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Effect of Modified Atmosphere Packaging on Microbial, Biochemical, Sensory properties and Shelf-life of Reef Cod (*Epinephelus diacanthus*) fillet during chilled Storage

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ABSTRACT: The objective of this study is to assess stability of chilling in combination with MAP to extend the selflife of chilled reef cod (*E. diacanthus*) fillet with minimum loss in quality. The assessment of fillet quality encompassed evaluation based on microbial, biochemical, and sensory attributes. The fresh reef cod fillets were packed under two conditions: T0 (Control: Air) and T1 (MAP: 40% CO_2 + 30% O_2 + 30% N_2) and stored at 4°C. Values of TVB-N, TMA-N, FFA, PV, and TBA, indicative of deterioration, increased throughout the chilled storage period. Biochemical values were relatively higher in reef cod fillets stored in air packs than in those stored using MAP. The Total Plate Count (TPC) exhibited higher final numbers in control (T0) sample compared to those in T1. *E. coli* and *S. aureus* were not detected in any of the samples. Sensory analysis (Texture, Color, Appearance, Odor, Overall quality) correlated well with the biochemical and microbiological analyses, indicating a shelf life of approximately 6 days for the control and 18 days for T1. In comparison between the treatments, MAP proved more effective in inhibiting biochemical, microbial, and sensory spoilage processes, extending the self life up to 18 days compared to T0. Overall, modified atmosphere packaging (MAP) showed a synergistic effect in extending shelf life as well as maintaining the quality of reef cod fillets under chilled storage.

Keywords: Reef cod Fillet, modified atmosphere packaging, Chilled storage, Physicochemical property, self-life.

INTRODUCTION

Fish is highly perishable, leading to a short shelf life for fresh fish. Fish is usually stored under refrigeration, presenting a very sort self life, or under freezing condition, exhibiting a longer self life but losing important quality properties (Eezek and Buchtová 2007). Consequently, the fishery industry has continuously sought new ideas and innovations to extend this shelf life. Both fish and shellfish have a limited shelf life during chilled storage. In recent decades, there has been a growing demand for chilled fish and fish products that can maintain quality over extended periods, allowing for storage and distribution in distant markets. Nowadays consumer demand for fresh, conveniently packed, boneless fish and fillet products on shelf service shelves that are also easy to prepare (Masniyom, 2011). Typical shelf-life under current icing and refrigerated storage conditions ranges from 2 to 14 days (Stammen *et al.,* 1990). The acceptability limit for fatty fish species has been reported to range from 4 to 9 days (Smith *et al.,* 1980; Simeonidou *et al.,* 1998; Erkan, 2003; Erkan and Ozden 2008). Therefore, fish processor must develop new methods that suit the current trend and lead to adequate self life extension of fish product while taking into consideration their sensory, and microbiological quality as well as safety (Masniyom, 2011). This study will focus on a technique involving changing the natural gas composition surrounding the fish fillet to delay quality changes and extend the freshness period of reef cod.

The main purpose of Modified Atmospheric Packaging (MAP) is to ensure the physicochemical and microbial quality and increase the shelf-life during chilled storage of fish fillets. MAP, also known as 'the enclosure of food products in gas-barrier materials, in which the gaseous environment has been altered' (Young *et al.,* 1988), aims to inhibit spoilage agents and either maintain higher quality within perishable food during its natural life or extend its shelf-life. The principle of MAP involves replacing air in the package with a different fixed gas mixture. Packaging fish fillets in a modified atmosphere allows the product to meet consumers' preferences for convenience while simultaneously delaying fish deterioration. MAP is a practical and economically viable packaging technique that enables the food industry and retailers to respond to market trends (Caleb *et al.,* 2012).

MATERIAL AND METHODS

Sample Preparation. Fresh reef cod obtained from Veraval fishing port was transported to the laboratory in iced condition. The samples were gilled, gutted, filleted, washed, and then stored on ice. The prepared reef cod fillets were divided into two treatment groups. In T0(C), reef cod fillets were packed in air using polyethylene

laminated trays (4×16×22 cm) and In T1 reef cod fillets was packed under modified atmosphere (40% CO_2, 30% N_2, 30% O_2) in polyethylene laminated tray of the same dimension and stored at chilled temperatures. The samples were taken at 0, 3, 6, 9, 12, 15, 18, 21, 24, 27, and 30 days after storage and analyzed for changes in microbial, biochemical, and sensory quality.

