

## The Impact of Glazing with Natural Preservatives on Some Qualitative Characteristics of Frozen-Stored Japanese Threadfin Bream *Nemipterus japonicus*

Shams A. Al-Hamdani<sup>1</sup>, Jalal M. Al-Noor<sup>2</sup>

<sup>1</sup>Departement of Fisheries and Marine Resources, University of Basrah, Iraq

<sup>2</sup>Unit of Aquaculture, College of Agriculture, University of Basrah, Iraq

\*Corresponding Author: [shamsfiras80@gmail.com](mailto:shamsfiras80@gmail.com)

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### ABSTRACT

The current study was conducted to determine the impact of immersion using garlic, sumac, and citric acid on certain qualitative characteristics of Japanese threadfin bream (*Nemipterus japonicus*) during frozen storage. Fish were immersed with 1% addition of each natural substance and stored at -18 °C for 150 days. The results revealed that immersion with garlic, sumac, and citric acid prevented lipid oxidation and reduced the production of free fatty acids (FFA), volatile nitrogen bases (TVN), and thiobarbituric acid (TBA) in the groups treated with natural substances compared to untreated and water-immersed groups. The lowest pH value was 6.11 at day zero for the frozen fish, which increased to 6.98 at the end of storage for the same treatment. The lowest TVN, FFA, and TBA values were observed in the garlic-treated fish at day zero: 4.2mg nitrogen/100g fish, 0.12%, and 0.27mg malondialdehyde/100g fish, respectively. These values increased significantly during the final storage period to 29.4mg nitrogen/100g fish, 0.88%, and 5.41mg malondialdehyde/100g fish for the frozen fish only at 150 days of storage, with significant differences among treatments and storage periods ( $P < 0.05$ ). The lowest percentage of lost fluids was 2.33% for the garlic immersion treatment at day zero, while the highest value was 5.49% for the frozen fish only at 150 days. In conclusion, the study demonstrated the potential use of these substances in immersion as natural antioxidants and inhibitors of enzymatic, microbial, and quality loss.

### INTRODUCTION

Fish are the second source of animal protein after poultry. Fish meat is highly favored by a huge number of populations due to its high protein content and low cholesterol levels. Additionally, fish meat contains various useful minerals, elements such as iodine, and important vitamins. Moreover, it has a lower content of connective tissues compared to red meat and higher water content, making it tender when cooked and easily digestible (Marques *et al.*, 2019). However, fish are prone to spoilage due to the natural flora on its surface, neutral pH, and high moisture content. The enzymes in fish tissues contribute to the autolysis of proteins and fats (Huss, 1995). Therefore, proper handling and storage methods are essential to maintain fish quality (Sone *et al.*,

2019). Freezing is commonly used to preserve the nutritional and qualitative value of fish and their products for an extended period by inhibiting microbial growth and reducing physical and chemical deterioration during storage (Malik *et al.*, 2021). However, frozen fish may undergo undesirable changes, such as flavor alterations like rancidity and undesirable fishy taste resulting from the formation of low molecular weight compounds due to lipid and protein oxidation. Changes in color, appearance, texture, loss of juiciness, and sponginess are also included (Park *et al.*, 2021). The quality of frozen fish can be preserved through packaging applying methods such as glazing (Wang *et al.*, 2020). Glazing involves applying a thin layer of ice to protect fish from oxidation, dehydration, and freezer burn during storage, maintaining flavor compounds and an attractive appearance (Shi *et al.*, 2019). Alternative packaging methods include the use of active packaging, incorporating components or substances with antioxidant and antimicrobial properties to extend product shelf life (Khalaf *et al.*, 2019; AL-Hilphy *et al.*, 2022). Spices and flavor enhancers have been used as effective additives in fish packaging to inhibit microbial growth, enzyme activity, and prevent color changes in fish slices (Klinmalai *et al.*, 2021). The study aimed to utilize Japanese sea bass (*N. japonicus*) as it is a marine fish available in Basra province and exploit its availability while preserving its high nutritional value suitable for marketing and human consumption, especially in remote areas, by using the glazing method to preserve it for long periods and prevent economic loss resulting from fish spoilage. Additionally, the study aimed to use natural food additives in fish meat to improve its nutritional value, extend its shelf life, and control spoilage by possessing effective antioxidant and antimicrobial properties, while avoiding artificial additives.

## MATERIALS AND METHODS

### Raw fish

Fresh *N. japonicus* samples were obtained from the marine fish market in Al-Faw City south of Basrah Governorate. Fish were averaged 19cm in length and were weighed 280gm. Samples were transported using an insulated container cooled with ice and upon arrival at the laboratory fish were thoroughly washed to remove any contaminants. A random sample was then taken to conduct qualitative analyses.

### Utilized food additives

Garlic powder, sumac powder, and citric acid were used as natural food preservatives added to the water glazing solution at a rate of 1%.

