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# Human Health Risk of Frozen Fish Species *Scomber scombrus* and *Merluccius capensis* Sold from Markets in Benin City, Edo State, Nigeria

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**Abstract**

*This study assessed heavy metal concentration and human health risks associated in consumption of fish fillets from two commercially imported frozen fish species sold in the markets of Benin City, Edo State. Concentrations of Iron (Fe), Cadmium (Cd), Copper (Cu), Zinc (Zn) and Lead (Pb) were quantified in Merluccius capensis and Scomber scombrus fish using Atomic Absorption Spectrometry (AAS) (Model AA 500L). The results revealed that all metals analysed in both fishes were below the recommended allowable limit. Mean fish fillet levels were accumulated in the order: Fe>Zn>Cu>Pb>Cd in both fish species. The estimated daily intakes of all the metals, were below the Provisional Tolerable Daily Intake (PTDI). The estimated Target Hazard Quotient (THQ) ranged between 0.007- 0.5146 and TCR values of all the metals were in the range of 1.89E-05 and 2.42E-05, which were lower < 1 indicating no carcinogenic risk. Cumulative cancer risk (ΣILCR) in both fish species did exceed the cancer benchmark. Health risk assessment to humans via dietary intake of fish indicates adverse health effects of fish intake. The heavy metal burden in the different varieties of frozen fish obtained from the markets demonstrates enormously worrisome risks for the population that consumes them. Thus, we recommend that the imported frozen fish species in the studied areas should be closely monitored to protect consumer health.*

## 1.0. Introduction

Nigeria is Africa's 3rd largest producer of fish with production at about 1,169,478 metric tons, accounting for 40% of its total annual fish demand of 3.4 million metric tons; while the rest of 60% of the annual fish demand is met through importation [1]. Consumption of imported frozen fish in Nigeria has superseded many sources of animal protein with massive frozen fish imports make the country Africa's top importer of frozen species [2]. Seasonal availability of indigenous species in Nigeria is not always, available particularly in light of the numerous reports of contaminated wild fisheries resources from aquatic systems. Thus, frozen fish are a preferred source of protein and are easily accessible to the populace [3]. Safety of frozen fish imported throughout the world is of concern as these fishery resources may be contaminated with toxic chemicals from polluted water ways [4,5,6]. Heavy metals are one of the significant and ubiquitous pollutants reported from aquatic systems, due to their potential toxicity and their capacity for bioaccumulation and biomagnification in aquatic ecosystems [7]. Metals, particularly Pb, Hg, Cd and As constitute a significant potential public health threat to humans, when consumed over a prolonged period of time. Heavy metals in fish are a recurrent problem that is constantly been addressed [8,9,10] but still arouses considerable interest. Food distributors now place a high value on factors like product quality and consumer safety guarantees for humans who consume these fishes. To detect current dietary intakes of heavy metals in developing countries, the Codex Committee on Food Additives and Contaminants recommends that dietary intakes of heavy metals of significant public concern be regularly monitored and promptly updated. Thus, this study investigates the concentration of heavy metals in the muscle of commercially consumed frozen fish sold in Benin market Edo state, to ascertain their safety for human consumption.

## 2.0. Materials and Method

### *2.1. Sample Collection and Preservation*

Imported frozen fish species of *Merluccius capensis* and *Scomber scombrus* commonly consumed were purchased from fishmonger at two major markets in Benin City metropolis. Fish samples (N=20) were collected in duplicates, from New Benin (6º21' 4.6728 5º37'48.3888E") and Oba market (6º22'26.2128N 5036'49.1688E") and transported to the laboratory in ice chest with sterile labelled polythene bags. Fish samples were weighted, before dissection to collect the muscle using the methods recommended by UNEP/FAO/IAEA/IOC [11].

### *2.2. Heavy Metal Analysis*

Samples of fish fillets from each fish species were separately then dried in an oven at 110ºC to get a uniform weight. The tissue samples were pulverised and homogenised by crushing them in a Teflon mortar. Two (2g) of fish flesh was digested by microwave in Teflon vessels using 6 mL concentrated $HNO_3$, 2 ml of $H_2O_2$ 30% and 2 mL of concentrated HF; HF was eliminated using $H_3BO_3$ [12]. The solution was transferred into a polyethylene volumetric flask and diluted to 100 ml with milliQ water. One milliliter of the solution was then diluted to 10 ml by adding $HNO_3$. All glassware and plastic containers were thoroughly cleaned with 10% nitric acid solution and milliQ water. Atomic Absorption Spectrometry (AAS) (Model AA 500L) was used to determine the heavy metal concentrations (Fe, Pb, Cd, Cu, and Zn) in the fish fillet samples.

