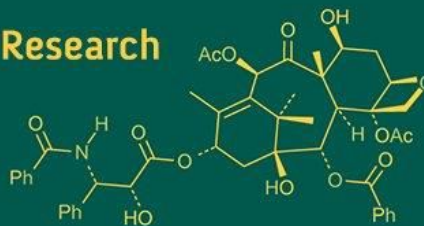


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Effect of refrigerated storage on the biochemical, microbial and functional properties of processed amur common carp (*Cyprinus carpio haematopterus*) meat products

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Abstract

This study investigated the biochemical, microbial, and functional stability of four processed meat products from Amur common carp (*Cyprinus carpio haematopterus*) i.e., minced meat (ACMM), surimi (ACS), flesh powder (ACFP), and high-protein surimi (HPAS) which was studied over 28 days of refrigerated storage. Significant ($p < 0.05$) increases were observed in spoilage indicators, including total volatile base nitrogen (TVBN), peroxide value (PV), thiobarbituric acid (TBA), free fatty acids (FFA), and total plate count (TPC). At the end of the 28-day storage period, ACMM exhibited the highest levels of TVBN (13.11 mg/100g), peroxide value (38.78 meq/kg), and free fatty acids (3.95%), indicating advanced spoilage. In contrast, HPAS demonstrated the greatest stability with the lowest values of TVBN (7.81 mg/100g), PV (2.91 meq/kg), and FFA (1.01%), reflecting the effectiveness of alkaline washing and cryoprotectant treatment. Functional quality indicators such as salt-soluble nitrogen (SSN) and non-protein nitrogen (NPN) declined across all samples, with HPAS retaining higher protein solubility. Microbial loads increased over time, but remained within acceptable limits, with ACFP exhibiting the lowest TPC due to its reduced water activity. These results underscore the superior shelf life and quality of HPAS and its potential for use in value-added fish product development.

Keywords: Refrigerated storage, amur common carp, surimi, biochemical changes, lipid oxidation, microbial stability

Introduction

India has witnessed a significant transformation in its fisheries sector over the past two decades. Inland fish production increased remarkably from 2.823 million metric tons (MMT) in 2000-01 to 13.11 MMT in 2023-24, marking nearly a 400% rise (DAHDF, 2023) ^[1]. This expansion is largely attributed to the rapid growth of aquaculture, which involves the cultivation and harvesting of fish in freshwater ecosystems such as lakes, ponds, rivers, and reservoirs. Between 2014 -15 and 2021-22, the sector recorded an impressive average annual growth rate of 8.61%. Inland fisheries now account for 75% of India's total fish production, while marine fisheries contribute the remaining 25%, reaffirming the strategic importance of inland aquaculture to national fish output. With an extensive inland water surface area of 314,400 km², India ranks among the top countries globally in terms of inland fishery resources (DAHDF, 2023) ^[1].

In recent years, attention has turned to the development of value-added fishery products such as surimi, a refined fish protein concentrates widely used in imitation seafood. Traditionally, marine fish like Alaska pollock and threadfin bream have been the preferred sources for surimi. However, with increasing raw material costs and supply limitations, the focus has shifted toward freshwater carps as promising alternatives due to their high availability, low production cost, and favourable processing characteristics. Several Asian carp species, including grass carp, silver carp, bighead carp, and black carp, are currently being explored for surimi production, particularly in China and Southeast Asia. Research by Endoo and Yongsawatdigul (2014) ^[2] highlighted the gel-forming potential of Asian carps,