A. Biochemical analysis

Determination of TMA-N. Preparation of Trichloro Acetic acid extract: The TMA-N and TVB-N content of the fish were determined using the Micro Conway diffusion method (AOAC, 2006). 5 gm. of accurately weighed samples were extracted with 20% trichloroacetic acid by grinding in a mortar and pestle. The content was then filtered through Whatman filter paper No. 1, and the filtrate was made up to 50 ml. The TCA extract was used to measure TMA-N and TVB-N.

Determination of TMA-N (mg %): Conway units were cleaned in chromic acid for 24 hours, soaked in water washed and dried. Cover plates were coated on under side with wax- grease. The units were kept ready before preparing the extract. Pipette carefully 1ml of standard N/100 sulphuric acid into inner chamber of the diffusion unit. To the outer chamber 1 ml TCA extract was added followed by 0.5 ml neutralized formaldehyde. The unit was then sealed with glass lid and incubate at 37 ºC for one hour. The inner chamber solution was titrated against standard N/100 sodium hydroxide with Tashiro's indicator. Similarly, a blank was also run. TMA-N was calculated and expressed as mg % of the samples using the following equation.

$$TMA{-}N = \frac{(Vs{-}Vb) \times 0.14 \times \text{Volume of extract}(mg/100g)}{\text{Vol. of sample taken} \times \text{weight of sample} \times 100}$$

Where,
Vs = Titre value of 0.01 N NaOH for the sample (ml)
Vb = Titre value of 0.01 N NaOH for the blank (ml)

Determination of TVB-N: Conway units were cleaned in chromic acid for 24 hours, soaked in water washed and dried. Cover plates were coated on under side with wax- grease. The units were kept ready before preparing the extract. 1ml of standard N/100 sulphuric acid was taken in the inner chamber of the diffusion unit. To the outer chamber 1 ml TCA extract was added followed by 1 ml of saturated potassium carbonate. The unit was then sealed with glass lid and incubate at 37 ºC for one hour. The amount of unreacted acid in the inner chamber was determined by titrating against standard N/100 sodium hydroxide with Tashiro's indicator. Similarly, a blank was also run. TVB-N was calculated and expressed as mg % of the samples using the following equation.

$$TVB{-}N = \frac{(Vs{-}Vb) \times 0.14 \times \text{Volume of extrac } (mg/100g)\text{t}}{\text{Vol. of sample taken} \times \text{weight of sample} \times 100}$$

Where,
Vs = Titre value of 0.01 N NaOH for the sample (ml)
Vb = Titre value of 0.01 N NaOH for the blank (ml) 23

Determination of FFA: The free fatty acid (FFA) content in the lipid extract was determined with improved titremetric method as described by Takagi *et al*. (1984). 100 g of fish muscle sample was taken and homogenized with 300 ml of chlorofoam-methanol mixture (2:1) for 2 min in a blender. Further 100 ml distilled water was added and bland for another 30 sec. then the homogenate was filtered through Buchner funnel under light suction. Residue was again blended with 100 ml chlorofoam and transferred to 500 ml separating funnel for equilibration. Bottom chlorofoam layer was collected and dry over anhydrous sodium sulphate. Extract volume was made up to 250 ml. 5 ml of methanol and 10 ml of isopropanol (Chloroform: Methanol: Isopropanol = 2:1:2) were added to 10 ml Chloroform extract and mixed in a 100 ml conical flask. Three drops of 0.5% metacresol purple was added as an indicator. The FFA was titrated to purple end point with 0.05 N aqueous sodium hydroxide. The percentage of free fatty acid was calculated as oleic acid percentage by following equation.

$$\% \text{ FFA} = \frac{(\text{ml of } 0.05 \text{ N NaoH}) \times (14.1225) \times \text{Volume of extract}}{\text{Weight of sample in g.} \times \text{aliquot volume}}$$

Determination of PV: The peroxide value (PV) of lipid was determined from the lipid extract according to Jacobs (1958) iodomatrically. 10 g sample was taken and ground well with 15 g anhydrous sodium sulphate. Homogenate was transferred to a 100 ml stopper flask and 30-50 ml chloroform was added and placed in dark place for about 15-20 minutes with occasionally shaking. 10 ml of chloroform extract, and 25 ml of solvent (2 volume of glacial acetic acid and one volume of chloroform) were added. The liberated iodine was titrated against 0.02N standard sodium thiosulphate solution using starch as indicator and expressed as milliequivalnt of peroxide/kg of lipid by following equation.

