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Shelf-life evaluation of fresh catfish (*Pangasianodon hypophthalmus*) fillets at different storage temperatures

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ABSTRACT

Shelf life of fish fillets from tra catfish (*Pangasianodon hypophthalmus*) was evaluated on the basis of chemical, microbiological and sensory quality during storage. The catfish fillets packed in polyamide package combined with vacuum level of 60% were stored at 0, 4, 8, 12±1.0°C. The quality of packed catfish fillets was assessed at the regular time intervals for a period of 25 days. The changes of quality were strongly dependent on the storage temperatures. The shelf-life of catfish fillets stored at 0, 4, 8, 12°C was, therefore, prolonged up to 21, 11, 7 and 3 days, respectively. In addition, total volatile basic nitrogen correlated well ($r = 0.953$) with the total microbial counts while the total microbial counts also correlated well ($r = 0.905$) with lactic acid bacteria counts. However, the weak correlation was shown between the total volatile basic nitrogen and the lactic acid bacteria counts ($r = 0.087$). The results obtained can be used as a reference tool to improve fishery quality management and to minimize the economic losses as well.

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1 INTRODUCTION

Pangasianodon hypophthalmus (tra fish or striped catfish) is a good source of various nutrients, namely protein quality, fatty acid, minerals, etc. that are required for human health (Karl *et al.*, 2010; Szlinder-Richert *et al.*, 2011; Usydus *et al.*, 2011; Ruiz-de-Cenzano *et al.*, 2013). This has become appreciated by consumers from different markets all over the world. Up to date, *Pangasianodon hypophthalmus* products, particularly frozen forms, are exported to 145 countries (VASEP, 2014). Besides frozen tra catfish, thawed tra catfish fillets considered as (re)fresh fillets have become increasingly popular in exported countries.

Product quality and shelf life of fishery products, rapid deterioration of fish are mainly caused by microbiological spoilage, leading to reduced shelf life and economic loss. Microbiological spoilage

can be manifested in visible growth, textural changes or off-odors and off-flavors (Gram, 2010). The production of these off-odors depends not only on the intrinsic characteristics related to chemical composition of fish, but also on the extrinsic parameters of storage temperature (Olafsdottir *et al.*, 2006). In general, fish is kept in chilled storage and preferable on ice. The storage temperature influences the growth of spoilage microbiology. More specifically, the growth of *Pseudomonas* spp., *Shewanella putrefaciens*, and *Psychrobacter immobilis* were promoted under chilled and iced conditions (Gennari *et al.*, 1999). In contrast, abusive temperature can create an optimal environment for strong microbiological spoilage such as *Photobacterium phosphoreum* or other microorganisms which are able to produce biogenic amines (Olafsdottir *et al.*, 2006).

In addition to temperature, storage conditions (i.e. aerobic storage, vacuum, modified atmosphere packaging) also impact on the shelf life of fish products. Modified atmosphere packaging (MAP), which employs elevated CO₂ and /or reduced O₂ concentration, is commonly applied storage method to extend the shelf life of fishery products. So far, only one study determined the shelf life of thawed tra catfish fillets stored under different MAP conditions at 4°C (Noseda *et al.*, 2012). On the other hand, the commercial aquaculture of tra catfish is still a young industry in Vietnam, which partly explains the little information concerning the shelf life and the quality of tra catfish during storage, particularly with the fresh tra catfish. The objective of the present study was to investigate the effect of temperature conditions on microbiological, physicochemical and sensory quality of fresh tra catfish fillets during storage.

2 MATERIALS AND METHOD

2.1 Experimental set-up

Twenty-five kg of *Pangasianodon hypophthalmus* fillets were bought from a company, located in Hau Giang province, processing tra catfish for export. After purchasing, the tra catfish fillets were stored in ice and transported in insulated boxes to the Laboratory of Microbiology and Biotechnology of Food Technology Department, Can Tho University for further storage experiments within 1 hour. At the laboratory, the fillets were washed in the batch consisting of a ratio fish to ice was 1:2. After draining, each fillet ranged from 150 to 200 g was then packed aseptically in PA (polyamide) package combined with vacuum level of 60% by means of vacuum packaging machine (Tecnovac 5100H, Italy). PA packages in size of 19x16 cm and thickness of 85 µm used for packing the samples were supplied by Cam Dao Company (District 3, Ho Chi Minh city, Vietnam). The control samples were fillets without packaging, stored under air. Considering the importance of the quality and shelf-life extension of fresh fish, this study was designed to evaluate the effect of storage temperatures (0°C, 4°C, 8°C and 12°C) on fish quality. The packaged fillets were stored under different simulated temperatures by adjusting the temperature in the refrigerator at 0, 4, 8, 12 ± 1.0°C. The temperature of refrigerators (Sanaky VH358W and Sanyo MPR-414F, Japan) was adjusted at different temperature levels accordingly. After adjusting the temperature levels, the set-up temperature was regularly checked from five to eight hours by thermometer. The packaged fillets of tra catfish were stored in the refrigerators at a set-up temperature for a period of 25 days. On a regular basis (day 0, 3, 5, 7, 9,

