Antimicrobial Quality Assessment of Seabass Fillets Treated with Pomegranate Peel Extract During Refrigerated Storage

İlknur UÇAK, Wadah ELSHEIKH^{*}

Nigde Ömer Halisdemir University, Faculty of Agricultural Sciences and Technologies, Nigde, Turkey

**Corresponding author: wadah988@gmail.com*

ORCID: 0000-0003-2506-2219^{*} - 0000-0002-9701-0824

Abstract

Maintaining the quality of seafood is a crucial concern because it is a highly perishable product. In this study, microbial changes in seabass (*Dicentrarchus labrax*) fillets treated with pomegranate peel extract (PPE) in different concentrations (0.5%, and 1%) were examined during storage at $4\pm1^{\circ}$ C. The total psychrophilic bacteria (TPB), total mesophilic bacteria (TMB), and total coliform bacteria (TCB) were determined. The findings indicated that PPE showed inhibitory effects on the growth of bacteria in seabass fillets. In both concentrations (0.5 and 1%) the usage of PPE performed better than the control group, where this was quite obvious. Therefore, it may be inferred that adding PPE to sea bass fillets during refrigeration prevented microbiological deterioration. Accordingly, adding PPE can be recommended to retain the microbial quality of seabass fillets.

Keywords: Seabass, pomegranate peel extract, shelf life, microbiological quality

Research article Received Date: 9 June 2023 Accepted Date:29 June 2023

INTRODUCTION

Fresh fish has great nutritional value, including a high concentration of omega-3 fatty acids, proteins, and vitamins, making it one of the most vital products. It is nevertheless one of the most perishable products, and storage makes it vulnerable to both chemical and microbial deterioration. Digestive enzymes, lipid oxidation, and bacteria all actively contribute to fish spoilage, which results in a decline in the quality of fish and fish products (Hassoun and Çoban, 2017).

Fish must therefore be preserved with good preservation practices to maintain its freshness. In the past, fish and fish products have been preserved using temperature-based methods, including chilling and freezing (Sampels, 2015). However, lipid oxidation, protein denaturation, and microbiological activity cannot be entirely inhibited by cooling or freezing alone. Because of this, combining different processing and packaging technologies is becoming rather popular (Ucak and Afreen, 2022).

Natural additives, particularly plant extracts and essential oils, have recently been demonstrated to improve the quality of fish products when added to composite and/or bilayer films (Ojagh et al., 2010). The phenolics and tannins found in pomegranate fruits (Punica granatum L.) support the fruit's antioxidant and antibacterial properties. They have a range of nutritional and biological functions as a result. The primary polyphenol source, pomegranate peel extract (PPE), has been linked to both antioxidant and antibacterial properties. Additionally, other studies have demonstrated that PPE has antibacterial action against both gram-positive and gram-negative bacteria (Akhtar et al., 2015). PPE's antioxidant activities delay or stop the commencement of lipid oxidation by blocking the beginning or continuing stages of oxidative chain reactions or by producing stable radicals (Shah et al., 2014). PPE has several biological effects in addition to its antioxidant capacity, including antibacterial activity (Viuda-Martos et al., 2010).

Numerous research have looked at how PPE's antibacterial and antioxidant qualities affect the quality characteristics of seafood products kept at low temperatures (Basiri et al., 2015; Topuz et al., 2015; Yuan et al., 2016; Berizi et al., 2018; Khojah, 2020; Yu et al., 2022). The current experiment aimed to assess the antimicrobial effects of PPE's on seabass fillets stored in refrigerated conditions.

MATERIAL and METHOD

Fresh seabass (*Dicentrarchus labrax*) were provided by the local fisherman in the Nigde city and were shipped to the laboratory in ice boxes. They were gutted, beheaded, and filleted before being washed. From nearby marketplaces, pomegranate peels were gathered.

Pomegranate peel extraction

Pomegranate peels were washed twice in tap water, then dried at 45°C for 48 hours before being pulverized into powder. For the extraction process, 10 g of powdered pomegranate peel and 100 mL of 80% ethanol were placed in a flask and sonicated for 1 hour at (25°C) using an ultrasonic bath (Ifesan et al., 2014; Ucak, 2020). The pomegranate peels were concentrated after the extraction process using a rotary evaporator (IKA, HB-10 digital, Germany) operating at 45°C under vacuum.

Preparation of pomegranate peel extracts and application to seabass fillets

The following solutions were used as separate treatments on fish fillet samples for 5 minutes: the control solution only comprised distilled water, while the others contained 0.5% and 1% PPE solutions, respectively. All samples were placed in strofoam plates and covered by stretch film. Every three days, microbiological analysis was conducted after 12 days of keeping all samples at 4° C.

