Postharvest Handling Practices and the Development of Histamine in Giant Trevally (*Aranxignobilis*) Fish: The Case of Fiji

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Abstract

This study aimed at assessing the common post-harvest handling practices of artisanal fishers in Fiji and its impact in the development of histamine in fish. The study involved in-depth interviews of postharvest handling practices, and its relationships in the development of histamine in Giant Trevally (*Aranxignobilis)* fish that were further exposed to three different temperatures; 0^0C, combination of 0^0C and 28^0C, and 28^0C. Results revealed that spear-gun fishers do not ice fish and rarely gut fish during their fishing trips, however only iced by either middlemen or vendors when fish are at the landing sites and are stored in old freezers. Fish are usually displayed un-gutted without ice on well drained tiled stalls and are recycled for display every two hours. Any unsold fish are returned into the old freezer for sale the next day. Histamine determination revealed that after 35 hours postharvest including further storage at 0^0C, and at combination temperatures of 5^0C and 28^0C (recycled every two hours) had histamine levels below the Food Drug Administration (FDA) permitted levels (50ppm). However fish stored at 28^0C showed histamine concentrations increased to 192.20 ppm at 15 hours post-harvest. The study demonstrates the importance of time and temperature control at postharvest handling of fish in tropical countries.

Key terms: *Histamine, Scombrotoxin, Calometric Test, Post-Harvest Handling Practices.*

INTRODUCTION

Fish is a perishable commodity (Bolta, 1995) and that postharvest handling is crucial in retaining its freshness, quality and safety. Quality parameters of fish are strongly related to its degree of freshness which warrants cold chain along the supply chain. It is understood that from the moment the fish is caught, deterioration process starts (Martinsdottir, 2002) which affects quality. These deterioration processes contribute to changes in the composition and structure of the fish due to biochemical, physical, enzymatic and bacterial reactions which negatively affects the sensory quality of fish and its related products. Good handling practices need to be seriously practiced in order to reduce fish spoilage, wastage and histamine production. Fish handling practices that increase risks of unsafe foods are public concerns which warrant serious action in the prevention of food-borne illness or fish poisoning (Ergönül, 2013) including Histamine Fish Poisoning (HFP).

HFP is a mild food-borne illness mainly caused by decarboxylation of histidine when exposed to high temperature over a long period of time mainly in scombroid fish species such as tuna, skipjack, bonito and mackerel. Some non-scombroid fish including sardines, herring, pilchards, marlin, mahi-mahi, mullet, kingfish and travelly also have been involved in outbreaks of scombrotoxin. Prevalent in the Asia-Pacific region as well as the United States (Patange *et al.*, 2004), scombrotoxin needs to be monitored and that histamine determination is an important part of food safety and international trade (Björnsdóttir-Butler *et al.*, 2009; Sánchez-Guerrero *et al.*, 1997; Wu *et al.*, 1997). Even though HFP may be common in the Pacific region, cases are rarely reported to medical authorities (WHO, 2011) and not officially recorded. Fish and fish products are regarded as one of the most commonly consumed protein sources in Fiji which showed 66% of the indigenous Fijian households consume fish either once or twice a week (National Food and Nutrition Centre, 2007). Because fish is highly perishable, handling of fish and fish products is crucial in the prevention and reduction of fish poisoning.

While histamine fish illness is associated with high intake of histamine levels greater than or equal to 50 mg/100 g fish (Lehane and Olley, 2000) it should be noted that real toxic dose of histamine to the human body is not easy to determine due non-homogeneity in the formation of histamine in the fish's body (Lerke *et al.*, 1978) and the presence of other biogenic compounds that potentiate toxicity (Shahidi and Simpson, 2004). Generally, the production of histamine in fish is related to various factors such as histidine content of the fish, presence of bacterial histidine decarboxylase (HD), and environmental conditions in which fish is stored (Ienistea, 1973). It is understood that during fish spoilage, certain bacteria produce decarboxylase enzymes, which act on free histidine and other amino acids in the fish muscle to form histamine and other biogenic amines (Eitenmiller and De Souza, 1984). The main bacteria responsible for histidine decarboxylation and HFP are reported to be members of the family Enterobacteriaceae (Frank *et al.*, 1985; Taylor and Sumner, 1986). Moreover, studies suggest that storage of longtail tuna at 25^0C for 1 day rapidly increased histamine levels to 284mg/kg compared to storage at 0^0C and 8^0C (Abdallah Al-Busaidi et al., 2011).
There are effective testing methods to identify likely toxic fish; hence their control and preventive measures are possible (Shahidi and Simpson, 2004). However, it is not possible to test all fish and thus a Hazard Analysis and Critical Control Points (HACCP) approach is the preferred management approach in the avoidance and reduction histamine development. The aim of this research was to assess the current handling practices of fish by artisanal fishers and its relationship with histamine development in Giant Trevally (*Aranxignobilis)* fish.

