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Changes in proximate, biochemical and microbiological characteristics of dried *Labeo gonius* fillets during storage at room temperature

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An experiment was carried out to assess the changes in proximate, biochemical and microbiological characteristics of dried *Labeo gonius* fillets stored at room temperature. Moisture, crude protein, total lipids, nitrogen free extract (NFE) and ash content of the product were 77.46, 17.94, 2.2, 1.59 and 0.81%, respectively. Total volatile base nitrogen (TVB-N) content was found below the range suggested by various researchers for fish and fish products. Total plate count (TPC) of 1.54 × 10^4 cfu/g was observed and the dominant genera of bacteria were *Pseudomonas* spp., *Micrococus* spp., *Streptococus* sp., *Barillus* spp. and *Vibrio* spp. found in higher percentage. There were no appreciable changes in salted dried *L. gonius* stored in two different packaging materials. Total lipid showed a progressive decrease in the entire salted dried sample during the storage period. An increase in TVB-N was found in dried products stored in gunny bag. The packaging material used had a little effect on peroxide formation. The most common fungi found during storage in the specimen studied were *Aspergillus* sp. and Pencellium.

Key words: *Labeo gonius*, fish, TVB-N, lipid, moisture.

INTRODUCTION

During the past few years, the sudden stride by India in Inland fish production has enabled it to achieve distinction of being the 2nd largest producer in the world. This phenomenal increase in the fish production during the last four decades has been recorded at 28.57% (CIFT, 1998). Although, world fish production is steadily increasing, preservation of the commodity still remains a challenging problem. Susceptibility of fish to rapid spoilage has been attributed to its intrinsic characteristics and to possibilities of microbial contamination from a variety of sources (Venugopal, 1997). Sun drying is one of the most traditional methods of drying since the prehistoric times. Considerable quantitative of dried fish are consumed in developing countries like India. Natural drying by exposure to sun and wind is widely spread out and is possibly the first method used for preserving seafood's. This is still applied to a large extent to preserve fish and squid (Sikorski et al., 1995). Fish curing is the cheapest and simplest way of fish preservation. The term fish curing, in its broader sense, includes all the methods of preservation of fish other than refrigeration and canning like sun drying, salting, pickling, smoking, artificial dehydration etc. Conventional air drying at relatively higher temperature is detrimental to fish muscle

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Abbreviations: KSC, Kursa, split bamboo rack sun dried and corrugated box stored; KSG, Kursa, split bamboo rack sun dried and gunny bag stored; **KOC,** Kursa, oven dried and corrugated box stored; **KOG,** Kursa, oven dried and gunny bag stored.

because, as a result of evaporation, more moisture and salts are diffused to the surface resulting in increase in salt concentration and changes in pH which affect solubility and water binding properties.

The traditional sun drying is subjected to contamination. The aim of this paper was to assess the changes in biochemical and microbiological characteristics of the *Labio gonus* fillets during oven and sun drying.

MATERIALS AND METHODS

Chemicals

Sodium chloride, anhydrous sodium sulphate, potassium iodide, trichloro acetic acid, ethanol and methanol were obtained from Merck (Damstadt, Germany). Plate count agar was obtained from Hi-media (Mumbai, India). Chloroform was procured from Merck (Damstad, Germany).

Fish preparation

Labeo gonius (Kursa) weight 0.75 ± 0.25 kg, caught from Nanak Sagar reservoir using gill net was used in this study. Fish was stored in ice with a fish/ice ratio of 1:2 (w/w) and transported within 1 h to the Department of Fish Processing, G.B. Pant University, India. Upon arrival, the fish was washed using tap water descaled and degutted. The descaled and degutted fish was then subjected to filleting. After washing, the fish was subjected to dry salting at a salt to fish ratio of 1:3 for 24 h. Before bolting, the fresh fish was subjected to biochemical and microbiological analyses.

Drying

The salted kursa was dried by following two methods:

1) Drying in sun shine on elevated rack made from split bamboo having dimensions of 1.0 × 1.5 m with mesh size of 4 cm and raised about 1 m above the floor.
2) The mechanical oven drying with the use of temperature controlled oven (Cole Parmer, Mumbai India).