although studies on their gelation properties compared to marine species remain limited. In India, Indian major carps such as *Catla catla*, *Labeo rohita*, and *Cirrhinus mrigala* dominate inland aquaculture due to their adaptability to low-input systems. These species contributed approximately 5.4 MMT to the country's inland fish production in 2023 (FAO, 2013) [3]. However, emerging carp species like the Amur common carp (*Cyprinus carpio haematopterus*) are gaining recognition for their superior growth performance and economic potential. Introduced by Karnataka Veterinary, Animal and Fisheries Sciences University (KVAFSU), Bidar, the Amur carp is a genetically improved strain of common carp characterized by 27% faster growth, delayed sexual maturation, greater feed conversion efficiency, and lower disease susceptibility (Vijayakumar *et al.*, 2020) [4]. Field trials across various ecological zones, including hilly areas like Meghalaya, demonstrated consistent growth and adaptability (Das, 2017; Sanjay Kumar, 2017) [5, 6]. India's total fish production reached 18.402 MMT in 2023-24, a substantial increase from 9.579 MMT in 2013-14. Inland fisheries contributed 13.9 MMT to this total, with Karnataka alone producing 1.225 MMT of carp in 2023 (GoI PIB, 2025) [7]. While specific production data for Amur carp is not yet officially reported, its cultivation is expanding, particularly in Karnataka's seasonal water bodies. The processing qualities of Amur carp make it especially suitable for surimi production. Its firm, white flesh is rich in myofibrillar proteins essential for gelation and textural integrity in surimi products. The low fat and oil content reduce the risk of lipid oxidation, enhancing shelf life and sensory quality. Its mild flavor allows for versatility in surimi-based applications. Moreover, the Amur carp's resilience and compatibility with sustainable farming practices make it an environment friendly choice. Lingaraju *et al.*, 2025 [8] noted that Amur carp combines economic viability, processing efficiency, and desirable sensory traits, making it a superior raw material for large-scale surimi production. Given these attributes, the Amur carp holds significant promise for enhancing the sustainability and profitability of India's value-added fish product sector. The aim of this study is to evaluate the biochemical, microbial, and functional changes in processed Amur common carp meat products during refrigerated storage. It investigates how storage duration affects quality parameters such as lipid oxidation, protein stability, and microbial load across various surimi-based formulations.

Materials and Methods

Farm raised freshwater Amur common carp was procured from a local fish farm located around Karnataka, India. The average length and weight of fish used in this study was measuring a standard-length of 35.24 cm, and average weight of fish 836.34 gm respectively.

Surimi Preparation

The fish was dressed, and fillets were separated from the both sides of the fish using a sharp knife. The fillets were fed to the reciprocating type meat picking machine. Picked meat was used in 4 batches. One batch of Picked meat (unwashed) was used as control (Amur Carp Minced Meat-ACMM). Minced meat was prepared from picked meat, mixed with cryoprotectants at the rate of sugar 4%, sorbitol 4%, sodium polyphosphate 0.2% and ascorbic acid 0.5%. Fish meat was packed in polyethylene bags (500g) frozen at

-35 °C by Plate freezer for ninety minutes and stored at -20 °C (Amur Carp Surimi-ACS). A part of minced meat was transferred to a bowl chopper, mixed and ground with dilute alkali solution sodium bicarbonate at rate of 0.2%, and washed thoroughly in chilled water, dewatered to prepare High protein amur surimi (HPAS). HPAS was mixed with cryoprotectants, packed and stored at -20° C. The last batch of minced meat was converted to Amur Carp Flesh Powder (ACFP) by suitable modification (Majewska *et al.*, 2025) [9] and stored.

Biochemical Analysis

Biochemical quality indicators were also evaluated during refrigerated storage. The pH of fish meat was measured using a digital pH meter after homogenizing 5 g of sample with 45 ml of distilled water (AOAC, 2010) [10]. Total volatile base nitrogen (TVB-N), which reflects the degree of spoilage, was determined by Conway's micro-diffusion technique (Conway, 1962) [11]. Peroxide value (PV), indicative of lipid peroxidation, was estimated by extracting fat with chloroform, followed by reaction with potassium iodide and titration with sodium thiosulphate (Jacobs, 1951) [12]. Free fatty acid (FFA) content was determined by titrating lipid extracts with sodium hydroxide using phenolphthalein as an indicator and was expressed as a percentage of oleic acid (Pearson, 1976) [13]. All analyses were conducted in triplicate, and results were expressed as mean ± standard deviation.

