Fishery Technology 55 (2018) : 114 - 119

Chilled Storage Studies of Depurated Cooked Clam Meat in two different Packaging Materials

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Abstract

The biochemical and microbiological parameters of depurated cooked meat of black clam (*Villorita cyprinoides*) (Gray, 1825) was assessed for the extension of shelflife in iced condition in two different packaging materials *viz*., Polyester polyethylene laminate (PEST-PE) and low density polyethylene (LDPE). Aerobic plate count revealed that the shelflife of black clam was 22 days in PEST-PE and 16 days in LDPE during which the count reached 5.38 log cfug^{-1} and 4.9 log cfug^{-1}, respectively. The clam meat stored in LDPE had a TBA value of 1.36 mg MDAkg^{-1} on 16th day and it was 1.23 mg MDAkg^{-1} for PEST-PE packed meat on the 22nd day. The values of TVB-N was 7.0 mg% and 7.4 mg% after 16 days and 22 days in the LDPE and PEST-PE, respectively. Since the value of APC was 5.38 log cfug^{-1} on the 16th day for clam meat packed in LDPE it was not analysed further as the microbiological count exceeded the limit of acceptability. From this study it is inferred that the shelflife of clam meat may be extended by proper icing after packaging in suitable packing material like PEST-PE.

Keywords: Clam meat, chilled storage studies, PEST-PE, LDPE

Introduction

Bivalves like mussels, oysters and clams play an important role in the food security of the population even though their contribution is very low when compared to the contribution from the finfishes. Apart from meeting the protein deficiency of the country, some of them command a very good price in the overseas market. They are mostly accessible due to their sedentary habit and ease of harvesting operations. Among the clam species, the important species in the Ashtamudi and Vembanad lakes of Kerala are yellowfoot clam (*Paphia malabarica*), *Meretrix casta* (yellow clam) and black clam (*V. cyprinoides)*. The black clam production in Vembanad lake during 2015 was estimated as 57905 t (CMFRI, 2015). It fetches low price compared to the yellow clam due to its low aesthetic appeal and presence of gritty materials (Bindu et al., 2007). The *Villorita* species became established as a new economic resources of the Perumbalam area of the Vembanad lake after the construction of Thanneermukkom bund (Thomson, 2009). The shell of clam is used mainly in the production of lime and a small amount goes to the ornamental industry. The clam meat contributes to the subsistence fishery of local coastal communities (Sathiadas et al*.,* 2004)

The fish and fishery products are susceptible to spoilage due to the growth of psychrotrophic spoilage organisms (Brody, 1989). Chilling by ice and mechanical refrigeration are the most common means of retarding microbial and biochemical spoilage (Gopal, 2005). If the chilled storage is carried out in appropriate packaging materials the shelf life will be prolonged further. The microcellular foams possess characteristics of light weight, thermal insulation and impact resistance in comparison to their solid counterparts (Baldwin et al., 1996; Forest et al., 2015; Tomsko et al., 2009). The low cost and good chemical resistance of polyethylene makes it a good packaging material for perishable food items (Maani et al., 2013; Vasile, 2000). Compared to the polyethylene films, polyester films have better application for packing frozen products due to its high tensile strength, low gas permeability and stability of wide range of temperature (60-220R”C) (Gopal, 2007). The present study compares extension of shelf life by chilled storage of *V. cyprinoides* in Polyester polyethylene (PEST-PE) laminate and Low-density polyethylene (LDPE) packaging materials.

Received 13 October 2017; Revised 27 March 2018; Accepted 28 March 2018

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The study was conducted to determine the storage stability of cooked clam meat in chilled condition for domestic trade.

Materials and Methods

The live clams were collected from Perumbalam island and transported to the laboratory. They were washed in potable water to remove the surface sand and other foreign materials. Further, they were loaded in perforated plastic trays with approximate capacity of 6 kg and immersed in water stored in tanks and well aerated for depuration. The ratio of water to clam was 10:1. The clams were steam shucked at 110°C for 10 min in a retort. The shells were opened up and the shucked meat was collected manually in a hygienic manner and packed in two different packing materials *viz*., LDPE and PEST-PE laminate. The tensile strength and elongation at break for the materials were determined as per IS 2508 (1984) and the overall migration residue (FDA, 1983) which aided in assessing the suitability of packaging materials for food contact application. Each pouch was filled with 50 g of clam meat, properly sealed and stored in an insulated ice box with flake ice in the ratio 1:1. It was stored in chill room at 2+1ÚC. The melt water was drained every day and supplemented with equal quantity of ice. The samples were subjected to biochemical and microbiological analysis at regular intervals of 3 days.

pH of the sample was analysed after making into a paste by dispersing 5 g homogenized clam flesh in 25 ml of distilled water and checking the pH using a digital pH meter as per AOAC (2005). The Thiobarbituric acid value was analysed using a spectrophotometer by measuring the absorbance at 538 nm as per Tarladgis et al. (1960). The TVB-N was determined from the clam meat extracted with TCA by microspace diffusion method (Conway, 1962).

