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Quality and Shelf Life Evaluation of Chitosan-coated Dried Bombay Duck (*Harpodon nehereus*)

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ABSTRACT

Background: Bombay duck (*Harpodon nehereus*) is one of the most abundant and preferable marine species in the Maharastra and Gujarat region. Bombay duck is mostly consumed in dried form. The major problem associated with traditionally sun-dried Bombay duck is contamination by insects or pests and uneven drying which leads to spoilage and poor end product quality. Chitosan and its derivatives are used as edible coating food applications due to its antioxidants and antimicrobial action. The present study was aimed to improve the quality of dried Bombay duck by applying chitosan coating and drying under a controlled temperature.

Methods: The five different lots of samples were prepared *viz*. i) control (without any treatment) ii) dip treated in1% acetic acid (AA) iii) dip treated in 0.5% chitosan dissolved in 1% acetic acid (CAA) iv) dip treated in 1% malic acid (MA) v) dip treated in 0.5% chitosan dissolved in 1% malic acid (CMA). All the samples were dried at 50°C. Biochemical, microbiological and sensory analyses were assessed out up to 4 months.

Result: The study revealed that the sample coated with chitosan which was pre-dissolved in either acetic acid or malic acid had lower TVB-N (86.5-115.25 mg%), TPC (5.3-5.5 log cfu^{g-1}) than control (TVB-163 mg%; TPC-7.4 log cfu^{g-1}). A similar trend was also observed for TBA values. Microbial analysis revealed that TPC crossed the limit of acceptability (5.2 log cfu^{g-1}) in 2nd month in control. However, AA, CAA, MA and CMA had acceptable level up to three months. Sensory analysis showed that the overall acceptability score was higher for fish coated with chitosan. Results suggested that chitosan coating and drying could improve the quality of dried Bombay duck.

Key words: Acetic acid, Bombay duck, Chitosan, Drying, Malic acid, Quality.

INTRODUCTION

Fish and fishery products are important sources of protein, fat, essential amino acids, minerals, vitamins and other nutrients. Hence, it has attracted considerable attention as a source rich in important nutritional components to the human diet. Around 20% of the artisanal catch is dried by traditional sun drying methods and consumed in the domestic market (Mukharjee *et al*., 1990). Bombay duck (*Harpodon nehereus*) is one of the most abundant and preferable marine species in the Maharastra and Gujrat region (Chakrabarti, 2010). Traditionally, drying of Bombay duck is done by interlocking their jaws on bamboo scaffolds which are fixed in the sand with bars tied with thick ropes. The major problem associated with traditionally sun-dried Bombay duck is contamination by insects or pests and uneven drying and longer drying time which leads to spoilage and poor end-product quality. Hence, a drying process under controlled temperature conditions has become most important in fish drying to achieve better quality of dried fish products. Although the drying process reduces the moisture content and prevents bacterial spoilage, the addition of antioxidants or antimicrobial agents will enhance the shelf life of dried fish products. There is an increasing interest in antioxidants or antimicrobial agents from natural sources for food application. Chitosan [β-(1,4)-2- amino-2-deoxy-D-glucopyranose] is a versatile biopolymer obtained by deacetylation of chitin, which is found in the shell of shrimp and crustaceans. The application of chitosan in sea foods has been studied by various researchers for shelf life extension and texture modification due to its intrinsic antimicrobial, antioxidant and good film-forming ability (Mohan *et al*., 2012; Jeyakumari *et al*., 2016a; Jeyakumari *et al*., 2016b; Remya *et al*., 2015; Renuka *et al*., 2016; Fabio Marcel *et al*., 2017; Ana Gabriela *et al*., 2017). There is a need for novel fish drying methods to control autolytic enzymatic and microbial spoilage of fish. Organic acids such as acetic acid, malic acid organic acids, such as citric acid and lactic acid suitable are used for various food preservation to inhibit metabolic reactions and microbial growth and are generally recognized as safe (GRAS) (Conor *et al*., 2018;

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Rey *et al*., 2012; Dibner and Buttin, 2002). Very few Studies have established the chitosan film wrapping for dried fish to improve the quality (Vimala devi *et al*., 2015; Agustini *et al*., 2007). Viji *et al*. (2021) studied the edible coating of carboxymethyl chitosan for quality and shelf life extension of dried anchovy. To the best of our knowledge no report on the application of chitosan coating and drying for quality improvement of Bombay duck. Based on the above information, the present work aimed to study the effect of chitosan coating and drying on the quality of dried Bombay duck in comparison with fish treated with only organic acids (acetic acid, malic acid).

