Indian J. Fish., 62 (1): 101-109, 2015

Nutritional composition, product development, shelf-life evaluation and quality assessment of pacu *Piaractus brachypomus* (Cuvier, 1818)

L. N. MURTHY, B. MADHUSUDANA RAO, K. K. ASHA* AND M. M. PRASAD

Research Centre of Central Institute of Fisheries Technology, Ocean View Layout, Andhra University P. O. Visakhapatnam – 530 003, Andhra Pradesh, India

**Central Institute of Fisheries Technology, Cochin – 682 029, Kerala, India*

e-mail: bmrcift@rediffmail.com

ABSTRACT

Red-bellied pacu *Piaractus brachypomus* has a characteristic pomfret like shape and is marketed as freshwater pomfret. Pacu flesh had adequate amounts of protein (17%) and fat (1.19%). Essential amino acids formed 80.5% of the total amino acids of pacu meat. Histidine (32.55%), lysine (25.77%) and phenyl alanine (8.88%) were the predominant essential amino acids. Pacu meat was rich in unsaturated fatty acids (85.5%) of which mono unsaturated fatty acids (MUFA) and poly unsaturated fatty acids (PUFA) constituted 53% and 32.5%, respectively. The predominant MUFA, PUFA and SFA were palmitoleic acid-C16:1 (44.3%), α-linolenic acid-C18:3 (15.6%) and stearic acid-C18:0 (13.1%), respectively. Eicosapentaenoic acid (EPA) and docosa hexaenoic acid (DPA) were relatively low at 1.87%, each. During iced storage of whole and gutted fish, moisture content increased and fat content decreased with storage period. The TVBN, TMA, AAN, PV, FFA, APC, H_2S producing bacteria showed an increasing trend during iced storage. Chemical and microbiological analyses showed that, both whole and gutted pacu can be stored under iced condition for a period of 13 days. Fish fingers prepared using pacu meat and batter had a moisture and protein content of 59.47% and 12.59% respectively and stored well for 2 months in air packing and 3 months in vacuum packing at -20°C. Pacu meat and fingers could serve as nutritionally good food item, but for the presence of fine spines in the flesh.

Keywords: Fish fingers, Gutted, Iced storage, Nutritional composition, Pacu, *Piaractus brachypomus,* Shelf life

Introduction

The red-bellied pacu (*Piaractus brachypomus*) is a natural fish in many American countries and is well accepted by consumers (Wicki *et al*., 2008). *Piaractus brachypomus* Cuvier 1818 was originally introduced in India in West Bengal and is currently distributed in Andhra Pradesh, Kerala and North Eastern states (Singh and Lakra, 2011). Mukherjee *et al.* (2012) recommended pacu as a new candidate species for commercial aquaculture. Pacu is commonly grown in polyculture with Indian major carps at a ratio of 1 pacu : 3 carps and attain maturity above 3 years of age under a stocking density of 2,000-2,500 individuals ha^{-1} (Chattarjee and Mazumdar, 2009). Polyculture with striped catfish, *Pangasianodon hypophthalmus* has also been reported from Andhra Pradesh (Krishna *et al*., 2011). Pacu has a characteristic pomfret-like shape and is often referred to as red pomfret or freshwater pomfret (Fig. 1). Using triangle and simple ranking test, Pullela *et al.* (2000) observed that purged and processed skinless fillets of pacu had the highest preference compared to other aquacultured fish, such as channel catfish, rainbow trout, tilapia and hybrid striped bass. Tanamati *et al.* (2009) reported that cultivated pacu possessed relatively higher lipid content (12.2%) compared to wild species (7.9%) but the *n*-3 polyunsaturated fatty acid concentrations was higher in wild pacu (485.1 mg g^{-1} flesh) than in the cultivated species (106.1 mg g^{-1}). De Castro *et al.* (2007) reported that tambacu, a hybrid of tambaqui (*Colossoma macropomum*) and pacu (*Piaractus mesopotamicus*) had a total lipid content of 1.3%, with a fatty acid profile of 37% SFA, 34% MUFA and 21% PUFA.

Fig. 1. Red-bellied pacu (*Piaractus brachypomus*)

There is paucity of information on the nutritional profile of pacu cultured in India. The present work was envisaged to generate information on the nutritional profile of cultured pacu, and to study the changes during iced-storage of whole and gutted pacu and to evalute the shelf life of frozen battered and breaded pacu fish fingers.

Materials and methods

Fish

Fresh pacu were procured from local market and transported immediately in chilled condition (<4°C) to the laboratory for analysis. Average length of the fish was 24.34 cm and had a mean weight of 289.1 g. The fisheries were divided into two lots, (a) whole and (b) gutted and were packed into insulated polystyrene boxes with adequate amount of flake ice, to maintain a temperature of <4°C. Whole and gutted pacu were sampled immediately (day 0), and at regular pre-determined intervals (3, 6, 10 and 13 d) for chemical, microbiological and organoleptic analysis. The fish: ice ratio was 1:1 and ice quantity was maintained, considering the weight of fish being removed for sampling and the weight of melting ice drained. Sampling in triplicate was continued over a 13 day storage period.

Preparation of battered and breaded pacu fingers

Frozen pacu fish fillets were cut into fish fingers (15±1g) and pre-dusted. Pre-dusted pacu fingers were coated with an adhesive type quick setting batter (maida 2000 g, corn flour 200 g, Bengal gram 200 g, salt 30 g, guar gum 5 g, turmeric powder 5 g and sodium tri-polyphosphate 10 g) formulated at CIFT, Cochin (Joseph, 2009). One part of the batter powder was mixed with two parts of water to get the required consistency. The batter coated fillets (about 30 g) were further coated with bread crumbs, packaged in pouches either in air or under vacuum, frozen and stored at −20°C. Vacuum packing was done using quick seal machine (Sevana Electrical Appliances Pvt Ltd, Kerala, India). The pacu fingers were analysed for biochemical and microbiological parameters at monthly intervals.