Microbiological Analysis

Total Plate Count (TPA)

The total plate count was determined according to the procedure described in APHA (2000) [14]. Enumeration of bacterial load was done using plate count agar (Hi-media Laboratories, Mumbai) by following spread plate technique. About 25 g of the sample prepared from Amur carp minced meat, surimi, flesh powder and high protein amur surimi sample was homogenized aseptically with 225 ml of sterile physiological saline (0.85% NaCl) for 5 min in a sterilized pestle and mortar. Appropriate dilutions were made from the homogenate, and 0.1 ml was placed on sterile total plate count (TPC) agar plates. The plates were incubated at 37 °C for 24 to 48 hours. The number of colonies were counted and expressed as number of colony forming units (CFU) per gram of sample.

Functional Characteristics

Estimation of non-protein nitrogen (NPN)

Non-protein nitrogen content was determined by the method as described by Velankar and Govindan (1958) [15]. About 3.0 g of meat sample was macerated with 10 ml of 20% TCA for 5 minutes using a pestle and mortar. The slurry was allowed to settle at refrigerated temperature (6-80 °C) for 30 min. The slurry was filtered and made up to 50 ml with distilled water and an aliquot of 5 ml was taken for nitrogen estimation by Kjeldahl method. The NPN content was expressed as mg N/100 g meat.

Protein Solubility

Solubility of proteins in extraction buffer (Phosphate buffer 50 mM, pH 7.5, 1.0 M NaCl) was determined for fresh and frozen stored samples. This buffer containing 1.0 M NaCl is termed as high ionic strength buffer. About 5 g of meat was homogenized with extraction buffer (EB) in the ratio of 1:10

(meat: EB) for 2 minutes using a homogenizer (Ultra Turrax T25, Janke and Kunkel, Staufen, Germany). The homogenate was centrifuged at $12000 \times g$ at 4°C for 15 min using high speed refrigerated centrifuge (Sorvall Legend XTR centrifuge, Thermo Fisher Scientific, New Hampshire, USA). The protein content of the supernatant was determined by Kjeldahl method. The solubility of the protein was expressed as percentage of total protein in the meat.

Statistical Analysis: All the experiments were performed in triplicates and expressed as the mean $\pm$ standard deviation ($n = 3$). One-way Analysis of Variance (ANOVA) followed by Tukey's test in SPSS statistics software (version 27.00) were performed to test the significance differences through the storage time.

Results and Discussion

This study evaluated the biochemical, microbial, and functional stability of Amur common carp (*Cyprinus carpio*

haematopterus) processed into four products ACMM, ACS, ACFP, and high-protein surimi (HPAS) during 28 days of refrigerated storage. Key quality indicators (pH, TVB-N, TBA, PV, FFA, TPC, NPN and SSN) were assessed to monitor spoilage, oxidation, and protein degradation.

Biochemical Analysis

The initial pH of all samples ranged from 6.67 to 6.81. HPAS showed the highest pH due to the use of dilute alkali (NaHCO_3) during alkaline washing, which neutralizes endogenous acids and stabilizes muscle proteins (Chotimarkorn & Yongsawatdigul, 2009) [16]. A gradual decline in pH was observed in all treatments over 28 days, particularly in ACMM and ACFP, which are more susceptible to microbial activity and proteolytic breakdown (Ojagh *et al.*, 2010) [17]. In contrast, ACS and HPAS maintained higher pH stability, consistent with reduced microbial substrates following surimi processing and the effect of cryoprotectants in suppressing pH-lowering microbial metabolism (Benjakul *et al.*, 2001) [18].

Table 1: Changes in the pH of refrigerated storage

Days of storage	ACMM	ACS	ACFP	HPAS	P<Value
Fresh	6.77 $\pm$ 0.02 ^{bC}	6.76 $\pm$ 0.01 ^{bC}	6.67 $\pm$ 0.005 ^{aD}	6.81 $\pm$ 0.01 ^{cD}	0.001
day 7	6.71 $\pm$ 0.005 ^{bB}	6.77 $\pm$ 0.01 ^{dC}	6.61 $\pm$ 0.005 ^{aC}	6.75 $\pm$ 0.005 ^{cC}	0.001
day 14	6.70 $\pm$ 0.005 ^{bB}	6.68 $\pm$ 0.01 ^{bB}	6.57 $\pm$ 0.01 ^{aB}	6.77 $\pm$ 0.01 ^{dC}	0.001
day 21	6.67 $\pm$ 0.005 ^{aA}	6.60 $\pm$ 0.01 ^{bA}	6.51 $\pm$ 0.01 ^{aA}	6.72 $\pm$ 0.02 ^{dB}	0.001
day 28	6.68 $\pm$ 0.005 ^{aA}	6.62 $\pm$ 0.005 ^{bA}	6.51 $\pm$ 0.005 ^{aA}	6.67 $\pm$ 0.005 ^{aA}	0.001
P<Value	0.001	0.001	0.001	0.001	

