Indian J. Fish., 71 (4): 109-114, 2024
doi:10.21077/ijf.2024.71.4.136115-14
Available online at: epubs.icar.org.in/index.php/IJF

Meat quality assessment of common carp (*Cyprinus carpio*) reared in inland saline water

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Abstract

The study was conducted to assess the meat quality of inland saline water reared common carp (ISWCC) with those compared with freshwater reared common carp (FWCC). Among the proximate parameters, protein, fat and ash contents showed significant differences ($p<0.05$) while the moisture content was non-significant ($p>0.05$). Potassium was the dominant mineral in both ISWCC (841 mg 100 g^{-1}) and FWCC (1114 mg 100 g^{-1}). Sodium was found higher in ISWCC (245 mg 100 g^{-1}) than in FWCC (137 mg 100 g^{-1}). The major amino acids in ISWCC were glycine, alanine, glutamic acid, and aspartic acid. Whereas glycine, lysine, alanine and histidine were the abundant amino acids recorded in FWCC. The polyunsaturated fatty acid content of ISWCC was 22.29% and FWCC was 34.55%. Therefore, it can be concluded that though there are slight variations recorded in the biochemical composition and nutrient profiling of ISWCC as compared with FWCC, taste and overall meat quality are not affected due to rearing in inland saline water (ISW) since most of the essential amino acids and fatty acids were recorded in ISWCC meat.

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Keywords:
Common carp, Inland saline water, Meat quality, Nutrient profile

Received : 08.05.2023
Accepted : 16.12.2024

Introduction

Inland soil salinity is one of the serious global issues and is a major environmental concern in many countries, posing a threat to profitable agricultural businesses, vulnerable habitats, important infrastructures, roads and buildings. There are approximately 1000 million ha of salt-affected soil spread over the world. In India, the issue of soil salinity has critically affected about 8.62 million ha of agricultural land. About 40% of India's inland saline soils are present in Haryana, Punjab, Rajasthan and Uttar Pradesh (Allan *et al*., 2009). These saline lands could be utilised for aquaculture practices to generate revenue from unused or underutilised resources. Hence, these lands have been converted into aquaculture ponds for farming practices called inland saline aquaculture (ISA). It is an adaptive solution to this environmental problem.

In 2020, global fish production was estimated to be around 178 million t. Of the total production, 37% (66 million t) was harvested in inland waters (83% from aquaculture and 17% from capture fisheries). The growth of aquaculture over the past few decades has contributed to an overall increase in productivity in inland waters. Aquaculture accounted for 49% (88 million t) of total production. The global demand for seafood is increasing, yet due to other land and water use activities, coastal aquaculture expansion is constrained in many locations. Inland aquaculture already contributes the most to overall aquaculture which is about 62.2% of total aquaculture production (SOFIA, 2022); however, ISA also has the potential to boost aquaculture productivity. Common carp is the world's third most widely introduced fish species. It is being investigated as a potential commercial aquaculture candidate in Asia and certain European nations due to its excellent adaptability to both environment and diet (Rahman, 2015). In 2020, common carp ranked fourth in terms of global finfish aquaculture production, providing 8.6% (4.24 million t) of total global finfish inland aquaculture production which is about 49.12 million t (SOFIA, 2022). It is one of the best candidate species for ISA as it can withstand moderate fluctuations in salinity and temperature (Whiterod and

Walker, 2006). However, fishes reared in an environment away from their original rearing conditions will be biologically stressed which may lead to changes in their meat quality parameters. There are no studies about the effect of salinity on common carp fish meat and the information on the fish meat quality is important for both the producers and consumers. Hence, this study was carried out to evaluate the meat quality parameters of common carp reared in ISW.

Materials and methods

Raw materials

Common carp cultured in ISW with a salinity of 4 ppt was collected and transported to the laboratory within 5 h from the Rohtak District of Haryana, India by Air cargo. The fish had an average weight of 450 to 500 g and a length of 28 to 30 cm. Fresh common carp reared in freshwater culture ponds was procured from a fish market in Dadar, Mumbai. The fish was immediately iced in icebox with fish:ice ratio of 1:1 (w/w) and transferred to the laboratory within one hour of procuring. The average size of fish was 497.33±2.52 g in weight and 28.56±1.20 cm in length.

