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Improving the Quality and Shelf-life of Catfish Burger using Nisin as Bio-Preservation During Refrigeration

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ABSTRACT

Nisin, used as a biological preservative agent, has emerged as a promising technique in recent years. Thus, this study was conducted to assess the effects of nisin treatment at different levels on the organoleptic, physico-chemical and microbiological properties of a burger sample at 4°C in the refrigerator. The physicochemical findings of the nisin-treated burger were significantly reduced compared to those of the untreated- nisin samples at different stages during refrigeration. Increasing the nisin medium causes a significant delay in microbiological results. According to the organoleptic attributes, the nisin-treated burger will remain fresh (12 days) in the refrigerator at 4°C without losing its acceptability for consumers, although the untreated-nisin shelf-life samples were only prolonged to 9 days. In addition, the corresponding microbiological properties confirm the sensory result of the increase in shelf life. The outcomes showed that nisin is a biological preservative that enhances the quality of catfish burgers and improves the shelf-life during refrigeration, and the best treatment was 0.8% of samples tread with nisin. Based on the outcomes, compared to the control samples, nisin was more successful in enhancing the physico-chemical, microbiological, and organoleptic qualities of burger samples during refrigeration.

INTRODUCTION

Consumers prefer freshwater fish due to their high nutritional content and the high quality of their components; mineral salts, abundant biologically valuable proteins, and polyunsaturated fatty acids (omega 6 and omega 3), as well as their delectable taste and affordable cost **(Mendivil, 2021; Xiao *et al.*, 2022)**. One of the fishery products with a limited shelf-life is fish burgers. The primary cause of spoiling is microbial activity, which produces noxious and unacceptably off-flavoring compounds such as sulfides, alcohols, ketones, aldehydes, nitrogen-containing substances, and organic acids **(Iacumin*et al.*, 2022)**. Frozen or refrigeration does not completely prevent microbial and enzymatic processes and causes the spoilage of fishery products. Seafood spoilage is usually due to lipid oxidation, increased enzyme activity, metabolic activity loss, or /and growth of the microorganism **(Li *et al.*, 2011)**. The microbial activity during storage leads to increases in alcohols, biogenic amines, ketones, sulphides, aldehydes, and organic acids, with unpleasant flavors and odors **(Luan *et al.*, 2023)**.

Natural or synthetic food preservatives are frequently used to stop or regulate the growth of microbes and undesirable substances in food. Several prevention techniques, including the application of chemicals, can inhibit or prevent microorganisms in seafood. The application of chemicals can alter the nutritional value, organoleptic characteristics, and quality of seafood, as well as posing hazards to humans **(Özogul*et al.*, 2016)**. Hence, food companies, scientists, and governments around the world have redoubled their efforts to improve the safety of food. Therefore, a novel and attractive strategy to enhance the quality of seafood is using the natural flora and/or their antimicrobial activity as biopreservatives. Among these agents, nisin is widely utilized in food preservation due to its superior antibacterial properties against foodborne pathogens **(Imade*et al.*, 2021)**. Nisin as a natural agent was used in preserving fish fillets and fish products, as reported by different researchers such as **Behnam *et al.* (2015)**, **Zhang *et al.* (2021)** and **Arafat *et al.*(2022)**. However, the nisin effects were addressed on organoleptic, microbiological, and physico-chemical of catfishburger kept in refrigerated conditions.

MATERIALS AND METHODS

Fish collection and preparation

A weight of 27kg of *Clarias gariepinus* was obtained from a private catfish farm in the Fayom province of Egypt. Catfish fresh specimens were purchased from a fisherman during spring 2023. Following purchase, samples were chilled and delivered on the same day to a marine biology lab for further evaluation. Catfish specimens were washed in potable water, then slaughtered gutted, the head removed, filleted, and re-washed, then carefully drained. The stock solution of nisin (Sigma Egyption office) was prepared following the method of **Ucar*et al.* (2020)**.

Processing seafood

Four treatments were created as follows: (1) Control-NT fillets soaked in deionized water; (2) TI-NT soaked in 0.2% of nisin solution; (3) T2-NT soaked in 0.4% of nisin solution, and (4) T3-NT soaked in 0.8% of nisin solution (1:1, W/V). Catfish burger was prepared under hygienic conditions from minced catfish obtained after the trimming and filleting process using the technique reported by **Abbas (2021)**. Nisin treatments (NT) were blended with other burger ingredients as described by **Abou-Taleb (2022)**, with minor modifications (Fig. 1), and burger samples were packed in plastic bags and kept in the refrigerator at 4°C. The size of each burger slice (50g) was 7.5cm in diameter and 0.50cm in thickness. Spices and other ingredients were purchased form the local market in Cairo, Egypt. Physicho-chemial, microbiological and sensory analyses were performed on days 0, 3, 6, 9, and 12 days. Fig. (2) illustrates the procedures for burger preparation.

