Effect of plant extracts on lipid and protein oxidation of mackerel (*Scomber scombrus*) mince during frozen storage

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Abstract The effects of different plant extracts [green tea extract (GTE), grape seed extract (GSE), and pomegranate rind extract (PRE)] at a level of 100 ppm equivalent phenolics and butylated hydroxytoluene (BHT) on the changes in quality of fish (*Scomber scombrus*) mince during 6 months frozen storage at -18 ± 1 °C were investigated. During storage, significant oxidative reactions in both the lipids and proteins were observed with the increase in thiobarbituric acid reactive substances (TBARS) and carbonyls and decrease in sulphhydryl groups and protein solubility. BHT and PRE effectively inhibited lipid oxidation as lower peroxide and TBARS values were observed. Moreover, antioxidants added to minced fish significantly reduced protein oxidation compared to control without any antioxidant. The minced fish containing PRE had lower carbonyl and higher sulphhydryl contents, but no significant differences for carbonyl and sulphhydryl contents were observed among antioxidant sources. Protein solubility decreased with increase in storage period. The loss of protein solubility was higher in control samples than in antioxidant treated ones. Among antioxidant sources, PRE was an excellent antioxidant toward both lipid and protein oxidations. Therefore, it could be a potential source of natural antioxidants in minced fish during frozen storage.

Keywords Green tea · Grape seed · Pomegranate rind · Oxidation · Frozen storage · Mackerel

Introduction

Oxidative reactions are the foremost factor for the consumer preference in seafood containing significant levels of polyunsaturated fatty acids (PUFAs), haem pigments (myoglobin and haemoglobin) and metals (like iron and copper). Depending on the changes of lipid and protein fractions, decrease the nutritional and sensory quality characteristics occur during storage of fatty fish species. For that reason fatty fish species can easily be exposed to oxidative reactions.

To prevent quality losses because of the oxidative reactions in fish meat throughout storage, natural antioxidant sources of plant origin have been studied largely, with their use chiefly growing in recent years. Natural antioxidant sources such as, green tea (*Camellia sinensis*), grape (*Vitis vinifera*) and pomegranate (*Punica granatum* L.) contain high levels of phenolic compounds. Therefore, these sources are widely used in research. The antioxidant properties of pomegranate peel (Naveena et al. 2008), grape seed (Pazos et al. 2005a, b) and green tea (Alghazeer et al. 2008) have been introduced in vivo or vitro studies. Pomegranate peel is an important source of anthocyanins, therefore it has both antioxidant and antimutagenic properties (Naveena et al. 2008). Grape seed, pulp and skins are rich source of polyphenolics (Pazos et al. 2005a, b). Grape seeds contain high amounts of catechin, epicatechin, phenolic acid and gallic acid (Palma and Taylor 1999). All these phenolic compounds have free radical scavenging activities and terminate the radical chain reactions (Sanchez-Alonso et al. 2008). Green tea leaves are a wealthy source of polyphenols, with catechins being the predominant group. The free radical scavenging capability and the metal-chelating capability of catechins obtained from tea are very high (Tang et al. 2002).

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Mackerel (*Scomber scombrus*) is tremendously prone to oxidative reactions due to its fatty acids distributions. Mackerel meat is also recognized to include high levels of haem proteins, which might act as an oxidation initiator to intent oxidative reactions. As a result, mackerel meat can easily deteriorate as a result of the application of insufficient preservation and storage conditions. Oxidative reactions and undesirable odour are known to reduce scores which obtained from sensory analysis. Therefore, the use of natural antioxidants is an effective method for controlling oxidation and off-odour development. The antioxidant efficiency of green tea, grape seed and pomegranate rind in frozen fatty fish was examined in this study. Mackerel was chosen as the fatty fish species due to its high industrial price. The efficiency of the extracts added directly to minced fish before freezing was monitored during a 6-month period of frozen storage. There are many studies on the effects of natural antioxidants to retard oxidation of lipids (Pazos et al. 2005a, b; Sanchez-Alonso et al. 2008; Alghazeer et al. 2008; Yerlikaya and Gökoglu 2010; Maqsood and Benjakul 2010). However, a limited research has been conducted on the protein oxidation in seafood (Medina et al. 2009; Sabeena Farvin et al. 2012). Vast researches have been undertaken on the antioxidant activities of green tea, grape and pomegranate extracts (Tang et al. 2002; Pazos et al. 2005a, b; Sanchez-Alonso et al. 2008; Naveena et al. 2008; Alghazeer et al. 2008; Yerlikaya and Gökoglu 2010), but the effects of these antioxidant sources on lipid and protein alteration have not been studied. Accordingly, this research focused on the effects of specific plant extracts on lipid and protein oxidation in minced mackerel during frozen storage.

