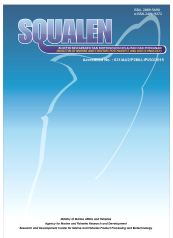

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Quality of Refrigerated Tilapia *(Oreochromis niloticus)* Slices under Vacuum and Modified Atmosphere Packaging

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Abstract

In order to develop a proper packaging system for sliced tilapia fish (*Oreochromis niloticus*), the biochemical and microbiological qualities under control (unsealed package), vacuum package and modified atmosphere packaging with 50% CO_2/50% N_2 (MAP-1) and 50% CO_2/50% O_2 (MAP-2) were evaluated every three days during 18 days of chilled storage (4±1°C). The pH value was not significantly different (p > 0.05) by treatments until day 9, but significantly lower values (p < 0.05) were observed on day 12 of the storage in all treated samples compared to the control. The total volatile base nitrogen (TVB-N) value progressively increased, but not significantly different (p > 0.05) in all treatments during the entire storage period. The thiobarbituric acid reactive substances (TBARS) amounts were significantly lower (p < 0.05) on day 12 in the vacuum and MAP-1 samples compared to the control sample, and were significantly higher (p < 0.05) on day 6, 9, and 12 of the storage period in MAP-2 samples compared to the control, vacuum and MAP-1 samples. The amounts of pH, TVB-N, and TBARS in all samples did not exceed the acceptable limit in almost the entire storage. The total viable count (TVC) progressively increased with storage time. Nevertheless, TVC values were lower (p < 0.05) on day 6, 9, and 12 of the storage periods in all treatments compared to the control. The TVCs exceeded the acceptable limit (7 log CFU/g) on days 6-9 for control, 9-12 for vacuum, day 12 for MAP-2, and 15 for MAP-1 sample during the storage period. Therefore, the MAP has shown promising results for shelf life extension that can be practiced to display the fishery products with prolonged shelf life.

Keywords: Tilapia, vacuum packaging, modified atmosphere packaging, quality, shelf life

1. Introduction

Tilapia (*Oreochromis niloticus*) is a freshwater fish commonly known as Nile Tilapia under the family of *Cichlidae*. Tilapia is native to Africa, and later it has been widely introduced for culture into tropical, subtropical, and even in temperate countries. It is a fast-growing species and has become the second most common farm-raised fish in the world (Gupta & Acosta, 2004). This fish is a popular cultured fish since it supports high-density culture system, resist hard environmental stress, tolerates low oxygen concentration, and is suitable for pond and cage cultures (DeLong, Losordo, & Rakocy, 2009). It contains several biochemical compositions in human diets. The proximate composition of tilapia fillets is moisture 77.6%, proteins 19.5%, total lipids 2%, and ash 1.8% on a wet basis (Bogard et al., 2015). The protein of tilapia is complete because it contains all essential amino acids. The omega-3 fatty acid is also found in tilapia muscle (Gonzales & Brown, 2006). It is regarded as a vitamin-rich fish containing vitamins A, D, E, B1, B3, B5, B6, B12, ascorbic acid, and folic acid. Several minerals are found in the tilapia fish body, including zinc, iron, iodine, calcium, selenium, magnesium, sodium, phosphorus, manganese, potassium, copper, sulfur, and chromium (Gonzales & Brown, 2006; Bogard et al., 2015).

With changing lifestyles, the buying behavior of the customers is also changed. The root causes of this change are economic development and social status. Nowadays, city-dwellers seek ready-to-cook

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or semi-prepared foods instead of raw foods due to their busy life. There are no such fish products available in the market of developing countries, including Bangladesh (Amanullah, 2019). To fulfill this demand, the superstores of the country could play an important role. However, the short shelf life is a limiting factor for perishable items like fish, even under the refrigerated condition in the superstores (Ashie, Smith, & Simpson, 1996). Extension of shelf life is significantly beneficial for the fish processing industries, as it can minimize the losses occurred at the stages of distribution and display. Some modern methods are developed for preserving the fish by technologists to fulfill the consumer demand for fresh fish with a reasonable price (Nagarajarao, 2016). In this respect, the packaging technology is receiving growing attention. Vacuum packaging and modified atmosphere packaging (MAP) are receiving increasing attention as preservation methods along with refrigerated storage to prolong the shelf life of fresh fish and meat (Narasimha Rao & Sachindra, 2002).

