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Preservative effect of ready to disperse bioactive edible coating powder from fish protein hydrolysate incorporated with chitosan and active clove oil on tuna fillets (*Thunnus albacares*) during chilled storage

K. Sathish Kumar[1,2*], J. Bindu[1], K. Elavarasan[1], Amjad K. Balange[2] and L. Narasimhamurthy[3]

[1]ICAR-Central Institute of Fisheries Technology, Willingdon Island, Kochi-682 029, Kerala, India
[2]ICAR-Central Institute of Fisheries Education, Versova, Andheri West, Mumbai-400 061, Maharashtra, India
[3]National Fisheries Development Board, Hyderabad-500 052, Telangana, India

Abstract

Developing bioactive edible coatings is an emerging trend to improve nutritional value, food quality, and safety. The present work was undertaken to develop ready to disperse bioactive edible coating (BEC) powder from fish protein hydrolysate incorporated with chitosan and clove oil, and to evaluate their preservative effect (10 and 20% level) on tuna fillets during chilled storage at 4°C. Microbiological, biochemical, and sensory evaluation were carried out during 18 days of storage study. Aerobic plate count of tuna fillets treated with 10 and 20% BEC solutions showed 5.45±0.007 log cfu g^{-1} and 5.33±0.009 log cfu g^{-1}, respectively at the end of storage, whereas the control sample reached 9.25±0.004 log cfu g^{-1}. Among the BEC solution treated samples, 20% BEC solution treated sample had the lowest TBARS (Thiobarbituric acid reactive substances) value (1.68±0.04 mg MDA kg^{-1}) followed by 10% BEC solution treated sample (1.93±0.02 mg MDA kg^{-1}) at the end of the storage, while the control sample exceeded the acceptable limit (2.388±0.167248 mg MDA kg^{-1}) on 9th day itself, which gave clear evidence that BEC solutions effectively extended the tuna fillet's shelf-life. The overall study revealed that the sample treated with BEC solutions showed an excellent preservative effect on tuna fillets, which indicates the potential of combined use of these agents in food products.

*Correspondence e-mail:
sathishcife@gmail.com

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Introduction

Fish is categorised under highly perishable commodity and generally fresh fish or shellfish are stored in ice or under refrigerated/chilled conditions during marketing. In these situations, shelf-life is very limited (depends on area of harvest, species, season and post-harvest handling) due to lipid oxidation and microbial deterioration (Bahram *et al.*, 2016). This leads to huge economic loss to fish traders and retailers (Reddy *et al.*, 1995; Getu *et al.*, 2015). Hence, fish processing industries and traders are actively looking for innovative and new technologies or methods to prolong the shelf-life of fresh fish/products during storage as well as marketing stage. In recent times, a variety of active packaging systems have emerged to improve the shelf-life and food safety. Among them, bioactive edible coating has gained more attention due to their several advantages such as edibility, biodegradability, bioactivities, barrier to moisture, gas and microorganisms (Sanchez-Gonzalez *et al.*, 2011; Rennie and Sunjka, 2018). Furthermore, edible coating enhances the nutritional value and sensory characteristics of food products.

Polysaccharides, proteins, and lipids are the three major materials commonly used for the development of edible coatings. Among them, protein based coatings have better advantage due to adherence to the hydrophobic food or meat surface and minimise the oxygen and CO_2 transmission while arresting the drip loss (Rodriguez-Turienzo *et al.*, 2011; Sanchez-Ortega *et al.*, 2014). Protein materials such as keratin and egg albumin (Kilincceker *et al.*, 2009), whey protein (Motalebi and Seyfzadeh, 2012), soy protein (Lin *et al.*, 2011), fish protein (Gulsum and Serkan, 2019), gelatin (Arfat *et al.*, 2015) and collagen (Dursun and Erkan, 2014) can be used for edible coating development. Generally, the seafood processing wastes namely, head, skin, viscera and bone contain high amount of protein, fat and minerals (Jayathilakan *et al.*, 2012). Extraction of protein from seafood waste could address the waste disposal and environmental pollution problems for the industries. It also increases the profit by conversion of waste into valuable fishery products.

Fish protein hydrolysate (FPH) is one of the most high value fishery product of the last decade, due to its application in various sectors such as food, cosmetics, pharmaceutical, nutraceutical and functional food industries. Edible portion or processing discards from finfish or shellfish can be used as a raw material for FPH production. Indian seafood processing industries generate a huge amount of waste which are rich in protein. It can be effectively converted into fish protein hydrolysate which will give higher returns to seafood processor (Elavarasan, 2018). There are several research studies carried out to characterise the functional and bioactive properties of FPH (Shabeena and Nazeer, 2013; Srikanya *et al.*, 2017; Henriques *et al.*, 2021; Undiganalu *et al.*, 2022). However, the studies on bioactive edible coating (BEC) development using FPH in combination with other active ingredients like chitosan and essential oils are rarely reported. FPH as coating material could enrich the nutritional value of food products with improved antioxidant properties (Najafian and Babji, 2012). The addition of essential oils into edible coatings is an effective and innovative method for enhancing their antimicrobial properties. Moreover, it minimises the impact on sensory characteristics of food products and also increases the bioactive substances availability in the food system (Donsì *et al.*, 2011, 2012).

