Reduction of aerobic and fecal coliform bacteria counts of fresh whole reef fish (*Epinephelus malabaricus*) using Lactic Acid

D.E.S. Abeywickrama^{1*}, P.Ginigaddarage², H.W. Cyril¹, K.S.Hettiarachchi² and G.J.G.Arachchi²

Abstract

The quality and safety of fish products is a critical factor to increase market value. Therefore, decontamination methods are used in the fish processing industry to reduce microbial count on fish, to gain maximum health and economic benefits. Reef fish (Epinephelus malabaricus) has a high demand in the export market. The objective of this study is to determine the effect of lactic acid to reduce the microbial load by monitoring Aerobic Plate Count (APC) and Fecal Coliform (FC) counts in uneviscerated Epinephelus malabaricus. Fresh whole reef fish were treated with separate chilled solutions of Lactic Acid (LA) containing 1, 2 and 3 % (w/v) by dipping for 5 and 10 minutes. Chilled water containing 0.5-1.0 ppm chlorine was used as a control. APC, FC count and sensory characteristics (odour, colour, texture and overall appearance) of fresh whole reef fish and cooked flesh samples were evaluated. Reduction in APC and FC (FC was analysed using Most Probable Number; MPN technique) counts after LA treatment was 5.97 CFU/g and 1.41 MPN/g for 5 minutes of dipping time and 5.58 CFU/g and 1.15 MPN/g for 10 minutes of dipping time in 2% LA treatment. Fish dipped in 2% LA solution showed significant (P < 0.05) reduction of APC and FC count. There was no significant (P>0.05) effect of dipping time on APC and FC counts. No significant (P>0.05) interaction between duration of dipping and LA concentrations of treatment on bacterial counts was observed. Colour of gills and transparency of eyes of fish were negatively affected by LA treatment. Redness value of gills reduced and converted to greenish brown colour and eyes of fish were converted in to whitish colour in treated fish. Sensory evaluation showed that there was no significant (P<0.05) effect of LA on sensory attributes of cooked fish samples.

Keywords: Lactic acid, *Epinephelus malabaricus*, Decontamination, Aerobic Plate Count, Fecal Coliform

Introduction

The quality of fish products is a critical factor affecting market demand and it is a major goal of the fisheries industry to provide safe, wholesome and acceptable fish products to the consumer (Frank and Koffi, 1990). Microbial spoilage plays an important role, and with respect to health and economic problems caused by these

¹Department of Animal Science, University of Peradeniya, Sri Lanka.

²National Aquatic Resources Research and Development Agency (NARA), Crow Island, Colombo-15, Sri Lanka.

^{*}Corresponding author: sachinuop@gmail.com

microbes, it is very important to reduce the initial microbial population on fish (Smulders and Greer, 1998). With the development of the industry, dilute solutions (1-3%) of organic acids (Acetic, Lactic and Propionic) are being recognized as a safe antimicrobial agent by the FDA (Food and Drug Administration) and have been approved as food additives by European Commission. It can be used without effect on desirable sensory properties of fish (Smulders and Greer 1998). The overall objective of the present research was to study the effect of lactic acid on microbial load and sensory qualities of fresh reef fish (*Epinephelus malabaricus*) fish.

Materials and methods

Sample collection: The samples were collected from a seafood processing and exporting company located at Ja-Ela in Sri Lanka. Fresh whole reef fish (*Epinephelus malabaricus*) of approximately same body weight were collected immediately after receiving to the processing plant and treated immediately.

Preparation of Lactic acid (LA) solution and treatments: Lactic acid solution was prepared by diluting 1%, 2% and 3% (w/v) of L (+) lactic acid (PURAC FCC 88, PURAC Thailand Ltd. Thailand) in chilled water. Separate groups of fresh whole reef fish were dipped for 5 min and 10 min in separate LA solutions containing 1, 2, and 3% (w/v) as treatment 1, 2, and 3 respectively. The chilled potable water that was treated with 0.5-.0 ppm chlorine was used as the control for each treatment. Flesh and skin samples from each fish were collected before and after each treatment.