In case of MAP, various gases such as N_2, CO_2, O_2, and CO have been used. Among these, CO_2, perhaps the most commonly used and effective gas along with other gases (DeWitt & Oliveira, 2016). Carbon dioxide is soluble in both water and lipids. It has a significant role as a bacteriostatic effect in MAP technology. It reduces the growth of microorganisms at the log phase (Alfaro, Hernández, Le Marc, & Pin, 2013). However, the effects are varied with CO_2 concentration, microbial load, storage temperature, and the type of packaged product (DeWitt & Oliveira, 2016). On the other hand, oxygen is used in MAP technology to prevent the growth of exclusively anaerobic bacteria and the toxin production by *Clostridium botulinum* type E (Pantazi, Papavergou, Pournis, Kontominas, & Savvaidis, 2008). Oxygen is also used to avoid the discoloration of the flesh (Soccol & Oetterer, 2003). Therefore, the application of MAP technology has become more popular with increasing consumer awareness and public demand (Sveinsdóttir, Martinsdóttir, Hyldig, & Sigurgísladóttir, 2010).

The MAP has been widely used as a packaging technique to extend the shelf life up to 25-400% of fish, meat, and various products in developed countries (DeWitt & Oliveira, 2016). The extension of shelf life in a vacuum packaging and MAP depends on raw materials, gas mixtures, temperature, and packaging materials. This packaging has been studied mostly for marine fishes (Ruiz-Capillas & Moral, 2001; DeWitt & Oliveira, 2016). However, not many studies were found on MAP for freshwater fishes such as tilapia, sutchi catfish, and carps (Li et al., 2017; Masniyom, Benjama, & Maneesri, 2013; Noseda et al., 2012). This form of packaging has not yet evolved in the market of developing countries, i.e., Bangladesh. Therefore, the current study was aimed to develop a proper vacuum packaging and MAP for this tilapia fish to extend the shelf life, which can ensure the supply of quality fishes conveniently.

2. Material and Methods

2.1. Sample Collection and Preparation

Fifty tilapia fishes (*Oreochromis niloticus*) with the individual weight of 0.5±0.1 kg were brought from a farmer in Puthia Upazila, Rajshahi - Bangladesh. The fish samples were transported in live condition to the Quality Control Laboratory of the Department of Fisheries, Rajshahi University. Then fishes were stunned by a percussive method and killed by cutting the head portions (Lines & Spence, 2012). Then fishes were dressed and cut into small slices with an average weight of 100±10 g. Fish slices were washed twice with running tap water at room temperature (25°C), and the last wash was performed with distilled water. After washing, all fish slices were kept in ice to preserve the freshness until packaging.

2.2. Packaging and Storage of Sample

Fish slices weighing approximately 200 g were placed in plastic pouches having high gas and moisture barrier properties. The multi-layered transparent pouch (polyethylene/polyamide/ polyethylene) having 100 µm densities was used for this purpose (Premiaflex Plastics Ltd., Dhaka). Four packaging systems were prepared as follow: (1) unsealed package as control; (2) vacuum package as treatment-1; (3) MAP-1 (50% CO_2/50% N_2) as treatment-2 and (4) MAP-2 (50% CO_2/50% O_2) as treatment-3. Vacuum packaging and MAP were carried out by the Multivac packaging unit (C100, Haggenmuller, Germany) fitted with a Gas Mixer (KM100-3 MEM, WITT, Germany) as instructed in the manual. After packaging, the amount of O_2, N_2, and CO_2 gas were randomly monitored in the headspace of some of the packaged samples by a gas analyzer (Oxybaby M+, WITT, Germany) to ensure whether the gas ratio was correct. Packaged samples were preserved at 4±1°C in a refrigerator (GL-C322RLBB-PZ, LG, South Korea) for 18 days. Three samples of each treatment were taken every three days for analysis in the laboratory.