Chitosan is one of the major polysaccharides used for edible films and coatings (Kanatt *et al.*, 2008; Mohamed *et al.*, 2013). The main reasons for adding chitosan into edible films/coatings is due to its ability to provide preservative effect on food products (Ouattara *et al.*, 2000; Jeon *et al.*, 2002). Furthermore, it has antioxidant and antimicrobial properties (Lopez-Caballero *et al.*, 2005), binding action (Shahidi *et al.*, 1999), biodegradability (Chillo *et al.*, 2008) as well as barrier properties against gas and aroma, which makes it more appropriate material for coatings and film development (Caner, 2005).

Incorporation of essential oils and chitosan into FPH based edible coatings will enhance the antioxidant and antimicrobial activity while improving the quality and safety of fishery products. Encapsulation of essential oil using FPH and chitosan could be an effective technique for the slow release of antimicrobial and antioxidant compounds to preserve the products for a longer period. Moreover, the ready to disperse encapsulated powder can be used whenever required by processors, retailer and consumers. With this background, the present work was aimed to prepare a microencapsulated BEC powder having a combination of clove essential oil, FPH and chitosan, and to study their preservative effect on tuna fillets during chilled storage (4°C).

Materials and methods

Freshly caught yellowfin tuna (*Thunnus albacares*) were procured from a fish market in Kochi, and transported to the research laboratory in iced condition. The fish (average weight 20±5 g) were filleted and washed thoroughly and used for storage study. Clove essential oil was purchased from Hi-Media, Mumbai, India. Chitosan was sourced from SRL, Mumbai, India. AR grade chemicals and reagents were used to carry out the present study.

Preparation of fish protein hydrolysate

Fish protein hydrolysate (FPH) was prepared from pink perch head waste based on the method described by Elavarasan and Shamasundar (2016). Initially, pink perch head waste was homogenised into paste form and mixed with distilled water at the ratio of 1:2 (w/v) and incubated in a water bath to attain the optimum temperature needed for the enzyme used. After pre-equilibration, the hydrolysis was initiated by adding the papain enzyme. For hydrolysis process, conditions such as enzyme to substrate ratio (E/S), temperature, pH and time of 0.63%, 55°C, 6 and 90 min, respectively were employed. Then the hydrolysis reaction was terminated by keeping the solution in boiling water bath for 15-20 min. Subsequently, the solution was allowed to cool at room temperature and then filtered using Whatman filter paper. Finally, the supernatant obtained was used for BEC powder preparation.

Preparation of BEC powder and treatment of fillets

To prepare the microencapsulated bioactive edible coating powder, 1% acetic acid (v/v) followed by 1% chitosan (w/v) was added to the FPH solution and stirred for 30 min using magnetic stirrer to achieve complete dispersion. Further, clove oil (1%) (v/v), followed by an emulsifying agent Tween 20 (0.2% of v/v of film forming solution) were added to the solution and continued the stirring for another 30 min. Finally, the bioactive solution was filtered and the residues were removed. The clear BEC solution was spray-dried and powder was stored in an airtight container for further study (Fig. 1).