Colour of the clam sample was determined using a Colorimeter for the minced meat. The instrument was first calibrated using standard white and black tiles under "C" illuminant condition in Hunter lab Miniscan XE plus spectro colorimeter (model No. D/ 8-S) with geometry of Diffuse/80 (sphere-8mm view) and an illuminant of D65/10 deg (Shah, 1991). The colour change was based on measurement of Lab values where L*a*b* system (Robertson, 1977) gives colour differences of samples during the chilled storag. The L*, a*, and b* values refer to the three axes of the system: a lightness axis (white–black; L*); and two axes representing both hue and chroma, one red–green (a*) and the other blue–yellow (b*).

About 25 g of the clam minced sample was homogenized with 225 ml sterile normal saline in a stomacher at 230 rpm for one minute for bacteriological analysis. The clam homogenates were serially diluted and plated on Tryptone soya agar for total aerobic mesophilic count. The colony-forming units after incubation was counted to determine the Aerobic plate count (APC) as per FDA (1998).

Results and Discussion

The physical properties of the packaging materials, PEST-PE and LDPE, used in the study are given in Table 1. The thickness of the packaging made of LDPE and PEST-PE are 1 μm and 0.68 μm, respectively. The tensile strength was 133 kgcm^{-2} in

Table 1. Physical properties of packaging materials

Parameters	PEST-PE	LDPE
Total thickness	250 gauge	170 guage
Tensile strength (machine direction)	133 kgcm^{-2}	60 kgcm-^{2}
Tensile strength (cross direction)	133 kgcm^{-2}	37 kgcm^{-2}
Elongation at break (machine direction)	83%	961%
Elongation at break (cross direction)	46%	597%
Overall migration residue		
Water extractives (49ºC for 24 h)	3 mgl^{-1}	8 mgl^{-1}
N-heptane (66ºC / 2 h)	12 mgl^{-1}	17 mgl^{-1}
Acetic acid	11 mgl^{-1}	13 mgl^{-1}

cross and machine direction for PEST-PE and 60 $kgcm^{-2}$ and 37 $kgcm^{-2}$ for LDPE respectively. The elongation at break was 83 and 46% in the machine direction and cross direction for PEST-PE and 961 and 597% for LDPE, respectively. Elongation at break determines the resistance of a packaging material to rupture and breakage when subjected to a tensile force. The levels of water, n-heptane and acetic acid extractives were 3 mgl^{-1}, 12 mgl^{-1} and 11 mgl^{-1} for PEST-PE packaging materials and these values were 8 mgl^{-1}, 17 mgl^{-1} and 13 mgl^{-1} for LDPE packaging materials. When food is packaged in plastic containers some non-polymeric components may leach out from plastic to food causing toxic hazards to the consumer (Gopal, 2005). The average water, heptane and acetic acid extractives of PEST-PE and LDPE packaging materials were below 50 mg l^{-1} (FDA, 1983) which makes it suitable for food contact application.

Changes in the pH values of the clam meat stored in two different packaging materials under chilled condition are illustrated in Fig. 1. The initial pH of the clam sample prior to the packaging was 6.5. The pH of the samples stored in PEST-PE gradually increased until 12 days of storage and then a slight fluctuation in the value was observed till 16^{th} day as pH approached a value of 7.0. In samples packed in LDPE, a steep increase in pH values during initial storage days and a sharp decrease after 12^{th} day was observed. Decrease in the pH could be due to the fermentative spoilage of glycogen present in the clam meat. A similar trend was shown in the studies of oysters in which the decrease in pH was attributed to the fermentative spoilage of the glycogen (Cao et al., 2009). The pH of oyster during the storage under various conditions are reported to vary between 5.6 and 6.3 (Araas et al., 2004). According to the studies of Pottinger (1948) the pH value below 5.2 is considered as sour or putrid in the case of the shucked oysters and the values above 6.2 are considered good. In the present study the level of the pH remained within these range during the entire storage period.