### Glazing process

Fish were frozen using a conventional freezer at a temperature of  $-18^{\circ}\text{C}$  for a period of 3 days in November 2022. Subsequently, glazing solutions were prepared with a concentration of 1% for each preservative, and the solutions were mixed thoroughly using a magnetic stirrer for 2 hours at a temperature of  $30^{\circ}\text{C}$  until complete dissolution. The glazing process was carried out using a wooden container with dimensions of  $90\times 60\text{cm}$  filled with crushed ice. The glazing solutions were placed in metal containers fixed in the middle of the ice. The frozen fish were then divided into five groups. The first group was left frozen as a control sample (A), while the second group was glazed with water only (B). The third group was glazed with a solution of water and garlic

powder (C), the fourth group was glazed with a solution of water and sumac powder (D), and the fifth group was glazed with a solution of water and citric acid (E). Fish were immersed in the glazing solutions individually for a period of 5 minutes, allowing the formation of a water layer or coating on the glazed fish surfaces. After completing the glazing process, samples were transferred to metal trays and stored frozen at a temperature of -18°C for a period of 150 days. During this period, changes in physicochemical characteristics were monitored.

### Estimation of physicochemical characteristics

#### Estimation of pH Value

The pH was measured using a pH meter according to the method described by Wong *et al.* (1991).

#### Estimation of total volatile nitrogen TVNB

Based on the method described by Egan *et al.* (1988), the collected distillate was titrated with sulfuric acid of 0.1 normality until the color changed from green-blue to light pink. The volume of acid used in titration was multiplied by 14 to obtain the TVNB (Total volatile nitrogen base) in milligrams nitrogen per 100 grams of fish meat, according to the following formula:  $100\text{g fish} = \text{ml } 0.1\text{N H}_2\text{SO}_4 \times 14/\text{TVNB mg N}$

#### Estimation of thiobarbituric acid (TBA)

The applied method was adopted from Egan *et al.* (1988), by using TBA reagent and the final solution measured spectrophotometer at a wavelength of 538 nanometers. The TBA number was calculated using the following formula:

$$\text{TBA mg malonaldehyd/kg fish} = 7.8 \times \text{Absorbance}$$

#### Estimation of acid value (AV)

The method described by Wong *et al.* (1991) was applied; the calculation of free fatty acids was then carried out based on oleic acid using the following formula :

$$\text{FFA} = \frac{\text{Volume of NaOH in milliliter} \times \text{Molarity} \times 28.2}{\text{Weight of the Sample (gm)}}$$

#### Estimation of drip loss (DL)

A piece of fish meat was weighed, tied with a thin cotton thread and hung in the refrigerator for 48 hours at a temperature of 4°C. Afterward, the sample was weighed after drying using a filter paper according to method mentioned by Rasmussein and Mast (1989):

$$\text{Drip loss} = \frac{\text{Original Weight of the Sample} - \text{Original Weight of the Sample after 48 hour}}{\text{Original Weight of the Sample}} \times 100$$

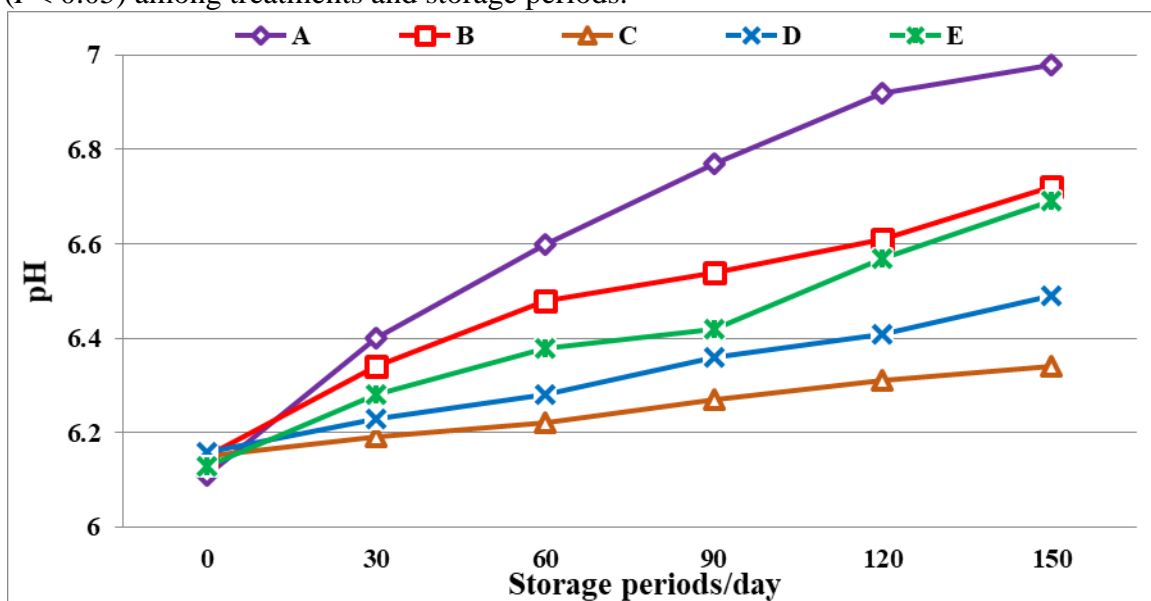
## RESULTS

The effect of immersion in natural preservatives and freezing storage duration on the physicochemical properties of examined fish was discussed in detail.

