RESEARCH ARTICLE Asian Journal of Dairy and Food Research, Volume 43 Issue 2: 313-319 (June 2024)

# Evaluation of Storage Stability and Quality Changes of Fish Muscle (*Pangasianodon hypothalamus*) Preserved in Fabricated Portable Solar Cooler and Domestic Refrigerator

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10.18805/ajdfr.DR-2075


## ABSTRACT

**Background:** This study was carried out to evaluate the quality changes and storage stability of fish muscle (*Pangasianodon hypothalamus*) preserved in fabricated portable solar cooler and domestic refrigerator.

**Methods:** Fish muscle from *Pangasianodon hypothalamus* were studied, keeping it in a fabricated portable solar cooler and in a domestic refrigerator at 5°±1°C to compare the shelf-life and quality of fish muscles. The fish muscle was packed separately in LDPE (low density polyethylene) packets and stored in a solar cooler and domestic refrigerator up to 7 days to compare the different physicochemical, microbiological and sensory parameters on 0, 3rd and 7th day.

**Result:** The results revealed that there were non-significant changes (P>0.05) observed for different parameters during the storage periods in both solar cooler and domestic refrigerator. In contrast to the domestic refrigerator requiring electricity for functioning, solar cooler requires green energy for the same which is abundant and free of cost, thus justifying its economical sustainability. Finally, from the present study it was concluded that the preservation of fish in a portable solar cooler system is thus economically suitable, feasible and acceptable up to 7 days as compared to domestic refrigeration method at controlled temperature.


**Key words:** Domestic refrigerator, Fish muscle quality, Portable solar cooler, Storage stability.

## INTRODUCTION

In India, fish is an integral part of daily diet for most of the people. The demand and per capita consumption of fish is also increasing. Besides, it is to note that fish is highly perishable item due to high content of moisture, protein, fat and other important nutritional parameters which all are conducive for the growth of microbes. Therefore, immediately after harvesting of fish, it should be stored in a proper temperature to avoid the spoilage. But it is a challenge to preserve fish in such rural, off-grid and catch-areas of fishes. So, one of the key interventions that can remediate the loss of quality due to spoilage and unhygienic storage conditions is through providing a cost-effective cooling solution to the fishers and fish vendors. Solar cooling is one such powerful and user-friendly solution, which is environment friendly and economical to use and operate as compared to traditional method of storage. Currently the conventional food preservation system is by use of refrigeration, which requires electricity and works under different harmful Freon gases. In present research, a solar cooler was fabricated and developed that could maintain the same temperature as refrigerator without using electricity and it has the potentiality to replace the conventional cooling and freezing methods *i.e.*, refrigeration.

Till date no such environment friendly and cheap cooler system were available to preserve such perishable products where utilization of green energy could be possible with a direct impact over global warming. This has been redressed fabrication of solar cooler with an attempt to use it replacing the conventional method of fish preservation by refrigeration. Besides, such facility can easily be utilized in the remote off-grid areas especially in the deep seashore soon after capturing and harvesting fishes.

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**How to cite this article:** Biswas, O. (2024). Evaluation of Storage Stability and Quality Changes of Fish Muscle (*Pangasianodon hypothalamus*) Preserved in Fabricated Portable Solar Cooler and Domestic Refrigerator. Asian Journal of Dairy and Food Research. 43(2): 313-319. doi: 10.18805/ajdfr.DR-2075.

**Submitted:** 03-02-2023 **Accepted:** 07-07-2023 **Online:** 11-08-2023

## MATERIALS AND METHODS

### Place and period of experiment

The present study was conducted at the Department of Agricultural Engineering, PalliSiksha Bhavana (Institute of Agriculture), Visva-Bharati, Santiniketan, West Bengal and Department of Livestock Products Technology, West Bengal University of Animal and Fishery Sciences, Kolkata from November 2018 to November 2020.

**Preparation of samples**

After collection of fish (*Pangasianodon hypothalamus*), it was thoroughly cleaned and washed with potable water. After removing the scales and skin, lengthwise fish muscle was cut parallel to the back bone and then these fillets were cut into smaller pieces and then packed with LDPE (low density polyethylene) and stored in the fabricated solar cooler (C1) and the domestic refrigerator (C2) up to 7 days. A totally six samples, from each C1 and C2 were kept in the solar cooler and refrigerator. Further, the samples of C1 and C2 were analyzed, for following physicochemical and microbiological studies up to 7 days.

