

The Effect of Powdered Cumin Sprinkling on Biochemical and Quality Attributes of Red Tilapia Fillets Stored in Ice

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Abstract: The effect of powdered cumin sprinkling on the biochemical and sensory properties of fresh and ice-stored tilapia fillets during 21 days was examined. The biochemical parameters were determined by measuring total Volatile Bases Nitrogen (TVB-N), Trimethylamine Nitrogen (TMA-N) and pH throughout storage in ice for 21 days. The sensory quality of cooked tilapia fillets was assessed by ten semi-trained panels at zero time and after 3, 6 and 9 days of ice storage. Initially, significant differences in pH, TMA-N, odour, flavour, appearance and overall acceptability were found between cumin-treated and untreated tilapia fillets at ($p < 0.05$). However, no significant differences were detected in TVB-N between cumin-treated and untreated tilapia fillets. Cumin powder addition (2% w/w) exhibited a preservative effect in treated fillets since significant lower levels of TMA-N were observed during ice storage. A continuous increase in TVB-N and TMA-N was observed throughout storage in ice for 21 days ($p < 0.05$). Both of TVB-N and TMA-N values were lower than acceptability limits during the storage period. The storage time had no effect ($p < 0.05$) on the sensory attributes of all cooked tilapia fillets. The present findings showed that cumin-treated tilapia fillets maintained their quality better than untreated fillets during the ice storage, since they achieved higher scores than untreated fillets.

Key words: Sensory assessment, treated fillets, TVB-N, TMA-N, pH

INTRODUCTION

Spices have in recent years gained importance as bio nutrients, both as functional food ingredients and nutritional supplements; they are used primarily for enhancing food flavour rather than shelf life extension (Milan *et al.*, 2008). Cumin (*Cuminum cyminum* L.) is a small annual plant belonging to the Apiaceae family. It is one of the popular spices regularly used as a flavouring agent (Thippeswamy and Naidu, 2005). Cumin's distinctive flavour and strong, warm aroma is due to its essential oil content which may be considered as an interesting source of antibacterial, antifungal and antioxidants components used as potent vagents in food preservation and for therapeutic or nutraceutical industries. Its main constituent and important aroma compound is cuminaldehyde (4-isopropylbenzaldehyde) (Hajlaoui *et al.*, 2010). Additionally and because of its greater awareness and safety concern regarding synthetic chemical additives, food preserved with natural additives has become more popular. For instance, the antimicrobial and antioxidant properties of essential oils and their active constituents derived from various plant organs have been

empirically recognised (Burt, 2004; Lee and Shibamoto, 2002). However, any processing technologies used in the production of such compounds have to prove their technical/scientific efficiencies and the product quality to meet the basic requirement of hygiene and safety standards. Therefore, it would be economically more suitable to use powdered spices or herbs as ingredients rather their extracts to preserve food including fish fillets (Smid and Gorris, 1999). In the seafood sector such preservative compounds including spices/herbs were equally used to extend the shelf life of fish fillets but at low levels to avoid strong flavours imparting unpleasant sensorial characteristics (Mejlholm and Dalgaard, 2000).

Fish is one of the most highly perishable food products, during handling and storage, quality deterioration of fresh fish rapidly occurs and limits the shelf life of the product. The quality of fish degrades due to a complex process in which physical, chemical and microbiological forms of deterioration are implicated (Sallam, 2007). The spoilage rate of tropical and subtropical species when stored in ice might differ considerably from those from cold waters. For fish caught

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in tropical or subtropical waters, the storage times in ice are relatively longer, e.g., from 2 to >3 weeks. This difference in storage time is due to the nature of the microflora initially present in fish at the moment of capture (El-Marrakch *et al.*, 1990). It is known that loss of freshness and spoilage pattern in fish markedly varies from species to species. Once the fish dies, several postmortem changes take place which are due to the breakdown of the cellular structure and biochemistry as well as to the growth of microorganisms that are either naturally associated with the fish or associated to contamination during handling (Ehira and Uchiyama, 1987). These postmortem changes that directly and strongly affect its quality and shelf life are associated with protein and ATP degradation, drop of pH, lipid oxidation, undesirable compounds production as Trimethylamine (TMA-N) and the molecular low weight volatile bases (TVB-N) which are produced by bacterial action. The tilapia (*Oreochromis*, *Surotherodon* and *Tilapia*) are species of major economic importance in tropical and sub-tropical countries throughout the world (Hussain *et al.*, 1995). Freshwater tilapia (*Oreochromis niloticus*) was chosen for this study for its good market acceptance and rusticity for handling. Tilapia is considered promising for aquaculture because of its rapid growth, late reproduction and high multiplication rate. It has a firm, consistent and tasty meat of great market acceptance.