#### pH values

The results in Fig. (1) highlight the impact of immersion in natural preservatives and freezing storage duration on the pH values of experimental fish. The findings revealed variability in the values across different treatments, with the highest value

recorded at 6.63 for frozen fish only. Conversely, fish infused with garlic showed the lowest pH at 6.24, while those infused with sumac exhibited a pH of 6.32. The results also indicated that fish immersed in water only and fish immersed in acid had pH values of 6.47 and 6.41, respectively. For the impact of storage duration on pH values, the data presented in Fig. (1) reveal a continuous change, with a gradual increase observed in the average pH values over storage periods for all treatments. The pH value in the freezing treatment started at 6.11 on day zero, and progressively increased to 6.6 and 6.77 after 60 and 90 days, reaching 6.92 and 6.98 after 120 and 150 days of storage, respectively. Similarly, the pH averages for the water-only immersion, sumac immersion, and acid immersion treatments in the final storage period of 150 days were 6.72, 6.49, and 6.69, respectively, compared to their initial values of 6.15, 6.16, and 6.13 on the first day of storage. For the garlic immersion treatment, which had the lowest pH value of 6.15 on day zero, an increase was observed over storage periods, reaching 6.22 after 60 days and gradually continuing to reach 6.34 at the end of the 150-day storage period. In general, the lowest overall average pH was 6.14 on day zero, and it began a gradual increase to reach an overall average of 6.39 and 6.47 for the storage periods of 60 and 90 days, respectively. The increase reached its maxima at the final storage period of 150 days, where the average was 6.64. Statistical analysis results indicate significant differences ( $P < 0.05$ ) among treatments and storage periods.

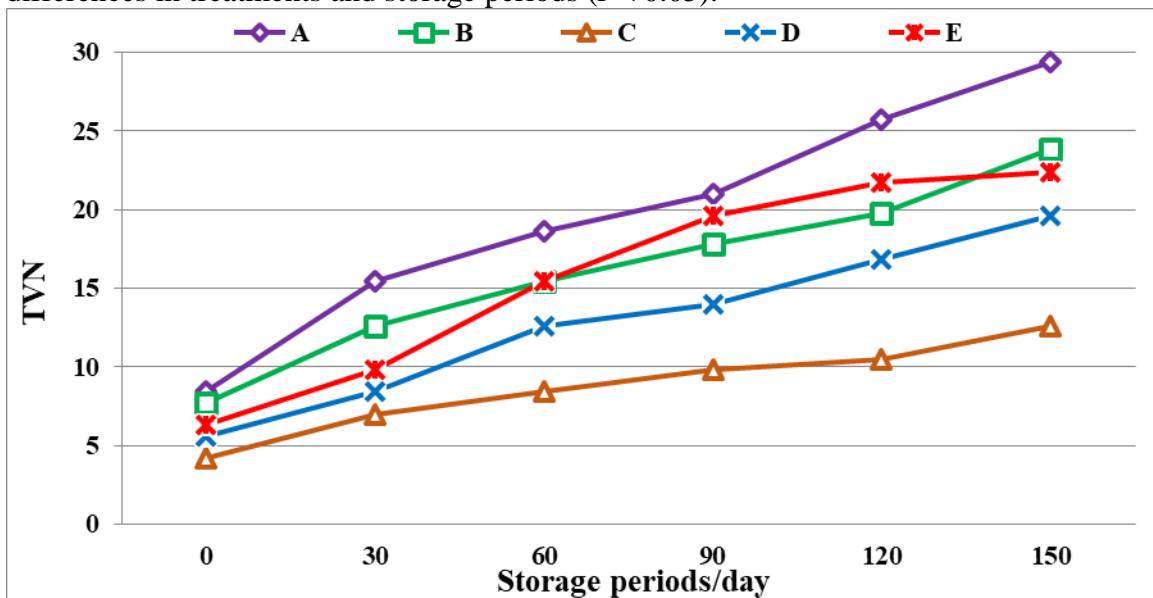


**Fig. 1.** The impact of various immersion treatments and freezing storage duration ( $-18 \pm 2$ ) on the pH values for bass fish: (A) Frozen as a control sample, (B) Glazed with water only, (C) Glazed with garlic, (D) Glazed with sumac, and (E) Glazed with citric acid