2.3. Biochemical and Microbiological Analyses

Various microbiological and biochemical analyses were performed to determine the shelf life of sliced tilapia fish stored at refrigerated temperatures. A 10 g of cut fish flesh was homogenized upon adding 50 mL of distilled water and then the pH of the homogenate was determined by a pH meter (HI2002, Hanna Inst, USA) (Binsi et al., 2015). Total volatile base nitrogen (TVB-N) was measured following European Commission (2005) method. According to Park et al. (2007), thiobarbituric acid reactive substance (TBARS) was estimated using the colorimetric method. Total viable count (TVC) was determined on plate count agar (Sigma-Aldrich, USA) by standard pour plate method following the decimal dilution technique by American Public Health Association (APHA) (1992) and expressed as log colony-forming units (log CFU/g). Plates were kept for 48 h in an incubator (Poleko, Poland) at 35°C, and later colonies were counted. The total coliforms and total fecal coliforms were enumerated by the method described by Food and Drug Administration (FDA) (2002) using nine tubes, the most probable number (MPN) test. In this case, only one sample from each packaging system was performed.

2.4. Statistical Analysis

All experiments were performed in triplicate. Data were subjected to analysis using one-way ANOVA. Tukey test was performed to measure the significant differences ($p < 0.05$) among treatments. Statistical analysis was conducted using the program SPSS Version 20 (IBM, IL, US).

3. Results and Discussion

3.1. pH Value

pH is an indicator factor for assessing the quality of fish and fishery products. The appropriate post mortem pH level is typically 6.8~7.0 (Metin, Erkan, Varlik, & Aran, 2001). In this study, the initial pH values of freshly sliced tilapia was 6.79. High initial pH values followed by declines were found for some treatments, and later increasing trends were evident. Nevertheless, until day 9 of the storage period, the pH values ($p > 0.05$) were not significantly affected by the treatments (Table 1). Significantly ($p < 0.05$) lower pH values were found in all treatments as compared to the control group on day 12 of the storage period. Similarly, no significant variations in pH values were observed under different packaging systems (air, vacuum, and MAP) at 1°C for tilapia fish (Cyprian et al., 2013) and at 4°C for rohu fish (Das, Alice, Hossain, Mehbub, & Islam, 2020). In another study, significantly no differences in pH values were observed for ready-to-cook (RTC) pangas curry among the treatments (unsealed package, MAP-1 with 50% CO_2/50% N_2, and MAP-2 with 75% CO_2/25% N_2) at 4°C during storage (Nayma, Das, Alice, Mehbub, & Islam, 2020). Lactic acid production via anaerobic glycolysis and the release of inorganic phosphates as a result of ATP degradation seemed to be the main reasons for this initial decreased pH values (Ayala et al., 2010). In 22 days of ice preservation, Ayala et al. (2010) have observed a similar trend of results for sea bream. On the other hand, the reaction between CO_2 and water forming carbonic acid that creates an acidic condition in the fillets may explain this decreasing pH (Velu, Abu Bakar, Mahyudin, Saari, & Zaman, 2013). pH value reached the acceptable limit on days 9-12 for the control, day 12-15 for vacuum, and day 18 of the storage period for MAP-2 sample (Table 1). Increased pH at the later storage stage could be due to the accumulation of amines and other volatile compounds through the autolytic and microbial activity on proteins and other compounds (Binsi, Shamasundar, & Dileep, 2007).

Table 1. pH value of sliced tilapia *(Oreochromis niloticus)* under vacuum packaging and MAP systems during storage at 4±1°C

Treatments	Storage Period (days)						
	0	3	6	9	12	15	18
Unsealed package (Control)	6.59±0.05^a	6.64±0.18^a	6.53±0.40^a	6.78±0.10^a	7.39±0.1^b		
Vacuum package	6.59±0.05^a	6.70±0.17^a	6.74±0.27^a	6.54±0.43^a	6.88±0.16^a	7.09±0.21	
MAP-1 (50% CO_2/50% N_2)	6.59±0.05^a	6.43±0.09^a	6.47±0.15^a	6.61±0.09^a	6.72±0.08^a	6.46±0.07	6.80±0.06
MAP-2 (50% CO_2/50% O_2)	6.59±0.05^a	6.56±0.09^a	6.33±0.23^a	6.18±0.07^a	6.67±0.01^a	6.81±0.18	7.00±0.04

Values (n=3) are expressed as mean ± SD. The values in the same column with various superscript letters reflect the significant difference at $p < 0.05$.

