Effect of Slaughter Method on Postmortem Changes of Grass Carp (Ctenopharyngodon idella) Stored in Ice

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ABSTRACT: The effect of 2 slaughter methods (immersion in ice-water slurry and electrical stunning followed by ice slurry asphyxiation) on the quality of grass carp (Ctenopharyngodon idella) stored in ice for 20 d was evaluated using sensory and chemical analysis. Electricity immediately stunned the fish and did not induce blood spots in the flesh. Fish killed by electricity showed a faster initial rate of ATP degradation and entered into rigor mortis earlier, but did not show significant differences in the sensory score when compared with fish killed by immersion in ice-water slurry. Thus, no differences were observed in the shelf life of carps between the 2 slaughter methods evaluated. The limit for acceptable quality of grass carp stored in ice was around 13 to 16 d. Grass carp accumulated more inosine than hypoxanthine, K, Ki, P, Fr, and H values were highly correlated with storage time and with the TFRU sensory scores in both groups; these could be used to assess the freshness quality of grass carp.

Keywords: Ctenopharyngodon idella, grass carp, fish slaughtering, K-value, sensory quality

Introduction

The production of freshwater fish has increased in the last few years in Brazil (Rangel 1995; IBAMA 2005). One of the main species farmed in southern Brazil is grass carp (Ctenopharyngodon idella). The intensive production of this fish has raised concerns over the quality of its products. Therefore, investigations on the freshness quality during handling, distribution, and storage in ice are of considerable interest (Scherer and others 2004).

Rigor mortis is one of the first postmortem changes. It has a major influence on the appearance and structure of fish muscle. When fish is killed, muscles are wholly relaxed, but as ATP (adenosine 5'-monophosphate) content decreases below a critical level, actin and myosin make an irreversible bond (acto-myosin) and muscle enters rigor mortis (Iwamoto and others 1987). After some time, fish muscle enters into a process of tenderization, possibly related to the degradation of connective tissue (Ando and others 1993) and breakage of Z-discs of myofibrils (Seki and Tsuchiya 1991) and acto-myosin junctions (Yamanoue and Takahashi 1988).

The pathway of ATP degradation in fish muscle has been extensively documented as the following sequence: ATP→adenosine 5'-diphosphate (ADP)→adenosine 5'-monophosphate (AMP)→inosine 5’monophosphate (IMP)→inosine (HxR)→hypoxanthine (Hx; Özogul and others 2000a). Various ratios of the concentration of ATP and its breakdown products, expressed as the K, Ki, G, P, H, and Fr values, are reliable chemical indicators of freshness quality in a variety of fish (Burns and others 1985; Özogul and others 2000b; Alasalvar and others 2001, 2002). Nevertheless, the pattern and rate of nucleotide degradation differs among fish species and body location and is affected by predeath handling, season, and storage conditions (Chiba and others 1991; Eriksson and others 1997). Hence, a ratio that is suitable for evaluating the freshness of 1 species may be not useful for another. G and P values are most useful with lean fish, because in fatty fish, rancidity may render the product undesirable before meaningful G and P values are attained (Shahidi and others 1994). Fr value may not be applicable to some species in which IMP levels drop rapidly in the early stage of ice storage (Greene and Bernatt-Byrne 1990). When ATP degrades to IMP soon after death, the Ki value that does not involve determination of ATP, ADP, or AMP may be used. However, in cases where ATP, ADP, and AMP remain detectable for even 2 wk (Karube and others 1984), the K value may be superior to the other values. For grass carp, only 1 report on the K value was found (Ehira and Uchiyama 1973).

Choosing the most suitable slaughter method is an important step for ensuring a good quality of fish products. A stressful slaughter may induce an earlier resolution of rigor mortis, a softer texture, gaping, and loss of shelf life (Lowe and others 1993; Robb and Kestin 2002). Stress and pain during handling and slaughtering is also a concern from an animal welfare point of view (Robb and Kestin 2002). A possible way of reducing premortem stress is the use of electrical stunning prior to slaughter (Marx and others 1997; Roth and others 2002; Lambooj and others 2004). Although the occurrence of broken vertebrae and hemorrhages (blood spots) has been reported after electrical stunning (Sharber and others 1994; Ainslie and others 1998; Roth and others 2003), it has been proposed that, with some improvements, electrical stunning could meet the requirements of meat quality and animal welfare (Marx and others 1997; Robb and Kestin 2002; Roth and others 2002; Lines and others 2003; van de Vis and others 2003). Most studies on electrical stunning employ an alternating current, which, however, is considered more injurious than direct current (Dolan and Miranda 2002;...
Effect of slaughtering on fish quality . . .