Biochemical and microbiological analyses

Moisture, protein, fat, ash, calcium, potassium, sodium and iron were determined as per standard methods (AOAC, 1990). Phosphorus was determined colourimetrically (Fiske and Subbarow, 1925). Total volatile base nitrogen (TVBN) and Trimethyl amine (TMA) were determined by the Conway micro diffusion method (Conway, 1950). Peroxide value (PV) was determined iodometrically (AOAC, 1990) and free fatty acid (FFA) value was determined as per AOAC (1975). Alpha amino nitrogen (AAN) was estimated following Pope and Stevens (1939). Mercury content was estimated using mercury analyzer (MA5840, Electronic Corporation of India, Hyderabad) which works on the principle that mercury vapour (atoms) absorbs resonance radiation at 253.7 nm. Cadmium, copper, zinc and lead were analysed following AOAC (2000) using atomic absorption spectrophotometer (Varian Spectra AA 220, Australia). Aerobic plate count (APC), MPN total coliforms, MPN faecal coliforms, MPN *Escherichia coli*, coagulase positive *Staphylococci*, *Vibrio cholera*, *Listeria* and *Salmonella* were determined as per standard methods (BAM, 1995). Hydrogen sulphide producing bacteria were determined using peptone iron agar (Gram *et al.,* 1987). The data was subjected to statistical analysis using SPSS 16 for windows.

Amino acid and fatty acid analyses

Total lipid was extracted (Folch *et al.,* 1957) and fatty acids were analysed according to the method of Metcalfe (1966), using gas liquid chromatography. Amino acid profile of the meat was determined as per Ishida *et al*. (1981) using High Performance Liquid Chromatography (Shimadzu LC 10AS). Tryptophan was determined spectrophotometrically as per the method of Sastry and Tammuru (1985).

Organoleptic analysis

Sensory characteristics mainly appearance, texture, odour, taste and colour were evaluated by five panelists. Scoring was done on a nine-point hedonic scale (Amerine *et al*., 1965), where score 9, is 'like extremely' and score 1 is 'dislike extremely'. Pacu meat was boiled in 1.5% NaCl solution for 10 min and presented for organoleptic evaluation.

Results and discussion

Proximate and mineral composition

The proximate composition of pacu flesh (Table 1) indicated moisture content of 80.86% and adequate amounts of protein (17%) and fat (1.19%). Ramos *et al.* (2008) reported 18% protein content in pacu (*Piaractus mesopotamicus*) flesh. The protein content of pacu was found to be lower than that of cultured Indian and exotic carps (Chakrabarti and Rao, 2008) but slightly higher than tilapia (Murthy *et al*., 2008). The fat content of pacu recorded in the present study was 1.19% which was similar to that of Indian major carps (Chakrabarti and Rao, 2008). Tanamati *et al.* (2009) reported 12.2% fat in cultured pacu and 7.9% in wild pacu from Brazil.

Table 1. Proximate and mineral composition of cultured pacu *P. brachypomus*

Parameters	Mean ± SD
Moisture (%)	80.86±0.22
Protein (%)	17.00±0.00
Fat (%)	1.19±0.005
Ash (%)	1.19±0.65
Sodium (mg%)	963±155
Potassium (mg%)	774±160
Calcium (mg%)	2110±55.2
Iron (ppm)	106±9.7
Phosphorus (mg%)	1650±89

* Results are average of triplicate determinations

Fatty acid content and composition also varies with the anatomical location of the sample used for anlysis (Kinsella *et al.,* 1977).

Amino acid profile

Pacu meat was found to be a rich source of essential amino acids (Table 2). Essential amino acids formed 80.5% of the total amino acids of pacu meat. Histidine (32.55%), lysine (25.77%) and phenyl alanine (8.88%) were the predominant essential amino acids which together constituted 67.2% of the total amino acid content of pacu. Glycine (9.34%) was the dominant non-essential amino acid. The amino acids arginine, cystine, proline, tryptophan and tyrosine were totally absent. Suseela (2009) reported that the meat of rohu and mrigal constituted higher proportion of glutamic acid and lysine. Mohanty and Kaushik (1991) reported that in the flesh of rohu, catla and mrigal fry; glutamic acid, aspartic acid and glycine were the dominant non-essential amino acids and phenyl alanine, lysine and leucine were the dominant essential amino acids.

Table 2. Amino acid profile of *P. brachypomus* (g 100 g $^{-1}$)

Amino acid	mg 100 g $^{-1}$	Percentage
Aspartic acid	449.953	3.71
Threonine*	506.427	4.18
Serine	237.346	1.96
Glutamic acid	482.731	3.98
Proline	0	0
Glycine	1132.285	9.34
Alanine	62.042	0.51
Cystine	0	o
Valine*	301.393	2.49
Methionine*	85.826	0.71
Isoleucine*	319.431	2.64
Leucine*	398.226	3.29
Tyrosine	0	0
Phenyl alanine*	1076.651	8.88
Histidine*	3944.572	32.55
Lysine*	3.122869	25.77
Arginine*	0	0
Tryptophan*	0	0

* Essential amino acids

Fatty acid composition of pacu

Pacu meat is rich in unsaturated fatty acids (85.5%) (Table 3). Of the total fatty acids, mono unsaturated fatty acids (MUFA) and poly unsaturated fatty acids (PUFA) constituted 53% and 32.5%, respectively. Saturated fatty acids (SFA) were found to be low (14.5%). The predominant MUFA, PUFA and SFA were palmitoleic acid-C16:1 (44.3%), α-linolenic acid-C18:3 (15.6%) and stearic acid-C18:0 (13.1%), respectively. The fatty acid profile of tambacu (hybrid of tambaqui *C. macropomum* and pacu *P. mesopotamicus*) was reported to be 37% saturated, 34% monounsaturated and 21% polyunsaturated (De Castro *et al*., 2007). Kinsella *et al.* (1977) analysed 18 species of freshwater fish fillets and observed that palmitic, palmitoleic, oleic, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DPA) were the most abundant fatty acids. Rao *et al.* (2012) reported that myristic acid (325.3 mg%) was the most prominent SFA and oleic acid (816.8 mg%) was the dominant MUFA in hilsa (*Tenualosa ilisha*) from river Godavari. Ramos *et al.* (2008) observed that in pacu (*P. mesopotamicus*), oleic acid is the predominant fatty acid, followed by palmitic acid and stearic acid. The difference in the levels of different fatty acids in pacu analysed in this study and other reported values may be attributed to the environment and type of feed, as these are known to influence the fatty acid composition of fish (Suzuki *et al.,* 1986; Moreira *et al.,* 2001).