Proximate composition

AOAC (2005) methods were used to determine the proximate composition of the fish meat. A hot air oven was used to measure the sample's moisture content. Inside a hot air oven adjusted to 100±2°C, the pre-weighed samples were left to dry for 16-18 h. The reduction in the weight of the samples after drying was measured and the moisture content was evaluated. The samples after drying were used for further proximate analysis. Crude protein was computed by multiplying the calculated nitrogen value using the Kjeldahl apparatus (Kelplus-KES12L VAI, Pelican, India) with the conversion factor of 6.25 (N x 6.25). The crude fat content was determined using soxhlet apparatus (EXPO Hi-TECH i-therm AL-7941, Mumbai, India) and the ash content by charring and incinerating the samples at a temperature around 600^0C in a muffle furnace (EXPO HI-TECH, i-therm AL-7941, Mumbai, India) for 6 h. The final values obtained were converted on wet weight basis.

Texture profile analysis

Texture profile analysis (TPA) on fish was carried out by using a texture analyser (Texan touch, Lamy Rheology, France) with a stainless-steel cylindrical probe of 50 mm diameter and 50 kg load cell. The entire fish was kept on the base plate and compressed twice by a probe connected to the drive system. Three measurements were done above the lateral line on both sides of the fish at 25, 50 and 75% of the total length. The sample was compressed to 30% of its original height at a deformation rate of 1 mm s^{-1}, with the down speed set to 1mm s^{-1}, the force to start set to 2.5 N, the wait position set to 5 mm, the delay set to 5 s and the up-speed set to 1 mm s^{-1}.

Sensory analysis

The non-trained panel members (n=10), who included staff and students from the Department of Post-Harvest Technology of ICAR-Central Institute of Fisheries Education (ICAR-CIFE), Mumbai, carried out the sensory evaluation. A 9-point hedonic scale was used to perform the sensory study (Meilgaard *et al*., 1999). The scale has scores ranging from 1 to 9, with 1 being the lowest (not recommended) and 9 being the highest score (highly acceptable). The sensory parameters studied were appearance, colour, odour, texture, flavour, taste and overall acceptability. The samples were cooked in steam for 10 min and provided to the panel members to test the taste of fish meat.

Amino acid profile analysis

High-resolution liquid chromatograph mass spectrometer (HR-LCMS) was used to analyse the amino acids according to the method given by Pattanaik *et al*. (2020). In vacuum-sealed tubes, samples (50 mg) were hydrolysed in 5 ml 6N HCl for 24 h at 110°C. The 6N HCl was evaporated after hydrolysis, leaving a 200 µl residue. 5 µl was collected from the residue and diluted with 500 µl of distilled water. The sampler used had a draw and eject speed of 100 µl min^{-1} and a 20 µl injection capacity. It was a Hi P sampler (G4226A). The flow rate was kept at 1.5 ml min^{-1} using a binary pump (G4220B, Agilent Technologies, Santa Clara, CA, USA), with a high-pressure limit of 1200 bar.

Fatty acid profile analysis

Fish muscle weighing 5 g was macerated in a 30 ml solution of a chloroform: methanol (2:1) ratio. To remove fat from the muscle, the Folch technique was employed (Folch, 1957). The extracted fat was collected in a flask with a flat bottom, and it was dried off by rotating vacuum evaporation at 55°C. Using the AOAC (1995) method, the dried lipid was collected and transformed into fatty acid methyl ester (FAME). The methylated fatty acids were separated using a GC-MS (QP2010, Shimadzu, USA) equipped with a DB wax capillary column (30 m 0.25 mm internal diameter 0.25 m film thickness) with helium as the carrier gas. A temperature of 250°C was chosen for the injector and detector. The injection was carried out using a 1 µl FAME injection volume in split mode (1:15). For two minutes, the initial column temperature was held at 50°C. It was intended for the temperature to increase by 10°C every minute until it reached 230°C and then allowed to stay there for 35 min. FAME was separated at a constant pressure of 82.5 Kpa. The peaks were located by comparing the mass spectra to the mass spectral database.

