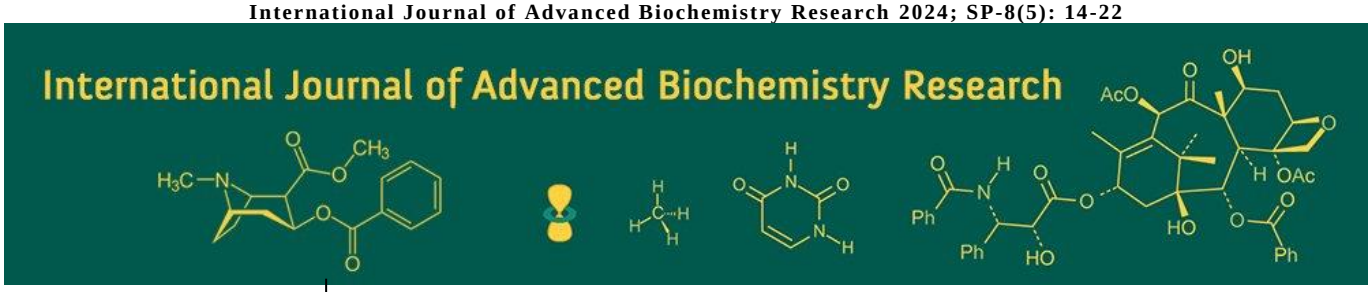


ISSN Print: 2617-4693
ISSN Online: 2617-4707
IJABR 2024; SP-8(5): 14-22
www.biochemjournal.com
Received: 24-02-2024
Accepted: 29-03-2024

Karunanithi Masilan
Department of Fish Processing Technology, Dr MGR Fisheries College and Research Institute, Tamil Nadu Fisheries University, Ponneri, Tamil Nadu, India

Neethirajan Neethiselvan
Department of Fishing Technology and Fisheries Engineering, Fisheries College and Research Institute, Tamil Nadu Fisheries University, Thoothukudi, Tamil Nadu, India

Arjunan Karthy
Department of Fishing Technology and Fisheries Engineering, Fisheries College and Research Institute, Tamil Nadu Fisheries University, Thoothukudi, Tamil Nadu, India

Lidiya Wilwet
Microbiology, Fermentation and Biotechnology Division, ICAR-Central Institute of Fishery Technology, Cochin, Kerala, India

Kavinesan Kumar
Department of Fish Processing Technology, Fisheries College and Research Institute, Tamil Nadu Fisheries University, Thoothukudi, Tamil Nadu, India

Corresponding Author:
Karunanithi Masilan
Department of Fish Processing Technology, Dr MGR Fisheries College and Research Institute, Tamil Nadu Fisheries University, Ponneri, Tamil Nadu, India

Physical properties and storage stability of fish scale gelatin based hydrogel under refrigerated conditions

Karunanithi Masilan, Neethirajan Neethiselvan, Arjunan Karthy, Lidiya Wilwet and Kavinesan Kumar

DOI: https://doi.org/10.33545/26174693.2024.v8.i5Sa.1101

Abstract
In this study, gelatin based hydrogels are prepared using fish scale discarded as waste by the fish processing plant. The physical quality and storage stability of the gel in refrigerated conditions were studied. In order to enhance their storage quality and safety, gels are prepared with the addition of permitted antifungal agent, sodium benzoate at three different concentrations *viz* 0.1%, 0.5% and 1%, and subsequently stored in refrigerated and room conditions. During storage, the parameters deciding physical quality of the gels such as weight loss, moisture loss, rate of shrinkage were found affected invariably in all the gels with different diameter i.e. 10 mm, 15mm and 20 mm Ø. However, the effects were highly pronounced in gel with 20 mm thickness Ø. During one month storage, about 20-24% weight loss was observed in 10 mm Ø and 15mm Ø. The result indicated that the type of storage influenced the growth of fungi. Addition of antifungal agent had significantly reduced the fungal growth and the gel treated with 1% sodium benzoate had much lower counts. This study helped as to prepare hydrogel utilizing fish scale that can be held safe at room temperature without any quality defect.

Keywords: Fish scale gelatin, hydrogel, physical quality, gel shrinkage

Introduction
Gelatin is dissolved in water at a specific temperature, it often forms a thermo reversible gel, which upon cooling hardens to create a hydrogel. According to Kim and Uyama (2007) [36], hydrogels are made of three-dimensional hydrophilic polymer networks that are encased in a lot of water. Due to their special characteristics, hydrogels are prospective materials that could be exploited in tissue engineering. The rate at which fish gelatin gels, its thermal stability, and its solubility are only a few of the criteria that restrict its application. Fish waste, including the skull, bones, fins, skins, and scales, can be used to create fish gelatin, a crucial biopolymer. Fish processing produces enormous amounts of trash, and today's worldwide issues include how to dispose of those wastes.
Globally, around 20 MMT of fish waste are produced each year, which accounts for 25% of all marine fish produced globally (FAO, 2014) [9]. Coastal contamination results from their direct dumping into the coastal waters close to fishing harbors (Govindharaj *et al*., 2019). So, in addition to lowering the risk of coastal contamination, the valorization of fish wastes into value-added products like gelatin would open the road for their optimal application. According to Gomez-Guillen *et al*. (2002), Muyonga *et al*. (2004), Badii and Howell (2006), and Huang *et al*. (2019) [37, 38, 39, 40] fish gelatin production is one method of utilizing processing waste from the seafood industry. It has good functional properties like gel strength, melting point, and gelling point, making it ideal for use in a variety of industrial applications such as the creation of stabilizer, gelling agent, and packaging film. Along with these applications it has also been applied to the development of artificial fish bait in the fishing business (Masilan and Neethiselvan, 2018, Karunanithi *et al*., a,b) [22, 23, 15-16]. With the help of other biopolymers like chitosan, carrageenan, and alginate, among others, a few studies have been started in various parts of the world to develop artificial hydrogel fish bait (Dellinger *et al*., 2016; Lokkeborg, 2014; Siikavupio *et al*., 2017; Karunanithi *et al*., 2018a,b; Masilan *et al*., 2018) [7, 20, 29, 15-16, 22-23].