Eicosapentaenoic acid (EPA) and docosahexaenoic acid (DPA) were relatively low in pacu meat at 1.87% and 1.87%, respectively (Table 3). Similar observation was made in several other studies. Ramos *et al*. (2008) reported that pacu had lower contents of ω-3 family fatty acids (1.13%). Gutierrez and da Silva (1993) reported that most freshwater fish from Brazil were found to be poor source of EPA and DHA. The fatty acid composition of pacu is strongly influenced by habitat and diet. Tanamati

Table 3. Fatty acid composition of pacu *P. brachypomus*

Fatty acid number	Common name	%*
C14:1	Myristooleic acid	4.37
C16:1	Palmitoleic acid	44.32
C17	Margaric acid	0.13
C18	Stearic acid	13.11
C18:3	α-linolenic acid (ALA)	15.61
C20	Arachidic acid	1.25
C20:3	Diomo- γ-linolenic acid	11.85
C20:4	Arachidonic acid	1.25
C20:5	Eicosa pentaenoic acid (EPA)	1.87
C22:6	Docosa hexaenoic acid (DHA)	1.87
C24:1	Nervonic Acid	4.37

* expressed as percentage of total fatty acid

et al. (2009) noticed that the *n*-3 polyunsaturated fatty acid concentrations were higher in wild pacu than in the cultivated strains.

Organoleptic characteristics of pacu

The organoleptic scoring on a 9-point scale was performed and the results are given in Table 4. Texture (8.13) scored the maximum. All other parameters scored more than 7 (like moderately) and the overall acceptability score was 8 (like very much) suggesting that pacu has good acceptance.

Table 4. Organoleptic evaluation of pacu

Organoleptic characteristics	Mean±SD
Appearance	7.88±0.23
Odour	7.5±0.19
Taste	7.13±0.13
Colour	7.63±0.26
Texture	8.13±0.23
Overall acceptability	8

Heavy metals in pacu

Cadmium, zinc and lead were detected in the edible part, skin, eye, liver and intestine of pacu (Table 5). Copper was detected only in the edible part and skin. Cadmium level was high in skin (3.04 ppm). Zinc content was highest in the intestine (54.3 ppm). Lead content was relatively higher in liver (0.81ppm) and intestine (0.66 ppm). Cadmium level in the pacu meat (0.77 ppm) exceeded the acceptable limit of 0.05 ppm, set by European Union whereas lead and mercury content in pacu meat were within acceptable limits (EC, 2005). Cd content of 0.75 ppm has been reported in frozen threadfin bream from the east coast of India (Prasad *et al.*, 2007).

Microbiological quality of pacu

Fresh pacu had an APC of 1,80,000 cfu g^{-1}. Total coliforms, faecal coliforms and *E. coli* were not detected (<3 MPN g^{-1}). Human pathogens *viz*., *Salmonella*, *Listeria*, *Vibrio cholera* and coagulase positive Staphylococci were not detected. H_2S producing bacteria were not detected indicating freshness. The results indicate that pacu meet the microbiological quality criteria of EIC (1995).

Changes in biochemical quality during iced storage of whole and gutted pacu

During iced storage of whole and gutted fish, moisture content increased and fat content decreased with storage period (Table 6). The increase in moisture was relatively higher in gutted fish and reached 84.55% at the end of 13 days of iced storage whereas in whole fish the moisture content reached 82.95% at the end of 13 days. Biochemical changes occurring during iced storage of whole and gutted pacu are presented in Table 7. The TVBN, TMA, AAN, PV and FFA showed an increasing trend during iced storage. Quality deterioration was relatively faster in gutted pacu. The increase in TVBN, TMA, PV and FFA was always higher in gutted pacu whereas the increase in AAN was slightly higher in whole pacu. Chintu *et al.* (2010) reported that freshwater fish *Labeo rohita* showed maximum NPN content of 17.1% of the total nitrogen, which was significantly higher compared to that of *Catla catla* and *Hypophthalmichthys molitrix*. *Etroplus suratensis* collected from both freshwater and brackishwater environments also showed significant difference (Chintu *et al.*, 2010). In whole pacu, the TVBN increased from 7.42 mg% to 19.97 mg%, while in gutted pacu the TVBN increased to 23.23 mg%

Table 5. Heavy metals in different body parts of pacu (Mean ±SD)

Heavy metals	Skin	Edible part	Eye	Liver	Intestine
Cadmium (ppm)	3.04 ± 0.13^{c}	0.77 ± 0.11^{b}	0.06 ± 0.01^{a}	0.07 ± 0.01^{a}	0.13 ± 0.01^{a}
Copper (ppm)	1.68 ± 0.1^{b}	3.5 ± 0.22^{c}	0^{a}	0^{a}	0^{a}
Zinc (ppm)	8.94 ± 0.28^{a}	12.67 ± 0.01^{a}	8.37 ± 0.28^{a}	2.83 ± 0.21^{a}	54.30 ± 10.65^{b}
Lead (ppm)	0.25 ± 0.02^{b}	0.04 ± 0.01^{a}	0.06 ± 0.01^{a}	0.81 ± 0.01^{d}	0.66 ± 0.02^{c}
Mercury (ppm)	0	0	0	0	0

Values bearing different superscripts in a row differ significantly ($p<0.5$)