Lines and others (2003). At present, most farmed grass carp in southern Brazil are slaughtered either by immersion in an ice-water slurry or asphyxiation in ice. These methods do not induce immediate loss of brain function and, therefore, are not considered humane (Robb and Kestin 2002; van de Vis and others 2003).

The present study was aimed at investigating the effect of different slaughter methods (immersion in ice-water slurry and electrical stunning followed by ice slurry asphyxiation) on postmortem changes in grass carp stored in ice and to evaluate the most reliable freshness quality indicators for grass carp, using K and related values.

Materials and Methods

Fish handling

Grass carps (Ctenopharyngodon idella) (average weight and length: 822 ± 147 g and 40.8 ± 2.8 cm, respectively) were raised at the Dept. of Zootecnia of Federal Univ. of Santa Maria in an earth pond (800 m²) at a density of approximately 1 fish/3.2 m². During the growth period (July 2002 to April 2003), chemical and physical quality of water was evaluated every 2nd day, using kit tests (Alfa Tecnocémica, Florianópolis, S.C., Brazil). Water alkalinity, pH, nitrite, ammonia, and transparency values remained almost constant (57.6 ± 2.6 mg CaCO₃/L, 7.5 ± 0.1, 0.06 ± 0.01 ppm, 0.49 ± 0.02 ppm, and 41.7 ± 7.6 cm, respectively) and within the range required for this species (Chen and others 1993). Dissolved oxygen content and temperature ranged between 4.3 and 9.2 mg/L and 15 to 29 °C, respectively. Fish were fed twice a day (in the morning and late in the afternoon) at a ratio of 5% of total biomass. The diet consisted of green fodder (Penisetum americanum and Pennisetum purpureum) and a commercial feed (Purina®, Paulínia, SP, Brazil) yielding 36% protein in the 1st month, 32% protein for the next 3 months, and then 28% protein up to the harvest. After catching, fish were fasted for 24 h before killing. Fish were randomly selected for one of the slaughter methods: immersion in ice-water slurry or electrical stunning followed by ice slurry asphyxiation. For ice-water slaughtering, fish were dipped in a polyethylene plastic tank containing 2 g NaCl/L to a depth of 50 cm. Two plate copper electrodes (40.0 × 2.5 × 1.2 cm) were vertically positioned in the tank of fish) of 2.0 m long, 1.0 m wide, 1.5 m deep) filled with fresh water (7.5 L/kg ice-water slurry or electrical stunning followed by ice slurry asphyxiation) on postmortem changes in grass carp stored in ice and to evaluate the most reliable freshness quality indicators for grass carp, using K and related values.

Analysis of ATP and its breakdown products

Four fish from each slaughter method were used in these assays. ATP and its breakdown products were extracted according to Ryder (1985). Briefly, 5 g of muscle from the anterior dorsal region of the fish were homogenized with 25 mL of 0.6 M perchloric acid at 0 °C for 1 min with an Ultra-turrax homogenizer (model T 18, IKA® Works Inc., Wilkinson, Del., U.S.A.). The homogenate was centrifuged at 2000 × g for 10 min, and 10 mL of the supernatant was immediately neutralized to pH 6.5 to 6.8 with 1 M potassium hydroxide. Potassium perchlorate was removed by filtration through a Millipore (0.22 µm) syringe filter and filtrate was stored at −20 °C for subsequent analysis.

A liquid chromatograph (Knauer, Berlin, Germany) equipped with a model K 1001 pump, a model K 5004 online degasser, a model K 2501 UV detector (set at 254 nm) and operated by Eurochrom 2000 software (Knauer, Berlin, Germany) was used for all analyses. Separation was achieved in a Europhase 5 µm 100 RP C18 column (250 × 4 mm of internal diameter) with a precolumn, using 0.04 M potassium dihydrogen orthophosphate (KH₂PO₄) and 0.06 M dipotassium hydrogen orthophosphate (K₂HPO₄) (pH 7.0) as mobile phase A and acetonitrile as mobile phase B. Buffer solutions were filtered through a 0.45-µm Millipore filter before use. Mobile phase was eluted at a flow rate of 0.8 mL/min following the gradient proposed by Vallé and others (1998), modified to increase peak resolution. A standard curve was prepared for each compound evaluated (ATP, ADP, AMP, IMP, HxR, Hx; ICN Biomedicals Inc., Aurora, Ohio, U.S.A.) in the range of 0.01 to 0.4 mg/mL. Samples were filtered through a 0.22-µm Millipore filter before injection, because additional potassium perchlorate precipitates during storage at −20 °C. All reagents used were of HPLC grade.