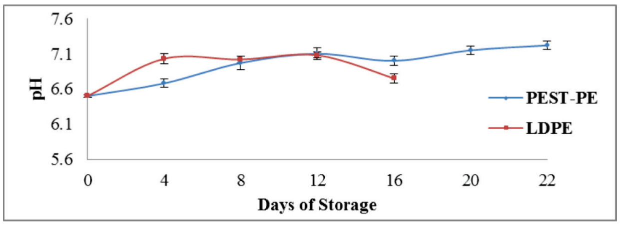

Fig. 1. Changes in pH during chilled storage

TBA is mainly used to evaluate the secondary lipid oxidation of clam meat, which is depicted in the Fig. 2. The TBA values of clam meat showed an increasing trend throughout the storage period and it increased from 0.31 mg malondialdehydekg^{-1} to 1.36 and 1.23 mg malondialdehydekg^{-1} for LDPE and PEST-PE samples, respectively during 16 and 22 days of storage period. Gradual increase in the lipid oxidation is observed during chilled storage and the proper packaging and icing can reduce the rate of lipid oxidation (Baldarati et al., 1982). Presence of oxygen in the air is the prime factor for lipid oxidation. The air packed samples of pearl spot fillets packed in PEST-PE samples got a shelf life of 12 days in the chilled storage (Manju et al., 2007). TBA value in the range of 1.5-2 mg malondialdehydekg^{-1} of sample is generally taken as the limit of acceptability (Lakshmanan, 2000). This study proved that proper packaging in the PEST-PE samples can reduce the rate of spoilage of clam meat compared to samples packed in LDPE. This may be attributed to low Oxygen transmission rate (OTR) and Water vapour transmission rate (WVTR) properties of PEST-PE films (Table 1).

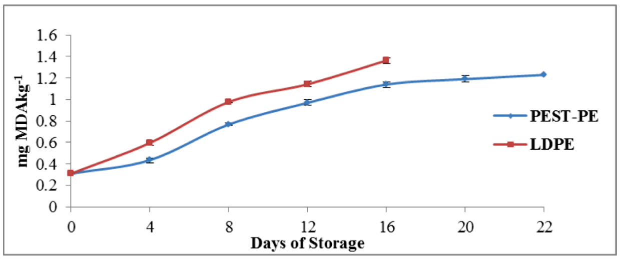

Fig. 2. Changes in TBA during chilled storage

The changes in the Nonprotein nitrogenous compounds namely Total Volatile Basic Nitrogen (TVB-N) during storage are considered as a chemical index for freshness evaluation of fish, crustaceans and molluscs (Lapa-Guimaraes et al., 2005). The TVB-N for the 0^{th} day was found to be 2.1 mg% which was found to slowly increase to 3.4 and 3.2 mg% in the PEST-PE and LDPE packaging after 8 days of storage. Thereafter the value increased to 7.0 and 7.4 mg% after 16 days and 22 days in the LDPE and PEST-PE, respectively as illustrated in Fig. 3. These results were similar to those found for other species of squids and cephalopods, in which the variations in TVB-N contents were small at the beginning of the storage (Vaz Pires et al., 2008). TVB-N has been considered useful indicator of spoilage, having little use as a freshness index (Yamanaka et al., 1987). The relatively low value of TVB-N in clam meat compared to fish is attributed to the high glycogen content which is converted to lactic acid during acidification. Even though the upper limit of spoilage is considered to be above a level of 30 mg N 100 g^{-1} TVB-N when the fishery products are concerned (Marrakchi et al., 1990; Harpaz et al., 2003), shelflife extension studies in mussels showed that TVB-N values were significantly lower than that of finfish and was found to reach only to a maximum of 15 mg 100 g^{-1}

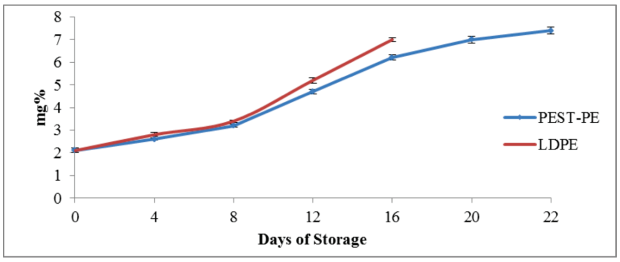

Fig. 3. Changes in TVB-N during chilled storage

The initial Aerobic Plate Count (APC) values for clam meat packed in PEST-PE and LDPE was 4.14 log $cfug^{-1}$ and 4.22 log $cfug^{-1}$, respectively. Thereafter APC increased only slightly to 4.3 log $cfug^{-1}$ on 8th day of storage in both cases. This can be considered as a lag phase of microbial growth. After 8 days of chilled storage the values increased drastically and reached 5 log $cfug^{-1}$ by 16th day in the case of LDPE. The increase was gradual in the PEST-PE packed meat, the count reaching 4.9 log $cfug^{-1}$ by 22nd day. This shows that the bacterial growth in PEST-PE was less compared to LDPE. The initial bacterial count was found to range from 10^2 to 10^5 log cfu g^{-1} in fish and shellfish even though subtle differences occurred (Cao et al., 2009). The initial lag phase of 8 days was observed in studies of raw oysters stored at 0°C. This lag phase was absent in chilled storage of raw oysters at 5 and 10°C (Cao et al., 2009). In processed shellfish requiring cooking (>70°C), the aerobic plate count <5 log $cfug^{-1}$ is referred as the limit of acceptability of clam (ICMSF, 1980).