Table 6. Changes in whole and gutted pacu during iced storage (Mean ±SD)

Parameter	Day 0	Day 3		Day 6		Day 10		Day 13	
	Whole	Whole	Gutted	Whole	Gutted	Whole	Gutted	Whole	Gutted
Moisture (%)	80.86 $\pm0.22^{a}$	80.89 $\pm0.11^{a}$	82.16 $\pm0.95^{b}$	82.52 $\pm0.05^{bc}$	82.51 $\pm0.28^{bc}$	82.45 $\pm0.33^{bc}$	83.55 $\pm0.23^{cd}$	82.95 $\pm0.83^{bc}$	84.55 $\pm0.24^{d}$
Protein (%)	17.00 $\pm0.00^{d}$	16.95 $\pm0.15^{d}$	16.55 $\pm0.25^{cd}$	16.50 $\pm0.3^{cd}$	16.50 $\pm0.3^{cd}$	16.15 $\pm0.05^{c}$	16.05 $\pm0.05^{bc}$	15.5 $\pm0.3^{ab}$	15.15 $\pm0.0.05^{a}$
Fat (%)	1.19 $\pm0.005^{ef}$	1.07 $\pm0.06^{de}$	1.02 $\pm0.65^{d}$	1.09 $\pm0.00^{de}$	0.98 $\pm0.01^{d}$	0.8 $\pm0.06^{c}$	0.66 $\pm0.01^{bc}$	0.62 $\pm0.42^{ab}$	0.48 $\pm0.03^{a}$

Values with different superscripts in a row differ significantly ($p<0.5$)

Table 7. Changes in biochemical parameters during iced storage of whole and gutted pacu (Mean ±SD)

Parameters	Day 0	Day 3		Day 6		Day 10		Day 13	
	Whole	Whole	Gutted	Whole	Gutted	Whole	Gutted	Whole	Gutted
TVBN (mg%)	7.42±0.5 [a]	8.55±0.29 [ab]	9.18±0.4 [b]	12.02±0.11 [c]	13.28±0.19 [d]	16.47±0.38 [e]	18.09±0.24 [f]	19.97±0.14 [g]	23.23±0.65 [h]
TMA (mg%)	5.52±0.41[a]	6.5±0.38[b]	6.91±0.08[b]	8.97±0.04[c]	9.81±0.19[c]	11.10±0.22[d]	12.44±0.43[e]	14.45±0.36[f]	16.66±0.34[g]
AAN(mg%)	42.54±0.61 [b]	46.53±0.48 [de]	43.95±.14 [bc]	46.85±0.51 [de]	45.25±1.1 [cd]	48.12±0.5 [ef]	46.84±.13 [de]	49.86±0.3 [f]	49.14±.57 [f]
PV(meq kg fat^{-1})	1.03±0.02 [a]	3.57±0.01 [c]	4.39±0.14 [d]	4.29±0.18 [d]	5.36±0.08 [e]	5.27±0.01 [e]	5.86±0.06 [f]	6.22±0.1 [f]	7.05±0.2 [g]
FFA (% of Oleic acid)	1.16±.06 [a]	9.63±0.23 [b]	11.28±0.33 [d]	9.9±0.04 [b]	13.08±0.08 [e]	10.59±0.02 [c]	14.32±0.22 [f]	11.4±0.12 [d]	15.15±0.11 [g]

Value with different superscripts in a row differ significantly ($p<0.5$)

during iced storage for 13 days. In whole pacu, the TMA increased from 5.52 mg% to 14.45 mg% but in gutted pacu the TMA increased to 16.66 mg% during iced storage for 13 days. Chintu *et al.* (2010) reported TMAO content of 11.25g% in *E. suratensis* collected from freshwater environment. Freshwater species *Micropterus salmonoides, Oreochromis mossambicus* and *Siniperca chuatsi* were found to contain TMAO in the range of 510-760, 85-720 and 400-640 mg kg^{-1} respectively (Chung *et al*., 2009). Chintu *et al.* (2010) reported AAN content in *C. catla, L. rohita* and *H. molitrix* as 26.25 g%, 28.52 g% and 24.01 g% respectively. Similarly PV increased from 1.03 to 6.22 meq kg fat^{-1} in whole pacu whereas in gutted pacu the PV increased to 7.05 meq kg fat^{-1}. FFA increased from 1.16 to 11.4% of oleic acid in whole pacu while FFA increased to 15.15% of oleic acid in gutted pacu, during iced storage for 13 days. However, the TVBN and PV values were within acceptable limits (Connell, 1975; EC, 1995) suggesting that, both whole and gutted pacu can be stored under iced condition for a minimum period of 13 days. Leitao *et al*. (1997) studied the shelf life and spoilage of pacu (*P. mesopotamicus*) during refrigerated storage at 5°C and noticed that pacu was highly stable during storage, with no evidence of deterioration after 14 days based on sensory, chemical or microbiological changes. Joseph *et al*. (1988) reported that rohu had an iced shelf life of 16 days, mrigal 15 days and catla 17 days. Surendran *et al*. (1989) reported that pearl spot and tilapia had shelf life of two weeks in ice. Gopal *et al*. (1990) found that catla fillet had an iced shelf life of 12 days. Manju *et al*. (2007) reported a shelf life of 8 days for air packaged gutted pearl spot during chilled storage. Chitra *et al*. (2008) reported that organoleptic scores of rohu fillets decreased from initial value of 9.17 to 6.17 after 14 days at 4 - 6°C and from 9.0 to 6.0 after 14 days at 1 - 2°C storage**.** Based on odour scores, Goulas and Kontominas (2007) reported that chill stored raw chub mackerel fillets remained acceptable up to 11 days. Papadopoulos *et al*. (2003) observed that TVBN values of gutted seabass were higher than whole fish stored in ice. The shelf life studies indicate that cultured pacu can be transported as whole fish to different markets in the country under iced condition.