### Total volatile nitrogen TVNB

The results in Fig. (2) show the levels of volatile nitrogen bases in glass-jarred fish treated with natural preservatives and stored under freezing conditions ( $-18 \pm 2^\circ\text{C}$ ) for varying durations. The percentage of nitrogen bases varied across treatments, with the highest percentage observed in the frozen fish treatment alone at 19.75mg nitrogen/ 100g fish. Afterward, the treatment involving immersion in water only showed a rate of 16.18. Conversely, the lowest values for nitrogen bases were recorded in fish immersed with

garlic, with a rate of 8.75. The remaining percentages for other treatments were 12.83 and 15.86 for fish immersed with sumac and fish immersed using acid, respectively. Regarding the impact of freezing storage periods on the values of volatile nitrogen bases, a gradual increase in values was observed with the extension of storage duration for all treatments. The highest values were reached in the freezing treatment, escalating from 8.4 on the first day to 29.4 on the last day of storage (150 days), compared to 18.6 and 21 for storage periods of 60 and 90 days, respectively. Conversely, the lowest values were in the garlic-infused treatment, showing the least increase in volatile nitrogen base values. On the first day (day zero) of storage, the values were 4.2, and they continued to increase to 8.4 and 9.8 for storage periods of 60 and 90 days, respectively. The averages reached 10.5 and 12.6 after 120 and 150 days of storage, respectively. Similarly, the values of volatile nitrogen bases in the water-only immersion treatment, sumac immersion treatment, and acid immersion treatment were affected by the storage durations. The results indicated a gradual increase in values with advancing storage, reaching 23.8, 19.6, and 22.4 on the last day (150) of storage, compared to 7.7, 5.6, and 6.3 on the first day of storage (day zero), respectively. In general, the lowest overall average for volatile nitrogen bases was 6.44 on the first day of storage, and the averages continued to increase until reaching the highest overall average of 21.56 at the end of the storage period (150 days), compared to overall averages of 16.44 and 18.9mg nitrogen/ 100g fish for the storage periods of 90 and 120 days, respectively. The results indicate significant differences in treatments and storage periods ( $P < 0.05$ ).

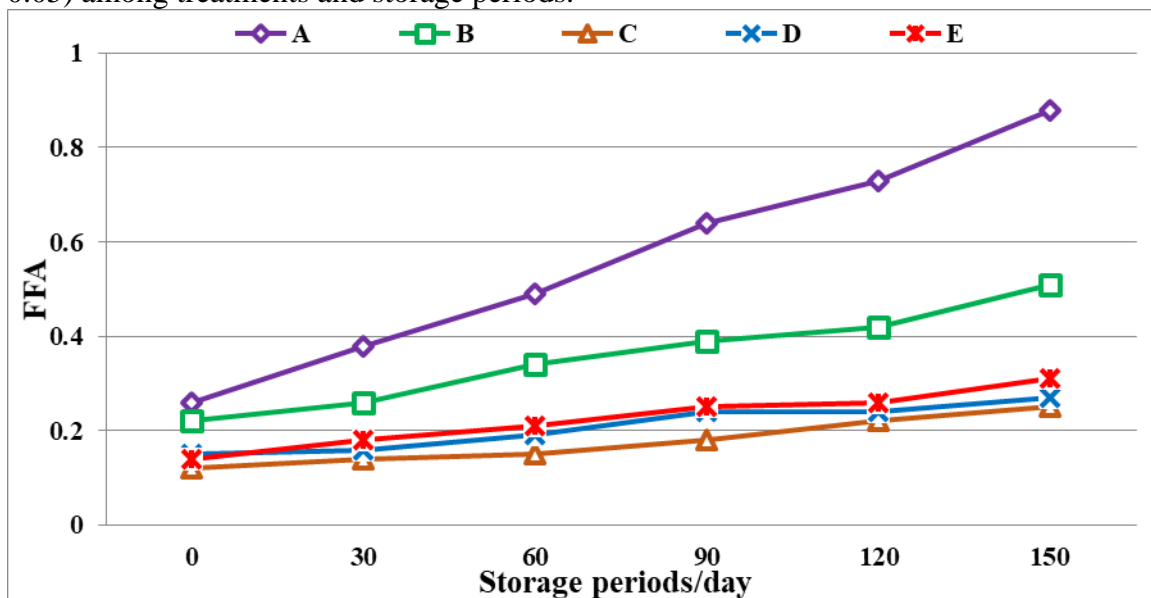


**Fig. 2.** The impact of various immersion treatments and freezing storage duration ( $-18 \pm 2$ ) on the TVN values mg nitrogen/ 100g fish for bass fish: (A) Frozen as a control sample, (B) Glazed with water only, (C) Glazed with garlic, (D) Glazed with sumac, and (E) Glazed with citric acid

### FFA values

The results in Fig. (3) illustrate the values of free fatty acids in bass fish immersed with natural preservatives and stored frozen ( $-18 \pm 2^\circ\text{C}$ ) for various storage periods. The results reveal variations in the average values of free fatty acids with different immersion treatments. The highest values were recorded in the samples frozen only, with an average of 0.56%, followed by samples immersed in water, with an average of 0.35%. The lowest

average for free fatty acids was 0.17% for fish immersed with garlic, followed by fish immersed with sumac at 0.2%; the percentage of free fatty acids in fish immersed with acid was 0.22%. The results revealed an increase in the levels of free fatty acids as storage periods progressed. The values were elevated in the freezing treatment from 0.26 at zero day to 0.49 and 0.64 after 60 and 90 days, respectively, continuing to increase to 0.73 and 0.88 after storage periods of 120 and 150 days, respectively. In contrast, the highest average for the water immersion treatment and the treatments involving immersion in sumac and immersion in acid in the final storage period of 150 days was 0.51, 0.27, and 0.31, respectively, compared to their values on the first day of storage (0.22, 0.15, and 0.14, respectively). Regarding the garlic immersion treatment, its lowest value was 0.12 at day zero, gradually increasing to 0.15 and 0.18 after storage periods of 60 and 90 days. The values continued to rise gradually, reaching 0.25 at the end of the 150-day storage period. The overall lowest average for free fatty acids was 0.17 for the zero-day storage period, steadily increasing to its highest overall rate of 0.44 at the end of the 150-day storage period, compared to 0.27 for the 60-day storage period and 0.34 for the 90-day storage period. Statistical analysis results indicate significant differences ( $P < 0.05$ ) among treatments and storage periods.