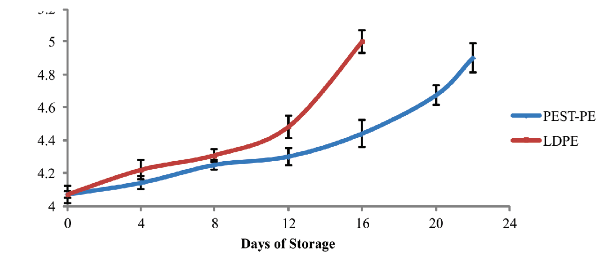

Fig. 4. Changes in APC during chilled storage

Fig. 5 shows the changes of colour (L*or lightness value) of the clam meat throughout the storage period. The colour of clam meat packed in both the packing materials (PEST-PE &LDPE) showed a gradual increase after 8th day of storage. This could be attributed to colour shift from dark to light due to leaching of nutrients. The L*-values increased with storage and developed a lighter and transparent appearance. The a*-value also increased significantly during storage denoting increase in redness. The b* value was found to increase with progress of chilled storage and developed a brighter and less transparent appearance. The colour loss in fish and shellfish products is primarily due to lipid oxidation by degradation of pigments namely carotenoids (Rodriguez-Amaya, 1993).

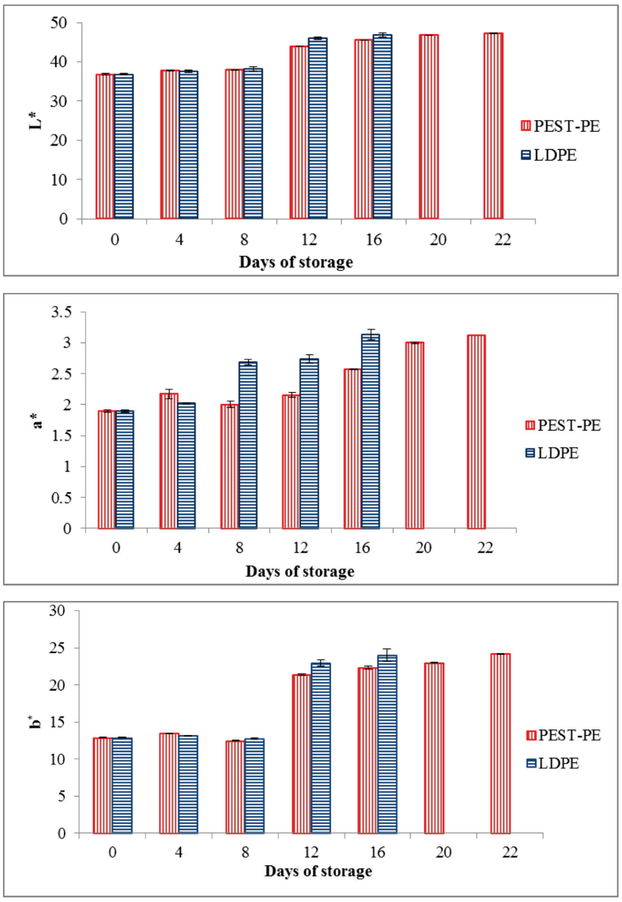

Fig. 5. Changes in Hunter (a) L*-Lightness, (b) a*-Redness and (c) b*-yellowness values in the samples packed in different packaging materials stored in chill room at temperature 2+1R"C. Error bars represent standard deviations of data from triplicate determinations.

This study showed that the shelf life of cooked clam packed in PEST-PE can be extended to 22 days in chilled condition. This information will benefit fishers and vendors to extend the shelf life of cooked clam using suitable packaging materials. The biochemical and microbiological parameters revealed that PEST-PE is more efficient than LDPE for packing of clam meat in chilled environment.

Acknowledgements

The study was carried out as a part of the project entitled "Development of clam cluster and clam processing facility at Perumbalam village, Thycatusherry block, Cherthala Taluk, Alappuzha district, Kerala" funded by DST-SEED, New Delhi and the financial assistance is gratefully acknowledged. The authors are thankful to the Director, ICAR-Central Institute of Fisheries Technology, Cochin for providing necessary facilities for this work and permission to publish the paper.

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