Changes in the microbiological quality of whole and gutted pacu during iced storage

APC of whole and gutted fish decreased during the first three days of iced storage and increased thereafter (Fig. 2). The initial decrease and subsequent increase in APC was greater in gutted pacu. The APC crossed the acceptable limit (EIC, 1995) on the 13th day of iced storage in whole (2.56 x 10^6 cfu g^{-1}) and gutted (3.4 x 10^6 cfu g^{-1}) pacu. Hydrogen sulphide (H_2S) producing bacteria showed an increasing trend during iced storage and crossed 1000 cfu g^{-1} by the end of 13 days of iced storage in gutted and whole pacu (Fig. 2). Hydrogen sulphide producing bacteria tolerate low temperatures and are extremely important in spoilage of proteinaceous foods during chilled storage. Barile *et al*. (1985) observed that

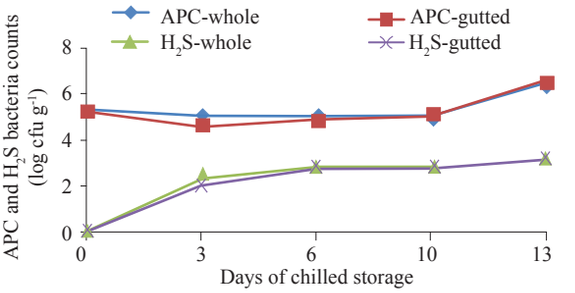

Fig. 2. Changes in the microbiological quality of whole and gutted pacu during iced storage
APC: Aerobic plate count, (log cfu g^{-1}); H_2S: Hydrogen sulphide producing bacteria (log cfu g^{-1}.)

on harvesting, mackerel had a predominantly mesophilic bacterial population but during iced storage psychrotrophs predominated the spoilage flora. Psychrotrophs can grow at 0 to 7°C even though they have optima between 20 and 30°C (Prescott *et al.*, 2005). Hydrogen sulphide producing bacteria accounted for 3 and 29% of the total plate count during storage of haddock at 1°C at day 0 and day 10 (Chai *et al.,* 1968). Rao and Khasim (2009) suggested that H_2S producing bacteria count of more than 3 log cfu g^{-1} appears to be the separating line between freshness and loss of freshness in the freshwater fish, rohu. Based on

microbiological parameters, spoilage appears to be relatively greater in gutted pacu compared to whole pacu during the later stages of iced storage. *E. coli*, *V. cholerae*, *Listeria*, *Salmonella*, and coagulase positive Staphylococci were not detected during iced storage of whole and gutted pacu. Papaopoulos *et al.* (2003) reported that pseudomonads and H_2S producing bacteria were the dominant bacteria at the end of the 16 day storage period in ice for both whole ungutted and gutted seabass. Mesophilic counts for gutted and ungutted fish exceeded 7 log cfu g^{-1} after 9 and 15 days of ice storage, respectively. Rao and Khasim (2009) observed a sharp increase in H_2S producing bacteria counts during the intermediate stage of spoilage of rohu stored at ambient temperature. Manju *et al.* (2007) noticed a continuous increase in APC which reached about 10^7 cfu g^{-1} on the 10^{th} day of chilled storage of pearl spot when the fish were deemed spoiled based on sensory scores.

Nutritional composition of pacu finger

Frozen fingers are products meant for relatively long term preservation. The pacu fingers had a moisture content of 59.47%, protein content of 12.59% and had appreciable levels of sodium, potassium, calcium and phosphorus (Table 8). Pacu fingers may serve as nutritionally good food item but due to the presence of fine spines in the meat, it could be preferred by carp consumers.

Table 8. Nutritional composition of pacu fingers

Parameters	Mean ± SD
Moisture (%)	59.47±0.29
Protein (%)	12.59±0.28
Fat (%)	5.21±0.15
Ash (%)	22.72±0.41
Sodium (mg%)	1390±26.25
Potassium (mg%)	454±8.85
Calcium (mg%)	167±0.6
Iron (ppm)	74.28±0.76
Phosphorus (mg%)	717±2.45

Changes in the biochemical quality of air and vacuum packaged frozen pacu fingers

The TVBN, TMA, PV and FFA showed an increasing trend during the three months of frozen storage of pacu fingers. Quality deterioration was relatively slower in vacuum packaged pacu fingers. The increase in TVBN, TMA, PV and FFA was always higher in air packaged pacu fingers (Table 9). In air packaged pacu fingers, the TVBN increased from 19.2 mg% to 29.15 mg%, while in vacuum packaged pacu fingers the TVBN increased to 27.39 mg% during frozen storage for 3 months. PV increased from 8.05 to 11.71 meq kg fat^{-1} in air packaged pacu fingers whereas in vacuum packaged pacu the PV increased to 8.08 meq kg fat^{-1}. However, the TVBN and PV values were within acceptable limits (Connell, 1975), both in air and vacuum packaging even at the end of three months of frozen storage. Results confirm the protective effect of vacuum packaging on fat. Vacuum packaging is widely used in food industry due to its effectiveness in reducing oxidative reactions in the products (Gopal *et al*., 1999). Devadasan *et al.* (1978) compared the frozen storage characteristics of six species of freshwater fishes and reported that carps had slightly better storage life than freshwater catfishes. Chitra *et al.* (2008) reported that TVBN content of rohu fillets increased from 75.17-94.73 mg% after 14 days at 4 - 6°C storage and 44.17-94.30 mg% after 14 days of storage at 1 - 2°C. Shahrooz and Abbas (2012) observed that freshness parameters *viz*., FFA, PV and TBA values increased marginally during frozen storage of vacuum packaged farmed cobia fillets. Nurhan (2007) revealed significant effects of cooking methods on proximate, amino acid and fatty acid contents of rainbow trout, but observed lack of negative influence of the cooking processes on the amino acid composition of rainbow trout fillets.

