Quality Changes in Intermediate Flying Fish, *Cheilopogon intermedius* Parin, 1961, During Ambient and Ice Storage

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Abstract

Intermediate flying fish, *Cheilopogon intermedius*, Parin, 1961, is a popular source of inexpensive animal protein in the Philippines, but it remains susceptible to deterioration during processing. The study evaluated the sensory, microbiological, and physico-chemical characteristics of flying fish, *C. intermedius* stored at ambient conditions (27-29 °C) and on ice (1-2 °C) storage. The storage duration of flying fish was 8 h at ambient temperature and around 12 days in ice. A ten-member taste panel rejected raw fish due to strong fishy to sour odours and soft texture. Similarly, cooked samples were rejected based on sour and ammoniacal odours, mushy texture, and a bitter taste. Results of sensory evaluation correlated with those of microbial and chemical analyses. Bacteria multiplied rapidly at ambient temperature with values of 11.05 log CFU.g⁻¹ and 7.55 log CFU.g⁻¹ (*P* < 0.05) for total viable count (TVC), and H₂S-producers count, respectively, at the end of the 20-h storage. In comparison, bacterial counts remained below rejection limits until the end of the storage period for samples stored in ice. Total volatile base nitrogen (TVB-N) and trimethylamine nitrogen (TMA-N) accumulated rapidly at ambient temperature, reaching 86.86 mg.100 g⁻¹ and 35.65 mg.100 g⁻¹, respectively, at the end of the storage period. In ice, TVB-N, TMA-N, and histamine values were slowed down, reaching rejection limits only at the end of the storage period. Moreover, the K value increased linearly at ambient storage, while a gradual increase was observed for samples stored in ice. Therefore, without the application of adequate icing, flying fish would undergo rapid decomposition at ambient temperature.

Keywords: fish spoilage, shelf life, freshness indicators, storage conditions

Introduction

Intermediate flying fish, *Cheilopogon intermedius* Parin, 1961, are an important pelagic fish caught in many coastal regions globally and consumed as fresh, processed, or baitfish in tuna fisheries. It comprises the traditional fishery resources in various Caribbean, Southeast Asian, and Southern Pacific regions and countries (Huang and Ou, 2012). Although it is one of the dominant catches in some areas of the Philippines, there is no exact production data on flying fish because the fishing method employed is not as developed as tuna, scads or sardines (Simora et al., 2016). As intermediate flying fish are mostly caught in nearshore areas, the crafts engaged in this fishery function as dayboats and, hence, do not have facilities to preserve the catch. Due to low quality, the landed flying fish cannot reach the prime market or processed into value-added products. It is important to determine the postmortem biochemical and quality changes to evaluate the impacts of postcapture handling practices on the quality of flying fish.

Postmortem changes in fish can be assessed with sensory, physicochemical and microbiological methods, which can be the basis for establishing the shelf life. Quality deterioration in fish occurs quickly and subsequently influences the shelf life of fresh fish. The initial loss of freshness in fish can be attributed to enzymatic and chemical reactions, while microbial activity is responsible for the obvious signs of spoilage (Gram and Huss, 1996). Fish spoilage...
patterns have been extensively studied for many marine fish species (Ababouch et al., 1996; Ozogul et al., 2011; Sardar et al., 2015). Unfortunately, the data available for the postmortem changes affecting the quality of fresh flying fish are scant. Fish processors consider flying fish as a vulnerable raw material for processing. Their muscle contains a significant amount of histidine (473 mg/100 g^-1) (Kung et al., 2015), which can be converted to histamine through the action of bacteria and endogenous enzymes. Histamine is a foodborne chemical hazard that can cause susceptible individuals allergy-like symptoms (Maintz and Novac, 2007). The conversion of histidine to histamine can be further accelerated if the raw material is subjected to temperature abuse (Kung et al., 2015).

