Research Article

Heavy metal levels and changes in trimethylamine content of smoked fish and meat under different storage conditions

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Abstract
Heavy metal (As, Cd, Co, Cu, Mn, Zn and Pb) levels and trimethylamine content of smoked fish and meat were investigated under different storage conditions using Atomic Absorption Spectrophotometry and Ultraviolet visible Spectrophotometry respectively. This was done to establish the best storage method that could guarantee longer shelf-life and assess the consumption safety of the smoked fish and meat. The smoked meat used for this study was purchased at Enuwa, Ile Ife, Osun State, Nigeria while the fish was purchased at Ile-Ife main market, Ile-Ife, Osun State, Nigeria. Mean concentrations of the heavy metals followed the order: Zn > Cu > Mn > Co > Cd > As > Pb in the fish products while it followed the order: Zn > Cu > Co > Mn > Cd > As > Pb in the meat products. The mean levels of trimethylamine (TMA) in fish and meat products according to storage conditions followed the order: Sundried > Oven dried > Freeze-dried and results indicated that TMA increased with the time of storage. The health risk assessment of heavy metals in smoked fish and meat indicated that there were no immediate health risks associated with their consumption. Long-term storage of fish and meat products should also be discouraged so as to avert increased levels of TMA.

Keywords Fish · Health risk · Heavy metal · Meat · Trimethylamine

1 Introduction

Fish is widely consumed in many parts of the world because it has high protein content and minerals [29]. The relatively low saturated fat content, high protein content and fatty acids, justifies the dietary consumption of fish because they are known to support good health [2]. According to FAO [17] and Gandotra et al. [18], fish provides 20% of the animal protein intake to about 2.6 billion people globally and at least 50% of animal protein intake for over 400 million in Asia and Africa. In developing countries, it provides only 13% of animal protein intake. Fish represents a major source of animal protein intake in Nigeria due to its availability and low cost.

Although, fish production is steadily increasing, preservation of the commodity still remains a challenging problem. The susceptibility of fish to rapid spoilage has been attributed to its intrinsic characteristics and high potential of microbial contamination from a variety of sources [36]. Fish and shellfish are highly perishable, and prone to vast variations in quality due to differences in species, environmental habitats, feeding habits and action of autolysis enzymes as well as hydrolytic enzymes of some microorganisms present in fish muscle [35].

The main purpose of freezing seafood is protecting the initial sensory and chemical properties of fresh products [13]. Shelf life and the quality of frozen fish depend on the handling conditions of the fish, the frozen amount of product, packing, freeze–thaw abuse, temperature of storage,

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temperature fluctuations and sustainability [30]. Quick freeze and thaw processes are widely used at home and restaurants [21]. The use of inaccurate freezing, storage and thawing processes on foods are largely responsible for microbiological, chemical and physical deteriorations [4].

The advent of civilization allowed the domestication of animals such as chickens, sheep, pigs and cattle, and eventually their use in meat production on an industrial scale [38]. Smoking is the process of flavouring, cooking or preserving food by exposing it to the smoke from burning or smouldering plant materials most often wood. Meats and fish are the most commonly smoked foods [20].

Heavy metals are elements of high atomic mass and a relatively higher density than that of the earth (5 g/cm^3). Unlike organic pollutants that are biodegradable, heavy metals are not biodegradable but can only be transformed from one oxidation state to the other or from one organic complex form to another [31]. As human activities increase, particularly with the application of modern technologies, pollution and contamination of human food chain has become inevitable. There are many health benefits accrued to the dietary intake of fish and meats, nevertheless, they possess a complex system that allows for the bioaccumulation of heavy metals. There are health threats associated with the intake of heavy metals via the consumption of fish and meat, ranging from osteoporosis to decreased sex drive to early aging to cardiovascular diseases to decreased mental function etc. [14].