Materials and methods

Materials

Mackerel (*S. scombrus*) was provided by a regional store in Iğne Ada (Kırklareli). They were stored on ground ice and transported to the laboratory within 6–8 h of being caught. Newly harvested green tea leaves (*Camellia sinensis*) were obtained from Rize. Kalecik karası grape seeds (*Vitis vinifera*) were supplied by a wine factory. Grape seeds were obtained from grape pomace, after a process of pressing and posterior maceration. After processing the grapes into wine, the seed by-product was used. Mature pomegranate (*Punica granatum* L., cv. Hicaznar) was provided by a fruit juice factory. After processing the pomegranate into fruit juice, the rind by-product was used. Butylated hydroxytoluene (BHT) was provided by Sigma-Aldrich Chemie, Steinheim, Germany.

Preparation of plant extract

Plant extracts were prepared utilizing the extraction system explained by Naveena et al. (2008). Plant materials were washed first. Then, pomegranate rind was split into 1 cm² pieces and grape seeds and green tea leaves were dried in their natural form. All plant material was dried at 60 °C in an oven, with a drying period of 3 h for green tea leaves, 12 h for grape seeds and 48 h for pomegranate rinds. In determining the drying times, the moisture content of the samples (about 60%) was taken into account so that grinding could easily be performed. Dried materials were cooled, powdered, packed and stored at −40 °C (MDF-U5411 Model, Sanyo, Korea) in high-density polyethylene bags until used. Water-soluble phenolics from plant materials were extracted in boiling water for 5 min and then filtered. Their concentrations were adjusted according to their phenolic contents.

Preparation of samples and storage conditions

Mackerels were purchased from a single lot of commercially harvested fisherman for two times in 1 week intervals. For each lot, the fish fillets (9 kg) were prepared after carefully deboned and eviscerated. The skinned fillets were ground through a 4 mm grinding plate and divided into five groups with 1.8 kg each; One group was separated as a control without antioxidant, other four groups were added GTE, GSE, PRE and BHT, separately. A-50 ml of water extracts of green tea, grape seed and pomegranate rind was individually added to minced fish samples in order to obtain a final concentration of 100 mg equivalent phenolics per kg of meat. BHT was added to contain 100 mg equivalent BHT per kg of meat after dissolved in 5 ml ethanol. The same volume of distilled water was added to control and BHT containing samples. Antioxidant level of 0.01% was chosen since it is the allowable level for BHT as an antioxidant by Food Codex in Turkey (Anonymous 2013). After the addition of antioxidant sources (GTE, GSE, PRE and BHT), each group was thoroughly mixed and formed into patties by hand. Totally, 200 patties of 90 ± 2 g were prepared. The samples were frozen to −18 °C for 2 h using a mobile air blast freezer (Frigoscandia Labofreeze, Sweden) at −35 °C and 2.5 ms^{−1} air flow and then frozen samples were placed in cartoon trays and stored at −18 °C for 6 mo.

Samples were taken before freezing (at day 0) and then at monthly intervals throughout frozen storage at −18 °C for a period of 6 months. At each analysis period, the samples were thawed at 4 °C for 6 h before evaluation.

Lipid oxidation

Lipid extraction

Total lipids were extracted from fish samples in accordance with the procedure detailed by Lee et al. (1996) as described by Soyer et al. (2010). Lipid content was calculated as percent.

Peroxide value (PV)

The peroxide value was determined according to the process of the International Dairy Federation (IDF) as described by Shantha and Decker (1994) and Soyer et al. (2010). The results were calculated in milliequivalents of peroxide per kg of lipid.

Thiobarbituric acid reactive substance (TBARS) value

TBARS were determined by the method outlined by McDonald and Hultin (1987) as described by Soyer et al. (2010). TBARS value was expressed as mg of malondialdehyde equivalents per kg of the sample.