Till date, there is no study on the effect of cumin extract or powdered cumin (*Cuminum cyminum* L.) on biochemical and sensory properties of tilapia fillets (*Oreochromis niloticus*) stored in ice. Additionally, no studies have so far been successfully conducted to produce such ice stored spiced tilapia fillet with cumin flavour. Although, there is potential for its palatability and acceptability in the local market in Malaysia, studies on its effect on the biochemical and sensory changes of fish fillets are not available. The main objective of this study was therefore, to produce ice stored spiced tilapia fillet with a new flavor (cumin) and to demonstrate its effect on the biochemical and sensory attributes of cumin-treated fish fillets.

MATERIALS AND METHODS

Preparation and treatment of fish samples: Live red tilapia (*Oreochromis niloticus*) was killed, scaled, headed, gutted and filleted. The fish fillets were divided into two batches. The first batch was treated and sprinkled with fine cumin at concentration (2% w/w) while the second batch was left without treatment as a control. Fillets were placed into polyethylene bags individually and stored in

sealed polystyrene boxes with flaked ice (2:1 w/w) in a cold room at 4°C. The filleted tilapia was sampled and subjected to chemical and sensory analyses after 0, 3, 6, 9, 12, 15, 18 and 21 days of storage.

Proximate analysis: The fish samples were analysed for proximate composition: Moisture content was determined by air-drying of a portion of minced fish fillet at 103±2°C for 24 h; crude protein by the Kjeldahl Method using potassium sulphate and copper (II) sulphate as the catalysts and 6.25 nitrogen to protein conversion factor; crude fat by petroleum ether extraction using the Soxhlet Method and ash by incineration in a muffle furnace at 550±°C for 24 h according to the method of Kirk and Sawyer (1991).

Determination of pH: The 10 g of each sample were homogenized in 100 mL of distilled water and the mixture was filtered. The pH of the filtrate was measured using a pH-meter at ambient temperature (Goulas and Kontominas, 2007).

Determination of total volatile bases nitrogen: TVB-N was determined by steam distillation of the Trichloroacetic Acid (TCA) extract using the modified method of Malle and Poumeyrol (1989) as described by Riquixo (1998); 200 mL of 7.5% aqueous TCA solution was added to 100 g of fish muscle and after homogenization, the mixture was filtered using Whatman filter paper. Following which 25 mL of filtrate was loaded into the distillation tube followed by 6 mL of 10% NaOH. Steam entrainment was performed using Struer automatic distillation. A beaker containing 15 mL of 4% boric acid and 0.04 mL of methyl red and bromocresol green indicator was placed under the condenser for the titration of TVBN. Distillation was started and steam entrainment continued until a final volume of 100 mL was obtained in the beaker. The results of TVB were obtained using the following Eq. 1:

$$\frac{\text{TVB mg-N}}{100\text{g}} = \frac{(\text{mL})(0.055 \text{ mol L}^{-1})}{8.333} (14 \text{ Ng/mol})100 \quad (1)$$

Determination of trimethylamine nitrogen: To assay TMA-N using TCA-extract by steam distillation (Malle and Poumeyrol, 1989), it was processed as for TVB-N (2.4.1) but with the addition of 20 mL 35% of formaldehyde to the distillation tube to block the primary and secondary amines whilst leaving only the tertiary amines to react. The results of TMA-N were obtained as follows in the Eq. 1. The results were expressed in mg nitrogen 100 g⁻¹ of sample.