**Fabrication of solar cooler**

Solar cooler was made following the protocol as outlined by Biswas *et al*. (2020; 2022). A cooling chamber with a volume of 25,000 cc was made using fruit grade plastic materials with provision of insulation. Two aluminum finned radiators in name of heat sinks were fitted inside and outside of the cooling chamber. These sinks helped to control heat of the cooling chamber. To regulate heat, two inbuilt small fans were fitted. The total heat transfer within the chamber takes place through a module called thermos-electric modules and the heat conductivity functions as per the principals called Peltier's effect. The solar cooler has a battery (12V, 10A, 10 Hrs, size 15.11 cm × 10.92 cm × 6.35 cm) and a polycrystalline type solar panel (12V, 80W, size 77.8 cm × 69 cm × 3 cm) were used. With these cooling devices a desired temperature up to (5°±1°C) could be reached within 6-7 hours of total time of day time exposure of the solar panel in a single day.

**Peroxide value (PV)**

Peroxide value (PV) was estimated as per procedure given by AOCS (1992), with slight modifications. Five grams of sample was weighed and mixed with 30 mL acetic acid-chloroform solution (3:2) in 250 ml glass-stoppered Erlenmeyer flask. Slurry obtained was gently swirled to extract lipid and then 0.5 ml saturated potassium iodide solution was added. After reaction for 1 minute with occasional shaking, 30 ml of distilled water and 0.5 ml of 0.5% starch solution were added. The mixed solution was titrated with 0.01 (N) sodium thiosulphate until intense blue color disappeared. A blank was also determined and subtracted from sample titration. The titration was persisted until the blue color disappeared ($T_1$ ml) and a blank was determined ($T_2$ ml). It is expressed in milliequivalents oxygen per 1000 g of sample.

**Thiobarbituric Acid Value (TBA) (mg malonaldehyde/kg)**

The TBA value was estimated for both of the samples, C1 and C2 on 0, 3$^{rd}$ and 7$^{th}$ day. The TBA number was determined as per the procedure by Strange *et al*. (1977) with slight modifications. 20 g minced samples was blended with 50 ml of precooled 20% TCA (Trichloroacetic acid) solution for 2 min. Than it was transferred to a beaker after rinsing with 50 ml of cold distilled water and mixed thoroughly. The mixture was filtered using Whatman-filter paper no 42 and the filtrate was named as TCA extract. 5 ml of freshly prepared 0.01(M) TBA solution was mixed with 5 ml of TCA extract in sterilized test tube and kept it in boiling water for 3 min. Another mixture of 5 ml of 20% TCA solution and 5 ml of 0.01(M) TBA solution was taken as blank solution. The absorbance at 532 nm was reported as TBA number.

**Tyrosine value**

For estimation of tyrosine value, the procedure of Teklemariam *et al*. (2015) was followed with slight modifications. 2.5 ml of TCA extract was diluted with equal amount of distilled water. 10 ml of 0.5 (N) freshly prepared sodium hydroxide and 3 ml of diluted phenol Folin- Ciocalteu reagent (1:2 with distilled water) were added. After 30 minutes, optical density (OD) was measured at 730 nm in a spectrophotometer. Tyrosine value was calculated by referring to the standard curve prepared as per the procedure of Abraha *et al*. (2018) and expressed as milligrams of tyrosine per g of sample.

**The pH value**

The pH value was estimated of the comminuted samples. Ten gram of sample was added with 50 ml of distilled water used for comminution using pestle and mortar. It was measured by digital pH meter (Systronic, Model 335).

**Microbiological studies**

Microbiological count *viz*. total plate count (TPC), total psychrophilic count (TPSC) and Total Coliform Count (TCC) in the sample was determined as per the method described in APHA (1992). All the agar mediums from Hi-media laboratories were used for all the microbiological assessments.

**Preparation of samples**

Samples were prepared according to APHA (1992). Ten gram of sample was transferred to 90 ml of normal saline solution and serial dilutions were prepared.

**Total plate count (TPC)**

It was determined by the APHA (1992) method using plate count agar. One ml of appropriate dilution of minced meat sample was transferred aseptically to sterile petri-plates in triplicate. The plates were then poured with 10-15 ml melted agar medium at 45°C. After solidification the petri-plates were incubated at inverted position at 37°C for 24-28 hrs. The colonies were counted by using colony counter. The average number of colonies was multiplied with dilution factor to obtain total count as colony forming unit (CFU) per gm of the sample. This count was then converted to total plate count of log CFU/g of sample.