$$\text{PV } (meq/Kg) = \frac{1000 \times (V{-}X) \times N \times \text{Volume of extract}}{W \times \text{aliquot sample vol. } 24}$$

Where,
W = Weight of sample
V = Vol. of sodium thiosulphate used for sample
X = Vol. of sodium thiosulphate used for blank
N = Normality of sodium thiosulphate solution

Determination of TBA: The Thiobarbituric acid (TBA) was determined according to AOAC (1990). 10 gram of fish sample was taken in to a distillation flask with 95 ml distilled water and 5 ml 4 N HCL. Distillation was done on hot plate at high temperature so that as much as 50 ml obtained. 5 ml of distillate obtained was transferred in screw cap test tube to where 5 ml TBA reagent was added. Tube was heated in water bath for 35 min till the pink colour developed. After the solution was cooled, Absorbance was recorded by spectrophotometer at a wavelength of 538. TBA was calculated and expressed mg malonaldehyd/kg by following equation.

$$TBA\ (MDAmg/kg) = Effectiveabsorbance \times 7.8$$

B. Microbiological Analysis

Preparation of Sample: 10 gm of fillet sample were taken in a sterile dish and transferred to a sterile mortar. The sample was homogenized with 90 ml of sterile phosphate buffer solution. Then, the homogenized sample was diluted tenfold (i.e., 10^{-1}). For further dilution, 1 ml from the 10^{-1} dilution was mixed with 9 ml of the appropriate medium for plating.

TPC (Total Plate Count): The microbiological characteristics of the fish fillet were assessed following

the standard method recommended by AOAC (2006). 1 ml. from each required dilution was transferred into separate serial petri dishes. Sterile Tryptone Glucose Beef Extract (TGBE) agar (0-15 ml) was spread onto the petri dishes, thoroughly mixed, and allowed to solidify. The petri dishes were then placed in the incubator and incubated at 37±1°C for 48 hours. After this period, the colonies were counted.

$$TPC\ (cfu/g) = \frac{No.\ of colonies \times Dilution}{\text{Weight of sample}}$$

E. coli: The determination of *E. coli* in fish fillet was conducted following the standard method described by AOAC (2006). 10 gm. of fish sample were taken aseptically and homogenized with 90 ml of physiological saline, resulting in a 10^{-1} dilution. 1 ml. of inoculum from the dilution was spread on sterile Tergitol-7 (T-7) agar plates (0-15 ml). The plates were then incubated at 37±1°C for 18-24 hours. E. coli colonies on T-7 agar plates manifest as circular, non-mucoid, flat, yellow-colored colonies with a pinkish tinge. The addition of Triphenyl tetrazolium chloride (TTC) allows for the confirmation of E. coli. Positive colonies were counted, and the results were expressed as the number of colony-forming units per gram.

***Staphylococcus aureus*:** The determination of *S. aureus* in the fish fillet sample was conducted following the standard method recommended by AOAC (2006). 1 ml. of inoculum from the dilution was spread on triplicate plates of Baird Parker agar (BPA) medium (0-15 ml each), distributing 1 ml of inoculum equally over each plate. The inoculum was spread on the agar surface using sterile bent glass streaking rods (hockey sticks). The plates were then incubated upright in a 35-37°C incubator for approximately 1 hour, followed by inversion and further incubation for 45-48 hours. The colonies were counted, calculated, and the results were expressed as the number of S. aureus per gram of the sample.

Sensory Characteristics. The sensory characteristics of the fish fillet were evaluated using a 9-point Hedonic scale (Joseph and Iyer 2006). The analysis was conducted on randomly selected samples retrieved from chilled storage. Each sample was assessed for appearance, color, odor, texture, and overall acceptability by five panelists using the 9-point Hedonic scale.

Statistical Analysis. Data were analyzed statistically as per factorial complete randomized design. Analysis of variance was worked out using standard statistical procedures as described by Snedecor and Cochran (1967). The analysis of variance (ANOVA) was carried out based on the experimental data using IBM BASIC windows release1.13 to know the significant difference for different treatment combination and to find the best treatment combination.