As made from different fish body parts, fish gelatin has physicochemical properties that vary. Fish scale gelatin (FSG) was found by Masilan *et al*. (2021) [24] to have a greater melting point (27.3 °C), higher gel strength (1.98 N), and a higher yield (16.82%) than gelatin made from the head, skin, and fins of Lethirinus spp. Despite FSG having good physicochemical properties, only a few attempts have been to make use of them for its value addition and their properties (Masilan *et al*., 2021, Masilan *et al*., 2022a,b) [24, 25-26]. A hydrogel-based fish bait appropriate for long line fishing was developed taking into account the superior functional and mechanical features of FSG. According to Lokkeborg (2014) [20], the substance chosen for creating artificial fish bait should have both binding and enticing capabilities. Since FSG has excellent binding abilities, it has been combined with other hydrolysates to create hydrogel. To create fish gelatin based hydrogel, needs to have a thorough understanding of the interactions between FSG and various temperatures. Therefore, in the present study, changes in the physico-chemical characteristics and microbiological analyses were examined when an FSG hydrogel was stored in different temperatures.

Materials and Methods

Designing of gel casting unit

Three different types of hydrogel were prepared using Teflon molds used as casting unit. Teflon castings were designed with 10cm length and three different diameters such as 10 mm Ø, 15mm Ø, and 20 mm. The casting units are illustrated in Fig. 1.

Casting of FSG Hydrogel

Castings were made using polypropylene beakers that had dimensions of 40 mm in height and 35mm in diameter. Distilled water was placed in a 50 ml glass beaker and heated to 80 °C using a hot plate magnetic stirrer (REMI, India) while being continuously stirred. Sucrose was added after the temperature reached 80 °C, then fish gelatin. A homogeneous hydrogel was created by continuously stirring the mixture. After the gelatin had been evenly dissolved, the hot hydrogel solution was poured into the casting units and refrigerated for 16 hours at a temperature of 6 °C to allow for gelation and subsequent testing (Masilan *et al*., 2022a) [25].

Effect of refrigeration storage on physical quality changes of FSG hydrogel

FSG hydrogel was prepared in three different sizes using Polyacrylic acid (PA) castings of cylindrical shape. The castings had the dimension of (i) 100 mm length and 10 mm dia, (ii) 100 mm length and 15mm dia, and (iii) 100 mm length and 20 mm dia were used. The FSG Hydrogel bait prepared in three different sizes is illustrated in Fig. 2.

Shrinking measurement of FSG hydrogel by area and volume

FSG hydrogel in three different sizes were weighed and measured using a vernier scale and stored in a refrigerator. The moisture content and weight of the baits were measured at an interval of three days of storage up to 30 days. Ten samples each in triplicate were used for the study.

The moisture loss and weight loss were calculated based on the following formulae:

$$\text{Moisture loss (\%)} = \text{Mi} - \text{Mf}$$

Where,

M_i and M_f are initial and final moisture content (%) of the bait.

$$\text{Weight loss (\%)} = \left\{\frac{\text{Wi}-\text{Wf}}{\text{Wi}}\right\} \text{x } 100$$

Where,

W_i and W_f are the initial and final weight (g) of the baits respectively.

The measurement of shrinkage was performed by measuring the dimensions of baits such as length and diameter were measured using a vernier scale. The surface area and volume of the baits were worked out using the formulae $A=2\pi rh$, and $V=\pi r^2 h$, respectively. The shrinkage both in terms of surface area and volume were calculated at an interval of three days up to thirty days under refrigerated condition. Ten samples each in triplicate were used for the study. Percentage of shrinkage was then calculated by:

$$\text{\% of shrinkage} = \frac{\text{Si}-\text{Sf}}{\text{Si}} \text{x} 100$$

Where,

S_i and S_f are initial and final surface area/ volume respectively.

Standardization of concentration of antifungal agent

The successfully evolved FSG hydrogel was prepared incorporating the antifungal agent, sodium benzoate (NaB) in artificial fish bait at four different concentrations such as 0%, 0.1%, 0.5%, and 1% and stored in a refrigerator for one month. Weekly samplings were made to analyse microbiological parameters such as total plate count (TPC) and total fungal count (TFC).

Statistical analysis

The quantitative and analytical values of the experiments were expressed as mean±SD. Triplicate samples were used for each of the experiment and the probability value of $p<0.05$ was considered as significant. One way ANOVA was accomplished for each experiment and mean values were compared using Duncan's post-hoc test. Turkey post-hoc test was performed for amino acid leaching analysis (SPSS, Version 20, SPSS Inc., Chicago, IL, USA).

Results and Discussion

Physical quality changes

Gel strength is an important physical parameter used for characterizing texture of any gel (Masilan *et al*., 2022b) [26]. During storage the parameters deciding physical quality of the gel such as weight loss, moisture loss, rate of shrinkage were found affected in gels with the diameter of 10 mm, 15mm and 20 mm thickness. However, the effects were highly pronounced in gels with 20 mm thickness. During one month storage, about 20 to 24% weight loss was found to occur in 10 mm and 15mm Ø gels and in the case of 20 mm bait, a high level of about 30% weight loss was observed (Fig. 3, Fig. 4 and Fig. 5). Regarding shrinkage volume, under refrigerated storage for 30 days, the values were 3365.66, 6688.35, and 10257.8 mm^3 for 10 mm, 15mm and 20 mm Ø baits respectively (Table 1, 2 and 3). The respective percentages of shrinkage in volume were 15.84%, 16.5% and 26.73% (Fig 3, Fig. 4 and Fig. 5). Similar

phenomenon was also noted with respect to Weight loss and loss of moisture content of the baits indicating positive correlation between them. The moisture loss were 217.90 mg, 354.64mg, and 594.23mg (Table. 1, 2 and 3) which accounted for the percentage loss of moisture content of 23.46%, 28.21% and 34.7% (Fig. 3, 4 and 5) for 10 mm, 15mm and 20 mm Ø baits respectively. In the case of weight loss, the reduction were 325.80 mg, 726.75 mg, and 1193.50 mg (Table. 1, 2 and 3) which accounted for the percentage loss of weight as 22.42%, 23.5% and 31.8% for 10 mm, 15mm and 20 mm Ø baits respectively. The relatively higher weight loss, moisture loss, and shrinkage in volume of the 20 mm Ø baits compared to other gels could be attributed to higher initial specific volume of 14,00 mm^3 /cm and specific surface area of 3,49 mm^2/cm which facilitated relatively higher degree of dehydration (Fig. 2, 3 and 4).