**Total psychrophilic count (TPSC)**

The plates were prepared similar to that of TPC but were incubated at 4±1°C for 7 days. The Colonies were counted and expressed as log CFU/ g.

### Total coliform counts (TCC)

Most probable number (MPN) method was used for the quantitative estimation for coliforms. Serial dilution of the samples was prepared as described earlier. Nine test tubes containing about 9 mL Lauryl Tryptose Broth (LTB) with inverted Durham's tube were sterilized. Three test tubes were inoculated with 1 mL from $10^{-1}$ dilution, another three test tubes were inoculated from $10^{-2}$ dilution and the remaining three test tubes were inoculated from $10^{-3}$ dilution. The inoculated tubes were incubated at 37°C for 48 hours. Test tubes showing positive results (gas production in Durham's tube) were counted and recorded as presumptive positive for coliforms.

### Determination of warner bratzler shear force value

The shear force value of the fish fillets during refrigerated storage was measured following the method of Bourne *et al*. (2004) using the TA-XT2i texture analyzer (Stable Micro Systems, Surrey, England) equipped with a Warner-Bratzler shear apparatus. The cylindrical longitudinal muscle samples of the fillet were cutout with a borer of 11 mm in diameter. The e-blade was pressed down at a constant speed of 2 mm/s through the sample, cutting the muscle fiber transversely. Shear force (N) was recorded from six measurements.

### Instrumental color

The color of cooked fish muscles (both C1 and C2 samples) was compared using a Lovibond Tintometer (Tintometer Ltd, Salisbury, UK). Samples from three different places of fish fillets were taken in the sample holder and secured against the viewing aperture. The sample color was matched by adjusting red (a*) and yellow (b*) units, while keeping the blue units fixed at 2.0. The corresponding color units were recorded. The hue and chroma (saturation) values were determined using the formula, $\tan^{-1}$ b/a and (a2+b2)1/2 (Hafez *et al*., 2019) respectively, where, a* is the red unit, b* the yellow unit.

### Statistical analysis

Statistical analysis of the data obtained was carried out using Two-way Analysis of Variance (ANOVA) technique by randomized block design. To compare the means, Tukey's HSD test was adopted by using SPSS-16 software package. Three replications of the study were carried out and measurements for all the parameters were taken in duplicate each.

## RESULTS AND DISCUSSION

### Physicochemical parameters

The mean±S.E. value of different physicochemical parameters *viz.* peroxide value, TBA, Tyrosine value and pH of fish fillets are shown in Table 1.

The mean peroxide values of C1 and C2 samples at different storage periods were presented in Table 1. The peroxide values of C1 and C2 were gradually increased along with the storage days. When comparisons were made between C1 and C2 values on each day, there were no significant ($p<0.05$) changes in peroxide value. But both C1 and C2 samples showed gradual significant ($p<0.05$) changes in peroxide value with advancement of storage period. On 0, 3rd and 7th day the values of C1 and C2 were 2.26 meq/kg, 2.69 meq/kg, 3.59 meq/kg and 2.84 meq/kg, 2.70 meq/kg, 3.47 meq/kg respectively. The peroxide value is a method to determine the early stages of fat oxidation. The product is considered rancid when Peroxide value of 20-40 meq/kg is reached (Economou *et al.,* 1991). Numerically, all the values of Table 1 were far below of 20 meq/kg. Therefore, on the basis of PV value it can be concluded that the materials kept in the different systems were sound irrespective of their period of preservation. Observation of the present study is supported with the findings of Strateva and Penchev (2020) and Rathod and Pagarkar (2013), who reported similar effect of storage period on the peroxide value of fresh and frozen/thawed fish sample.

