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Quality Changes of Salted Kass (*Hydrocynus forskalii*) During Storage at Ambient Temperature (37±1°C)

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Abstract: In the present study, the chemical and microbiological quality changes in salted (25% of the fish weight) *Hydrocynus forskalii* was carried out during storage at +37±1°C. Moisture, ash, protein, lipid, fiber and pH were analyzed to determine chemical quality and total viable counts of bacteria (TVC), total Staphylococcus-Micrococcus and yeast-mould were measured to determine microbial quality during the storage period. Reduction of chemical quality was found statistically significant ($p<0.05$). No yeast and mould were detected for the period of storage. Microbial analysis demonstrated that the salted techniques reduced the microbial counts of the salted fish, whereas it retarded the microbial growth during the last two months. Based on the data, the optimal shelf life was found to be three months for salted *Hydrocynus forskalii*.

Key words: *Hydrocynus forskalii*, salted, quality, storage, ambient temperature

INTRODUCTION

Salting process is considered as one of the oldest methods of fish preservation and this process is still been used in several places around the world. The effect of salt is to obstruct or destroy the growth of the microorganism, where in this end the fish meat gets it's way to durability. The preservation period of product is linked to the amount of salt added, there fore a straight proportion is present between the amount of salt used and the preservation period (Bahri *et al*., 2006). Salted fish products are popular in many countries around the globe. As these have been proven to be safe for millenniums, even in developed countries (Turan *et al*., 2007).

Through generations waters of Sudan (100.000 km^2 fresh water and 750 km length of coastal marine waters on the Red Sea) have been fished for centuries. Estimated that 26000-29000 tons of fish have been taken from them annually (Yousif, 1988). This represents about 29% of the estimated annual potential i.e. 104,000 tons (Henderson, 1975). More recent estimates of production were in the range of 110,000 ton/year (Federal Fisheries Administration Department, Annual Reports, 1996). In the Sudan, nearly 70% of the total fish landings are consumed in forms of fresh fish; the rest is cured either by salting, fermentation or sun-drying. Only few of the local fish supply is smoked, except in the southern Sudan where smoked and very dry fermented fish products are very popular among the local community (FAO, 1992). Salted fish is always made from *Hydrocynus* spp "Kass" which belong to the family Characidae (Idris, 1981).

The major reason given by the processors for choosing this species for salting is that this type of fish is relatively lean, Sudanese consumer prefers little fat in the salted products (Dirar, 1993). Other reasons given for the choosing of this fish for salting, include a belief that it is tastier than that from other fish types, moreover after treatment, the still preserve the original shape, colour and flesh, whereas other fish types become soft, lose firmness and fastly liquefy. The objective of this study was to investigate the effect of storage time on the nutritive value of the preserved *Hydrocynus forskalii* using dry traditional salting method.

MATERIALS AND METHODS

Collection of samples: Samples of fresh fish were brought from local fishermen market, namely Kass (*Hydrocynus forskalii*). These samples were kept in polyethylene bags with crushed ice and transported to the Fisheries Research Center, where samples for chemical and microbiological analysis were immediately carried out.

Processing: Fresh fishes were always washed, eviscerated, washed again and transferred to baskets to dry up while covered by a thin cloth to prevent insects invade. Then fish were weighed to the nearest gram using a dial balance (KRUPS type 875), for the purpose of salting, a total weight of salt estimated 25% of the fish weight was used. The procedure used is called dry salting. In this method salt was applied by hand and brushing of the fish surface, the inner lining of eviscerated abdominal cavity and the gills chambers.

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This process was conducted by separating the fish layers by coarse salt mattresses inside a plastic container. The stack of the fish and salt are left about 7 days to let the salt penetrate the muscles. When the salt has penetrated the fish, it extracts the fish fluids through plasmolysis. The extracts fluid (pickle) was allowed to drain continuously. Used salt is removed from the fish surfaces and the fish restacked with new dry salt between the layers once during the ripening process. Salted product, was packed in polythene bags and stored at ambient temperature (37±1°C) for six months.

Chemical analysis: Moisture, protein, lipid, fiber and ash contents were determined according to AOAC (1996). Hydrogen ion concentration (pH) was determined by using of one gram of fish sample added to ten ml of distilled water and put into Heraeus CHRIST for digestion of the sample and then placed into a buffer tube of pH meter (JENNAY 3015) for reading.