Changes in the microbiological quality of frozen pacu fingers

APC showed gradual reduction till the third month both in air and vacuum packaged pacu fingers (Fig. 3) and all values were less than the maximum permissible level of 5,00,000 cfu g^{-1} (EIC, 1995). Faecal coliforms were detected during the first two months of frozen storage, both in air and vacuum packaged products (Table 10). *E.coli* was not detected in vacuum packaged fingers but was detected in air packaged pacu fingers till the end of 2 months of frozen storage but at a low level (3.6 MPN g^{-1}). *V. cholerae*, *Listeria*, *Salmonella* and coagulase positive Staphylococci were not detected; both in freshly prepared and frozen stored pacu fingers during frozen storage. Microbiological quality of pacu fingers was good and the quality of vacuum packaged fingers was relatively better.

Table 9. Changes in biochemical parameters of pacu fingers during frozen storage period (Mean ±SD)

Parameters	Fresh	1 Month		2 Months		3 Months	
	Air	Air	Vacuum	Air	Vacuum	Air	Vacuum
TVBN (mg%)	19.20±.32 ^{b}	23.08±.44 ^{c}	19.66±.14 ^{b}	27.59±.63 ^{d}	27.19±.46 ^{d}	29.15±.27 ^{d}	27.39±1.16 ^{d}
TMA (mg%)	13.07±0.75 ^{b}	14.40±.17 ^{b}	13.20±.32 ^{b}	19.57±.34 ^{d}	17.07±.75 ^{c}	20.02±.09 ^{d}	17.03±.41 ^{c}
PV (meq kg fat^{-1})	8.05±0.45 ^{a}	9.64±0.6 ^{bc}	8.21±0.05 ^{a}	10.21±0.01 ^{c}	8.62±0.03 ^{ab}	11.71±0.5 ^{d}	8.08±0.03 ^{a}

Value with different superscripts in a row differ significantly ($p<0.5$)

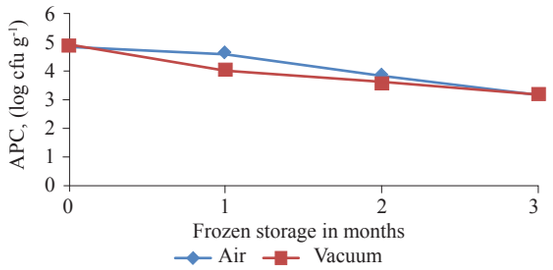

Fig. 3. Changes in Aerobic plate count of pacu fish fingers during frozen storage

It has been well documented that bacteria grow more quickly in fish stored in air compared to that stored under vacuum at 0°C (Leung *et al*., 1992; Lyon and Reddmann, 2000; Ozugul *et al*., 2004; Manju *et al*., 2007).

Red-bellied pacu (*P. brachypomus*) cultured in freshwater, had adequate amounts of protein, higher proportion of essential amino acids and unsaturated fatty acids. Chemical and microbiological results show that, both whole and gutted pacu can be stored under iced condition for a period of 13 days. Pacu fish fingers stored well for a minimum period of three months under air/vacuum packaging at -20°C. Pacu meat and fingers may serve as nutritionally good food item but due to fine spines in the flesh, it may be better preferred by carp consumers.

Table 10. Changes in counts of faecal indicator bacteria in pacu fish fingers during frozen storage

Storage period	Faecal coliforms (MPN g^{-1})		*E. coli* (MPN g^{-1})	
	Air	Vacuum	Air	Vacuum
1 month	15	3.6	3.6	<3
2 month	9.2	3.6	3.6	<3
3 month	<3	<3	<3	<3

Acknowledgements

We express our sincere thanks to Dr T. K. Srinivasa Gopal, Director, CIFT, Cochin for the encouragement. Technical assistance rendered by K.V. S.S.S.K. Harnath, B. K. Panda, A. K. Panigrahi, N. Venkata Rao, P. Radha Krishna, Prasanna Kumar, S. N. Disri, G. Bhushanam and technical staff of Biochemistry and Nutrition Division, CIFT, Cochin is gratefully acknowledged.

References

AOAC 1975. *Official methods of analysis,* 12th edn. Association of Official Analytical Chemists, AOAC International,Washington DC, USA.

AOAC 1990. *Official methods of analysis,* 15th edn. Association of Official Analytical Chemists, AOAC International, Washington DC, USA

AOAC 2000. *Official methods of analysis,* 17th edn. Association of Official Analytical Chemists, AOAC International, Washington DC, USA.

Amerine, M. A., Pongborn, R. H. and Roescler, E. B. 1965. *Principles of sensory evaluation of food*. Academic Press, New York, 602 pp.

BAM 1995. *Bacteriological analytical manual.* Food and Drugs Administration, 8th edn, AOAC International, USA.

Barile, L. E., Mille, A. D., Reilly, A. and Villadsen, A. 1985. Spoilage pattern of mackerel (*Rastrelliger faughni*) 2. Mesophilic and psychrophilic spoilage. Spoilage of tropical fish and product development. *FAO Fisheries Report. No. 317 supplement*, FAO, Rome, p. 146-149.

Chai, T., Chen, C., Rosen, A. and Levin, R. E. 1968. Detection and incidence of specific species of spoilage bacteria in fish. II. Relative incidence of *Pseudomonas putrifaciens* and fluorescent pseudomonads in haddock fillets. *Appl. Environ. Microbiol*., 16: 1738-1741.

Chakrabarti, R. and Rao, B. M. 2008. The suitability of muscle of *Cirrhinus mrigala* in the formation of gel: a comparative electrophoretic study of six tropical carp meats. *Intl. J. Food Sci. Technol.*, 43: 179-184.

Chattarjee, N. R. and Mazumdar, B. 2009. Induced breeding of pacu (*Piaractus brachypomus*) in captivity with pituitary extract. *Aquacult. Asia*, 14: 23.

Chintu, M. R., Suseela Mathew and Mathew, M. 2010. Comparative evaluation of non-protein nitrogenous compounds in fishes of fresh and brackishwater environments. *Fish. Technol.,* 47: 161-166.

