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Research Article:

Determination of freshness quality changes using selective methods: A comparative study for eclipse parrotfish (*Scarus russelii*) and spangled emperor (*Lethrinus nebulosus*)

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Abstract

The present study was carried out to determine freshness quality and remaining storage time of eclipse parrotfish and spangled emperor stored in ice on days 0, 2, 3, 5, 7, 9, 12, 15, 19 and 21 using selective methods. A score of 11-12 coincided with the level at which the fish were considered unacceptable by a sensory panel of six experienced assessors. The demerit score of eclipse parrotfish and spangled emperor increased linearly (R^2=0.9563 and 0.9624, respectively) with storage time. The limit for acceptability of eclipse parrotfish and spangled emperor stored in ice was 9 and 19 days respectively. Total volatile base nitrogen value (45.87±1.10 mg/100g) for parrotfish stored in refrigerator exceeded limit of acceptability at the time of sensory rejection (9th day) while the value (5.93±0.28 mg/100 g) for spangled emperor was remained below the limit of acceptability after 9 days and increased to 54.96±1.80 mg/100g at the 19th day. The pH for both fishes showed significant (p<0.05) increases during the storage. Total viable count in spangled emperor analyzed were found to be below the acceptability limits (6 log10 cfu/g) by 15 days while maximum level of acceptability of eclipse parrotfish was until 7 days. Shelf-life of both fishes was different. Sensory score, value of TVB-N, pH and TVC provide evidence that the limit for acceptability was 7 days for eclipse parrotfish and 15 days for spangled emperor stored at 0 - 2^0C.

Keywords: Eclipse parrotfish, Freshness quality, Spangled emperor

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Introduction

Fish freshness is the most important attribute when assessing the quality of fish and fishery product, can be explained by some objectives sensory, biochemical, microbial and physical parameters (Venugopal, 2006). Securing post-harvest benefits through control of fish loss has long been concern of development practitioners committed to improving the livelihoods of fishermen, processors and traders (Gbola and Yvette, 2010). Fish chilling is one of the most efficient ways to extent the freshness of fish and is very important component in the Sri Lankan fishery production.

Marine fisheries in Sri Lanka play a vital role in contributing directly to foreign exchange earnings. Exporting marine fish fauna provide a greater commercial value to the country economy, which was LKR 24,716 Million in year 2015 (Fisheries Statistics, 2016).

Eclipse parrotfish (Scaridae) *Scarus russelii* (Valenciennes, 1840) are important food and brightly coloured fish species which found in relatively shallow tropical oceans. It is not a major targeted commercial catch, but may form a significant component of artisanal fisheries and are frequently found in local fish markets (Choat *et al.*, 2012). Even though they are highly popular food fishes in some areas, catch data on parrotfishes are not currently reported to Food and Agricultural Organization of the United Nations. They are usually caught in traps, gill nets, or by spearing. Even though it is not commonly consumed in Sri Lanka, a commercial fishery exists in the Indo-Pacific Ocean. Spangled emperor (Lethrinidae) *Lethrinus nebulosus* (Forsskål, 1775) are commercially exporting aquatic fauna in Sri Lanka by their tasty and meaty. It is major interest to fisheries (De Bruin *et al.*, 1994).

Few studies have focused on freshness assessment of fishes in Sri Lanka. Jayasinghe and Rajakaruna (2005) identified the bacterial contamination of tuna fish sold in fish markets in the central province of Sri Lanka. A comparative quality assessment of five types of selected fishes such as yellowfin tuna, sailfish, sardine, shrimp and squid collected from retail market in Sri Lanka investigated by Jianadasa *et al.* (2014). Quality attribute of spangled emperor (*Lethrinus nebulosus*) in Saudi Arabia is recorded by Shady *et al.* (2016). Microbial assessment of spangled emperor collected from Dammam fish market in Saudi Arabia is conducted by Sami *et al.* (2016).

Though there are some studies on freshness analysis for different fishes, studies on eclipse parrotfish and spangled emperor in Sri Lanka are still not elucidated so far. As the market demand for fresh eclipse parrotfish and spangled emperor is quite increased, the present study will be useful to both retailers and consumers. Therefore, the aim of the present study was to analyze the freshness changes in terms of

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sensory, chemical, physical and microbiological changes during ice storage. In addition, it is also anticipated that the freshness quality of both fishes be served as a base for further comparison of the quality of these fish handled under commercial conditions.