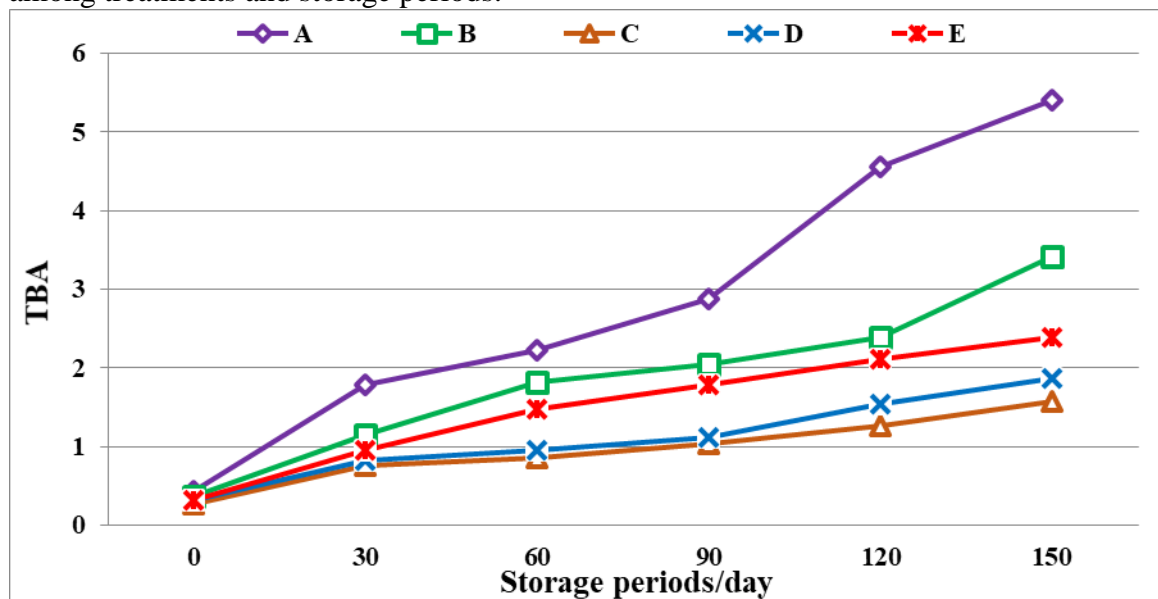
**Fig. 3.** The impact of various immersion treatments and freezing storage duration ( $-18 \pm 2$ ) on the FFA values % for bass fish: (A) Frozen as a control sample, (B) Glazed with water only, (C) Glazed with garlic, (D) Glazed with sumac, and (E) Glazed with citric acid

### TBA values

Fig. (4) displays the effect of freezing with natural preservatives and storage duration on the averages of thiobarbituric acid values for examined fish. The results showed that the highest percentage of thiobarbituric acid was observed in fish frozen without any glazing, with an average of 2.88mg malondialdehyde/ 100g fish, following in value were the fish glazed with water, with an average of 1.86mg malondialdehyde/ 100g fish. In contrast, fish glazed with garlic obtained the lowest average for thiobarbituric acid values at 0.95mg malondialdehyde/ 100g fish. The remaining treatments varied in values, reaching 1.1 and 1.5mg malondialdehyde/ 100g fish for fish treated via glazing with sumac and acid, respectively. With respect to the effect of storage duration on the



thiobarbituric acid values, continuous changes were observed from the beginning until the end of the storage period. A gradual increase was noticed during freezing treatment, starting from 0.43mg malondialdehyde/ 100g fish at day zero to reaching 2.23 and 2.87mg malondialdehyde/ 100g fish after 60 and 90 days, respectively. The values continued to rise, reaching 4.56 and 5.41mg malondialdehyde/ 100g fish at the end of the storage periods of 120 and 150 days. In contrast, the highest averages for the treatment with water glazing alone, glazing with sumac, and glazing with acid in the last storage period of 150 days were 3.42, 1.86, and 2.39mg malondialdehyde/ 100g fish, respectively, compared to their values on the first day of storage (0.36, 0.32, and 0.32, respectively). Regarding the treatment with garlic glazing, the lowest average for thiobarbituric acid was recorded at day zero (0.27), and the values increased to 0.86 after 60 days of storage. The values continued to gradually increase with the progression of the storage period, reaching 3.42mg malondialdehyde/ 100g fish at the end of the 150-day storage period. The lowest overall average for thiobarbituric acid was 0.34 for the zero-day storage period, and it continued to increase, reaching its highest overall rate of 1.57mg malondialdehyde/ 100g fish at the end of the 150-day storage period. This was compared to an overall rate of 1.46 for the 60-day storage period and 1.77 for the 90-day storage period. Statistical analysis results indicate significant differences ( $P < 0.05$ ) among treatments and storage periods.