Microbiological examination: Decimal dilutions (up to 10^{-6}) of fish samples were prepared using sterile % 0.1 peptone water solution. The appropriate dilutions were pour-plated on appropriate media for enumeration of bacteria or yeast-mold (Harrigan, 1998). The microbiological media and incubation conditions used for enumeration of microorganisms were Plate Count Agar (PCA) for Total Viable Counts (TVC) (at 37±1°C, 48 h). Mannitol Salt Agar (MSA) was used for counting *Staphylococcus-Micrococcus* spp. (37±1°C 36-48 h), the numbers of *Staphylococcus aureus* were determined by applying coagulase test on bright yellow halo colonies on Mannitol Salt Agar. Potato dextrose agar (mark 0130) was used for counting mold and yeast (22±1°C 5 days).

Sensory evaluation: For examination purposes, end products were submitted on 20 persons test panel. From Fisheries Research Center staff, fishermen and some students of Department of Fisheries, College of Natural Resources, University of Juba and judged in comparison with salted fish. Comparison was carried out in terms of organoleptic characteristics, such as colour, flavour, taste, texture and general appearance. The panel was requested to rate each organoleptic feature of the end products according to a 10 point scale (9 = excellent; 8-9 very good; 6.5-7.9 good; 5-6.4 fair; <5 bad), using the score method as reported by (Afolbi *et al*., 1984).

Statistical analysis: The mean and standard deviation (mean ± SD) for the results obtained were calculated using SPSS software (Version 10).

RESULTS AND DISCUSSION

The results of chemical composition of fresh fish used in salted fish preparation, namely *Hydrocynus forskalii* (Kass) was given in Table 1. The moisture contents have recorded high value 70.21±0.101%, similar results were obtained by various researchers, namely Abdullahi (2000) who worked on fresh water fish species: *Alestes nurse, A macrolepidotus*, *Hydrocynus brevis* and *Hepsetus odoe* and Clucas (1981) on *Hydrocynus vittatus*. The protein content was 20.20±0.368% on wet basis, this is probably due to the high moisture content. These results agreed with those obtained by other investigators for common Nile fishes, (Mahmmoud, 1977; Iskander, 1982; Ssali, 1988). Lipid content was 1.84±0.113% on wet basis .It was obvious that *Hydrocynus forskalii* belongs to the category of low fat fish classified by Ackman (1989) having fat content below 5%. Ash and fiber content were 1.9±0.368% and 1.93±0.110% respectively. These results are in accordance with those obtained by (Mahmmoud, 1977; Ssali, 1988).

Table 1: Chemical composition of fresh *Hydrocynus forskalii* (g/100 g)

Parameters (%)	Mean±SD
Moisture	70.21±0.101
Ash	1.90±0.368
Protein	20.20±0.368
Lipid	1.84±0.113
Fiber	1.93±0.110
pH	6.6±0.370

The moisture content changes were determined to be significant difference ($p<0.05$) and decreased progressively during the six months storage period (Table 2). The moisture contents of salted samples were determined least 19.31±0.101%, highest 63.41±0.103%. The reduction in moisture contents during storage can be attributed to protein denaturation and consequent loss of water-holding capacity of the protein in the used fish samples. Dry salting produced considerable loss of constituent water due to heavy uptake of salt (Martínez-Alvarez and Gomez-Guillén, 2006). The findings of present study are similar to the findings of Kucukoner and Akyuz (1992), on dry salted horse mackerel and Bahri *et al*. (2006) on salted Grey Mullet. Changes in protein content of salted *Hydrocynus forskalii* samples were observed to be significant ($p<0.05$). The protein content of samples was determined least 11.68±1.06 %, highest 18.00±0.100% (Table 2). It is evident that the protein content of processed fish has decreased after the course of salting. Loss of protein during processing is extremely variable. In our results, the losses of protein during storage period were averaged to 6.37%. Salting of fish was usually accompanied by protein losses, as water is drawn out a meal brine is formed, some protein is

Table 2: Changes in chemical composition of salted *Hydrocynus forskalii* (g/100 g) during storage (37±1°C)