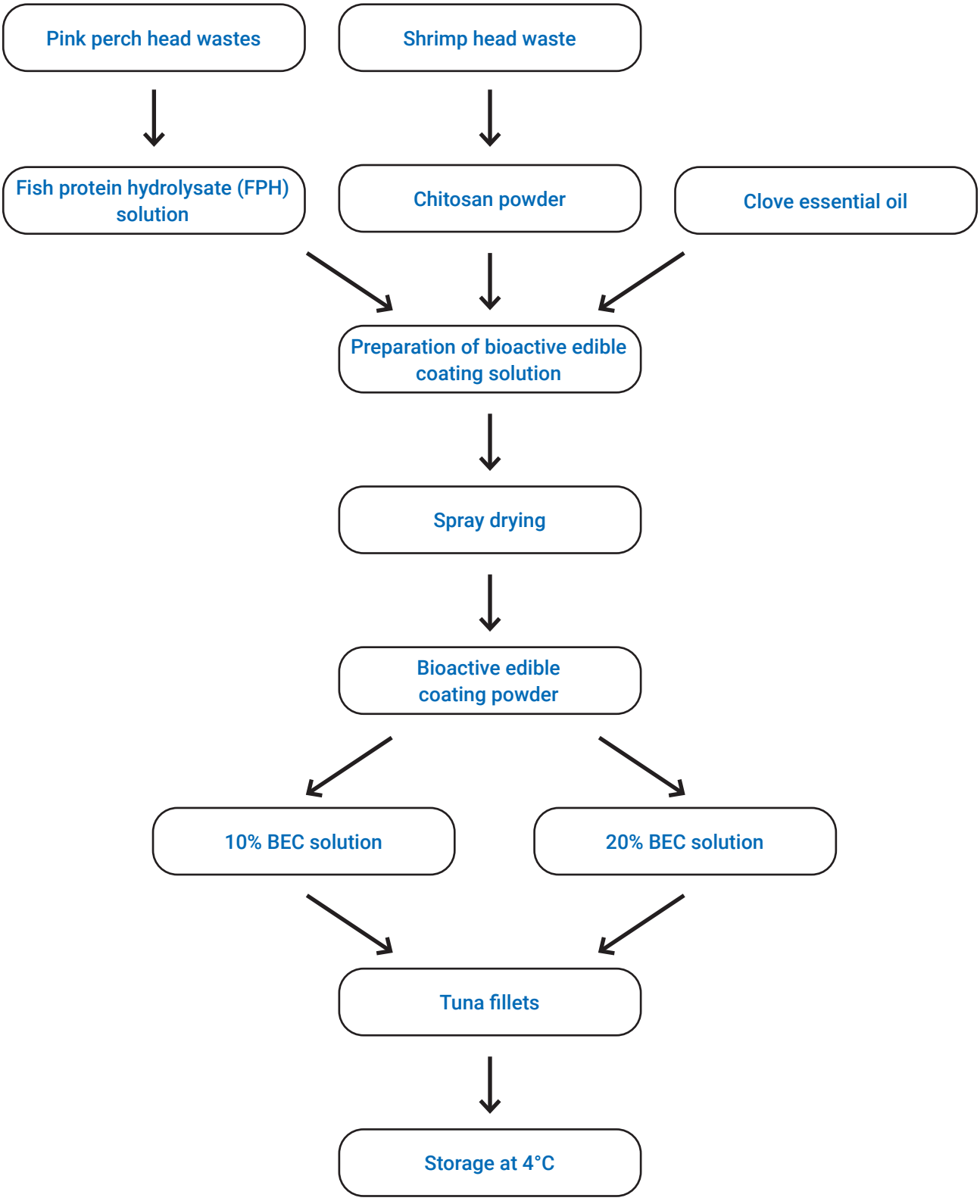

Fig. 1. Schematic diagram for the preparation of BEC powder

For the storage study, tuna fillets were dipped in the BEC solutions at 10 and 20% for 5 mins. Then the fillets were taken out from the solution and the excess solution was allowed to drain off. Finally, the bioactive edible coated tuna fillets were packed in sterile polyethylene pouches and stored at 4°C to study the preservative effect. Samples without BEC solution treatment served as control.

Biochemical analysis

The quality indices such as pH of tuna muscle was measured using a pH meter (Eutech Cyberscan 510, Singapore) by homogenising 5 g of tuna muscle in distilled water (1:5 w/v). Thiobarbituric acid (TBA) value was estimated as per the method prescribed by Tarladgis *et al.* (1960) and the values were expressed as mg of malonaldehyde (MDA) kg^{-1} of lipid. Total volatile base nitrogen (TVBN) and trimethylamine (TMA) were determined based on the micro diffusion method (Conway, 1962). Peroxide value (PV) was calculated according to the AOAC methods (AOAC, 2000) and the values were expressed as meq kg^{-1} of lipid.

Microbiological analysis

Spread plate technique was carried out to estimate the aerobic plate count (APC) (Hitching *et al.*, 1995). Approximately 10 g

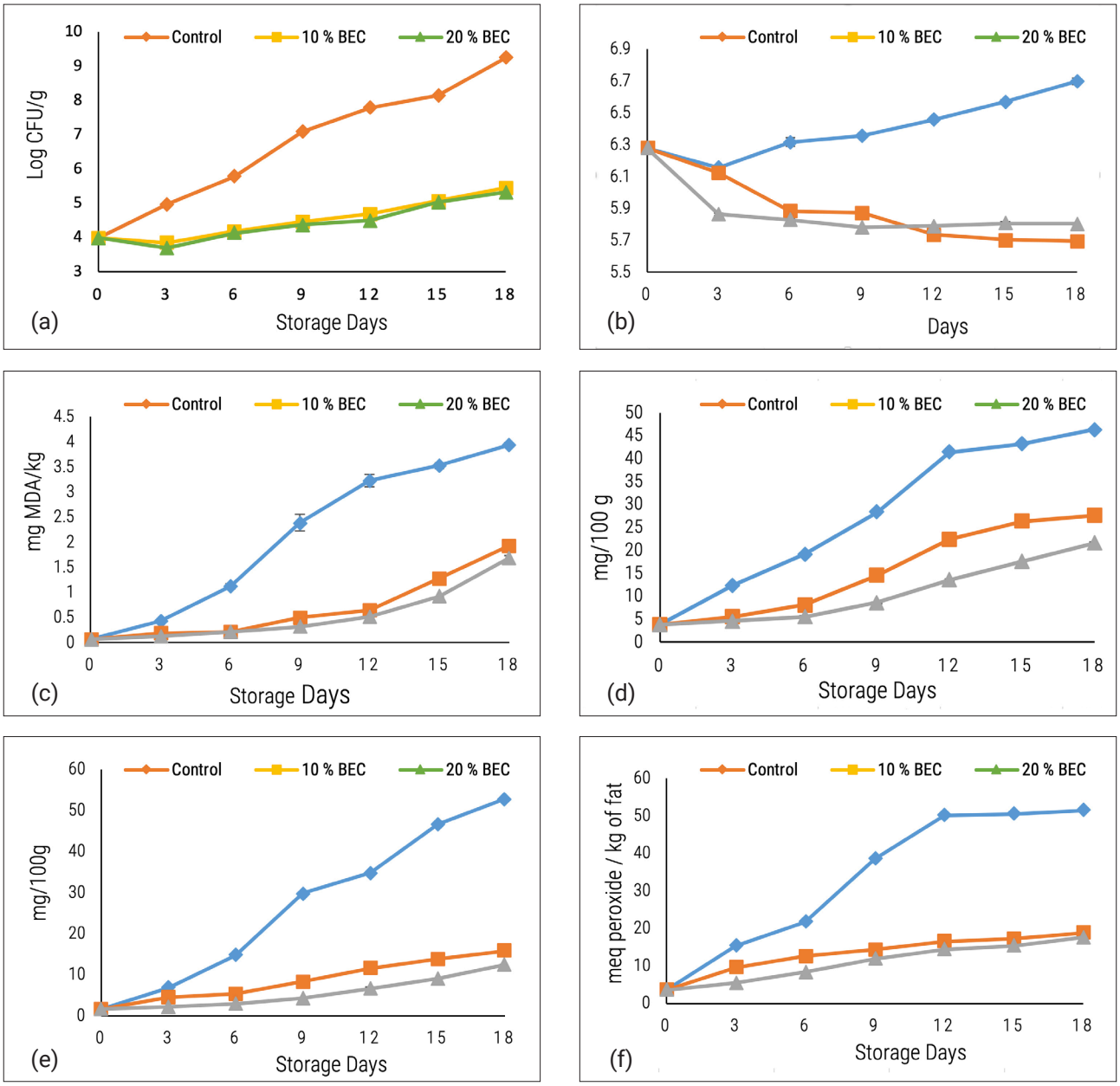

Fig. 2. Microbial and biochemical changes of tuna fillets during chilled storage (a) APC, (b) pH, (c) TBARS, (d) TVB-N, (e) TMA and (f) PV