Storage

The salted dried kursa was packed in high density polythene bags of 40 gauge size and stored at room temperature for a period of six months. The stored fish was subjected to biochemical and microbial analysis at a regular interval of every one month for a period of six months.

Analyses

The stored fish was subjected to the following analyses

Proximate composition

For fresh fish, the proximate composition was determined from the body muscle tissues whereas, in case of salted sun and oven dried, it was carried out from dried flesh. The analysis was carried in triplicate and the average values were calculated and expressed on mean ± SD of triplicate observation.

Moisture

Moisture content was determined following the AOAC (2000). 5 to 10 g of sample was taken in a petriplate and dried in an electric oven at 100 ± 2°C for 16 to 18 h. The samples were kept in a dessicator until weighing. The weight loss in the process was expressed as % moisture content in the sample.

Protein

Total nitrogen of the dry sample was determined following Micro Kjeldhal method (AOAC, 2000).

Nitrogen free extract (NFE)

N.F.E. extract was calculated by using the following expression:

N.F.E. (%) = 100 - (crude protein % + crude fibre + total lipid + total ash %)

Fat free sample was used for the estimation of crude fibre.

Total volatile base nitrogen (TVB-N)

The total volatile base nitrogen was determined by Conway's microdiffusion analysis (Osman et al., 2001). In the procedure, the trichloro acetic acid (TCA) extract prepared sample was treated with potassium carbonate, ammonia liberated and absorbed by boric acid. The quantity of ammonia absorbed was volumetrically determined by titrating the ammonium borate against standard sulphuric acid. The TVB-N content was calculated and expressed as milligram nitrogen per 100 g sample.

Peroxide value

Peroxide value was estimated by the method described by Ozogul et al. (2011). First lipid extract was prepared. The sample was homogenized in a mixture of chloroform and methanol in such proportion that a miscible system is formed with water of the sample. Dilution with chloroform and water separates the homogenate into layers. The lipids dissolve in chloroform layer while non-lipids are contained in methanol layer. This extraction was also used for the estimation of free fatty acids (FFA). For the estimation of peroxide value, 10 ml of lipid extract was used and its oxygen content estimated idometrically. The sample was treated with potassium iodide and iodine which was liberated by the peroxides was titrated against sodium thiosulpahte solution. Peroxide value of the samples was expressed as mill moles of oxygen per kilogram of fat.

Free fatty acids (F.F.A)

Free fatty acid content of the samples was estimated by improved titrimetric method of Ke (1976) as described by Takagi et al. (1984). The 10 ml of lipid extract, 5 ml of methanol and 10 ml of isopropanol were added and mixed thoroughly. It was titrated against standard sodium hydroxide (0.05 N) with addition of metacresal indicator. The end point was recorded with the development of purple colour. FFA content was expressed as a percentage of total lipid as oleic acid and empirically calculated as follows:

$$\text{FFA as oleic acid (\%)} = \frac{\text{Millilitre of NaOH for titration} \times \text{Normality of NaOH}}{\text{Weight of sample}} \times 28.2$$

Table 1. Biochemical and microbiological characteristics of fresh kursa.

Biochemical characteristic	Kursa
Moisture (%)	77.46 (1.645)
Crude protein (%)	17.94 (1.223)
Total lipids (%)	2.42 (0.502)
Nitrogen free extract (NFE) (%)	1.59 (0.523)
Ash (%)	0.805 (0.116)
Total volatile base nitrogen (TVB-N) (mg/100g)	11.37 (1.799)
Peroxide value (PV) (millimoles of O_2/kg fat)	19.54 (2.694)
Free fatty acid (FFA) (% of total lipid as oleic acid)	7.55 (0.856)
Microbiological characteristic*	
Total plate count (total plate count (TPC)) (cfu/g)	1.50×10^4 (5.377)
Total fungal count (TFC) (cfu/g	1.80×10^2 (4.379)

*Mean value for 4 samples. Figures in parenthesis indicate standard deviation.

Sodium chloride content

Salt content of the sample was determined by the method prescribed for salted fishery products (FAO, 2008). 5 g of the muscle sample was treated with distilled water against standard silver nitrate solution using potassium chromate as indicator.

