

Quality and Shelf – Life Assessment of Variously Processed Catfish and Tilapia Stored at Ambient Temperature

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ABSTRACT

Aims: The effects of oven – drying (OVD), brining and oven – drying (BOVD) and smoking (SMK) on quality and shelf – life of Catfish (*Chrysichthys nigrodigitatus*) and Tilapia (*T. zillii*) were evaluated.

Study design: Completely randomised block design.

Place and Duration of Study: Department of Environmental Management & Toxicology, Federal University of Agriculture Abeokuta, Nigeria, between February 2011 and August 2012.

Methodology: Life fish were collected and processed using three local preservation methods. Processed fish samples were stored in perforated plastic containers for 11 successive weeks at ambient temperature (25-32°C) and assessed weekly for levels of trace metals (Fe, Zn, Cd, Pb) and physical attributes of odour, flavour and texture. Microbial load was assessed by total viable count (TVC) and biochemical activity by total volatile nitrogen (TVN).

Results: This study showed that brined/ oven-dried fish was the most accepted followed by smoked fish. The quality of processed fish under ambient conditions decreased with increasing storage time in the order OVD>SMKD<BOVD. Statistical analysis showed that no significant ($P=0.05$) differences existed between the trace metal contents of the samples from the three processing methods. Also, samples processed using the three processing methods contained levels of trace metals below the recommended limits for trace metals in fish and are therefore safe for human consumption. However, method of processing had significant effects ($P=0.05$) on the TVC and TVN content of fish.

Conclusion: Brined/oven dried samples were most organoleptically accepted and deteriorated the least. *Chrysichthys nigrodigitatus* deteriorated faster in odour and flavour than *Tilapia zillii*. Caution should be exercised in the consumption of processed fish stored on open shelf for very long weeks (above 5 weeks) as they could contain higher amounts of microbial cells.

Keywords: Trace metals, Quality, Brining/Oven drying, Smoking, Total viable count, Total volatile nitrogen

1. INTRODUCTION

Among the food resources of the world, fish and fishery products are very important as sources of protein, especially in countries that are unsuitable for livestock production. Fish is a highly

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proteinous food consumed by a larger percentage of populace because of its availability and palatability [1]. However, quality of harvest is markedly affected by the ease of deterioration and spoilage in fresh fish. As such, processing provides a way out to avoid losses due to quality deterioration and spoilage.

Although fish is a good source of some essential nutrients, processing and processing practices could cause modifications in proximate composition, fatty acids and amino acids as well as changes in the nutritional quality of fish and trace metal composition [2].

The study of mineral elements present in fish products is of biological importance; since many of such elements take part in human metabolic processes and are known to be indispensable to all living things [3]. A deficiency in these principal nutritional mineral elements induces a lot of malfunctioning; as it reduces productivity and causes diseases, such as inability of blood to clot, osteoporosis, anaemia etc. [3, 4]. Also, the measurement of quality is often necessary to ensure that fresh and processed fish meets the requirements of food regulations and commercial specifications [5].

Therefore, considering the possible health risks and the nutritional benefits associated with fish consumption; this study investigated the effect of three local processing methods on the quality and shelf life of *Chrysichthys nigrodigitatus* and *Tilapia zillii*, two common fresh water fish species readily available and consumed in Nigeria.

2.0 MATERIALS AND METHODS

2.1 Sample Collection

Life fish, Bagrid catfish and Tilapia (*Chrysichthys nigrodigitatus* and *Tilapia zillii* respectively) with average weights of $250 \pm 15\text{g}$ were obtained from fishermen at Ibaro village along the Oyan Lake, Ogun State, South West Nigeria. The fishes were stunned immediately after capture by applying a blow to the head. They were immediately gutted and washed with clean portable water. The fish were cut into about 50 g pieces and washed again with distilled water. The samples were then separated into three parts, packed in ice packs and transported to the laboratory.