Microbiological Analysis: Fish samples collected in sterile plastic bags were packed in ice and transported to the Laboratory of Institute of Post-Harvest Technology (IPHT) at National Aquatic Resources Research and Development Agency (NARA) in less than two hours. APC and FC counts were determined according to the SLS 516 Part 1:1991 and SLS 516 Part 3:1982 respectively.

Sensory analysis: Fresh whole reef fish and cooked samples (control and treatments) were evaluated through Hedonic scale test. A trained panel made up of 9 panelists was selected. Samples were cooked in polypropylene pouches in boiling water for 15 min and cooled. Samples were evaluated using a five point hedonic scale.

Statistical Analysis: Analysis of Variance (ANOVA) performed using SPSS Version 15.0 and comparison of means were made using Tukeys test at the 95% confidence level. Sensory data was analyzed by Friedman non- parametric statistics using Statistical Analysis System (SAS).

Results and Discussion

Table 1: Effect of Lactic acid concentration and duration of dipping time on reduction of APC and FC count of fish (Mean \pm SD)

	Reduction APC (log ₁₀ CFU/g)		Reduction of FC count (MPN/g)	
Duration of	5 minutes	10 minutes	5 minutes	10 minutes
Control	5.37 ±0.42	5.27±0.10	0.84±0.22	0.84±0.13
1% LA treatment	5.17±0.04	5.13±0.08	0.75±0.27	0.81±0.18
2% LA treatment	5.97±0.01	5.58±0.08	1.41±0.01	1.15±0.02
3% LA treatment	4.9 7 ±0.23	5.16±0.11	0.65±0.24	0.8±0.03

Table 1 shows all the treatments reduced the APC and FC count of fish. Kim and Marshall (2000) also reported that lactic acid was effective in suppressing bacteria on refrigerated catfish fillets. But significantly (P<0.05) highest APC and FC count reduction was observed in 2% LA treatment. According to the Dan Rotaru and Dalea (2008), possible explanation for these results are, there are some biochemical reactions at bacterial membrane level, with some precipitation reactions in the superficial areas, fact that couldn't permit the penetration of concentrated acid solutions inside the microbial cell, keeping its viability

There was no significant (P>0.05) effect of duration of dipping time on reduction of APC and FC count of fish. Kim and Marshall (2000) mentioned firmly attached microbe cells were less sensitive to lactic acid than loosely attached cells. It gives positive explanation for results of the present study that duration of dipping was not significantly (P>0.05) affected on reduction of APC and FC count. No significant (P>0.05) interaction was observed between LA concentration and dipping duration in the experiment.

According to sensory evaluation of fresh fish, control sample had the highest scores for gill appearance, odour of gills, texture, and overall score than the samples dipped in LA for 5 and 10 minutes. Development of brown colour of gills and reduced transparency of eyes were due to the LA treatments. Appearance of skin and texture had approximately same scores in both control and treatments dipped for 5 and 10 minutes. According to sensory scores of cooked samples, all treatments and controls had approximately same scores for appearance, colour, odour, texture, flavour, and overall score. Lactic acid treatments were not negatively affected on sensory characteristics of cooked samples.

Conclusion

There was a significant (P<0.05) reduction of APC and FC on fresh reef fish at 2% LA (w/v). According to the results 2 % LA (w/v) solution was more efficient than the 1 and 3 % LA (w/v) solutions.

References

- Dan, S., Rotaru, O. and Dalea, I. (2008). The effects of lactic and acetic acid treatment on the psychrotrophic germ growth from the surface of beef and pork. 245–250pp.
- Frank, J.F., and Koffi, R.A. (1990). Surface-adherent growth of Listeria monocytogenes is associated with increased resistance to surface sanitizers and heat. Journal of Food Protection, 53: 550-554.
- Kim, C.R. and Marshsall, D.L. (2000). Quality evaluation of refrigerated chicken wings treated with organic acids. Journal of Food Quality, 23: 327-335.
- Smulders, F. J. and Greer, G.G. (1998). Integrating microbial decontamination with organic acid in HACCP programs from muscle foods: prospects and controversies. International Journal of Food Microbiology, 443: 149-69.