11, 13, 15, 17, 19, 21, 23 and 25 of storage), three samples of tra catfish were randomly selected for the assessment of pH, the total volatile compound content, microbiological quality and sensory quality.

2.2 Analytical methodology

On a regular basis of storage periods, different parameters of pH, total volatile basic nitrogen content, microbiological analysis, and sensory quality were determined in triplicate. The analytical methodology was based on TCVN 8338:2010 (TCVN, 2010). pH was measured with a pH-electrode (Mettler Toledo, Schwerzenbach, Switzerland). Total volatile basic nitrogen (TVB-N) was based on steam distillation of an alkalized samples and absorbed by boric acid solution (1%) and further titration using sulfuric acid solution (0.1N) described by Antonacopoulos and Vyncke (1989). For microbiological analysis, a 25 g composite sample from different parts of each fillet sample was transferred aseptically to a stomacher bag by means of sterile scalpels and tweezers. 225 ml of sterile Maximum Recovery Diluent (MRD, Merck, Darmstadt, Germany) was added and the mixture was homogenized for 1 minute. Further decimal dilutions were prepared in MRD. The total mesophilic counts and lactic acid bacteria counts were determined by pour plating the decimal dilutions on Plate Count Agar (PCA, Merck, Darmstadt, Germany) and on de Man Rogosa Sharpe agar (MRS, Merck, Darmstadt, Germany) (with an additional over layer), respectively. The pour plates were incubated for 3 days at 37°C to determine the total mesophilic and lactic acid bacteria counts. With regards to sensory evaluation, there were nine panellists that participated in the sensory tests. The sensory evaluation was assessed using a scoring scale from 1 to 5 including odor, color and texture (Meenakshi *et al.*, 2010).

2.3 Statistical analysis

All experiments were performed in triplicate. The results of microbiological analysis of the fillets were expressed as log CFU/g. Results are reported as mean value ± standard deviation of these triplicate analyses. Cross-correlation were tested by means of the non-parametric Spearman rank order correlation coefficients (*r*) two tailed test ($\alpha = 0.05$) in SPSS version 20 (IBM Inc., Chicago, Ill., USA).

3 RESULTS AND DISCUSSION

The protein, lipid and water content of tra catfish fillets samples using in the present study were 16.23%, 1.71% and 80.56%, respectively (data not shown). It was a good source of nutrient for micro-

bial growth and subsequently induced physicochemical and sensory changes.

3.1 Effect of storage at different temperatures on microbiological quality

The evolution of total mesophilic counts and lactic acid bacteria is showed in Fig. 1A and 1B, respectively. The growth rate of those microbial counts depended on the temperatures stored. During storage, the lower temperature would have resulted in the lower microbial evolution. The initial total mesophilic count was 4.7 ± 0.1 log CFU/g at day 0 and on day 5 this level increased to 5.1 ± 0.1 , 5.3 ± 0.0 , 5.8 ± 0.3 and 7.0 ± 0.1 log CFU/g on samples stored under 0, 4, 8 and 12°C, respectively. The initial total mesophilic count was consistent with the previous study of thawed tra catfish (Noseda *et al.*, 2012; Tong Thi *et al.*, 2016). The total mesophilic count upper limit of acceptability, which is 7.0 log CFU/g for freshwater fish as tra catfish (ICMSF, 2002) was exceeded after 25, 15, 9 and 5 days of storage at 0, 4, 8 and 12°C, respectively.