2.2 Fish Processing

For the oven – drying treatment (OVD), one part of samples were dried in an oven at an initial temperature of 45°C for two (2) hours, then at 105°C to a moisture content of $15 \pm 5\%$. For brining and oven - drying treatment (BOVD), a part of samples were soaked in a 36% cooking salt solution [6]. for 30 minutes. The samples were drained on white cardboard sheets in the laboratory and dried in an oven at an initial temperature of 45°C for two (2) hours, then at 105°C to a moisture content of $15 \pm 5\%$. For smoking treatment, another part of samples were drained of water and smoked to a moisture content of $15 \pm 5\%$ in a neatly prepared traditional open drum and fired from below with wood. Iron mesh were placed 20cm from the top of the drum on which the samples were placed to be smoked. The drum was covered with aluminium foil to conserve heat and smoke. All samples cooled and exposed to ambient temperatures to air-dry. The samples were stored in perforated clean plastic containers at ambient temperature ($25 - 32^{\circ}\text{C}$) and labelled accordingly.

2.3 Determination of Freshness Quality

2.3.1 Organoleptic Assessment

Processed fish were placed in transparent polyethylene bags and sample coded. Consumer acceptance of odour, flavour, and texture of samples were assessed by a panel of assessors. Total sensory score obtained from an addition of individual sensory score was graded using a 15 point

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hedonic scale modified from literature [6, 7]. Grades were allotted depending on their quality as follows: $12 < S \leq 15$ = very fresh, $9 < S \leq 12$ = fresh, $6 < S \leq 9$ = poor, $3 < S \leq 6$ = bad, ≤ 3 = very bad.

2.3.2 Total Viable Count

1g of fish muscles was weighed aseptically and homogenised in 10 ml sterile peptone water. Serial dilutions of the mixture were prepared and 0.1 ml of diluent was spread on already prepared plates of nutrient agar. Duplicate plates were incubated at $25 \pm 5^\circ\text{C}$ for 24 hours. The total colonies were counted to represent the total number of bacterial cells (TVC) capable of forming colonies.

2.3.3 Total Volatile Nitrogen

Total volatile nitrogen was determined using the colorimetric method [8]. 0.5 g of fish muscle was digested using 30 ml Sulphuric acid and Selenium tablet. The digests were made up to 100 ml in clean volumetric flasks. 100 ml standard solutions of NH_4SO_4 (0, 5, 10, 15, 20 and 25 ml of the 100 $\mu\text{g/ml}$ NH_4^+ -N) were prepared from NH_4SO_4 .

0.1 ml each of the standards and samples were measured using pipette into clean test-tubes and 5.0 ml of reagent N1 was added to each test tube, mixed well and left for 15 minutes. Then 5.0 ml of reagent N2 was added to each test tube, mixed well and left for 1 hr for full colour development. The absorbance of standards and samples were read at 655 nm using a Shimadzu UV- 1800 spectrophotometer. Calibration curve of absorbance against standard concentration was plotted and the TVN concentrations determined from the curve. Reagents were prepared at least 24 hours before analysis.

Reagent N1: 34 g sodium salicylate, 25 g sodium citrate and 25 g sodium tartrate were dissolved in 750 ml water. 0.12 g sodium nitroprusside was added and when dissolved made up to 1000 ml with distilled water.

Reagent N2: 30 g sodium hydroxide was dissolved in 750 ml water. The solution was allowed to cool and 10 ml sodium hypochlorite solution was added and made up to 1000 ml with distilled water.

Both reagents were prepared at least 24 hours before analysis.
All chemicals and reagents used for analysis were of analytical grade.

2.3.4 Trace Metal Analysis

1 g of fish muscle was weighed and mixed with 20 cm^3 of the digestion mixture (Nitric acid/Hydrogen peroxide - 3:1). The mixture was heated until fuming ceased. The resulting digest was filtered using Whatman filter paper No. 42 and made up to 100 cm^3 with de-ionised distilled water. The digest were analysed for Zn, Fe, Cd and Pb using a Buck 205 AA Atomic Absorption Spectrophotometer.

2.3.5 Statistical Analysis

Data from laboratory analysis are expressed using illustrative charts. Results are expressed as mean of triplicate trials. Data were analysed by one-way analysis of variance and correlation analysis. Least significance difference (LSD) test was used for means separation for statistical significance at 95% ($P=0.05$) confidence level.

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3.0 RESULTS AND DISCUSSION

All processing methods followed the usual procedures used to prepare fish for table consumption in Nigeria and fresh batches of fish muscles were analysed to serve as baseline for comparison (Table 1).