of tuna fillet sample was aseptically homogenised with 90 ml of sterile normal saline using mortar and pestle for 1 min. Then, the supernatant (1 ml) was transferred into a tube containing 9 ml sterile normal saline and mixed thoroughly using vortex mixer. Further serial dilutions and spread plate technique was carried out in plate count agar and incubated at 37°C for 48 h and the average count was expressed as cfu g^{-1} of sample.

Sensory analysis

Sensory evaluation was carried out based on the 9-point hedonic scale defined by Meilgaard *et al.* (1999). The control and BEC solution treated samples were taken out randomly and provided to panelists with a sensory evaluation sheet to evaluate the overall acceptability. Sensory attributes like colour, appearance, odour and textural values were used to evaluate the overall acceptability. An overall acceptance score of 6 and above was considered acceptable. The average overall acceptability value of the 15 semi-trained panelists for each sample was calculated and the values were expressed as Mean ± Standard Deviation (n=15).

Results and discussion

Biochemical analysis

Changes in the pH of tuna fillets dipped in BEC solutions during chilled storage are shown in Fig. 2b. The pH of 10 and 20% BEC solutions dropped from the initial value of 6.28±0.00 to 5.69±0.005 and 5.80±0.005, respectively. The control sample showed an immediate dip on 3rd day of storage (6.15±0.005), later increased gradually to 6.69±0.02 on 18th day of chilled storage whereas BEC solutions treated samples showed a decrease in pH value throughout the storage period. A significant decrease in pH was observed in both 10 (5.69±0.005) and 20% BEC solutions (5.80±0.005) treated samples during 18 days chilled storage, which could be due to the production of lactic acid through anaerobic glycolysis process and the release of inorganic phosphate, a product of ATP degradation. The control sample reached a pH of 6.69±0.02 on the 18th day of storage, which may be due to the enzymatic degradation of the cellular contents of the muscle (Bo Lan *et al.*, 2018). According to Salgado *et al.* (2011), pH increase during storage period due to production of alkaline compounds, like biogenic amines and ammonia, by the action of microbial and endogenous enzymes, are responsible for fish spoilage. Compared to control samples, a lower pH value was observed in tuna fillets treated with BEC solutions, which reveals that BEC solutions effectively minimise the formation of alkaline compounds during storage.

TBARS value is most commonly used to measure the lipid oxidation (Al-Kahtani *et al.*, 1996; Yanar *et al.*, 2006). Changes in the TBARS value of BEC solutions treated tuna fillets at chilled storage (4°C) are shown in Fig. 2c. Initial TBARS value of tuna fillets was 0.068±0.00 mg MDA eq kg^{-1} muscle. As the storage period progressed, rapid lipid oxidation was observed in the control samples, in which TBARS value amplified into 2.38±0.16 and 3.94±0.04 mg MDA kg^{-1} muscle on Day 9 and Day 18, respectively. While, the TBARS values of 10 and 20% BEC solution treated samples gradually increased from 0.068±0.00 to 1.93±0.02 mg MDA kg^{-1} and 1.68±0.04 mg MDA kg^{-1} respectively on 18th day of storage. Among the treatments, fillets treated with 20% BEC solution treated samples showed the lowest TBARS value (1.68±0.04 mg MDA kg^{-1}) followed by 10% BEC solution treated samples (1.93±0.02 mg MDA kg^{-1}), which indicated that tuna fillets dipped with BEC solutions effectively delayed oxidation of lipids during storage. The acceptable limit of TBARS values is 1-2 mg MDA kg^{-1} of fish muscle (Connell, 1990). In this present study, TBARS values of the sample treated with 10 and 20% BEC solutions were within the acceptable limit at the end of 18 days storage, while the control exceeded the limit (2.388±0.167248 mg MDA kg^{-1}) on the 9th day itself.

TVB-N value of BEC solutions treated tuna fillets during chilled storage are shown in Fig. 2d. TVB-N values of samples treated with 10 and 20% BEC solution increased gradually from 3.71±0.00 to 27.61±0.27 mg 100 g^{-1} and 21.59±0.19 mg 100 g^{-1} respectively, compared to control value of 46.31±0.09 mg 100 g^{-1} after 18 days of storage. Generally, the recommended TVB-N level for iced fish is 30-35 mg TVB-N 100 g^{-1} of muscle (Connell, 1995). In the present study, none of the samples treated with BEC solutions, exceeded the acceptable limit during the storage. However, the control sample exceeds the limit (41.35±0.24 mg 100 g^{-1}) on the 12th day itself. Generally, TVB-N values are used to assess the quality index of fishery products, particularly as it is a product of bacterial spoilage (Lannelongue *et al.*, 1982). The present study showed that BEC solutions effectively minimised bacterial growth during chilled storage and extended the shelf-life of tuna fillets.