*Different super script in capital letter (A, B, C) and small letters (a, b, c) indicate significant difference ($p < 0.05$) amongst the rows and columns. Values are expressed as mean $\pm$ standard deviation ($n=3$)

TVB-N levels increased significantly ($p < 0.05$) in all samples during storage, with the highest values observed in ACMM (13.11 mg/100g), indicating advanced protein degradation due to microbial deamination and autolysis

(Hosseini *et al.*, 2010) [19]. HPAS maintained the lowest TVB-N values (7.81 mg/100g), validating that alkaline-washed surimi removes non-protein nitrogenous compounds, reducing spoilage potential (Liu *et al.*, 2013) [20]. The TVBN levels in Amur carp surimi and flesh powder reached 10.08% and 9.46%, respectively. These findings are consistent with the acceptability threshold of 20 mg/100g for fish products (ICMSF, 2011) [21].

Table 2: Changes in the total volatile base nitrogen during refrigerated storage

Days of storage	ACMM	ACS	ACFP	HPAS	P<Value
Fresh	7.85 $\pm$ 0.25 ^{aC}	2.07 $\pm$ 0.32 ^{aB}	7.88 $\pm$ 0.29 ^{aC}	1.43 $\pm$ 0.12 ^{aA}	0.001
day 7	10.40 $\pm$ 0.55 ^{bD}	3.84 $\pm$ 0.19 ^{bB}	7.72 $\pm$ 0.46 ^{aC}	2.89 $\pm$ 0.10 ^{bA}	0.001
day 14	10.91 $\pm$ 0.36 ^{bC}	5.88 $\pm$ 0.51 ^{cB}	8.77 $\pm$ 0.32 ^{bC}	4.57 $\pm$ 0.34 ^{cA}	0.001
day 21	11.56 $\pm$ 0.55 ^{cD}	7.77 $\pm$ 0.43 ^{dB}	8.89 $\pm$ 0.22 ^{bcC}	6.31 $\pm$ 0.20 ^{dA}	0.001
day 28	13.11 $\pm$ 0.2dD	10.08 $\pm$ 0.27 ^{eC}	9.46 $\pm$ 0.35 ^{cB}	7.81 $\pm$ 0.23 ^{eA}	0.001
P<Value	0.001	0.001	0.001	0.001	

*Different super script in capital letter (A, B, C) and small letters (a, b, c) indicate significant difference ($p < 0.05$) amongst the rows and columns. Values are expressed as mean $\pm$ standard deviation ($n=3$)

Peroxide Value (PV) increased markedly in ACMM (from 2.71 to 38.78 meq O_2/kg), confirming its vulnerability to primary lipid oxidation due to higher fat and haemoprotein content (Shahidi & Zhong, 2005) [22]. Surimi products (ACS

and HPAS) exhibited much lower PVs (4.44 and 2.91 meq O_2/kg , respectively), due to removal of lipid substrates during washing, which is known to improve oxidative stability (Park *et al.*, 2006) [23]. ACFP remained the most stable (1.44 meq O_2/kg) due to low water activity and reduced enzymatic and microbial activity (Rawdkuen *et al.*, 2008) [24].