METHODOLOGY

This research employed two different approaches; an in-depth interview using piloted questionnaires that gathered the post-harvest handling practices of fish and the determination of histamine in *Giant Trevally* fish that were exposed to three different sets of temperatures.

Post-Harvest Handling Practices

An in-depth interview was conducted on six fishermen and their associated vendors and retailers that identified the supply chain and the associated handling practices of fish they caught, handled and/or processed and auctioned. The handling practices data related to time and temperature were used to set up a time-temperature abuse pilot experiment to demonstrate the relationship between time-temperature exposure discussed below and the development of histamine in *Giant Trevally* prior to histamine determination.

Time-Temperature Exposure and Sample Preparation

A batch of *Giant Trevally* containing a total of 5 fish was purchased from the major local fish market in the Capital City; Nabukalou Creek, Suva. The history and temperature of the batch of fish that was purchased was gathered through in-depth interview discussed above prior to transportation of fish to the postharvest laboratory at the University of the South Pacific. In the lab, the fish were quickly cleaned, filleted and diced into 1cm cubes and stored at three different sets of temperatures; 0^0C, 28^0C and temperature combination of 5˚C and 28˚C respectively for up to 35 hours in total, prior to histamine determination using the Histamine Test Kit purchased from AltiMed Australia Pty Ltd discussed below.

Histamine Determination

In brief, from a 3g of homogenized *Giant Trevally* fish tissue, 1g was transferred to a heat resistant test tube and added with 24ml of EDTA buffer and boiled for 20 minutes. This was placed on ice to cool to <20˚C and then filtered. The liquid phase was used for histamine analysis following the instruction in the test kit. Absorbance was measured at 470 nm with the UV Spectrophotometer Brand Kyoritsu Chemical; Corporation, Model ABS-B470.

Results and Discussion

Post-Harvest Handling Practices

Hygiene and good handling practices on board the fishing vessel, at the landing site, during transportation and processing of fish, remain the highest priority in avoiding contamination and cross-contamination which may contribute to the reduction of histamine production in fish species high in histidine. Figure 1 shows the current postharvest handling practices of fish caught by artisanal fishermen through spear-gun method and then auctioned or sold by their associated vendors and retailers. In-depth interviews revealed that most artisanal fishermen go out fishing overnight; usually from 7pm (after the sunsets) and returns around 7am (before the sunrises). This shows approximately 12 hours of fishing trip in small on-board vessels without ice by artisanal fishers. According to fishers, 12 hours overnight fishing is a short fishing duration hence no ice is required for chilling their catch. Fish in this case fish are usually stored loose on pallets on-board the vessel overnight without ice.

According to fishers, knowing the best time and site to catch fish reduce the duration of fishing trip and hence spear – gun fishers prefer to catch fish when the fish are asleep around 10pm to 5am, which approximately 6-7 hours depending on weather and species of fish. After catching fish early in the morning, fish are sort by size and threaded in bundles without gutting and placed back on loose pallets for storage without ice. The fish are then transported to the fish landing sites in Suva, in time for the arrival of middle-men and retailers/vendors around 7am.

<u>Figure 1: Post Harvest Handling Practices of Fish from Harvest to Auction/Sale</u>

At the landing site, the bundled fish are transferred without gloves from the fishing vessel to old freezers containing ice by the middle-men, retailers or vendors. This shows that icing or chilling of fish only takes place upon landing and that quantity of ice used is generally not sufficient to thoroughly cool all the fish that have been bought by middlemen. Furthermore, most old freezers look unhygienic and do not have drain - holes that allow melted-water to drain away. This contributed to accumulation of drip from fish which usually tainted with blood and that provide good source of nutrients for bacterial growth. When auctioned, some fish are displayed on tiled and well drained stalls un-gutted and without ice, which sometimes are kept for hours before being sold. Majority of the fish vendors recycled the fish on display by putting the fish back in the old freezer container every two hours, together with the melted-water. Any unsold fish for the day are returned into the old freezer for sale the next day. It is observed that melted-water is not usually removed from the containers, which appears to be a common practice by the vendors. Instead, new ice is simply added onto the fish containing melted- water mixture. This practice of recycling of fish at the combination temperatures of 5^0C and 28^0C every two hours was adopted as part of the experiment discussed below prior to the determination of histamine.