TMA values of BEC solutions treated tuna fillets stored in chilled storage are shown in Fig. 2e. TMA values of samples treated with 10 and 20% BEC solutions increased from 1.55±0.00 to 15.80±0.24 mg 100 g^{-1} and 12.35±0.06 mg 100 g^{-1}, respectively at the end of storage as compared to control sample which ranged from 1.55±0.00 to 52.77±0.14 mg 100 g^{-1}. TMA level of 10 to 15 mg 100g^{-1} is recommended as acceptable limit for fish (Connell, 1995). In the present study, tuna fillet treated with 10% BEC solution slightly crossed the limit (15.80±0.24 mg 100 g^{-1}) on the 18th day of storage, but 20% BEC solution treated samples were within the acceptable limit (12.35±0.06 mg 100 g^{-1}). Control samples crossed the acceptable limit on 9th day of storage. The presence of TMA is due to the bacterial degradation of trimethylamine oxide (TMAO) which is naturally present in marine fishes. TMA level is used as a specific index of bacterial spoilage. The study revealed that BEC solutions effectively reduce TMA production during storage as compared with control samples.

Peroxide value is one of the indicators for oxidative rancidity (Shahidi and Wanasundara, 1996). The peroxide values of

tuna fillets treated with 10 and 20% BEC solutions increased from 3.51±0.00 to 18.82±0.02 meq kg^{-1} of fat and 17.51±0.05 meq kg^{-1} of fat, respectively as compared to control having PV value of 51.44±0.06 meq kg^{-1} of fat after 18 days storage (Fig. 2f). In the present work, it was observed that the BEC solution effectively reacted with oxygen present in the environment leading to reduction in hydroperoxide formation (Bensid *et al.*, 2014), which delayed the lipid oxidation during storage.

Microbiological analysis

Changes in the total bacterial counts of tuna fillets treated with BEC solutions increased steadily during chilled storage (Fig. 2a). APC of tuna fillets treated with 10 and 20% BEC solution increased from the initial level of 3.99±0.005 to 5.45±0.007 log cfu g^{-1} and 5.33±0.009 log cfu g^{-1}, respectively on 18th day of storage, whereas control sample reached 9.25±0.004 log cfu g^{-1} on 18th day of storage. Bacterial counts were low at the beginning of storage due to the freshness of the fish. The initial APC of tuna fillet (3.99±0.004 log cfu g-1) was within acceptable limit of fresh fish (5 x 10^5 cfu g^{-1}) (ICMSF, 1986), and it gradually increased as storage period proceeded in all the samples. Control sample reached the APC of 5.78±0.004 log cfu g^{-1} on the 6th day of storage, whereas the samples treated with BEC solutions reached >5 log cfu g^{-1} on the 15th day of storage. This is a clear evidence that BEC solutions effectively delayed the bacterial growth during chilled storage by prolonging its lag phase. Among the two BEC solutions, 20% BEC solution indicated low APC value of 5.33±0.009 log cfu g^{-1} followed by 10% BEC solution (5.45±0.007 log cfu g^{-1}) at the end of the storage period, which demonstrated the potential of BEC powder as a natural antimicrobial agent. The active component like eugenol is the main reason behind the antimicrobial properties of clove oil which was incorporated in the BEC solution. It disrupts the cytoplasmic membrane of bacterial cells and causes cell damage or rupture which leads to cell death (Broxton *et al.*, 1983; Gill and Holley, 2006). Clove oil possesses antimicrobial activity against both Gram negative bacteria such as *Escherichia coli*, *Salmonella* sp., *Pseudomonas aeruginosa*, *Aeromonas hydrophila* and Gram positive bacteria like *Staphylococcus aureus*, *Listeria* sp. and *Bacillus* sp. (Gislene *et al.*, 2000; Qiao Hu *et al.*, 2018). Similar results were observed in other studies related to bioactive edible coating combined with chitosan and/or other essential oils on rainbow trout (Ozlem *et al.*, 2018) and cold-smoked sardine (Gomez-Estaca *et al.*, 2007).

Sensory evaluation

Sensory evaluation of tuna fillets dipped in BEC solutions are shown in Fig. 3. There was a gradual decrease in overall acceptability score of all samples during the storage study. Initially, the tuna fresh fillets had a sensory value of 8.5±0.005 which decreased gradually as the storage days prolonged, according to the BEC treatments. Control samples exceeded the acceptable limit on the 6th day of storage (<6.00), whereas the sample treated with 10 and 20% BEC solutions crossed the acceptable limit on 9th (5.29±0.39) and 12th (4.91±0.28) days of storage, respectively. The result of sensory value directly correlated with the microbial evaluation. This indicates that the BEC solutions effectively extended the shelf-life of tuna fillets as compared with control sample.