Table 3: Changes in the peroxide value during refrigerated storage

Days of storage	ACMM	ACS	ACFP	HPAS	P<Value
Fresh	2.71 $\pm$ 0.25 ^{bA}	0.00 $\pm$ 0.00 ^{aA}	0.00 $\pm$ 0.00 ^{aA}	0.00 $\pm$ 0.00 ^{aA}	0.001
day 7	8.70 $\pm$ 0.18 ^{cB}	1.55 $\pm$ 0.12 ^{bB}	0.00 $\pm$ 0.00 ^{aA}	0.00 $\pm$ 0.00 ^{aA}	0.001
day 14	21.74 $\pm$ 0.34 ^{bC}	1.46 $\pm$ 0.04 ^{aB}	1.47 $\pm$ 0.06 ^{aB}	1.53 $\pm$ 0.05 ^{aB}	0.001
day 21	35.53 $\pm$ 0.24 ^{cD}	2.98 $\pm$ 0.13 ^{bcC}	1.46 $\pm$ 0.04 ^{aB}	2.92 $\pm$ 0.08 ^{bcC}	0.001
day 28	38.78 $\pm$ 0.32 ^{dE}	4.44 $\pm$ 0.20 ^{cD}	1.44 $\pm$ 0.01 ^{aB}	2.91 $\pm$ 0.09 ^{bcC}	0.001
P<Value	0.001	0.001	0.001	0.001	

*Different super script in capital letter (A, B, C) and small letters (a, b, c) indicate significant difference ($p < 0.05$) amongst the rows and columns. Values are expressed as mean $\pm$ standard deviation ($n=3$)

The thiobarbituric acid reactive substances (TBA-RS) values showed a statistically significant increase ($p < 0.005$) across all product categories during the storage period, reflecting the progression of lipid oxidation. Initially, the highest TBA value was observed in ACMM (0.40 mg MDA/kg), while HPAS exhibited the lowest (0.12 mg MDA/kg), suggesting superior lipid stability in HPAS likely due to better washing efficiency or endogenous antioxidant retention (Zhang *et al.*, 2022) [25]. All samples showed marked oxidative advancement by day 7, with HPAS rising to 0.44 mg MDA/kg and ACMM reaching 0.62 mg MDA/kg. At the day 14, ACMM and ACFP further escalated to 0.71 and 0.66 mg MDA/kg respectively,

indicating increased susceptibility of flesh powder to lipid peroxidation (Chakraborty *et al.*, 2009) [26]. An accelerated oxidation phase was evident on day 21, as ACMM, ACFP, and HPAS displayed converging TBA values between 0.75-0.92 mg MDA/kg, suggesting depletion of protective factors and intensified oxidative stress (Murthy *et al.*, 2021) [27]. On day 28, the peak TBA values were recorded ACMM at 1.24, ACFP at 0.97, ACS at 0.96, and HPAS at 0.85 mg MDA/kg. Although HPAS started with lower TBA levels, its relatively high end-point oxidation implies that even highly processed surimi types are vulnerable over time, depending on storage conditions and antioxidant dynamics. These results align with established understanding that TBA values correlate with lipid degradation in fish-based systems and demonstrate the need for targeted preservation strategies in surimi products (Janero, 1990; Shahidi, 1990) [28, 29].

Table 4: Changes in TBA value during refrigerated storage of Amur Carp surimi

Days of storage	ACMM	ACS	ACFP	HPAS	P<Value
Fresh	0.4 $\pm$ 0.00 ^{aC}	0.25 $\pm$ 0.00 ^{aB}	0.23 $\pm$ 0.00 ^{aA}	0.12 $\pm$ 0.00 ^{aB}	0.001
day 7	0.62 $\pm$ 0.00 ^{bB}	0.33 $\pm$ 0.02 ^{bA}	0.31 $\pm$ 0.00 ^{bA}	0.44 $\pm$ 0.00 ^{bA}	0.001
day 14	0.71 $\pm$ 0.00 ^{cD}	0.52 $\pm$ 0.01 ^{cA}	0.66 $\pm$ 0.01 ^{cC}	0.61 $\pm$ 0.00 ^{cB}	0.001
day21	0.92 $\pm$ 0.00 ^{dB}	0.72 $\pm$ 0.01 ^{dA}	0.75 $\pm$ 0.01 ^{dA}	0.76 $\pm$ 0.01 ^{dA}	0.001
day28	1.24 $\pm$ 0.02 ^{eC}	0.96 $\pm$ 0.01 ^{eB}	0.97 $\pm$ 0.01 ^{eA}	0.85 $\pm$ 0.01 ^{eB}	0.001
P<Value	0.001	0.001	0.001	0.001	