Microbiological quality changes

Spoilage of baits by Fungi belonging to the genera *Candida* spp and *Aspergillus* spp were observed during the end of fourth week of storage even after incorporation of Sodium benzoate (NaB) at the concentrations of 0.5% and 1% levels. Fungal infections could be observed at the end of third week in the baits treated with 0.1% of NaB. However, no fungal infections could be observed in the gels incorporated with NaB at 0.5% and 1% till third week of storage (Fig. 6). Irrespective of the levels of incorporation of NaB, fungal infection was evident in all the experimental gels after third week of storage. Storing gels in refrigerator without the addition of antifungal agent could withstand without fungal attack up to one week. However, fungal infection could be visible on eighth day onwards. The gels added with NaB at 1% level did not show any visible impact with respect to fungal growth compared to that treated with 0.5% NaB till third week (Fig. 7). Hence, it was concluded that in the economic point of view addition of NaB at 0.5% level would be sufficient to delay the onset of fungal infection up to three weeks. In room temperature all the baits lost their quality completely within 7 days. The addition of NaB at different levels did not have any visible impact on TPC. The TPC increased to 0.5 log CFU/g, 1 log CFU/g, 2 log CFU/g, and 3 log CFU/g on 1st, 2nd, 3rd, and 4th week of storage irrespective of the gels the levels of addition from 0.1% to 1%. However, a general reduction in TPC could be observed in the NaB incorporated gel throughout the storage period. The TPC levels of control gel on 1st, 2nd, 3rd, and 4th week of storage were 1 log CFU/g, 4 log CFU/g, 6 log CFU/g, and 7 log CFU/g respectively (Fig. 8).

Table 1: Shrinking characteristics of FSG hydrogel 10 mm Φ bait on refrigerated storage

Storage days	Loss of weight (mg)	Loss of moisture content (mg)	Shrinking of Volume (mm^3)	Shrinking of area (mm^2)
0^{th}	420±0.11	226.8±0.15	3380.0±0.07	1570.0±0.02
3	415.8±0.10	217.9094±0.14	3365.466±0.05	1565.133±0.02
6	411.6±0.10	209.291±0.10	3273.767±0.05	1549.119±0.01
9	406.812±0.11	201.3825±0.14	3167.533±0.07	1519.917±0.05
12	397.194±0.12	195.7057±0.15	3099.933±0.09	1474.701±0.03
15	390.726±0.10	192.213±0.14	3010.6±0.10	1445.028±0.02
18	382.0824±0.10	186.4296±0.17	2944.453±0.07	1428.7±0.05
21	367.08±0.20	184.9327±0.15	2905.347±0.09	1409.232±0.07
24	345.03±0.18	178.8998±0.17	2875.873±0.10	1373.122±0.05
27	330.0024±0.15	173.5927±0.17	2844.507±0.10	1346.432±0.04
30	325.8024±0.11	217.9094±0.14	3365.466±0.07	1565.133±0.04

Notes: All values are mean standard deviation ± of triplicate analysis

Table 2: Shrinking characteristics of FSG hydrogel 15mm Φ bait on refrigerated storage

Storage days	Loss of weight (mg)	Loss of moisture content (mg)	Shrinking of Volume (mm^3)	Shrinking of area (mm^2)
0^{th}	950.00±0.25	494±0.17	8010.0±0.11	2550.0±0.09
3	930.525±0.20	479.6246±0.17	7841.79±0.13	2503.335±0.05
6	900.03±0.21	464.7552±0.20	7776.909±0.13	2489.82±0.05
9	875.045±0.21	451.1208±0.18	7691.202±0.10	2461.77±0.09
12	859.56±0.21	443.9084±0.18	7623.918±0.17	2444.94±0.04
15	835.525±0.25	428.9896±0.19	7503.768±0.13	2399.04±0.05
18	802.085±0.20	414.1696±0.18	7343.568±0.13	2364.87±0.07
21	781.09±0.20	399.5472±0.20	7143.318±0.15	2321.775±0.09
24	763.515±0.22	381.6644±0.21	6967.899±0.17	2253.945±0.09
27	743.09±0.21	370.8952±0.20	6822.918±0.17	2194.275±0.11
30	726.75±0.21	354.6426±0.20	6688.35±0.15	2164.95±0.14

Notes: All values are mean standard deviation ± of triplicate analysis

Table 3: Shrinking characteristics of FSG hydrogel 20 mm Φ bait on refrigerated storage

Storage days	Loss of weight (mg)	Loss of moisture content (mg)	Shrinking of Volume (mm^3)	Shrinking of area (mm^2)
0^{th}	1750.00±0.27	910.00±0.21	14000.0±0.19	3490.0±0.11
3	1650.075±0.30	848.939±0.28	13482.0±0.17	3416.012±0.13
6	1584.275±0.30	819.364±0.28	13045.2±0.17	3314.104±0.13
9	1525.125±0.31	791.154±0.28	12602.8±0.15	3226.854±0.15
12	1490.825±0.35	767.221±0.31	12439±0.15	3167.873±0.13
15	1434.3±0.35	735.007±0.30	12119.8±0.17	3116.989±0.10
18	1378.475±0.38	703.066±0.33	11628.4±0.15	3063.173±0.11
21	1319.15±0.35	681.408±0.30	11363.8±0.17	3003.843±0.11
24	1263.325±0.35	652.015±0.31	10984.4±0.13	2939.976±0.10
27	1225.35±0.38	626.808±0.32	10607.8±0.15	2864.243±0.13
30	1193.5±0.38	594.23±0.33	10257.8±0.15	2739.65±0.13