This study was carried out to investigate the sensory, microbiological and physicochemical changes in intermediate flying fish, *C. intermedius*, in ambient and ice storage. The intermediate flying fish, *C. intermedius*, is a prominent species caught in Philippine waters but is considered a low-value fish. The results of this study aim to help establish guidelines and criteria using simple and direct methods to determine the shelf life of fresh flying fish and improve the utilisation of this important fishery resource.

Materials and Methods

Ethical approval

No live animals were used in the conduct of the experiment. The samples and methods were conducted according to the Guidelines for the Use of Fishes in Research (American Fisheries Society, 2014) and were approved by the Office of the Vice Chancellor for Research and Extension (Project SP15-07) of the University of the Philippines Visayas (3/31/2017).

Fish storage conditions

Fresh flying fish, *C. intermedius*, (Fig. 1) was purchased from a fishing village market in San Jose, Antique, Philippines, within 4 h postcapture. Fish were packed in polystyrene boxes with enough flake ice and immediately transported to the laboratory within 1 h. The fish samples’ average length and body weight were 14.33 ± 6.26 cm and 60.02 ± 9.02 g, respectively. The average dressing percentage was 45.04 ± 1.86 %. Two storage conditions were carried out with 20 kg of each lot. The first lot was kept in a basket at an ambient temperature (27–29 °C). Samples were withdrawn every 2 h for sensory, microbiological, and physicochemical change analyses. The second lot was stored in an insulated ice box with ice to fish ratio of 1:1, and the temperature was maintained at 1–2 °C. The fish were re-iced every 2 days or as needed. Samples were withdrawn every 2 h for ambient storage and every 2 days for ice storage and subsequently analysed to evaluate for sensory, microbiological, and physicochemical changes evaluations.

Sensory evaluation

The sensory analysis was conducted by a panel of 10 semi-trained laboratory panellists according to the EEC freshness rating system (Huss, 1988) for raw fish and according to the method of Jorgensen et al. (1988) for cooked fish. The samples were blind-coded with 3-digit random numbers, and the sensory attributes examined for raw fish were rancid odours, general appearance of the mucus, skin, eyes, gills, odour of the gills, adherence of the scales and flesh condition using the 10-1 scoring scale. A high score (10-7) was given to fish devoid of off-odours or off-flavours, while those exhibiting flat and neutral odours or flavours received a score of 6. Any fish with off-odour or off-flavour were allocated a score below 6. Scores below 4 corresponded to unacceptable quality. For cooked fish, fillets were steamed for 20 min, and the eating quality (cooked flavour) was assessed using the same scoring scale as in raw fish, but with particular attention to the changes in characteristic seaweed odour/flavour to a flat, neutral odour/flavour, to the sensation of off-odour and off-flavour characteristic of spoiled fish.

Microbiological analysis

Twenty-five grams of fish meat was aseptically weighed and homogenised in 225 mL of 0.1 % peptone water diluent (pH 7 ± 0.1), Serial dilution was prepared. One mL of each decimal dilution was pipetted into sterile plates. Total viable count (TVC) expressed as colony-forming units per gram of fish muscle (CFU.g^-1) of the representative samples were determined by standard plate count methods on Nutrient Agar (Difco, USA), while a selective count of H2S-producing bacteria was determined on Iron Agar (Difco, USA) as described by Gram et al. (1987). All counts were performed in triplicate, and data were transformed into logarithms of the number of colony-forming units per gram of a sample (log CFU.g^-1).

Physicochemical analyses

pH was measured according to the method described by Woyewoda et al. (1988). Water holding capacity
(WHC) was determined according to the method described by Eide et al. (1982) with slight modifications. Briefly, 2 g of homogenised fish sample was weighed into a 50-mL conical tube inserted with Whatman No. 1 filter paper and immediately centrifuged at 750 \( \times g \) for 5 min. The liquid loss was expressed as the percentage of weight lost during centrifugation. The weight loss was divided by the water content of the sample and expressed as % WHC.