The K, G, P, H, and Fr values were calculated according to Saito and others (1959), Karube and others (1984), Burns and others (1985), Gill and others (1987), Shahidi and others (1994), and Luong and others (1992), respectively:

\[ K(\%) = \frac{(HxR + Hx)}{(ATP + ADP + AMP + IMP + HxR + Hx)} \times 100 \]
\[ Kl(\%) = \frac{(HxR + Hx)}{(IMP + HxR + Hx)} \times 100 \]
\[ G(\%) = \frac{(HxR + Hx)}{(AMP + HxR + Hx)} \times 100 \]
\[ P(\%) = \frac{(HxR + Hx)}{(AMP + IMP + HxR + Hx)} \times 100 \]
\[ H(\%) = \frac{(Hx)}{(IMP + HxR + Hx)} \times 100 \]
\[ Fr(\%) = \frac{(IMP + HxR + Hx)}{(IMP + HxR + Hx)} \times 100 \]

Sensory assessment

The sensory assessment of grass carp was conducted using the Tasmanian Food Research Unit (TFRU) freshness assessment scheme, a system developed at CSIRO Div. of Food Research (Branch and Vail 1985), with minor modifications for grass carp (Table 1). At each day of analysis, 1 fish from each slaughter method was assessed by an expert panel of 7 to 14 panelists. Each panelist was given up to 4 simple descriptors, scoring demerit points from 0 to 3 where 0 indicated best quality and any higher score indicated poorer quality. The scores for the separate characteristics were summed to give an overall sensory score. Panelists were also asked to state whether the fish were acceptable or unacceptable for consumption. This judgment was used to determine fish shelf life.

Color analysis

Fish (five specimens from each slaughter method) were filleted (93 h post mortem), and the flesh color was assessed using a CR-300 Chromometer (Minolta, Osaka, Japan), according to the Intl. Commission on Illumination (CIE 1976 L° a° b°), using a standard illuminant D65, with a 10° supplementary standard observer and a standard calibration plate (number 15233011). Color of fillets was measured at 3 positions above the lateral line: close to the head (anterior), in the middle of the fillet (middle), and in the tail area (pos-
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Table 1—Modified Tasmanian Food Research Unit (TFRU) sensory assessment scheme for grass carp

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Demerit points*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
</tr>
<tr>
<td><strong>General</strong></td>
<td></td>
</tr>
<tr>
<td>Appearance</td>
<td>Very bright</td>
</tr>
<tr>
<td>Firmness</td>
<td>Firm</td>
</tr>
<tr>
<td>Slime</td>
<td>Absent</td>
</tr>
<tr>
<td>Stiffness</td>
<td>Preigor</td>
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<tr>
<td>Odor</td>
<td>Fresh</td>
</tr>
<tr>
<td><strong>Eyes</strong></td>
<td></td>
</tr>
<tr>
<td>Clarity</td>
<td>Clear</td>
</tr>
<tr>
<td>Shape</td>
<td>Convex</td>
</tr>
<tr>
<td>Localization in the eye socket</td>
<td>Prominent</td>
</tr>
<tr>
<td>Iris</td>
<td>Visible</td>
</tr>
<tr>
<td><strong>Gills</strong></td>
<td></td>
</tr>
<tr>
<td>Color</td>
<td>Dark red</td>
</tr>
<tr>
<td>Mucus</td>
<td>Absent</td>
</tr>
<tr>
<td>Odor</td>
<td>Fresh</td>
</tr>
<tr>
<td><strong>Belly</strong></td>
<td></td>
</tr>
<tr>
<td>Firmness</td>
<td>Firm and elastic</td>
</tr>
<tr>
<td>Vent Condition</td>
<td>Normal</td>
</tr>
<tr>
<td>Odor</td>
<td>Fresh</td>
</tr>
</tbody>
</table>

*Total demerit points (0 to 40).

Statistical analysis

The effect of the slaughter method on the rigor index was analyzed by analysis of variance (ANOVA) with time considered as repeated measure. The effects on the color of fish fillets (2 slaughter methods versus 3 positions) and the effects on the sensory score, nucleotide concentration, and on K and related values (2 slaughter methods versus storage time) were analyzed by two-way ANOVA. Post-hoc analysis was carried out using Duncan’s test. The correlations between sensory score or K and related values with the time of storage and sensory scores with K and related values were evaluated by a simple linear correlation. Differences were considered to be significant when \( P < 0.05 \). Data were analyzed using the Statistica 6.0 software package.