Notes: All values are mean standard deviation ± of triplicate analysis

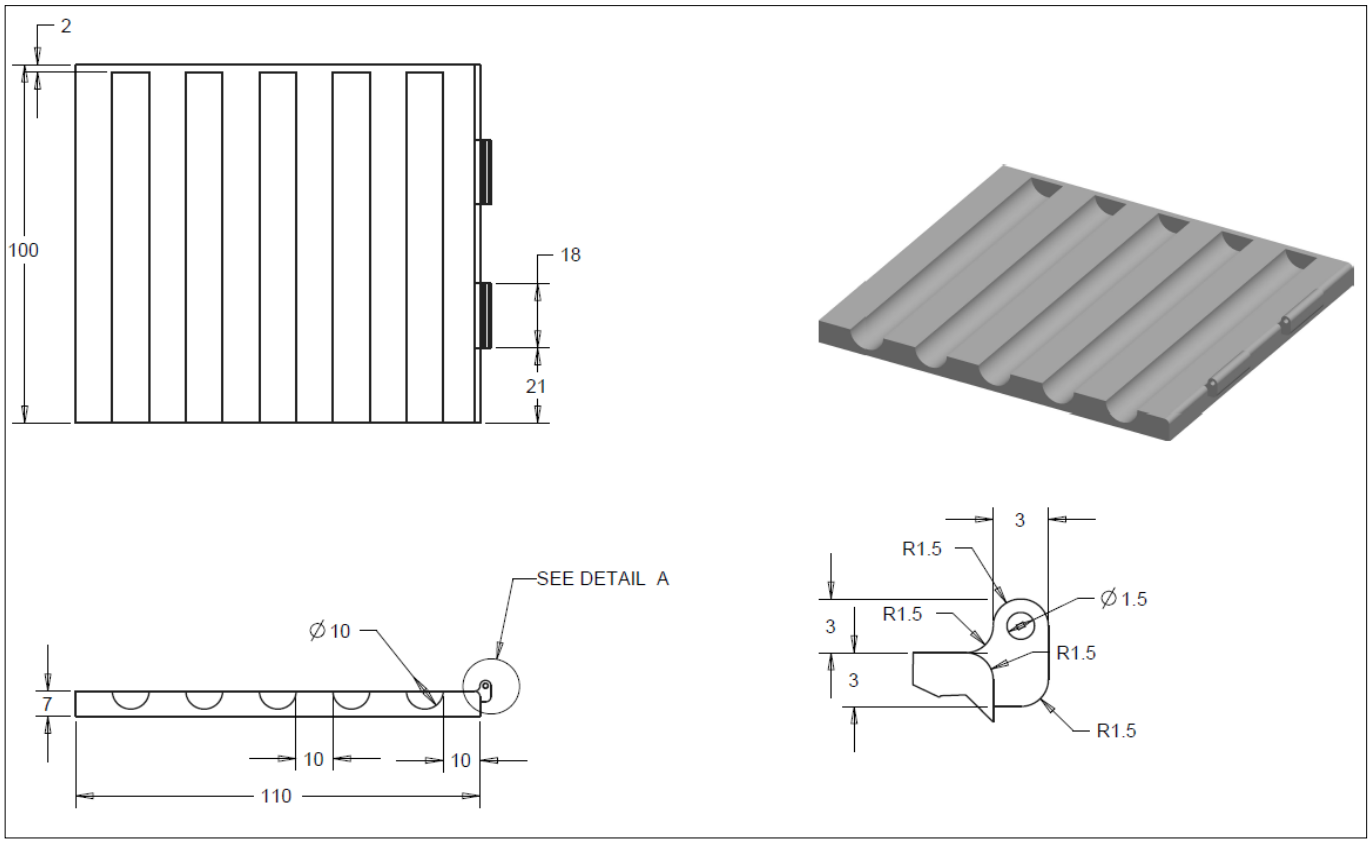
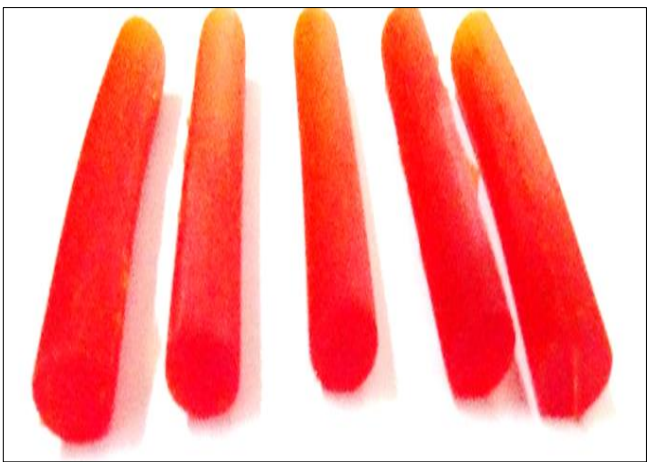