Development of bioactive edible coating from fishery waste materials is one of the emerging trends to reduce environmental pollution. This study revealed that BEC powder developed from fish protein hydrolysate combined with chitosan and clove essential oil was more effective to maintain the quality of tuna fillets during chilled storage. The quality and safety analysis clearly indicated that the samples treated with BEC solutions were highly efficient and extended the shelf-life of tuna fillets compared to control samples, which indicated the potential of combined usage of these agents. However, the potential application of fish protein hydrolysate and clove oil should be further studied, especially in terms of the sensory acceptability, since these

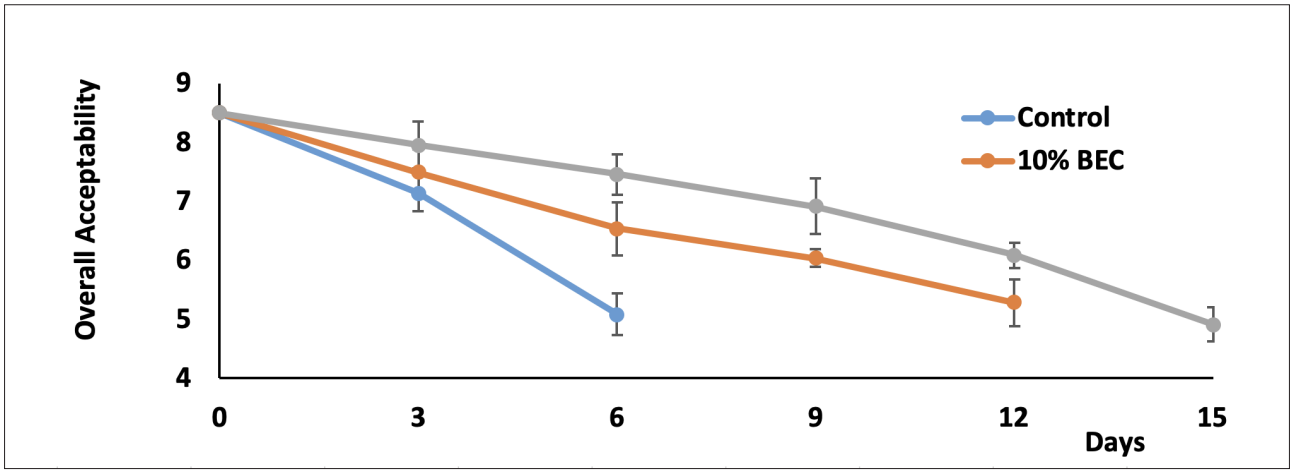

Fig. 3. Sensory changes of tuna fillets during chilled storag

materials may impart unpleasant attributes (bitter taste and/or strong odour) to fish or fishery products.

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References

Al-Kahtani, H. A., Abu Tarboush, H. M., Bajaber, A. S., Atia, M., Abou Arab, A. A. and El Mojaddidi, M. A. 1996. Chemical changes after irradiation and post-irradiation storage in tilapia and Spanish mackerel. *J. Food Sci*., 61: 729-733.

AOAC 2000. Peroxide value in oils and fats/Pearson's composition and analysis of food, *Official methods of analysis of Association of Official Analytical Chemists*, 17th edn. Association of Official Analytical Chemists (AOAC) International, Maryland, USA, 641 p.

Arfat, Y. A., Benjakul, S., Vongkamjan, K., Sumpavapol, P. and Yarnpakdee, S. 2015. Shelf-life extension of refrigerated seabass slices wrapped with fish protein isolate/fish skin gelatin ZnO nanocomposite film incorporated with basil leaf essential oil. *J. Food Sci. Technol.,* 52: 6182-6193. https://doi.org/ 10.1007/s13197-014-1706-y

Bensid, A., Ucar, Y., Bendeddouche, B. and Ozogul, F. 2014. Effect of the icing with thyme, oregano and clove extracts on quality parameters of gutted and beheaded anchovy (*Engraulis encrasicholus*) during chilled storage. *Food Chem*., 145: 681-686. https://doi.org/10.1016/j.foodchem.2013.08.106

Bo Lan, Jennifer, A. M., Michael, J. O'Sullivan, Chan, Y. P., Jae, H. K., William, C. C., James, P. B., and Jin-Ah, P. 2018. Airway epithelial compression promotes airway smooth muscle proliferation and contraction. *Am. J. Physiol. Lung Cell Mol. Physiol*. 315: L645-L652. https://doi.org/10.1152/ajplung.00261.2018

Broxton, P., Woodcock, P. M. and Gilbert, P. A. 1983. Study of the antibacterial activity some polyhexamethylene biguanides towards *Escherichia coli* ATCC8739. *J. Appl. Bacteriol*., 54: 345-353. https://doi.org/10.1111/j.1365-2672.1983.tb02627.x

Caner, C. 2005. The effect of edible eggshell coatings on egg quality and consumer perception. *J. Sci. Food Agric*., 85: 1897-1902. https://doi.org/10.1002/jsfa.2185

Chillo, S., Flores, S., Mastromatteo, M., Conte, A., Gerschenson, L. and Del Nobile, M. A. 2008. Influence of glycerol and chitosan on tapioca starch based edible film properties. *J. Food Eng*., 88: 159-168. https://doi.org/10.1016/j.jfoodeng.2008.02.002

Connell, J. J. 1990. Methods of assessing and selecting for quality. In: Connell, J. J. (Ed.), *Control of fish quality*. Fishing News Books, Oxford, UK, pp. 122-150

Connell, J. J 1995. *Control of fish quality,* 4th edn. Churchill Livingstone, Edinburg, Scotland, 245 p.

Conway, E. J. 1962. *Micro diffusion analysis and volumetric error*, 5th edn. Crosby Lockwood and Son Ltd., London, UK.

Donsì, F., Annunziata, M., Sessa, M. and Ferrari, G. 2011. Nanoencapsulation of essential oils to enhance their antimicrobial activity in foods. *LWT-Food Sci. Technol*., 44: 1908-1914. https://doi.org/10.1016/j.lwt.2011.03.003

Donsì, F., Annunziata, M., Vincensi, M. and Ferrari, G. 2012. Design of nanoemulsion-based delivery systems of natural antimicrobials: Effect of the emulsifier. *J. Biotechnol.,* 159: 342-350. https://doi.org/10.1016/j.jbiotec.2011.07.001

Dursun, S. and Erkan, N. 2014. The effect of edible coating on the quality of smoked fish. *Italian J. Food Sci*., 26(4): 370-382.