Table 1: TVC, TVN and Trace metal concentrations of fresh fish muscle.

	<i>Chrysichthys nigrodigitatus</i>	<i>Tilapia zillii</i>
Total viable count (cfu/g)	5.5×10^5	4.0×10^5
Total volatile nitrogen (mg N/ 100g)	3.340	2.870
Iron (Fe) (mg/Kg)	1.931	2.241
Zinc (Zn) (mg/Kg)	3.753	2.574
Cadmium (Cd) (mg/Kg)	0.013	0.045
Lead (Pb) (mg/Kg)	< 0.050	< 0.050

3.1 Organoleptic Assessment

The sensory scores of processed fish are presented in figs 1 and 2. Method of processing had no significant ($P=0.05$) effect on the sensory acceptance of processed samples (Table 1). There was a general decline in the physical attributes such as odour, flavour and texture of fish as the weeks progressed in the order : Brined/Oven dried > Smoked > Oven dried (i.e. BOVD > SMKD > OVD). The reduction in the sensory qualities with increase in the storage period of processed fish could be attributed to higher activities of the spoilage agents. Similar trend was observed in smoked fish [9] and crustaceans (Oyster and Shrimps) under storage [10].

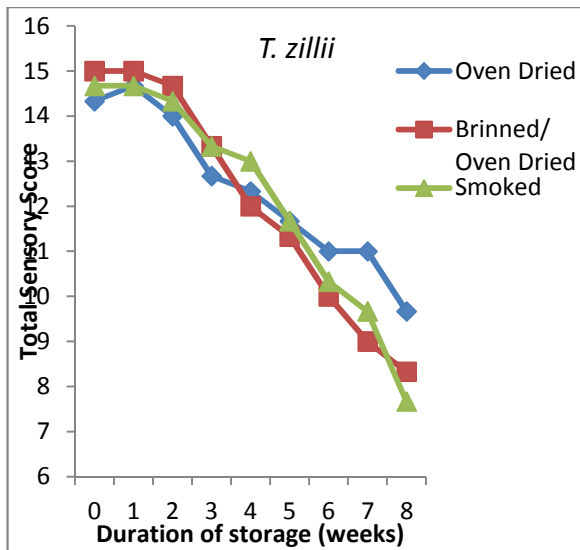
Among the three processing methods, BOVD samples though most organoleptically accepted reduced the most in texture and odour as the duration of storage increased. Hence, it can be assumed that BOVD samples absorbed moisture the most from the surrounding air. Reduction in texture could be due to the possibility of magnesium chloride impurities in the salt used for processing. Magnesium chloride is hygroscopic and if present in salted fish makes it damp paving way for bacterial and mould spoilage [11] Also, in the BOVD samples, salt has the potential to accelerate fat oxidation [11] accounting for the decrease in odour.

Based on sensory assessment, the processed fish samples were unacceptable by the end of the 6th week. Fluffy woolly mat of moulds was noticed on fish samples from the 6th week of storage. There was also a significant colour change in all three groups as from the 7th week.

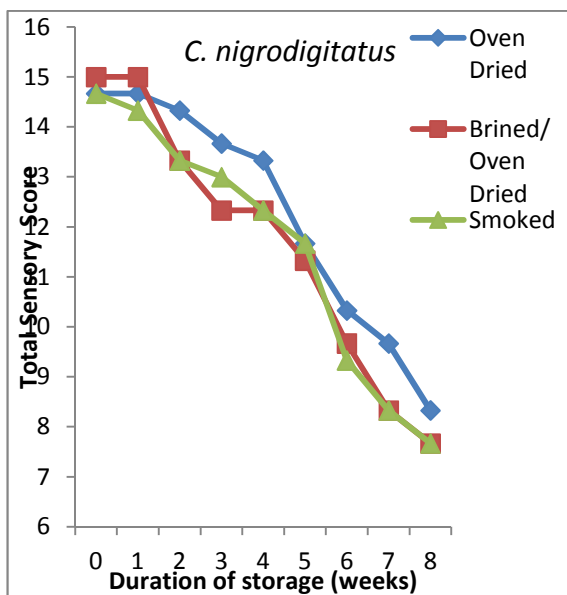
Chrysichthys nigrodigitatus decreased greatly in odour and flavour as the duration of storage increased compared with *Tilapia zillii*.