*Different super script in capital letter (A, B, C) and small letters (a, b, c) indicate significant difference ($p < 0.05$) amongst the rows and columns. Values are expressed as mean $\pm$ standard deviation ($n=3$)

Free Fatty Acids (FFA) serve as indicators of hydrolytic rancidity. FFA levels rose in all samples, with ACMM showing the highest increase (from 2.42% to 3.95%), primarily due to endogenous lipase activity and microbial lipolysis (Maqsood & Benjakul, 2010) [30]. The FFA level of minced meat was 2.42%, while it was reduced to 1.50% in surimi. Surimi's lower TBA values are attributed to the reduction of pro-oxidants like sarcoplasmic proteins and lipids (Zhang *et al.*, 2022) [25]. Alkaline washing also improves antioxidant capacity by promoting the removal of

oxidation catalysts (Park *et al.*, 2006) [23]. Negligible FFA levels were observed in Amur carp flesh powder and high protein surimi. After four weeks of refrigerated storage, an increase in FFA levels was noted. Minced meat exhibited the highest FFA percentage at 3.95%, followed by surimi at 2.53%. However, both high-protein surimi and Amur carp flesh powder maintained FFA levels within acceptable limits, recording 1.01% and 0.47% of oleic acid, respectively. HPAS and ACFP maintained significantly lower FFA accumulation ($< 1.1\%$), consistent with previous findings showing reduced lipid hydrolysis in low-fat or washed surimi products (Gómez-Guillén & Montero, 2005) [31].

Table 5: Changes in the free fatty acid during refrigerated storage

Days of storage	ACMM	ACS	ACFP	HPAS	P<Value
Fresh	2.42 $\pm$ 0.052 ^{aA}	1.5 $\pm$ 0.01 ^{bA}	0.16 $\pm$ 0.008 ^{aA}	0.164 $\pm$ 0.002 ^{aA}	0.001
day 7	2.36 $\pm$ 0.027 ^{dA}	1.74 $\pm$ 0.03 ^{cB}	0.23 $\pm$ 0.003 ^{aB}	0.403 $\pm$ 0.004 ^{bB}	0.001
day 14	2.94 $\pm$ 0.04 ^{dB}	2.17 $\pm$ 0.07 ^{cC}	0.243 $\pm$ 0.002 ^{bB}	0.486 $\pm$ 0.006 ^{bC}	0.001
day21	3.54 $\pm$ 0.07 ^{dC}	2.4 $\pm$ 0.04 ^{dD}	0.328 $\pm$ 0.006 ^{cC}	0.729 $\pm$ 0.008 ^{bD}	0.001
day28	3.95 $\pm$ 0.02 ^{dD}	2.53 $\pm$ 0.04 ^{dD}	0.474 $\pm$ 0.006 ^{dD}	1.01 $\pm$ 0.02 ^{bE}	0.001
P<Value	0.001	0.001	0.001	0.001	

*Different super script in capital letter (A, B, C) and small letters (a, b, c) indicate significant difference ($p < 0.05$) amongst the rows and columns. Values are expressed as mean $\pm$ standard deviation ($n=3$)

Microbiological Analysis

Total Plate Count (TPC): Initial microbial counts across all treatments (10^{-2} CFU/g) reflected high hygienic quality

(APHA, 2017). However, microbial proliferation increased significantly ($p < 0.05$) during storage, particularly in ACMM and HPAS (2.6×10^{-3} CFU/g at day 28), due to higher moisture and nutrient availability. ACS and ACFP demonstrated better microbial stability. ACFP, being freeze-dried, retained the lowest TPC, aligning with previous studies emphasizing the microbial inhibitory effect of low water activity (Ojagh *et al.*, 2010; ICMSF, 2011) [17, 21].