Microbiological analyzes

Using the spread plate approach, plate count agar (PCA) was used to calculate the total numbers of mesophilic and psychrophilic bacteria (ICMSF, 1982). For the total mesophilic bacteria counts and psychrophilic bacteria counts the plates were incubated at 37°C for 24-48 hours and at 8°C for 7 days, respectively. Violet red bile agar (VRBA) was used for the total coliform bacteria count in accordance with Anonymous (1998) method. Pour plating was carried out by incubation at 37°C for 24-48 hours.

Statistical analysis

Statistical analysis was done using SPSS software, and the Duncan multiple comparison test (one-way ANOVA) was used to analyze the results.

RESULTS and DISCUSSION

The primary factor causing the quality of fresh or scarcely preserved fish to deteriorate is microbial decomposition, which, in extreme situations, can lead to an up to 25% loss of marketable fish. Fish and fish products deteriorate for a variety of reasons, one of which is bacterial development (Tavares et al., 2021). The parts of fish supply a variety of nutrients for the exponential growth of microorganisms that the organism no longer controls, especially bacteria that are tolerant of a wide range of temperature conditions (Zhuang et al., 2021). Psychotropic bacteria spoil the bulk of the food stored in refrigerators and other aerobic storage conditions.

Figure: 1 shows changes in total psychrophilic bacteria counts of Sea-bass fillets and the impact of pomegranate peel extract on the microbial quality of sea bass fillets during refrigerated storage. The amount of TPB was 2.15 log CFU/g at the start of storage and grew in all groups throughout the course of that time. It was found that during storage, the amount of TPB in fish fillets treated with 1% PPE emulsion was considerably (P<0.05) lower than that in the control and 0.5% PPE groups. While the TPB counts of the control and the 0.5% PPE groups were 6.46 and 5.29 log CFU/g on the 12th day of storage, they were 5.06 log CFU/g in the 1% PPE group. The lowest APR count values during storage were observed in sea bass fillets treated with 1% PPE emulsion.

As reported by Ucak et al. (2020), rainbow trout had a TPB of 1.93 log CFU/g at the beginning of storage. The initial TPB was 2.59 log CFU/g in the rainbow trout fillets according to Ucak (2019), however Uçak et al. (2018) discovered that it was 2.47 log CFU/g and that it rose over time. The initial TPB was 3.48 log CFU/g in the fresh Shabout, according to Duman and Özpolat (2015), both of which are greater values than the one from the current investigation. 1.75 log CFU/g of TPB were initially found in sea bass fillets. The control group had the highest values, whereas the groups covered with films containing citrus seed extract had the lowest values, according to Ucak et al. (2021). This is similar to the quantity discovered in the current investigation.

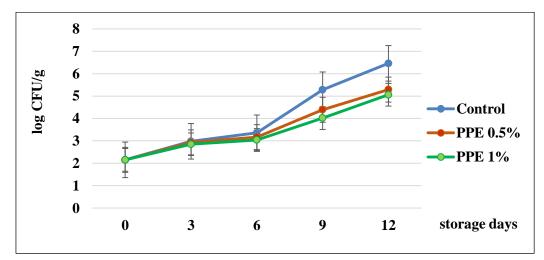
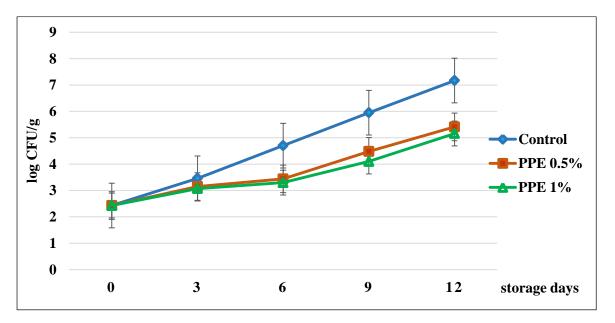


Figure 1. Changes in total psychrophilic bacteria counts of seabass fillets during storage period

Eurasian Journal of Agricultural Research 2023; Vol: 7, Issue: 1, pp: 37-43

The changes in total mesophilic bacteria (TMB) count of seabass fillets treated with PPE are presented in Fig. 2. At the beginning of the storage period, the TMB count of sea bass fillets was found to be 2.43 log CFU/g. This value increased in all groups throughout the storage period and showed the highest value as 7.17 log CFU/g in the control group, while it was 5.4 and 5.16 in 0.5% PPE and 1% PPE, respectively, at the end of the storage. The lowest (5.16 log CFU/g) TMB count was observed in the fish treated with 1% PPE at the end of storage, and it was also low during all periods of storage compared with other groups.