Materials and methods

Sampling and storage

A total of 106 freshly caught *S. russelii* (40-50 cm in total length and 450-550g in total weight) and *L. nebulosus* (40-55 cm in total length and 250-350g in total weight) were obtained from the auction market in Jaffna Peninsula during the period of April 2016 to February 2017 and transported to the laboratory. Fish samples were washed and cleaned in running tap water and held un-gutted in sterile Reynolds zipper polythene bags and distributed uniformly in freezer compartment of refrigerator at 0 - 2^{o}C with fish/crushed ice ratio of 1: 2 (W/W).

Sensory, chemical, physical and microbiological analysis for each was performed on days 0, 2, 3, 5, 7, 9, 12, 15, 19 and 21. On the day of analysis, six of each fishes were taken randomly. The specimens were allowed to thaw by water immersion method (Archer *et al.*, 2008). Of these six specimens, three were used for sensory analysis and reused to chemical and physical analysis. The other three were used for microbiological analysis.

Determination of sensory score (Hyldig *et al.*, 2007)

Sensory analysis on the basis of skin appearance, body stiffness, belly condition, eye clarity and shape, gill appearance, gill-mucus and odour of each fishes was performed by a sensory panel of six experienced assessors. The guidelines used to assess the quality are given in Table 1. The sensory scale is based on the freshness quality grading system developed by Hyldig *et al.* (2007) and it was applied on whole samples from day 0 to the day on which fish is spoiled, in order to obtain quality index score. Each attribute was scored from 0 to 3, where 0 represented prime freshness and any higher score indicated sign of spoilage. The scores for all attributes were then summed to give an overall sensory score called as quality index.

Determination of Total volatile bases Nitrogen (TVB-N) (Malle and Tao, 1987)

Hundred g of flesh of each sample were homogenized with 200ml of 7.5% (v/v) aqueous trichloroacetic acid (TCA) solution in a blender for 1 min. The homogenate sample was centrifuged at 3000 rpm for 5 min and the supernatant liquid was filtered using suction pump. 25 milliliter of the filtrate was loaded into a Kjeldahl distillation tube, followed by 5 ml of 10% (w/v) aqueous NaOH solution. Steam-distillation was performed using a vertical steam-distillation unit, and the distillate was received into a beaker containing 15 ml of 4% (v/v) aqueous boric acid solution

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up to a final volume of 50 mL. The titration was performed against aqueous 0.5 N Sulphuric acid solutions.

Table 1: Sensory assessment scheme for *Eclipse parrotfish and* Spangled emperor.

Parameters assessed	Demerit points[a]			
	0	**1**	**2**	**3**
Skin appearance	Bright shining	Bright	Dull	----
Body stiffness	Stiff	Elastic	Firm	Soft
Belly condition	Firm	Soft	Belly burst	----
Smell	Fresh	Neutral	Musty	Rancid
Eye clarity	Clear	Cloudy	----	----
Eye shape	Normal	Plain	Sunken	----
Gill appearance	Red	Faded	Discoloured	----
Gill- Mucus	Absent	Transparent, gelatinous	Milky	Transparent, watery
Gill-Odour	Sea weedy	Neutral	Rotten and/or metallic	----

[a] Demerit point scale -20

Determination of pH value (Ozogul *et al*., 2006)

The samples were homogenized in distilled water in the ratio of 1: 10 (W/V). The pH of fillet was determined using a pH meter (Eco TestrpH 2).

Determination of TVC (SLS, 1991)

A 25g of flesh sample was transferred to a blender. Peptone water (225 ml of 0.1%) with salt (NaCl, 0.85% [W/V]) was added, and the mixture was homogenized for 60s. Serial dilution was carried out and 0.1 ml of samples was spread on Nutrient Agar Medium under aseptic conditions. Plates were incubated at 37°C for 24-48 hrs. Three replicates were also made. Aerobic bacterial count was made after incubation and the results were expressed as log 10 cfu/g.

Statistical analysis

The data were statistically analyzed using SPSS Software, version 10 (Stat Soft, Inc. 1995, Tulsa, OK, USA). The data were checked for normal distribution with one sample Kolmogorov-Smirnov test and the variances were checked in the Levene's test for homogeneity. Then the data were analyzed by one-way Analysis of Variance (ANOVA). Significant differences were defined as $p<0.05$.

Results

Comparison of quality index score

Changes of sensory demerit score of eclipse parrotfish and spangled emperor over the 21 days storage in refrigerator are shown in Figure 1. In this scheme, 0 represents absolute freshness of fish and 20 represent completely spoiled fish. A score of 11-12 coincided with the level at which the fish were considered unacceptable by members of the panel. The demerit score of eclipse parrot fishes and spangled emperor increased linearly (R^2=0.95 and 0.96 respectively) with storage time. The

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limit for acceptability of eclipse parrotfish and spangled emperor stored in refrigerator was 9 and 19 days.