Obviously, temperature is one of the factors that affect greatly the growth rate of the microorganism (Dalgaard *et al.*, 1997). On the contrary, the number of lactic acid bacteria appeared somewhat no lag phase during storage as the initial lactic acid bacteria was 2.7 ± 0.1 log CFU/g followed by a rapid increase to 3.2 ± 0.0 ; 3.6 ± 0.3 ; 3.9 ± 0.1 ; 4.0 ± 0.1 log CFU/g on samples after 5 days of storage under 0, 4, 8 and 12°C, respectively. The lactic acid bacteria may result in cross contamination and environmental contamination during processing (Tong Thi *et al.*, 2013). The present study, tra catfish fillets were packed in PA packages under vacuum of 60%. As a result, the lactic acid bacteria became dominant towards the end of shelf life. A strong correlation was observed between the total mesophilic counts and lactic acid bacteria ($r = 0.905$). The lactic acid bacteria which are tolerant to CO₂ considered to dominating spoilage organisms on MAP freshwater fish as tra catfish (Arkoudelos *et al.*, 2007; Noseda *et al.*, 2012).

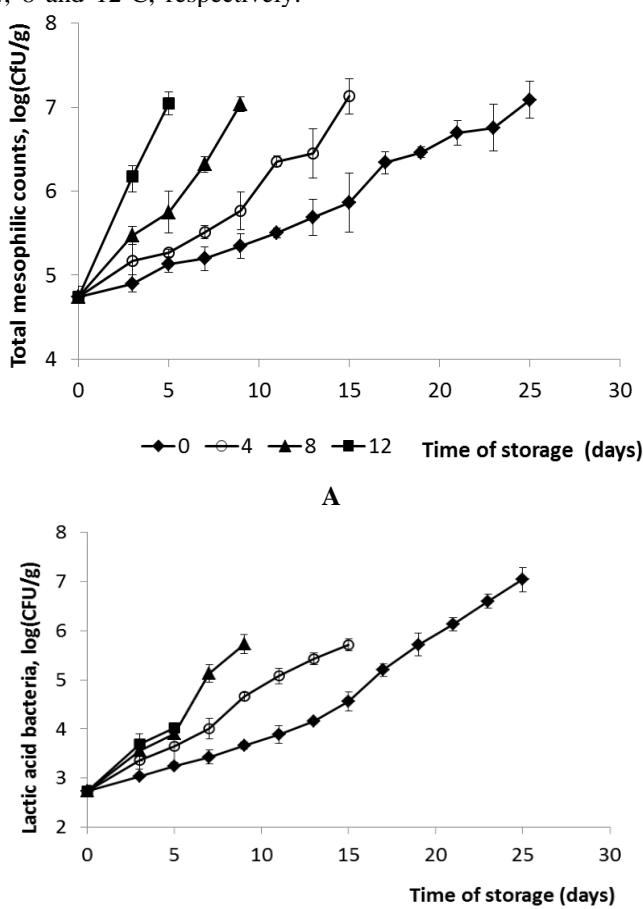


Fig. 1: Evolution of total mesophilic counts (A) and lactic acid bacteria (B) of fresh tra catfish stored at 0°C (◆); 4°C (○); 8°C (▲) and 12°C (■). Bars represent the standard deviation of three independent samples

3.2 Effect of storage at different temperatures on physicochemical quality

The evolution of TVB-N content and pH of tra catfish fillets samples stored at 0, 4, 8, 12°C is depicted in Fig. 2A and Fig. 2B, respectively. The initial pH was 6.72 and decreased slightly between 6.46 and 6.68 (Fig. 2A). Thereafter, the pH increased throughout the storage period. The rate of change in pH increased with the temperature storage, i.e. pH of 6.43 (at 0°C), 6.48 (4°C), 6.58 (8°C) and 6.92 (12°C) for a period of 5 days. The TVB-N is a quality parameter indicating spoilage of fish during storage (Connell, 1995). The TVB-N content of tra catfish samples kept under different temperatures is depicted in Fig. 2B. Similar results were observed in pH evolution, the TVB-N change was higher at high temperature than at low temperature. The value of TVB-N was 12.84 ± 0.81 mg/100g at day 0. At day 5, the TVB-N content

increased to 14.71 ± 1.21 , 16.34 ± 0.81 , 22.41 ± 1.85 and 30.58 ± 1.07 mg/100 g of tra catfish fish during storage at 0, 4, 8 and 12°C, respectively. Afterward, they continued to increase to 27-31 mg/100 g at the end of shelf life. The results obtained can be explained that microbiological growth with a shorter lag phase at higher temperature condition inducing the faster evolution of TVB-N. As the TVB-N content was associated with fishery product spoilage, an increase in the TVB-N content in those samples indicated the stage of substantial spoilage of the muscle (Masniyom *et al.*, 2013). A noticed finding in the present study is the TVB-N content correlated well with the total mesophilic counts ($r = 0.953$). In contrast, a weak correlation was observed between the TVB-N content and lactic acid bacteria ($r = 0.087$). It is suggested that the TVB-N can be used as an index to predict the total microbial counts on tra catfish during storage.