Sensory evaluation: The sensory quality of cooked cumin-treated and untreated tilapia fillets was assessed by ten semi-trained panels at zero time and after 3, 6 and 9 days of ice storing according to Sallam (2007). The fillets were grilled on both sides (Grilling machine). Immediately after grilling, samples were provided to the sensory panel using a code with random numbers of three digits. Drinking water was provided to cleanse the mouth cavity between testing each sample. Panellists were asked to assess the odour and flavour intensity, juiciness, hardness and tenderness by using an eight-point hedonic scoring scale with 8 = extremely intense/juicy/tender, 7 = very intense/juicy/tender, 6 = moderately intense/juicy/tender, 5 = slightly intense/juicy/tender, 4 = slightly bland/dry/tough, 3 = moderately bland/dry/tough, 2 = very bland/dry/tough, 1 = extremely bland/dry/tough was employed for odour and flavour intensity, juiciness, hardness, chewiness and tenderness, respectively. While a nine-point hedonic scale from extremely acceptable (9) to extremely unacceptable (1) was used for evaluation of the appearance. Moreover, a six-point scoring scale with 6 = no-detected off-odour/off-flavour, 5 = barely detected off-odour/off-flavour, 4 = slight off-odour/off-flavour, 3 = moderate off-odour/off-flavour, 2 = strong off-odour/off-flavour, 1 = extreme off-odour/off-flavour was utilized for the assessment of the off-odour and off-flavour. Tilapia samples receiving overall scores of >4 were considered acceptable while a score of between 3 and 4 was considered as borderline acceptability. Additionally, a space was provided for further flavour description and additional comments. These attributes were selected to reflect possible sensorial changes in the tilapia fillets which could be easily detected by the panellists.

Statistical analysis: All experiments were carried out in triplicate. The data obtained on proximate, chemical and sensory evolution were subjected to Analysis of Variance (ANOVA) by using Minitab Version 16. The difference among the mean values of the various treatments and storage period were determined by the Least Significant Difference (LSD) test and the significance was defined at $p < 0.05$. The results of chemical and sensory analyses were reported as mean values \pm standard error.

RESULTS AND DISCUSSION

Proximate analysis: The data in Table 1 illustrate the proximate analyses results at time zero for cumin-treated and untreated tilapia fillets. Generally, there was a significant difference in moisture content and ash while the protein and total lipid contents did not differ significantly ($p < 0.05$) between the treated and untreated

Table 1: Proximate composition of cumin-treated and untreated tilapia fillets

Composition	Cumin-treated fillets	Untreated fillets
Fat ^A	17.96 \pm 0.14 ^a	17.71 \pm 0.09 ^a
Protein ^B	18.74 \pm 0.32 ^a	16.81 \pm 0.86 ^a
Moisture ^C	74.35 \pm 0.17 ^a	73.45 \pm 0.23 ^b
Ash ^D	1.60 \pm 0.03 ^a	1.27 \pm 0.03 ^b

^{a,b}Mean values of 3 replicates \pm standard error. Mean values within the same row with the same letters are not significantly different ($p < 0.05$); ^A (g/100 g flesh fish) on base dry weight; ^{B-D} (g/100 g flesh fish) on base wet weight

Table 2: Changes in pH of cumin-treated and untreated tilapia fillets stored in ice

Ice (days)	Untreated fillets	Cumin-treated fillets
0	6.07 \pm 0.03 ^{aC}	6.01 \pm 0.01 ^{aC}
3	6.23 \pm 0.03 ^{aB}	6.23 \pm 0.02 ^{aB}
6	5.89 \pm 0.03 ^{bD}	6.17 \pm 0.06 ^{aB}
9	6.25 \pm 0.01 ^{aB}	6.25 \pm 0.01 ^{aB}
12	6.40 \pm 0.02 ^{aA}	6.40 \pm 0.01 ^{aA}
15	6.43 \pm 0.01 ^{aA}	6.43 \pm 0.01 ^{aA}
18	6.21 \pm 0.01 ^{aB}	6.20 \pm 0.01 ^{aB}
21	6.38 \pm 0.02 ^{bA}	5.97 \pm 0.01 ^{aC}

^{a,b}Mean values of each parameter individually within a row with the same letter are not significantly different ($p < 0.05$); ^{A-D}Mean values within a column with the same letter are not significantly different ($p < 0.05$)

tilapia fillets. A similar study was conducted on the effect of thyme on wild and farmed gilthead sea bream fillets stored in ice. This study showed that thyme treatment did not reveal any effect in lipid content on any of the sea bream fillets (Attouchi and Sadok, 2010). The average moisture content was determined as being (74.35 \pm 0.17 and 73.45 \pm 0.23%) for the cumin-treated and untreated tilapia fillets, respectively. The proximate composition data for untreated fish in Table 1 agrees well with the data presented by Yanar *et al.* (2006), Justi *et al.* (2003) and Puwastien *et al.* (1999).