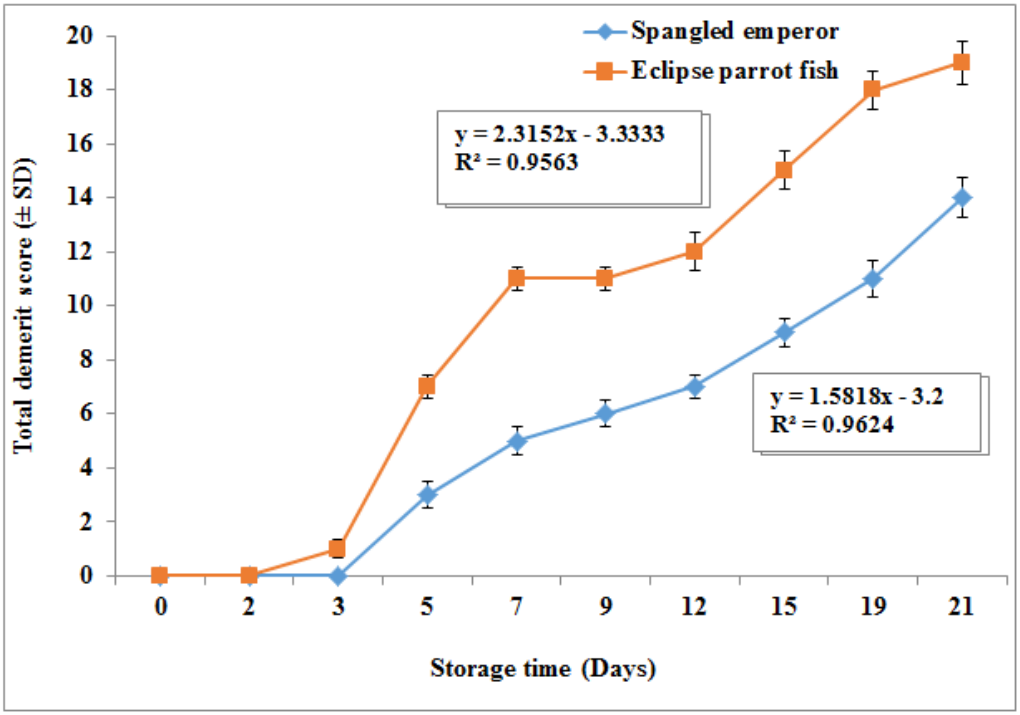


Figure 1: Changes of sensory demerit score of eclipse parrotfish and spangled emperor over the 21 days storage in ice.

Comparison of TVB-N

Value of TVB-N in eclipse parrotfish and spangled emperor stored in refrigerator are shown in Figure 2. Initial TVB-N values were 12.62±0.05 mg/100g and 9.22 ± 1.63 mg/100g for eclipse parrotfish and spangled emperor, respectively and thereafter the values increased to 71±3.16 mg/100g and 100.87±1.21 mg/100g after 21-days storage. Value of TVB-N for spangled emperor was not changed significantly (p<0.05) in the first three days and recorded between 9.22±1.63 to 10.64±1.16 mg/100g. TVB-N value of 45.87 (±1.10 mg/100g) for eclipse parrotfish stored in ice exceeded limit of acceptability at the time of sensory rejection (9th day) while TVB-N value of 5.93 (±0.28 mg/100g) for spangled emperor was remained below the limit of acceptability after 9 days in ice storage and increased to 54.96± 1.80 mg/100g at 19th day (sensory rejection time).

Comparison of pH values

The pH values for eclipse parrotfish and spangled emperor were 6.72±0.05 and 6.92±0.05 respectively at the beginning of the storage (Fig. 3). Although all pH values of studied fresh fish were below 7, there were significant differences between both fish species in pH value. There was a slight increase in pH over storage, but changes in the pH values

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between the fish species during the entire storage trial were not statistically significant (p>0.05). The pH values in eclipse parrot fishes and spangled emperor (8.07±0.05 and 7.77±0.05 for eclipse parrotfish and spangled emperor, respectively) showed statistically significant increases at the end of the storage period (p<0.05).