Water activity (aw)

In order to enumerate the influence of water on the flourishment of microbial fishery, the water activity of the sample was determined at room temperature using water activity value analyzer model no. 5803 (Glufft Gmbh and Co, Stuttgart-1). The instrument consists of a thermocole case, holding two sample containers each with a detachable sensor head, which indicates water activity directly on the dials at also the required level of temperature. Calibration of the a_w meter was carried out by keeping a filter paper soaked with the standard solution of barium chloride in the sample container (a_w = 0.9 at 20°C). The holding period was 3 h to get the a_w at room temperature. The instrument was manually calibrated to get the correct water activity.

Microbiological analysis

Total plate count (TPC)

10 g meat was homogenized aseptically with 90 ml of sterile physiological saline (0.85% NaCl) for 3 min. Appropriate dilutions were made from the homogenate in 0.85% saline and planted onto nutrient (plate count) agar plates by spread plate technique. The plates were incubated for 24 h at room temperature. Total plate count (TPC) was enumerated and were expressed as number of colony forming units/g.

Halophillic bacterial count

Halophillic bacterial count was determined using 3.5% sodium chloride solution as diluent. Planting was done onto nutrient (plate count) agar with 10% salt plates by spread plate technique. The colonies developed in the planter were counted and expressed as number of colony forming units/g of sample.

Total fungal count

10 g of fish sample was weighed aseptically and homogenized with 90 ml of physiological saline solution. Appropriate dilutions were made from the 9.0 ml physiological saline and plated onto Potato dextrose agar plate containing antibiotics or tartaric acid solution. The plates were incubated at room temperature for four days and TFC were enumerated and expressed as number of colony forming units/g.

RESULTS AND DISCUSSION

Biochemical characteristics

Fresh kursa used in the study contained 77.46% moisture, 17.94% crude protein, 2.42% total lipid, 1.59% nitrogen free extract (NFE) and 0.81% ash. The total volatile base nitrogen (TVB-N) was 11.37 mg/100 g of sample. The fresh raw material had a peroxide value (PV) value of 19.54 millimoles of oxygen per kg of fat and free fatty acid content of 7.55% of total lipid as oleic acid. Lidabati et al. (1999) have reported proximate composition of kursa *(L. gonius*) as 78%, moisture; 18.6%, crude protein; 1.30%, fat and 0.7%, ash. This study is in close approximation to that of *L. gonius* found during this study. Proximate composition of fish varying with Species sea, body size, season, environmental factors and nutritional status (Sankar and Ramachandra, 2001). The contents of nitrogenous substances as total volatile base nitrogen (TVB-N) in fresh kursa are shown in Table 1. TVB-N content for kursa was found below the levels suggested by different researchers for various fish and fish products. A value of 35 mg/100 g of TVB-N has been suggested as border line (Ghaly, 2010). TVB-N is mainly contributed by ammonia in the muscle produced by deamination of muscle proteins (Chaijan et al., 2006). The chemical assessment of oxidative and hydrolytic rancidity was carried out on fresh fish and results are shown in Table 2.

Connell (1976) has suggested that if PV is above 20 millimoles of oxygen per kg of fat, then the fish may show off odour and taste rancid. The carbohydrate content as NFE in fresh kursa is shown in Table 2 and was less in amount as compared to protein and lipid. Love (1980) described that fish store most of their carbohydrate reserves in liver as glycogen and the levels in muscle as glucose and glycogen.

Microbiological characteristics of raw martial

Bacterial and fungal counts of fresh kursa are presented in Table 2. In the present study, the Total plate count (TPC) of 1.54×10^4 of sample was observed in kursa. In fresh kursa, 1.29×10^5 cells/g Total plate count (TPC) and 0.63×10^2 cells/g TFC has been reported by Litlabati et al. (1999). Some dominant genera of bacteria were identified. Among those *Pseudomonas* spp., *Micrococus* spp., *Streptococus* sp., *Barillus* spp. and *Vibrio* spp. were found in higher percentage. Surendran and Thampurah (2002) has observed that *Aeromonas* spp. and *Pseudomonas* spp. were the main microorganisms associated with fresh water fish *E. coli* and salmonella were not observed during fresh condition of fish. The dominant fungi isolated from the sample of kursa were *Aspergillus* spp. and *Mucor* spp.