Preservation of fish and meat can be achieved by various methods, including refrigeration, freezing, salting, canning (wet salting), icing, smoking, glazing, drying, frying, etc. Trimethylamine is a reduction product of trimethylamine oxide during spoilage and ammonia is mainly formed as a product of protein breakdown. Trimethylamine is one of the volatile amines plus ammonia which can be used as an index of spoilage [8]. Therefore, the quality of fish and meat processed by the various methods cannot be the same, and hence, its subsequent effect on the fish and meat shelf life also varies. It has been observed that different processing methods have different effects on the nutritional compositions of fish and meat [2]. It is therefore important to investigate the effect of storage conditions on the levels of Trimethylamine (TMA) and heavy metals present in fish and meat.

2 Materials and methods

2.1 Sample collection and preparation

Freshly smoked meat (*Bubalus bubalis*) sample used for this study was purchased at Palace road, Enuwa, Ile Ife, Osun State, Nigeria while the Fish (*Clarias gariepinus*) was purchased at Sabo market, Oja Tuntun, Ile Ife. Three (3) samples each of the fish and meats were obtained from the markets and subjected to the various storage conditions (sun-drying, freeze-drying and oven-drying). The entire fish and meat samples were ground into fine powder using the ceramic mortar and pestle to increase the surface area of the samples and to ensure the homogenity and uniformity of the samples. Each time measurement of Trimethylamine (TMA) and determination of heavy metal were to be done, a part of the ground fish and meat samples were taken and subjected to appropriate procedures. The measurement of trimethylamine (TMA) and determination of heavy metal were carried out in triplicates.

2.2 Measurement of TMA Content in Fish and Meat

The procedure for determination of TMA concentration in meat was modified from those reported by Ward et al. [37]. A 0.50 g sample of the ground meat or fish was placed in a 20 mL tube and 5 mL of 10% trichloroacetic acid (TCA) was added and homogenized quickly. This was centrifuged at 3000 revolutions per minute (rpm) and the supernatant was decanted into 25 mL test tube. Thereafter, 1.0 mL of 10% formaldehyde solution, 10 mL anhydrous toluene, 3.0 mL 1:1 potassium carbonate solution was added and the mixture shook vigorously for 10 min. The toluene phase was transferred into a plastic tube and 1.0 g anhydrous sodium sulphate was added and mixed properly. After mixing, 1 mL liquid was transferred to a new plastic tube and 5 mL 0.02% picric acid was added, forming the yellow TMA-N picrate complex. It was then transferred into a UV–Vis cuvette and the absorbance–wavelength was read at its maximum absorption wavelength of 410 nm using a UV Shimadzu 1800 spectrophotometer in which 10% TCA served as blank.

2.3 Procedure for acid digestion

Accurately weighed 1 g of the sample was carefully placed into a 100 mL Pyrex beaker, and 5 mL of concentrated nitric acid (HNO_3) was added. The mixture was placed on a hot plate inside the fume cupboard and boiled gently for about 45 min, additional 5 mL of HNO_3 was added when the mixture was about to dry during the boiling process. After the sample has completely dissolved on boiling with concentrated HNO_3, the mixture was removed from heat and 1 mL of perchloric acid ($HClO_4$) was added. The mixture was further boiled gently for about 30 min and was removed from the hot plate and cooled. After cooling, the content of the beaker was transferred quantitatively into a 25 mL volumetric flask and made up to the mark by adding distilled water. The mixture was poured into the plastic vial and the heavy metals concentrations in the

digested mixture were analysed using Atomic Absorption Spectrophotometer (AAS) Model Alpha Star 4 (ChemTech Analytical) available at the Centre for Energy Research and Development, Obafemi Awolowo University, Ile-Ife.