Chitra, C. P., Ganesh, K., Vasudev, V., Sridhar, N., Hemaprasanth, K. and Raghunath M. R 2008. Efficacy of sanitisers on the shelf life of rohu steaks stored at two refrigeration temperatures. *Fish. Technol.,* 45: 197-208.

Connell, J. J. 1975. *Control of fish quality*. Fishing News Books, Ltd., Surrey, England, p. 136- 137.

Conway, F. 1950. *Microdiffusion and analysis and volumetric error*, 3rd edn., Crosby Lockwood and Sons, London, p. 87-107.

Chung, S. W. C and Chan, B. T. P. 2009. Trimethylamine oxide, dimethylamine, trimethylamine and formaldehyde levels in main traded fish species in Hong Kong. *Food Addit. Contam. Part B Surveill*., 2: 44-51.

De Castro, F. A. F., Sant Ana, H. M. P., Campos, F. M., Costa, N. M. B., Silva, M. T. C., Salaro, A. L. and Franceschini, S. C. C. 2007. Fatty acid composition of three freshwater fishes under different storage and cooking processes. *Food Chem.*, 103: 1080-1090.

Devadasan, K., Varma, P. R. G. and Venkataraman, R. 1978. Studies on frozen storage characteristics of fillets of six species of freshwater fishes. *Fish Technol.*, 15: 1-6.

EC 1995. Commission decision 95/149/EEC of 8th March 1995 relating to total volatile basic nitrogen (TVB-N) limit values for certain categories of fishery products and specifying the analysis methods to be used. *Official J. Eur. Communities*, L 97: 84-87.

EC 2005. Commission regulation (EC) NO 78/2005 of Jan 2005 amending regulation No. 466/2001 as regards heavy metals. *Official J. Eur. Union*, L 16, p. 43-45.

EIC 1995. *Quality assurance and monitoring system manual of export of fresh, frozen processed fish and fishery products*, Export Inspection Council of India, GOI, New Delhi.

Fiske, C. H. and Subbarow, Y. 1925. The colorimetric determination of phosphorus. *J. Biol. Chem.*, 66: 375-400.

Folch, J. and Less, Stanley, H. A. 1957. A simple method for isolation and purification of total lipids from animal tissues. *J. Biol. Chem.,* 226: 497-499.

Gopakumar, K. 1997. *Biochemical composition of Indian food fish.* Central Institute of Fisheries Technology, Cochin, p. 1-44.

Gopal, T. K. S., Nambiar, V. N., Bhattacharya, S. K., Joseph, J. and Prabhu, P. V. 1990. *Chilling and freezing*, New Fish Products, IIF-IIR Commission C2, Aberdeen, UK.

Gopal, T. K. S., Joseph, J. and Balachandran, K. K. 1999. Development of fish products employing hurdle technology. In : *Preservation of food by hurdle technology*. DFRL. Mysore, p. 93-103.

Goulas, A. E. and Kontominas, M. G. 2007. Effect of modified atmosphere packaging and vacuum packaging on the shelf-life of refrigerated chub mackerel (*Scomber japonicus*): biochemical and sensory attributes. *Eur. Food Res. Technol.*, 224: 545-553.

Gram, L., Trolle, G. and Huss, H. H. 1987. Detection of specific spoilage bacteria from fish stored at low (0°C) and high (20°C) temperatures. *Intl. J. Food Microbiol.*, 4: 65-72.

Gutierrez, L. E. and da Silva, R. C. M. 1993. Fatty acid composition of commercially important fish from *Brazil. Sci. Agric. Piracicaba.*, 50: 478-483.

Ishida, Y., Fujita, T. and Arai, K. 1981. New detection and separation method for amino acid by high performance liquid chromatography. *J. Chromatogr.,* 204: 143-148.

Joseph, A. C. 2009. Coated products from freshwater fish and shellfish. In: Joseph, J., Ravishankar, C. N., Zynudheen, A. A., Bindu, J., Ninan, G. and Mohan, C. O. (Eds.). *Post harvest technology of freshwater fish.* Central Institute of Fisheries Technology, Cochin.

Joseph, J., Surendran, P. K. and Perigreen, P. A. 1988. Studies on the iced storage of cultured rohu (*Labeo rohita*). *Fish. Technol.*, 25: 105.

Joseph, J. 2002. Present status and future prospects of freshwater fish processing. In: Boopendranath, M. R., Meenakumari, B., Joseph, J., Sankar, T. V., Pravin, P. and Edwin, L. (Eds.), *Riverine and reservoir fisheries of India.* Society of Fisheries Technologists (India), Cochin, p. 281-287.

Kinsella, J. E., Shimp, J. L., Mai, J. and Weihrauch, J. 1977. Fattyacid content and composition of freshwater fish. *J. Am. Oil Chem. Soc.*, 54: 424-429.

Krishna, R. R., Rao, K. V. P. and Reddy, D. M. 2011. The princess of aquaculture and the plights of the fish farmers. *Aquacult. Asia.*, XVI: 12-16.

Leitao, M. F. F., Rios, D. P. F. A., Guimaraes, J. G. L., Baldini, V. L. S. and Mainardes-Pinto, C. S. R. 1997. Chemical and microbiological changes in pacu (*Piaractus mesopotamicus*) stored under refrigeration at 5°C. *Cienc. Tecnol. Alimentos,* 17: 160–166.

Leung, C. K., Huang, Y. W. and Harrison, M. A. 1992. Fate of *Listeria monocytogenes* and *Aeromonas hydrophila* on packaged channel catfish fillets stored at 4°C. *J. Food Prot.*, 45: 440-444.

Lyon, W. J. and Reddmann, C. S. 2000. Bacteria associated with processed crawfish and potential toxin production by *Clostridium botulinum* type E in vacuum packaged and aerobically packaged crawfish tails. *J. Food Prot.*, 63: 1687-1696.