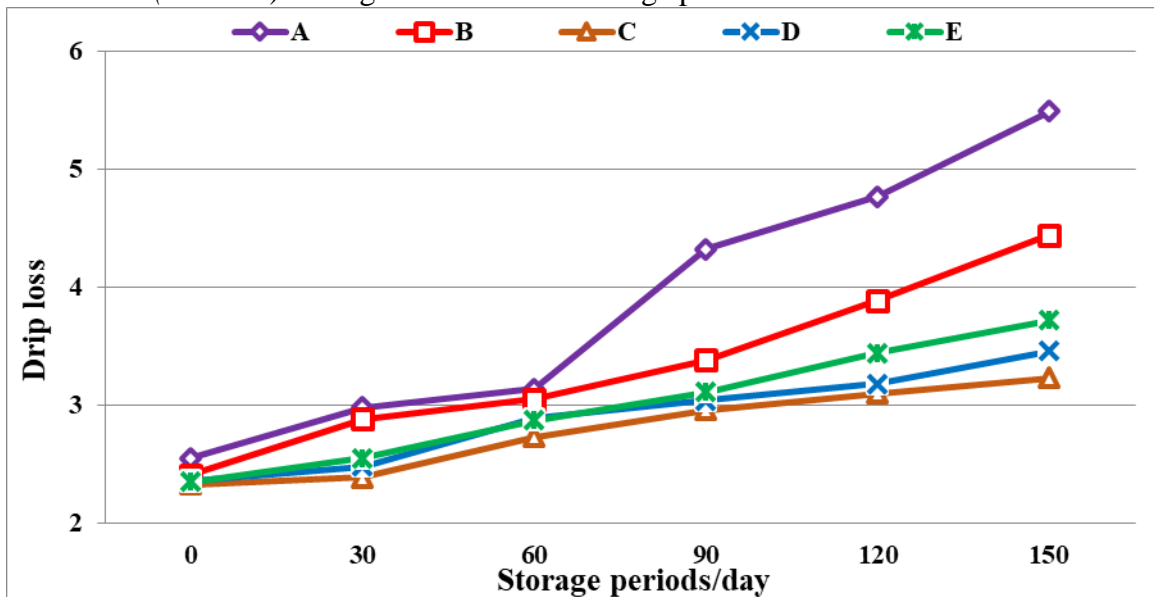


**Fig. 4.** The impact of various immersion treatments and freezing storage duration ( $-18 \pm 2$ ) on the TBA values mg malondialdehyde/100g fish for bass fish: (A) Frozen as a control sample, (B) Glazed with water only, (C) Glazed with garlic, (D) Glazed with sumac, and (E) Glazed with citric acid

### Drip loss

The results in Fig. (°) illustrate the percentage of lost liquids for glazed bass fish with natural preservatives stored by freezing ( $-18 \pm 2^\circ\text{C}$ ) for different periods. The results showed variation in the means depending on the type of treatment used. The highest rate was recorded in frozen samples at 3.87, and this value decreased to 3.34, 3, and 2.9 in samples glazed with water only, glazed with acid and glazed with sumac, respectively. In

comparison, the treatment with garlic glazing had the lowest overall average at 2.79. Regarding the impact of storage duration on the percentage of lost liquids, a gradual increase was observed with the extended storage period for all treatments. In the freezing treatment, the percentages increased from 2.55 on the first day to 5.49 on the last day of storage (150 days), with intermediate values of 3.14 and 4.32 for the storage periods of 60 and 90 days, respectively. However, the treatment with garlic glazing exhibited the least increase in the percentage of lost liquids as the storage period advanced. The percentage on the first day (day zero) of storage was 2.33, and the values continued to increase, reaching 2.73 and 2.96 for the storage periods of 60 and 90 days, respectively. The averages reached 3.23 on the last day of storage after 150 days. The glazing treatments with water, acid and sumac were affected by the storage duration, showing a gradual increase in values with the progress of storage. On the last day (150) of storage, the percentages were 4.44, 3.72, and 3.46, respectively, compared to the first day of storage (day zero) when they were 2.41, 2.35, and 2.36, respectively. In general, the lowest average percentage of lost liquids was 2.4 for the zero-day storage period, and the averages continued to increase, reaching their highest overall rate of 4.06 at the end of the 150-day storage period. This was comparable to overall rates of 3.36 and 3.67 for the 90 and 120-day storage periods, respectively. Statistical analysis results indicate significant differences ( $P < 0.05$ ) among treatments and storage periods.