2.4 Quality assurance and control

The major quality control parameter used in this study was the inclusion of two blank samples of the same concentrations. This was intended to ascertain the reliability of the AAS performance. Recovery analysis was done to ascertain the accuracy of the analytical procedure used by spiking 1 g each of different samples with 5.0 µg/g standard mixture of the available heavy metal solutions (Cd, Cu, Pb and Zn). Standard metal solutions were used to fortify the samples with the specified metal, digested and taken for AAS analysis. The percentage recovery (% R) for the heavy metals was determined as given below:

$$\%R = \left[\left(C - C'\right) / A\right] \times 100 \quad (1)$$

where C = heavy metal concentration in the spiked sample, C′ = heavy metal concentration in the unspiked sample and A = the amount of heavy metal used for spiking [23].

2.5 Data treatment

Descriptive statistics (mean and standard deviation) were used to interpret the data. Human health risk assessment such as estimated daily intake, target hazard quotient, cancer risk and relative risk were carried out to determine the health risks associated with exposure to the samples.

2.5.1 Estimated daily intake (EDI)

Estimated daily intake (EDI) of heavy metals via ingestion route was calculated using Eq. (2)

$$EDI = \frac{C \times IR \times EF \times ED}{BW \times AT} \quad (2)$$

where C = metal concentration in fish and/or meat (mg kg^{-1}); IR = ingestion rate (kg $person^{-1}$ day^{-1}), (0.08 kg $person^{-1}$ day^{-1} for meat while 0.06 kg $person^{-1}$ day^{-1} for fish); EF = exposure frequency (360 days $year^{-1}$); ED = exposure duration (70 years for adults), equivalent to the average lifetime; BW = average body weight (kg), (70 kg for adults); AT = average exposure time for noncarcinogens (365 days $year^{-1}$×ED), total THQ in this study was treated as the arithmetic sum of the individual metal THQ values [32].

Assumptions for the health risk calculations were:

a. Ingested dose is equal to the absorbed pollutant dose [33].
b. Cooking has no effect on the pollutants [7].
c. The average body weight of a Nigerian adult is assumed to be 70 kg

2.5.2 Target hazard quotient (THQ)

The THQ, the ratio of the exposure dose to the reference dose (RfD), represents the risk of noncarcinogenic effects. If it is less than 1, exposure level is less than the RfD. This indicates the daily exposure at this level is unlikely to cause adverse effects during a person lifetime, and vice versa. The dose calculations were performed using standard assumptions from the integrated USEPA risk analysis [34].

The target hazard quotient (THQ) was finally calculated using Eq. (3).

$$THQ = \frac{EDI}{mathrmRfD} \quad (3)$$

In this study, the total THQ was expressed as the arithmetic sum of the individual metal THQ values according to the method of Chien et al. [5]:

$$\text{Total THQ (TTHQ)} = \text{THQ (toxicant 1)} + \text{THQ (toxicant 2)} + \cdots \text{THQ (toxicant n)} \quad (4)$$

2.5.3 Cancer risk (CR)

The CR over a lifetime of exposure to the heavy metals was estimated using the cancer slope factor according to Eq. 5 ([24, 28]):

$$CR = \frac{EF \times ED \times IR \times CF \times C \times CSF}{BW \times AT} \times 10^{-3} \quad (5)$$

where CSF is the cancer slope factor (mg/kg/day), while the other parameters have been defined previously. The US Environmental Protection Agency set an acceptable lifetime carcinogenic risk of 10^{-5} [26].

2.5.4 Relative risk (RR)

Yu et al. [39] defined the RR of contaminants for both carcinogenic and non-carcinogenic effects, which can be helpful to recognize the most harmful contaminants. The RR equation is as follows:

$$RR = \frac{C}{RfD} \quad (6)$$

where all parameters have been previously defined [39].

3 Results and discussion

3.1 Validation of analytical procedure

The reliability of the analytical procedures used in this study was tested in terms of percentage recovery (%R) and linearity of calibration (R^2) with their values listed in Table 1. A quantitative agreement was observed with the recovery values of potentially toxic metals in the samples ranging from 84.65% in Mn to 99.35% in Zn. The %R values obtained were within the 70–110% range for recovery as stipulated by the EU Guidelines for evaluating accuracy and precision of a method [15]. For the AAS used, the standard calibration curves obtained showed high level of linearity with R^2 values ranging from 0.9885 for Cd to 0.9993 for Mn. These values were adjudged reliable to give precise and accurate metal concentrations in the samples.