MATERIALS AND METHODS

Bombay duck (200-250 g size) were procured from a local fish market at Vashi and brought to the laboratory in iced condition. Fresh Bombay duck was cleaned, split opened and washed with potable water. Then, it was subjected to dip treatment at 5% salt solution for 5 min to get an ideal salt content in the finished product. The five different lots samples were prepared *viz*. i) control (without any treatment) ii) dip treated in1% acetic acid (AA) iii) dip treated in 0.5% chitosan dissolved in 1% acetic acid (CAA) iv) dip treated in 1% malic acid (MA) v) dip treated in 0.5% chitosan dissolved in 1% malic acid (CMA). All samples were given dip treatment for 10 min and dried at 50°C for 14 hrs. All the dried fish were packed in polythene bags and kept in an atmospheric condition. Biochemical, microbiological and sensory analyses were carried out for up to 4 months.

Proximate composition and salt were analyzed as per the AOAC (2015). Total volatile base nitrogen (TVB-N) values were evaluated by the Conway micro diffusion method (Conway, 1950). The peroxide value (PV) was estimated by a titrimetric method of (AOAC, 2015). Thiobarbituric acid (TBA) values were analyzed as per the method followed by Tarladgis *et al*., (1960). Sensory evaluation of dried fish samples was done after frying at a temperature of 160-180°C for two minutes. The panelist was asked to evaluate the appearance, color, flavor, texture, taste and overall acceptability by using nine points hedonic scale. A score between 7.0 and 9.0 indicated, “extremely liked,” scores between 5 and 7 indicated “liked” and a score below 5 was the limit of acceptability (Tahra *et al*., 2018). Total plate count (TPC) and *Staphylococcus aureus* were evaluated according to FAO (1992). *Escherichea coli* were determined as per the BAM method (2002). The data obtained were analyzed by one-way analysis of variance (ANOVA) using SPSS software version 16.0 (SPSS Inc, Chicago, Illinois, USA). All mean separations were tested at a significance level of 5%.

RESULTS AND DISCUSSION

Proximate composition

Proximate composition is one of the major factors that determine the nutritional value of food. Generally, the proximate composition of fish varies between species and also from species to species. This variation is influenced by various factors such as size, sex, age, geographical location and season. In the present study, the moisture content of dried Bombay duck ranged between 11.25-11.58%. Protein and lipid content varied from 69.35-71.02%, 3.8-4.8%, respectively. Results are in accordance with previous reports for sun-dried Bombay duck (Jamil *et al*., 2017; Bhattacharya *et al*., 2016). The protein content observed in this study confirms that fish is primarily a good source of protein. The highest ash content was found in control, AA and CAA. Results are in accordance with previous reports for dried Bombay duck (Jamil *et al*., 2017; Hossain *et al*., 2015). Except for the control (10.25%), all samples had a salt content (6.10-6.5%) within the acceptable limit (7.5% salt) as per Indian standards for fish products (IS 14950). The variation of salt content between samples may be due to different treatment processes followed during sample preparation.

Changes in biochemical quality

Moisture and pH

The moisture content of the dried samples showed an increased trend during storage. Mozzammel Hoque *et al*. (2018) also observed a gradual increase in the moisture content of dried fish during storage. It was observed that the control had a higher moisture content (14.5%) at the end of the fourth month and others followed in the order of MA>AA>CMA>CAA. However, the moisture content in all the samples were within the acceptable limit of 15% (IS 14950) throughout the storage period. The measurement of pH value of fish muscle indicates the quality of the fish. The Initial pH of dried Bombay duck varied between 5.8-6.54. Farzana *et al*. (2014) observed an increasing trend in pH value of dried fish stored at room temperature due to an increase of basic compounds. In the present study, there is no significant difference in pH of dried fish during storage. However, there is a significant difference ($p<0.05$) in pH of dried Bombay duck between treatments (Table 1). It might be due to differences in the composition of treatment used in the study.