Changes in proximate composition

The moisture content of the products, stored in gunny and corrugated boxes showed an increase in moisture during the storage period (Table 2). The moisture uptake during the storage period was significant in the products stored in gunny bags. Moisture absorption in such products is obvious during monsoon due to high relative humidity difference. The low moisture uptake in products despite high relative humidity indicates the advantage of packing salted-dried fish in high density polythene bags and storing it in corrugated box. Santhnaraj et al. (1973) has recorded a moisture content of 9.2 to 11.6% after sun drying anchovies before storing. There were no appreciable changes in salted-dried kursa and stored in two different packages during the storage period. After drying and before storing, the protein content of kursa was around 52.52 and 50.60% in split bamboo rock drying and oven drying, respectively; and it was slightly decreased during storage (Bhat et al., 2010).

The total lipid content showed a progressive decrease in the entire salted dried sample during storage period and it was probably due to oxidative deterioration, thereby affecting lipid extraction (Gantotra et al., 2012). The high degree of unsaturation in the form of multiple double bonds in fatty acids renders fish highly susceptible to oxidative rancidity (Obemeata et al., 2011). The higher value of total ash content in salted dried kursa was attributed to high salt content. Similar levels of ash content in salted dried fish were noticed by several workers (Kiin-Kabari et al., 2011).

Changes in physiochemical characteristics

In the present study, the initial salt content of KS was 17.53% and in case of KO was 19.02%; but at the end of storage period the values decreased to 12.71, 12.98, 14.48 and 1.438% in case of KSC, KSG, KOC and KOG, respectively (Table 3). This increase in salt content can be attributed to uptake of moisture due to hydrostatic moisture of salt during the storage period (Dewi et al., 2011). The a_w (water activity) of products remained almost constant till 2nd month of storage in both salted-dried products. Later, a_w increasing was observed and it was found significant in gunny bag stored up to 0.81 and 0.80 in case of KSG and KOG, respectively. Kalaimani et al. (1988) have recorded a_w of 0.75 in case of salted dried *Cynoglossus macrostomus* and 0.63 to 0.82 in case of unsalted anchovies.

Changes in biochemical characteristics

Total volatile bases (TVB-N)

TVB-N is present in very small quantity in fresh fish and produced mainly due to bacterial action and is mainly constituted by ammonia in the muscle produced by deamination of muscle adenylic acid and by process leading to denaturation of muscle protein. In spoiling fish, volatile bases are produced by putrefactive process and are determined as a measure of content of spoilage which the fish has undergone. Wallace (2000) has pointed out that TVB-N is better index of spoilage. In the present study, salted dried kursa carp showed an increase in TVB-N (Table 8). Though, there is a marked difference in increase between split bamboo rack and oven dried products, the type of package appears to be critical as the gunny bag stored samples showed a higher TVB-N content throughout the storage period.

Joseph et al. (1983) have recorded a TVB-N content of 55.58, 74.94 and 105.50 mg/100 g of sample in case of salted dried sole along Karwar. In the present study, increase in TVB-N throughout the storage period may be due to microbial activity, absorption of moisture and relative decrease in salt content. It is difficult to fix the limit of TVB-N for cured products due to variety and diversity of products and their processing procedure.

Lipid deterioration

Factors that influence lipid oxidation were reported by Burlakova (1988). They found that oxidisability of fish lipids correlated with the content of PUFA, the content of

Table 2. Changes in proximate composition of salted sun and oven dried Kursa stored at room temperature.