**Fig. 5.** The impact of various immersion treatments and freezing storage duration ( $-18 \pm 2$ ) on the drip loss values % for bass fish: (A) Frozen as a control sample, (B) Glazed with water only, (C) Glazed with garlic, (D) Glazed with sumac, and (E) Glazed with citric acid

## DISCUSSION

### pH values

The reason for the elevated pH values in frozen fish samples alone may be attributed to the formation of nitrogen compounds resulting from enzyme and microbial activity, leading to protein degradation (Badee *et al.*, 2013). The decrease in pH values in samples treated with freezing and natural preservatives could be a result of their inhibitory role, aiding in reducing microbial contamination in meat, protein breakdown,



and the accumulation of ammonia, which reflected in the decrease in pH values (Souza *et al.*, 2019). These results are consistent with the findings of Tan *et al.* (2019) who observed that, freezing with the use of preservatives effectively contributed to lowering pH values and preserving water retention in frozen hake for six months compared to control samples. Similarly, Pezeshk *et al.* (2015) found that, the use of natural preservatives led to a reduction in microbial activity, subsequently resulting in a decrease in pH values in the treated samples. For the impact of storage duration on pH values, the figure revealed a continuous change, with a gradual increase observed in the average pH values over storage periods for all treatments, the reason for the increase in pH values suggest a role of microorganisms in breaking down proteins into amino acids, releasing nitrogen. Consequently, the accumulation of ammonia occurs as a byproduct of amino acid breakdown, leading to an elevation in pH values (Souza *et al.*, 2019). Daneshi *et al.* (2023) showed that compared to the unglazed control samples, glazing treatment reduced the quality loss of shrimp during the 150 days of frozen storage. Results also illustrated that Spirulina glazed shrimp samples had lower TVB-N, PV, TBA, and higher textural and sensory properties compared to the other treatments.

#### **Total volatile nitrogen TVNB**

The increase in total volatile nitrogen content in all frozen and glass-jarred fish samples may be attributed to hydrolysis of proteins by protease enzymes produced by protein-degrading bacteria (Duarte *et al.*, 2020). It was observed that, the use of natural preservatives during immersion helped better preserve fish meat by containing effective natural substances that prevent protein degradation, thereby reducing the production of total volatile nitrogen compared to freezing-only samples (Kaba *et al.*, 2014). The current results are consistent with the study conducted by Fadiloğlu and Çoban (2019), who demonstrated a reduction in TVN values for glass-jarred slices of trout stored in freezing conditions (-18°C) for 6 months compared to the control treatment. Similarly, Hosseini *et al.* (2022) found that the combined effect of glass-jarring, aqueous extract, and coriander leaf extract gave the lowest values for volatile nitrogen bases in mackerel fish stored under freezing conditions at -18°C for 6 months compared to the control treatment, with a measured shelf life of 180 days. These findings align with those of Wang *et al.* (2020) who found that, using glass-jarring with rosemary acid reduces the auto-oxidation process and lowers the levels of free nitrogen bases in glass-jarred tuna fish stored under freezing conditions for 180 days compared to frozen-only samples. The results coincide with the study by Hui *et al.* (2023) regarding the use of essential oils from citrus peel waste and their role in inhibiting microorganisms and improving the glazing process. This led to a reduction in volatile nitrogenous bases levels in frozen tilapia fish at -18°C, as well as a decrease in the level of biogenic amines. Regarding the impact of freezing storage periods on the values of volatile nitrogen bases, a gradual increase in values was observed with the extension of storage duration for all treatments. The highest values were reached in the freezing treatment, the subsequent increase in the values of volatile nitrogen bases during the progression of storage periods can be attributed to the accumulation of alkaline compounds (such as ammonia and trimethylamine) resulting from protein degradation by enzymes (Duarte *et al.*, 2020). These findings are consistent with those of Fadiloğlu and Coban (2018) who showed that, the use of sumac reduced the values of volatile nitrogen bases in *Oncorhynchus mykiss* during refrigerated storage for 12 days.

### FFA values

The increase in free fatty acid percentages in fish treated with immersion and natural substances was lower compared to the control treatment. The decrease in free fatty acids can be attributed to the reduction in bacterial counts due to the presence of natural substances, especially garlic and sumac, containing antibacterial compounds. These compounds inhibit the production of lipase enzyme, which is secreted by some bacteria involved in the enzymatic spoilage of meat. Conversely, the FFA levels increased in the control treatment, contributing to increased enzymatic spoilage due to bacterial enzymes (Mehrabi *et al.*, 2021). These findings match those of Fadiloğlu and Coban (2019) who reported that, the use of sumac powder reduced the production of free fatty acids in frozen trout slices stored for 6 months compared to the control treatment. It was observed that, acidity values varied with different treatments. The use of natural preservatives during frozen storage was found to be more effective in extending the shelf life and preserving quality. This effectiveness could be traced back to the presence of active compounds such as phenolic compounds and flavonoids in these preservatives. These compounds were found to inhibit the growth of fat-degrading bacteria, thus reducing acidity through their impact on the bacteria secreting lipase enzyme, which plays a role in lipid hydrolysis (Mei *et al.*, 2019). Furthermore, Méndez *et al.* (2023) demonstrated that, using glazing with natural preservatives resulted in lipid stabilization for frozen stored hake fish, inhibiting degradation and reducing the release of free fatty acids.