The smallest quantity of an analyte that can be detected by the instrument is described as the limit of detection (LOD). It was evaluated as three times background noise produced from the analysis of blank samples. On the other hand, the limit of quantification (LOQ) is defined as the smallest concentration that is analyzed with reasonable reliability. The LOD and LOQ values obtained in this study were calculated as the concentrations of the analytes at which the signal to noise ratio (S/N) was equal to 3 and 10 respectively.

3.2 Elemental concentrations of fish and meat

The levels of As, Cd, Co, Cu, Mn, Pb and Zn were determined in the oven-dried, sun-dried and freeze-dried fish and meat samples, and they are presented in Tables 2 and 3 respectively. Variations exist in the elemental concentrations of the smoked fish and meat samples depending on the storage conditions. In the oven-dried fish samples, the levels of the potentially toxic metals ranged from 0.20 to 3.15 µg/g, while in sundried fish samples from 0.15 to 1.73 µg/g. The freeze-dried fish samples had concentrations ranging from 0.17 to 1.68 µg/g. Generally, the mean concentrations of the potentially toxic metals in the fish samples are in the following order: Zn >

Table 1 Calibration parameters of AAS and % recovery of potentially toxic metals

Metal	Current (mA)	Wavelength (nm)	Calibration curve (R^2)	Amount used for spiking (µg/g)	Amount recovered (µg/g)	Percentage recovery (% R)	LOD (µg/g)	LOQ (µg/g)
As	7	210.1	0.9965	20	19.62	98.10	0.01	0.03
Cd	8	228.6	0.9885	20	18.91	94.55	0.01	0.03
Co	7	211.5	0.9979	20	17.63	88.15	0.05	0.15
Cu	6	325.1	0.9981	20	18.10	90.53	0.03	0.09
Mn	10	280.0	0.9930	20	16.93	84.65	0.005	0.015
Pb	10	282.9	0.9978	20	19.25	96.25	0.013	0.04
Zn	8	214.2	0.9976	20	19.87	99.35	0.005	0.015

Table 2 Levels (µg/g) of potentially toxic metals in the fish

Drying method	As	Cd	Co	Cu	Mn	Pb	Zn	Total Metal
Oven-dried	0.25 ± 0.05	0.43 ± 0.03	1.47 ± 0.08	2.73 ± 0.32	2.30 ± 0.23	0.20 ± 0.02	3.15 ± 0.20	10.53 ± 0.93
Sundried	0.20 ± 0.03	0.15 ± 0.02	0.97 ± 0.06	1.73 ± 0.11	1.73 ± 0.11	0.22 ± 0.01	1.50 ± 0.05	6.50 ± 0.39
Freeze-dried	0.25 ± 0.02	0.33 ± 0.01	1.38 ± 0.03	1.13 ± 0.32	1.33 ± 0.10	0.17 ± 0.03	1.68 ± 0.13	6.27 ± 0.64
Mean ± SD	0.23 ± 0.03	0.30 ± 0.02	1.27 ± 0.05	1.86 ± 0.17	1.78 ± 0.14	0.19 ± 0.02	2.11 ± 0.13	7.74 ± 0.56