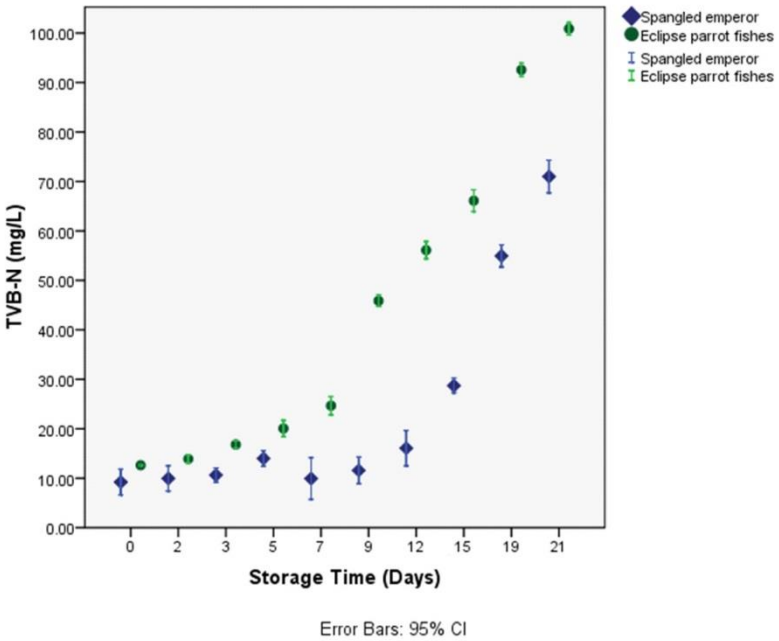


Figure 2: Changes of TVB-N value (mean ± SE) for eclipse parrotfish and spangled emperor over 21 days storage in ice.

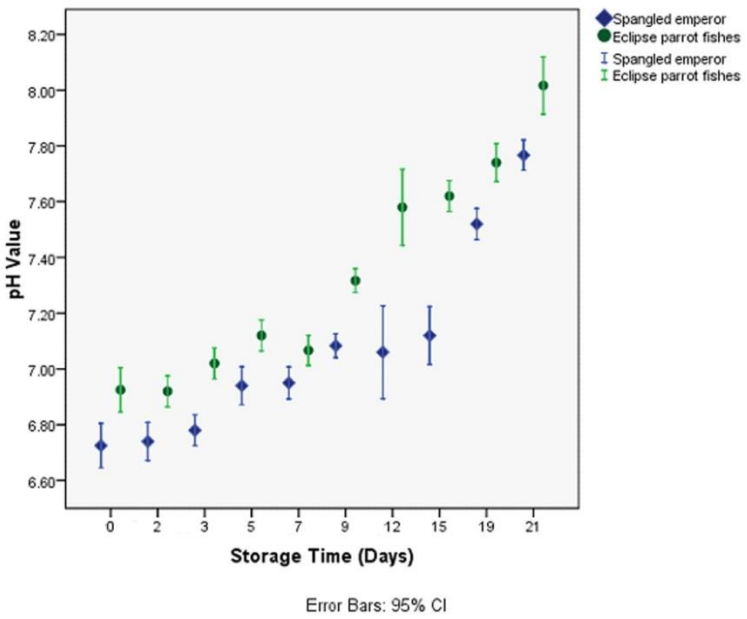


Figure 3: Changes of pH value (mean ± SE) for eclipse parrotfish and spangled emperor over 21 days storage in ice.

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Determination of total viable count

Changes of microbial counts in the eclipse parrotfish and spangled emperor kept in ice are shown in Figure 4. There were increases in total viable counts (TVC) over the period of storage in the present study. Initial aerobic bacterial count was recorded as 3.57 and 3.62 log 10 cfu/g in eclipse parrotfish and spangled emperor, respectively. TVC levels in spangled emperor analyzed were found to be below acceptability limits (6 log10 cfu/g) by 15 days while maximum level of acceptability of eclipse parrot fishes was until 7 days.