3.2. Total Volatile Base Nitrogen (TVB-N) Value

In general, the chemical degradation of food is estimated by measuring changes in TVB-N content, which is the sum of all chemicals including trimethylamine, dimethylamine, and ammonia. It is commonly used as an indicator for predicting bacterial spoilage of fish (Wu & Bechtel, 2008). The TVB-N amount is generally considered to be an acceptability limit for ice-stored fish at 30–35 mg N/100 g fish (Connell, 1995).

In the current study, the TVB-N content was initially 1.68±0.40 mg/100 g and then progressively increased over time for the whole storage period in all samples. The maximum TVB-N content reached 7.56±1.98 mg/100 g on day 12 of the storage for the control sample (Table 2). Increasing TVB-N value in refrigerated storage could be due to the production of basic compounds via microbiological action on protein and non-protein nitrogenous compounds. On and after 9 days of storage, the TVB-N rise becomes apparent when pH values were also comparatively higher. This condition might be correlated with deaminase activity in which favored under alkaline medium (Soccol, Oetterer, Gallo, Spoto, & Biato, 2005). However, the increase in TVB-N contents of all packaged samples did not exceed the acceptable limit (30-35 mg/100g) (Table 2). Similarly, TVB-N values did not cross the acceptable limit in the case of sliced rohu fish (Das et al., 2020) and goonch fish (*Bagarius bagarius*) (Alice, Amanullah, Karim, Hossain, & Islam, 2020) under the same packaging and storage conditions. During storage, the MAP samples are likely to have lower TVB-N content than the control and vacuum pack samples. TVB-N content was not significantly influenced ($p > 0.05$) by treatments during each of the storage days (Table 2). Soccol et al. (2005) found similar results with no significant differences in TVB-N contents of tilapia fish among treatments (air package, vacuum package, and a MAP with 60% CO_2/40% O_2) during 20 days storage at 1°C. Significantly higher levels of TVB-N content (more than 30 mg/100 g) were observed in tilapia fish under air, vacuum and MAP (60% CO_2, 10% O_2 & 30% N_2) at the end of 18 days of storage at 4°C (Masniyom et al., 2013) compared with the current study.

3.3. Thiobarbituric Acid Reactive Substance (TBARS) Value

TBARS is an important parameter for measuring secondary lipid oxidation products, including malonaldehyde (MDA). TBARS content of 2 mg of MDA/kg fish is the acceptable limit, and an unpleasant odor and taste have emerged in fish beyond this limit (Connell, 1995). In this study, the TBARS amount was initially 0.26 mg MDA/kg tilapia fish. The TBARS amounts remained nearly steady, with some fluctuation for vacuum and MAP-1 samples over the entire storage period. On the other hand, TBARS amounts gradually increased until day 3 for MAP-2 and day 9 for the control sample and rose sharply in the rest of the storage. The TBARS amounts were significantly ($p < 0.05$) influenced by treatments in the storage period. The TBARS amounts of the vacuum and MAP-1 (50% CO_2/50% N_2) samples were lower at the end of the storage than the control and MAP-2 samples (Table 3). A similar trend of TBARS value was observed for sliced goonch fish under the same packaging and storage conditions (Alice et al., 2020). It has possibly occurred for the absence of O_2, which delayed the oxidation of the polyunsaturated fatty acids (PUFA). This finding is agreed with Masniyom

Table 2. TVB-N value (mg/100g) of sliced tilapia *(Oreochromis niloticus)* under vacuum packaging and MAP systems during storage at 4±1°C

Treatments	Storage Period (days)						
	0	3	6	9	12	15	18
Unsealed package (Control)	1.68±0.40^a	2.24±0.40^a	3.50±0.99^a	4.76±0.40^a	7.56±1.98^a		
Vacuum package	1.68±0.40^a	2.80±0.00^a	3.64±0.40^a	5.88±0.40^a	6.44±0.20^a	7.14±0.20	
MAP-1 (50% CO_2/ 50% N_2)	1.68±0.40^a	2.10±0.59^a	3.36±0.00^a	2.66±0.59^a	5.04±0.40^a	5.32±0.40	6.10±0.48
MAP-2 (50% CO_2/ 50% O_2)	1.68±0.40^a	2.24±0.00^a	3.22±0.20^a	3.80±0.62^a	4.70±1.27^a	6.10±0.68	5.74±0.99

Values (n=3) are expressed as mean ± SD. The values in the same column with various superscript letters reflect the significant difference at $p < 0.05$.