Mineral profile analysis

Mineral content was assessed using an inductively coupled plasma atomic emission spectrophotometer (ICP-AES: Model Thermo Electron IRIS INTREPID II XSP DUO, Germany). Concentrated HNO_3 was used to homogenise the dried fish sample and the homogenate was digested in a microwave digester (Milestone, Shelton, Italy). The processed sample was inhaled into the flame, where the distinctive radiation absorption by each element was recorded. The mineral concentration was determined and represented in mg 100 g^{-1} of sample.

Statistical analysis

The SPSS 16.0 statistical program was used to statistically evaluate the results (SPSS, 2000). The statistical significance

($p \leq 0.05$) among the triplicates was determined for each of the above experiments using Duncan's multiple range tests. The results were expressed as mean±standard deviation.

Results and discussion

Comparative proximate analysis between ISWCC and FWCC

Table 1 indicates the proximate composition of ISWCC and FWCC. From the results, it was found that ISWCC had a significantly higher ($p<0.05$) protein content (16.34%) than FWCC (15.55%). However, the ash content in FWCC (1.35%) was significantly higher ($p<0.05$) compared to ISWCC (0.97%). The fat content of FWCC (2.02%) was similarly found to be significantly higher ($p<0.05$) than that of ISWCC (1.86%). The moisture content of the samples did not show significant difference ($p>0.05$). Since the composition of fish meat varies substantially depending on age, sex, size, growth stage, diet, environment and season, the proximate composition of fish meat varies greatly from one species and one individual to another (Venugopal and Shahidi, 1996). The results of the present study were closely related to the findings of Dembele *et al*. (2010). Kumar *et al*. (2016) reported that as the salinity increased, the percentage of protein content increased with a declining trend in ash content which is similar to the findings of this study. The findings of the current study reveal that the protein content of common carp fish meat is not affected by salinity. The reduction in the fat content of ISWCC indicated a higher energy expenditure in maintaining osmoregulation. Ohta and Watanabe (1996) also recorded a reduction in lipid content with increasing salinity. The present investigation indicates that the proximate composition of *C. carpio* varies with increased salinity in ISW.

Comparative texture profile analysis between ISWCC and FWCC

Since many fish species lack a distinct flavour, texture becomes extremely vital for consumer acceptance and it is one of the most significant quality factors of fish (Hyldig and Nielsen, 2001). The texture is influenced by various intrinsic and extrinsic factors (Barroso *et al*., 1998). The textural parameters of ISWCC and FWCC meats are shown in Table 2. The hardness value was observed significantly higher ($p<0.05$) in FWCC (10.71) than in ISWCC (8.03). The cohesiveness was found to be significantly higher ($p<0.05$) in ISWCC (1.44) than in FWCC (0.96). The elasticity value in FWCC (1.92) was significantly higher ($p<0.05$) compared to ISWCC (1.69). The slight variations in the textural properties could be attributed to the method of fish culture, which can impact the texture of fish flesh (Jobling, 2001). The variations in the textural parameters between ISWCC and FWCC may be due to myosin, an important component in the texture profile quality assessment which can alter the hardness level in the fish (Ingolfsdottir, 1997).

Table 1. Comparative proximate analysis between ISWCC and FWCC (wet weight basis)

Parameters (%)	ISWCC	FWCC
Moisture	80.34 ± 0.13^a	80.92 ± 0.52^a
Protein	16.34 ± 0.09^a	15.55 ± 0.05^b
Fat	1.86 ± 0.01^a	2.02 ± 0.05^b
Ash	0.97 ± 0.03^a	1.35 ± 0.03^b

† ISWCC-Inland saline water reared common carp, FWCC- Freshwater reared common carp
‡ Data expressed as mean±SD (n=3), the mean value in the same row with different superscripts are significantly different. *$p<0.05$

Table 2. Comparative texture profile analysis between ISWCC and FWCC

Parameters	ISWCC	FWCC
Hardness	8.03 ± 0.60^a	10.71 ± 0.32^b
Cohesiveness	1.44 ± 0.19^a	0.96 ± 0.03^b
Elasticity	1.69 ± 0.12^a	1.92 ± 0.30^a