The total volatile base nitrogen (TVB-N) and trimethylamine nitrogen (TMA-N) of flying fish were measured by the Conway micro diffusion method (Woyewoda et al., 1986) and expressed as mg.100 g\(^{-1}\) of fish muscle.

Histamine content was analysed using the fluorometric method (AOAC, 2012) with modifications. Briefly, a 5-g sample was transferred into 50 mL polypropylene tubes and homogenised with 40 mL methanol for 1 min. The homogenates were placed in a water bath at 60 °C for 15 min, cooled at room temperature, and then transferred to volumetric flasks, and methanol was added to the final volume of 50 mL. The homogenates were filtered, and the extract was subjected to fluorometric (Trilogy® Turner, USA) reading. Histamine content was expressed as mg.100 g\(^{-1}\) of the sample.

**K value determination**

**K** value was determined according to the high-performance liquid chromatography (HPLC) procedure of Yokoyama et al. (1992) with some modifications. Five grams of fish muscle were homogenised for 1 min with 10 mL chilled 0.6 M perchloric acid at 4 °C. The obtained homogenate was centrifuged for 10 min with a revolving speed of 7188 \( \times g \) at 4 °C. A 5-μL filtrate obtained from the sample extracts was injected into the HPLC system (Shimadzu LC-10VP, Japan) equipped with a UV/VIS detector and a low gradient pump (Shimadzu LC-10ATVP, Japan) with a four-channel mixer (Shimadzu FVC-10ALVP, Japan) after filtration through a 0.45 μm filter membrane. The separation of the nucleotide products was obtained by a 5 μm C18 column (250 × 4.60 mm i.d.) equilibrated at 30 °C. The correlation coefficient of peak areas against nucleotide standard concentrations and coefficients of variation for each degradation compound were calculated after injecting several replicates of each nucleotide standard solution. The K value was then calculated using the equation below:

\[
K \text{ value} (\%) = \left( \frac{HxR + Hx}{ATP + ADP + AMP + IMP + HxR + Hx} \right) \times 100
\]

where ATP, ADP, AMP, IMP, HxR, Hx represent the concentration (μmoL.g\(^{-1}\)) of adenosine triphosphate (ATP), adenosine diphosphate (ADP), adenosine monophosphate (AMP), inosine monophosphate (IMP), inosine (Hx-R), and hypoxanthine (Hx).

**Statistical analysis**

The effect of storage time on various parameters was analysed using one-way analysis of variance (ANOVA). Given the various parameters, Tukey’s multiple comparisons test was used to determine the differences between treatment means and compare the two storage conditions, ambient and iced. Analyses were performed using R software (R Core Team, 2014), and figures were made in Microsoft Excel 2020. P values of 0.05 or less were considered statistically significant.

**Results**

**Changes in sensory qualities**

The overall sensory acceptability scores of the raw and cooked flying fish samples stored at ambient and ice storage are shown in Figure 2. As the storage time progressed, the sensory quality scores for samples stored in ambient and on ice declined. At ambient storage, the samples were found in acceptable conditions up to 10 h and 6 h for raw and cooked samples, respectively (Fig. 2A). On the other hand, fish samples stored in ice were found acceptable for up to 16 days and 8 days for raw and cooked samples, respectively (Fig. 2B). Rejection of raw fish by the taste panelists at the 12th hour for ambient storage and 20 th day of ice storage was mainly characterised by strong fishy to sour odours and soft texture. Cooked samples gave sour and ammoniacal to faecal odours with a mushy texture. The predominant flavour was bitter taste, observed on the 8th hour and 10th day for ambient and ice storage, respectively.