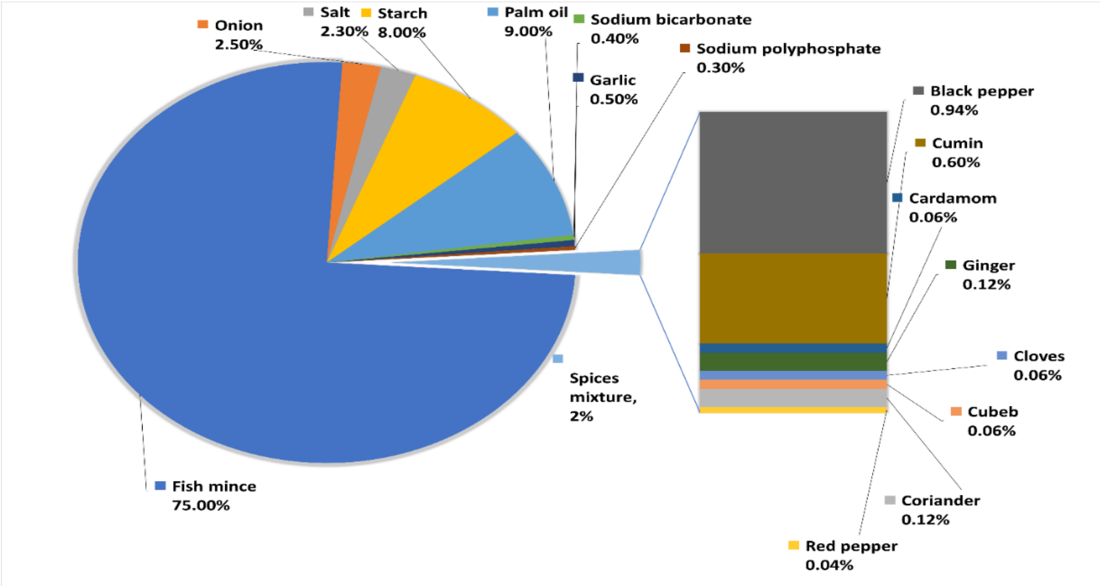

Fig., 1. The percentages of ingredients and spices for the catfish burger recipe.

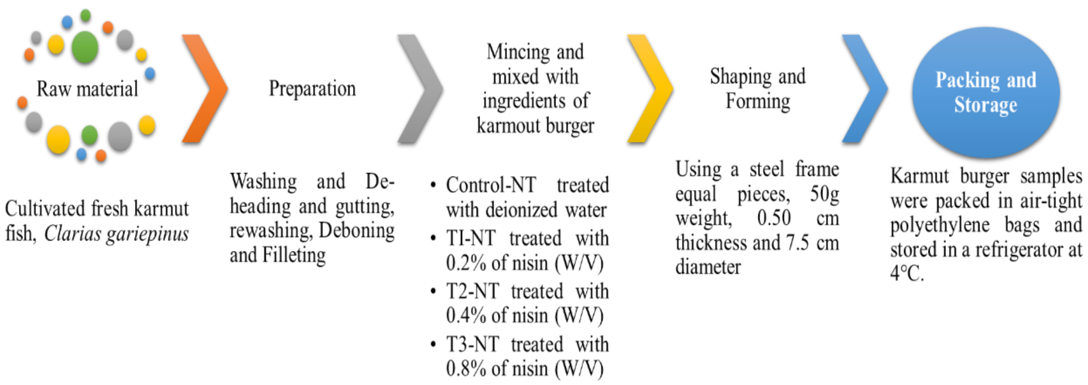

Fig. 2. Catfish burger treatments preparation scheme

Methodology

The microbial investigations (total bacterial count, TBC), psychrophilic bacteria (PB), lactic acid bacteria, (LAB), yeasts and molds (YM, **Difco Manual, 1984)**); thiobarbituric acid (TBA, **Ke *et al.*, 1984)**); total volatile bases nitrogen (TVB-N, **Pearson, 1993**); physico-chemical indices (pH value, **Vareltzis *et al.*, 1997)**); proximate composition (Carbohydrates, crude fat, ash, crude protein, moisture, and calorific value **(AOAC, 2012)**, and trimethylamine nitrogen (TMA-N,

AOAC, 2012) were carried out at regular intervals every 3 days on (zero time), 3, 6, 9, and 12 days with refrigeration storage.

Organoleptic evaluation

The burger slices (NT treatments) were fried on a prepared electric fryer for 5 minutes (each side for 2.5min.) at 170°C. The burger samples were sensory-evaluated by ten panelists, who examined the fried burger both immediately after processing and regularly every three days during refrigeration. According to **El-Sherif and Abd El-Ghafour (2016)**, panelists score on a hedonic scale of 0-3.9, 4-4.9, 5.0-6.5, 6.6-7.5, 7.6-8.5, and 8.6-10 (very bad, poor, accepted, good, very good, excellent, respectively) for taste, appearance, texture, odor, and overall acceptability.To verify, the effect of NT and time of storage on the burger sample shelf-life and quality, studies on physicochemical, microbiological, and organoleptic attributes were performed.

Statistical analysis

The data were statistically checked using SPSS 22, a statistical program. An analysis of variance was performed on proximate composition, physicochemical indices, and organoleptic evaluations, followed by Duncan's test. The level of significance was adjusted to $P< 0.05$, while the findings were tabulated as average±SD with superscript letters denoting a significant difference.