Fig 1: Design layout of mould

A. 10 mm Φ (100 mm x 10 mm)



B. 15mm Φ (100 mm x 15mm)

C. 20 mm Φ (100 mm x 20 mm)

Fig 2: The gelatin based fish bait in different sizes

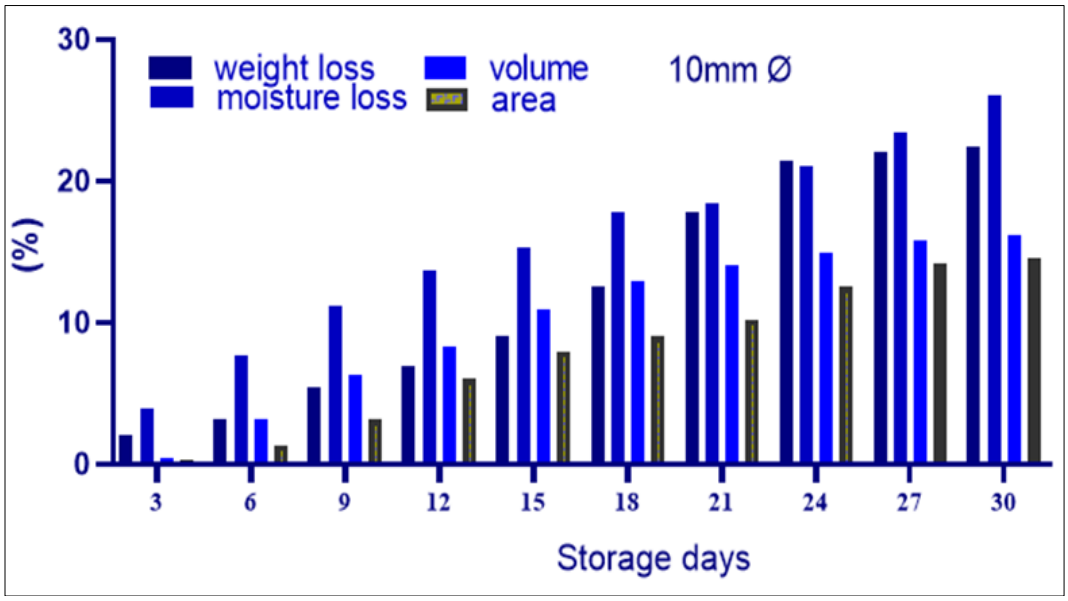


Fig 3: Shrinking characteristics of 10 mm Ø bait on refrigerated storage

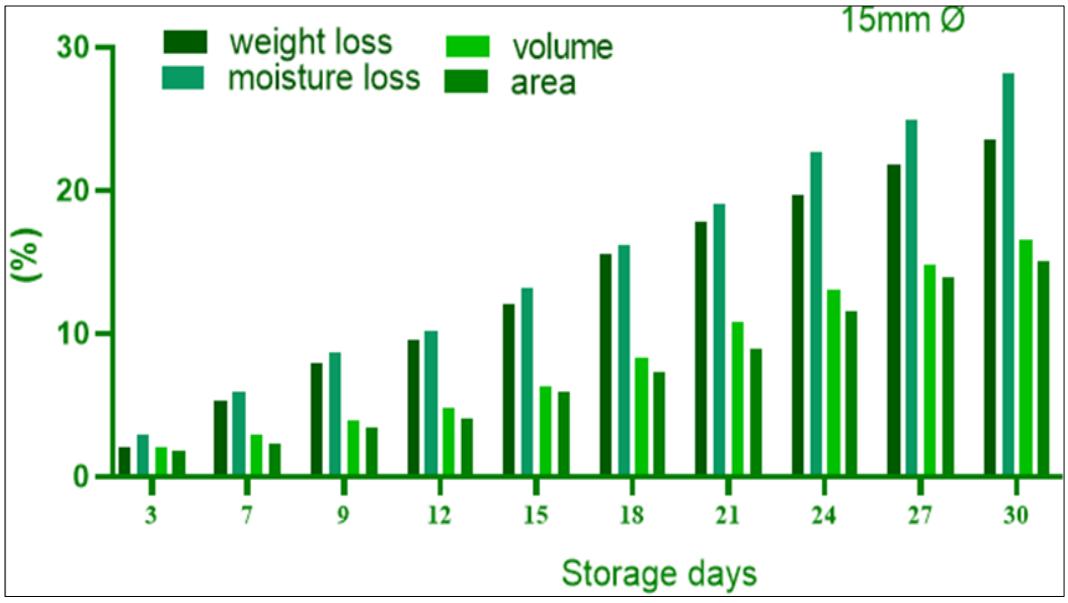


Fig 4: Shrinking characteristics of 15mm Ø bait on refrigerated storage

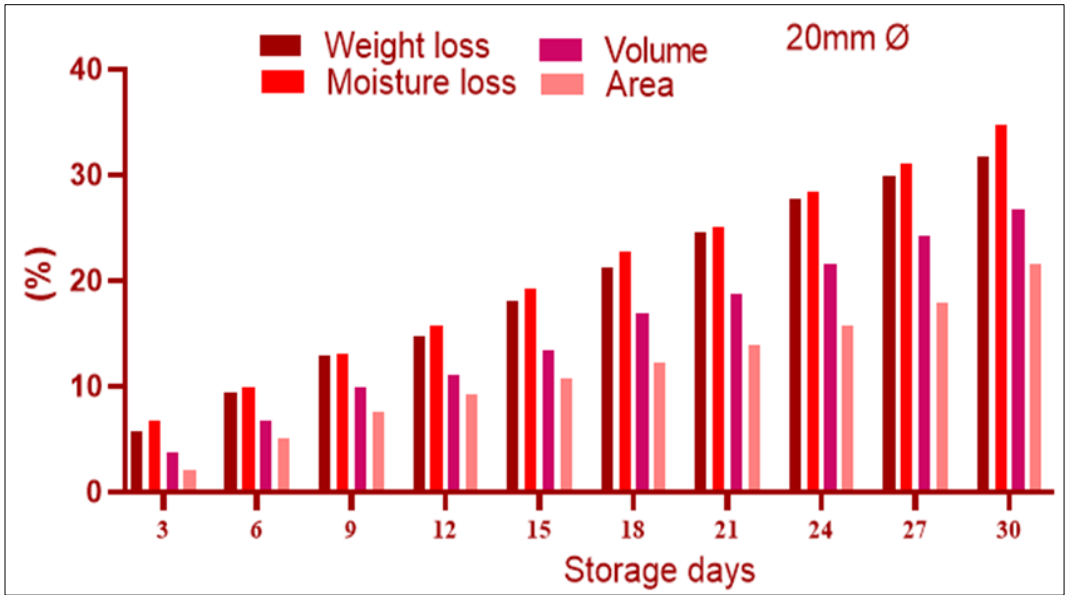


Fig 5: Shrinking characteristics of 20 mm Ø bait on refrigerated storage