![Fig. 2. Changes in the general acceptability scores for raw and cooked samples of intertermEDIATE flying fish, Chelopogon intermedius at ambient (A) and ice (B) storage. Values represent the mean ± SD of three replicates.](image-url)
**Microbiological changes**

Changes in total viable counts (TVC) and hydrogen sulphide (H₂S)-producing bacterial counts were determined in flying fish samples stored in ambient and in ice (Fig. 3). The initial TVC and H₂S-producers count in samples stored at ambient temperature were 3.68 and 2.74 log CFU.g⁻¹, respectively (Fig. 3A). These counts significantly increased (P < 0.05) to 11.05 and 7.55 log CFU.g⁻¹ for TVC and H₂S-producers, respectively, at the end of the storage period (20 h). At this stage, it exceeded the limit value of 7.0 log CFU.g⁻¹ set by the International Commission for Microbiological Safety of Foods (ICMSF, 1986). For samples stored in ice, the values for TVC and H₂S-producers rose to still acceptable values of 6.55 and 5.45 log CFU.g⁻¹, respectively, at the end of the 20-day storage period from the initial count of 2.12 log CFU.g⁻¹ for both TVC and H₂S-producers (Fig. 3B). Overall, at the end of the storage period for ambient and iced conditions, the log counts of samples stored in ice for TVC and H₂S-producers were significantly lower (P < 0.05) than those obtained in ambient storage.

The TVB-N contents of the flying fish samples stored at ambient temperature showed a slow increase during the early storage stages. However, at the later stages of ambient storage, TVB-N levels increased significantly (P < 0.05) and reached a maximum of 86.86 mg.100 g⁻¹ at the end of the 20-h storage period. On the other hand, there was also a slow increase in the TVB-N levels of samples stored in ice until the 16th day of storage, but an abrupt increase was observed on the 20th day, reaching a maximum of 46.84 mg.100 g⁻¹.

TMA-N levels for samples stored at ambient temperature increased significantly (P < 0.05) from an initial value of 4.35 mg.100 g⁻¹ to a maximum value of 35.65 mg.100 g⁻¹ at the end of the 20-h storage period. The same trend was observed for TMA-N levels during storage in ice, but there was a slow increase. An abrupt increase (P < 0.05) was observed at the end of the storage period at 15.84 mg.100 g⁻¹ from the initial value of 4.35 mg.100 g⁻¹. The TVB-N and TMA-N values obtained for samples stored in ice were significantly lower (P < 0.05) than those stored at ambient temperature at the end of the storage period.

Low histamine levels were detected at ambient temperature until the 8th hour of storage. However, the results show that ambient-stored samples reached the defect action level of > 5 mg.100 g⁻¹ (FDA, 1995) on the 10th hour of storage and onwards, with histamine content ranging from 5.21 to 10.16 mg.100 g⁻¹. On the other hand, low histamine levels were observed in the ice-stored samples, reaching a maximum value of 6.05 mg.100 g⁻¹ at the end of the 20-day storage period. The histamine level obtained for samples stored in ice was significantly lower (P < 0.05) than those stored at ambient temperature at the end of the storage period.

**Changes in K value**

Variations in the K value of the flying fish samples during ambient and ice storage are shown in Figure 4, with the initial K value at 25.21 %. K value increased linearly with storage time in flying fish stored at ambient temperature, reaching 94.55 % at the end of the 20-h storage period. For ice-stored samples, the K value significantly increased (P < 0.05) at reasonably stable rates, reaching a maximum of 85.53 % after 16 days. According to Ehira and Uchiyama (1987), a K value of 60 % is the rejection level for most fish species. In this study, ambient-stored samples reached the rejection limit on the 8th hour of storage at 61.02 %, while ice-stored samples reached the rejection limit after 6 days of storage at 68.15 %. At the end of the storage period for both conditions, the

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**Table 1** shows the physicochemical changes in flying fish during ambient and ice storage. An initial pH value of 5.64 was observed at the beginning of the storage study. It significantly increased (P < 0.05), reaching pH 7.56 for ambient storage and pH 7.23 for ice storage, by the end of the storage period. On the other hand, the WHC of the flying fish muscle decreased significantly (P < 0.05) throughout the storage period for both ambient and ice storage conditions. Initially, the WHC of the fresh muscle was 38.03 %, but the value significantly (P < 0.05) decreased to 30.06 % at the end of the 20-h ambient storage. The percentage WHC obtained for samples stored in ice was 33.77 % at the end of the 20-day storage period, which was significantly higher (P < 0.05) than the WHC of those samples obtained at ambient storage.