Results and Discussion

Welfare implications

Fish killed by immersion in ice-water slurry showed strong averse movements in the 1st min, which may indicate stress and pain. Movements finished after approximately 10 min, due to cold paral- ysis, but fish remained immersed for 20 min until dead. Few studies evaluating the welfare of fish killed by ice were found in the literat- ure. The loss of sensibility resulting from hypothermia is said to be painful in humans. Conversely, it has been proposed that cooling fish in ice slurry could be painful, because immersion in iced water has been used as a torture method in humans (Robb and Kestin 2002). Despite this controversy, slaughtering by immersion in ice-water slurry is considered to have a highly negative impact on animal welfare (Robb and Kestin 2002). Gilt-head seabream take about 5 min to lose brain function in ice slurry, while rainbow trout take about 10 min (Robb and Kestin 2002; van de Vis and others 2003). One of the reasons cited by producers for using this method is that rapid chilling promotes flesh quality by reducing both autolytic degradation and muscular activity immediately before death (Robb and Kestin 2002). However, Mochizuki and Sato (1994) reported that lactic acid concentrations in fish killed in ice slurry were higher than in fish killed by severing the spinal cord, indicat- ing greater muscular activity pre-mortem.

In contrast to the slaughtering by immersion in ice-water slurry, grass carps slaughtered by electricity ceased movements immediately after starting electrical treatment. Hence, from a welfare point of view, electricity seems to be more suitable to stun/kill grass carp when compared with immersion in ice-water slurry, due to the im- mediate stunning that probably reduces stress and pain. These results are in agreement with those of other studies that consider electrical stunning as having a very low impact on animal welfare (Robb and Kestin 2002).

Rigor index and ATP degradation

Fish slaughtered by electricity entered earlier into rigor mortis \( (P < 0.05) \). Onset of rigor occurred within 8 and 15 h of death for elec- tricity and ice-slaughtered fish, respectively (Figure 1). We found no studies comparing immersion in ice-water slurry and electrical slaughtering, but some authors observed that fish killed by electric- ity entered earlier into rigor mortis than fish killed by percussion (Marx and others 1997; Morzel and others 2003). However, others found no difference in rigor development between fish stunned by electricity or concussion and earlier rigor development for CO₂- stunned fish (Marx and others 1997; Roth and others 2002).

Two hours after death, ATP concentration was higher in fish killed by immersion in ice-water slurry than in fish killed by electricity \( (P < 0.05) \); Figure 2). Accordingly, at this time, IMP concentration was high-
er in fish killed by electricity than in those killed by immersion in ice-water slurry \((P < 0.05; \text{Figure 2})\). The faster depletion of ATP may have contributed to the earlier onset of rigor in fish killed by electricity. Chiba and others (1991) observed that the tetanus during exposure to electricity depletes creatine phosphate and ATP.

In the study of Abe and Okuma (1991), carps achieved full rigor mortis after 24 h on ice. Maximum muscle contraction \((I_R = 100\%)\) was observed 20 h after death in fish killed by electricity, whereas in fish killed by immersion in ice-water slurry, maximum muscle contraction \((I_R = 92\% \text{ to } 97\%)\) was observed 24 to 48 h after death (Figure 1).

Wang and others (1998) reported that muscle enters a full rigor mortis as ATP decreases to about 1 \(\mu\text{mol/g}\). Twenty-four hours after death ATP concentration in fish muscle was around 0.1 \(\mu\text{mol/g}\), regardless of the slaughter method (Figure 2). For both groups, rigor mortis was not fully resolved even after 93 h post mortem (Figure 1).

