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EFFECT OF DIFFERENT SALT CONCENTRATION ON TOTAL BACTERIAL COUNT AND HEAVY METAL COMPOSITION OF THE FISH *Hydrocynus spp.*

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ABSTRACT: *This piece of work was done in an attempt to evaluate the issue of the traditional fish salting practice in the Sudan. Fassiekh was selected as one of the widely consumed salted fish product, of great preference among Sudanese consumers. The study was directed towards the study of the heavy metal concentration and the microbiological analysis of fresh fish and fassiekh to compare the effect of the different salt concentrations. One kind of fish species preferable by Sudanese consumers in fasseikh making was selected for this study namely hydrocynus spp (kass). Samples were taken from Elmawrada fish market, and subjected to three salt concentration levels (15%, 20%and 25%) by weight to achieve the goals of the study. Fresh fish were carefully handled throughout the preparation process; they were eviscerated and cleaned up and divided in to two groups then three sup groups to be treated with different salt concentration. After the fermentation process sample were taken to do the heavy metal concentration analysis and microbiological analysis. It was observed that the heavy metal (Arsenic, Cadmium and lead) concentration were not significant. But the microbiological analysis result showed significant decrease in total bacterial count in all concentrations.*

ORIGINAL ARTICLE

Key words: *Heavy Metal, Bacteria, Fish, Salt.*

INTRODUCTION

Fish is widely consumed in many parts of the world by humans because it has high protein content, low saturated fat and also content omega fatty acids known to support good health. Marine foods are very rich sources of mineral component (Sikorski et al., 1990). The global contribution of fish as a source of protein is high, ranging from 10% to 15% of the human food basket across the world (Wilson et al., 2007). Despite the fact that the nutritional value of fish is well known, it nevertheless plays only a limited role in the diet of many countries. Therefore, it would seem appropriate to find new processing methods for this compared valuable raw material so as to increase consumer interest. Compared to mammalian meat, fish meat has more water and less connective tissue, which contains very little elastin (Kolakowska, 2001).

Fish is most important source of meat that may play an appreciable role in solving food problem in the world especially in the developing countries (FAO, 1991). Fish and fishery products are highly nutritious, in addition to the high percentages of animal protein, they provide several other nutrients such as vitamins A and B especially in the liver, and E and K vitamins, and they are good sources of some minerals like Calcium, Phosphorus and Iron (Lunven, 1982).

Salting is one of the earliest techniques for preserving fish. Salting preserves by lowering the moisture content of the fish to the point where bacterial and enzymatic activities are retarded. Spoilage organisms generally can not survive long at salt contents of 6 to 8 (Wet basis of the fish) or higher (Wheaton and Lawson, 1985).

The major goal for the food processing industry is to provide safe, wholesome and acceptable food to the consumer. Control of microorganisms is essential in meeting this goal. This control is partly exerted through processing and preservation techniques that eliminate microorganisms or prevent their growth. It is also required that the basic hygienic level during processing is high and that efficient cleaning and disinfecting procedures that eliminate spoilage and bacteria are used. Many food pathogenic and spoilage bacteria are able to attach to food contact surfaces (Fonnesbech Vogel et al., 2001).

The objectives of this study were to assess:

1) The safety of the product by the microbiological parameters of both fresh *Hydrocynus spp* and *fessiekh.*

2) More details about fish salting and to know the effect of salting on the heavy metal concentration.

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MATERIALS AND METHODS

Sample collection

Fresh fish samples were collected from EL Mawrada fish market the samples was kass (*Hydrocynus spp*). A total of 6 kg each have about 100-110g stored In Iced container and transferred to the fisheries laboratory in Sudan university department of fisheries and wildlife biology for preparing and processing. Random samples were taken to do the chemical and microbiological analysis of fresh fish.

Preparation and processing of fish

Preparing fassiekh takes place as follows: first fresh fishes were individually gutted and washed by tap water and placed on a plastic dishes to dry and then weighted on sensitive balance (FEJ-2000B).the addition of salt is made relevant to weight of the fish. To get the effect of salting of fish after processing, fish were devided in to tow group each have three kg, the first group for take the effect of salting on the product weight and the second to do the chemical and microbiological examination, each group divided in to three groups each group was about (1) Kg then subjected to a different salt concentration (15%, 20%, and 25%). Each fish was salted separately, with coarsely ground salt, applied all over the body with especially on the gill area and the cavity of the gutted specimens. Then the fish were stacked in layers separated by layers of salt on plastic container and covered with a heavy cover. The fish are thus left to undergo fermentation for (10) days.

Random sample were taken on the 10th day to do microbiological and chemical analysis

Bacteriological examinations: Fresh and salted fish was analyzed for the determination of the total count of bacteria.

Culture media used

Aerobic Plate Count: It is a simple media. Is to be prepared according to manufactures instructions by suspending 20.1 grams of the powder in one liter of distilled water, boiling until dissolved completely and then sterilizing by autoclaving at 121 C^{0}for 15 min.