RESULTS

Proximate composition

The minced and burger proximate composition and physicochemical indices are displayed in Table (1). Minced catfish recorded a moisture content of 79.01±1.12%, crude protein of 17.56±0.58%, fat content of 1.88±0.04%, ash content of 1.17±0.07%, carbohydrate 0.38±0.01%, and a calorific value of 88.68±0.98 kcal $100g^{-1}$. Additionally, the TMA, TBA, pH, and TVB of the outcomes were 0.39±0.02 mg $100g^{-1}$, 0.28 ± 0.010 mg kg^{-1}, 6.65±0.24, and, 3.97±0.14 mg $100g^{-1}$, respectively.

After processing, the minced catfishwas characterized by significantly higher moisture, crude protein, and pH values compared to the burger samples. However, the minced catfishhad significantly lower crude fat, ash content, carbohydrate, and calorific values than the burger samples. In refrigeration, the initial values of the proximate composition, and the physicochemical indices of the burger treated with nisin showed non-significant differences at zero-time. Such changes between minced catfish and burger samples could be caused by ingredients in the burger during its preparation. Similar findings were suggested by **HassabAlla *et al.* (2009)** and **Emir-Çobanand Tuna-Keleştemur (2017)**. Similarly, **Lima-Veeck *et al.* (2018)** reported that the silver catfish, *Rhamdia quelen*, had a moisture content of 78.10 ± 0.60 %, ash of 1.20 ± 0.10 %, and 12.90 ± 1.10 % for protein; while for fat, it recorded a value of 4.20 ± 1.00 %.

Table 1. Proximate composition and physicochemical indices of minced and burger catfish at zero time

	Minced catfish	Catfish burger treatments			
		Control-NT	T1-NT	T2-NT	T3-NT
Proximate composition					
Moisture (%)	79.01±1.12[a]	66.65±0.99[b]	65.75±1.05[b]	65.99±1.08[b]	65.69±0.91[b]
Crude protein (%)	17.56±0.58[a]	15.92±0.98[b]	15.99±0.85[b]	16.03±0.75[b]	15.95±0.48[b]
Crude fat (%)	1.88±0.14[b]	2.88±0.17[a]	2.90±0.15[a]	2.87±0.12[a]	2.85±0.19[a]
Ash (%)	1.17±0.11[b]	3.66±0.20[a]	3.60±0.24[a]	3.59±0.31[a]	3.71±0.24[a]
Carbohydrate (%)	0.38±0.01[b]	10.89±0.85[a]	11.76±0.67[a]	11.52±0.95[a]	11.80±0.55[a]
Calorific value (kcal 100 g^{-1})	88.68±2.98[b]	133.16±3.25[a]	137.10±3.25[a]	136.03±2.55[a]	136.65±1.89[a]
Physicochemical indices					
pH value	6.65±0.24[a]	6.50±0.27[b]	6.51±0.19[b]	6.53±0.24[b]	6.48±0.24[b]
TVB (mg $100g^{-1}$)	3.97±0.14[b]	5.35±0.09[a]	5.29±0.85[a]	5.30±0.64[a]	5.34±0.58[a]
TBA (mg Malo./kg)	0.28±0.01[b]	0.34±0.01[a]	0.33±0.01[a]	0.35±0.01[a]	0.33±0.01[a]
TMA-N (mg $100g^{-1}$)	0.39±0.02[b]	0.49±0.01[a]	0.46±0.02[a]	0.48±0.01[a]	0.473±0.01[a]

*The significant variations at *P<0.05* are represented in each row with different lowercase letters, while the different uppercase letters in a column represent significant variations (*P*<0.05).

Physicochemical indices

One of the most significant parameters for preventing seafood spoilage and bacteria growth is pH **(Anvari *et al.*, 2013)**. The pH value in burger samples under refrigeration condition is illustrated in Fig. (3A). The pH values in the burger samples showed 6.50, 6.51, 6.53, and 6.48 for control-NT, T1-NT, T2-NT, and T3-NT, respectively, at zero-time. During refrigeration, it was gradually increased with increasing storage time and exhibited the highest value (8.02) in the untreated-NT burger samples, while they were arranged as T1-NT (7.00) > T2-NT (6.85) > T3-NT (6.77) on the 12th day. Our results are nearly similar to those of **Aref *et al.* (2016)**, **Rani *et al.* (2017)** and **El-Lahamy *et al.* (2018)**. According to **Saleem *et al.* (2019)**, the pH of fish and seafood products ranges between 6.3 and 7.0, where 7.0 is the accepted upper limit. During storage, the pH levels of the NT-treated burger samples reached a very acceptable level on the 12th of storage. In the NT-untreated samples; however, its value was rejected after 9 days (7.15) and 12 days (8.02) of storage. However, the pH increment in the burger sample was suggested as a result of the formation of nitrogen derivatives due to physicochemical alterations under refrigeration conditions. This observation agrees with those of **El-Sherif *et al.* (2011)** and **Saleem *et al.* (2019)** and disagrees with that of **Abou-Taleb (2022)** who observed a decrease in the pH value of fish burger during refrigeration.