† ISWCC-Inland saline water reared common carp, FWCC- Freshwater reared common carp
‡ Data expressed as mean± SD (n=3), the mean value in the same row with different superscripts are significantly different. *$p<0.05$

Comparative sensory scores of ISWCC and FWCC

Comparative sensory scores of ISWCC and FWCC are given in Table 3. According to the findings, all the sensory parameters in both samples received excellent scores, implying that the raw material used in the study was quite fresh. There was no significant difference ($p>0.05$) found in the sensory parameters between ISWCC and FWCC. Geosmin and 2-methylisoborneol are primarily responsible for the "earthy" and "musty" off-flavours, respectively detected in fish (Kumari *et al*., 2021). Further, it can be attributed that culturing common carp in ISW will not increase the incidence of geosmin and 2-methylisoborneol related off-flavours in common carp. According to the findings of this study, common carp can be cultivated in salt-affected areas without affecting their sensory properties.

Comparative mineral profiles of ISWCC and FWCC

Minerals, which are made up of the ash left over after biological components have been entirely burnt or oxidised, are recognised to be necessary for cellular metabolism in all animals, including fish. Table 4 presents the mineral profile of ISWCC and FWCC. The mineral content of ISWCC was found in the following order Cu<Zn<Fe<Mg<Ca<Na<P<K, whereas the mineral content of FWCC was observed in the order Zn<Fe<Mg<Na<P<Ca<K. The total mineral content was higher in FWCC meat (2156.5 mg 100 g^{-1}) than in ISWCC meat (1978.24 mg 100 g^{-1}). It correlates with the higher ash content observed in the meat of FWCC than in ISWCC meat. Since the minerals can be obtained by the fish from both their diet and surrounding water, variations in the mineral profile were observed

Table 3. Comparative sensory scores of ISWCC and FWCC

Parameters	ISWCC	FWCC
Appearance	9.00 ± 0.00^a	8.93 ± 0.12^a
Colour	8.96 ± 0.10^a	8.84 ± 0.20^a
Texture	9.00 ± 0.00^a	8.75 ± 0.27^a
Odour	8.95 ± 0.10^a	8.93 ± 0.12^a
Taste	8.88 ± 0.21^a	9.00 ± 0.00^a
Flavour	8.84 ± 0.20^a	8.97 ± 0.08^a
Overall acceptability	8.96 ± 0.10^a	8.84 ± 0.20^a

† ISWCC-Inland saline water reared common carp, FWCC-Freshwater reared common carp
‡ Data expressed as mean± SD (n=8), the mean value in the same row with different superscripts are significantly different. *$p<0.05$

Table 4. Comparative mineral profiles of ISWCC and FWCC

Minerals	ISWCC (mg 100 g^{-1})	FWCC (mg 100 g^{-1})
Potassium	841	1114
Phosphorus	756	290
Sodium	245	137
Calcium	69	432
Magnesium	61	109
Iron	4.3	72
Zinc	1.9	2.5
Copper	0.04	ND
Total	1978.24	2156.5

† ISWCC-Inland saline water reared common carp, FWCC- Fresh water reared common carp
ND – Less than 0.01 ppm

between ISWCC and FWCC meats. Potassium (K) was found to be the most abundant mineral in both ISWCC (841 mg 100 g^{-1}) and FWCC (1114 mg 100 g^{-1}) meat. Schaeffer *et al*. (2012) also recorded higher content of K in the meat of common carp. FWCC is having a higher content of both K and Mg than ISWCC. Reduced level of Mg and K in ISWCC than FWCC is due to the mineral deficiency in the ionic profiles of ISW. ISW is deficient in K and Mg (Roy *et al*., 2007). The concentration of most ions in ISW bodies is similar to or greater than those of marine water, except for K (Prangnell and Fotedar, 2005). ISWCC had higher sodium (Na) content (245 mg 100 g^{-1}) than FWCC (137 mg 100 g^{-1}) due to the high content of Na in ISW than in freshwater. Therefore, it is concluded that the level of aqueous minerals has been correlated to the level of minerals in fish meat.