No significant differences were observed in ADP and AMP concentrations between the slaughter methods evaluated (Figure 2). AMP concentration remained almost constant during storage both in fish slaughtered by electricity \((0.21 \pm 0.01 \mu\text{mol/g})\) and in ice-slaughtered fish \((0.20 \pm 0.03 \mu\text{mol/g})\). ADP concentration decreased in the 1st 24 h in fish slaughtered by electricity \((\text{decrease from } 0.75 \pm 0.11 \text{ to } 0.16 \pm 0.01 \mu\text{mol/g}; P < 0.05)\) and in ice-slaughtered fish \((\text{decrease from } 0.86 \pm 0.14 \text{ to } 0.18 \pm 0.01 \mu\text{mol/g}; P < 0.05)\) and then remained almost constant until the end of storage. IMP concentration increased \((P < 0.05)\) in the 1st 24 h in both groups, due to rapid ATP degradation. IMP concentration declined from the 4th day of storage onwards in fish slaughtered by electricity, while in ice-slaughtered fish IMP decline started on the 7th day \((P < 0.05; \text{Figure 2})\). Several researchers have reported that the conversion of ATP into IMP is usually completed within 1 day (Alasalvar and others 2001; Huidobro and others 2001; Mendes and others 2001; Lougovois and others 2003) and is presumed to be totally autolytic. On day 0, fish killed by electricity had higher IMP values than those killed on ice \((P < 0.05)\), due to the faster ATP degradation. Inosine \((\text{HxR})\) concentration increased \((P < 0.05)\) until the 7th day of storage in both groups \((P < 0.05)\). From the 4th day of storage onwards, fish killed by electricity showed lower \(\text{HxR}\) and higher \(\text{Hx}\) concentration than fish killed by immersion in ice-water slurry \((P < 0.05; \text{Figure 2})\).

The rate of nucleotide degradation differs among species (Martin and others 1978; Hattula and others 1993; Mendes and others 2001) and is dependent on factors such as predeath conditions, stress during capture, slaughter method, season, and storage conditions (Chiba and others 1991; Lowe and others 1993; Erikson and others 1997; Grigorakis and others 2003). Grass carp seems to accumulate \(\text{HxR}\), but exhibits small quantities of \(\text{Hx}\) (Figure 2). Thus the use of \(\text{Hx}\) to evaluate the freshness of grass carp is limited.

**Sensory assessment**

Changes in the TFRU sensory score of grass carp over the 20 d of storage in ice are shown in Figure 3. The sensory score increased linearly for fish killed by immersion in ice-water slurry and by electricity \((r = 0.98\) and \(r = 0.95\), respectively, \(P < 0.05)\) during storage (Figure 3).

There was no significant difference in the sensory score between groups. Panelists indicated that fish were unacceptable at 16 d. Hence, shelf life of grass carp stored in ice estimated by sensory evaluation was around 13 to 16 d. That is in good agreement with results based on microbiological criteria (Scherer 2004; Scherer and others 2004). At the limit of acceptability, the sensory scores of grass carp ranged from 23 to 25, for both groups (Figure 3). Alasalvar and others (2001) found a score around 20 when *Sparus aurata* was considered unacceptable \((17-18 \text{ d of storage in ice})\) and a score around 20-22 when *Dicentrarchus labrax* was considered unacceptable \((16 \text{ to } 18 \text{ d of storage in ice}; \text{Alasalvar and others 2002})\).

Demerit points of all parameters evaluated in the TFRU sensory assessment scheme changed during storage \((P < 0.05, \text{data not shown})\). Some parameters, such as gills (odor, mucus, and color), belly firmness, and eye shape were found to be good indicators of freshness for grass carp, because they showed the greatest changes in demerit points during storage. However, other parameters such...
as general appearance, slime, and blood in the eyes showed only slight changes during the storage.

According to the sensory evaluation, grass carp freshness was indicated by dark red gills, fresh fish odor, little fluid mucus, convex and clear eyes, and firm belly, whereas poor quality was indicated by brown gills, cloudy and sunken eyes, and soft belly.

**Color**

Although electrical stunning has been considered more ethical than other stunning methods, exposure to electricity may lead to broken vertebrae and hemorrhages (blood spots), reducing the commercial value of fish. Roth and others (2003) observed that these consequences depend on the electric field strength and current duration. Hemorrhages may cause fish killed by electricity to have redder and darker flesh (higher $a^*$ and lower $L^*$ values, respectively) than fish killed by percussion (Morzel and others 2003). Occurrence of broken vertebrae was not evaluated in the present study. The use of electricity did not induce blood spots in flesh or in the skin, as observed by visual inspection (data not shown). Accordingly, color analysis did not show significant differences in $a^*$ and $L^*$ values between the slaughtering methods evaluated (Table 2). Similarly, Bogess and others (1973) also found no differences in $L^*$ values (Hunter Lab color space) between fish killed in ice slurry or by electricity (direct current). However, ANOVA revealed that fish killed by electricity showed a significantly higher $b^*$ value than those killed in ice (Table 2). Post hoc analysis revealed that only the $b^*$ value of the middle position of ice-slaughtered fish was significantly different from the anterior position of fish killed by electricity. Changes in muscle color resulting in the yellowing of flesh have been observed in red meat animals subjected to high levels of activity prior to slaughter (Warriss 1996). These changes are caused by the rapid drop in pH postmortem, leading to protein denaturation and loss of water, resulting in changes in the reflection of light. However, this hypothesis seems unlikely to explain differences in $b^*$ value observed in the present study, because no significant difference was observed in the pH value of fish slaughtered by ice when compared with those slaughtered by electricity either immediately after death (6.63 ± 0.06 against 6.57 ± 0.06) or when color was evaluated (93 h after death; 6.80 ± 0.04 against 6.82 ± 0.03). Because color measurements were performed 93 h after death, we cannot rule out that the observed differences in $b^*$ value could also be a result of time (approximately 4 d of storage) and not only the slaughter method.