### *2.3. Quality Control and Quality Assurance*

The reagents utilised in this study were from Sigma-Aldrich company. All experiments used double-distilled and de-ionized water, unless otherwise specified. To avoid contamination, the instrument was thoroughly cleaned before use. The contamination of the reagents was tested using reagent blanks. A spike-recovery test was performed on random fish samples to ensure the analytical technique and digestion were accurate and precise. The laboratory conducts intra-analyst comparisons (IACs) at least twice a year, as well as proficiency testing and inter-laboratory comparisons. The total recoveries for heavy metals (Fe, Pb, Cd, Cu and Zn) were greater than 80%, and the detection limit for all metals tested was 0.006 mg/kg.

### 2.4. Human Risk Assessment in Fish Muscle

#### 2.4.1. Estimated Daily Intake (EDI)

Estimated daily intake (EDI) of metal concentration (Fe, Zn, Cu, Pb and Cd) through fish consumption was estimated using the equation from USEPA [13].

$$\mathbf{EDI = Cm \times IR \times EF \times ED / BW \times AT} \qquad \mathbf{(1)}$$

Cm = Concentration of metal, IR = Ingestion rate (0.0255 kg/day for adults, EF = Exposure frequency (350 day/year), ED = Exposure duration (30 years), BW = Body weight (70kg), AT = (70) × 365 day/year) 25550 days.

### 2.4.2. Target Hazard Quotient (THQ)

**THQ =EDI/RFD (2)**

THQ ≤ 1 is acceptable risk level and THQ >1 indicate a potential risk level. Oral reference dose RFD; Fe, 0.007, Pb, 0.0035, Cd, 0.0005, Cu,0.04 and Zn, 0.3 respectively [14]

### 2.4.3. Hazard Index (HI)

Hazard index (HI) estimates the potential risk of all the metals collectively and is calculated as sum of THQ [15]

**HI = THQFe + THQPb + THQCd + THQCu +THQZn (3)**

If the HI value obtained is below 1, indicates no adverse effect to human health.

### 2.4.4. Incremental Lifetime Cancer Risk (ILCR)

Incremental Lifetime Cancer Risk (ILCR) is a calculation to estimate the likelihood of an individual to develop cancer during their lifetime as a result of exposure to a given carcinogenic substance.

**ILCR = EDI × CSF (4)**

Incremental lifetime cancer risk (the safe limit for cancer risk is ≤ 0.000001, the considerable risk limit is close to 0.000001-0.0001, and the serious cancer risk threshold is ≥ 0.0001).

**CSF = Cancer slope factor (for Pb, Cd and Cu are 0.0085, 0.38, and 0.04mg/kg/day)**

**Total Carcinogenic Risk (∑ILCR)**

**∑ ILCR = ILCRFe+ ILCR Pb + ILCRCd+ ILCRCu+ ILCRZn**

Total carcinogenic risk equal to the sum ILCR of individual metals

### 2.4.5. Statistical Analysis

Descriptive statistics were conducted to determine the mean and Standard Error of the Mean (S.E.M) of investigated toxic metals in the fish samples. The test of significance and one-way ANOVA were also carried out on the data. Statistical analysis was carried out using SPSS 16 and Microsoft Excel 16 packages.

## 3.0. Results and Discussion

### 3.1. Metal Concentrations in Different Fish Species

Table 1 shows, frozen fish fillet samples from Merluza (*Merluccius capensis*) and Titus (*Scomber scombrus*) were examined for Fe > Zn > Cu > Pb > Cd. Statistical no significant differences (p>0.05) were observed between the two species and metals. On average the metal concentration in the studied frozen fish species samples followed an increasing trend of Fe > Zn > Cu > Pb > Cd. It was observed that Fe had the highest concentration resulting in the dominant metal in the two fish species. The mean value of Fe, Pb, Cd, Cu, and Zn in the fish tissues of *Merluccius capensis* were 24.06mg/kg, 0.17 mg/kg, 0.10 mg/kg, 2.24 mg/kg, 14.12 mg/kg and *Scomber scombrus* 23.40 mg/kg, 0.21 mg/kg, 0.10 mg/kg, 3.09 mg/kg, 16.51mg/kg. Metal concentrations assessed were within the Joint FAO/WHO committee's recommended maximum acceptable limits The current study's heavy metal accumulation was compared to similar studies done in different places, and the findings are shown in Table 2. Iron is an essential element for many species and its deficiency causes anaemia in humans [16]. The mean concentration of Fe was below the corresponding values mentioned by Abubakar *et al.* [2] and Ekere *et al.* [5] and the mean values did not exceed the FAO/WHO limit of 100 mg/kg. Consequently, the study suggest that frozen fish samples were not hazardous when Fe contamination was taken into account. However, this low level of Fe may have come from contaminated rivers. Thus, through the consumption of these fish, these metals may be posed a threat. Toxicity due to excessive Fe can led to gills damage and respiratory stress in fish [29]. Pb is a hazardous trace element that enters the human body through fish ingestion [17]. Overdose and excess levels of Pb in the body system can disturb metabolic activities can result in memory loss, retardation, neurotransmitter problems, cardiovascular illnesses and kidney and liver damage [18]. Mean Pb concentrations of the studied fillets were (0.17mg/kg) for *Merluccius capensis*, 0.21 mg/kg for *Scomber scombrus*. The highest Pb content was found in *Scomber scombrus* (0.21 mg/kg). Mean Pb concentration of Trachurus *trachurus* and *Otolithes ruber* was found as 0.1604 mg/kg and 3.64 mg/kg respectively by Sangiuliano *et al.* [30] and Khafeef and Hantosuh [31]. Our obtained values were well within the maximum permissible limit in fish which approximates 2.0 mg/kg [19].