Storage period (month)	Moisture (%)				Crude protein (%)			
	KSC	KSG	KOC	KOG	KSC	KSG	KOC	KOG
0	6.42±0.84		4.93±0.24		52.52±1.56		50.60±1.18	
1	7.14±0.72	7.90±0.33	6.39±0.55	8.62±0.84	51.32±1.51	51.40±1.45	50.4±1.11	49.82±1.56
2	9.81±0.58	11.47±0.59	9.34±1.12	11.41±0.97	49.33	50.85±0.82	49.49±1.42	48.72±1.48
3	11.50±0.43	13.93±0.56	11.39±0.72	13.57±0.80	48.08±1.32	50.15±1.02	49.13±0.88	48.24±1.37
4	13.59±0.70	19.34±0.88	12.40±0.77	19.65±0.80	47.35±0.77	48.29±0.89	48.34±1.44	47.41±1.20
5	15.49±0.55	18.43±1.14	14.39±0.39	19.10±1.67	47.11±0.84	47.45±1.31	47.26±0.98	46.74±1.44
6	15.07±0.86	-	14.12±0.29	-	46.33±1.47	-	46.48±1.43	-
Storage period (month)	**Total lipids (%)**				**NFE (%)**			
	KSC	KSG	KOC	KOG	KSC	KSG	KOC	KOG
0	9.48±0.64		10.33±0.87		3.30±0.10		3.27±0.08	
1	8.89±0.85	8.65±0.53	9.53±0.41	9.57±0.90	3.14±0.17	3.12±3.05	3.23±0.12	3.27±0.15
2	8.16±0.92	7.88±1.27	8.50±1.16	8.87±1.14	3.06±0.09	2.99±0.04	3.13±0.10	3.10±0.04
3	7.35±0.45	7.26±0.60	7.65±0.87	7.96±0.83	2.79±0.08	2.63±0.09	2.81±0.08	2.75±0.05
4	6.96±0.97	6.99±0.85	7.18±0.78	7.44±0.75	2.66±0.11	2.58±0.10	2.57±0.07	2.49±0.06
5	6.59±0.80	6.25±0.79	6.70±1.31	6.53±0.44	2.09±0.08	2.04±0.08	2.19±0.06	2.14±0.06
6	5.78±0.68	-	6.24±0.94	-	1.98±0.07	-	2.12±0.08	-

KSC: Kursa, Split bamboo rack sun dried; corrugated box stored. KSG: Kursa, Split bamboo rack sun dried; gunny bag stored. KOC: Kursa, oven dried; corrugated box stored. KOG: Kursa, oven dried; gunny bag stored.

Table 3. Changes in physiochemical characteristics sun and oven dried kursa stored at room temperature.

Storage period (month)	Salt content (%)				Water activity				Rehydration capacity (%)			
	KSC	KSG	KOC	KOG	KSC	KSG	KOC	KOG	KSC	KSG	KOC	KOG
0	17.53±0.70		19.02±1.20		0.67±0.09		0.68±0.14		21.84±1.19		19.42±2.09	
1	17.50±0.88	16.80±1.27	18.57±0.42	18.61±0.22	0.96±0.08	0.69±0.09	0.69±0.09	0.70±0.09	22.12±2.46	21.17±1.19	20.26±0.99	19.75±2.47
2	16.96±0.74	15.58±1.53	17.40±1.14	16.37±1.27	0.70±0.04	0.71±0.06	0.70±0.08	0.72±0.07	22.48±2.34	22.38±1.99	20.69±0.99	20.38±1.33
3	15.67±1.02	14.74±0.65	16.36±1.74	15.62±1.34	0.72±0.08	0.73±0.05	0.74±0.09	0.74±0.09	23.14±1.50	22.77±1.99	21.43±1.70	20.42±1.42
4	14.74±1.14	13.66±1.33	15.71±1.83	14.77±1.23	0.76±0.04	0.75±0.05	0.77±0.04	0.80±0.06	23.97±2.63	23.39±1.90	21.56±1.85	21.36±1.59
5	13.54±1.27	12.98±1.47	15.05±2.23	14.32±2.69	0.80±0.10	0.81±0.08	0.80±0.09	0.87±0.08	24.59±1.30	23.77±1.91	22.85±1.06	22.16±1.65
6	12.74±1.08	-	14.48±1.22	-	0.81±0.02	-	0.82±0.06	-	25.88±0.97	-	23.58±1.77	-

KSC: Kursa, Split bamboo rack sun dried; corrugated box stored. KSG: Kursa, Split bamboo rack sun dried; gunny bag stored. KOC: Kursa, oven dried; corrugated box stored. KOG: Kursa, oven dried; gunny bag stored.

phospholipids and the content in the latter of phosphatidyl ethanolamine and cordiolipid (%). Bernardez (2005) stated that double bonds of unsaturated fatty acids are highly susceptible to oxidation and this leads to the production of carbonyls and other secondary oxidation products which impart the characteristic rancid off flavor to the product. Oxidation rancidity is most often measured by 2-thiobarbituric acid (TBA), peroxide value (PV) or carbonyl value (Gray, 1978). Lipid hydrolysis by itself has no nutritional significance but the accumulation of free fatty acids (FFA) in fish oils in undesirable amount due to secondary reaction catalyzed, such as increased susceptibility to oxidation and consequent development of off flavours (Nair et al., 2000). There has also been a long debate over claims that lipid hydrolysis and associated accumulation of FFA contributes to accelerated protein denaturation in fish (Sikorski, 1976).