**K and related values**

There is limited information on the behavior of the commonly used indicators of fish freshness, such as $K$, $Ki$, $G$, $P$, $H$, and Fr values for grass carp (Ehira and Uchiyama 1973). Figure 4 displays changes of these values in grass carpets slaughtered by ice-water slurry and electricity over 20 d of storage.

$K$, $Ki$, $P$, and $H$ values showed a linear increase during storage ($r = 0.96$), while $G$ increased only in the 1st 4 d ($r = 0.67$), and Fr linearly decreased ($r = 0.97$) during the storage. No significant differences were observed in $K$ and related values between the 2 slaughter methods evaluated. Similarly, no significant differences were observed in the $K$ value of gilt-head seabream ($Sparus aurata$) killed by asphyxiation, percussion, or immersion in ice-water slurry (Huidobro and others 2001). In contrast, rainbow trout killed in ice slurry had higher $K$ value than those killed by percussion (Özogul and Özogul 2004). The modified TFRU sensory scores for grass carp had high correlations with $K$ ($r = 0.98$), $Ki$ and $P$ ($r = 0.97$), $Fr$ ($r = 0.98$), and $H$ ($r = 0.91$). TFRU sensory score was also significantly correlated with $G$ value, but with a lower correlation coefficient ($r = 0.78$). Alasalvar and others (2002) found high correlations between $K$, $Ki$, $G$, $P$, or Fr and storage time ($r > 0.98$), indicating that these values offered a better indication of sea bass (Dicentrarchus labrax) freshness than the $H$ value.

The $K$ value limit for consumption differs among species. For example, Alasalvar and others (2001) found that the $K$ value of sea bream ($Sparus aurata$) was 39% when fish were considered unacceptable, and Lakshmanan and others (1996) found limited $K$ values amounting to 70.5% and 54.9% for mullet (Lisa corsula) and pearl-spot ($Etroplus suratensis$), respectively. In the limit of acceptability (13 to 16 d), $K$ values of grass carp killed by ice-water slurry and by electricity ranged from 72% to 79% and 73% to 80%, respectively (Figure 4). These values are higher than the previously reported $K$ value for grass carp (35%; Ehira and Uchiyama 1973). The average $Ki$, $P$, $H$, and $Fr$ values for grass carp stored in ice. Grass carp were killed by immersion in ice-water slurry (a) or by electricity (b). Results are means ± standard errors ($n = 5$). Means within the same column that have no common letters are significantly different ($P < 0.05$).

![Figure 4](image-url)
Ex values in the limit of acceptability ranged from 75% to 83%, 73% to 81%, 15% to 17%, and 24% to 16% for fish carcasses killed by immersion in ice-water slurry, and 79% to 86%, 76% to 83%, 24% to 30%, and 20% to 13% for carp carcasses killed by electricity (Figure 4).

Conclusions

The limit for the acceptability of grass carp stored in ice was around 13 to 16 d. The modified TFRU scheme sensory score had a highly positive correlation with storage time. Gills, eye shape, and belly firmness were good indicators of the freshness of grass carp. K, P, F, and H values had high correlations with storage time and with the TFRU sensory scores and could be used to assess the freshness quality of grass carp. Slaughtering by electricity accelerated the initial rate of ATP degradation and the development of rigor mortis, but did not affect the shelf life of grass carp stored in ice when compared with slaughtering by immersion in ice-water slurry. Electricity did not yield blood spots in the flesh.

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Furthermore, the use of process waters (Ramanathan and others 2000b) and the use of methods based on sensory, chemical, and microbiological quality of rainbow trout (Onchorhynchus mykiss) stored in ice and MAP. Eur Food Res Technol 219:211–6.