Cd is an extremely toxic element, and the accumulation of this substance in the body can be detrimental to both fish and humans who consume them. Exposure to cadmium has been associated with liver disease, kidney damage, and an elevated risk of cancer in animals and humans alike [20]. Table 2 reveals that the mean value of Cd in this study (0.10 - 0.10 mg/kg) was lower than the corresponding mean value mentioned by Abubakar *et al.* [2] and Ekere *et al.* [5] but higher than the values reported by Oluyemi and Olabanji [21] and Wangboje and Onojesio [22].The average Cu value of (2.24-3.09 mg/kg) in the two fish species was two-and-half-three-fold higher than those reported by Ahmad and Al-subeihi *et al.* [23] and below the concentration threshold set by the FAO/WHO (1.0 mg/kg). Cu is a vital dietary trace element needed by all organisms, including fish. It acts as a cofactor for specific proteins and enzymes involved in various metabolic processes, including antioxidant reactions, electron transport, and the synthesis of heamoglobin and collagen [24]. The US EPA recommends 40 $g/kg^{-1}$ RfD for Cu [25] and excessive quantities might cause health concerns such as liver and renal disorders [20]. Zinc is considered essential for normal growth, and developmental functions in all animal species however, zinc concentrations can become toxic at high consumption [26]. The mean value of Zn in this study was 14.12 mg/kg and 16.51 mg/kg in both fish species, values exceed Wangboje and Onojesio [22] and Bolawa *et al*. [32] and were below the limit of 100 mg/kg set by FAO/WHO committees. Lower Zn concentrations may enhance the fish's metabolic activity, which are critical for their development. Kumar *et al*. [33] found that greater Zn concentrations in fish tissues inhibited metabolism and growth. Furthermore, low Zn levels may produce toxicity, changing the physiological processes of the fish. Overall, Zn was considerably bioaccumulated in the muscle of the fish.

**Table 1: Heavy Metals Concentration (mg/kg) in Frozen Fish Species from Markets in Benin City**

| Heavy metals | Fish species | Origin | Muscle (mg/kg) | FAO/WHO Limits 1989 |
|---|---|---|---|---|
| **Fe** | *Merluccius capensis* | Chile | 24.06 ± 2.13 | 100 |
| | *Scomber scombrus* | USA | 23.40 ± 1.88 | |
| **Pb** | *Merluccius capensis* | Chile | 0.17 ± 0.04 | |
| | *Scomber scombrus* | USA | 0.21 ± 0.03 | 2.00 |

| | | | | |
|---|---|---|---|---|
| **Cd** | *Merluccius capensis* | Chile | 0.10 ± 0.00 | |
| | *Scomber scombrus* | USA | 0.10 ± 0.00 | 1.0 |
| **Cu** | *Merluccius capensis* | Chile | 2.24 ±1.04 | |
| | *Scomber scombrus* | USA | 3.09 ± 1.07 | 30 |
| **Zn** | *Merluccius capensis* | Chile | 14.12 ± 0.82 | 100 |
| | *Scomber scombrus* | USA | 16.51 ±2.50 | |