Parameters (%)	0 (week)	Month 1	Month 2	Month 3	Month 4	Month 5	Month 6
Moisture	63.41±0.103[a]	56.82±0.108[b]	38.42±0.103[c]	25.71±0.10[d]	22.900±0.10[e]	20.00±00.100[f]	19.31±0.100[g]
Ash	11.20±0.10[a]	12.22±0.106[b]	13.00±0.36800[c]	13.21±0.100[d]	13.51±0.103[e]	13.83±0.113[f]	13.98±0.113[g]
Protein	18.00±0.10[a]	16.21±0.101[b]	15.72±0.108[c]	14.51±0.101[d]	13.23±0.103[e]	12.510±1.06[f]	11.68±1.06[g]
Lipid	1.41±0.101[a]	1.200±0.368[b]	1.00±0.100000[c]	0.830±0.113[d]	0.712±0.104[e]	0.512±0.103[f]	0.410±0.101[g]
Fiber	1.31±0.103[a]	1.200±0.101[b]	0.927±0.110[c]	0.832±0.113[d]	0.523±0.108[e]	0.340±0.716[f]	0.367±0.104[g]
pH	6.600±0.10[a]	6.5000±0.10[a]	6.200±0.100[a]	6.100±0.155[a]	5.900±0.100[a]	5.600±0.104[a]	5.5±0.106[g]

Values are shown as mean ± standard deviation of triplicate measurements. Different superscript letters in the same row indicate significant differences between groups ($p<0.05$)

dissolved into the brine (Clucas, 1981). Generally the quantity of protein lost depends on the exact nature and duration of the salting process and the conditions of fish when salted (Eltom, 1989). The lipid contents changes were determined to be of significant difference ($p<0.05$), least 0.41±0.101% and highest 1.41±0.101%. It is clear from the present results that lipid content was decreased, this might due to the leaching out of some substances during processing as there were correlation between transfer rate of lipid from muscle and salt concentration in muscle. The transfer rate increased with high salt concentration. Changes of ash content of salted *Hydrocynus forskalii* samples were found significant ($p<0.05$) least 11.20±0.100%, highest 13.98±0.113%. It is known that the oozing of fish juice during salting usually is accompanied by losses of minerals thus the relatively high ash content observed in salted samples can be attributed to the salt penetration into fish flesh during the curing process. The findings of present study are lower than findings of (Salma *et al*., 1977) who reported that the ash content in salted sardine ranged between 14-18%. This difference may be attributed to the different in fish species, storage time and technological procedures. Changes of fiber content of salted *Hydrocynus forskalii* samples were found significant ($p<0.05$) least 0.367±0.104% and highest 1.313±0.103%. There were no significant differences ($p>0.05$) in mean pH levels during storage time for salted *Hydrocynus forskalii.* The pH values of samples were determined least 5.5±0.11, highest 6.6±0.10, (Table 2). Similar results were reported in salted fish by other researchers (Gokoglu *et al*., 1994; Kucukoner and Akyuz, 1992; Bahri *et al*., 2006). It has been noted that pH values in samples of trout, anchovy and mirror carp fish were found between 6.41-6.70 at the beginning and then changed depending on the storage period and varied between 5.34-6.81 (Bahri *et al*., 2006).