While Ucak (2020) reported the initial viable count of trout burgers as 2.92 log CFU/g, Keser and İzci (2020) discovered that the total bacterial count of the trout meatballs made with laurel and rosemary essential oils was found to be significantly higher (5.24 log CFU/g) than the current study. According to Zhuang (2019), PPE increases the shelf-life of carp fillets and prevents spoilage bacteria.



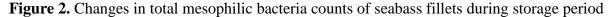


Figure 3 demonstrates the variations in the total coliform bacteria counts of sea bass fillets as well as the effect of pomegranate peel extract on the microbiological analysis of sea bass fillets at $4\pm1^{\circ}$ C. The total amount of coliform bacteria is recognized as a good predictor of fish hygiene. Trout meat was found to contain 1.80 log cob/g of coliform bacteria at zero day. Coliform bacteria growth, which grew until the end of storage, was considerably reduced (P<0.05) in the groups to which 1% pomegranate peel extract was applied. The total number of coliform bacteria per day at the ending of storage in the control group was 5.47 log cob/g, while it was found to be 4.42 and 4.12 log cob/g in the PPE 0.5% and PPE 1% groups, respectively. Similar studies have shown that the growth of total coliform bacteria in fish and fish-derived products is inhibited by natural extracts (Ucak et al., 2018; Frangos et al., 2010; z, 2018; Rezaeifar et al., 2020).

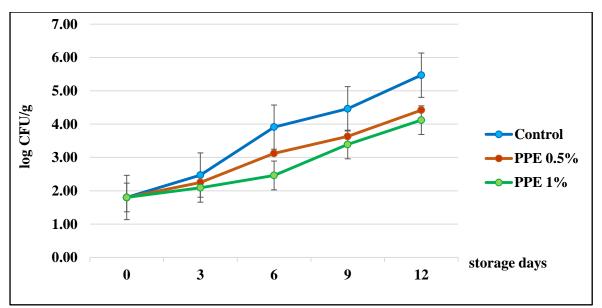


Figure 3. Changes in total coliform bacteria counts of seabass fillets during storage period

CONCLUSION

One of the most important concerns in recent years has been the examination of industrial food waste and the extraction and addition of antioxidant and antibacterial compounds to food from these wastes. Pomegranate peel is another of the by-products that make up a substantial portion of the potent antioxidants. The results obtained from this study show that pomegranate peel extract has positive effects on the characteristics of quality and microbial content of the seabass fillets. Microbial spoilage in fish fillets was delayed, and compared to the untreated group, the shelf-life was increased which clearly reflects the effectiveness of the antioxidants contained in pomegranate and their effectiveness in reducing the deterioration of quality in fish products.

REFERENCES

- Akhtar S., Ismail T., Fraternale D. & Sestili, P. 2015. Pomegranate peel and peel extracts: Chemistry and food features. *Food chemistry*, *174*, pp.417-425.
- Anonymous 1998. Bacteriological Analytical Manual (8thedn, rev.A). Association of Official Analytical Chemists, Gaithersburg, ch.28.
- Basiri S., Shekarforoush S.S., Aminlari M. & Akbari S. 2015. The effect of pomegranate peel extract (PPE) on the polyphenol oxidase (PPO) and quality of Pacific white shrimp (*Litopenaeus vannamei*) during refrigerated storage. *LWT-Food Science and Technology*, 60(2), pp.1025-1033.
- Berizi E., Hosseinzadeh S., Shekarforoush S.S. & Barbieri G. 2018. Microbial, chemical, textural and sensory properties of coated rainbow trout by chitosan combined with pomegranate peel extract during frozen storage. *International Journal of Biological Macromolecules*, 106, pp.1004-1013.
- Duman M. & Özpolat E. 2015. Effects of water extract of propolis on fresh shibuta (Barbus grypus) fillets during chilled storage. *Food Chemistry*, 189, pp.80-85.
- Frangos L., Pyrgotou N., Giatrakou V., Ntzimani A. & Savvaidis I.N. 2010. Combined effects of salting, oregano oil and vacuum-packaging on the shelf-life of refrigerated trout fillets. *Food microbiology*, 27(1), pp.115-121.