Table 3. TBARS value (mg MDA/kg) of sliced tilapia *(Oreochromis niloticus)* under vacuum packaging and MAP systems during storage at 4±1°C

Treatments	Storage Period (days)						
	0	3	6	9	12	15	18
Unsealed package (Control)	0.26 ± 0.08^{a}	0.28 ± 0.0^{a}	0.46 ± 0.19^{ab}	0.62 ± 0.08^{a}	1.59 ± 0.03^{b}		
Vacuum package	0.26 ± 0.08^{a}	0.30 ± 0.03^{a}	0.29 ± 0.08^{a}	0.60 ± 0.05^{a}	0.41 ± 0.09^{a}	0.71 ± 0.07	
MAP-1 (50% CO_2/ 50% N_2)	0.26 ± 0.08^{a}	0.36 ± 0.01^{a}	0.53 ± 0.17^{ab}	0.53 ± 0.03^{a}	0.52 ± 0.15^{a}	0.77 ± 0.11	1.04 ± 0.07
MAP-2 (50% CO_2/ 50% O_2)	0.26 ± 0.08^{a}	0.34 ± 0.04^{a}	0.84 ± 0.16^{b}	1.79 ± 0.14^{b}	1.96 ± 0.12^{c}	2.83 ± 0.24	4.21 ± 0.19

Values (n=3) are expressed as mean ± SD. The values in the same column with various superscript letters reflect the significant difference at $p < 0.05$.

et al. (2013), who noted that modified atmosphere and vacuum packaged tilapia fish had the lower MDA in the absence or lower of O_2 compared to the control sample. On the other hand, the products packed under MAP-2 in the current study (which contains 50% of O_2) presented higher ($p < 0.05$) TBARS value on day 6, 9, and 12 of storage compared to the control (which contains approximately 21% of O_2 (data not shown)), vacuum and MAP-1 samples (Table 3). Except for the MAP-2, no samples crossed the acceptable limit (2 mg MDA/kg) over the entire storage time. Similar results were observed for tilapia fillets under control, vacuum, and MAP with 50% CO_2 /50% N_2 conditions at the same storage temperature (Lázaro, Monteiro, & Conte-Junior, 2020). The MAP-2 (50% CO_2/50% O_2) sample exceeded the limit on and after day 12 of the storage (Table 3). The higher amounts of TBARS in the MAP-2 sample may be due to the higher rate of secondary lipid oxidation relative to the other packed samples since it contained a higher amount of O_2 (50%) along with CO_2. Ruiz-Capillas and Moral (2001) found similarly higher TBARS amounts for CO_2-enriched packaging systems. It was probably the result of a synergistic reaction between CO_2 and O_2, which eased the autoxidation of PUFA (Ruiz-Capillas & Moral, 2001). However, the levels of TBARS may not indicate the actual degree of fat oxidation because MDA may interact with other fish flesh compounds such as amines, nucleic acid and nucleosides, amino acids, proteins, and other aldehydes (Domínguez et al., 2019).

3.4. Total Viable Count (TVC)

In the current study, the initial TVC of sliced tilapia was 4.17 log CFU/g, which indicates an acceptable fresh quality. The bacteria counts of 2 - 6 log CFU/g are considered the satisfactory initial standard of freshly caught freshwater fish reported for tilapia, rainbow trout, silver perch, and sea bass (Gelman, Glatman, Drabkin, & Harpaz, 2001). This initial TVC value was lower than that of recently studied tropical fishes such as 4.32 log CFU/g for pangasius catfish (Islam et al., 2020), 4.29 log CFU/g for rohu fish (Das et al., 2020), and 5.22 log CFU/g for goonch fish (Alice et al., 2020). With increasing storage time under all treatments, the TVCs of sliced tilapia gradually increased. The TVCs of the control sample quickly raised to 7 log CFU/g within eight days from the initial value. TVCs were initially not significantly influenced by the treatments ($p > 0.05$). However, significantly ($p < 0.05$) lower TVCs were found on day 9 and 12 of the storage period in all treatments in comparison to the control (Figure 1). International Commission of Microbiological Specification for Food (ICMSF) (1986) suggested that the TVC value of 7 log CFU/g is recommended for fresh and frozen fish as the upper acceptability limit. The TVCs surpassed the 7 log CFU/g on days 6-9 for control, days 9-12 for vacuum, day 12 for MAP-2, and day 15 during the storage period for MAP-1 sample (Figure 1). Based on the acceptable limit of TVC, the shelf life was estimated at 6-9 days for control, 9-12 days for vacuum package, 12 days for MAP-2, and 15 days for MAP-1 sample. In the previous studies, comparatively lower shelf life for sliced goonch fish (Alice et al., 2020) and higher shelf life for rohu fish (Das et al., 2020) were observed under the same packaging and storage conditions. Lower TVCs of the samples kept under MAP directed that 60% CO_2 effectively inhibited the microbial growth as CO_2 shows more effects as an antibacterial agent at a higher concentration (Farber, 1991). The high CO_2 levels inhibited psychotropic bacteria's growth during