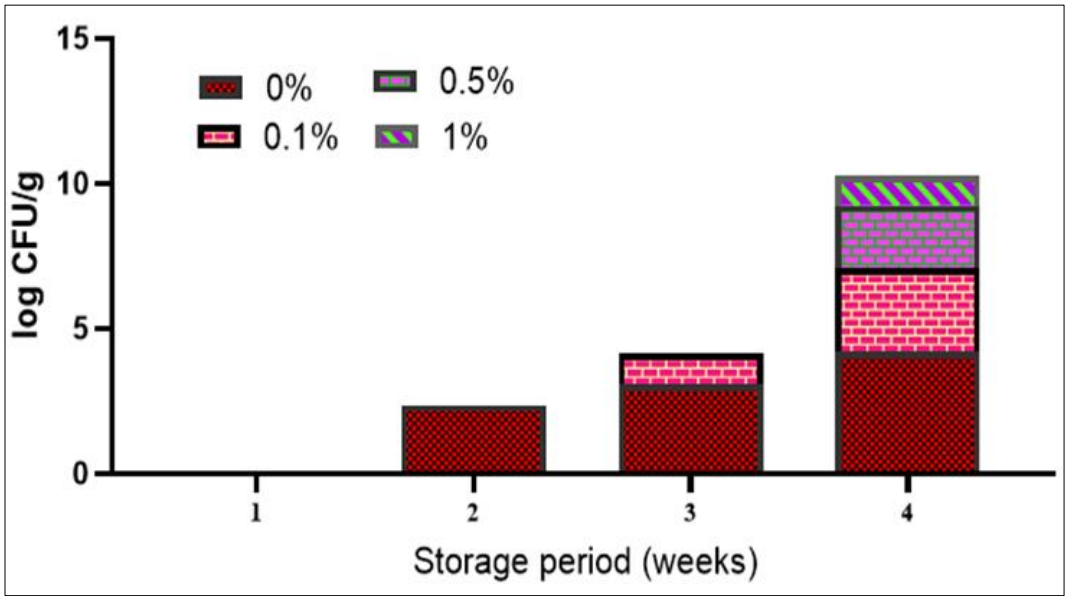


Fig 6: Trend in fungal infection on baits incorporated with different levels of NaB under refrigerated condition

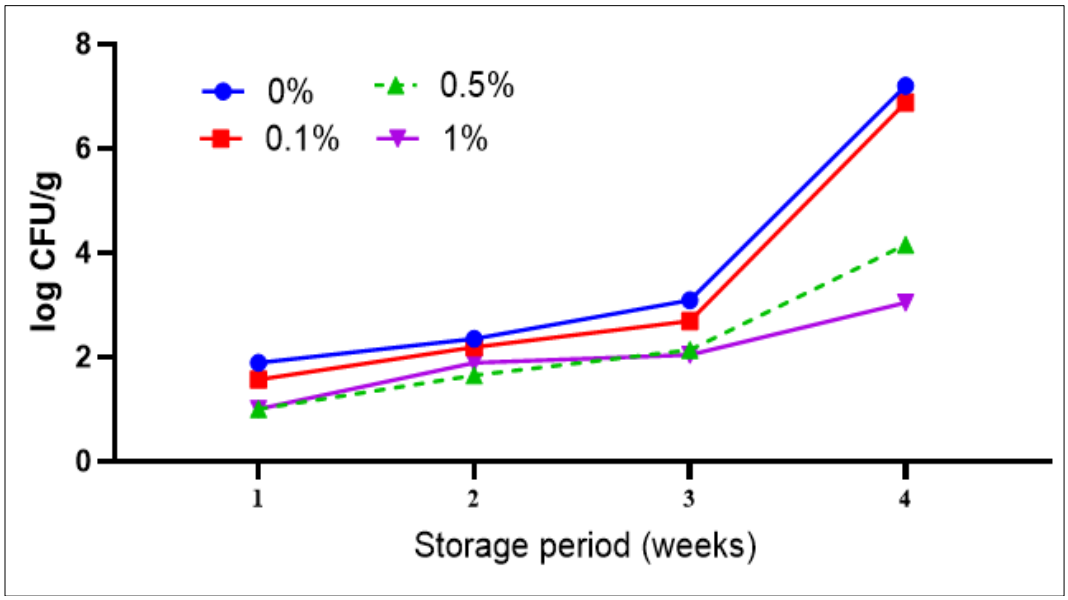


Fig 7: Trend in fungal infection on baits incorporated with different levels of NaB in room temperature.

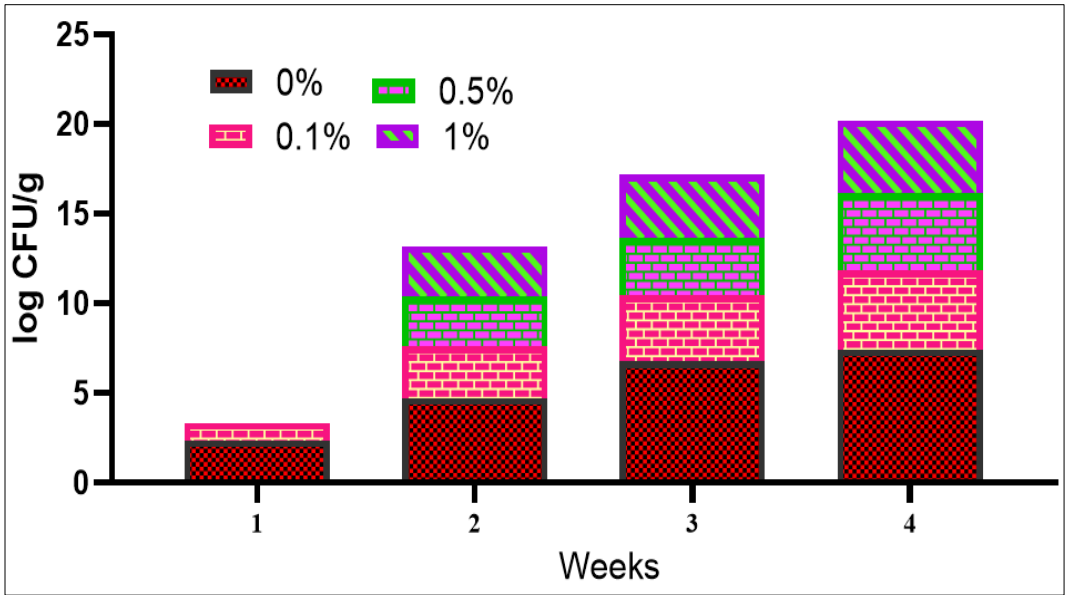


Fig 8: Trend in bacterial infection on baits incorporated with different levels of NaB under refrigerated condition