- Hassoun A. & Çoban Ö.E. 2017. Essential oils for antimicrobial and antioxidant applications in fish and other seafood products. *Trends in Food Science & Technology*, 68, pp.26-36.
- ICMSF 1982. Microorganisms in foods. Their significance and methods of enumeration, 2nd ed.; London: Univ. Toronto Pres.
- Ifesan B.O.T., Fadipe E.A. & Ifesan B.T. 2014. Investigation of antioxidant and antimicrobial properties of garlic peel extract (*Allium sativum*) and its use as natural food additive in cooked beef. *Journal of Scientific Research & Reports*, 3(5), pp.711-721.
- Keser E. & İzci L. 2020. Gökkuşağı Alabalığı (Oncorhynchus mykiss)'ndan elde edilen balık köftelerinde biberiye ve defne uçucu yağlarının mikrobiyolojik ve duyusal kaliteye etkisi. Acta Aquatica Turcica, 16(1), 13-21.
- Khojah S.M. 2020. Bio-based coating from fish gelatin, K-Carrageenan and extract of pomegranate peels for maintaining the overall qualities of fish fillet. *Journal of Aquatic Food Product Technology*, 29(8), pp.810-822.
- Ojagh S.M., Rezaei M., Razavi S.H. & Hosseini S.M.H. 2010. Effect of chitosan coatings enriched with cinnamon oil on the quality of refrigerated rainbow trout. *Food chemistry*, 120(1), pp.193-198.
- Rezaeifar M., Mehdizadeh T., Mojaddar Langroodi A. & Rezaei F. 2020. Effect of chitosan edible coating enriched with lemon verbena extract and essential oil on the shelf life of vacuum rainbow trout (*Oncorhynchus mykiss*). *Journal of Food Safety*, 40(3), p.e12781.
- Sampels S. 2015. The effects of processing technologies and preparation on the final quality of fish products. *Trends in Food Science & Technology*, 44(2), pp.131-146.
- Shah, M.A., Bosco, S.J.D. and Mir, S.A. 2014. Plant extracts as natural antioxidants in meat and meat products. *Meat science*, *98*(1), pp.21-33.
- Tavares J., Martins A., Fidalgo L.G., Lima V., Amaral R.A., Pinto C.A., Silva A.M. & Saraiva J.A. 2021. Fresh fish degradation and advances in preservation using physical emerging technologies. *Foods*, 10(4), p.780.
- Topuz O.K., Yerlikaya P., Uçak İ., Gümüş B., Büyükbenli H.A. & Gökoğlu N. 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, pp.625-632.
- Ucak I. 2020. Determination of antioxidant and antimicrobial effects of pomegranate peel extract in trout burgers stored at cold temperatures (4±1oC). *Ege Journal of Fisheries and Aquatic Sciences*, 37(4), 415-422
- Ucak I. 2019. Physicochemical and antimicrobial effects of gelatin-based edible films incorporated with garlic peel extract on the rainbow trout fillets. *Progress in Nutrition*, 21(1), pp.232-240.
- Ucak I., Abuibaid A.K., Aldawoud T.M., Galanakis C.M. & Montesano D. 2021. Antioxidant and antimicrobial effects of gelatin films incorporated with citrus seed extract on the shelf life of sea bass (*Dicentrarchus labrax*) fillets. *Journal of Food Processing and Preservation*, 45(4), p.e15304.
- Ucak I. & Afreen M. 2022. Effect of Chitosan Coating Enriched with Peppermint Essential Oil Emulsion on the Microbiological Quality of Fish Meatballs. *Eurasian Journal of Food Science and Technology*, 6(1), pp.60-68.
- Ucak I., Khalily R., Abuibaid A.K. & Ogunkalu O.A. 2018. Maintaining the quality of rainbow trout (*Oncorhynchus mykiss*) fillets by treatment of red onion peel extract during refrigerated storage. *Progress in Nutrition*, 20(4), pp.672-678.

- Ucak I., Khalily R., Carrillo C., Tomasevic I. & Barba F.J. 2020. Potential of propolis extract as a natural antioxidant and antimicrobial in gelatin films applied to rainbow trout (*Oncorhynchus mykiss*) fillets. *Foods*, 9(11), p.1584.
- Viuda-Martos M., Fernández-López J. & Pérez-Álvarez J.A. 2010. Pomegranate and its many functional components as related to human health: a review. *Comprehensive Reviews in Food Science and Food Safety*, 9: 635-654.
- Yu D., Zhao W., Dong J., Zang J., Regenstein J.M., Jiang Q. & Xia W. 2022. Multifunctional bioactive coatings based on water-soluble chitosan with pomegranate peel extract for fish flesh preservation. *Food Chemistry*, 374: 131619.
- Yuan, G., Lv, H., Tang, W., Zhang, X. and Sun, H. 2016. Effect of chitosan coating combined with pomegranate peel extract on the quality of Pacific white shrimp during iced storage. *Food Control*, 59: 818-823.
- Zhuang S., Hong H., Zhang L. & Luo Y. 2021. Spoilage-related microbiota in fish and crustaceans during storage: Research progress and future trends. *Comprehensive Reviews in Food Science and Food Safety*, 20(1): 252-288.
- Zhuang S., Li Y., Jia S., Hong H., Liu Y. & Luo Y. 2019. Effects of pomegranate peel extract on quality and microbiota composition of bighead carp (*Aristichthys nobilis*) fillets during chilled storage. *Food microbiology*, 82: 445-454.