Total volatile base nitrogen (TVB-N) and Tri methylamine nitrogen (TMA-N)

TVB-N measures biochemical quality indices of fish and fishery products. The initial TVB-N content of dried Bombay duck had 35.5-60.5 mg%. The variation in TVB-N content may be due to different treatments followed in the study. Several authors observed a higher value of TVB-N (71-115 mg%) for dried Bombay duck collected from local fish markets (Vijayan and Surendran, 2012; Hossain *et al*., 2015; Jamil *et al*., 2017). The lower TVB-N obtained in the present study indicates that fish were dried under controlled conditions. In the present study, Total volatile base nitrogen showed an increased trend during storage. The increase in TVB-N content in dried Bombay duck during storage was also observed by previous researchers (Das *et al*., 2018; Bhattacharya *et al*., 2016). In the present study, CAA had

lower TVB-N values (86.5 mg%) than other treatments and control. It might be due to the fact that chitosan coating helps in reducing the capacity of bacteria for the oxidative deamination of nonprotein nitrogen substances (Fabio Marcel *et al*., 2017; Remya *et al*., 2015). There is a significant difference ($p<0.05$) in TVB-N content, of dried Bombay duck during storage (Table 1). Connell (1995) has suggested a limit of 200 mg% TVBN for salted and dried fish. Accordingly, TVB-N values were within the limit of acceptability in all samples during the storage period.

Peroxide value (PV) and Thiobarbituric acid reactive substances (TBARS)

PV and TBA value is most commonly used to assess lipid oxidation in food and is regarded as useful index to measure the degree of oxidative rancidity. Initial peroxide value ranged between 0.98-1.74 meq O_2 kg^{-1} of fish. In the present study, peroxide value showed an increased trend during storage (Fig 1). Highest PV was observed in the control (10.5 meq O_2 kg^{-1} of fish) and lower PV was found in CAA (6.2 meq O_2 kg^{-1} of fish) followed by CMA (6.8 meq O_2 kg^{-1} of fish) end of storage period. A peroxide value of more than 20 meq O_2 kg^{-1} oil for fish usually gives a rancid smell and taste (Reza, 2006). Accordingly, in the present study, all the samples showed PV within the limit of acceptability. Unlike, PV, TBA values also showed an increased trend during storage (Fig 2). Result showed that the control had TBA values of 1.7 mg malonaldehyde/kg at the end of the storage period. However, CAA and CMA had lower TBA values of 0.82-0.85 mg malonaldehyde kg^{-1}. Vimaldevi *et al*. (2015) observed lower PV and TBA values for dried anchovies wrapped in chitosan-based film. TBA values less than 3.0 mg malonaldehyde kg^{-1} in cured fish are generally considered an acceptable limit. Accordingly, all the samples had a TBA value within the acceptable limit throughout the storage period.

Overall acceptability

Sensory analysis showed that scores for all dried fishes ranged from 6.8 to 9 on a nine-point hedonic scale. No insect infestation or broken pieces in all dried samples were observed throughout the storage period. Results indicated that the control had characteristic color with a slight rancid odor during storage. Moreover, chitosan treated sample (CAA/CMA) had no rancid odor and fetched a higher score

Table 1: Changes in moisture, pH, total volatile base nitrogen (TVB-N) content in bombay duck during storage.