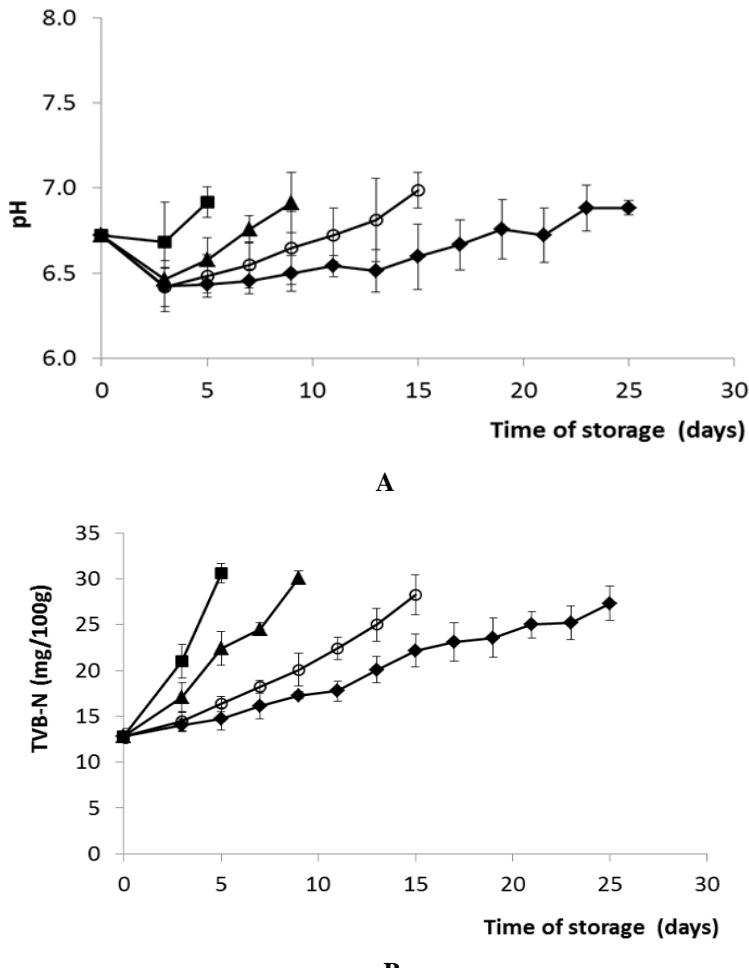
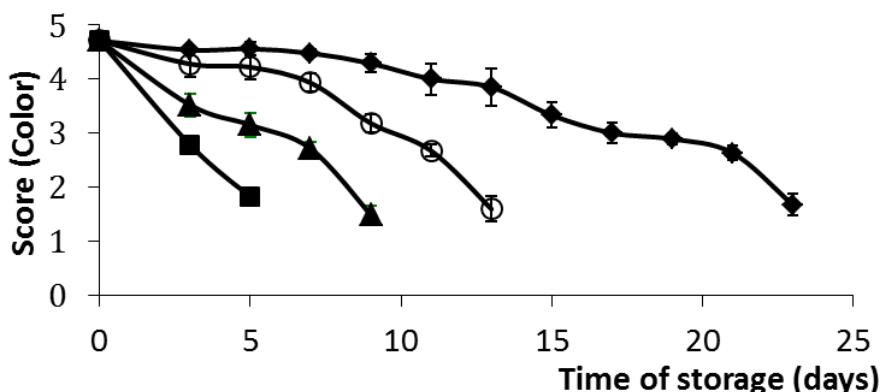


Fig. 2: Evolution of pH (A) and total volatile basic nitrogen (TVB-N) (B) of fresh tra catfish stored at 0°C (◆); 4°C (○); 8°C (▲) and 12°C (■). Bars represent the standard deviation of three independent samples