Protein oxidation measurement

Protein carbonyls

Carbonyl measurement is probably the most commonly used procedure for following oxidation of protein. Protein carbonyls were determined using the method described by Zakrys et al. (2008). Protein concentration was determined using the Biuret method (Gornall et al. 1949). Carbonyl concentration was calculated on the DNPH-treated sample using an extinction coefficient of $21.0 \text{ mM}^{-1} \text{ cm}^{-1}$ at 370 nm for protein hydrazones and expressed as nmol carbonyls/mg protein.

Sulphydryls

Sulphydryl groups (thiol content) were measured using the method outlined by Srinivasan and Hultin (1997) as described by Soyer et al. (2010). The sulphydryl content was determined using a molar extinction coefficient of $11,400 \text{ M}^{-1} \text{ cm}^{-1}$ and expressed as nmol sulphydryl groups/mg protein.

Protein solubility

Protein solubility was determined in accordance with the process of Farouk and Swan (1998) with some modifications. Two grams of samples were weighed in a 30-ml test tube and homogenised with 20 ml of 1.1 M KI in 0.1 M

phosphate buffer (pH 7.4) for 20 s at excessive velocity using an Ultra Turrax (T25, IKA, Germany) and centrifuged (Z3226 K, Hermle, Germany) at 4 °C and $6000 \times g$ for 15 min. The supernatant contained the total soluble protein (TSP) was separated from the pellet. The protein concentration of the supernatants was determined using the Biuret method. Protein solubility was expressed as a percent.

SDS-PAGE

Preparation of myofibrillar proteins

Samples (5 g) were homogenised with 40 ml 0.03 M phosphate buffer (pH 7.4) for 2 min at high speed using an Ultra Turrax (T25, IKA, Germany). The homogenate was centrifuged at 4 °C and $10,000 \times g$ for 20 min. The supernatant included sarcoplasmic proteins was discarded. Myofibrillar proteins were obtained from the produced pellet by homogenising it with a solution containing 8 M Urea and 1% (w/v) β -mercaptoethanol for 2 min using an Ultra Turrax. The homogenate was centrifuged at 4 °C and $10,000 \times g$ for 20 min. The protein concentration of myofibrillar extracts was determined using the Biuret method. The concentration was adjusted with deionised water to produce a final concentration of 2 mg/ml and then heated at 100 °C for 5 min prior to electrophoresis.

Gel electrophoresis

SDS-PAGE was carried out as described earlier by Laemmli (1970). A 12% separating gel with 4% stacking gel was used for myofibrillar proteins. 20 μl samples were loaded onto the gel. After the run, Coomassie Brilliant Blue R-250 (0.1%) in fixative (40% methanol, 10% acetic acid) were used for staining and 40% methanol and 10% acetic acid were used for destaining the gels. The bands on the gels were identified using the molecular weight standard band for each gel. All calibration standards were purchased from Sigma Chemical, St. Louis, MO, USA.

Statistical analyses

The data were analysed using two-way analysis of variance (ANOVA) to determine the effects of antioxidant treatments (control, GTE, GSE, PRE and BHT) and storage time (from 0 to 6 months) on the alteration of lipids and proteins of minced fish meat. The experiments were replicated twice and all parameters were measured in triplicate. The statistical model was a completely random design. Duncan's multiple range test (Steel 1980) was used to evaluate the differences between means and determine

the significance of the mean values for multiple comparison at ($P < 0.01$ and $P < 0.05$).

Results and discussion

Lipid oxidation

The impact of different antioxidant sources on peroxide value and TBARS values in minced fish during 6 months of frozen storage at $-18\text{ }^{\circ}\text{C}$ is shown in Fig. 1a, b. The variance analysis for the PV data indicates that PVs were significantly ($P < 0.01$) affected by both the antioxidant treatment and storage period. PV peaked in the control, GTE, GSE and PRE samples at 4 months of storage, then started to decline rapidly. Of the five groups, GTE-applied samples showed the highest PV during the 4-month period. Compared with other natural extracts, PRE samples had the

lowest PV while BHT-added samples showed lower increases for the period of storage. Samples mixed with GTE had higher PV than control samples during storage (Fig. 1a), which indicating that green tea was not effective in controlling oxidation. This situation possibly explained by means of pro-oxidant effects of green tea, which involves high amount of chlorophyll pigments (Alghazeer et al. 2008). The data presented here are in agreement with prior findings. Alghazeer et al. (2008) showed that using in a higher concentration of green tea may have a pro-oxidant influence. Variations in the capacity to inhibit PV formation among antioxidants are defined by their structural differences (Maqsood and Benjakul 2015). PV determinations are not reliable in fatty products due to the fact that the peroxides that initially form are changeable and rapidly react to form secondary lipid oxidation products. A reduction peroxide value indicates oxidation progressing, formation of secondary oxidation products (Undeland

