Asian Fisheries Society, Selangor, Malaysia

Quality Assessment of *Labeo rohita* and *Labeo calbasu* sold in Commercial Outlets of Sambalpur, Orissa, India

JYOTIRMAYEE SAHU¹, SEEMARANI SHASINI¹ and M. M. PRASAD^{1*}

¹Burla Research Centre of Central Institute of Fisheries Technology Burla-768 017, Sambalpur, Orissa, India

Abstract

Bacteriological, biochemical, and sensory qualities of commercial sold *Labeo calbasu* and *Labeo rohita* were reported. The moisture, total volatile nitrogen (TVN), Peroxide Value and α -Amino nitrogen were 73.46 %, 9.65 mg % N, 23.15 meq. Kg⁻¹ fat and 29.06 mg^{-100g}, respectively, for *L. calbasu* and 79.80 %, 6.73 mg % N, 23.02 meq. kg⁻¹ fat and 29.16 mg^{-100g}, respectively, for *L. rohita*. The bacteriological analysis of *L. calbasu* showed that total viable bacteria were at 4.82 log CFU g⁻¹, the sample was free from coliforms and faecal streptococci, although it harboured Group-D Streptococci and Staphylococci. The bacteriological analyses of L. rohita before and after washing indicate reduction in the counts of different bacteria groups by 1 to 2 log CFU g⁻¹ of the sample. Variation in counts of different bacteria was also observed from edible meat portion to gut sample of the same variety of fish. The fresh *L. calbasu* and *L. rohita* scored an average of 9.75 and 9.2 in overall sensory evaluation by ten point Hedonic scale indicating the excellent quality. In addition, the cooked *L. calbasu* and *L. rohita* scored 8.58 and 7.9 out of ten points, respectively, which confirm that both the fishes were excellent in condition at the time of sale. In view of the occurrence of hygiene indicator bacteria more number of studies is necessitated.

Introduction

Fish form a rich source of animal protein available at an affordable price to all sections of the society and provide a means to tide over the nutritional difficulties of man. Importance of fish as a source of high quality, balanced, and easily digestible protein is well understood (Nair and Mathew 2000). Fish was one of the important items of bartered foods and was also exported outside the country in ancient times. From earning a few million rupees in initial years of Indian independence to a staggering gain of foreign exchange beyond billion dollars (US\$) or 72,000 million rupees, fishing industry is playing a pivotal role in Indian economy. Fish is also providing jobs to more than four million people (Prasad and Seenayya 1998). Annual production of inland fish has increased from 0.2 to 2.8 million t (14 folds) during the last five decades. During

^{*}Corresponding Author. Tel: +91-663-2430503

Email : prasadmothadaka@yahoo.com

this period fish production systems in the inland waters have expanded, diversified, intensified and technologically advanced. Inland open water systems such as rivers, estuaries, lagoons, flood plains, wetlands, and reservoirs contribute to nearly one million tons of fish (Sugunan 2002).

The information on quality of freshwater fish such as physical, biochemical, bacteriological, and sensory characteristics is scanty. The major food fishes of India include species of *Labeo, Catla, Cirrhina, Mystus, Wallago, Notopterus, and Ophiocephalus*. Importance of any fish as a food depends on its well-balanced chemical composition and it determines its nutritional quality (Shasini 2004). Reliable data on the nutritional quality of fish is therefore very essential in aiding the nutritionists and the technologists in dietary formulation, processing, and product development, as well as, nutrition labeling (Nair and Mathew 2000; Sahu 2004, Prasad et al. 2005).

In the light of the above, studies were undertaken on physical, biochemical, microbiological, and sensory characteristics of two freshwater *Labeo species viz.*, *Labeo calbasu* and *Labeo rohita* sold in wet fish market of Sambalpur, Orissa. Both the *Labeo species* are common varieties of IMC that are sold in many parts of Orissa.

Materials and methods

Collection of sample

Fish samples were collected from the local outlets of Sambalpur, Orissa in fresh condition in sterile polythene bag (200 gauge) for immediate analysis in the laboratory.

Physical characters of fish.