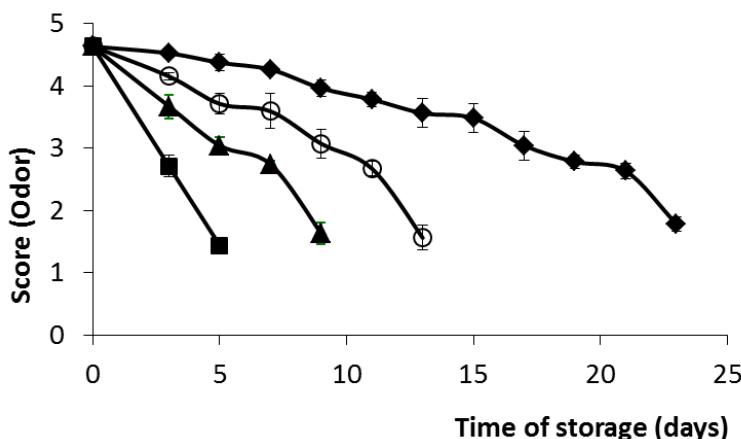
3.3 Effect of storage at different temperatures on sensory quality

The results of sensory attributes (color, odor, and texture) of tra catfish fillets are shown in Fig. 3A, 3B, and 3C. Fresh tra catfish had white color, no off-odor and hard texture were therefore considered to possess very high acceptability (score of 4.6-4.7 at day 0). Among them, the samples stored at 0°C were preferable with the highest score compared to those other samples stored at higher temperature (i.e. 4, 8 and 12°C) during storage time. The differences in the quality scores during storage conditions are possibly due to the effect of temperature to the enzymatic and microbial activities in fish samples, whereas low temperatures delayed the enzymatic and microbial activities. Graham *et*

al. (1992) also reported that fish quality maintained for 2 days (at 15°C), 4 days (5°C) and 10 days (0°C) before becoming unacceptable for human consumption. The sensory quality, is normally applied in estimating the freshness of fish, had a correlation with chemical and microbiological quality (Karungi *et al.*, 2004). Moreover, the sensory and chemical quality agreed well with the microbiological quality (Ghaly *et al.*, 2010). Therefore, in addition to the sensory quality, the limits of acceptability were based on chemical quality (i.e. TVB-N of 35 mg N/100 g) and microbiological quality (i.e. total aerobic counts of 7 log CFU/g) as a tool to estimate the shelf life of fresh tra catfish during storage. The shelf-life of tra catfish fillets stored at 0, 4, 8, 12°C was up to 21, 11, 7 and 3 days, respectively.



A



B

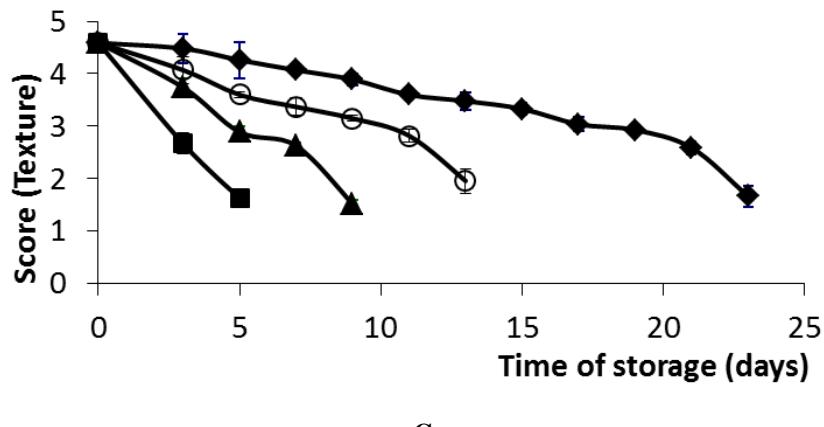


Fig. 3: Sensory evaluation of color (A), odor (B) and texture (C) of fresh tra catfish during storage at 0°C (◆); 4°C (○); 8°C (▲) and 12°C (■). Bars represent the standard deviation of three independent samples

4 CONCLUSION

Temperature had a great impact on the shelf life of tra catfish fillets stored in PA packaging with vacuum level of 60%. The use of low temperature might retard the microbiological and physicochemical changes, leading to decrease in spoilage and prolongation of the shelf life of fresh tra catfish fillets. The tra catfish stored at 0°C was observed the optimal temperature for shelf life extension. More importantly, the present study shows that TVB-N value is valid quality index for prediction the microbiological quality of tra catfish fillets.

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