pH: The effect of cumin and storage time on the pH of tilapia fillets during ice storage for 21 days are shown in Table 2. All tilapia fillets showed an initial low pH. Towards the end of storage (days 21), lower pH values in cumin-treated samples were recorded in comparison to untreated samples which may be due to cumin inhibitory effects on microbial growth which delay the formation of basic nitrogen compounds. The pH was significantly changed ($p < 0.05$) between cumin-treated and untreated fillets. The variation in pH possibly reflects the different nutritional states of fish as the lower pH values could possibly be due to the higher initial levels of muscle glycogen in well fed farmed fish (Kristoffersen *et al.*, 2006; Kyraana *et al.*, 1997). Storage time had a significant ($p < 0.05$) effect on the pH values of both fillets. During storage in ice, the pH of untreated fillets increased at different rates to reach a value of 6.38 \pm 0.02 at the end of 21 days of storage. The increase of pH values during the storage period may be attributed to the production of basic compounds such as ammonia, trimethylamine as well as other biogenic amines by fish spoilage bacteria (Ozyurt *et al.*, 2009; Kyraana *et al.*, 1997).

Total volatile bases nitrogen: TVB-N and TMA-N are the most useful indices for spoilage in fresh and lightly preserved seafood (Dalgaard, 2000). Fish decomposition is a progressive proteolysis of the muscle tissue brought about primarily by the action of microorganisms and to a lesser extent by autolytic enzymes. Because, the changes during the decomposition are known to be very complex, a single chemical index may not be a reliable indicator for a particular sample of fish (Castillo-Yanez *et al.*, 2007). TVB-N and TMA-N analysis are accurate in predicting the fish spoiling process. These analyses have been traditionally used as indicators of quality in fish products stored on ice. TVB-N is a term that includes the measurement of trimethylamine, dimethylamine, ammonia and other compounds associated with seafood spoilage and increases as spoilage progresses (Gokoglu *et al.*, 1998). The variation of TVB-N contents for untreated and cumin-treated tilapia fillets are presented in Fig. 1. The initial TVB-N values in fillets tilapia analyzed ranged from 4.97 ± 0.12 to 10.78 ± 0.15 mg N 100 g^{-1} flesh for cumin-treated tilapia fillets and from 4.85 ± 0.13 to 10.94 ± 0.15 mg N 100 g^{-1} flesh for untreated fillets during the 21 days storage period. During ice storage, had no significantly ($p < 0.05$) changes in TVB-N contents between cumin-treated fillets and untreated fillets. The TVB-N values increased according to time of storage. Generally, the amount of TVB-N was low throughout the iced storage. It has been shown that initial TVB-N values of a particular fish species is affected by fish feeding type, catching season and fish size (Goulas and Kontominas, 2007). Moreover, this change is related to age, harvesting area and culture method (Kyranas *et al.*, 1997) and sex of fish (Kilinc and Cakli, 2005). The level of TVB-N for white fish is generally considered to be fresh if the TVB-N is < 20 mg N 100 g^{-1} sample. If the TVB-N reaches 30 mg N 100 g^{-1} , most authorities consider the fish to be stale while at a level of 40 mg N 100 g^{-1} , the fish is regarded as unfit for consumption. A level of 30 mg muscle TVB-N 100 g^{-1} has been considered the upper limit above which some fishing products are considered spoiled and unfit for human consumption (Gokoglu *et al.*, 1998). Lang proposed that the quality classification of fish and fish products regarding TVB-N values would be high quality up to 25 mg N 100 g^{-1} , good quality up to 30 mg N 100 g^{-1} , limit of acceptability up to 35 mg N 100 g^{-1} and spoil above 35 mg N 100 g^{-1} . Varga *et al.* (1980) using an experienced sensory panel and a chemical analysis to evaluate the freshness of cod fillets, showed a TVB-N value of 20 mg N 100 g^{-1} for fresh fish, 60 mg N 100 g^{-1} for marginal fish and > 80 mg N 100 g^{-1} for inedible fish. In this study, the final TVB-N content of treated (10.78 ± 0.15 mg N 100 g^{-1}) and untreated (10.97 ± 0.15 mg N 100 g^{-1}) was lower than these limits indicating that the fillets of tilapia maintained at a good quality during the storage. Regardless of treatment, the results indicate that