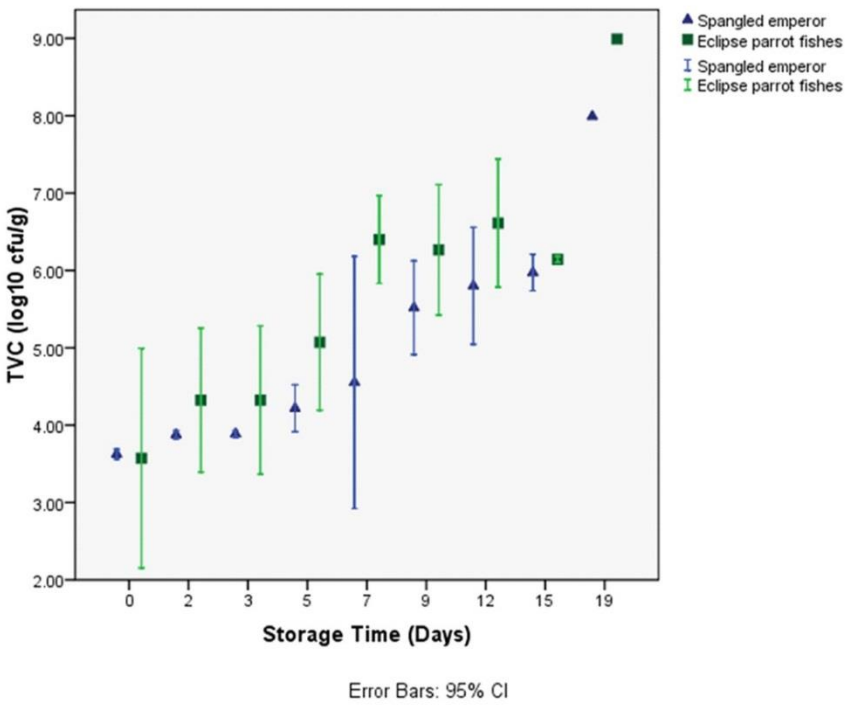


Figure 4: Changes of TVC (log10 cfu/g) for eclipse parrotfish and spangled emperor over 21 days storage in ice.

Discussion

Quality index score method has been considered a reliable sensory tool for assessing the freshness of fishery product. A sensory score of 11-12 coincided with the level at which the fish were considered unacceptable by members of the panel. Different rates of quality deterioration between different fish species is previously reported (Huis In't Veld, 1996; Chytiri *et al.*, 2004; Boulter *et al.*, 2006). Although the initial sensory score of both fishes was the same on day 3, this score for eclipse parrotfish was significantly higher than that for spangled emperor after 3 days. This could possibly be explained by a soft flesh texture in the eclipse parrotfish that might cause faster deterioration of its flesh.

Total volatile basic nitrogen (TVB-N) quantifies the content of ammonia,

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trimethylamine (TMA) and dimethylamine (DMA) in fish (Howgate, 2009). The concentrations of TVB-N are widely used to evaluate fish quality deterioration. Özyurt *et al.* (2009) proposed that quality classification of fish and fish products based on TVB-N values would be ‘‘high quality” up to 25 mg/100g, ‘‘good quality” up to 30 mg/100g, ‘‘limit of acceptability” up to 35 mg/100g, and ‘‘spoilt” above 35 mg/100g.

In the present study, the value of TVB-N for spangled emperor was not changed significantly (p<0.05) in the first three days and recorded between 9.22±1.63 to 10.64±1.16 mg/100g. These results therefore agree with previous records by Boițeanu *et al.* (2014), which was 10.2 mg/100g for emperor sea bream. In addition, Jeyasekaran *et al.* (2004) reported initial amounts of 11.5 mg/100g for *Lethrinus miniatus*. In the present study, initial value of TVB-N for spangled emperor was lowest than those reported by Shady *et al.* (2016), which was 24.35±2.81 mg/100g. In previous studies, the value of TVB-N for blue-spotted emperor was at the highest EU limit of acceptability after 5 days storage at 4°C (Fuentes-Amaya *et al.*, 2016).

A pH value of fresh fish ≤ 6.2 indicates good quality (FAO, 2015). If pH values are more than 7, fish is spoiled, this is because some chemical changes after death occurred and some basic products released make rise in pH value. Although all pH values of studied fresh fish were below 7, there were significant differences between both fish species in pH value. The pH values are in agreement with reported postmortem pH range of 6.0-6.8 (Howgate, 2009). Increases in the pH of both fishes are related to accumulation of alkaline compounds, such as ammonia mainly derived from microbial action during fish muscle spoilage.

Initial aerobic bacterial count was recorded from 3.57 to 3.62 log 10cfu/g in eclipse parrotfish and spangled emperor respectively, which was lower value than reported by Chummun and Neetoo (2016). At the beginning of storage, microbiological levels may vary among finfish species, which depends on harvest water temperature, handling and storage situation (Huis In’t Veld, 1996; Chytiri *et al.*, 2004). TVC for fish exceeding 7 log 10cfu/g is considered as the maximum level for acceptability. In the present study, TVC levels in analyzed spangled emperor were found to be below the acceptability limit (6 log 10cfu/g) by15 days while maximum level of acceptability of eclipse parrot fishes reached in 7 days. It can be concluded that bacteria grew more quickly in eclipse parrotfish than spangled emperor during the storage periods. This conclusion implies that sensory analysis correlated well with the microbiological analysis of the fish.

In conclusion, shelf-life of both fishes was different. The data on sensory

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score, value of TVB-N, pH and TVC provide evidence that the limit for acceptability was 7 days for eclipse parrotfish and 15 days for spangled emperor when stored in ice.

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