Elavarasan, K. 2018. Protein hydrolysates from fish processing waste: Health benefits and their potential application. In: Bindu, J., Sreejith, S. and Sarika, K. (Eds.), *Protocols for the production of high value secondary products from industrial fish and shellfish processing*, ICAR-Central Institute of Fisheries Technology, Kochi, India, pp. 1-17.

Elavarasan, K. and Shamasundar, B. A. 2016. Effect of oven drying and freeze drying on the antioxidant and functional properties of protein hydrolysates derived from freshwater fish (*Cirrhinus mrigala*) using papain enzyme. *J. Food Sci. Technol.,* 53(2): 1303-1311. https://doi.org/10.1007/s13197-015-2084-9

Getu, A., Misganaw, K. and Bazezew, M. 2015. Post-harvesting and major related problems of fish production. *Fish. Aquac. J.,* 6: 4. https://doi.org/DOI: 10.4172/2150-3508.1000154

Gill, A. O. and Holley, R. A. 2006. Disruption of *Escherichia coli*, *Listeria monocytogenes* and *Lactobacillus sakei* cellular membranes by plant oil aromatics. *Int. J. Food Microbiol*., 108: 1-9. https://doi.org/DOI:10.1016/j.ijfoodmicro.2005.10.009

Gislene, G. F., Paulo, C. and Giuliana, L. 2000. Antibacterial Activity of Plant Extracts and Phytochemicals on antibiotic resistant bacteria. *Braz. J. Microbiol*., 31: 314-325. https://doi.org/10.1590/S1517-83822000000400003

Gomez-Estaca, J., Montero, P., Gimenez, B. and Gomez-Guillen, M. C. 2007. Effect of functional edible films and high pressure processing on microbial and oxidative spoilage in cold-smoked sardine (*Sardina pilchardus*). *Food Chem*., 105: 511-520. https://doi.org/10.1016/j.foodchem.2007.04.006

Gulsum, B. M. and Serkan, K. 2019. Effects of edible coatings based on ultrasound-treated fish proteins hydrolysate in quality attributes of chilled bonito fillets. *J. Aquat. Food Product Technol.*, 28(10): 999-1012. https://doi.org/10.1080/10498850.2019.1681572

Henriques, A., Vázquez, J. A., Valcarcel, J., Mendes, R., Bandarra, N. M. and Pires, C. 2021. Characterization of protein hydrolysates from fish discards and by-products from the North-west Spain fishing fleet as potential sources of bioactive peptides. *Mar. Drugs*, 19: 338. https://doi.org/10.3390/md19060338

Hitching, A. D., Feng, P., Matkins, W. D., Rippey, S. R. and Chandler, L. A. 1995. Aerobic plate count. In: Tomlinsion, L. A. (Eds.), *Bacteriological analytical manual*, 18th edn. Association of Official Analytical Chemists (AOAC) International, Maryland, USA. pp. 4.01-4.29.

ICMSF 1986. Microorganisms in foods. In: *Sampling for microbiological analysis: Principles and scientific applications*. International Commission on Microbiological Specifications for Foods (ICMSF), University of Toronto, Toronto, Canada, 188 p.

Jayathilakan, K., Sultana, K., Radhakrishna, K. and Bawa, A. S. 2012. Utilization of byproducts and waste materials from meat, poultry and fish processing industries: A review. *J. Food Sci. Technol*., 49(3): 278-293. https://doi.org/10.1007/s13197-011-0290-7

Jeon, Y. I., Kamil, J. Y. V. A. and Shahidi, F. 2002. Chitosan as an edible invisible film for quality preservation of herring and Atlantic cod. *J. Agric. Food Chem*., 20: 5167-5178. https://doi.org/10.1021/jf011693l

Kanatt, S. R., Chander, R. and Sharma, A. 2008. Chitosan and mint mixture: A new preservative for meat and meat products. *Food Chem*., 107: 845-852. https://doi.org/10.1016/j.foodchem.2007.08.088

Kilincceker, O., Dogan, I. S. and Kucukoner, E. 2009. Effect of edible coatings on the quality of frozen fish fillets. *LWT-Food Sci. Technol*., 42(2): 868-873. https://doi.org/10.1016/j.lwt.2008.11.003

Lannelongue, M., Hanna, M. O., Finne, G., Nickelson, R. and Vanderzant, G. 1982. Storage characteristics of finfish fillets (*Archosarqgus probatocephalus*) packed in modified gas atmospheres containing carbon dioxide. *J. Food Protect*., 45: 440-444. https://doi.org/10.4315/0362-028X-45.5.440

Lin, L., Wang, B. and Weng, Y. 2011. Quality preservation of commercial fish balls with antimicrobial zein coatings. *J. Food Quality*, 34: 81-87. https://doi.org/10.1111/j.1745-4557.2011.00370.x

Lopez-Caballero, M. E., Gomez-Guillen, M. C., Perez-Mateos, M. and Montero, P. 2005. A chitosan-gelatin blend as a coating for fish patties. *Food Hydrocoll*., 19: 303-311. https://doi.org/10.1016/j.foodhyd.2004.06.006

Meilgaard, M., Civille, G. V. and Carr, B. T. 1999. *Sensory evaluation techniques*, CRC Press, Boca Raton, Florida, USA, 387 p.