Table 3 Levels (µg/g) of potentially toxic Metals in the meat

Drying method	As	Cd	Co	Cu	Mn	Pb	Zn	Total Metal
Oven-dried	0.45 ± 0.13	0.63 ± 0.20	2.28 ± 0.23	3.05 ± 0.23	2.85 ± 0.11	0.38 ± 0.23	4.15 ± 0.40	13.79 ± 1.53
Sundried	0.38 ± 0.01	0.70 ± 0.15	2.50 ± 0.20	2.60 ± 0.10	2.50 ± 0.43	0.45 ± 0.43	3.85 ± 0.23	12.98 ± 1.55
Freeze-dried	0.40 ± 0.15	0.25 ± 0.55	2.85 ± 0.34	3.55 ± 0.30	0.05 ± 0.23	0.20 ± 0.13	4.63 ± 0.13	11.93 ± 1.83
Mean ± SD	0.41 ± 0.43	0.52 ± 0.43	2.54 ± 0.43	3.06 ± 0.43	1.80 ± 0.43	0.34 ± 0.40	4.21 ± 0.25	12.88 ± 0.40

Cu > Mn > Co > Cd > As > Pb. Cadmium (Cd) is known to be an endocrine disturbing substance and may lead to the development of prostate cancer and breast cancer in humans [25]. In fish, arsenic (As) causes neoplastic alterations and bizarre morphological alterations in the early life stages. The immediate death can be the major result for acute exposures. Chronic exposures can lead to severe health risks [3]. Despite the relatively high mean concentration observed for zinc (Zn) in the fish samples, its values still fall below the FAO maximum guideline of 30 µg/g of zinc (Zn) for safe human consumption [16]. However, increased concentration due to bioaccumulation may induce toxicity, characterized by symptoms of irritability, muscular stiffness and pain, loss of appetite, and nausea [22]. The levels of copper (Cu), zinc (Zn), lead (Pb), arsenic (As) and cadmium (Cd) in this study was higher than those reported by [11] while the levels of copper (Cu), zinc (Zn), lead (Pb), manganese (Mn) and cadmium (Cd) in this study are lower than the values reported by [12].

In the case of oven-dried meat samples, the levels of the potentially toxic metals ranged from 0.38 to 4.15 µg/g; 0.38–3.85 µg/g in the sundried meat samples; and 0.05–4.63 µg/g in the freeze-dried meat samples. Generally, the concentrations of the metals in the meat samples followed the order: Zn > Cu > Co > Mn > Cd > As > Pb. Increased levels of manganese (Mn) can cause non-carcinogenic hazards such as neurologic disorder. Also, a very high intake of copper (Cu) can cause health problems, such as liver and kidney damage [1].

Storage conditions was observed to play an important role on the levels of the potentially toxic metals in both fish and meat samples. The levels of the potentially toxic metals are of the order: Oven dried > Sun-dried > Freeze-dried. This could be as a result of higher water and other volatile contents in the samples in the order Freeze-dried > Sundried > Ovendried.

3.3 Health risk assessment of the potentially toxic metals in the fish and meat samples

The results of the health risk assessment of the investigated potentially toxic metals in the fish and meat samples are presented in Table 4. In the fish samples, the estimated daily intakes (EDIs) for As, Cd, Co, Cu, Mn, Pb and Zn were all lower than the oral reference dose (RfD). A similar trend was observed for the meat samples except for As whose EDI was higher than its RfD. The target hazard quotient (THQ) for the investigated metals in the fish samples followed the trend: As > Cd > Co > Pb > Cu > Mn > Zn while in meat samples: As > Cd > Co > Pb > Cu > Zn > M n. The total target hazard quotient (TTHQ) of the metals upon consumption of the fish and meat samples are 1.05 and 2.48, respectively. These values are higher than unity (1) which indicated that a daily exposure at this level may likely cause adverse health effects over prolonged reliance on the consumption of the fish and meat samples investigated.