RESULTS AND DISCUSSION

Effect of MAP on Biochemical changes of Reef cod fillet: The changes in TMA-N, TVB-N, FFA PV, TBA content in air packed (T0) and MAP packed (T1) sample showed progressive increase trends with increasing chilled storage period (Table 1) TMA-N values of reef cod fillets are presented in (Table 1). TMA-N content is often used as a biochemical index to assess keeping quality and shelf-life of fish (Connell, 1990). In marine fish, TMA which is formed from trimethylamine oxide (TMAO) because of bacterial enzyme activity, is the main component responsible for an unpleasant ‘‘fishy’’ odour (Connell, 1990; Debevere & Boskou 1996; Sivertsvik *et al.,* 2002: Shakila *et al.,* 2003). The TMA-N value increase to 13.75±0.44 in T0, and 11.45±0.38 in T1, on the last day of storage period. After day 15 of storage. The level of TMA-N found in fish rejected by Huss (1995) is typically around 10–15 mg TMA-N/100 g. TMA-N values of the control samples were significantly higher and increased with a significantly higher rate than the corresponding values T1 this clearly indicates the effectiveness of MAP in T1.

TVB-N is widely used as an indicator of fish quality, with its increase being linked to the activity of spoilage bacteria and endogenous enzymes (Ozogul *et al.,* 2004; Ruiz *et al.*, 2005). The action of these enzymes leads to the formation of compounds such as ammonia, monoethylamine, dimethylamine, and trimethylamine (Debevere & Boskou 1996), imparting characteristic off-flavors to fish. The TVB-N value of reef cod fillets progressively increased during storage and showed statistical significance ($p < 0.05$). Air-packed fillets (T0) recorded the highest value of 46.25±1.26 mg/100g, while MAP-packed fillets (T1) had a value of 43.50±0.58 mg/100g. However, like TMA-N, the TVB-N level exceeded acceptable limits in air-packed fillets after 6 days of storage and in MAP-packed reef cod fillets after 18 days of storage. This phenomenon might be attributed to the presence of lactic acid dissolved in the fillet, causing a notable pH change, and contributing to the rise in TVB-N value (Genç *et al.,* 2013). Notably, the slowest increase was observed in the MAP (T1) group. This is primarily because the higher CO_2 content in MAP could suppress bacterial activity and delay chemical reactions (Sivertsvik *et al.,* 2003). The TVB-N, like the TMA-N value, increased with storage time at 4°C (Fig. 1), aligning with findings reported in various studies by different researchers (Tejada and Huidobro 2002; Grigorakis *et al.,* 2003; Ozogul *et al.,* 2005). Lipid deterioration is a limiting factor in the shelf life of fish. The formation of free fatty acids (FFA) during seafood storage is a key factor contributing to the deterioration of protein quality. Glycerides, glycolipids, and phospholipids are hydrolyzed by lipases to form FFA, which then undergo further oxidation to produce low molecular weight compounds, such as aldehydes and ketones. These compounds are responsible for the off-flavor, off-odor, and taste of fish (Ozogul *et al.,* 2005). The FFA content showed a slow increasing trend in both treatment samples during chilled storage periods, exhibiting statistically significant differences ($p < 0.05$) (Table 1 and Fig. 1). Ozogul *et al.,* 2011 reported a similar increasing trend in FFA in common sole (*Solea solea*) during ice storage. At the end of the storage period, the FFA values were found to be 0.94±0.03 in T0 and 0.78±0.08 in T1. Peroxide value (PV) is used to indicate the oxidative state of lipid-containing foods,

measuring the initial stage of oxidative rancidity (Balachandran, 2001). Pearson (1970) suggested that the acceptable level of PV ranges from 20 to 40 m.equ/kg of fat. As oxidation progresses, peroxides break down into aldehydes or combine with proteins (Woyewoda and Bligh 1986). In this study, the changes in PV value in all chilled samples showed increasing trends, like FFA and TBA values, with statistically significant results ($p < 0.05$) during the storage period (Table 1), albeit remaining within the acceptable limits (Fig. 1). At the end of the chilled storage period, the peroxide value (PV) was 7.08±0.17 m.equ/kg in T0 and 4.68±0.35 m.equ/kg in T1. Similar PV results were obtained by Mohan *et al.* (2008) for sea bream.