Immediately after bringing the sample, length, breadth and weight was recorded in aseptic conditions.

Assessment of freshness of fish

Intellectron-Fish Tester VI (German make) was used to assess freshness of fish samples. The measuring positions (base of tail and abdominal cavity) for *L. calbasu* and *L. rohita* were as follows: at the head region below operculum, in the mid region above the abdomen and at the tail region above caudal fin. The measured data were expressed as the "degree of freshness scale".

Sensory evaluation

For sensory evaluation of the raw and cooked fish, a group of expert panelists (15) were chosen and the same group carried out all the sensory evaluation throughout

the study. A small amount of fish samples were used for cooking in brine (2%) for five minutes. After bringing the temperature to ambient temperature, sensory evaluation tests were carried out (Sahu 2004; Shasini 2004). The sequence of observation included the general appearance, appearance of flesh, including, belly flaps, odour, and texture. The scoring ranged from a minimum of zero to maximum up to ten in a ten point Hedonic scale, where eight to ten was excellent, six to eight very good, four to six good, two to four average and two and below considered to be a spoiled case in which fishes are not fit for consumption. The statistical analysis of the data was carried out using standard methods (Visweswara Rao 1996).

Chemical composition

The fishes were analyzed for physical and biochemical quality parameters such as moisture, pH, fat, peroxide value (PV) and α -amino nitrogen (alpha NH₂) by standard methods (AOAC 1995). The Total Volatile Nitrogen (TVN) content of the fresh water fish samples were estimated by the method of Conway (1947).

Bacteriological examination of fish

Both the freshwater fish samples *Labeo calbasu* and *Labeo rohita* were screened for bacteriological quality that included total viable bacteria, coliforms, faecal streptococci, and staphylococci by standard methods (ICMSF 1978).

The bacteriological examination of the fresh water fish samples for total viable bacteria (TVB) were done by Miles and Mishra's method and spread plate method. The sterile poured plates were inoculated with sterile (Gamma irradiated) calibrated disposable pipettes (Volac-John Poulten Ltd, Barking, England).

Plate Count Agar (Standard Method Agar) was used to assess the load of TVB in the fish samples (ICMSF 1978).

Violet Red Bile Agar (1.2 percent) was used for the detection of coliforms present in the edible meat portion of the fish samples. Baird Parker Agar, a selective medium used for detection of coagulase positive staphylococci (ICMSF 1978). The media was sterilized in autoclave followed by addition of concentrated egg-yolk emulsion along with Potassium Tellurite solution and mixed well before pouring on the plates. KF Streptococcal Agar was used for the enumeration of faecal streptococci (ICMSF 1978). The required amount of KF Streptococcal Agar was boiled in distilled water to dissolve all the constituents followed by the addition of 1 ml (10%) of Triphenyl Tetrazolium Chloride to each 100 ml of sterile medium and mixed thoroughly. Kanamycin Aesculin Azide Agar, a selective media for the cultivation and isolation of Group-D streptococci was used.

Results and Discussion

The present study dealt on the quality aspects of the freshwater *Labeo calbasu* and *Labeo rohita*. The countries of distribution of *L. calbasu* and *L. rohita* include India, Pakistan, Bangladesh, Burma, Thailand, and South China (Sahu 2004). The principal freshwater *Labeo species*, consumed as food in India are *L. bata* (Bata), *L. boga* (Burmese fish or Jamuna fish), *L. boggut*, *L. calbasu* (Kalbasu), *L. dero* (Bongsa), *L. fimbriatus*, *L. gonius* (Cursa), *L. kontius*, *L. rohita* (Rohu) (Sashni 2004). The main *Labeo species* found in Orissa are *L. bata*, *L. boggut*, *L. calbasu* and *L. rohita* (Khanna 1999). In the present study, the fish used for experimental purposes were *Labeo calbasu* and *Labeo rohita*, which are bottom feeders, utilizing the decayed vegetation of benthic animals, plants and epiphytic plankton from the bottom of the pond. In Orissa, these two *Labeo species* are found in the river Mahanadi and its branches, and in ponds.