**Table 1:** Mean±SE values of physicochemical properties of fish muscle stored in refrigeration (C1) and Solar cooler storage (C2) at 5°±1°C.

| Parameters \ Days | 0 day | 3rd Day | 7th Day |
|---|---|---|---|
| **Peroxide value (meq/kg)** | | | |
| C1 | $2.26\pm0.01^{aA}$ | $2.69\pm0.05^{bA}$ | $3.59\pm0.02^{cA}$ |
| C2 | $2.48\pm0.06^{aA}$ | $2.70\pm0.07^{bA}$ | $3.47\pm0.06^{cA}$ |
| **Thiobarbituric acid value (mg malonaldehyde/g)** | | | |
| C1 | $0.18\pm0.02^{aA}$ | $0.19\pm0.02^{aA}$ | $0.21\pm0.05^{bA}$ |
| C2 | $0.18\pm0.01^{aA}$ | $0.20\pm0.05^{aA}$ | $0.22\pm0.02^{bA}$ |
| **Tyrosine value (mg/g)** | | | |
| C1 | $0.31\pm0.004^{aA}$ | $0.43\pm0.007^{bA}$ | $0.47\pm0.003^{cA}$ |
| C2 | $0.31\pm0.005^{aA}$ | $0.44\pm0.005^{bA}$ | $0.48\pm0.007^{cA}$ |
| pH | | | |
| C1 | $5.96\pm0.04^{aA}$ | $6.11\pm0.08^{bA}$ | $6.18\pm0.86^{bA}$ |
| C2 | $5.94\pm0.03^{aA}$ | $6.29\pm0.04^{bA}$ | $6.16\pm0.07^{bA}$ |

*Row wise Means bearing subscript indicates significant and non-significant difference at ($p<0.05$). Column wise means bearing superscript indicates significant and non-significant difference at ($p<0.05$).

The Thiobarbituric acids (TBA) values (mg malonal dehyde/kg) of C1 and C2 samples showed a gradual increment after 3rd day of storage. In compare to C1 and C2 values, they did not show any significant ($p<0.05$) changes both in day wise and in between samples up to 3rd day of storage. On 7th day of storage, the values were showing significant ($p<0.05$) change from previous storage days but not in the comparative values of the same day. On 0, 3rd and 7th day the values of C1 and C2 were 0.18 mg malonaldehyde/g, 0.19 mg malonaldehyde/g, 0.21 mg malonaldehyde/g and 0.18 mg malonaldehyde/g, 0.20 mg malonaldehyde/g, 0.21 mg malonaldehyde/g respectively. The Thiobarbituric acids (TBA) values (mg malonaldehyde/kg) of C1 and C2 samples showed a gradual increment after 3rd day of storage. This is due to the fat degradation products are higher in Pangus fish (Somboonyarithi, 1990). The observation of the present study could collaborate the above note in respect to TBA values. Further to note that The Variation in TBA values was used to describe the degree of lipid oxidation as second stage auto- oxidation during chilled storage as opined by Mazandrani *et al*. (2015) and the present work in regard to the variations of TBA value could be explained in regard to this explanation. Similar increasing trend of TBA value was noticed by Mazandrani *et al*. (2015) and Nag *et al*. (1998).

The Tyrosine values of C1 and C2 were gradually increased. In compare to C1 and C2 values on each day did not show any significant ($p<0.05$) changes. But both C1 and C2 samples showed a gradual significant ($p<0.05$) change in values with advancement of storage period. On 0, 3rd and 7th days the values of C1 and C2 were 0.31 mg/g, 0.43 mg/g, 0.47 mg/g and 0.31 mg/g, 0.44 mg/g, 0.48 mg/g respectively. In a similar work of the estimation of tyrosine value, Ozden (2005) found a same type of results while working with marinated fishes. Tyrosine value can effectively monitor the muscle quality to indicate proteolysis. Tyrosine value increases rapidly when muscle stored at low temperature (nearing to 7°C). Tyrosine value was used as one of the methods for detecting microbial spoilage of muscle food (Jay, 1996). In the present study the gradual increment of Tyrosine value attributed to the fact that there was proteolysis of this amino acid in course of storage period.

The pH of C1 and C2 samples showed a gradual increment in storage days. The C1 and C2 values, they did not show any significant ($p<0.05$) change in day wise on 0 day. On 3rd and 7th day of storage. The values showed significant ($p<0.05$) changes within the preservation systems between 0, 3rd and 7th day storage periods. Storage time tends to increase the pH values which can be associated with the production of basic components such as ammonia, volatile alkali and trimethylamine due to internal enzymatic activity and the growth of bacteria (Chamarana *et al*., 2012). The gradual increment of the pH value as observe in the present study could substantiate the above observation of gradual increment of pH in both the systems of preservation and along with the period of preservation. The results are supported by the findings of Guimarães *et al.* (2015) and Rathod and Pagarkar (2013).

**Microbiological counts**

The mean±SE value of different microbiological parameters *viz*. TPC, TPSC and TCC of fish fillets are shown in Table 2.