Storage days	Samples	Moisture (%)	pH	TVB-N (mg%)
0	Control	11.58 ± 0.50^{aA}	$6.54\pm.1^{cA}$	60.5 ± 1.5^{cA}
	AA	$11.33\pm.25^{aA}$	$5.8\pm.06^{Aa}$	45.2 ± 3.5^{bA}
	CAA	11.25 ± 0.40^{aA}	$6.1\pm.15^{aB}$	36.5 ± 1.2^{aA}
	MA	11.25 ± 0.45^{aA}	$5.9\pm.18^{abA}$	48.6 ± 3.5^{bA}
	CMA	$10.8\pm.50^{aA}$	$6.1\pm.08^{abA}$	35.5 ± 2.5^{aA}
1st Month	Control	11.9 ± 0.60^{aA}	$6.56\pm.05^{cA}$	75.5 ± 2.8^{dB}
	AA	11.65 ± 0.30^{abA}	$5.85\pm.08^{aA}$	58.6 ± 4^{bcBA}
	CAA	11.58 ± 0.60^{aA}	$6.05\pm.1^{abA}$	48.6 ± 2^{aB}
	MA	11.45 ± 0.55^{abA}	$5.9\pm.15^{aA}$	65.8 ± 4^{cB}
	CMA	11.5 ± 0.55^{abA}	$6.2\pm.15^{bA}$	52.4 ± 1.8^{abB}
2nd Month	Control	12.45 ± 1.2^{aA}	$6.58\pm.1^{cA}$	97.5 ± 4^{cC}
	AA	$12.35\pm.55^{abcA}$	$5.85\pm.09^{aA}$	63.2 ± 5.5^{aB}
	CAA	$11.9\pm.55^{aA}$	6.1 ± 0.09^{abA}	62.6 ± 1.8^{aC}
	MA	$11.9\pm.35^{abA}$	$5.9\pm.07^{aA}$	84.2 ± 5.5^{bC}
	CMA	$11.9\pm.45^{bcA}$	$6.25\pm.06^{bA}$	67.25 ± 3^{aC}
3rd Month	Control	13.85 ± 1.2^{bAC}	$6.62\pm.06^{cA}$	145 ± 3.5^{dD}
	AA	12.65 ± 0.55^{bcAB}	$5.9\pm.15^{aA}$	82.4 ± 4.2^{abC}
	CAA	$12.04\pm.55^{aA}$	$6.1\pm.15^{abA}$	75.5 ± 2.5^{aD}
	MA	$12.8\pm.35^{bcAB}$	$5.85\pm.05^{aA}$	92.55 ± 4.2^{cC}
	CMA	$12.25\pm.45^{bcA}$	$6.3\pm.04^{bA}$	84.25 ± 1.2^{bD}
4th Month	Control	$14.5\pm.4^{bC}$	$6.62\pm.15^{cA}$	163 ± 5.2^{aE}
	AA	$13.1\pm1.2c^{AB}$	$5.95\pm.12^{aA}$	110.65 ± 3.8^{aD}
	CAA	$12.25\pm.8^{aA}$	$6.15\pm.12^{abA}$	86.5 ± 2^{aE}
	MA	$13.65\pm.45^{cBC}$	$5.9\pm.1^{aA}$	125.2 ± 3.8^{aD}
	CMA	$12.8\pm.55^{cAB}$	$6.3\pm.08^{bA}$	115.25 ± 1.8^{aE}

Results are mean±SD (n= 3), values with different small, capital superscript letters are significantly ($P<0.05$) different respect to between treatment, storage days, respectively. Where, Control- Without acid or chitosan treatment; AA- 1% Acetic acid treated; CAA- 0.5% Chitosan dissolved in 1% acetic acid treated; MA-1% malic acid treated; CMA- 0.5% chitosan dissolved in 1% malic acid treated).

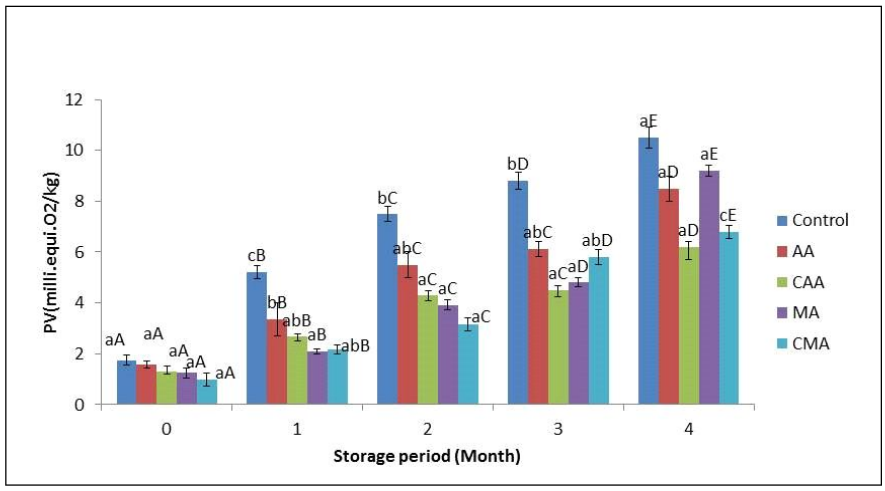

Fig 1: Changes in peroxide value (PV) of dried bombay duck during storage.