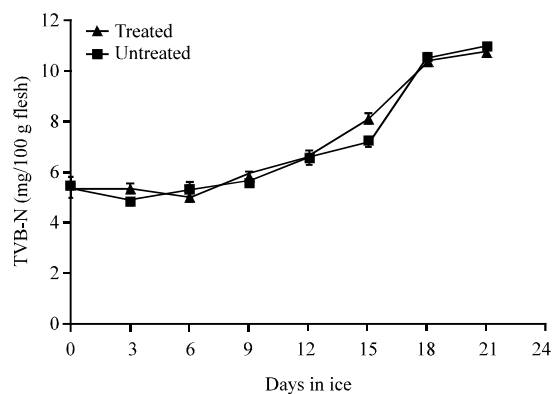


Fig. 1: The changes in TVB-N of cumin-treated and untreated tilapia fillets during the 21 days storage period in ice

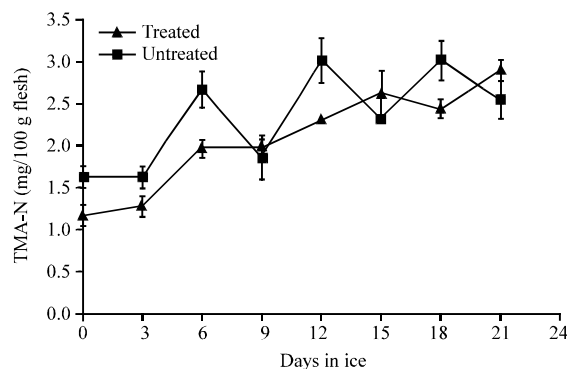


Fig. 2: The changes in TMA-N of cumin-treated and untreated tilapia fillets during the 21 days storage period in ice

from the beginning of the storage for tilapia fillets, the amounts of TVB-N were low during the edible storage period and only increased slightly when the fish was near to rejection (not exceed 10.97). Therefore, the TVB-N is considered to be unreliable for estimating the degree of freshness in the early stages of the storage of tilapia fillets as it does not reflect the degree of spoilage in the later stages.

Trimethylamine nitrogen: TMA-N is often used as an index in evaluating the shelf-life and keeping quality of fish products because it rapidly accumulates in the muscle under refrigerated conditions. The TMA-N production in fish tissue during cold storage could be used as an indicator of bacterial activity and is an accepted measure of deterioration. The pungent odour of spoiled fish has often been related to the TMA-N tissue levels also with the number of spoilage organisms present in many fish species, the rejection limit is usually from 5-10 mg

TMA-N 100 g^{-1} muscle (El-Marrakch *et al.*, 1990). The changes in TMA-N content in cumin-treated and untreated tilapia fillets stored in ice are shown in Fig. 2. Initially, TMA-N values of treated and untreated fillets were 1.16 ± 0.13 and $1.62 \pm 0.13 \text{ mg N } 100 \text{ g}^{-1}$ flesh, respectively by the day 6 and 9 of storage, the TMA-N values of treated samples remained steady, attaining a final value of $2.90 \pm 0.12 \text{ mg N } 100 \text{ g}^{-1}$ flesh by the end of the storage period (day 21). The highest value of $3.01 \text{ mg N } 100 \text{ g}^{-1}$ flesh was detected for untreated tilapia fillets on 12 and 18 days which was however within the limit of acceptability. In this study, the production of TMA-N in cumin-treated and untreated fillets increased significantly ($p < 0.05$) with time. Cumin powder exhibited a preservative effect in cumin-treated tilapia fillets since significant lower levels of TMA-N were observed during ice storage which may be attributed to the antibacterial properties of phenolic compounds of this herb. The relatively small increase in TMA-N over the storage period in ice reflects the low level of Trimethylamine Oxide (TMAO) in the flesh of fillets tilapia. In addition, it reflects the low number of spoilage organisms present in raw fish. TMA-N is produced by the decomposition of TMAO caused by bacterial spoilage and enzymatic activity. Both TVB-N and TMA-N values remained below the limit of acceptability throughout the entire storage period. Ehira and Uchiyama (1987) pointed out that spoilage odour in fish kept at 0°C did not appear before day 17,

where the viable bacterial count was in the order of 10^5 CFU g^{-1} , the minimum of the range considered to be the threshold for spoilage. From a review of the literature, a drastic increase in TMA-N associated with a Viable Bacterial Count (VBC) higher than 10^6 CFU g^{-1} of muscle was found which confirms the usefulness of TMA-N as an indicator of the bacterial spoilage onset rather than an indicator of freshness. The International Commission on Microbiological Specifications for Food (ICMSF) recommends that during fish storage on ice total Aerobic Plate Counts (APC) should never exceed 10^7 CFU g^{-1} wet weight (ICMSF, 1978). Both TVB-N and TMA-N values remained below the limit of acceptability throughout the entire storage period.