Conclusion

During storage, the parameters deciding physical quality of the gels such as weight loss, moisture loss, rate of shrinkage were found affected invariably in all the gels with different diameter i.e. 10 mm, 15mm and 20 mm. Regarding shrinkage in volume under refrigerated storage for 30 days, the volume were 3365.66, 6688.35, and 10257.8 mm^3 for 10 mm, 15mm and 20 mm Ø gels respectively. The respective percentages of shrinkage in volume were 15.84%, 16.5% and 26.73%. Similar phenomenon was also noted with respect to weight loss and loss of moisture content of the gels indicating positive correlation between moisture content and shrinkage in volume. The addition of NaB at different levels did not have any visible impact on TPC. Fungal infections could be observed at the end of third week in the gels treated with 0.1% of NaB. However, no fungal infections could be observed in the gels incorporated with NaB at 0.5% and 1% till third week of storage. Irrespective of the levels of incorporation of NaB, fungal infection was evident in all the gels after third week of storage. Storing gels in refrigerator without the addition of antifungal agent could withstand without fungal attack up to one week. However, fungal infection could be visible on eighth day onwards. The gel added with NaB at 1% level did not show any visible impact with respect to fungal growth compared to that treated with 0.5% NaB till third week**.** Hence, it was concluded that in the economic point of view addition of NaB at 0.5% level would be sufficient to delay the onset of fungal infection up to three weeks.

References

1. Ahuja I, Dauksas E, Remme JF, Richardsen R, Loes AK. Fish and fish waste-based fertilizers in organic farming-with status in Norway: A review. Waste Management. 2020;115:95-112. https://doi.org/10.1016/j.wasman.2020.07.025
2. Archdale MV, Anasco CP, Tahara Y. Catches of swimming crabs using fish mince in "teabags" compared to conventional fish baits in collapsible pots. Fisheries Research. 2008;91:291-298. https://doi.org/10.1016/j.fishres.2007.12.005
3. Archdale VM, Kawamura G. Evaluation of artificial and natural baits for the pot fishery of the sand crab *Ovalipespunctatus* (De Haan, 1833). Fisheries Research. 2011;111(3):159-163. https://doi.org/10.1016/j.fishres.2011.07.006
4. Arias-Moscoso JL, Maldonado-Arce A, Rouzaud-Sandez O, Márquez-Ríos E, Torres-Arreola W, Santacruz-Ortega H, *et al*. Physicochemical characterization of protein hydrolysates produced by autolysis of jumbo squid (*Dosidicus gigas*) byproducts. Food Biophysics. 2015;10(2):145-154. https://doi.org/10.1007/s11483-014-9374-z
5. Cavalheiro JMO, Souza EO, Bora PS. Utilization of shrimp industry waste in the formulation of tilapia (*Oreochromis niloticus*, Linnaeus) feed. Bioresource Technology. 2007;98:602-606. https://doi.org/10.1016/j.biortech.2006.02.018
6. Dale T, Siikavuopio SI, Aas K. Development of formulated bait for edible crab (*Cancer pagurus* L.) using by-products from the fisheries and aquaculture industry. Journal of Shellfish Research. 2007;26(2):597-602. https://doi.org/10.2983/0730-8000(2007)26[597:DOFBFE]2.0.CO;2
7. Dellinger A, Plotkin J, Duncan B, Robertson L, Brady T, Kepley C. A synthetic crustacean bait to stem forage fish depletion. Global Ecology and Conservation. 2016;7:238-244. http://dx.doi.org/10.1016/J.GECCO.2016.07.001
8. Ellingsen OF, Døving KB. Chemical fractionation of shrimp extracts inducing bottom food search behavior in cod (*Gadus morhua* L.). Journal of Chemical Ecology. 1986;12:155-168. https://doi.org/10.1007/BF01045599
9. FAO. The State of World Fisheries and Aquaculture. Food and Agriculture Organization of the United Nations, Rome, Italy; c2014.
10. FAO. Food and Agriculture Organization. The state of world fisheries and Aquaculture-(SOFIA). Technical Report; c2019 http://www.fao.org/publications/sofia/en/. Accessed April 5, 2020.
11. Fathiraja P, Gopalrajan S, Karunanithi M, Nagarajan M, Obaiah MC, Durairaj S, *et al*. Response surface

