.

## 2.5. Statistical analysis

All measurements were replicated three times for each lot and mean values ± standard deviations were reported for each case. One-way ANOVA was used and mean comparison was performed by Duncans' new multiple range test. Statistical analysis was prepared using the SPSS statistical software, (release 18.0) for Windows (SPSS Inc. Chicago, IL). Significant differences were considered at the 95% confidence level ( $P < 0.05$ ).

# 3. Results and Discussion

## 3.1. Peroxide value

Lipid in the fish is susceptible to oxidation and PV measures the amount of hydroperoxides formed i.e., the primary lipid oxidation products in fish muscle. Nevertheless, their decay leads to the formation of a wide range of carbonyl compounds, hydrocarbons, furans and other products that contribute to the rancid taste of decaying food (Yanishlieva & Marinova, 2001). The PV values of treatments during the refrigerated storage are shown in Fig 1. The PV values of samples significantly increased ( $p < 0.05$ ) during storage time, until day 8, and then decreased gradually until the end of storage time. After the maximum value was reached, a decrease in PV values was observed. Increasing in PV shows lipid oxidation and formation of hydroperoxides with a rate higher than that of their decomposition and decrease in it after reaching the maximum value may result from the lower availability of substrate and instability of peroxide molecules, which leads to the formation at the rate of decomposition (Pereira de Abreu et al., 2011; Abdollahi et al., 2014).

Moreover, the PV in the treated samples with fennel extracts and BHT were significantly ( $P < 0.05$ ) lower than the control during the storage. These results could be attributed to the antioxidant activity of fennel extract and BHT. Moreover the results indicated that lower PV could be due to high phenolic content present in the extracts. It has been well confirmed that phenolic contents are able to donate a hydrogen atom to the free radicals thus stopping the propagation chain reaction during lipid oxidation process (Rathore et al., 2011).

However, the lowest PV during the storage period was observed in the 1.5% FE samples, which indicated that a high concentration of fennel extract led to a reduction of the PV value. It has been reported that a higher concentration of extract may act as a better pro-oxidant (Sarah et al., 2010; Bagheri et al., 2015).

## 3.2. Thiobarbituric acid (TBA)

Lipid Shelf life of fish is limited because of the oxidation of lipid (Ozogul et al., 2005). TBA value has been widely used to evaluate of degree of secondary lipid oxidation (Nishimoto et al., 1985), and the presence of TBA reactive substances is due to the second stage auto-oxidation during which peroxides are oxidized to aldehyde and ketone (Lindsay, 1991). In the present study, changes in TBA values of different treatment groups were shown in Fig 2. The TBA values of all treatment increased during storage. This increase may be attributed to the partial dehydration of fish and the increased oxidation of unsaturated fatty acids (Song et al., 2011). According to Connell (1990), a TBA value of 2 mg MDA/kg of fish sample is regarded as the limit of acceptability. The initial TBA value of the fish samples was 0.63 mg MDA/kg and this value increased to 2.87, 1.78, 1.39, 1.06 and 1.78 mg MDA/kg at the end of storage for the Control, 0.5% FE, 1% FE, 1.5% FE and 200 ppm BHT, so only control samples exceeded the maximal permissible limit. The lowest TBA during the storage period was observed in the 1.5% FE samples. This indicated that fennel extract may have retarded lipid oxidation. This property can be due to the phenolic content of fennel extract. This observation was in agreement with what reported by Alipour et al. (2015) about the antioxidant properties of fennel extract on the TBA of silver carp fish.

Hasani and Javadian (2015) compared antioxidative activities among ethanol extract of bitter orange peel extract and BHT on common carp by measuring both primary and secondary oxidation. Their results showed that, higher concentrations of the extract made more decrease in TBAS and PV levels than the BHT during whole of storage period. Their results are in agreements with our results.

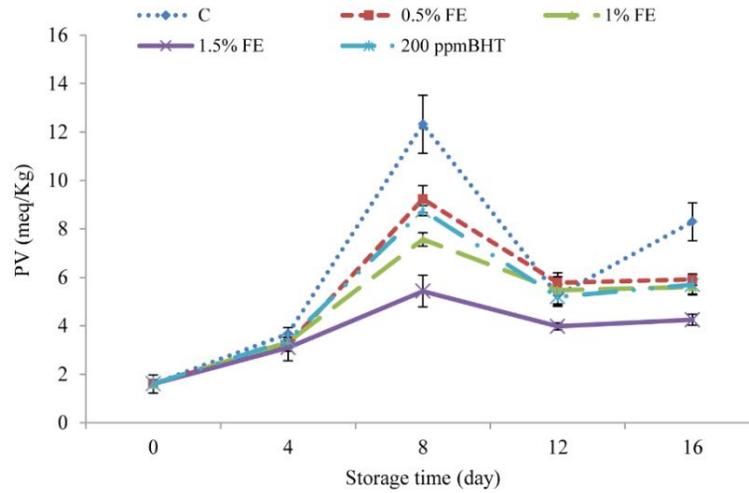


Fig. 1. Change in peroxide value (PV) of minced silver carp during refrigerator storage. (C: control, without treatment, FE 0.5%: treatment with 0.5% fennel extract, FE 1%: treatment with 1% fennel extract, FE 1.5%: treatment with 1.5% fennel extract and BHT 200: treatment with 200 ppm BHT).