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Figs 1: Sensory scores of processed *Tilapia zillii*



Figs 2: Sensory scores of processed *Chrysichthys nigrodigitatus*.

3.2 Total Viable Count

The mean total viable count (TVC) of processed fish is presented in figs 3 and 4. Results showed a significant ($P=.05$) difference between the total viable count of BOVD, SMKD and OVD (Table 1). BOVD had the lowest TVC out the processing methods studied. The introduction of heat during all the processing methods employed would not only kill microorganisms but will also reduce the moisture content of the fish muscles making the environment less favourable for microbial growth. However, heating does not destroy all organisms as thermophiles may survive in dried fish after

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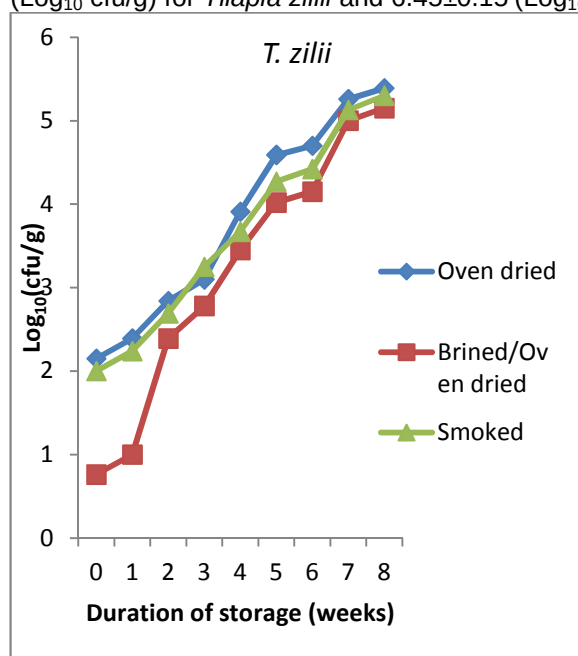
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heating, accounting for the higher microbial count of oven dried samples. The inclusion of salt (in case of BOVD samples) and smoke (SMKD samples) along with heating usually provides a more efficient method of processing, accounting for the lower microbial count in brined/oven dried and smoked samples [12].

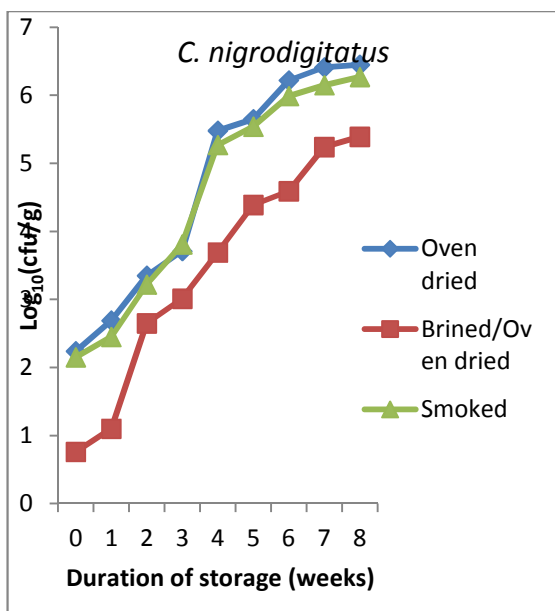
TVC of fish muscles increased significantly ($P=0.05$) with increase in the duration of storage. As the duration of storage increase, processed fish samples may absorb small amounts of moisture from surrounding atmosphere providing enabling environment for microbial growth [11]. Smoked samples also had relatively lower total microbial count when compared with oven dried samples as the duration of processing increased. This can be explained by the bactericidal effect of smoke constituents such as acids, aldehydes and phenols [11]. It has also been reported that the addition of the phenolic fraction of wood in the concentration of 45mg/kg resulted in inhibitory effects on *Staphylococcus aureus* [13]. Yeast and moulds are however more resistant to the inhibitory effect influence of smoke even up to a concentration of 60 mg/kg.