Mohamed, C., Clementine, K. A., Didier, M., Gerard, L. and Noelle, D. C. M. 2013. Antimicrobial and physical properties of edible chitosan films

enhanced by lactoperoxidase system. *Food Hydrocoll.*, 30: 576-580. https://doi.org/10.1016/j.foodhyd.2012.07.018

Motalebi, A. A. and Seyfzadeh, M. 2012. Effects of whey protein edible coating on bacterial, chemical and sensory characteristics of frozen common kilka (*Clupeonellia delitula*). *Iranian J. Fish. Sci*., 11(1): 132-44.

Najafian, L. and Babji, A. S. 2012. A review of fish-derived antioxidant and antimicrobial peptides: Their production, assessment, and applications. *Peptides*, 33: 178-185. https://doi.org/10.1016/j.peptides.2011.11.013

Ouattara, B., Simard, R. E., Piette, G., Begin, A. and Holley, R. A. 2000. Inhibition of surface spoilage bacteria in processed meats by application of antimicrobial films prepared with chitosan. *Int. J. Food Microbiol*., 62: 139-148. https://doi.org/10.1016/s0168-1605(00)00407-4

Ozlem, P. C., Halil, Y. and Ali, A. 2018. Effects of chitosan coating and rosemary oil on rainbow trout (*Oncorhynchus mykiss*, W. 1792) filets. *Indian J. Animal Res*., 52(1): 160-166.

Qiao Hu, Meifang Zhou and Shuyong Wei 2018. Progress on the antimicrobial activity research of clove oil and eugenol in the food antisepsis field. *J. Food Sci.,* 83(6): 1476-1483. https://doi.org/10.1111/1750-3841.14180

Reddy, N. R., Villanueva, M. and Kautter, D. A. 1995. Shelf-life of modified-atmosphere-packaged fresh tilapia fillets stored under refrigeration and temperature-abuse conditions. *J. Food Prot.,* 58: 908-914.

Rennie, T. J. and Sunjka, P. S. 2018. Modified atmosphere for storage, transportation and packaging. In: Pareek, S. (Eds.), *Novel postharvest treatments of fresh produce,* CRC Press/Taylor and Francis Group, Boca Raton, Florida, USA, 433-480 p.

Rodriguez-Turienzo, L., Cobos, A., Moreno, V., Caride, A., Vieites, J. M. and Diaz, O. 2011. Whey protein-based coatings on frozen Atlantic salmon (*Salmo salar*): Influence of the plasticiser and the moment of coating on quality preservation. *Food Chem*., 128: 187-194. https://doi.org/10.1016/j.foodchem.2011.03.026

Salgado, P. R., Fernandez, G. B., Drago, S. R. and Mauri, A. N. 2011. Addition of bovine plasma hydrolysates improves the antioxidant properties of soybean and sunflower protein-based films. *Food Hydrocolloids*, 25: 1433-1440. https://doi.org/10.1016/j.foodhyd.2011.02.003

Sanchez-Gonzalez, L., Vargas, M., Gonzalez-Martınez, C., Chiralt, A. and Chafer, M. 2011. Use of essential oils in bioactive edible coatings. *Food Eng. Rev.,* 3: 1-16.

Sanchez-Ortega, I., García-Almendárez, B. E., Santos-Lopez, E. M., Amaro-Reyes, A., Barboza-Corona, J. E. and Regalado, C. 2014. Antimicrobial edible films and coatings for meat and meat products preservation. *The Sci. World J*., 1-18.

Shabeena Y. N. and Nazeer, R. A. 2013. Antioxidant and functional properties of protein hydrolysates from pink perch (*Nemipterus japonicus*) muscle. *J. Food Sci. Technol*., 50(5): 972-978. https://doi.org/ 10.1007/s13197-011-0416-y

Shahidi, F., Arachchi, J. K. V. and Jeon, Y. J. 1999. Food applications of chitin and chitosans. *Trends Food Sci. Technol*., 10(2): 37-51. https://doi.org/10.1016/S0924-2244(99)00017-5

Shahidi, F. and Wanasundara, U. N. 1996. Review-methods for evaluation of the oxidative stability of lipid-containing foods. *Food Sci. Technol. Intl*., 2: 73-81.

Srikanya, A., Dhanapal, K., Sravani1, K., Madhavi, K. and Praveen Kumar, G. 2017. A study on optimization of fish protein hydrolysate preparation by enzymatic hydrolysis from tilapia fish waste mince. *Intl. J. Current Microbiol. Appl. Sci*., 6(12): 3220-3229. https://doi.org/10.20546/ijcmas.2017.612.375

Tarladgis, G. B., Watts, M. B. and Younathan, T. M. 1960. A distillation method for the quantitative determination of malonaldehyde in rancid foods. *J. Am. Oil Chemists Soc.,* 37: 44-50. https://doi.org/10.1007/BF02630824

Undiganalu, G. Y.., Sneha, V. and Mamatha B. S. 2022. Functional properties of protein hydrolyzate from ribbon fish (*Lepturacanthus savala*) as prepared by enzymatic hydrolysis. *Int. J. Food Propert.*, 25(1): 187-203. https://doi.org/10.1080/10942912.2022.2027964

Yanar, Y., Celik, M. and Akamca, E. 2006. Effects of brine concentration on shelf-life of hot-smoked tilapia (*Oreochromis niloticus*) stored at 4°C. *Food Chem*., 97: 244-247. https://doi.org/10.1016/j.foodchem.2005.03.043