Comparative amino acid profiles of ISWCC and FWCC

The amino acid (AA) profiles of ISWCC and FWCC are provided in Table 5. Both samples included a total of 15 AAs, of which nine were essential amino acids (EAA), which are crucial for maintaining human health. EAAs cannot be made by the human body; therefore, people need to receive them from a variety of sources, including fish meat and are vital for wound healing (Jais *et al*., 1994). From the results, it was found that the abundant AAs in ISWCC were glycine, alanine, glutamic acid and aspartic acid. Whereas glycine, lysine, alanine and histidine were the abundant AAs recorded in FWCC. These AAs constituted more than 50% of the total AAs recorded in the samples. Glycine was present in a higher amount in both ISWCC (18.87%) and FWCC (20.49%). Kadhim *et al*. (2019) also recorded a higher amount of glycine than other AAs in common carp. Depending upon the factors such as location, seasons, size of fish, food, age and reproductive status, the types and concentrations of AAs in fish tissues will vary (Limin *et al*., 2006). In this study, the contents of aspartic acid and glutamic acid were found to increase in ISWCC than in FWCC. Umami AAs, such as aspartic acid and glutamic acid are free AAs that are recognised to impart umami flavour, which influences the taste and quality of the meat, as well as consumer preference (Liyana-Pathirana *et al*., 2002). According to this result, increased aquaculture salinity can improve the flavour of common carp fish meat. These findings imply that ISW mostly changed the number of free AAs in common carp meat, which changed the flavour more than the nutritional value of meat.

Table 5. Comparative amino acid (%) profiles of ISWCC and FWCC

Amino acids	ISWCC	FWCC
Alanine*	13.01	11.62
Arginine **	5.21	7.73
Aspartic acid*	9.49	1.37
Glutamic acid*	12.75	4.81
Glycine *	18.87	20.49
Histidine**	2.14	9.21
Isoleucine **	2.97	2.01
Leucine **	6.45	8.82
Lysine**	7.64	12.41
Methionine**	1.33	4.48
Phenylalanine**	2.84	2.67
Serine *	6.49	5.04
Threonine **	4.83	4.29
Tyrosine*	2.10	3.02
Valine **	3.88	2.02

† ISWCC-Inland saline water reared common carp, FWCC- Freshwater reared common carp
**- Essential amino acid, *- non-essential amino acid

Table 6. Comparative fatty acid profiles of ISWCC and FWCC

Fatty acid	Name	ISWCC	FWCC
C 12:0	Lauric acid	0.14±0.00^a	0.34±0.01^b
C14:0	Myristic acid	1.82±0.05^a	4.40±0.02^b
C15:0	Pentadecyclic acid	0.35±0.01^a	1.01±0.80^b
C16:0	Palmitic acid	20.50±0.10^a	15.26±0.27^b
C17:0	Margaric acid	0.32±0.00	-
C18:0	Stearic acid	5.75±0.19^a	5.94±0.09^a
SFA		28.88	26.95
C16:1 (n-7)	Palmitoleic acid	7.43±0.47^a	10.72±0.38^b
C18:1(n-9)	Oleic acid	35.27±0.16^a	19.44±0.30^b
C20:1(n-9)	Gondoic acid	5.63±0.03^a	7.60±0.71^b
MUFA		48.33	37.76
C18:2 (n-6)	Linoleic acid	12.75±0.03^a	10.72±0.44^b
C20:4 (n-6)	Arachidonic acid	3.67±0.02^a	4.33±0.52^b
C22:4 (n-6)	Adrenic acid	0.68±0.03^a	0.87±0.09^a
C20:2 (n-6)	Dihomo-γ-linolenic acid	1.47±0.01^a	1.68±0.08^b
C20:5 (n-3)	Eicosapentaenoic acid	0.67±0.01^a	3.12±0.29^b
C22:6(n-3)	Docosahexaenoic acid	2.42±0.00^a	3.83±0.57^b
C18:3 (n-3)	α- Linolenic acid	0.95±0.0^a	4.87±0.00^b
C18:4 (n-3)	Stearidonic acid	0.17±0.00^a	1.24±0.00^b
C22:5 (n-3)	Docosapentaenoic acid	1.51±0.02^a	2.49±0.45^b
C18:2 (n-7)	Rumenic acid	-	1.40±0.02
PUFA		22.29	34.55
Miscellaneous		0.50	0.74
Total		100	100