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**Chemical changes**

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Good quality characteristics based on sensory attributes primarily influence consumer acceptability and preferences. The changes in the eating quality of flying fish were very distinct, from a characteristic seaweed odour and flavour to the sensation of off-odour or off-flavour characteristic of spoiled fish. The perceived changes in the eating quality of flying fish as storage time progressed were linear. Similar trends were reported for other fish species stored at ambient temperature or ice storage (Massa et al., 2005; Okoro et al., 2010; Ozogul et al., 2011). Based on the results of the sensory assessment, the maximum storage life of *C. intermedius* ranged from 6–8 h at ambient temperature and 10–12 days in ice. A similar pelagic fish, *Sardina pilchardus* (Walbaum, 1792), was reported to have a storage time of 21–27 h at ambient temperature and 8–11 days in ice based on sensory qualities (Ababouch et al., 1996). The shorter shelf life observed in the present study reflects the vulnerability of flying fish to spoilage if stored at

### Discussion

Based on our previous studies on *C. intermedius* it is a potential raw material for processing into value-added products because its low lipid content of 1.21 % makes it less prone to oxidation during storage. At the same time, it has a high protein content of 21.01 %, making it an inexpensive source of animal protein (Simora et al., 2019). Therefore, it is necessary to determine the kinetics of microbiological, biochemical and sensory changes in flying fish stored under different conditions to predict the shelf life for its maximum utilisation.

Good quality characteristics based on sensory changes in *pH*, water holding capacity (WHC), total volatile base nitrogen (TVB-N), trimethylamine nitrogen (TMA-N) and histamine of intermediate flying fish, *Cheilopogon intermedius* during ambient and ice storage.