Physical characteristics of the fish

The length, breadth in three different regions of the body and weight of both the fish samples were taken in aseptic conditions. In the present study, the average (35 fishes) length of the fish *Labeo calbasu* (33.1 cm) was slightly higher to the observations made earlier (Bandhyopadhyay et al. 1985) in which the average length of *Labeo calbasu* was 30 cm. Whereas the length of *Labeo rohita* (33 cm) and weight (400g) were on lower side to the previous reports (Bandhyopadhyay et al.1985) where the average length and weight of *Labeo rohita* were 35 cm and 600g, respectively.

Freshness of fish samples

The samples of *Labeo calbasu* taken for experimental purpose showed maximum Intellectron reading at head portion (behind gills) (100), mid portion (belly region) (100) and end portion (caudal region) (100). The Intellectron reading for *Labeo rohita* were 72, 80 and 82 for head, mid and tail portions, respectively. The lowest indicator for freshness is '0' and the maximum is '100'. The readings of the present study indicate the freshness of both the fishes is of excellent category. In a study to assess the freshness of freshwater fish sold in wet fish market of Burla, the Intellectron readings varied from 17 to 75 and 47 to 72 for iced riverine/ reservoir fish and un-iced pond reared fish, respectively (Anon, 2004). In comparison with the above-mentioned study, the freshness of the fishes of the present study showed a marked improvement.

Sensory evaluation of fresh and cooked fish

Results of sensory characteristics of *L. calbasu* and *L. rohita*, of the present study are shown in Table 1.

Table 1. Physico-chemical quality characteristics of *Labeo rohita* : A comparison of present study with previous studies

Sl No Quality parameters		Comp	Comparison of results with earlier studies					
		Present	study S	Sahoo (2006)*	Bandypadhyay et al. (1985)			
		L. calbasu	L. rohita					
1	Moisture (%)	73.46	79.8	80.06	80.57			
2	Total volatile nitrogen (mg%N)	9.65	6.73	13.4	8.8			
3	Peroxide Value (m.e.q.Kg ⁻¹ fat)	23.15	23.02	33.32	18			
4	αNH_2 (mg g-100)	29.06	29.16	53.41	30.00			
5	Sensory evaluation (10 point	9.75 (fresh)	9.2 (fresh)) 7.5 (fresh)	9.9 (fresh)			
	hedonic scale)	8.58 (cooked)	7.9 (cooke	d) 7.2 (cooked)	ND			

ND: Not Done

In the present study the raw fish *L. calbasu* scored an average of 9.75, whereas *L. rohita* scored an average of 9.2 out of a ten point hedonic scale in overall appearance, flesh quality, odour and texture indicating the excellent quality. The average scoring of cooked *L. calbasu* and *L. rohita* was 8.58 and 7.9, respectively. This shows that the sensory quality of fishes of the present study were in excellent category.

Physical and Biochemical quality of the freshwater fishes

Details of different physical and biochemical quality analysis of *Labeo calbasu* and *Labeo rohita* collected from the local outlet are presented in Table 1. The compositions vary widely depending upon several factors such as species, size, sex, maturity and many others (Nair and Mathew 2000).

The moisture content of the fish *L. calbasu* was 73.46% while that of *L. rohita* was 79.80% (Table 1). The moisture content of *L. calbasu* is 2.5% (Jose Joseph 2002) and 7% (Bandhyopadhyay et al. 1985) lesser to the earlier reported data of the same variety of fish. Whereas the moisture content of *L. rohita* is 0.8% more to the earlier reports (Bandhyopadhyay et al. 1985). Jose Joseph (2002) reported the moisture in *L. rohita* in the range of 76.4 to 78.0%, which is slightly lower in comparison to the present study.

The fat content of the species *L. calbasu* and *L. rohita* were 0.85% and 4.44%, respectively, on dry weight basis. The earlier study of the same variety of fish, the fat percent varied from 1.80 to 4.20% (*L. rohita*) and 0.6% (*L. calbasu*) (Jose Joseph. 2002). The fat percent of *L. calbasu* of the present study is higher in comparison with same variety of fish reported by Jose Joseph (2002), while that of *L. rohita* is similar to

the higher range reported by the same author.