Serial dilution: 1 gram of fish was added to warm 37 C^0 (9) ml normal saline then shook well to distribute all the organism within test tube. Then one ml from previous mixture (fish +normal saline) was transferred with a sterile 1 ml graduated pipette to 9 ml sterile normal saline in screw – capped bottle (Bijout bottles) and then mixed thoroughly. Using another sterile pipette, one ml of the dilution prepared was transferred to a second bottle and the process was repeated to make ten fold dilutions as described by Harrigan and MacCance (1976).

Total Bacterial Counts

Total bacterial count in the fresh and salted samples was determined a sterile pipette was used to transfer 1 ml of the selected dilution into duplicate sterile plates. Nutrient agar was used. The plates were incubated at 37 C^0 for 48 hours; Colonies were counted.

Determination of minerals

The heavy metal (lead, cadmium, arsenic) were determined by (perkin Elmer A (Analyst 700) Atomic Absorption Spectrophotometer following the method described by the manufacture.

Statistical analysis

Results were analyzed using the SPSS computer program / one way (ANOVA).

RESULTS

Table 1 shows Heavy metals content (mg/100g) of fresh fish and *fassiekh* with different salt concentration (15%, 20%and 25%) expressed on wet matter basis.

Table 1 - The effect of salt concentration level on heavy metal contents of *hydrocynus spp.*

Salt (%) / Parameters	0	15	20	25	Significance
Arsenic	0.199±0.13	0.201±0.05	0.201±0.05	0.204±0.00	NS
Cadmium	0.175±0.00	0.181±0.04	0.179±0.00	0.182±0.03	NS
Lead	0.324±0.05	0.326±0.05	0.326±0.05	0.326 ±0.05	NS

NS: not significant

Microbiological analysis

Table 2 shows the total bacterial count of fresh *(Hydrocynus spp.)* fish and *fassiekh* with different salt concentration (15%, 20%and 25%; After 10 days) by CFU/gm.

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Table 2 - The effect of salt concentration level on the total bacterial count of *hydrocynus Spp.*

Parameter / Salt (%)	Total plate count (TPC)
0	$58.1\times10^3 \pm 21.1\times10^3$ [a]
15	$10\times10^3 \pm 1\times10^3$ [b]
20	$7.8\times10^3 \pm 0.76\times10^3$ [b]
25	$4\times10^3 \pm 1\times10^3$ [b]
Significance	*

* Significant at (P≤0.05). [a,b] means within the same column followed by the different superscript are significantly (P≤0.05) different. The total bacterial count of fresh fish is 58.1×10^3 and in fassiekh with (15%, 20%and 25%) salt concentration is 10×10^3, 7.8×10^3 and 4×10^3, respectively.

DISCUSSION

Arsenic content

The arsenic content of fresh fish and salted fish (15%.20%, and 25%) under investigation were 0.199 and 0.0201, 0.201, and 0.204 mg/100g respectively. this result is lower than that reported by (Sivaperumal et al., 2007) who found the arsenic content in *Etroplus ssuratensis* about 1.515 mg/kg. Salting and fermentation not affected significantly (P<0.05) the arsenic content in the product.

Cadmium content

The cadmium content of fresh fish and salted fish (15%, 20% and 25%) under investigation were 0.175 and 0.181, 0.179, 0.182 mg/100g respectively the permissible limit for Cadmium in sea bass is 0.1 mg/kg wet weight (EC, 2001). Salting and fermentation treatment not affected cadmium value at (P<0.05).

Lead content

The lead content of fresh fish and salted fish (15%, 20%25%) under investigation were 0.324and 0.330, 0.477, and0.464 mg/100g respectively. This result is lower than that reported by (Dural et al., 2007) who found that the lead content about 0.40–2.44 mg kg^{-1} for muscles and 1.41–3.92 mg kg^{-1} for livers of fish from Tuzla Lagoon . Salting and fermentation treatments not affected lead value at (P<0.05).

Microbiological parameters

Table 2 shows the total bacterial count values of fresh *hydrocynus spp* fish and *fassiekh* with (15%, 20% and 25%). The data showed significant different, the highest value (58.17×10^3) was reported in fresh fish, while the lowest value (4×10^3) was reported in fassiekh with 25% salt concentration after 10 days .this result is lower than that reported by (Vishwanath et al., 1998) who found the total plate count of bacteria in fresh *Monopterus albus* about (1.2×10^{-6} 1.0×10^{-7}). This decrease in total bacterial count may be due to the presence of high salt concentration in fessiekh so the pathogenic microorganism growth is controlled.

Also this result is in agreement with the findings of (El-Tom, 1989 and Abu Giddeire 2001) who reported that the count of microorganism increased rapidly during first fermentation days and began to decrease later. And in agreement with Ahmed (2006).

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