<table>
<thead>
<tr>
<th>Storage condition</th>
<th>Storage time</th>
<th>pH</th>
<th>WHC (%)</th>
<th>TVB-N (mg.100 g⁻¹)</th>
<th>TMA-N (mg.100 g⁻¹)</th>
<th>Histamine (mg.100 g⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Ambient</strong></td>
<td>0 h</td>
<td>5.64 ± 0.05a</td>
<td>38.03 ± 2.35a</td>
<td>6.85 ± 1.12a</td>
<td>4.35 ± 1.02a</td>
<td>0.10 ± 0.01a</td>
</tr>
<tr>
<td></td>
<td>2 h</td>
<td>6.62 ± 0.01b</td>
<td>36.60 ± 3.64c</td>
<td>8.54 ± 0.58a</td>
<td>9.60 ± 1.00b</td>
<td>1.89 ± 0.21b</td>
</tr>
<tr>
<td></td>
<td>4 h</td>
<td>6.82 ± 0.02c</td>
<td>36.30 ± 2.25c</td>
<td>14.77 ± 1.73c</td>
<td>14.74 ± 1.26c</td>
<td>1.75 ± 0.15c</td>
</tr>
<tr>
<td></td>
<td>6 h</td>
<td>6.46 ± 0.11b</td>
<td>35.98 ± 2.59b</td>
<td>27.94 ± 1.40c</td>
<td>18.24 ± 1.35cd</td>
<td>2.43 ± 0.02b</td>
</tr>
<tr>
<td></td>
<td>8 h</td>
<td>6.82 ± 0.05b</td>
<td>35.97 ± 1.30c</td>
<td>48.58 ± 0.88a</td>
<td>14.89 ± 1.83c</td>
<td>3.10 ± 0.08b</td>
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<tr>
<td></td>
<td>10 h</td>
<td>6.65 ± 0.05b</td>
<td>35.90 ± 1.57c</td>
<td>46.58 ± 1.06d</td>
<td>17.80 ± 1.01cd</td>
<td>5.21 ± 0.01b</td>
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<tr>
<td></td>
<td>12 h</td>
<td>6.76 ± 0.10b</td>
<td>33.91 ± 0.72b</td>
<td>50.36 ± 3.93a</td>
<td>21.39 ± 0.40ab</td>
<td>6.01 ± 0.12c</td>
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<tr>
<td></td>
<td>16 h</td>
<td>7.27 ± 0.02c</td>
<td>32.57 ± 0.16a</td>
<td>52.11 ± 0.09a</td>
<td>25.42 ± 3.43a</td>
<td>8.57 ± 0.09a</td>
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<tr>
<td></td>
<td>20 h</td>
<td>7.56 ± 0.02c</td>
<td>30.06 ± 0.79a</td>
<td>86.86 ± 0.81a</td>
<td>35.65 ± 0.81a</td>
<td>10.18 ± 0.45a</td>
</tr>
<tr>
<td><strong>Ice</strong></td>
<td>0 d</td>
<td>6.64 ± 0.05a</td>
<td>38.03 ± 2.35a</td>
<td>6.85 ± 1.12a</td>
<td>4.35 ± 1.02a</td>
<td>0.10 ± 0.03a</td>
</tr>
<tr>
<td></td>
<td>2 d</td>
<td>6.18 ± 0.03a</td>
<td>37.03 ± 1.04a</td>
<td>12.58 ± 1.59a</td>
<td>4.77 ± 0.99a</td>
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<td>4 d</td>
<td>6.39 ± 0.03a</td>
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<td>35.98 ± 2.59b</td>
<td>13.27 ± 0.40a</td>
<td>5.44 ± 1.75a</td>
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<tr>
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<td>35.97 ± 2.36b</td>
<td>8.42 ± 2.80a</td>
<td>7.25 ± 0.67a</td>
<td>1.81 ± 0.69a</td>
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<tr>
<td></td>
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<td>34.36 ± 0.72b</td>
<td>14.73 ± 0.62c</td>
<td>7.83 ± 0.37a</td>
<td>2.62 ± 0.45a</td>
</tr>
<tr>
<td></td>
<td>12 d</td>
<td>7.10 ± 0.03c</td>
<td>35.48 ± 2.16c</td>
<td>15.66 ± 0.03a</td>
<td>8.18 ± 0.01a</td>
<td>3.06 ± 0.15a</td>
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<tr>
<td></td>
<td>16 d</td>
<td>7.22 ± 0.05c</td>
<td>33.95 ± 0.97a</td>
<td>16.97 ± 1.80a</td>
<td>8.66 ± 0.24a</td>
<td>3.04 ± 0.05a</td>
</tr>
<tr>
<td></td>
<td>20 d</td>
<td>7.23 ± 0.02c</td>
<td>33.77 ± 0.80c</td>
<td>46.84 ± 1.83d</td>
<td>15.84 ± 0.89b</td>
<td>6.05 ± 0.23a</td>
</tr>
</tbody>
</table>

The values represent the mean ± SD of three replicates. Different letters in the same column are statistically significant (P < 0.05).

**Fig. 4.** Changes in the K value of intermediate flying fish, *Cheilopogon intermedius* at ambient and ice storage. Values represent the mean ± SD of three replicates.
ambient temperature for prolonged periods. This further indicates that exercised care to keep the fish temperature to around 0 °C or proper icing should be imposed during postharvest.