The TVB content is one of the most employed fish and seafood quality indices (**Kostaki*et al.*, 2009**). **Li *et al.* (2012)** revealed that the maximal allowable limit for TVB in fish and seafood products ranges between 25 and 40 mg 100 g^{-1}, based on species. The TVB levels in burger samples under refrigeration condition is illustrated in Fig. (3B). The contents of TVB in burger samples at zero-time were 5.35, 5.29, 5.30, and 5.34 mg100 g^{-1} of control-NT, T1-NT, T2-NT, and T3-NT, respectively. During refrigeration, TVB levels in burger samples treated with NT remained below the acceptable limit throughout the entire refrigeration period at 4°C, while it was higher than this limit for the control-NT samples (36.98 mg $100g^{-1}$) and became rejected on the 12^{th} day. TVB content of burger samples increased sharply within the refrigeration period. The TVB of control-NT samples under refrigeration exhibited the highest content compared to NT-treated samples. Similar findings were stated by **Mahmoudzadeh *et al.* (2010)**, **Vanitha *et al.* (2013)**, **Çoban and Tuna-Keleştemur (2017)**, **El-Lahamy*et al.* (2018)** and **Abou-Taleb (2022)**. In this work, the increment in TVB-burger contents may be due to the ammonia formation, amino-acids deamination, and the production of volatile amines during refrigeration. The formation of alkaline media is related to the breakdown of nitrogenous substances, leading to an elevation in pH value of seafood during storage **(Sallam *et al.*, 2007**). Additionally, **Özogu*et al.* (2006)** revealed that the increase in TVB levels in fish and seafood during storage may be due to bacteria and enzymic spoilage.

Thiobarbituric acid (TBA) is a lipid rancidity indicator in fishery products and is often recorded as evidence of lipid oxidation **(Rezaei & Hosseini, 2008)**. The TBA levels in burger samples under refrigeration are represented in Fig. (3C). The initial values of TBA in burger samples at zero-time were 0.34, 0.33, 0.35, and 0.33 mg malonaldhyde/kg of control-NT, T1-NT, T2-NT, and T3-NT, respectively. During refrigeration, it increased gradually with increasing the storage periods, and the NT groups had lower TBA values than untreated groups during the different storage periods. Similar outcomes were suggested by **El-Hanafy *et al.* (2011), Yarnpakdee*et al.* (2012)**, **Gao *et al.* (2014)**, **Çoban and Tuna-Keleştemur (2017)** and **Abou-Taleb (2022)**. TBA indices in burger samples may be elevated due to fat degradation and production of secondary products at cold temperatures **(Amin, 2012)**. The ascending order of TBA values in the burger samples under refrigeration is as follows: control-NT > T1-NT > T2-NT > T3-NT. The elevation of TBA in burger samples with refrigeration time may be due to microbial spoilage, lipid autoxidation, and/or oxidative rancidity **(Osman & Zidan, 2014)**. According to **EOS (2010)**, the higher allowable limit for TBA in fish and seafood products is 4.5 mg malonaldhyde/kg. Based on this limit, TBA-burger levels for all nisin treatments were below the allowed limit at different refrigeration times; however, they were unacceptable in untreated samples (control-NT) at 9 and 12 days of storage (4.51 and 5.66 mg malonaldhyde/kg, respectively).

The TMA levels in burger samples under refrigeration are shown in Fig. (3D). The TMA levels in burger samples were 0.49, 0.46, 0.48, and 0.47 mg $100g^{-1}$ in the control-NT, T1-NT, T2-NT, and T3-NT, respectively, at zero-time. During refrigeration, the control-NT burger showed the maximal value of TMA (10.23 mg $100g^{-1}$) whereas, it was ordered in the NT samples in the following sequence of T1-NT (8.54 mg $100g^{-1}$) > T2-NT (7.39 mg $100g^{-1}$) > T3-NT (6.99 mg $100g^{-1}$) on the 12th day. According to **EOS (2010)**, the higher allowable limit for TMA in fish and seafood products is 10 mg100 g^{-1}. Based on this limit, TMA-burger contents in samples treated with NT were acceptable at the close of the 12^{th} day, while the NT-untreated samples were rejected on the 12th day. The increment in TMA-burger contents during refrigeration may be due to the breakdown of TMAO to trimethylamine via mechanism of non-enzymatic, either through native cell enzymes or through enzymes (trimethylaminase) that are not completely inhibited. Similar outcomes were mentioned by **Daboor and Ibrahim, (2008)**, **Saleem *et al.* (2019)**, **Abou-Taleb (2022)** and **Arafat *et al.* (2022)**.