Manju, S., Jose, L., Srinivasa Gopal, T. K., Ravishankar, C. N. and Lalitha, K. V. 2007. Effects of sodium acetae dip treatment and vacuum-packaging on chemical, microbiological, textural and sensory changes of pearl spot (*Etroplus suratensis*) during chill storage. *Food Chem.*, 102: 27-35.

Metcalfe, L. D., Schimitz, A. A., Petha, J. R. 1966. Rapid determination of fatty acid esters from lipids for gas chromatographic analysis. *Anal. Chem.*, 38: 514-515.

Mohanty, S. S. and Kaushik, S. J. 1991. Whole body amino acid composition of Indian major carps and its significance. *Aquat. Living Resour.,* 4: 61-64.

Moreira, A. B., Visentainer, J. V., Souza, N. E. and Matsushita, M. 2001. Fattyacids profile and cholesterol contents of three Brazilian *Brycon* freshwater fishes. *J. Food Comp. Anal.*, 14: 565–574.

Mukherjee, M., Das, S. and Borah, A. K. 2012. Successful breeding and seed production of pacu (*Piaractus brachypomus*) in captivity. *Fishing Chimes,* 32: 57-60.

Murthy, L. N., Panda, S. K. and Shamasundar, B. A. 2008. Physicochemical and functional properties of proteins of tilapia (*Oreochromis mossambicus*). *J. Food Process. Eng.,* 34: 83–107.

Nurhan, U. 2007. Change in proximate, amino acid and fatty acid contents in muscle tissue of rainbow trout (*Oncorhynchus mykiss*) after cooking. *Intl. J. Food Sci. Technol.*, 42: 1087–1093.

Ozugul, F., Polat, A. and Ozugul, Y. 2004. The effects of modified atmosphere packaging and vacuum packaging on chemical, sensory and microbiological changes of sardines (*Sardinella pilchardus*). *Food Chem.*, 85: 49-57.

Papadopoulos, V., Chouliara, I., Badeka, A., Savvaidis, I. N. and Kontominas, M. G. 2003. Effect of gutting on microbiological, chemical, and sensory properties of aquacultured seabass (*Dicentrarchus labrax*) stored in ice. *Food Microbiol.*, 20: 411-420.

Pope, C. G. and Stevens, M. E. 1939. The determination of amino nitrogen using copper method. *Biochem. J*., 33: 1070-1077.

Prasad, M. M., Khasim, D. I, Bandyapadhyay, J. K and Gupta, S. S. 2007. Bacteriological quality and heavy metals in edible meat portion of Japanese threadfinbream (*Nemipterus japonicus*). *J. Food Sci. Technol.*, 44: 319-322.

Prescott, L. M., Harley, J. P. and Klein, D. A. 2005. *Microbiology*, 6th edn., McGraw-Hill, New York, p. 119-124.

Pullela, S. V., Fernandes, C. F., Flick, G. J., Libey, G. S., Smith, S. A. and Coale, C. W. 2000. Quality comparison of aquacultured pacu (*Piaractus mesopotamicus*) fillets with other aquacultured fish fillets using subjective and objective sensorial traits. *J. Aquat. Food Prod. Technol.*, 9: 65-76.

Ramos Filho, M. M., Ramos, I. L., Hiane, P. A. and Souza, E. M. T. 2008. Lipid profile of four species of fish from the pantanal region of Mato Grosso do Sul. *Cienc. Tecnol. Alimentos,* 28: 361-365.

Rao, B. M, and Khasim, D. I. 2009. Hydrogen sulphide producing bacteria as indicators of spoilage of freshwater rohu (*Labeo rohita*) fish. *J. Food Sci. Technol*., 46: 377-379.

Rao, B. M., Murthy, L. N., Suseela Mathew, Asha, K. K., Sankar, T. V. and Prasad, M. M. 2012. Changes in the nutritional profile of Godavari hilsa shad, *Tenualosa ilisha* (Hamilton, 1822) during its anadromous migration from Bay of Bengal to the River Godavari. *Indian J. Fish.*, 59: 125-132.

Sastry, C. S. P. and Tammuru, M. K. 1985. Spectrophotometric determination of tryptophan in protein. *J. Food Sci. Technol.*, 22: 146-147.

Singh, A. K. and Lakra, W. S. 2011. Risk and benefit assessment of alien fish species of the aquaculture and aquarium trade into India. *Rev. Aquacult.,* 3: 3-18.

Shahrooz, T. and Abbas A. M. 2012. Influence of vacuum packaging and long term storage on some quality parameters of cobia (*Rachycentron canadum*) fillets during frozen storage. *Am. Eurasian J. Agric. Environ. Sci.,* 12: 541-547.

Surendran, P. K., Joseph, J., Shenoy, A.V., Perigreen, P. A., Iyer, K. M. and Gopakumar, K. 1989. Studies of spoilage of commercially important tropical fishes under iced storage. *Fish. Res.*, 7: 1-9.

Suseela, M. 2009. Biochemical composition of freshwater fish in comparison to marine and brackishwater. In: Joseph, J., Ravishankar, C. N., Zynudheen, A. A., Bindu, J., Ninan, G. and Mohan, C. O. (Eds.), *Post harvest technology of freshwater fish*. Central Institute of Fisheries Technology, Cochin.

Suzuki, H., Okazahi, K., Hayakama, S., Wada, S. and Tamura, S. 1986. Influence of commercial dietary fattyacids on polyunsaturated fattyacids of cultured freshwater fish and comparison with those of wild fish of the same species. *J. Agric. Food Chem.*, 34: 58–60.

Tanamati, A., Stevanato, F. B., Visentainer, J. E. L., Matsushita, M., de Souza, N. E. and Visentainer, J. V. 2009. Fatty acid composition in wild and cultivated pacu and pintado fish. *Eur. J. Lipid Sci. Tech.,* 111: 183-187.

Wicki, G., Rossi, F., Merino, O. and Luchini, L. 2008. Optimising production of pacu through polyculture. *INFOPESCA International*, 34: 29-35.

Date of Receipt : 14.11.2012
Date of Acceptance : 05.08.2014