The estimated factors for the cancer risk of As, Cd and Pb are also presented in Table 4. The cancer risk associated

Table 4 Health risk assessment of potentially toxic metals in the fish and meat samples

Heavy metal (µg/g)		EDI	RfD	THQ	CR	RR (%)
As	Fish	0.000194	0.0003	0.648141	2.92E−06	61.63
	Meat	0.000462		1.540509	6.93E−06	62.06
Cd	Fish	0.000254	0.001	0.25362	9.64E−07	24.12
	Meat	0.000586		0.586145	2.23E−06	23.61
Co	Fish	0.001074	0.02	0.053683	–	5.10
	Meat	0.002863		0.143155	–	5.76
Cu	Fish	0.001572	0.04	0.039311	–	3.74
	Meat	0.003449		0.086231	–	3.47
Mn	Fish	0.001505	0.14	0.010749	–	1.02
	Meat	0.002029		0.014493	–	0.58
Pb	Fish	0.000161	0.004	0.040157	1.37E−08	3.82
	Meat	0.000383		0.095812	3.26E−08	3.86
Zn	Fish	0.001784	0.3	0.005946	–	0.56
	Meat	0.004746		0.015818	–	0.63
TTHQ	Fish			1.051607		
	Meat			2.482162		

EDI estimated daily intake, *RfD* oral reference dose, *THQ* target hazard quotient, *TTHQ* total target hazard quotient, *CR* cancer risk and *RR* relative risk

with the intake of As, Cd and Pb in the fish and meat samples are lower than the set tolerable limit of 10^{-5} by USEPA [10]. This suggests that there is no carcinogenic risk for As, Cd and Pb from the consumption of the fish and meat samples. The non-carcinogenic relative risk values from the consumption of both the fish and meat samples follow the same trend as observed for the THQ. In both fish and meat samples, the highest concern of consumption is related to As.

3.4 Levels of trimethylamine content (TMA) in fish and meat samples

The consumption of meat and meat products mostly depends on colour, appearance, flavour and taste [9]. Meat natural quality such as appearance, acceptability and TMA content are mostly dependent on storage time of meat. In this study, trimethylamine content was determined with the aim of evaluating the influence of acceptability of fish and meat storage at different drying method. Cat Fish and Bufallo meat were used to test trimethylamine contents at different drying method between 1 and 5 weeks.

The calibration curve (presented in Fig. 1) was developed using serially diluted 0, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50 ppm of TMA standard solution with UV–Visible Spectrophotometer at λ = 410 nm with R^2 value of 0.9959. Hence, the values of TMA obtained in the fish and meat samples could be relied upon as being satisfactorily accurate. The levels of the TMA in the fish and meat samples are presented in Table 5. Results showed that the meat TMA value obtained in freeze-dried sample for 1 week (0.200 ± 0.001) was lower than sundried (0.210 ± 0.002) which was also lower than oven dried (0.250 ± 0.01) and the same trend was observed in the subsequent weeks. Similarly, in fish, it was discovered that in week one of storage, the concentration of TMA (0.225 ± 0.001) in freeze dried sample was lower than sun dried (0.250 ± 0.002) which was lower than oven dried (0.375 ± 0.002). A general increase in the levels