Table 6: Changes in total plate count (CFU/g) during refrigerated storage

Days of storage	ACMM	ACS	ACFP	HPAS	P<Value
Fresh	1.36 $\pm$ 0.10 $\times 10^{-2}$	1.35 $\pm$ 0.43 $\times 10^{-2}$	1.21 $\pm$ 0.12 $\times 10^{-2}$	1.55 $\pm$ 0.69 $\times 10^{-2}$	0.001
day 7	1.73 $\pm$ 0.34 $\times 10^{-2}$	1.85 $\pm$ 0.50 $\times 10^{-2}$	1.55 $\pm$ 0.32 $\times 10^{-2}$	1.19 $\pm$ 0.35 $\times 10^{-2}$	0.001
day 14	2.31 $\pm$ 1.2 $\times 10^{-2}$	2.05 $\pm$ 1.23 $\times 10^{-2}$	1.34 $\pm$ 0.90 $\times 10^{-2}$	1.69 $\pm$ 1.5 $\times 10^{-2}$	0.001
day21	2.29 $\pm$ 0.20 $\times 10^{-3}$	1.84 $\pm$ 0.14 $\times 10^{-3}$	2.83 $\pm$ 0.80 $\times 10^{-3}$	2.29 $\pm$ 0.65 $\times 10^{-3}$	0.001
day28	2.62 $\pm$ 0.12 $\times 10^{-3}$	2.21 $\pm$ 1.30 $\times 10^{-3}$	2.62 $\pm$ 0.60 $\times 10^{-3}$	2.54 $\pm$ 1.32 $\times 10^{-3}$	0.001
P<Value	0.001	0.001	0.001	0.001	

Non-Protein Nitrogen (NPN)

Non-Protein Nitrogen (NPN) content declined in all products over time, most notably in ACFP (230.01 to 125.67 mg/100g), likely due to protein breakdown followed by microbial utilization of peptides and amino acids (Chaijan *et al.*, 2006) [32]. ACMM had the highest initial NPN, a reflection of endogenous enzymatic hydrolysis and retained

sarcoplasmic proteins (Maqsood & Benjakul, 2011) [33]. ACFP's lower NPN compared to ACMM and surimi products is attributed to heat treatment, which reduces enzymatic activity (Gokoglu *et al.*, 2004) [34]. In contrast, HPAS and ACS had significantly lower NPN content due to repeated washing, which removes water-soluble nitrogenous substances (Benjakul *et al.*, 2003) [35].

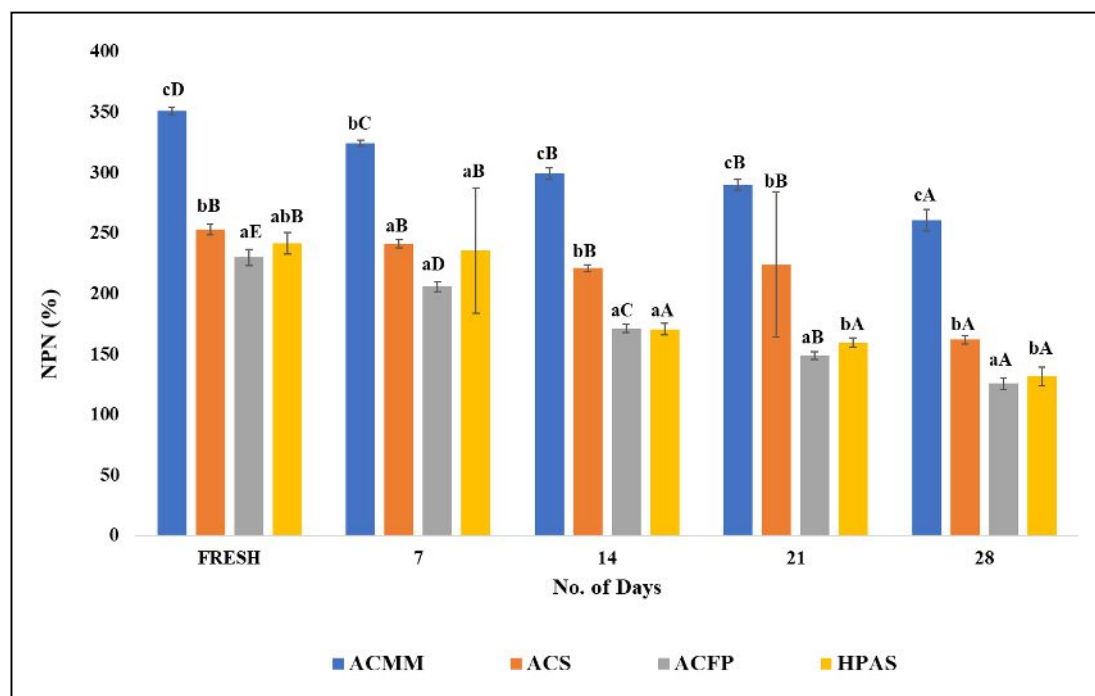


Fig 1: Changes in the non-protein nitrogen during refrigerated storage

Salt-Soluble Nitrogen (SSN)