Sensory evaluation: Fresh red tilapia is generally considered to have very high acceptability for consumers. However, fresh fish is susceptible to spoilage caused by both microbiological and chemical reactions. The changes in sensory attributes of cooked cumin-treated and untreated tilapia fillets through 9 days of storage are shown in Fig. 3. The characteristic odour and flavour of cumin-treated and untreated tilapia fillets slightly decreased in intensity during ice storage. Despite the long storage time (9 days), the sensory attributes of cooked untreated and cumin-treated fillets remained similar to those of untreated and treated tilapia fillets at zero time. The storage time had no effect ($p < 0.05$) on the sensory attributes of ice all cooked tilapia fillets. No significant

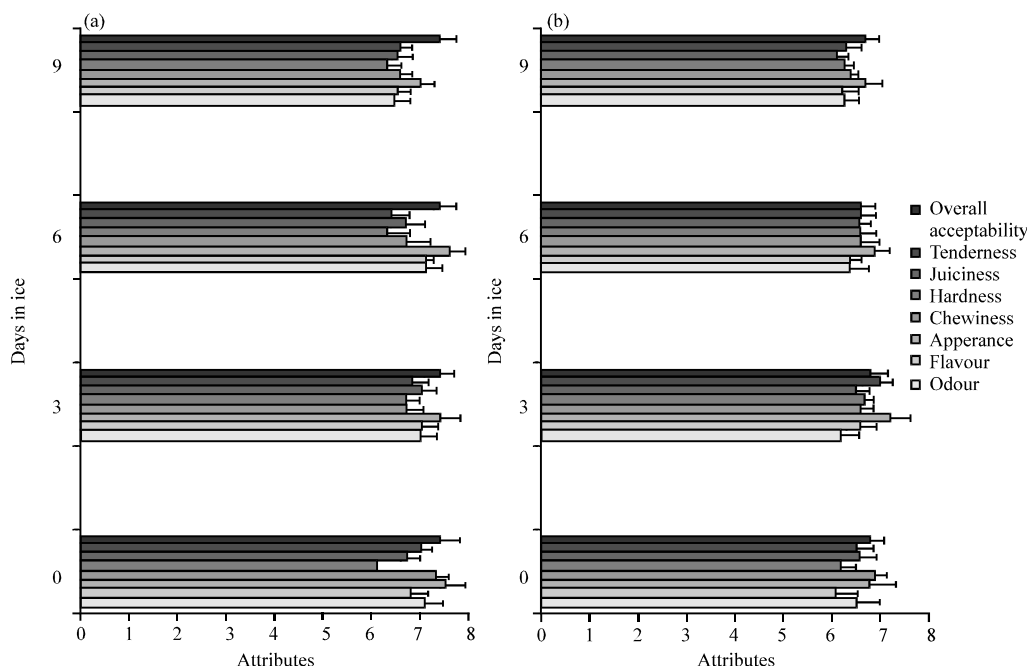


Fig. 3: Changes in sensory attributes scores of cooked fillets cumin-treated and untreated fillets during iced storage; a) Cooked cumin-treated tilapia fillets; b) Cooked untreated tilapia fillets