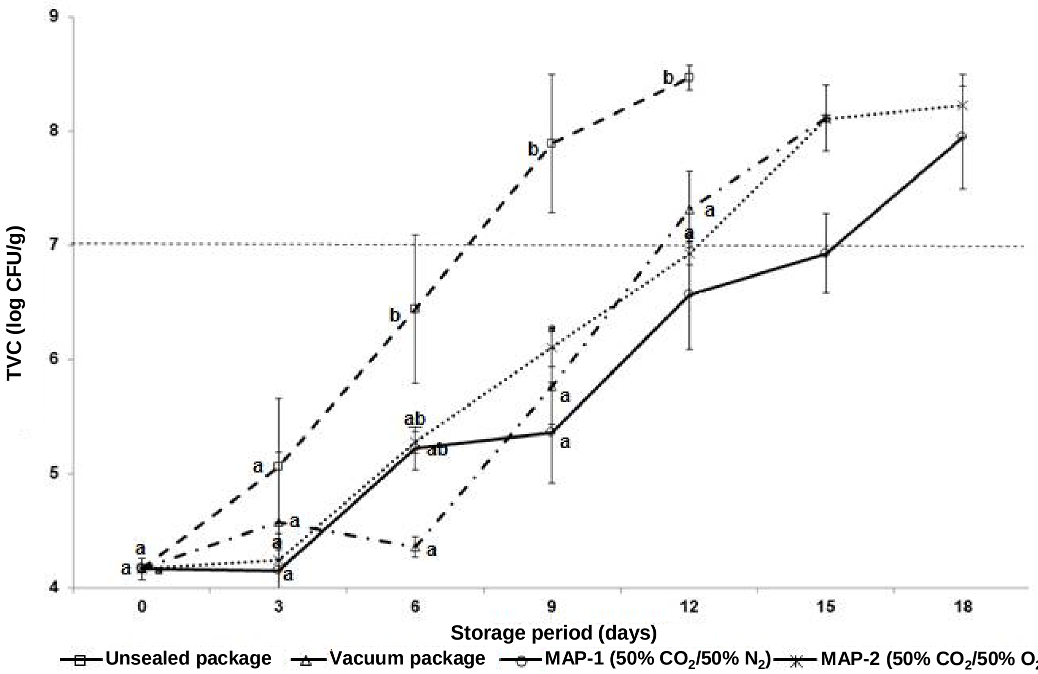

Figure 1. Changes in total viable counts (log CFU/g) of sliced tilapia *(Oreochromis niloticus)* under vacuum packaging and MAP systems during storage at 4±1°C. Values (n=3) are expressed as mean ± SD. The values on the same day with different letters reflect the significant difference at $p < 0.05$

fish storage at low-temperature, indicating the high CO_2 sensitivity of those bacteria (Masniyom, Benjama, & Maneesri, 2011). On the other hand, lower TVCs of samples kept under vacuum packaging compared to that of air indicated that the absence of oxygen could hinder the growth of aerobic spoilage bacteria, predominantly *Aeromonas* spp. and *Pseudomonas* spp. (Masniyom et al., 2011). Our result was in agreement with Masniyom et al. (2013), who stated that TVC remained low when tilapia kept in MAP with 60% CO_2. It was also reported that 50% CO_2 inhibited the bacterial growth in chub mackerel (*Scomber colias*) (Stamatis & Arkoudelos, 2007), 40% CO_2/30% N_2/30% O_2 in swordfish (*Xiphias gladius*) (Pantazi et al., 2008) and 50% CO_2/50% N_2 in *Pangasius hypophthalmus* fillets (Noseda et al., 2012) during refrigerated storage.