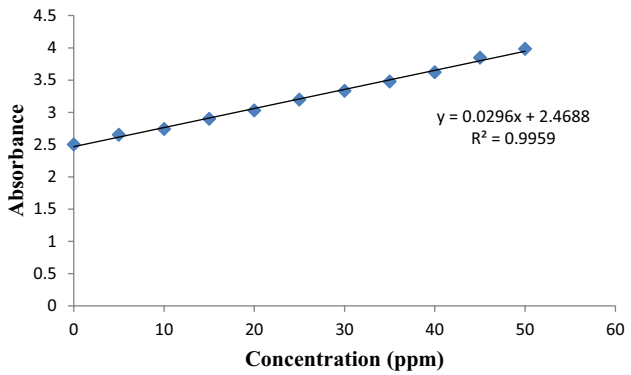

Fig. 1 Working calibration curve for trimethylamine

Table 5 Levels (μg/g) of TMA in Fish and Meat

Samples	Week	Sun dried	Oven dried	Freezing
Fish	1	0.250 ± 0.001	0.210 ± 0. 002	0.200 ± 0.001
	2	0.425 ± 0.002	0.325 ± 0.004	0.250 ± 0.002
	3	1.475 ± 0.006	1.375 ± 0.001	0.975 ± 0.003
	4	2.725 ± 0.002	1.725 ± 0.006	1.125 ± 0.001
	5	2.300 ± 0.002	1.725 ± 0.018	1.325 ± 0.002
	Mean ± SD	1.435 ± 0.003	1.072 ± 0.006	0.775 ± 0.002
Meat	1	0.375 ± 0.002	0.250 ± 0.002	0.225 ± 0.001
	2	2.100 ± 0.001	0.625 ± 0.003	0.550 ± 0.001
	3	2.500 ± 0.001	2.275 ± 0.006	0.400 ± 0.003
	4	2.850 ± 0.003	2.475 ± 0.002	0.700 ± 0.004
	5	2.800 ± 0.003	2.500 ± 0.002	1.400 ± 0.001
	Mean ± SD	2.125 ± 0.002	1.600 ± 0.002	0.655 ± 0.003

SD standard deviation

of TMA was observed within the 5 weeks of conducting the study for fish and meat samples using different preservation methods. The rate of spoilage increased with the time of storage. This is consistent with the findings of Horsfall et al. [19] and Sallam [27] who reported that the formation of biogenic toxic substances (TMA and total volatile bases) increased with the time of storage. The higher TMA levels observed for both fish and meat sundried samples during week 4 was probably due to the low intensity of solar radiation during the study period. The increasing order of mean TMA (μg/g) in fish sample was freeze dried (0.775) < oven dried (1.072) < sundried (1.435). A similar trend was observed for meat samples in which mean TMA levels (μg/g) in freeze dried sample (0.855) < oven dried (1.600) < sundried sample (2.065). The recommended level of TMA value for human consumption is 10–15 mg N/100 g [6]. The TMA values in fish and meat samples within the 5 weeks fell within the acceptable TMA range. The level of protein decomposition was higher upon the use of sun as a drying method, followed by the use of oven, and lastly the use of refrigeration. This is probably due to the relatively higher bacterial and thermal action during the period of storage.

4 Conclusion

The results of this study showed that the concentrations of the metals in the samples were within the acceptable limits set by regulatory agencies. The health risk assessment of the potentially toxic metals equally showed that there were no carcinogenic and non-carcinogenic risks associated with the consumption of the fishes and meats. However, the relative risk indicated that the highest concern of risk is related to As. Therefore, the smoked fish

and meats should be consumed moderately to prevent As induced health hazards. The levels of TMA present in the analyzed fish and meat samples were equally within acceptable limits. The relatively low TMA levels observed during the onset of storage are not reliable parameters to estimate the degree of freshness in the analyzed fish and meat samples; their upsurge in TMA levels at the latter stages of storage only reflect a degree of spoilage. Among the storage methods investigated, freeze-drying was the best method of preservation. However, regardless of the method of storage, preservation of fish and meat for long periods should be discouraged so as to prevent protein decomposition which subsequently increases TMA levels and protein denaturation. Also, the extent of formation of TMA as a function of time can be exploited in a bid to reducing the incidence of fish and meat poisoning.

Compliance with ethical standards

Conflict of interest The authors declare that there is no conflict of interest.