Fig. 1 Effect of different antioxidant sources on lipid oxidation products **a** peroxide value and **b** TBARS in minced fish during frozen storage. Bars represent the standard deviation

2001). For this reason, PV should be used in conjunction with different ways.

Changes in TBARS values of minced fish during frozen storage are shown in Fig. 1b. Secondary oxidation products presented low values at the beginning of the storage and gradually increased for all groups throughout storage. Variance analysis of TBARS values revealed the significant ($P < 0.01$) impacts of natural extract sources and storage time, along with significant interactions between these two factors. The level of TBARS in control samples increased speedily from 2.80 to 8.25 mg MDA/kg for the duration of the preliminary 3 mo of storage, then increased up to 14.25 mg MDA/kg at the end of storage. PRE-treated samples showed a slight increase in TBARS values from 2.34 to 7.57 mg MDA/kg throughout 6 mo of storage. In BHT-added samples, TBARS values increased slowly from 2.88 to 5.33 mg MDA/kg during 6 months of storage. Eventually, after storage period, TBARS values for all the samples treated with antioxidants were significantly ($P < 0.01$) lower than the values of the control samples. This data showed that green tea, grape seed, pomegranate rind extracts and BHT were efficient in retarding the increase in TBARS values during frozen storage. The activities of antioxidants differed from one to other at the same time in frozen storage but BHT and PRE were more effective than GTE and GSE. The results obtained from this research showed that PRE was much more effective than other plant extracts in terms of inhibiting the lipid oxidation in minced fatty fish. The total phenolic compounds and distribution of phenolics in the extracts played an important role due to their antioxidant activity (Sabeena Farvin et al. 2012). Variations between antioxidant-added samples may also be due to different stages of decomposition of peroxide, the formation of carbonyl compounds and the interaction of certain substances such as free amino acids, peptides, proteins, aminated phospholipids in the muscle structure (Sanchez-Alonso and Borderias 2008). The data presented here are in agreement with formerly reported data (Alghazeer et al. 2008). Lau and King (2003) indicated that GSE added at the levels of 10 and 20 g/kg to turkey thigh meat reduced TBARS values nearly tenfold in comparison with the control samples. Schormuller (1969) asserted 7–8 mg MDA/kg meat as the higher restrict of ideal freshness. In our study, TBARS values in PRE and BHT samples were within acceptable limits during frozen storage for 6 months.

The quantity of oxidation products formed demonstrated that all natural extracts were effective to retard lipid oxidation during storage. The effectiveness of antioxidants depended on various factors such as dissolution rate, efficiency in lipid phases and stability during storage.

Protein oxidation

Continuous monitoring of the carbonyl and sulphydryl content as indicators for protein oxidation during frozen storage allowed understanding the mechanism of protein damage in exhaustively. The carbonyl and sulphydryl content of minced fish treated without and with antioxidant sources during storage are given in Fig. 2a, b. The protein carbonyls occur with the progression of protein oxidation. The carbonyl content of all samples generally increased during storage. This increase in the control is much faster than others. The carbonyl content of the control samples increased from 1.03 to 1.47 nmol/mg protein during the first month of storage, then reached to 2.41 nmol/mg protein after 6 mo. Antioxidant treated samples showed the same trend but the level of increase was significantly ($P < 0.01$) lower than control samples. At the end of 6 months frozen storage, the carbonyl contents of all samples applied with antioxidants were significantly lower than those of the control samples. All antioxidant sources showed considerable efficiency at preventing the formation of protein carbonyls. Similar finding was documented by Vuorela et al. (2005) using plant phenolics in cooked pork meat patties which were reported to lower carbonyl formation for antioxidant treated samples. The carbonyl content of muscle food shows differences rely on muscle type, the status of oxidative reaction and solubility of proteins (Fagan et al. 1999). Lund et al. (2007) advocate the extent of 2–14 nmol carbonyls/mg protein to be the restrict of oxidation. In this research, the carbonyl content of GTE, GSE, PRE and BHT-treated samples was < 2 nmol/mg protein after 6 mo of storage, but the control sample exceeded 2 nmol/mg protein after 5 mo of storage. No significant difference was observed among antioxidant sources for each storage period. These results indicated that protein oxidation in minced fish was limited by antioxidant addition.