The major issues identified in the current post-harvest practices with fishermen include;

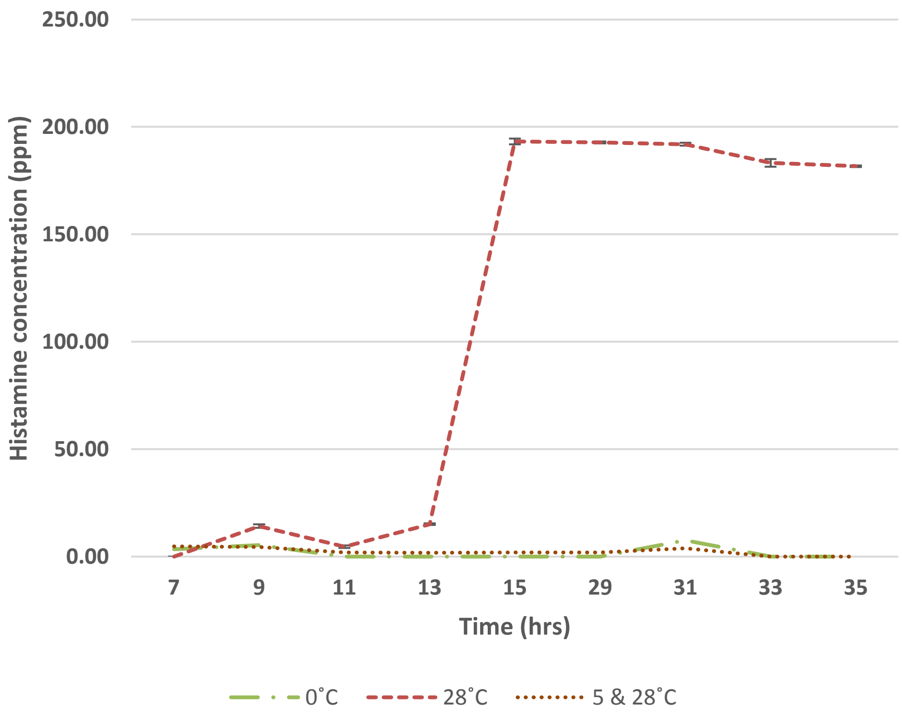
- The fishing method in the use of spear-guns or sharp gear cause open injury and bruises to fish which enhances and accelerates deterioration and spoilage of fish. The impact of such fishing method becomes more complex during long fishing trips especially when ice is not used.
- Fish are openly exposed on pallets on-board fishing vessels without proper cool storage or ice. The problem is aggravated when the fishing vessels are un-hygienically dirty without proper cleaning and sanitizing prior to the fishing trip.
- Chilling fish in the dirty old freezers with melting ice, and addition of new ice without removing melting water increase the chances of contamination.
- Displaying of fish for auction or sale without ice and recycling the fish every two hours intervals back into the old dirty melted ice when not sold. Any unsold fish are returned into the old freezer for sale the next day which increase chances of contamination and recontamination.

Time-Temperature Exposure and Histamine Determination

Histamine is considered to be principal toxic component of scombrotoxin, although other compounds such as biogenic amines including putrescine and cadaverine may also play a role in scombrotoxin (Histamine – Scombrotoxin; www.atuna.com/index.php/health/histamine). Food regulations in most countries allow counts up to 50 or 100ppm, as safe levels (FDA, 1998). In countries with small coastal vessels, a lack of supply of ice and insufficient cold supply chain with poor hygienic circumstances, histamine is unfortunately still frequently detected at high levels. Acceleration of histamine development to unacceptable levels takes place when fish is transported in un-chilled trucks or open air in tropical temperatures (Histamine – Scombrotoxin; www.atuna.com/index.php/health/histamine).

Figure 2 shows the histamine level of *Giant Trevally* that were further exposed at three different temperatures; 0^0C, 28^0C and combination of 5^0C and 28^0C after 7 hours of post-harvest exposure. The three temperatures selected was based on the information gathered from the in-depth interview discussed above in which recycling of fish at auction or display of fish for sale occurs between 5^0C and 28^0C at intervals of 2 hours. Results revealed that *Giant Trevally* stored at 28^0C only had elevated growth of histamine levels over 31 hours post-harvest compared to 0^0C only and combination temperatures between 5^0C and 28^0C recycle. The results obtained in this study confirmed that low temperature; 0^0C and 5^0C are effective in controlling, limiting and suppressing histamine formation which had been supported by several studies (Guizani *et al.,* 2004; Silva *et al.,* 1998) which showed negligible levels of histamine detected at storage temperature of 0^0C and below. This means that histamine development is suppressed during storage at such low temperatures. Further results at 28^0C revealed the highest concentration of histamine at 193.16ppm after 15 hours of exposure. This data appears to be in agreement with Mu¨ ller *et al.,* (1992) and Arnold *et al.,* (1980) that reported the highest histamine level detected in *trevally* species stored at 28^0C was detected unacceptable highest histamine concentration after two days of storage. This may be due to the optimal temperature for most bacteria that grow at 20-30^0C, although some histamine producing bacteria such *vibrio* species have temperature optima below 10^0C (McMeekin *et al.*, 1993). It is interesting to observe that histamine level at 28^0C exceeded the FDA permitted levels of 50ppm fresh weight at 13.5 hours of post-harvest and exceeded the EU permitted levels of 100ppm fresh weight at 14.0 hours post-harvest duration.