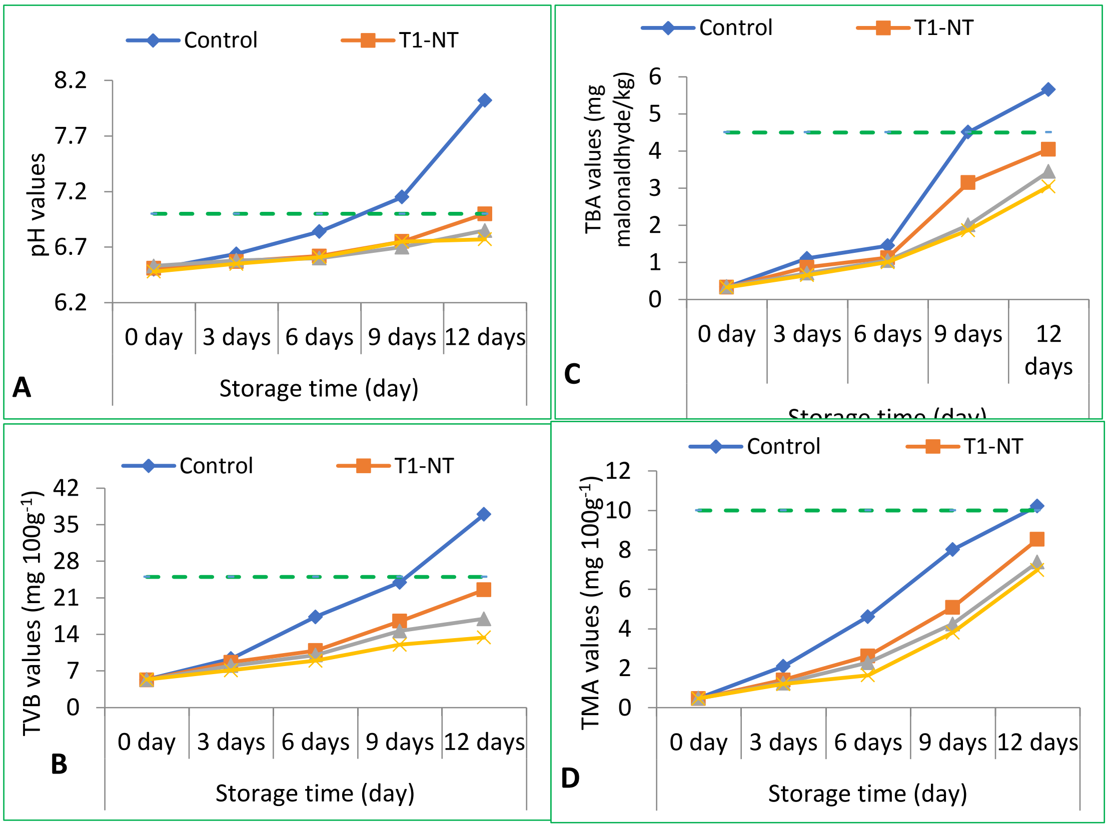

Fig. 3. Changes in physicochemical indices of catfish burger treated by NT during refrigeration at 4°C: **A)** pH, **B)** TVB, **C)** TBA, D) TMA.

Microbiological indices

TBC is used as an indicator of the acceptability of seafood because of the bacterial effect on the spoilage of seafood (**Emir-Çoban & Tuna-Keleştemur, 2017**). Fig. (4A) represents the TBC levels in burger samples under refrigeration. Initial values of TBC in burger samples at zero-time were 3.46, 2.59, 2.53, and 2.48 log-cfu g^{-1} of control-NT, T1-NT, T2-NT, and T3-NT, respectively, indicating a high burger quality. Similar outcomes were mentioned by **Kose *et al.* (2009)** and **Ehsani *et al.* (2014)**. During refrigeration, TBC-burger samples were increased at the end of storage to record 10.35, 5.95, 5.49, and 5.03 log-cfu g^{-1} of control-NT, T1-NT, T2-NT, and T3-NT, respectively. Consequently, compared to untreated-NT samples, using nisin for burger samples during refrigeration was more effective in reducing the rate of increasing TBC levels and extending the shelf-life. Nisin is effective on Gram-positive species, including microbial microbe spores **(Hampikyan & Ugur, 2007)**.

These outcomes are similar to those of **Lyhs *et al.* (2001)**, **Behnam *et al.* (2015)**, **Lima-Veeck *et al.*(2018)**, **Abou-Taleb (2022)** and **Arafat *et al.*(2022)**. According to **ICSMF (1986)**, the higher allowable limit for TBC in fishery products is 7 log-cfu g^{1}. Based on this limit, TBC-burger levels in all treatments with nisin were accepted on the 12^{th} day, while the untreated-NT samples were rejected after the 12^{th} day of storage (10.35 log-cfu g^{-1}). In accordance with this observation, the higher TBC-burger level during refrigeration can be explained by the high content of fatty acids and nitrogen components produced by the proteins and fats breakdown by fish enzymes, thus resulting in an optimal time for bacterial growth **(Aref *et al*., 2016)**.

Psychotropic bacteria, which are aerobic Gram-negative, are among the most common species in microbial spoilage at low temperatures **(Barkhordari & Bazargani-Gilani, 2021)**. Fig. (4B) represents the psychrophilic bacteria levels in burger samples under refrigeration. Initial values of psychrophilic bacteria in burger samples were significantly higher in control samples (2.87), compared to those treated with NT groups which in the following sequence, T1-NT (1.98 log-cfu g^{-1}) > T2-NT (1.87 log-cfu g^{-1}) > T3-NT (1.65 log-cfu g^{-1}). This delay in the rate of increasing TBC value in NT-treated burger samples compared to untreated burger could be due to nisin's more potent antibacterial activity. During refrigeration, the count of psychrophilic bacteria increased to recorded 6.85, 5.19, 4.79, and 4.11 log-cfu g^{-1} of control-NT, T1-NT, T2-NT, and T3-NT, respectively at 12^{th} days. However, psychrotrophic viable counts in burger samples showed increasing trends with an increase in storage under refrigeration. Furthermore, the decline in PVC values in NT-treated burger samples compared to the NT-untreated ones could be explained by the nisin's antibacterial action in the burger samples kept at cold temperatures. Similar findings were suggested by **Behnam *et al.* (2015)**, **Veeck *et al.* (2018)** and **Arafat *et al.* (2022)**.