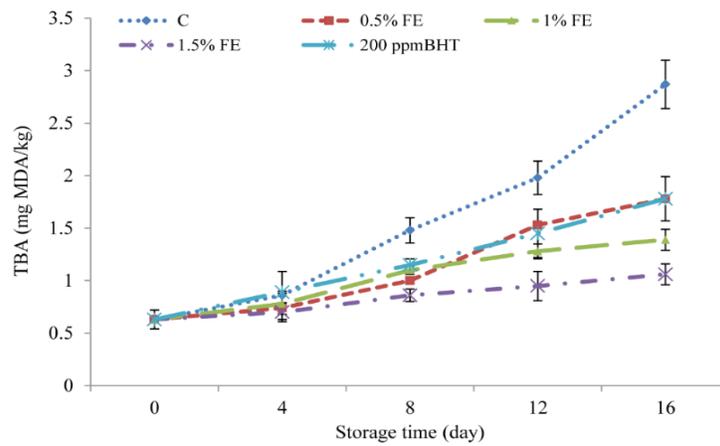


Fig. 2. Change in thiobarbituric acid (TBA) of minced silver carp during refrigerator storage. (C: control, without treatment, FE 0.5%: treatment with 0.5% fennel extract, FE 1%: treatment with 1% fennel extract, FE 1.5%: treatment with 1.5% fennel extract and BHT 200: treatment with 200 ppm BHT).

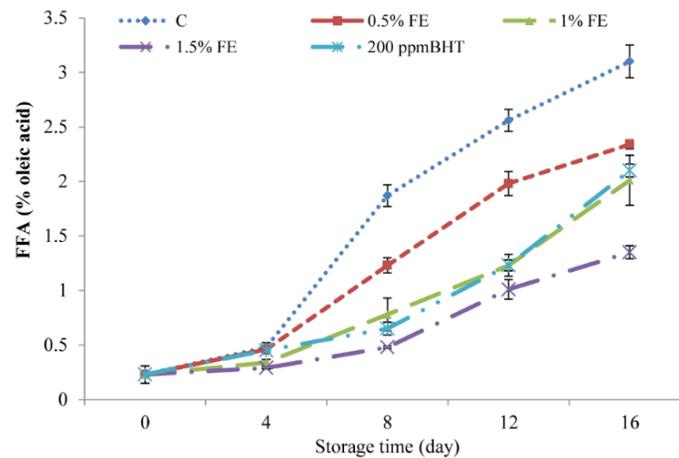


Fig. 3. Change in free fatty acid (FFA) of minced silver carp during refrigerator storage (C: control, without treatment, FE 0.5%: treatment with 0.5% fennel extract, FE 1%: treatment with 1% fennel extract, FE 1.5%: treatment with 1.5% fennel extract and BHT 200: treatment with 200 ppm BHT).

### 3.3. Free fatty acid (FFA)

The presence of FFAs is caused by the hydrolysis of lipids. Triglyceride in the depot fat is cleaved by triglyceride lipase originating from the digestive tract or excreted by certain microorganisms (Viji et al., 2015). FFA value of various samples during refrigerator storage is depicted in Fig. 3. The FFA values of all treatment increased during storage. Increasing in FFA displays hydrolytic oxidation in the samples caused by internal or bacterial enzymes and the decrease might be related to the interaction of triacylglyceride products with proteins (Pereira de Abreu et al., 2011).

FFA of fresh fish was approximately 0.23 % oleic acid sample, until it reached a maximum (Control= 3.1, 0.5% FE= 2.34, 1% FE= 2.01, 1.5% FE= 1.35 and 200 ppm BHT= 2.1) on the day 16.

## 4. Conclusion

Generally, results of the present study apparently indicated that fennel extract exhibited strong antioxidant effect on the quality of minced silver carp during refrigerator storage which was almost more effective than the antioxidant activity of synthetic antioxidants (BHT). Therefore, the fennel extract (at 1.5% concentrations) can be promising alternatives to reduce synthetic materials in food formulation and moreover it can use as the natural products of potent preservative in fish preservation.

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