Determination of TVN forms the most widely used test for fish spoilage. The TVN content in the present samples of *L. calbasu* and *L. rohita* are 9.65 mg N % and 6.73 mg N %, respectively (Table 1). High TVN values were found to correlate with high bacterial activity and spoilage of fish (Sanjeev & Surendran, 1996). However, the level of TVN in the present study indicates that fishes sold in wet fish market of Sambalpur, are in good condition. According to Bandhyopadhyay et al. (1985) fresh *L. calbasu* and *L. rohita* showed the TVN of 8.8 and \geq 9.0 mg N % indicating a lower value (in *L. calbasu*) and a higher value (in *L. rohita*) in comparison to the present study.

The PV of *L. calbasu* and *L. rohita* after storing the samples at -8° C for three weeks and after subjecting the edible meat to ambient temperature for few hours were 23.15; 41.39 and 23.047; 36.90 meq/Kg fat, respectively (Table 1). The PV of other studies in *L. calbasu* was 18 meq/Kg fat and *L. rohita* was \geq 30 meq/Kg fat (Bandhyopadhyay et al. 1985).

Similar results were also observed in the analyses of the edible meat portion of the fish for á-amino groups. The á-amino nitrogen in the samples of *L. calbasu* and *L. rohita* stored at -8° C for 3 weeks and when exposed to ambient temperature were 29.06; 43.42 and 29.16; 64.80 mg g-100, respectively.

Microbiological examination of fish

The results of bacteriological analysis *viz.*, total viable bacteria, coliforms, faecal streptococci and staphylococci of both the *Labeo species* are shown in the Table 2.

No of samples		Bacterial counts before washing g-1				
Group D						
	TVB	С	FS	FS	Staphylococci	
1	58×10 ⁴	37×10 ²	94×10 ²	53×10 ²	85×10 ³	
2	14×10 ³	24×10 ²	17×10 ³	NG	40×10^{2}	
3	27×10 ³	23×10 ²	98×10 ²	18×10 ²	78×10 ³	
4	34×10 ³	28×10 ²	87×10 ²	25×10 ²	93×10 ³	
5	31×10 ³	19×10 ⁴	92×10 ²	34×10 ²	27×10 ⁴	

Table 2. Occurrence of total viable bacteria (TVB), hygiene indicator and other groups of bacteria in commercial sold *Labeo rohita*

No of samples		Bacter	rial counts afte				
Group D							
	TVB	С	FS	FS	Staphylococci		
1	27×10 ³	88×10 ²	80×101	50×101	50×10 ²		
2	14×10 ³	60×101	20×101	<101	30×10 ²		
3	19×10 ³	43×10 ²	16×10 ¹	80×101	CG		
4	22×10 ³	16×10 ²	56×101	11×10^{2}	34×10 ²		
5	20×10 ³	30×10 ³	57×10 ²	14×10 ²	CG		
No of samples	-	Bacterial counts of gut g ⁻¹					
	TVB	С	Group D FS	FS	Staphylococci		
1	88×10 ⁵	30×10 ⁵	<101	$< 10^{1}$	<101		
2	72×10 ⁵	38×10 ⁵	<101	26×10 ²	60×10 ²		
3	24×104	18×10 ³	<101	34×10 ²	44×10^{2}		
4	28×10 ⁵	12×10 ⁵	<101	42×10 ²	61×10 ²		
5	22×104	24×10 ³	20×101	33×10 ²	10×10^{2}		

TVB: total viable bacteria; CG: crowded growth; C: coliforms; Group D FS: Group D Faecal streptococci; FS: Faecal streptococci. Bacterial vounts are averages of triplicate determinations.

Where the occurrence was less than one log g⁻¹, the same projected as $< 1 \log g^{-1}$ owing to the experimental limitation that included the media employed, dilution factor, fastidious nature of some bacteria, pH, temperature, and size of inoculum. Fish is harvested from relatively cleaner environments, however, during subsequent handling spoilage and pathogenic bacteria come in contact with the fish (Chichester and Graham 1973; Prasad and Rao 1993). Hence, it is essential to screen the fish for spoilage and hygiene indicator bacteria.