Lactic acid bacteria, as pathogens classified as Gram-positive, are anaerobes with facultative characteristics that may grow in both aerobic and anaerobic environments **(Mexis *et al*., 2009)**. Data in Fig. (4C) reveal that the initial counts of LAB in burger samples were significantly higher in the control samples (3.14 log-cfu g^{-1}), compared to NT-treated groups which in the following sequence, T1-NT (2.12 log-cfu g^{-1}) > T2-NT (2.06 log-cfu g^{-1}) > T3-NT (2.00 log-cfu g^{-1}). During refrigeration, the maximum counts of LAB in the NT-untreated burger samples was 6.22 log-cfu g^{-1}, while NT-treated burger groups varied from 2.94 to 4.56 log-cfu g^{-1} on the 12^{th} day. Data previously noted that the bactericidal effect of nisin focused on bacteria of Gram-positive, hence, the pattern seen in counts of LAB in NT-treated burger samples was to be expected. In general, the results agree with those of **Mexis*et al.* (2009)** and **Behnam *et al.*(2015)**.

Yeast and mold counts in refrigerated burger samples are represented in Fig. (4D). Initial values for yeast and mold count in the burger samples were 3.14, 3.08, 3.10 and 3.09 log-cfu $g^{-1}$of control-NT, T1-NT, T2-NT, and T3-NT, respectively. During refrigeration, the number of yeasts and molds increased on day 12 to 8.22, 6.33, 5.04 and 4.71 log-cfu $g^{-1}$of control-NT, T1-NT, T2-NT, and T3-NT, respectively. The counts of yeast and mold in the burger groups increased with increasing refrigeration period, and they declined in NT-treated burger groups compared to NT-untreated samples. Similar outcome was suggested by **Abou-Taleb (2022)** and **Arafat *et al*. (2022)**. Based on the microbial indices, they reported that the burger samples have a significant effect on the reduction of microbial growth stored under refrigeration.

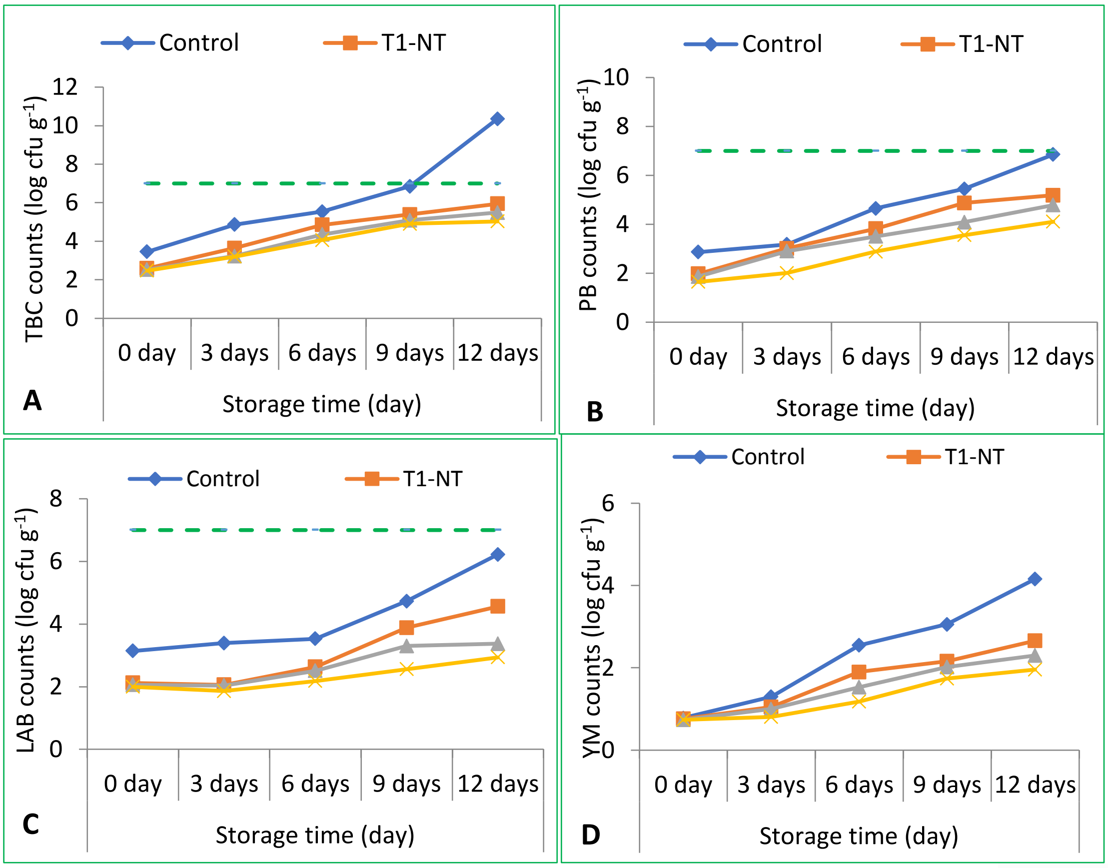

Fig. 4 Changes in microbiological indices of catfish burger treated by different treatments of nisin during refrigeration at 4°C: **A)** TBC, **B)** PB, **C)** LAB, **D)** YM.