Total plate count (log cfu/g) of C1 and C2 samples grew over time significantly ($p<0.05$). At 0 day, there was no significant difference ($p>0.05$) observed between C1 and C2 samples but beyond 3rd day of storage this comparison became significant ($p<0.05$). On 0, 3rd and 7th day the counts of C1 and C2 were 1.24 log cfu/g, 2.32 log cfu/g, 3.36 log cfu/g and 1.20 log cfu/g, 2.41 log cfu/g, 3.48 log cfu/g respectively. Total psychrophilic counts were not detected up to 3rd day of storage and total coliform counts were not detected throughout the storage periods. In the present study it is also to highlight that such increments of peroxide value, TBA value, Tyrosine value and pH might be due to the enhanced microbial load, enhanced production of proteolytic enzymes in the late logarithmic phase of microbial growth

**Table 2:** Mean±SE values of Microbiological properties of fish muscle stored in Refrigeration (C1) and Solar cooler storage systems (C2) at 5°±1°C.

| Parameters \ Days | 0day | 3rd Day | 7th Day |
|---|---|---|---|
| **TPC (log cfu/g))** | | | |
| C1 | $1.24\pm0.02^{aA}$ | $2.32\pm0.92^{bA}$ | $3.36\pm0.08^{cA}$ |
| C2 | $1.20\pm0.02^{aA}$ | $2.41\pm0.11^{bB}$ | $3.48\pm0.12^{cB}$ |
| **TPSC (log10 cfu/g)** | | | |
| C1 | ND | ND | 1.64±0.08A |
| C2 | ND | ND | 1.40±0.06A |
| **TCC MPN/g** | | | |
| C1 | ND | ND | ND |
| C2 | ND | ND | ND |

*Row wise Means bearing subscript indicates significant and non-significant difference at ($p<0.05$). **Column wise means bearing superscript indicates significant and non-significant difference at ($p<0.05$).

ND= Not detected.

which were altogether responsible for autolysis and bacterial proteolysis (Gram and Huss, 1966). When this study was extended towards the evaluation of microbiological values in terms of TPC, TPSC, TCC, the explanation as noted above was found to be very much parenting and seems to be justified observation in relation to the present study. The TPC of the both C1 and C2 samples were far below the maximum permissible limit *i.e.,* $\log_{10}$ 6 cfu/g (Jay, 1996). However, throughout the storage, no significant differences ($p>0.05$) were noticed between the C1 and C2 samples indicating comparable efficacy of solar cooler with the conventional refrigerator. Such findings are collated with several earlier reports of Waghmare *et al.* (2017), Rathod and Pagarkar (2013) and Nag *et al.* (1998). However, the results as observed in terms of microbes were well within the range of acceptability when observed in different categories of systems and periods.

**Warner bratzler shear force value (N) and instrumental color**

Results obtained for Shear force value (N) and instrumental colors (LAB colors) of fish fillets are graphically presented under Fig 1 and Fig 2 (a, b and c). Significant increase in Shear force value was during advancement of storage period which indicates the textural properties or rather tenderness of fish fillets got tougher with the progress of time under refrigerated storage. L* value of both the sample was increased but in a non-significant manner ($p>0.05$). Redness value (a*) decreased and Yellowness value (b*) increased with the progress of time and this was significant ($p<0.05$) beyond 3rd day of storage. However, throughout the storage period, C1 and C2 sample never differ significantly among themselves. The sensory attributes of the samples as observed through Warner Bratzler Shear