In the present study changes in PV and FFA of the lipid fraction of salted-dried fish has shown an appreciable increase in all the products initially up to 3 months in salted-dried kursa and thereafter showed a decrease. The package material used seems to have a little effect in peroxide formation. The results show that corrugated boxes did not possess any advantage over gunny bags with regard to gas permeability. Prior to salting, the PV values were 19.54 mill moles of O_2 per kg of fish fat in kursa and due to changes during salting and drying, the value increased to a maximum of 73.53 millimoles of O_2 per kg of fat in kursa. The result indicates that the salting and drying conditions accelerate lipid oxidation and this is in agreement with the results of previous workers (Waterman, 1976; Smith, 1988; Smith et al., 1988).

Microbiological changes

Total plate counts

In the present study, the total plate count (TPC) (Table 4) showed an overall increasing trend in salted dried kursa and increase was greater in gunny bag stored products. Joseph et al. (1983) have recorded a Total plate count (TPC) of 3.63×10^3, 3.23×10^3 and 10.93×10^3 cells/g in case of salted dried sole along Karwar, Mangalore and Calicut coast, respectively. These counts are nearer to the counts observed in the present study. The significant counts were recorded during storage probably due to increased water activity. The inter correlation matrix of TPC, TVB-N a_w and overall acceptability scores are shown in Table 5. It showed a positive correlation of total plate count (TPC) with a_w and TVB-N but was significantly negatively correlated with overall acceptability.

Halophillic count

Halophiles actually require salt for growth and will not grow unless salt is present and grow mostly in salted dried fish products (Khan et al., 2005). They have stated that the halophiles are aerobic and usually not found in pickled fish where only limited oxygen assess is possible through the brine. In the present study, it is evident from Table 4 that the halophillic bacteria could not be detected initially, but gradual increase was observed from 5^{th} month of storage due to higher moisture and salinity in the product. The counts of more than 1×10^2 cells/g could be due to their higher water activity. AOAC (2005) observed that a_w for a saturated salt solution is 0.75 and if the equilibrium relative humidity is greater than 75% of the salted products it will take up moisture from the atmosphere increasing the O_2 and consequently introducing the possibility of storage by group of microbes.

Total fungal count (TFC)

Not all fungi which recur in fish are considered deleterious. Moulds are one of the important causes of spoilage of salted dried fish products and they produce mycotoxins and they are able to grow in salt concentrations between 5 and 26% (Reilly, 1986). In the present study, Table 6 shows the fungal count of salted dried kursa. It is clear from the results that an increasing trend was shown during the storage period. TFC was more in gunny bags stored products as compared to corrugated boxes. A significant fungal growth was recorded after 4^{th} month of storage and this may be due to increase in a_w, moisture content and salt content. Products deteriorate by growth of moulds if the water content is approximately 15% (Gandotra et al., 2012). These observations were in close agreement to the present study. The rapid reduction in the water activity ($a_w < 0.75$) is the most important factor in controlling fungi/mould contamination of the fishery products during storage (Kolakowska, 2002).

Dominant bacterial and fungal flora during storage period

Table 7 shows the dominant bacterial and fungal flora. *E. coli* and salmonella bacteria were not detected during the storage period. Drying of fish is reported to impart a degree of microbiological stability to the product which is a function of reduced water activity and heating. Surendran and Thampurah (2002) had observed that *Aeromonas* and *Pseudomonas* are the main microorganisms associated with fresh water fish. The most common fungi found during storage in the specimen studied were *Aspergillus* sp. and Pencellium. Turkkan et al. (2008) have reported the production of aflatoxin when dried and smoked tilapia was inoculated with *Aspergillus flavus*. Although, there are no reported increases of consumes being poisoned by mycotoxins in fishery products; there is a definite risk to human health considering how fish are traditionally processed.