**Table 2: Heavy Metal Concentrations in the Present Study and Comparing with other Studies**

| Fish species /Area | Fe | Pb | Cd | Cu | Zn | Reference |
|---|---|---|---|---|---|---|
| *Merluccius capensis* | 24.06 | 0.17 | 0.10 | 2.24 | 14.12 | Present Study |
| *Scomber scombrus* | 23.40 | 0.21 | 0.10 | 3.09 | 16.51 | Present Study |
| *Scomber scombrus* | - | 0.43 | 0.07 | - | - | Oluyemi and Olabanji, 2011 |
| *Trachurus Murphyi* | 48.417 | 7.542 | 1.400 | - | - | Abubakar *et al.*, 2014 |
| *Sardinella sindesis* | 45.06 | - | 0.150 | 4.89 | - | Ekere *et al.*, 2014 |
| *Trachurus Trachurus* | 26.90 | 7.30 | 1.050 | - | - | Abubakar *et al*., 2015 |
| *Sepia officinalis* | - | 0.03 | 0.17 | 1.10 | - | Sangiuliano *et al.*, 2017 |
| *Scomber japonicas* | 340.0 | - | - | 2.30 | 150.0 | Bolawa *et al*., 2018 |
| *Merlucclus sp.* | 520.00 | - | - | 2.10 | 390.00 | |
| *Pangasius hypophthalmus* | - | - | - | 0.746 | 16.0 | Ahmad and Al-subeihi *et al.*, 2023 |
| *Trachurus trachurus* | - | 0.1604 | 0.0289 | 0.793 | 0.0289 | Wangboje and Onojesio, 2023 |
| *Otolithes ruber* | - | 3.46 | 0.34 | - | 0.18 | Khafeef and Hantosuh, 2023 |

### 3.2. Health-Risk Assessment for Fish Consumption

Fish is an important dietary item for most human populations since, it is high in protein, and its consumption is highly advised. However, residues of toxic substances in its muscle may cause a variety of severe metabolic alterations in humans [27]. The health risk assessment of heavy metals in frozen fish species consumed in Benin City is shown in Table 2. The computed EDI values for the populations ranged between 5.75E-06 -45E-05 mg/kg/day. The calculated EDI values were generally higher in the population that consumed *Scomber scombrus* fish, but lower than the Provisional Tolerable daily Intakes (PTDIs), showing a situation of no risk for the consumers of the investigated fish.

**Table 3: The health risk indices (EDI, THQ, HI, TCR and ILCR) for metals through fish consumption (mg/kg day$^{-}$1 bw)**

| FISH SPECIES | METALS | EDI | PTDI | THQ | ILCR |
|---|---|---|---|---|---|
| *Merluccius capensis* | **Fe** | 0.0036 | 0.8 | 0.5146 | |
| | **Pb** | 2.55E-05 | 0.0005 | 0.0073 | 2.16E-07 |
| | **Cd** | 1.5E-05 | 0.5 | 0.0299 | 5.24E-06 |
| | **Cu** | 0.0003 | 0.5 | 0.0083 | 1.34E-05 |
| | **Zn** | 0.0021 | 0.3 | 0.007 | |
| **HI** | | | | **0.57** | |
| **∑ILCR** | | | | | **1.89E-05** |
| *Scomber scombrus* | **Fe** | 0.0035 | 0.8 | 0.5004 | |
| | **Pb** | 3.14E-05 | 0.0005 | 0.0089 | 2.67E-07 |

| | | | | | |
|---|---|---|---|---|---|
| | **Cd** | 1.5E-05 | 0.5 | 0.0299 | 5.39E-06 |
| | **Cu** | 0.0005 | 0.5 | 0.0116 | 1.85E-05 |
| | **Zn** | 0.0025 | 0.3 | 0.0082 | |
| **HI** | | | | **0.56** | |
| **∑ILCR** | | | | | **2.42E-05** |

The estimated THQ ranged between 0.007- 0.5146 and the TCR values of all the metals were in the range of 1.89E-05 and 2.42E-05, which were lower < 1 indicating no carcinogenic risk. The HI value for all the metals was less than 1 (Table 3), revealing an absence of possible health hazards for the population in Benin City from these elements. The findings of the current study showed that ILCR for Pb, Cd and Cu during a lifetime consumption of contaminated fish was 2.16E-07, 5.24E-06 and 1.34E-05 respectively. USEPA established a tolerable limit of lifetime carcinogenic risk within the range $1 \times 10^{-6}$ to $1 \times 10^{-4}$ [28]. The overall carcinogenic risk in this study was higher than the USEPA acceptable level, indicating that those who eat frozen fish fillets are at risk for cancer.

## 4.0. Conclusion

Metal intake from fish consumption is a food safety concern that has a negative impact on consumer health, thus information regarding metal concentrations in frozen products is required to determine the possible harm to the population's health. Overall, the amounts of metals examined in the various fish species were typically lower than the maximum allowable levels specified by food safety guidelines but collectively can pose risk to consumers' health. Thus, constant monitoring is recommended to avoid an unexpected health hazard that can occur over a long period of consuming these frozen fish species.

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