The loss of sulphydryl groups has been used as a marker of protein oxidation in meat products since cysteine residues show high susceptibility to oxidation (Lund et al. 2011). Sulphydryl contents decreased during frozen storage for all samples indicating the formation of disulphide bonds (Fig. 2b). The loss of sulphydryl groups was affected by the use of antioxidant sources and storage time. The highest decrease was observed in control samples (from 75.13 to 27.53 nmol/mg protein) during the entire storage period. After 6 months frozen storage, the sulphydryl contents for GSE and PRE treated samples was significantly higher than the those of GTE, BHT and control samples. These data suggested that GSE and PRE have great potential to be used as natural antioxidants. These results suggested that the applied antioxidants showed a significant protective effect towards protein oxidation during frozen storage.

Fig. 2 Effect of different antioxidant sources on protein oxidation products **a** carbonyl content and **b** sulphhydryl content in minced fish during frozen storage. Bars represent the standard deviation

Fig. 3 Effect of different antioxidant sources on protein solubility in minced fish during frozen storage. Bars represent the standard deviation

Fig. 4 SDS-PAGE profile of myofibrillar proteins throughout the frozen storage: **a** first day of storage, **b** third month and **c** sixth month. S1: low molecular weight protein standards; S2: high molecular weight protein standards

Figure 3 shows changes in total protein solubility (TPS) obtained in fish mince during 6 mo of storage. A variance evaluation of protein solubility showed significant effects of treatment, storage time as well as important interactions between these two factors. Protein solubility decreased gradually during extended storage. Antioxidant treated samples had significantly higher solubility than the control one. There was a considerably loss in the control sample, from 25.81 to 9.78 (62% loss) during 6 mo of storage. PRE and BHT samples had higher solubility than GTE and GSE samples at the end of storage period. Jiang and Lee (1985) reported that the muscle protein of frozen mackerel was unstable during frozen storage at -20°C and protein insolubility increased during prolonged storage since both myosin and actin became insoluble. Antioxidants protect proteins by limiting protein oxidation. Therefore, the TPS ratios of the samples added with antioxidants were higher than the control sample.

The electrophoretic patterns of myofibrillar proteins in minced fish samples at different stages of storage are given in Fig. 4a–c. There was a rigid alteration in specific protein bands when compared between fresh fish samples at the beginning of the storage time and frozen samples stored for 6 months. Generally, myofibrillar protein fractions were altered and the amount of high molecular weight protein fraction decreased. The 200 kDa (myosin from pig) and 116 kDa (β -galactosidase from *E. coli*) bands were observed at the beginning of the frozen storage in all groups. In the control samples, at the beginning of the storage, 45–14.2 kDa bands were also determined. The intensity of the 66 kDa (albumin bovine serum) band increased after 3 months of storage. The intensity of the 97 kDa (phosphorylase *b* from rabbit) band increased after 6 months of storage. The degradation of structural proteins in meat could be a result of the higher degree of protein oxidation and proteolysis reaction (Maqsood and Benjakul 2015).

The results showed that the oxidative stability of mackerel mince during frozen storage at -18°C was improved with the addition of antioxidants. Depending on the prolonged frozen storage, oxidative reactions occurred in lipids and proteins. The rates of lipid and protein oxidation and the quantity of oxidation products formed depended on the type of antioxidant used. BHT and PRE were more effective in retarding oxidative reactions. Compared with the other natural antioxidant sources, PRE was determined to be more effective in protecting the quality of mackerel mince during frozen storage. These results showed that PRE could be utilized as a natural antioxidant in raw minced fish tissue to extend storage time. GTE was not effective against lipid and protein oxidation. The results indicated that natural extracts (such

as PRE) obtained from plant sources could be used as antioxidant in the frozen storage of mackerel mince.