Total viable bacteria

The TVB count in all fish samples varied three to four logs g^{-1} . In all the samples excepting the sample two the counts have decreased upon washing of the fish (Table 2). The TVB in gut of all the fish samples were higher by 1 to 2 log CFU g^{-1} in comparison with edible meat portion of the same sample. The fish flesh containing 8 log CFU g^{-1} TVB are considered unfit for human consumption (Alamas 1981) and the values in the present study is below the hazardous level.

Coliforms

Washing of the fish samples reduced the counts of coliforms and guts of all the

fish harboured high counts (Table 2). The presence of coliforms indicates level of hygiene of the product (Sahu 2006). Occurrence of coliforms in the guts could be due to feeding habits of the fish.

Group D faecal Streptococci

The counts of group D streptococci reduced from 1 to 2 log CFU g^{-1} in four out of five samples upon washing and the guts of the fish did not harbour this group of bacteria except in sample five where the occurrence is 200 CFU g^{-1} of the sample (Table 2).

Faecal streptococci

Washing of fish decreased number of streptococci. However, no significant difference is seen in their number in edible meat portion and in the guts (Table 2). The presence of faecal streptococci is hazardous to human health as they are implicated in infections of endocarditis in aged persons and pregnancy related problems in young women (Devriese 1992).

Staphylococci

The Staphylococcal count decreased from 1 to 2 log CFU g⁻¹ upon washing the fish (Table 2). The fish samples harboured more number of staphylococci in edible meat portion than in guts (Table 2). The studies showed that fish and fishery products are good sources for staphylococcal food poisoning (Sanjeev and Surendran 1996). The acceptable level of staphylococcal counts in fish product is 6 log CFU g⁻¹ and above (Bergdoll 1979) and in the present study the counts of staphylococci were below level of human health hazard.

Conclusions

The analysis of physicochemical quality parameters such as moisture, TVN, PV and α -Amino nitrogen of *Labeo calbasu* and *Labeo rohita* revealed that fishes were of good quality and on par with the quality of fishes of same varieties of other studies.

The raw fish *L. calbasu* and *L. rohita* scored an average of 9.75 and 9.2 in overall appearance, flesh quality, odour, and texture in the sensory evaluation by Hedonic scale indicating the excellent quality. And the cooked *L. calbasu* and *L. rohita* scored 8.58 and 7.9 out of a maximum of ten points, respectively, confirm that both the fishes were excellent in condition at the time of sale. The freshness test by using Intellectron-Fishtester VI showed that both the fishes were of excellent quality.

The bacteriological analysis before and after washing of the fish samples indicate reduction in the counts of different bacteria groups by 1 to 2 log CFU g^{-1} of the sample.

528

Variation in counts of different bacterial groups was also seen from edible meat portion to gut sample of the same variety of fish. In view of occurrence of hygiene indicator bacteria, more number of studies is necessitated especially in the absence of surveillance programs.

Acknowledgements

Authors wish to thank Dr. K Devadasan, Director, CIFT, Kochi, for permission to obtain guidance for M.Sc Dissertation work from Burla Research Centre of CIFT. M.M. Prasad, wishes to thank Director, CIFT, for permission to present the work at 8th Asian Fisheries Forum, Kochi.

References

- Alamas, A.K. 1981. Chemistry and microbiology of Fish and Fish Processing. Dept. of Biochemistry. Norweign Institute of Technology, University of Trontheim. Norway.
- Anon. 2004. CIFT (Central Institute of fisheries Technology) Annual Report-2003-2004. ISSN-0972-0667. Matsyapuri, P.O. Cochin 682 029.
- AOAC. 1995. Association of Official Analytical Chemists, 16th edn., Washington, D. C. USA.
- Bandhyopadhyay, J. K., A.K. Chattopadhaya and S.K.Bhattacharyya. 1985. Studies on the ice storage characteristics of commercially important fresh water fishes. In: Harvest and Post Harvest Technology of Fish. (ed. K. Ravindran, P. A. Perigreen, P. Madhavan, A. G. P. Gopalakrishnan, P. A. Panicker and Mary Thomas), pp381-

385. Society of Fisheries Technologists of India. Cochin, India.

Bergdoll, M. S. 1979. In: Food borne infections and intoxications (ed. H. Reiman and F.L. Bryan), pp 444. Academic Press, New York, USA.