methodology model to optimize concentration of agar, alginate and carrageenan for the improved properties of biopolymer film. Polymer Bulletin; c2021. p. 1-27. https://doi.org/10.1007/s00289-021-03797-5
12. Hara TJ. Smell, taste and chemical sensing. In: Farrell AP, editor. Encyclopedia of Fish Physiology: From Genom to Environment, Academic Press, Elsevier; 2011;1:183-235.
13. Ikeda I, Hosokawa H, Shimeno S, Takeda M. Identification of feeding stimulant for jack mackerel (*Trachurus japonicas*) in its muscle extract. Bulletin of the Japanese Society of Scientific Fisheries; c1988a. Available from: https://agris.fao.org/agris-search/search.do?recordID=JP8805846
14. Ikeda I, Hosokawa H, Shimeno S, Takeda M. Identification of feeding stimulant in the krill extract for jack mackerel. Nippon Suisan Gakkaishi. 1988b;54(2):235-238.
15. Karunanithi M, Neethirajan N, Padmanaban V, Robinson JS, Wilwet L. Evolving artificial fish bait using shrimp processing and poultry wastes. Journal of Entomology and Zoology Studies. 2018a;6(2):2863-2871.
16. Karunanithi M, Neethirajan N, Padmanaban V, Robinson JS. Development of Dry Artificial Fish Bait for Trap Fishing Using Tuna Red Meat and Shrimp Head Wastes. Journal of Aquatic Food Product Technology. 2018b;27(9):1009-1022. https://dx.doi.org/10.1080/10498850.2018.1518944
17. Kasumyan AO, Døving KB. Taste preferences in fishes. Fish and Fisheries. 2003;4:289-347. https://doi.org/10.1046/j.1467-2979.2003.00121.x
18. Kawamura G, Matsuoka T, Tajiri T, Nishida M, Hayashi M. Effectiveness of a sugarcane-fish combination as bait in trapping swimming crabs. Fisheries Research. 1995;22(1-2):155-160. https://doi.org/10.1016/0165-7836(94)00314-M
19. Lee CM. Seafood flavor from processing by-products. In: Maximizing the Value of Marine By-products. Wood head publishing; c2007. p. 304-327. https://doi.org/10.1533/9781845692087.2.304
20. Lokkeborg S, Siikavuopio SI, Humborstad OB, Utne-Palm AC, Ferter K. Towards more efficient long line fisheries: fish feeding behaviour, bait characteristics and development of alternative baits. Reviews in Fish Biology and Fisheries. 2014;24(4):985-1003. http://dx.doi.org/10.1007/S11160-014-9360-Z.
21. Mackie AM, Grant PT, Shelton RGJ, Hepper BT, Walne PR. The relative efficiencies of natural and artificial baits for the lobster *Homarus gammarus*: laboratory and field trials. Journal of the International Council for the Exploration of the Sea. 1980;39:338-345. https://doi.org/10.1093/icesjms/39.2.123
22. Masilan K, Neethiselvan N. A review on natural and artificial fish bait. International Journal of Fisheries and Aquatic Studies. 2018;6(2):198-201.
23. Masilan K, Neethiselvan N, Wiwet L, Jeyashakila R, Vijay K. Storage stability of artificial dry fish bait. International Journal of Current Microbiology and Applied Sciences. 2018;7(5):1177-1183. https://doi.org/10.20546/ijcmas.2018.705.143
24. Masilan K, Neethiselvan N, Jeyashakila R, Sivaraman B, Ravikumar T, Nagarajan M, *et al*. Valorization of discarded industrial fish processing wastes for the extraction of gelatin to use as biodegradable fish bait matrix using RSM. Peer Journal Materials Science. 2021;3:e14. https://doi.org/10.7717/peerj-matsci.14
25. Masilan K, Neethiselvan N, Shakila RJ, Karthy A, Arisekar U, Muralidharan N, *et al*. Development of fish gelatin-based artificial fish baits incorporating bioattractants from seafood processing waste. Journal of the Indian Chemical Society. 2022;99(3):100376. https://doi.org/10.1016/j.jics.2022.100376
26. Masilan K, Neethiselvan N, Shakila RJ, Muralidharan N, Karthy A, Ravikumar T, *et al*. Investigation on the coacervation of fish scale gelatin hydrogel with seafood waste hydrolysates for the development of artificial fish bait: Physico-chemical, thermodynamic, and morpho-structural properties. Journal of the Indian Chemical Society. 2022;99(11):100783. https://doi.org/10.1016/j.jics.2022.100783
27. Miranda CL, Viana MT. Development of artificial lobster baits using fish silage from tuna by-products. Journal of Shellfish Research. 2000;19(1):259-264.
28. MPEDA. MPEDA press release export statistics 2014-15 and RGCA. Marine Product Export Development Authority, Cochin, India; c2015.
29. Siikavuopio SI, Whitaker DR, Martinsen G, Sather BS, Stormo SK. Testing baits prepared from by-product of the shrimp and snow crab industry in the pot fishery for *Gadus morhua* (Linnaeus, 1758) and *Pollachius virens* (Linnaeus, 1758). Journal of Applied Ichthyology. 2017;33(6):1153-1157. https://doi.org/10.1111/jai.13468
30. Siikavuopio SI, James P, Stenberg E, Evensen T, Sather BS. Evaluation of protein hydrolysate of by-product from the fish industry for inclusion in bait for longline and pot fisheries of Atlantic cod. Fisheries Research. 2017;188:121-124. http://dx.doi.org/10.1016/J.FISHRES.2016.11.024.
31. Suresh AV, Kumaraguruvasagam KP, Nates S. Attractability and palatability of protein ingredients of aquatic and terrestrial animal origin, and their practical value for blue shrimp, *Litopenaeus stylirostris* fed diets formulated with high levels of poultry byproduct meal. Aquaculture. 2011;319(1-2):132-140. https://doi.org/10.1016/j.aquaculture.2011.06.039
32. Toften H, Jobling M. Feed intake and growth of Arctic charr, *Salvelinus alpinus* (L.), fed diets supplemented with oxytetracycline and squid extract. Aquaculture Nutrition. 1997;3(4):255-259. https://doi.org/10.1046/j.1365-2095.1997.00098.x
33. Utne-Palm AC, Bogevik AS, Humborstad OB, Aspevik T, Pennington M, Løkkeborg S. Feeding response of Atlantic cod (*Gadus morhua*) to attractants made from by-products from the fishing industry. Fisheries Research. 2020;227:105535. https://doi.org/10.1016/j.fishres.2020.105535
34. Westerberg H, Westerberg K. Properties of odour plumes from natural baits. Fisheries Research. 2011;110(3):459-464. https://doi.org/10.1016/j.fishres.2011.06.002.
35. Yacoob SY, Suresh AV. Attractants: The basics. Global Aquaculture Advocate; c2003. Available from: file:///C:/Users/HP/Downloads/attractants-the-basics.pdf
36. Kim YJ, Uyama H. Biocompatible hydrogel formation of gelatin from cold water fish via enzymatic

networking. Polymer journal. 2007 Oct;39(10):1040-1046.
37. Gómez-Guillén MC, Turnay J, Fernández-Dıaz MD, Ulmo N, Lizarbe MA, Montero P. Structural and physical properties of gelatin extracted from different marine species: a comparative study. Food Hydrocolloids. 2002 Jan 1;16(1):25-34.
38. Muyonga JH, Cole CG, Duodu KG. Fourier transform infrared (FTIR) spectroscopic study of acid soluble collagen and gelatin from skins and bones of young and adult Nile perch (*Lates niloticus*). Food chemistry. 2004 Jul 1;86(3):325-332.
39. Badii F, Howell NK. Fish gelatin: structure, gelling properties and interaction with egg albumen proteins. Food hydrocolloids. 2006 Jul 1;20(5):630-640.
40. Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, *et al*. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. The lancet. 2020 Feb 15;395(10223):497-506.