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Assessing the quality of sardine based on biogenic amines using a fuzzy logic model

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ABSTRACT

There is an increasing concern about the quality and quality assessment procedures of seafood. In the present study, a model to assess fish quality based on biogenic amine contents using fuzzy logic model (FLM) is proposed. The fish used was sardine (*Sardinella* sp.) where the production of eight biogenic amines was monitored over fifteen days of storage at 0, 3 and 10 °C. Based on the results, histamine, putrescine and cadaverine were selected as input variables and twelve quality grades were considered for quality of fish as output variables for the FLM. Input data were processed by rules established in the model and were then defuzzified according to defined output variables. Finally, the quality of fish was evaluated using the designed model and Pearson correlation between storage times with quality of fish showed r = 0.97, 0.95 and 1 for fish stored at 0, 3 and 10 °C, respectively.

1. Introduction

The shelf life of fish under storage and especially in elevated temperatures is very short and, therefore, assessment and quality measurement of fish is necessary (Duflos, Dervin, Malle, & Bouquelet, 1999). Many investigators have proposed methods for evaluation of fish quality. Among the most widely used indicators are quantification of total volatile basic nitrogen (TVBN), trimethylamine (TMA) content, the ratio of TMA/TVBN, quantification of nucleotide derivatives and assessment of biogenic amines (Duflos et al., 1999; Ruiz-Capillas & Jiménez-Colmenero, 2010). However, there is still no complete agreement as to which proposal should be applied for all species of fish or different fish products. The absence of a rejection limit in most of proposed methods and a wide variety in fish species are some drawbacks (Duflos et al., 1999).

Assessment of biogenic amines is one of the most applied methods to evaluate fish quality. Research and development, control of processing steps, raw materials quality control, end product control and fermentation processes monitoring are some applications of analysis of biogenic amines (Ruiz-Capillas & Jiménez-Colmenero, 2010; Önal, 2007). They are found in many foods including fish and fishery products and the levels of biogenic amines usually increase during storage of fish (Ruiz-Capillas & Jiménez-Colmenero, 2010; Silla Santos, 1996). Furthermore, they indicate a wide range of toxicity and hazardous effects on human health (Prester, 2011). Histamine, the most important biogenic amine, has frequently been reported as the main agent of scombroid fish poisoning while other biogenic amines such as putrescine and cadaverine are suggested as potentiators of histamine (Hungerford, 2010). Apart from the role of biogenic amines in scombroid fish poisoning, they have been reported as potential carcinogenic compounds where formation of nitrosamines from biogenic amines and especially putrescine and cadaverine has been investigated by many researchers (Al Bulushi, Poole, Deeth, & Dykes, 2009; Ruiz-Capillas & Jiménez-Colmenero, 2010). At present, the legal maximum level of biogenic amines in fish products has been established only for histamine and not for the other biogenic amines. The Food and Drug Administration (FDA) of the United States of America has established a guidance level for histamine at 50 mg/kg for assuring the safe consumption of scombroid or scombroid-like fish (FDA, 2009). The European Union established 100 mg/kg as the maximum level of histamine for Scombridae and Scomberesocidae families (European Community, 1991). For total biogenic amines, a maximum level of 300 mg/kg has been suggested by the European Community (1991). However, estimation of histamine alone has not been deemed adequate as an indicator of decomposition (Al Bulushi et al., 2009; Mietz & Karmas, 1978). Histamine

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accumulation during storage is frequently lower than other biogenic amines such as putrescine and cadaverine, even in fish containing high contents of free histidine. Moreover, formation of histamine in fish is highly dependent on fish species and storage temperature (Al Bulushi et al., 2009; Ruiz-Capillas & Jiménez-Colmenero, 2010). Therefore, finding a trustworthy method to include other biogenic amines in fish quality could elevate the standards of fish quality assessment.

Some investigators have proposed multiple biogenic amines as quality indicators to evaluate fish quality. Mietz and Karmas (1978) introduced a chemical index ranging from 0 to 10 to evaluate fish spoilage using histamine, putrescine, cadaverine, spermine and spermidine. They estimated the quality of salmon, tuna and rock fish, and considered 10 as the limit for acceptation of fish. On the other hand, Veciana-Nogués, Mariné-Font, and Vidal-Carou (1997) proposed the sum of putrescine, cadaverine, histamine and tyramine as another quality indicator and reported a good correlation between organoleptic value or time of storage and this quality indicator. An amine index was also introduced by Duflos et al. (1999) to evaluate the quality of plaice and Whiting. However, endeavors to correlate fish quality to these indicators faced some drawbacks including the absence of some biogenic amines during the early stages of storage, differences between fish species, lack of a clear limit for rejection of the fish in some of them, different patterns of formation of some biogenic amines during storage and allocating equal weight to histamine and other biogenic amines (Al Bulushi et al., 2009; Duflos et al., 1999). An ideal quality indicator should consider different weights for histamine and other biogenic amines because of differences in toxicity and hazardous effects. It should also be able to distinguish rejected and non-rejected fish by score.