Table 1: Changes in TMA-N, TVB-N, FFA, PV, TBA in reef cod fillets (*E. diacanthus*) during chilled storage.

Storage period (days)	Packaging condition of reef cod fillets during chilled storage									
	TMA-N		TVB-N		FFA		PV		TBA	
	Air packed (Control) (T0)	Air packed (Control) (T1)	Air packed (Control) (T0)	Air packed (Control) (T1)	Air packed (Control) (T0)	Air packed (Control) (T1)	Air packed (Control) (T0)	Air packed (Control) (T1)	Air packed (Control) (T0)	Air packed (Control) (T1)
0	2.43±0.06	2.48±0.05	12.48±0.95	12.00±0.82	0.22±0.02	0.22±0.01	1.03±0.05	1.00±0.00	0.82±0.02	0.82±0.02
3	5.33±0.19	3.40±0.28	25.80±0.89	22.15±0.70	0.27±0.03	0.24±0.02	2.13±0.25	1.43±0.13	1.72±0.48	1.27±054
6	8.15±0.13	6.18±0.10	35.53±2.00	28.38±1.82	0.29±0.01	0.21±0.01	2.25±0.17	1.65±0.12	1.69±0.48	1.37±0.11
9	8.58±0.45	6.85±0.10	37.50±1.00	31.50±1.00	0.57±0.06	0.29±0.03	4.18±0.05	1.98±0.10	2.47±0.53	1.65±0.54
12	9.28±0.10	7.30±0.35	41.00±1.15	34.25±1.50	0.65±0.03	0.25±0.05	4.67±0.43	1.98±0.10	3.34±0.50	2.79±0.93
15	10.40±0.82	8.40±0.23	41.00±1.41	35.50±0.58	0.66±0.03	0.40±0.01	5.35±0.45	2.03±0.06	4.51±0.24	3.49±0.29
18	10.82±0.83	8.98±0.25	41.25±0.96	40.50±.0.58	0.70±0.01	0.50±0.07	5.80±0.51	2.34±0.51	4.78±0.10	3.97±0.49
21	11.38±0.97	9.75±0.10	42.25±0.50	39.50±1.00	0.64±0.16	0.50±0.14	6.20±0.09	3.13±0.50	5.67±0.36	4.46±0.37
24	12.36±0.95	9.73±.0.43	43.50±0.58	40.25±0.96	0.68±0.17	0.59±0.09	6.85±0.25	3.45±0.27	6.30±0.29	5.40±0.64
27	12.83±0.55	11.03±0.62	44.75±0.50	42.25±0.50	0.71±0.03	0.63±0.03	6.96±0.56	4.30±0.18	6.92±0.82	5.48±0.60
30	13.75±0.44	11.45±0.38	46.25±1.26	43.50±0.58	0.94±0.03	0.78±0.08	7.08±0.17	4.68±0.35	6.98±0.15	5.76±0.17