The total viable count of bacteria in fresh fish used as raw material (*Hydrocynus forskalii*), ranged between 10^2 and 10^4 cfu/g. The fish is more susceptible to microorganisms after catching. The number of bacterial counts may be possibly explained by contamination of fish during catching, handling, transportation and exposure to the surrounding environment. Shewan (1962), Liston (1980) and Gram (1989) noted that the bacterial flora on newly caught fish depends on the environment, in which it was caught rather than on the fish species. The total viable count of salted samples were varied during storage time. The total of viable counts of bacteria were found least <100 cfu/g, highest $6.5x10^3$ cfu/g (Table 3). There was a general trend of marked increase in case of total viable counts of bacteria in zero days (1st week), then after that the counts begin to decrease as salting proceeded. This could be explained on the basis that in a short processing period as that in the present case, it is hard to believe that the substrate for microbial growth comes from the degradation products of the protein. It is more likely that the microbial growth occurs as a result of attacking proteinaceous and other soluble nitrogenous compounds that exists in the fish juice. Also the early increase occurred while fish was wet and the provision of salt promoted the growth of halotolerant and halophilic bacteria in the fish. As the fish became drier, there was a decrease in water activity and this together with the accumulated salt in the flesh, resulted in suppression of bacterial growth. In this work 10^4 cfu/g of the total of viable counts bacteria was used as the limit for the evaluation of microbial spoilage. When aerobic plate counts reach 10^6 cfu/g, the food product was assumed to be at or near spoilage (Pascual and Calderón 2000; Arashisar *et al*., 2004; Ozogul *et al*., 2004). In this study, the microbial growth was lower in salted samples and hasn't reach 10^6 cfu/g. However, by the end of storage period, growth was not detected. Count of *Staphylococcus-Micrococcus* were determined least <100 cfu/g, highest 3.4 x 10^3 cfu/g. Similar result was obtained by (Bahri *et al*., 2006) from salted Grey Mullet. In present study *Staphylococcus aureus* was determined in samples. Small number of *Staphylococcus aureus* in water products do not cause any health problems. However, this microorganism can reach high levels (>5 $\log_{10}$ cfu/g) in products prepared by hand under bad conditions and can cause food poisoning (Varnam and Evans, 1991). The values of *Staphylococcus aureus* was still within the limit of 10^3 cfu/g recommended by ICMSF (1978) in good manufacturing practices. No yeast or mould was detected in our fresh and salted samples.

The organoleptic properties of the salted *Hydrocynus forskalii* that the products were acceptable according to the panel's evaluation, though statistically there was

Table 3: Changes in microbiological quality of salted *Hydrocynus forskalii* during storage (+37±1°C)

Microorganisms	0 (week)	Month 1	Month 2	Month 3	Month 4	Month 5	Month 6
Total Viable Counts (TVC) (cfu/g)	6.5×10^3	4.5×10^3	2×10^3	1.5×10^3	<100	•	•
Staphylococcus-Micrococcus (cfu/g)	3.4×10^3	2.5×10^3	1.5×10^3	<100	•	•	•
Staphylococcus aureus (cfu/g)	2.5×10^2	1.5×10^2	<100	•	•	•	•
Yeast-Moulds	•	•	•	•	•	•	•

• Not detected

Table 4: Sensory evaluation of salted *Hydrocynus forskalii* by taste panel

Parameters	0 (week)	Month 1	Month 2	Month 3	Month 4	Month 5	Month 6
Odour	9.1 ± 0.368^a	8.5 ± 0.368^b	8.2 ± 0.368^c	6.5 ± 0.10^d	6.0 ± 0.10^e	5.6 ± 0.10^f	4.2 ± 0.10^g
Taste	9.3 ± 0.10^a	8.1 ± 0.368^b	7.2 ± 0.368^c	6.6 ± 0.10^d	5.8 ± 0.10^e	5.5 ± 0.10^f	4.2 ± 0.10^g
Colour	9.5 ± 0.368^a	8.7 ± 0.368^b	8.2 ± 0.368^c	7.2 ± 0.368^d	6.4 ± 0.10^e	6.2 ± 0.10^f	5.7 ± 0.10^g
Texure	8.7 ± 0.10^a	7.2 ± 0.10^b	7.0 ± 0.100^c	6.5 ± 0.368^d	5.9 ± 0.10^e	5.3 ± 0.10^f	4.2 ± 0.10^g
General appearance	9.0 ± 0.368^a	8.4 ± 0.368^b	7.3 ± 0.368^c	6.2 ± 0.10^d	5.5 ± 0.10^e	5.2 ± 0.10^f	4.5 ± 0.10^g

Values are shown as mean ± standard deviation of triplicate measurements. Different superscript letters in the same row indicate significant differences between groups ($p<0.05$)

significant difference ($p<0.05$) in the sensory evaluation during storage period based on the panel's score (Table 4). In the present experiment, scores are the average of 20 panel taste sheets. It could be noticed that salted samples at zero (week) has received higher scores, followed by one month. Samples, has received lowest scores at three months and at six months the product was rejected. This wide range indicated the diversity in the final quality and can be largely attributed to the effect of various conditions upon the salting agents and activities. It is seen that the main factor affecting the quality is time of storage.

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