Delays in drying fish during the rainy season, the use of heavily contaminated brines and low standards of hygiene

Table 4. Changes in total plate count (TFC) and halophilic count (HC) of salted sun and oven dried Kursa.

Storage period	Total plate count (TPC) (cfu/g)				HC (cfu/g)			
	KSC	KSG	KOC	KOG	KSC	KSG	KOC	KOG
0	2.94×10^2		2.53×10^2		Estd 1×10^1		Estd 1×10^1	
1	4.31×10^2	6.49×10^2	3.91×10^2	5.8×10^2	1×10^1	1×10^1	1×10^1	1×10^1
2	6.8×10^2	7.11×10^2	4.50×10^2	6.21×10^2	$1\times10^1\times$	1×10^1	1×10^1	1×10^1
3	7.90×10^2	9.10×10^2	6.11×10^2	8.04×10^2	1×10^1	1×10^1	1×10^1	1×10^1
4	9.60×10^2	1.50×10^3	6.30×10^2	1.05×10^3	1×10^1	1×10^1	1×10^1	1×10^1
5	1.30×10^3	3.50×10^3	9.82×10^2	2.20×10^3	1.04×10^2	1.21×10^2	6.42×10^2	1.10×10^2
6	2.10×10^3	-	1.69×10^3	-	1.52×10^2	-	1.31×10^2	-

KSC: Kursa, Split bamboo rack sun dried; corrugated box stored. KSG: Kursa, Split bamboo rack sun dried; gunny bag stored. KOC: Kursa, oven dried; corrugated box stored. KOG: Kursa, oven dried; gunny bag stored.

Table 5. Correlation coefficient (r) and regression equation (Y = a + b X) of salted sun and oven dried kursa.

Relation	Correlation coefficient (r)				Regression equation (Y=a + b X)			
	KSC	KSG	KOC	KOG	KSC	KSG	KOC	KOG
Moisture and salt	- 0.961**	- 0.982**	- 0.989**	-0.959 **	Y= 21.26-0.508 X	Y= 19.42 – 0.326 X	Y= 21.52 – 0.467 X	Y=20.63 – 0.325 X
a_w and Total plate count (TPC)	0.927**	0.958**	0.768*	0.967**	Y=-6655.88 + 10.320.71 X	Y=-15091.71 + 22503.91 X	Y=-3098.24 + 5056.38 X	Y= -6040.87 + 9257.98 X
a_w and HC	0.848*	0.822*	0.706 ns	0.815*	Y= 627.42 + 912.17 X	Y= -517.70 + 751.70 X	Y= -301.19 + 445.81 X	Y= -324.29 + 466.85 X
a_w and TFC	0.966**	0.987**	0.918**	0.979**	Y= -5979.65 + 9109.91 X	Y= -8646.65 + 13029.39 X	Y= -3759.77 + 5811.06 X	Y= -4494.34 + 6812.43 X
TVB-N and Total plate count (TPC)	0.966**	0.973**	0.912**	0.947**	Y= -221.09 + 27.77 X	Y= -1081.46 + 59.11 X	Y= -260.96 + 20.99 X	Y= -515.59 + 31.93 X
TVB-N and HC	0.917**	0.844*	0.821*	0.770 ns	Y= -6249 + 2.55 X	Y= -50.54 + 1.99 X	Y= -49.26 +1.81 X	Y= -42.99 + 1.55 X
TVB-N and TFC	0.961**	0.980**	0.997**	0.995**	Y= -253.51 + 23.41 X	Y= -504.50 + 33.46 X	Y= -402.88 + 22.06 X	Y= -467.69 + 24.37 X
TVB-N and overall acceptability	- 0.966**	- 0.966**	- 0.842**	- 0.938**	Y= 8.80 – 0.55 X	Y= 8.97 – 0.69 X	Y= 7.92 – 0.17 X	Y= 8.66 – 0.43 X
PV and FFA	- 0.135 ns	- 0.387 ns	0.426 ns	0.941**	Y= 25.08 – 0.60 X	Y=25.76 – 0.97 X	Y= 15.74 + 0.43 X	Y= 13.72 + 071 X
PV and overall acceptability	- 0.169 ns	0.329 ns	- 0.195ns	- 0.742ns	Y= 6.91 – 0.24 X	Y= 5.81 + 0.21 X	Y= 7.32 – 0.11 X	Y= 6.89 – 0.97 X
FFA and overall acceptability	0.595 ns	0.573 ns	0.364 ns	0.102 ns	Y= 2.02 + 0.19 X	Y= 5.88 + 0.14 X	Y= 6.63 + 0.20 X	Y= 6.33 + 0.18 X
Total plate count (TPC) and overall acceptability	- 0.933**	- 0.933**	- 0.982**	- 0.987**	Y= 8.24 – 0.19 X	Y= 7.62 – 0.11 X	Y= 7.74 – 0.88 X	Y= 7.97 – 0.13 X
TFC and overall acceptability	- 0.872*	- 0.948**	- 0.854*	- 0.905*	Y= 7.98 – 0.21 X	Y= 7.87 – 0.20	Y= 7.61 – 0.79 X	Y= 7.80 – 0.17 X