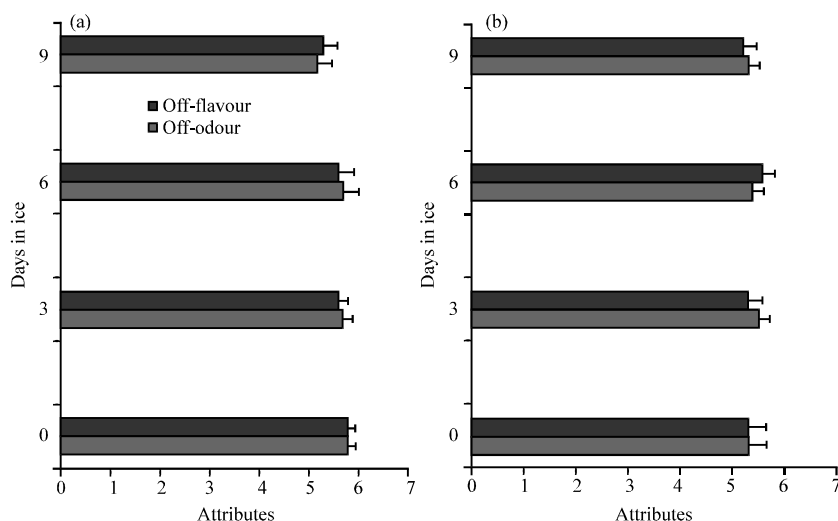


Fig. 4: Changes in off-odour and off-flavour intensity of cumin-treated fillets and untreated fillets during ice storage; a) Cooked treated tilapia fillets; b) Cooked untreated tilapia fillets

difference ($p < 0.05$) was detected for the juiciness, tenderness, chewiness, hardness between the control and cumin-treated samples. However, significant differences were found in odour, flavour, appearance and overall acceptability between untreated and cumin-treated fillets. At the end of the present study; the cumin-treated tilapia fillets showed the highest score (7.4 ± 0.30) of overall acceptability while the score for untreated fillets was (6.7 ± 0.26). The sensory assessment of all cooked tilapia fillets showed that no score for the fillet attributes dropped to the limit of acceptability, since all mean acceptability scores were still above 4 by the end of the evaluation. The present findings showed that cumin-treated tilapia fillets maintained their quality better than untreated fillets during the ice storage, since they achieved higher scores than the untreated fillets.

Generally, the off-odour or off-flavour in fish and fish products causes a major reduction in acceptability for consumers or makes them unsuitable for sale (Robin *et al.*, 2006). The formation of secondary lipid oxidation products is one of the main causes of the development of undesirable odours in fish muscle especially the fishy odour (Thiansilakul *et al.*, 2010). The compounds associated with off-odour or off-flavour are generated by enzymatic reactions, lipid auto-oxidation, microbial action and environmentally or thermally derived reaction (Selli *et al.*, 2009). Amongst these reactions, lipid oxidation is closely related to the alteration of odour and flavour in fish (Maqsood and Benjakul, 2011). Figure 4 shows the changes in off-odour and off-flavour of cooked cumin-treated and untreated tilapia fillets through 9 days of storage. At the beginning of the study, the off-odour

and off-flavour of cumin-treated and untreated fillets had higher scores and then slightly decreased ($p < 0.05$) with time. The off-odour and off-flavour values of cooked cumin-treated and untreated fillets at day 0 were 5.80 ± 0.13 , 5.30 ± 0.33 and 5.80 ± 0.13 , 5.30 ± 0.33 , respectively whereas at 9 days of iced storage, they were 5.20 ± 0.25 , 5.30 ± 0.21 and 5.30 ± 0.26 , 5.20 ± 0.25 , respectively. No significant differences ($p < 0.05$) were identified for the intensity of the off-odour and off-flavour scores between the control and fillets treated with cumin. The formation of TMA depends on the content of TMAO which are primarily found in marine fish and results in a detectable fishy odour (Gram and Huss, 1996). In this study, the small detectable amounts of TMA-N and TVB-N in red tilapia, a freshwater fish, could reflect the good quality attributes of all tilapia fillets which kept an edible quality during at least 21 days of storage, since they were evaluated with higher scores.

CONCLUSION

No significant differences ($p < 0.05$) in TVB-N parameter exist between cumin-treated and untreated fillets. However, cumin inhibitory effect was more marked in TMA-N values and sensory attributes for treated than untreated fillets. Regardless of treatment, the results indicate that from the beginning of the storage for tilapia fillets, the amounts of TVB-N and TMA-N were low during the edible storage period and only increased slightly at the end of storage (not exceed 10.97 and 3.01, respectively). Therefore, the TVB-N and TMA-N are considered to be unreliable for estimating the degree of

freshness in the early stages of the storage of tilapia fillets as they do not reflect the degree of spoilage in the later stages. The sensory assessment of cooked tilapia fillets showed that no score of the fillet attributes dropped to the limit of acceptability, since all mean acceptability scores were still above 4 by the end of the evaluation. The present findings showed that cumin-treated tilapia fillets maintained their quality better than untreated fillets during the ice storage since they achieved higher scores than untreated fillets. Further studies are needed to identify specific spoilage bacteria in tilapia fish that could be responsible for fish quality differences.

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