Chichester, C. O and D. Graham. 1973. Microbial safety of Fishery Products. Academic Press, New York, AC. USA.

- Conway, E. J. 1947. Microdiffusion analysis and volumetric error. Cross by. Lock Wood and Sons. London. U. K.
- Devriese, L. A., M.D. Collins and R. Wirth. 1992. The Genus Enterococcus. In the Prokaryotes: A Handbook on the Biology of Bacteria, Eco-physiology, Isolation, Identification and Applications (ed. A.Balows, H.G. Truper, M. Warkin,
- W. Harder and K.H. Scheleifer), pp 465-1481. Springer-Verlog, New York.
- ICMSF. 1978. Microorganisms in Foods. 2nd Edition. International Commission on Microbiological Specifications for Foods. University of Toronto Press, Toronto, Canada.
- Jose Joseph. 2002. Present Status and future prospects of fresh water fish processing. In: Riverine and Reservoir Fisheries of India. Proceedings of the National Seminar on Riverine and Reservoir Fisheries – Challenges and Strategies, 23-24, May, 2001 (eds M. R. Bhoopendranath, B. Meenakumari, Jose Joseph, T. V. Shankar, P. Pravin and Leela Edwin), pp 281-287. Cochin, India.

Khanna, S. S. 1999. An Introduction to Fishes (3^{rd} Edn). Central Book Depot. Allahabad, India.

- Nair, P.GV and S. Mathew. 2000. Biochemical composition of fish & shell fish. CIFT. Technology Advisory Series. Central Institute of Fisheries Technology, Cochin 682029 (India). pp 14.
- Prasad, M. M and C.C.P.Rao. 1993. Food borne infections intoxications and their control. In: Proceedings of the

National Seminar on Health and Environment, JNTU, Kakinada, Andhra Pradesh, India. pp 89-95.

- Prasad, M. M and G. Seenayya. 1998. Fish as food in ancient India. Fishing Chimes 17 (10): 39-41.
- Prasad, M. M., J.K. Bandhyopadhyay and Prem Kumar. 2005. Biodiversity utilization and conservation in Hirakud reservoir and Mahanadi River System: Fresh water prawn scenario. In: Abstracts of the National Symposium on Environmental Biotechnology and Biodiversity Conservation pp 23.
- Sahu, J. 2004. Quality assessment of commercial Freshwater Fish: Labeo calbasu. M. Sc. Dissertation. G. M. College

(Auto), Sambalpur, Orissa, India.

- Sahoo, R.S. 2006. "Quality characteristics of *Labeo rohita* sold in Burla fish market, with special reference to antibiotic susceptibility of Staphylococcal isolates" M.Sc., Dissertation work submitted to the School of Life-sciences, Sambalpur University, Orissa, India.
- Sahu, J. 2006. Quality evaluation Commercial Smoke cured Freshwater Fish and Shellfish. M. Phil. Dissertation. G. M. College (Auto), Sambalpur, Orissa, India.
- Sanjeev, S and P.K. Surendran. 1996. Fate of enteropathogenic staphylococci in fish subjected to curing. Fishery Technology 33 (1): 66-68.
- Shasini, S. 2004. Bacteriological, Biochemical and Sensory characteristics of Commercial fresh water fish: *Labeo rohita*. M. Sc. Dissertation. G. M. College (Auto), Sambalpur, Orissa, India.
- Sugunan, V. V. 2002. Inland Fisheries enhancement, the prospects and paths ahead. ISFP Newsletter, Jul-Aug, 2002, 4 (3):1-14.
- Visweswara Rao, K. 1996. Biostatistics. A Manual of Statistical Methods for use in health, nutrition and anthropology. Jaypee Brothers Medical Publishers (P) Ltd. New Delhi.

Received: 29 December 2007; Accepted: 31 October 2008