Figure 2. Histamine levels (ppm) of *Giant Trevally* stored at 0^0C, 28^0C & combination between 5^0C & 28^0C



According to Brinker *et al.,* (1995), histamine level in a composite sample of fish or fish products, other than crustaceans or molluscs, must not exceed 100mg/kg. If histamine levels are above 100mg/kg it may indicate that fish had been mishandled during storage and processing. Moreover, histamine-forming bacteria are capable of growing and producing histamine over a wide temperature range, however more rapid at high-abuse temperatures of 21.1^0C and higher than moderate-abuse temperatures of 7.2^0C (Emborg *et al.*, 2004). It has been argued that histamine growth is particularly rapid at temperatures near 32.2^{0}C (FDA, 2011). It has been observed that histamine is the result of high temperature spoilage for long-term, while relatively low-temperature spoilage is commonly associated with organoleptically detectable decomposition (FDA, 2011). Handling fish at high temperatures such as 20^{0}C contributes towards spoilage of fish with regards to histamine level mainly in the subtropical countries. Emborg *et al*., (2004) argued that formation of microbes on histamine and biogenic amines in chilled fresh tuna stored at 2^{0}C vacuum-packed (VP) cause histamine poisoning at a concentration of >7000mg/kg. Histamine ELISA kit and plate count agar (Auerswald *et al.*, 2005) have proven histamine to be strongly dependent on temperature-time (Yesudhason *et al.*, 2012).

Interestingly this this study with *Giant Trevally* samples stored at the combination temperatures between 5^{0}C and 28^{0}C recycled every two hours showed histamine levels well below FDA and EU permitted levels. The highest recording was 4.8ppm at 7 hours of storage. This result appears to be in agreement with that of Lopez-Sabater *et al.,* (2005) who observed the growth of histamine stopped in bigeye tuna when it reached its maximum level. Similarly Sato *et al.*, (1994) also observed that initially, histamine accumulated in fish stored at 5^{0}C and then levels start decreasing when the count of histamine decomposing bacteria exceeded 10^6 cells/g. These results are comparable with the study conducted by Auerswald *et al.*, (2005), who reported that histamine level in the *trevally* sample stored at 5^{0}C only (Sato *et al.,* 1994) increased only marginally from 1.7 to 5.3ppm during four days. However, because the current research was only conducted for a duration of 35 hours post-harvest observation, data could not be matched. Nonetheless, one could predict that combination temperatures of 5^{0}C and 28°C could assist delay the formation of histamine below FDA permitted levels to a certain degree only for short duration and expected to increase during long storage. The result of this study confirmed that temperature and time control are essential in the prevention of histamine development. There may be other factors that contribute to the rate of histamine growth at certain temperatures including species which may be due to biological make-up (Kanki *et al*., 2004) and as well as normal microbial flora on fish and post-catching contamination on board fishing vessels. The key to keeping bacterial numbers and histamine levels low is the rapid cooling of fish after catching and the maintenance of adequate refrigeration during handling and storage as observed at 0^{0}C which delayed spoilage.

Although many countries have set guidelines for maximum permitted levels of histamine in fish including EU and FDA at 50ppm and 100ppm respectively (FDA, 1998), histamine concentrations within a spoiled fish are extremely variable, as in the threshold toxic dose. Despite the huge expansion in trade in recent years, great progress has been made in ensuring the quality and safety of fish products mainly through the introduction of international standards of food hygiene and the application of risk analysis and hazard analysis and critical control point (HACCP) principles (Huss *et al.*, 2004). These safety standards and HACCP need to be strengthened and implemented in Fiji in order to reduce and avoid scombrotoxicity.

Conclusion

The current handling practices of scombroid fish when recycled at 2 hours interval between 5^0C and 28^0C up to 35 hours of post-harvest appear to have safe levels of histamine. However, continuous recycle at 28^0C only after 13hrs of post-harvest of scombroid fish revealed unsafe levels of histamine, contributing to unnecessary levels of histamine poisoning. Further investigation of histamine development in fish exposed to 2 hours interval recycle between 5^0C and 28^0C beyond 35 hours post-harvest especially when fish are not sold after 3-4 days.

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