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References

- Alghazeer R, Saeed S, Howell NK (2008) Aldehyde formation in frozen mackerel (*Scomber scombrus*) in the presence and absence of instant green tea. Food Chem. doi:[10.1016/j.foodchem.2007.08.067](https://doi.org/10.1016/j.foodchem.2007.08.067)
- Anonymous (2013) Turkish food codex: regulation on food additives. Republic of Turkey Ministry of Food, Agriculture and Livestock, Ankara
- Fagan JM, Slecicka BG, Sohar I (1999) Quantitation of oxidative damage to tissue proteins. Int J Biochem Cell Biol. doi:[10.1016/S1357-2725\(99\)00034-5](https://doi.org/10.1016/S1357-2725(99)00034-5)
- Farouk MM, Swan JE (1998) Effect of muscle condition before freezing and simulated chemical changes during frozen storage on protein functionality in beef. Meat Sci. doi:[10.1016/S0309-1740\(98\)00035-7](https://doi.org/10.1016/S0309-1740(98)00035-7)
- Gornall AG, Bardawill CJ, David MM (1949) Determination of serum proteins by means of the biuret reaction. J Biol Chem 177:751–766
- Jiang ST, Lee TC (1985) Changes in free amino acids and protein denaturation of fish muscle during frozen storage. J Agric Food Chem 33:839–844. doi:[10.1021/jf00065a018](https://doi.org/10.1021/jf00065a018)
- Laemmli UK (1970) Cleavage of structural proteins during the assembly of the head of bacteriophage T4. Nature. doi:[10.1038/227680a0](https://doi.org/10.1038/227680a0)
- Lau DW, King AJ (2003) Pre- and post-mortem use of grape seed extract in dark poultry meat to inhibit development of thiobarbituric acid reactive substances. J Agric Food Chem. doi:[10.1021/jf020740m](https://doi.org/10.1021/jf020740m)
- Lee CM, Trevino B and Chaiyawat M (1996) A simple and rapid solvent extraction method for determining total lipids in fish tissue. J AOAC Int. PMID: 8920137
- Lund MN, Lametsch R, Hviid MS, Jensen ON, Skibsted LH (2007) High-oxygen packaging atmosphere influences protein oxidation and tenderness of porcine *Longissimus dorsi* during chill storage. Meat Sci. doi:[10.1016/j.meatsci.2007.03.016](https://doi.org/10.1016/j.meatsci.2007.03.016)
- Lund MN, Heinonen M, Baron CP, Estévez M (2011) Protein oxidation in muscle foods: a review. Mol Nutr Food Res. doi:[10.1002/mnfr.201000453](https://doi.org/10.1002/mnfr.201000453)
- Maqsood S, Benjakul S (2010) Comparative studies of four different phenolic compounds on in vitro antioxidative activity and the preventive effect on lipid oxidation of fish oil emulsion and fish mince. Food Chem. doi:[10.1016/j.foodchem.2009.06.004](https://doi.org/10.1016/j.foodchem.2009.06.004)
- Maqsood S, Benjakul S (2015) Lipid oxidation, protein degradation, microbial and sensorial quality of camel meat as influenced by phenolic compounds. LWT Food Sci Technol. doi:[10.1016/j.lwt.2015.03.106](https://doi.org/10.1016/j.lwt.2015.03.106)
- McDonald RE, Hultin HO (1987) Some characteristics of the enzymic lipid peroxidation system in the microsomal fraction of flounder skeletal muscle. J Food Sci. doi:[10.1111/j.1365-2621.1987.tb13964.x](https://doi.org/10.1111/j.1365-2621.1987.tb13964.x)
- Medina I, González MJ, Iglesias J, Hedges ND (2009) Effect of hydroxycinnamic acids on lipid oxidation and protein changes as well as water holding capacity in frozen minced horse mackerel white muscle. Food Chem. doi:[10.1016/j.foodchem.2008.10.031](https://doi.org/10.1016/j.foodchem.2008.10.031)
- Naveena BM, Sen AR, Kingsly RP, Singh DB, Kondaiiah n (2008) Antioxidant activity of pomegranate rind powder extract in cooked chicken patties. Int J Food Sci Technol. doi:[10.1111/j.1365-2621.2007.01708.x](https://doi.org/10.1111/j.1365-2621.2007.01708.x)