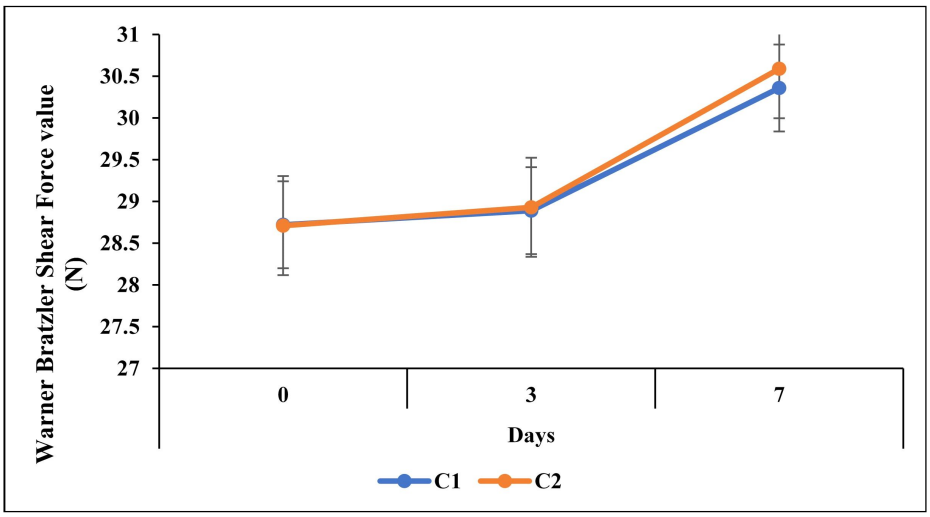


**Fig 1:** Mean±SE of changes in warner bratzler shear force values (N) of fish (*Pangasianodon hypothalamus*) muscle stored in refrigeration (C1) and solar cooler storage (C2) at 5°±1°C.

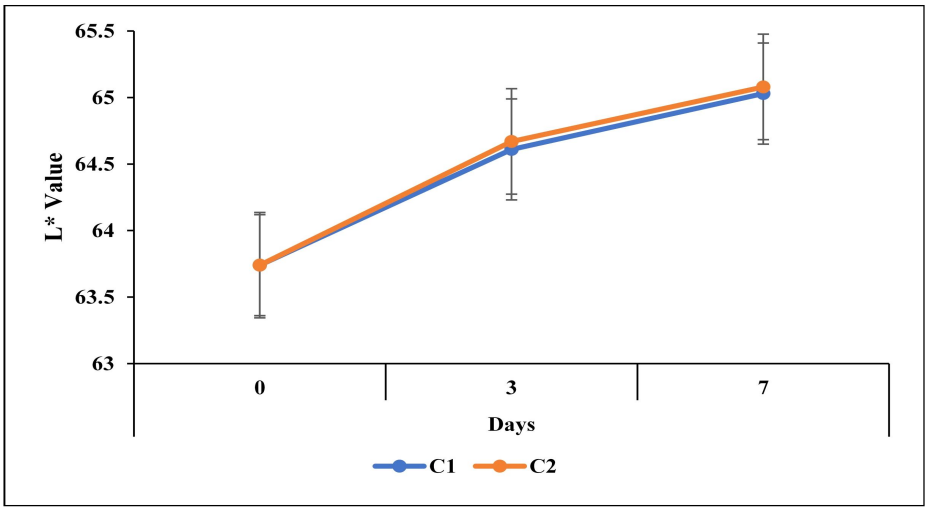


**Fig 2(a):** Mean±SE of changes in instrumental colour (L* Value) of fish (*Pangasianodon hypothalamus*) muscle stored in refrigeration (C1) and solar cooler storage (C2) at 5°±1°C.

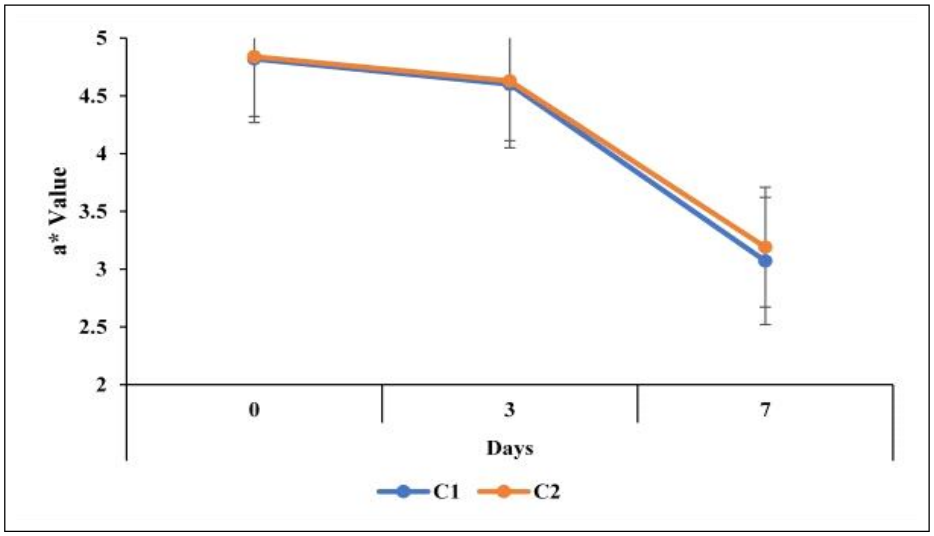


**Fig 2(b):** Mean±SE of changes in instrumental colour (a* Value) of fish (*Pangasianodon hypothalamus*) muscle stored in refrigeration (C1) and Solar cooler storage (C2) at 5°±1°C.