Organoleptic evaluation

The most crucial quality criteria for burger samples are presumed to be their organoleptic characteristics, which also have a significant effect on the acceptability of consumer.The organoleptic evaluation (texture, taste, odour, appearance, and overall acceptability) of burger treated with NT (T1-NT, T2-NT, and T3-NT) and NT-untreated (control-NT) samples were cooking by frying the KN samples immediately after processing and every 3 days of refrigeration to evaluate the effect of NT and refrigeration on the quality aspects. Outcomes of Table (2) reveal that the scores of odor, appearance, texture and taste reported non significant difference ($P < 0.05$) between samples of NT-treated and untreated at zero-time and were of excellent quality. However, it was a significant decline in sensory scores during refrigeration for NT-treated and untreated burger samples. As a result, the burger samples performed both very good and excellent scores on the first three days of storage before deteriorating to display moderate scores on days 6 through 9 of refrigeration, while the NT-untreated burger samples were rejected at the end of 12 days, whereas the NT-treated burger samples continued to be of an acceptable quality until the storage ended at that point. This is possible because nisin is a potent antimicrobial agent that can be used to prevent the spread of food spoilage pathogens. The outcome suggests that as the nisin level increases, the storage efficiency of catfish burger also improves. These findings agree with those of **Zakipour*et al.* (2013)** and **Zhang *et al.* (2021)** who postulated that, the nisin-treated fillets of rainbow trout enhance the sensory aspects as well as extend the shelf-life.

Table 2. The texture, taste, odor, and appearance, as organoleptic evaluation of KB treated with NT during refrigeration at 4°C

Organoleptic evaluation		KB treatments			
		Control-NT	T1-NT	T2-NT	T3-NT
Appearance					
	0 day	9.82±0.23^{aA}	9.70±0.35^{aA}	9.80±0.65^{aA}	10.00±0.00^{aA}
	3 days	8.56±0.54^{bB}	9.00±0.23^{aB}	8.97±0.54^{aB}	9.00±0.41^{aB}
	6 days	7.52±0.62^{cC}	8.60±0.54^{bC}	8.54±0.74^{aB}	8.54±0.61^{aC}
	9 days	6.50±0.65^{dD}	7.50±0.57^{cD}	7.80±.0.84^{bC}	8.20±0.42^{aC}
	12 days	5.60±0.47^{dE}	7.00±0.68^{cE}	7.50±0.65^{bD}	7.90±0.74^{aD}
Odor					
	0 day	9.50±0.40^{aA}	9.60±0.12^{aA}	9.70±0.13^{aA}	9.60±0.32^{aA}
	3 days	8.25±0.56^{bB}	8.90±0.26^{aB}	9.00±0.10^{aB}	8.90±0.45^{aB}
	6 days	7.50±0.74^{cC}	8.60±0.34^{bC}	8.60±0.33^{bC}	8.80±0.85^{aC}
	9 days	6.30±0.64^{dD}	7.60±0.51^{cD}	7.80±0.45^{bD}	8.20±0.32^{aD}
	12 days	4.70±0.82^{dE}	6.70±0.47^{cE}	7.30±0.51^{bE}	7.60±0.48^{aE}
Taste					
	0 day	9.60±0.21^{aA}	9.70±0.10^{aA}	9.60±0.10^{aA}	9.70±0.21^{aA}
	3 days	8.00±0.64^{bB}	8.85±0.42^{aB}	8.85±0.33^{aB}	9.00±0.45^{aB}
	6 days	7.50±0.56^{cC}	8.00±0.42^{cC}	8.30±0.66^{bC}	8.50±0.25^{aC}
	9 days	6.00±0.51^{cD}	7.60±0.38^{bD}	7.60±0.32^{bD}	8.20±0.65^{aC}
	12 days	4.50±0.89^{dE}	6.70±0.66^{cE}	7.00±0.35^{bE}	7.50±0.47^{aD}
Texture					
	0 day	9.70±0.11^{aA}	9.60±0.09^{aA}	9.70±0.10^{aA}	9.80±0.15^{aA}
	3 days	8.45±0.50^{bB}	8.70±0.39^{aB}	8.70±0.26^{aB}	8.80±0.14^{aB}
	6 days	7.70±0.74^{dC}	7.90±0.88^{cC}	8.00±0.66^{bC}	8.30±0.85^{aC}
	9 days	6.50±0.50^{dD}	7.60±0.47^{cC}	7.80±0.58^{bC}	8.00±0.45^{aC}
	12 days	5.00±0.28^{cE}	7.00±0.60^{bD}	7.00±0.49^{bD}	7.50±0.36^{aD}

*Hedonic scale of very bad, poor, accepted, good, very good, and excellent 0-3.9, 4-4.9, 5.0-6.5, 6.6-7.5, 7.6-8.5, and 8.6-10, respectively. However, the significant variations at $P<0.05$ are represented in each row with different lowercase letters, while the different uppercase letters in a column represent significant variations.