Findings in the sensory assessment correlated well with microbial analysis. Results of microbial analysis of flying fish samples stored at ambient temperature revealed that it retained the freshness of raw fish only for 8 h when the TVC was 5.67 log CFU.g⁻¹, and by the 10th hour, the value increased to 7.05 log CFU.g⁻¹ which is the rejection limit set by ICMSF (1986) for human consumption. On the other hand, a long lag phase was observed for samples stored in ice, representing those bacteria adapting to the cold environment before multiplication. Until the end of the storage period, the TVC and H₂S-producers count was still below the rejection limit. The same findings were also observed in other fish species that were stored in ice (Adoga et al., 2010; Ozogul et al., 2011). Moreover, the H₂S-producers count was generally lower than the total viable counts throughout the storage period. H₂S-producing bacteria of the genus Shewanella, such as S. putrefaciens and S. baltica, are commonly isolated in marine fish and cause spoilage in both fresh and packed fish, producing off-odours even at low cell numbers (Dalgaard, 1995). The count of H₂S-producers can specifically indicate spoilage, causing an increase in bacterial load (Rao and Khasim, 2009). As observed in this study, the sensory scores for cooked fish samples stored at ambient temperature dropped to unacceptable levels at the 8th hour, coinciding with the exponential increase of H₂S-producer counts starting at the 8th hour of the storage period. This indicates that H₂S-producing bacteria can be a suitable indicator to detect spoilage in fresh flying fish.

A gradual increase in pH was observed for samples in ambient and ice storage, which may be attributed to the production of volatile basic compounds such as ammonia and trimethylamine by specific spoilage organisms (SSO) present in fish (Tsogas et al., 2019). Changes in pH at postmortem may vary from pH 6.0 to 7.1, depending on the season, species and other factors (Duarte et al., 2020). Similarly, a decrease in WHC throughout the storage period can be caused by the water-protein interactions in fresh raw fish muscle, which are partly replaced by protein-protein interactions during storage. The muscle cell shrinks as spoilage progresses, causing the liquid to leak out of the cells to the intercellular space of the fish meat, thereby reducing the water-holding capacity of the fish muscle (Løje et al., 2017).

An increase in TVB-N levels beyond the maximum permissible level of 35 mg.100 g⁻¹ (European Commission, 1995) towards the end of ambient and ice storage may be attributed to bacterial spoilage after the bacterial population has grown since TVB-N is produced mainly by bacterial decomposition of fish flesh (Bahmani et al., 2011). TVB-N is normally low during the edible storage period, and increasing levels were found in fish at near rejection levels (Adoga et al., 2010). In this study, TVB-N might be considered a reliable indicator of flying fish freshness at ambient temperature. However, its reliability diminishes in ice storage, because only the results of samples stored at ambient-stored align with sensory and microbial assessments, indicating an acceptability window of approximately 6 hours at ambient temperature. Moreover, total volatile bases (TVB) during storage are mostly due to the production of trimethylamine (TMA) (Bekhit et al., 2021). TMA-N particularly contributes to the characteristic ammonical off-odour and “fishy” off-flavour in spoiled fish (Gram and Huss, 1996). A good quality fish contains <1.5 mg.100 g⁻¹TMA-N, and the limit of acceptability is 10–15 mg.100 g⁻¹ (FAO, 1995). Results of TMA-N for ambient-stored samples coincided with the rejection levels for sensory and microbial analysis. TVB-N and TMA have been found to parallel the increase in the total viable count during fish storage (Bekhit et al., 2021), and this was observed in this study and others (Aubourg et al., 2005; Ozogul et al., 2011). For ice-stored samples, TVB-N and TMA-N reached rejection levels on the 20th day. This also seems to corroborate the sensory and microbial data in which the flying fish samples were in acceptable condition for 16 days in ice.

The rate of histamine development in fresh flying fish was effectively decelerated by icing, while a faster rate of histamine accumulation was observed in ambient-stored samples. This showed the effectiveness of icing in the growth inhibition of spoilage bacteria capable of producing histamine. Histamine values of >20 mg.100 g⁻¹ are indicative of a decomposed sample, while a US Food and Drug Administration (USFDA) recommended level of >5 mg.100 g⁻¹ indicates decomposition for scombroid fish and products, hence, unsuitability for human consumption (FDA, 1995). The data show that toxic or unacceptable histamine levels can develop at ambient temperature (10 h), correlating with the sensory assessment and total viable count. This indicates the high free histidine inherently present in flying fish muscle and the presence of highly prolific bacteria capable of histidine decarboxylation (Lehane and Olley, 2000). A group of bacteria with important histidine decarboxylase activity, especially at ambient temperature, has also been confirmed in sardines (S. pilchardus) (Ababouch et al., 1991).