Salt-Soluble Nitrogen (SSN), an indicator of myofibrillar protein functionality, showed a gradual decrease across all treatments. ACMM declined from 80.19% to 62.16%, consistent with denaturation and aggregation during storage (Shahidi, 1994) [36]. Surimi products retained higher SSN,

particularly HPAS (75.46% to 69.62%), due to the alkaline solubilization and cryoprotectants, which preserve protein integrity and functionality (Park *et al.*, 2006; Wang *et al.*, 2022) [23, 37]. ACFP showed minimal change, possibly due to low proteolytic activity in the dry state (Rawdkuen *et al.*, 2008) [24].

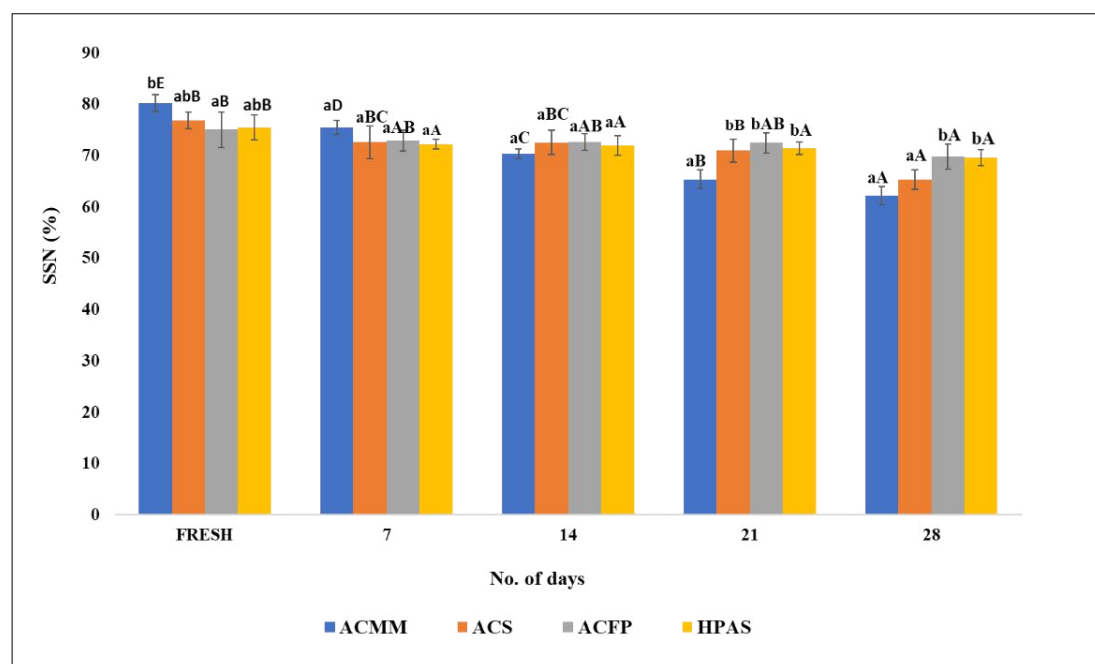


Fig 2: Changes in the salt soluble nitrogen during refrigerated storage

Conclusion

The study demonstrates that processed Amur common carp meat products exhibit varying degrees of biochemical, microbial, and functional stability during refrigerated storage, with high-protein surimi (HPAS) consistently showing superior performance. Surimi-based formulations, particularly HPAS and ACS, maintained lower levels of lipid oxidation, protein degradation, and microbial proliferation compared to minced meat and flesh powder, confirming the efficacy of washing and cryoprotectant treatments. These findings support the suitability of Amur carp for value-added processing and reinforce its potential as a sustainable and high-quality raw material for India's surimi industry.

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