Generally, the overall acceptability of NT-treated and untreated burger samples exhibited high grades in the first refrigeration periods and gradually declined until the end of storage periods (Fig. 5). During refrigeration, the burger samples were of highly acceptable quality even until the storage ended (12 days), while the NT-control samples were spoiled on day 9 of refrigeration period. Moreover, the shelf-life of NT-treated burger samples was exhibited to be 12 days while 9d for the NT-untreated groups. According to the present study outcomes, nisin treatment can effectively reduce microbial growth, reduce chemical spoilage, preserve/improve sensory features, and prolong the shelf-life of burger groups throughout 12 days under refrigeration.

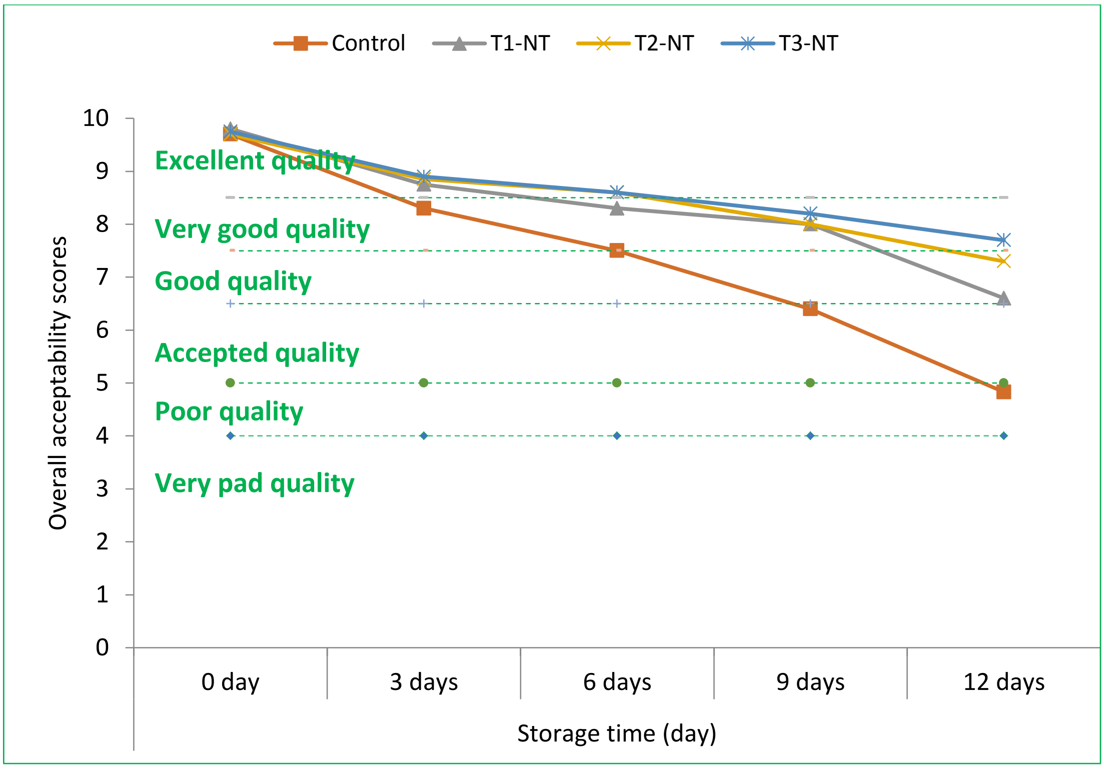

Fig. 5. The overall acceptability of catfish burger treated by NT during refrigeration at 4°C

CONCLUSIONS AND RECOMMENDATION

The indices of physicochemical and microbiological spoilage were performed, followed by addressing the organoleptic characteristics of burger during refrigeration (4°C) in order to comprehend the nisin influence, as a bio-preservative, on the shelf-life and qualities aspects of catfish burger. Nisin-treated burger samples recorded a significant decrease in the physicochemical indices ($P<0.05$), comared to NT-untreated burger groups, following the sequential order of T3-NT < T2-NT < T1-NT < control-NT. However, the pH, TVB, TBA, and TMA contents in the burger control samples exceeded the allowed limit by day 12, whereas the NT-treated burger samples remained within the permissible limit. The results obtained from the analysis of the TBC, PB, YM, and LAB as microbiological indices are in an excellent agreement with the physico-chemical findings, where the NT-control groups showed a significant (*P*<0.05) increase in the microbial growth compared to nisin-treated samples. The corresponding microbiological assessment also confirmed the reduced shelf-life of the NT-untreated burgers. The results of this study showed that nisin extended shelf-life to day 12 for storage, compared to the 9day- period of storage for Nt-untreated burger groups. According to the results of this study, the studied nisin can be considered a potential source of antimicrobial agents in seafood and pharmaceuticals.

HIGHLIGHT

- Nisin application preserves catfish burger's organoleptic quality maintained and increases shelf life.
- Nisin reduced bacterial growth on catfish burger during refrigeration.
- Nisin-ureated fish burgerwere found to be of a higher quality than the untreated burgers.
- Physico-chemical outcomes showed that the nisin application has reduced oxidative rancidity.

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