While the microbial load of *Tilapia zillii* did not exceed the recommended maximum bacterial count for good quality fish, fluffy woolly mat of moulds were noticed on the fish samples by the 7th week. However, by the 6th week, oven dried and smoked samples of *Chrysichthys nigrodigitatus* had total viable counts exceeding 5.7 ($\text{Log}_{10} \text{cfu/g}$).

The highest total viable count was recorded at the 8th week in oven dried samples: 5.39 ± 0.09 ($\text{Log}_{10} \text{cfu/g}$) for *Tilapia zillii* and 6.45 ± 0.15 ($\text{Log}_{10} \text{cfu/g}$) for *Chrysichthys nigrodigitatus*.



Figs 3: Total viable count ($\text{Log}_{10} \text{cfu/g}$) of processed *Tilapia zillii*



Figs 4: Total viable count (Log₁₀(cfu/g)) of processed *Chrysichthys nigrodigitatus*

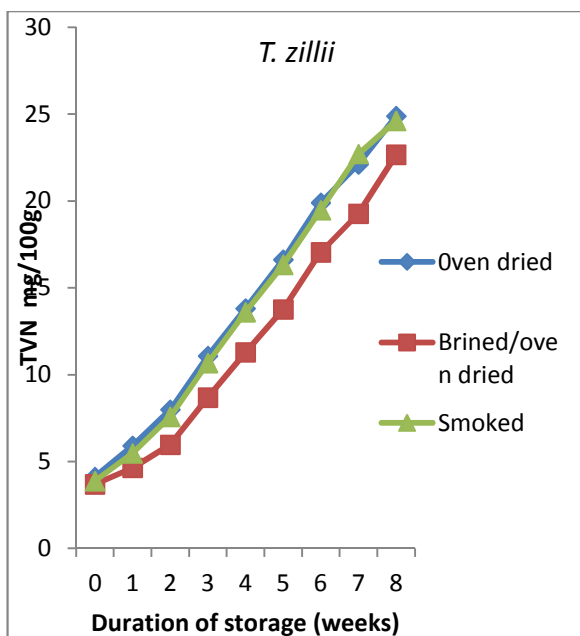
3.3 Total Volatile Nitrogen

Mean total volatile nitrogen (TVN) of processed fish samples are presented in figs 5 and 6. Method of processing has a significant ($P=.05$) effect on the total volatile nitrogen content with brined/oven dried fish samples having the least total volatile nitrogen content out of the three processing methods used (Table 1). The variation in total volatile nitrogen content is partly related to the microbial load of the processed samples analysed each week. The viable count of processed fish samples was in the order: BOVD < SMKD < OVD. As such, the microbial breakdown of fish protein is expected to be of same trend.

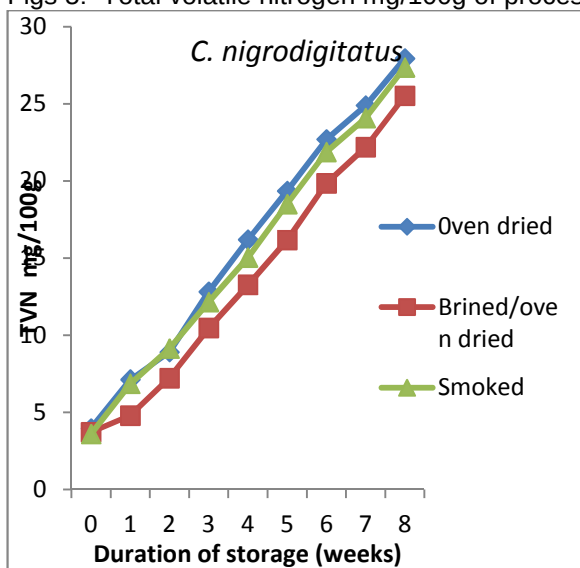
The lower TVN content in BOVD can also be due to the presence of salt which apart from reducing microbial growth also reduces autolytic enzyme activities thereby reducing protein breakdown [14]. The highest total volatile nitrogen content was recorded at the 8th week in oven dried samples: 24.89 ± 0.01 mg/100g for *Tilapia zillii* and 27.94 ± 0.14 mg/100g for *Chrysichthys nigrodigitatus*. TVC had a strong positive correlation ($P=.05$) with the TVN of fish samples, e.g. ($r = 0.973$ OVD, $r = 0.926$ for BOVD and $r = 0.985$ for SMK).