- Palma M and Taylor LR (1999) Extraction of polyphenolic compounds from grape seeds with near critical carbon dioxide. J Chromatography A. PMID: 10444839
- Pazos M, Gallardo JM, Torres JL, Medina I (2005a) Activity of grape polyphenol as inhibitors of the oxidation of fish lipids and frozen fish muscle. Food Chem. doi:[10.1016/j.foodchem.2004.07.036](https://doi.org/10.1016/j.foodchem.2004.07.036)
- Pazos M, González MJ, Gallardo JM, Torres JL (2005b) Preservation of the endogenous antioxidant system of fish muscle by grape polyphenols during frozen storage. Eur Food Res Technol. doi:[10.1007/s00217-004-1113-0](https://doi.org/10.1007/s00217-004-1113-0)
- Sabeena Farvin KH, Grejsen HD, Jacobsen C (2012) Potato peel extract as a natural antioxidant in chilled storage of minced horse mackerel (*Trachurus trachurus*): effect on lipid and protein oxidation. Food Chem. doi:[10.1016/j.foodchem.2011.09.056](https://doi.org/10.1016/j.foodchem.2011.09.056)
- Sanchez-Alonso I, Borderias AJ (2008) Technological effect of red grape antioxidant dietary fibre added to minced fish muscle. Int J Food Sci Technol. doi:[10.1111/j.1365-2621.2007.01554.x](https://doi.org/10.1111/j.1365-2621.2007.01554.x)
- Sanchez-Alonso I, Jimenez-Escrig A, Saura-Calixto F, Borderias AJ (2008) Antioxidant protection of white grape pomace on restructured fish products during frozen storage. LWT Food Sci Technol. doi:[10.1016/j.lwt.2007.02.002](https://doi.org/10.1016/j.lwt.2007.02.002)
- Schormuller J (1969) Handbuch der Lebensmittelchemie, vol IV. Springer, Heidelberg
- Shantha NC and Decker EA (1994) Rapid, sensitive, iron-based spectrophotometric methods for determination of peroxide values of food lipids. J AOAC Int. PMID: 8199478
- Soyer A, Özalp B, Dalmış Ü, Bilgin V (2010) Effects of freezing temperatures and frozen storage time on lipid and protein oxidation in chicken meats. Food Chem. doi:[10.1016/j.foodchem.2009.11.042](https://doi.org/10.1016/j.foodchem.2009.11.042)
- Srinivasan S, Hultin HO (1997) Chemical, physical and functional properties of cod proteins modified by a nonenzymic free-radical-generating system. J Agric Food Chem. doi:[10.1021/jf960367g](https://doi.org/10.1021/jf960367g)
- Steel RGD (1980) Principle and procedures of statistic: a biometrical approach. McGraw-Hill Text, New York, p 481
- Tang SZ, Kerry JP, Sheehan D, Buckley DJ (2002) Antioxidative mechanisms of tea catechins in chicken meat systems. Food Chem. doi:[10.1016/S0308-8146\(01\)00248-5](https://doi.org/10.1016/S0308-8146(01)00248-5)
- Undeland I (2001) Lipid oxidation in fatty fish during processing and storage. In: Kestin SC, Warriss PD (eds) Farmed fish quality. Fishing News Book, Oxford, pp 261–275
- Vuorela S, Salminen H, Makela M, Kivikari R, Karonen M, Heinonen M (2005) Effect of plant phenolics on protein and lipid oxidation in cooked pork meat patties. J Agric Food Chem 53(22):8492–8497. doi:[10.1021/Jf050995a](https://doi.org/10.1021/Jf050995a)
- Yerlikaya P, Gökoğlu N (2010) Inhibition effects of green tea and grape seed extracts on lipid oxidation in bonito fillets during frozen storage. Int J Food Sci Technol 45:252–257. doi:[10.1111/j.1365-2621.2009.02128.x](https://doi.org/10.1111/j.1365-2621.2009.02128.x)
- Zakrys PI, Hogan SA, O’Sullivan MG, Allen P, Kerry JP (2008) Effects of oxygen concentration on the sensory evaluation and quality indicators of beef muscle packed under modified atmosphere. Meat Sci. doi:[10.1016/j.meatsci.2007.10.030](https://doi.org/10.1016/j.meatsci.2007.10.030)