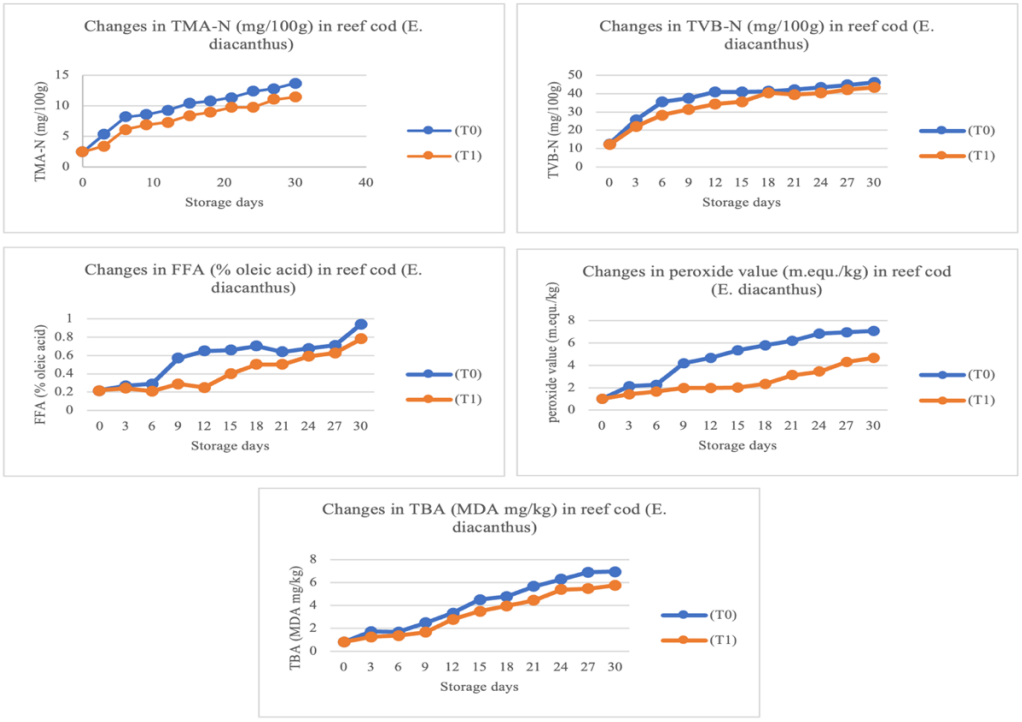

Fig. 1. Changes in TMA-N, TVB-N, FFA, PV, TBA in reef cod fillets (*E. diacanthus*) during chilled storage.

The TBA value serves as an index of advanced lipid oxidation, measuring malondialdehyde (MDA) content. MDA is formed through hydroperoxides, which represent the initial reaction product of polyunsaturated fatty acids with oxygen (Fernandez *et al.*, 1997). The changes in TBA content in reef cod fillets packed under air (T0) and MAP (T1) samples showed progressive increasing trends with the increasing chilled storage periods (Table 1). A statistically significant difference ($p < 0.05$) was observed in TBA values. A positive correlation was noted between FFA and TBA (Fig. 1), indicating a trend of progressive increase with an increasing period of storage. It is evident that the liberated FFA entered an advanced stage of oxidation, albeit slowly, resulting in an increased TBA value. The TBA value in T1 exhibited a low growth rate in the late storage period (20-30 days), suggesting that CO_2 and N_2 could effectively inhibit lipid oxidation in fillets, limiting microbial growth and the release of lipolytic enzymes (Kimbuathong *et al.*, 2020).

Effect of MAP on Microbial changes of Reef cod fillet: The initial values of TPC in each sample were more or less the same, demonstrating an increasing trend but remaining within the acceptable limit as the storage period increased (Table 2 and Fig. 2). The TPC of reef cod fillets stored in air (T0) reached 6.48 log CFU/g on day 30 of storage, while MAP-packaged samples (T1) reached 6.33 log CFU/g on day 30. None of the samples exceeded the value of 7 log CFU/g, which is considered the upper acceptable

microbiological limit for marine species (ICMSF, 1986) during the 30-day storage period. Hence, it is evident that MAP samples have a longer shelf life concerning the bacterial spoilage limit. The limitation of bacterial growth in MAP may be attributed to the inhibitory effect of CO_2. It has been reported that aerobic spoilage microorganisms such as *pseudomonads* and *Shewanella putrefaciens* are generally sensitive to CO_2; therefore, MAP (T1) delayed the spoilage of fish (Stammen *et al.*, 1990; Gill and Mollin 1991). CO_2 extended the lag phase, reduced the growth rate of microorganisms in their logarithmic phase, and inhibited the activity of succinate dehydrogenase and malate dehydrogenase enzymes in the Krebs Cycle of microorganisms (Singh *et al.*, 2014; Zhang *et al.*, 2017). Throughout the study, *E. coli* and *Staphylococcus aureus* were not detected. The absence of *E. coli* and *S. aureus* indicated a microbiologically safe and stable product at the end of the storage period.

Table 2: Changes in TPC log(cfu/g) in reef cod fillets (*E. diacanthus*) during chilled storage.