The Fuzzy Logic Modelling (FLM) is a successful tool to predict uncertain issues. The theory of fuzzy logic was first developed by Zadeh (1975, 1983). The theory discusses about parameters in systems with impossible precise definition. In fuzzy logic theory, unlike standard theory, the belonging of an object to a set is not absolute. It means that an object could belong to a set partially. Since biological systems are not crisp or exact and many objects in biological systems are explained as a range of values, FLM could successfully be applied to explain them. Although biogenic amines usually appear during storage of fish and could be suitable indicators for assessment of fish quality, their increase during storage indicates a vast fluctuation and variation and, species-dependent even at identical temperatures and times of storage. Furthermore, the actual toxicity level or hazardous level of biogenic amines is unclear because of their various hazard roles as potentiators of histamine, carcinogenic and toxic effects. Thus, they could be an appropriate subject for FLM. FLM allows different roles for any biogenic amines as independent variables to be defined. However, little information exists regarding application of FLM on the fish quality assessment using biogenic amines. Therefore, in the present study, finding a compatible model for assessment of fish quality by biogenic amines using FLM was examined on sardine under storage as one of the most popular templates in fish experiments.

2. Materials and methods

2.1. Chemicals

Standard biogenic amines including histamine dihydrochloride, 2-phenylethylamine hydrochloride, spermine tetrahydrochloride, spermidine trihydrochloride, cadaverine dihydrochloride, tryptamine hydrochloride, putrescine dihydrochloride, tyramine hydrochloride were purchased from Sigma Aldrich (St. Louis, MO, USA). HPLC grade chemicals including methanol, acetonitrile, ammonium acetate and other chemicals with analytical grades were obtained from Merck (Darmstadt, Germany).

2.2. Sample preparation

Sardine (*Sardinella* spp.) weighing between 50 and 75 g was obtained in the early morning from a local wet market in Serdang, Selangor, Malaysia. They had been kept in ice since their capture less than 12 h before purchase. After purchase, the fish were placed in ice and transferred to the laboratory within one hour. All fish were immediately rinsed under running tap water and divided to three groups. Each group was placed in a rectangular plastic container, covered with aluminum foil and then stored at 0 ± 1, 3 ± 1 and 10 ± 1 °C for up to 15 days. During storage, six pieces of fish were withdrawn at random, eviscerated and rinsed under tap water. Eviscerated fish at Day 0 served as the control. Then, the fish meat was removed with a knife and homogenized in a Warring blender (Model 32BL79, USA), transferred into glass bottles, capped and stored at −80 °C. Before to analysis, bottles were removed from frozen storage and kept at ambient temperature until the fish meat has thawed.

2.3. Extraction of biogenic amines

To extract biogenic amines, 5 g of homogenized fish muscle was vortexed with 20 ml of 6% (w/v) trichloroacetic acid in a 50 ml centrifuge bottle for 3 min (Zare, Muhammad, Bejo, & Ghazali, 2013). The extract was then centrifuged for 10 min at 8000*g* and 4 °C. The supernatant was filtered through a Whatman No. 2 filter paper (Whatman, Maidstone, UK) and the volume of extract was adjusted to 50 ml with 6% (w/v) trichloroacetic acid. Biogenic amine derivatization was accomplished with 2 ml aliquot of extracted solutions.

2.4. Biogenic amines determination and HPLC conditions

Derivatization of biogenic amines in the extracts obtained above, chromatography conditions, injection of standards and calculation of biogenic amines concentration were performed according to Zare et al. (2013), Zare, Muhammad, Bejo, and Ghazali (2015). The HPLC separation column was an endcapped Purospher STAR RP18 (5 μm, 250 × 4.6 mm I.D., from Merck, Darmstadt, Germany) equipped with a Purospher RP-18e guard (4 mm × 4 mm I.D., Merck). The HPLC apparatus was Waters 2695 Alliance HPLC (Waters Corp., Milford, MA, USA) system. The system had been equipped with an online degasser, two channel UV detector (Waters 2478), two HPLC pumps (Waters 515) and an autosampler.

2.5. Algorithms and computational methods

In FLM, three main steps namely fuzzification, fuzzy processing and defuzzification are necessary (Zadeh, 1983). The designing of these three main steps in the present study is explained as follow.

2.5.1. Fuzzification

In the first step of FLM, it is necessary to define the vague desired input variables (Zadeh, 1983). Within this step, the input variables are interpreted by the fuzzy system. Fuzzification is performed by definition of two main components which consists of membership and labelling. To evaluate fish quality, input variables and memberships were defined according to levels of biogenic amines during storage of sardine at different temperatures, which were evaluated in present study, along with extensive literature review and evaluation of other similar studies as will be explained in Section 3. Three biogenic amines, histamine, putrescine and cadaverine, were selected as input variables and three different levels of low, medium and high were defined for each one as