The results revealed an increase in the levels of free fatty acids as storage periods progressed. The elevation in the levels of free fatty acids with the progression of storage periods may be attributed to the activity of lipolytic enzymes, which break down ester bonds, releasing free fatty acids as the final product of the hydrolysis of fats by lipase and phospholipase enzymes produced by fat-degrading bacteria (Mei *et al.*, 2019). The findings agree with the observations of Al-Magsosy (2014) and those of Fadiloğlu and Coban (2018), who noted an increase in free fatty acid levels in glazed fish using certain natural preservatives during frozen storage at -18°C for 120 days.

### TBA values

The increase in TBA values and secondary products of fat oxidation in fish is owing to the breakdown of peroxides during frozen storage (Tingting *et al.*, 2012). However, samples glazed with preservatives showed a lower increase in TBA values. This can be attributed to the antioxidant activity of the natural additives which contain phenolic compounds with a high efficacy in inhibiting free radicals. Additionally, their antimicrobial activity and their ability to alter the permeability of bacterial cell membranes by increasing free amino groups contribute to reducing the impact on the bacterial cell wall's negative charge (Shahbazi & Shavisi, 2018). These results confirm that the use of natural preservatives was effective in preserving glazed fish samples from oxidation for a longer period compared to control samples. This effectiveness could be ascribed to the presence of various active compounds, such as phenols and flavonoids, which have the ability to capture free radicals and halt the oxidative reaction chain. This is achieved through the contribution of hydroxyl groups that break down the oxidation chain and inhibit the lipid oxidation process by suppressing the activity of free radicals. This, in turn, extends the initial stage of the oxidation process, slowing down the formation of peroxides and subsequently reducing the formation of hydroperoxides.

Consequently, the amount of malondialdehyde, which is one of the secondary products of fat oxidation and the breakdown of peroxides in meat and its products, decreases (**Zhang et al., 2019**). These results are consistent with the findings of **Tan et al. (2019)** who observed that, glazing with preservatives effectively contributed to reducing the values of thiobarbituric acid in frozen-stored catfish for a period of six months. In addition, the results align with the study conducted by **Fadılođlu and Coban (2019)** on the impact of using sumac at 5 and 10%, reducing TBA levels for frozen-stored trout fillets at  $-18^{\circ}\text{C}$  for six months compared to the control treatment. Additionally, **Dai et al. (2015)** demonstrated that glazing with plant extracts was more effective in reducing fat oxidation, attributing this to the resistance to oxygen diffusion compared to samples glazed with water and frozen alone. For the effect of storage duration on the thiobarbituric acid values, continuous changes were observed from the beginning until the end of the storage period. The reason for the increase in thiobarbituric acid values is attributed to the occurrence of auto-oxidation and the production of aldehydes and ketones (**Tingting et al., 2012**). These results coincide with the results of **Al-Magsosy (2014)**, who observed an increase in thiobarbituric acid values for glassed fish using certain natural preservatives during frozen storage at  $-18^{\circ}\text{C}$  for 120 days.

#### **Drip loss**

The reason for the reduced water loss from the treated samples is attributed to the use of natural preservatives which reduces moisture transfer between the food and the external environment. The moisture barrier property increases with the use of natural preservatives (**Qin et al., 2019**). Additionally, the presence of aromatic rings in natural substances leads to a reduction in free volume and an elongation of the water vapor transfer path, resulting in lower water loss (**Yong et al., 2019**). These results are congruent with the findings of **Tan et al. (2019)** who observed that, glazing with the use of preservatives effectively contributed to preserving water retention capacity of frozen-stored catfish for a period of six months, the results were also consistent with the study of **Elghazali et al. (2023)** who found that, the use of gelatin with rosemary preserved the total muscle protein and sarcoplasmic proteins of frozen stored tilapia fish at  $-18^{\circ}\text{C}$  for six months. **Yue et al. (2023)** concluded that, using glazing with natural preservatives led to a reduction in water loss susceptibility and prevented deterioration in the muscle protein characteristics of fish ball products. Regarding the impact of storage duration on the percentage of lost liquids, a gradual increase was observed with the extended storage period for all treatments. **Zhang et al. (2021)** observed a decrease in water-holding capacity (WHC) values attributed to the growth of ice crystals during frozen storage. The reason for weight loss differences between treatments over the storage periods can be related to water sublimation during frozen storage. Glazing can act as a protective barrier against water vapor, preventing sample drying. Consequently, the water content of the samples gradually decreased. The removal of water from the squid was more challenging through centrifugation, leading to a gradual increase in WHC during the late stage of frozen storage. These results align with the findings of **Tan et al. (2019)** who observed that, glazing with the use of preservatives is effectively contributed to reducing moisture loss and maintaining water retention capacity for frozen-stored catfish over a six-month period.

## CONCLUSION

The current research highlights the feasibility of using natural food additives in fish meat to improve its nutritional value, extend its shelf life, and control spoilage by possessing effective antioxidant and antimicrobial properties, while avoiding artificial additives.

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