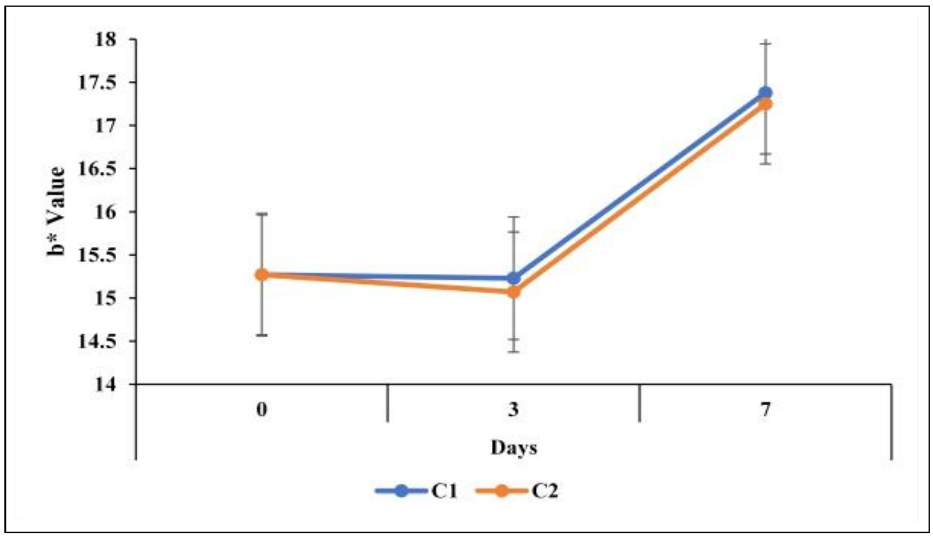


**Fig 2(c):** Mean±SE of changes in instrumental colour (b* Value) of fish (*Pangasianodon hypothalamus*) muscle stored in refrigeration (C1) and solar cooler storage (C2) at 5°±1°C.

Force value (N) and Instrumental colors (lab colours) were found at the positive side of enhancement in muscle quality. Pornpimol and Thamarak (2018) studied Shear force values of fish fillets under frozen storage and reported similar increasing trend. The changes in instrumental color values with the storage time agree with the observation reported by Waghmare *et al*. (2017) and Pornpimol and Thamarak (2018).

Therefore, when this study considered inclusively of all the parameters including physiochemical, microbiological and sensory attributes, the role of solar cooler and domestic refrigerator were found equally effective. However, there is scope to prolong this study along with investigation some more related parameters to declare that, such fabricated solar cooler is a perfect replacer of domestic refrigerator in course of fish preservation. The observation of Biswas and Kandaswami (2021) can be considered here to develop the solar cooler with further technological inputs. Nevertheless, the potentiality of fabricated solar cooler cannot be ignored in terms of its efficiency for extending the shelf-life of perishable muscle food item.

## CONCLUSION

Based on the studies, it was observed that '*Pangasius* fish fillet' stored in controlled refrigerator storage at 5±1°C (C1) and in solar cooler storage at 5±1°C (C2) showed non-significant difference in terms of peroxide value, texture profile analysis and instrumental color throughout the storage over time for 7 days period. In contrast to the domestic refrigerator requiring electricity for functioning, solar cooler requires green energy for the same which is abundant and free of cost, thus justifying its economical

sustainability. In addition, the fabrication of solar cooler is done by locally available materials which are economic as compared to the domestic refrigerator. Till date no such environment friendly and cheap cooler system in a form of cooler box is readily available to preserve fish or meat products particularly in Indian market. So, from the results it could be concluded that solar cooler storage can be used as a substitute of refrigeration storage. Further research is needed on solar coolers, so that solar cooler can replace conventional refrigerator for the storage of perishable products particularly at the places of fish harvesting. This would thus boost the better use of unconventional "green" source of energy.

## ACKNOWLEDGEMENT

The author acknowledges the help of the departmental laboratories with the financial support of DST, Govt. of India through Inspire Fellowship (DST/INSPIRE Fellowship/ IF170433) to the first author.

### Conflict of interest

All authors declare that they have no conflict of interest.

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