K value is widely utilised as an index of freshness before bacterial spoilage begins in the fish muscle (Huss, 1988). During postmortem, nucleotides and their related compounds degrade in a series of stages due to endogenous biochemical changes in the fish muscle (Brenner et al., 1988). In this study, K value increased linearly at ambient storage, while a gradual increase was observed in samples stored in ice. The results correlated well with sensory quality when panellists judged the ambient-stored samples as unacceptable at the 8th hour. The K value also reached...
the rejection limit of >60 %. A similar trend was observed in mullet Liza corsula (Hamilton, 1822) and pearlspot Etroplus suratensis (Bloch, 1790), where the K value increased linearly with storage time (Lakshmanan et al., 1996). However, the K value reached the rejection limit after 6 days for ice-stored samples. This result did not correspond with the sensory, microbial and chemical data in which the unacceptable levels were detected from 12–16 days in ice storage. This may be due to the high initial level of K value detected in the samples (25,21 %). This initial K value is comparable to that obtained in Nile perch Lates niloticus (Linnaeus, 1758), with an initial K value of 25 % (Williams et al., 1993). In general, K values of 20 % represent an excellent degree of freshness suitable for consuming raw fish at sashimi grade (Tejada, 2009). Variation of initial nucleotide contents is associated with differences among species, season, catching gear, and stress during fish death, among other factors (Huss, 1995). Following the high initial nucleotide content in the flying fish muscle, rapid nucleotide degradation resulting in the initial nucleotide content in the flying fish muscle, and other factors. Huss, 1995). Following the high initial nucleotide content in the flying fish muscle, rapid nucleotide degradation resulting in the formation of products associated with spoilage, such as hypoxanthine and inosine, must have occurred during ice storage. Ababouch et al. (2010) also found unstable K values for amberjack Seriola dumerili (Risso, 1810) and red sea bream Pagoz major (Temminck and Schlegel, 1843) stored in ice. Thus, the K value provided a useful indicator for freshness in flying fish stored at ambient temperature but not necessarily at iced storage.

Existing preservation methods for flying fish are limited to salting and drying. Adequate postharvest technologies for effective utilisation of this important fishery resource to increase its market value are essential. Considering the short shelf life at ambient temperature, C. intermedius can be processed as a marinated product, where the combined effect of acid, spices and salt can retard the action of spoilage bacteria and enzymes. Flying fish can also be utilised in fish mince or in surimi to produce value-added products such as fish nuggets, fish balls, fish cakes, imitation crab sticks, and many more.

Conclusion

The present study provides data on the sensory, microbiological, and physico-chemical changes during the storage of flying fish at ambient temperature and in ice. Results revealed that C. intermedius was found in edible condition for 8 h at ambient temperature and around 12 days in ice. Sensory and microbial data supported the results of chemical analyses such as TVB-N, TMA-N and histamine, with a drop in the sensory scores. K value provided a useful indicator for freshness in flying fish stored at ambient temperature but not in ice, which may be attributed to the high initial nucleotide content of the flying fish muscle. Overall, the spoilage pattern of flying fish stored at ambient and in ice can be used to assess the effects of postharvest handling, especially the delays in fish icing.

Acknowledgements

This research was supported by grants from the Philippine Commission on Higher Education – National Agriculture and Fisheries Education System and from the Office of the Vice Chancellor for Research and Extension (Project SP15-07). University of the Philippines Visayas. The authors thank the Bureau of Fisheries and Aquatic Resources Region 6 for the assistance in the K value analysis.

Conflict of interest: The authors declare that they have no conflict of interest.

Author contributions: Rhoda Mae C. Simora: Concept, experimental design, data analysis, drafting and revising of manuscript. Cindy D. Armada: Experimentation, data analysis, revising of manuscript. Karmelie Jane M. Monaya: Experimentation, revising of manuscript.

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