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Figs 5: Total volatile nitrogen mg/100g of processed *Tilapia zillii*



Figs 6: Total volatile nitrogen mg/100g of processed *Chrysichthys nigrodigitatus*

3.4 Trace Metal Content

Mean metal concentrations in fish samples are expressed in table 2. Method of processing had no significant ($P=0.05$) effect on the trace metal content of fish samples. Among the three processing methods studied, SMKD samples had the highest trace metal content. The slightly higher concentration of concentrations in SMKD can be due to weighing error or the possibility of contamination from smoke particles used for the processing process [15]. The higher concentration of Fe and Zn in the fish samples analysed can be due to the natural abundance of these

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metals in Nigerian soils and since the source of metal depositories are aquatic [16]. The Cd content of the fish samples was lower than Fe and Zn with the highest concentration of 0.081 ± 0.004 mg/Kg in smoked *Chrysichthys nigrodigitatus*. A possible explanation to this could be due to the fact that Cd is easier to leach from fish muscle than Fe and Zn [17]. Pb was below 0.05 mg/Kg in both fresh and processed samples of all fish species.

The content of all Fe, Zn, Pb and Cd in all processed fish samples were lower than the maximum allowable limit [18, 19] and had no correlation with the duration of storage remaining almost constant throughout the period of study.

Table 2: Sensory score, TVC, TVN and trace metal concentrations of processed fish samples

	Sensory score	TVC $\log_{10}(\text{cfu/g}) \pm \text{SD}$	TVN mg N/ 100 g	Fe mg/kg	Zn mg/kg	Cd mg/kg	Pb mg/kg
<i>Tilapia zillii</i>							
OVD	14.33 ± 0.58^a	2.15 ± 0.15^a	4.117 ± 0.003^a	3.712 ± 0.003^a	7.214 ± 0.002^a	0.023 ± 0.002^a	< 0.05
BOVD	15.00 ± 0.00^a	0.76 ± 0.02^b	3.690 ± 0.009^b	3.713 ± 0.002^a	7.221 ± 0.002^a	0.024 ± 0.003^a	< 0.05
SMKD	14.67 ± 0.58^a	2.00 ± 0.00^a	3.884 ± 0.014^c	3.714 ± 0.006^a	7.246 ± 0.013^a	0.024 ± 0.002^a	< 0.05
<i>Chrysichthys nigrodigitatus</i>							
OVD	14.67 ± 0.58^a	2.15 ± 0.15^a	3.966 ± 0.125^a	4.640 ± 0.000^a	5.161 ± 0.000^a	0.079 ± 0.000^a	< 0.05
BOVD	15.00 ± 0.00^a	0.76 ± 0.02^b	3.724 ± 0.550^b	4.652 ± 0.003^a	5.162 ± 0.004^a	0.079 ± 0.000^a	< 0.05
SMKD	14.67 ± 0.58^a	2.00 ± 0.00^a	3.605 ± 0.162^c	4.713 ± 0.001^a	5.164 ± 0.002^a	0.081 ± 0.004^a	< 0.05

Values with different superscripts in the same column for each specie indicate significant difference at $P=.05$

Values are means $\pm$ Standard deviations of triplicate determinations.

4.0 CONCLUSION

In conclusion, fish samples processed by the methods of oven drying, brining/oven drying and smoking underwent loss in quality with duration of storage. However, brined/oven dried samples were most organoleptically accepted and deteriorated the least. *Chrysichthys nigrodigitatus* deteriorated faster in odour and flavour than *Tilapia zilli*. From results, the optimum storage period of the processed fish species is 4-5weeks. Therefore, in spite of being physically firm, caution should be exercised in consuming processed fish stored on open shelf for very long weeks as they could contain higher amounts of microbial cells.

Results of this study also showed that consumption of smoked and brined/oven dried fish is safe with respect to Fe, Zn and Cd concentrations. However, since metal concentration of processed

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fish muscle is dependent on fresh content of fish, regular monitoring of fish is required to detect sudden changes in trace metal concentration of fresh fish muscle.

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