*Significant at 5%; **significant at 1%; ns, non significant.

KSC: Kursa, Split bamboo rack sun dried; corrugated box stored. KSG: Kursa, Split bamboo rack sun dried; gunny bag stored. KOC: Kursa, oven dried; corrugated box stored. KOG: Kursa, oven dried; gunny bag stored.

Table 6. Changes in total fungal count (TFC) of salted sun and oven dried kursa.

Storage period (month)	TFC (cfu/g) of kursa			
	KSC	KSG	KOC	KOG
0	0.95×10^2		0.81×10^2	
1	1.80×10^2 (3.56)	2.20×10^2 (3.86)	1.41×10^2 (2.99)	1.75×10^2 (2.50)
2	4.11×10^2 (4.57)	5.89×10^2 (4.86)	3.61×10^2 (4.20)	4.24×10^2 (5.06)
3	7.76×10^2 (5.06)	9.25×10^2 (3.11)	5.80×10^2 (3.86)	7.15×10^2 (4.03)
4	9.84×10^2 (4.27)	1.29×10^3 (5.56)	7.96×10^2 (4.65)	1.01×10^3 (2.50)
5	1.11×10^3 (2.99)	1.81×10^3 (4.19)	1.08×10^3 (3.92)	1.35×10^3 (2.99)
6	1.50×10^3 (5.56)	-	1.32×10^3 (5.25)	-

KSC: Kursa, Split bamboo rack sun dried; corrugated box stored. KSG: Kursa, Split bamboo rack sun dried; gunny bag stored. KOC: Kursa, oven dried; corrugated box stored. KOG: Kursa, oven dried; gunny bag stored.

Table 7. Dominated bacteria and fungi in salted sun and oven dried kursa stored at room temperature.

Dominated bacteria	Dominated fungi
Aeromonas spp.	*Aspergillus niger*
Flavor bacterium	*A. flevipes*
Micrococcus	*Pencillium* spp.
Pseudomonas spp.	*Rizopus niger*
Vibrio spp.	*Mucor*
Streptococcus spp.	
Staphyloccus spp.	

Table 8. Changes in TVB-N (mg/100 g) of salted sun and oven dried kursa stored at room temperature.

Storage	Kursa			
	KSC	KSG	KOC	KOG
0	19.63 ±1.63		23.14±0.73	
1	22.31±1.49	25.68±1.02	25.77±1.87	28.35±1.63
2	28.57±1.66	32.59±1.80	31.80±1.42	36.46±1.72
3	34.73±1.38	37.48±1.83	43.53±2.09	45.19±1.30
4	45.67±1.86	49.68±1.04	55.70±0.91	59.59±0.92
5	64.67±1.49	72.73±1.94	68.42±1.33	76.64±1.22
6	75.38±3.21	-	77.31±2.58	-

KSC: Kursa, Split bamboo rack sun dried; corrugated box stored. KSG: Kursa, Split bamboo rack sun dried; gunny bag stored. KOC: Kursa, oven dried; corrugated box stored. KOG: Kursa, oven dried; gunny bag stored.

all contribute to contamination by toxinogenic moulds.

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