† ISWCC-Inland saline water reared common carp, FWCC- Freshwater reared common carp
SFA-Saturated fatty acid, MUFA-Monounsaturated fatty acid, PUFA-Polyunsaturated fatty acid
‡ Data expressed as mean±SD (n=3), the mean value in the same row with different superscripts are significantly different. *$p<0.05$

Comparative fatty acid (%) profiles of ISWCC and FWCC

Table 6 represents the fatty acid (FA) profiles of ISWCC and FWCC. From the results, monounsaturated fatty acids (MUFAs) were found as the major FAs in both ISWCC (48.33%) and FWCC (37.76%). Among the MUFAs, oleic acid was abundant in both ISWCC (35.27%) and FWCC (19.44%). All vertebrates, including fish and humans, require polyunsaturated fatty acid (PUFA) for appropriate growth, development and reproduction. Since vertebrates cannot synthesise n-3 or n-6 FAs, they must be obtained through diet (Tenyang *et al*., 2020). The total lipid content and FA levels in fish are greatly dependent on the diet consumed (Memon *et al*., 2011). The PUFA content was higher in FWCC (34.55%) than in ISWCC (22.29%). There were marked differences found between ISWCC and FWCC regarding the individual PUFA content. The largest differences were recorded in the contents of EPA and α-linolenic acid with FWCC characterised by greatly increased amounts of both the FAs compared with ISWCC. Among the PUFAs, linoleic acid which is an essential FA generally required in the diets of freshwater fish was present in a higher amount in both ISWCC (12.75%) and FWCC (10.72%). EPA and DHA contents were 0.67 and 2.42% in ISWCC and 3.12 and 3.83% in FWCC fat samples respectively. The decreased levels of long-chain PUFAs (EPA and DHA) in ISWCC than FWCC might be due to less FA synthesis from 18-carbon FAs like linoleic acid and α-linolenic acid. It may be inferred that in freshwater conditions, the production of long-chain PUFAs from 18-carbon FA precursors in common carp meat was substantially active, but not in inland saline conditions.

According to research, the two most helpful omega-3 FAs are EPA and DHA, which have a variety of health benefits. These include lowering myocardial infarction risk, lowering blood pressure and lipid levels in the blood, boosting the immune system and maintaining appropriate brain function in humans (Swanson *et al*., 2012). The percentage of EPA+DHA found in this study for ISWCC (3.09%) and FWCC (6.95%) was higher than this reported by Skalecki *et al*. (2016) in their work, where the EPA + DHA percentage in common carp was 2.72%. The lesser content of arachidonic acid in ISWCC is related to its physiological functions. Arachidonic acid is a key precursor for eicosanoids (leukotrienes, prostaglandins, and thromboxanes), which are formed in response to environmental stress (Tocher, 2003). In this study, it was discovered that ISW conditions did not provide environmental stress on the FA synthesis in freshwater originating common carp since they did not need to raise the amount of arachidonic acid in their body to cope with the stress of living in ISW.

The baseline information generated in this study indicates that the body composition of *C. carpio* varies with the increased salinity in ISW. However, the mineral, AA and FA compositions were found to be comparable, with only minor differences in protein, fat and ash contents and textural parameters between ISWCC and FWCC were recorded. These variations indicate that rearing common carp in ISW has an impact on the body composition, which could be associated to the physiological adaptation of the fish to salinity. The presence of most of the essential AAs and FAs and high content of protein indicate that common carp gives better quality finished products when reared in ISW. The outcome of this research will be useful for fish farmers as well as fish processors in the way that the fish farmers can produce common carp by utilising salt-affected inland areas and processors can use the meat to produce different value-added products as the meat quality is not affected by the inland saline environment.

Acknowledgments

We sincerely acknowledge Director, ICAR-CIFE, Mumbai, for providing financial support and facilities for undertaking this study.

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