Storage period (days)	Packaging condition of reef cod fillets during chilled storage	
	TPC log (cfu/g)	
	Air packed (Control) (T0)	Air packed (Control) (T1)
0	3.08±0.15	3.08±0.15
3	3.39±0.10	3.39±0.10
6	4.93±0.03	4.79±0.07
9	4.96±0.02	4.84±0.01
12	4.98±0.03	4.90±0.04
15	6.08±0.05	5.92±0.03
18	6.31±0.04	6.10±0.07
21	6.30±0.03	6.17±0.06
24	6.35±0.07	6.29±0.03
27	6.46±0.01	6.32±0.01
30	13.75±0.44	11.45±0.38

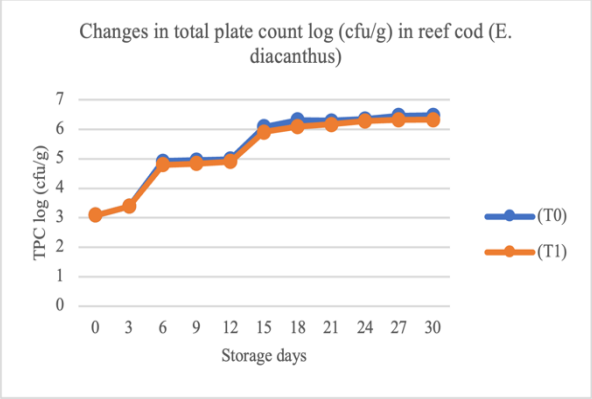

Fig. 2. Changes in TPC log(cfu/g)in reef cod fillets (*E. diacanthus*) during chilled storage.

Effect of MAP on Sensory Characteristics of Reef cod fillet: Sensory evaluation is considered the most reliable test for both raw materials and processed fishery products (Ryder *et al.*, 1993). The sensory analysis of both treatments was assessed over the 30-day storage period, as depicted in Table 3 and Fig. 3 for the experiment.

Table 3: Average panel scores for overall acceptability of reef cod (*E. diacanthus*) fillets during chilled storage.

Storage period (days)	Packaging condition of reef cod fillets during chilled storage	
	Average panel score	
	Air packed (Control)(T0)	Air packed (Control)(T1)
0	8.50±0.58	8.75±0.50
3	7.25±0.50	8.75±0.50
6	6.00±0.00	7.00±0.00
9	5.50±0.58	7.00±0.00
12	5.00±0.00	6.50±0.58
15	4.00±0.00	5.00±0.00
18	3.25±0.50	5.00±0.00
21	3.00±0.00	5.00±0.00
24	2.00±0.00	4.25±0.50
27	1.75±0.96	3.50±0.58
30	0.75±0.50	2.75±0.50

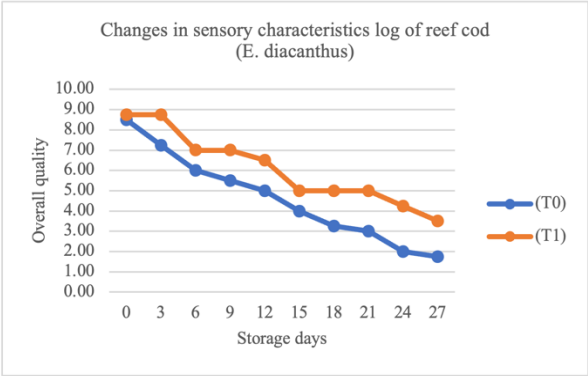

Fig. 3. Changes in overall acceptability during chilled storage period.

Up to 12 days of storage MAP packed reef cod fillet samples received an overall acceptability score of 6 significantly ($p<0.05$) higher than the lower acceptability limit for control. The results of the present study agree with those reported by Mejlhom and Dalgaard (2002) for cod fillets. The fillets colour, odour, texture, appearances, and overall acceptability scores of all packaged fillet samples decreased gradually (Fig. 3) with the time of refrigerated storage with a relatively similar decrease rate as the texture and odour scores. Finally, the total shelf-life extension, which was achieved in the present study between control and MAP reef cod fillets, was 18 days according to organoleptic score.

CONCLUSIONS

In this present study biochemical, microbial and sensory property and self life of reef cod fillet in air and MAP with (40% CO_2 + 30% O_2 + 30% N_2) ware evaluated during chilled storage. The result showed that MAP could effective slow down the increase of TMA-N, TVB-N, FFA PV, TBA value. Based on microbial and sensory analysis the use of MAP (T1) extends the self-life of reef cod filler up to 18 days against 6 days of air packed (T0). Thus, it is evident that MAP of reef cod fillet is preferred over air packed in terms of longer self-life. MAP will be advantages to retail consumer for

storage the reef cod fillets for further processing and cooking.

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