MAP-1 treatment (50% CO_2 /50% N_2) in the current study showed the highest shelf life (15 days) for tilapia fish, which is similar to that of Masniyom et al. (2013) under MAP (60% CO_2/10% O_2/30% N_2) at 4°C. Raising the amount of CO_2 in the package from 50% to 75% along with N_2, the shelf life of ready-to-cook pangas fish curry at 4°C increased from 12 to 15 days (Nayma et al., 2020). Reddy, Schreiber, Buzard, Skinner, and Armstrong (1994) recorded a longer shelf life of tilapia fillets (>25 days) in the MAP storage than the current studies of MAP (50% CO_2/50% N_2) stored at 4°C. The explanation for this might be the use of a significant amount of carbon dioxide and nitrogen together. Cyprian et al. (2013) recorded a shelf life of packaged tilapia fillets for 23 days under MAP (50% CO_2/50% N_2) stored at 1°C, which is also higher than current research. This result might be due to the difference in storage temperature where the 1°C is colder than 4°C of the current study. The highest shelf life of 15 days was observed in the case of MAP-1 (50% CO_2 /50% N_2) due to the slightly slower growth of bacteria during the storage period than other packaging systems. MAP-1 created anaerobic conditions within the pack, which reduced the growth of aerobic bacteria and thus increased the shelf life. Also, CO_2 has a bacteriostatic effect and prevents oxidative rancidity as well (Farber, 1991).

3.5. Total Coliforms and Fecal Coliforms

Total coliform bacteria are usually observed in the aquatic environment, soil, and vegetation, and those are commonly harmless. Fecal coliform bacteria are a type of total coliform bacteria found in the intestines and feces of humans and animals. They are regarded as sanitary index organisms for foods and water. Their presence in the food indicates the probable occurrence of pathogenic bacteria of fecal origin (Li & Liu, 2019). Total coliforms and fecal coliforms have acceptable limits of <100 MPN / g and <10 MPN / g for fish and fishery products, respectively (ICMSF, 1986).

In the current study, total coliforms and fecal coliforms showed irregular growth behavior and were unsuccessful in remarking a progressive increase in total coliform and fecal coliform counts. There is an agreement with the study of Abelti (2013), who also found irregular growth behavior of total and fecal coliforms while studying ice stored lobsters (*Pacifastacus* spp). However, the total coliform counts of sliced tilapia fish ranged from <3 to 93 MPN/g, and fecal coliform counts ranged from <3 to 9.2 MPN/g under all packaging systems in the 18 days of storage period (data not shown). Total coliform counts were not affected by the treatments in the storage period, which agrees with that of Soccol et al. (2005). However, the total coliforms and fecal coliform counts did not exceed the acceptable limit under all packaging systems, which directed that the samples were taken from a non-polluted area and maintained aseptic conditions throughout the preparation of packaged products by handlers. The low growth of coliforms may have observed because coliforms are mesophilic and, therefore, are hindered by refrigerated storage at 4°C (Soccol et al., 2005). It was noted that CO_2 more than 50% delayed coliform growth in tilapia fillets stored under MAP (Reddy, Schreider, Buzard, Skinner, & Armstrong, 1994).

4. Conclusion

The above discussion led to the conclusion that all treatments obtained adequate shelf life during the storage period, except for the control sample showing total bacterial counts crossed the acceptable levels on days 6-9 of the storage. Vacuum packaging extended the shelf life by 9-12 days. On the other hand, MAP-1 with 50% CO_2/50% N_2 extended the shelf life by 15 days and MAP-2 with 50% CO_2 /50% O_2 increased the shelf life by 12 days. Therefore, the MAP-1 demonstrated the highest shelf life of sliced tilapia fishes based on bacterial counts. The processors or superstores may use this packaging technology to display the fish and fishery products under refrigeration conditions, which will increase the product's shelf life, value, convenience, and grade.

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