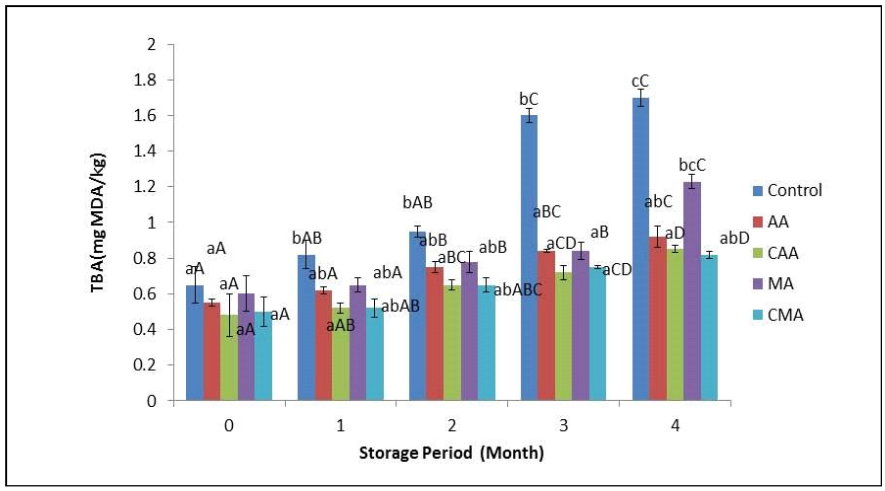

Fig 2: Changes in thiobarbituric acid value (TBA) of dried bombay duck during storage.

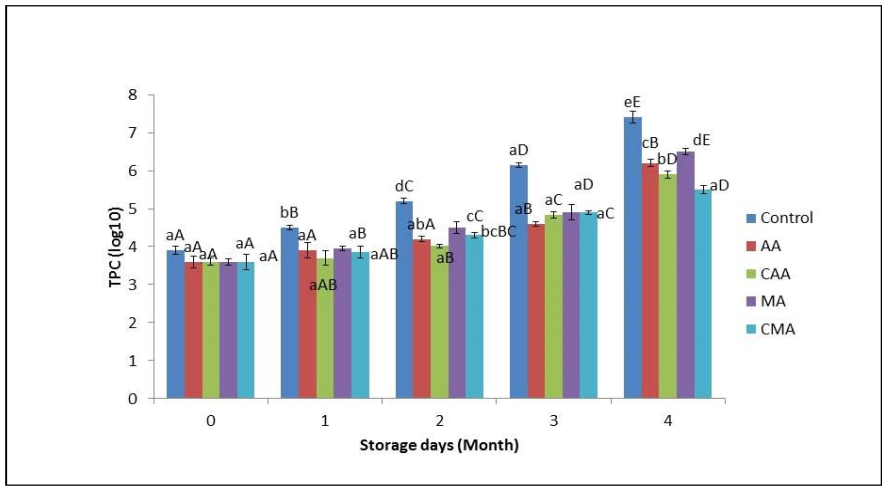

Fig 3: Changes in total plate count (TPC) of dried bombay duck during storage.

for appearance and overall acceptability during storage. Results are in agreement with the previous report for chitosan/ thyme oil-coated dried fishes (Tahra *et al*., 2018; Gore *et al*., 2019; Suliemen and Allaahmed, 2012).

Changes in microbial quality

The initial total plate count in dried Bombay duck ranged between 3.6-3.9 log cfu g^{-1} and it showed an increasing trend during storage (Fig 3). The recommended total plate count for dried fish products for human consumption is 5 $\log_{10}$ (IS 14950). In the present study, TPC crossed the acceptable limit in control during 2nd month of storage. However, AA, CAA, MA and CMA had acceptable level up to three months. Results indicated that the sample coated with chitosan which was pre-dissolved in either acetic acid or malic acid had lower TPC than acetic acid or malic acid treated alone and control. It might be due to the antibacterial activity of chitosan and organic acids (Nicholas, 2003; Vimaldevi *et al*., 2015; Agustini *et al*., 2007; Viji *et al*., 2021). *Staphylococcus aureus*, *E. coli*, was found to be absent throughout the storage period which indicates fish were cleaned and dried in hygienic conditions.

CONCLUSION

It can be concluded that the combined effect of chitosan coating and drying of Bombay duck at controlled conditions revealed that the process method applied in the study could able to produce superior quality of dried fish products with extended shelf life. Further, the chitosan-coated sample showed less TVB-N, PV, TBA and TPC value than acetic acid or malic acid treated alone and control. Sensory analysis showed that the overall acceptability score was higher for fish coated with chitosan. Results suggested that chitosan coating and drying could reduce the oxidation, total bacterial count and extend the shelf life of dried Bombay duck. Results from the study will be more helpful to dry fish processors to produce quality dried Bombay duck with improved shelf life.

Conflict of interest

All authors declare that they have no conflicts of interest.

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