References

1. Agency for Toxic Substances and Disease Registry (ATSDR) (2013) Toxicological profile for copper. U.S. Department of Health and Human Services, Washington, DC
2. Al- Reza S, Karmaker S, Hasan M, Roy S, Hoque R, Rahman N (2015) Effect of traditional fish processing methods on the proximate and microbiological characteristics of Laubuka dadiburjori during storage at room temperature. J Fish Aquat Sci 10:232–243
3. Ashraf M, Maah M, Yusoff I (2011) Bioaccumulation of heavy metals in fish species collected from former tin mining catchment. Int J Environ Res 6(1):209–218
4. Bulduk S (2002) Food technology, 1st edn. Detay Publication. No: 02-06-0395-HY40, pp 147–149, ISBN 975-8326-43-0
5. Chien LC, Hung TC, Choang KY, Yeh CY, Meng PJ, Shieh MJ, Han BC (2002) Daily intake of TBT, Cu, Zn, Cd and As for fishermen in Taiwan. Sci Total Environ 285:177–185
6. Connell JJ (1995) Control of fish quality, 4th edn. Wiley, London, England. ISBN 13:9780852382264
7. Cooper CB, Doyle ME, Kipp K (1991) Risk of consumption of contaminated seafood, the Quincy Bay case study. Environ Health Perspect 90:133–140
8. da Silva (2002) Lead and human health: an update. American Council on Science and Health. http://www.acsh.org/wpcontent/uploads/2012/04/20040402_LeadHealth. Accessed 17 Dec 2019
9. Davoli R, Braglia S (2007) Molecular approaches in pig breeding to improve meat quality. Brief Funct Genom Proteom 6(4):313–321
10. Durodola SS, Ayinuola O, Ore OT, Makinde OW (2019) Assessment of human health risk of potential toxic metals in herbal concoction teas commonly consumed in Nigeria. World J Appl Chem 4(3):41–47. https://doi.org/10.11648/j.wjac.20190403.12
11. Eboh L, Mepba H, Ekpo M (2006) Heavy metal contaminants and processing effects on the composition, storage stability and fatty acid profiles of five common commercially available fish species in Oron Local Government, Nigeria. Food Chem 97(3):490–497. https://doi.org/10.1016/j.foodchem.2005.05.041
12. Edosomwan EU, Ainerua MO, Izevbizua DO (2016) Heavy metals levels in dried fish consumed in Benin and Warri Metropolis; levels and health risk assessment. Afr Sci 17(2):93–96
13. Ekinci R, Yapar A (2004) Alabalıkların Donma ve Çözünme Süreleri Üzerine Dondurma Sıcaklığı ve Hava Sirkülâsyonunun Etkileri. FÜ Fen Müh Bil Derg 16:61–68
14. Ellen G, Loon JW, Tolsma K (1990) Heavy metals in vegetables grown in the Netherlands and in domestic and imported fruits. Z Lebensm Unters Forsch 190(1):34–39. https://doi.org/10.1007/bf01188261
15. EU 2006. Directive 2002/32/EC of the European Parliament and the council. http://eur-lex.europa.eu/LexUriServ/site/en/consleg/2002/L/02002L0032-20060224-en.pdf. Accessed 18 Dec 2019
16. FAO (2014) Sustainable fisheries and aquaculture for food security and nutrition (p. 119). Food and Agriculture Organization of the United Nations. Retrieved from http://www.fao.org/documents/card/en/c/63d4de9f-50df-49b1-b6d2-b7d0368744ea. Accessed 18 Dec 2019
17. FAO (2008) Fisheries and aquaculture report. No. 889. Food and Agriculture Organization of the United Nations (FAO), Cairo, p 61
18. Gandotra R, Koul M, Gupta S, Sharma S (2012) Change in proximate composition and microbial count by low Temperaturepreservation in fish muscle of Labeo *Rohita* (Ham-Buch). IOSR J Pharm Biol Sci 2:13–17
19. Horsfall M Jr, Kinigoma BS, Spiff AI (2006) Evaluation of the levels of total volatile bases and trimethylamine formed in fish stored at low temperature. Bull Chem Soc Ethiop 20(1):155–159
20. McGee H (2004) On food and cooking (revised edition). Scribner Publisher, New York, pp 427–429
21. Mol S, Özden Ö, Erkan N, Baygar T (2004) Determination of quality parameters of different solubility terms for imports mackerel. Turk J Vet Anim Sci 28:1071–1077
22. NAS-NRC (1982) National, drinking water and health. Academy of Sciences-National Research Council National Academic Press, Washington D.C
23. Oyewole FG, Adebiyi FM (2017) Total and speciation analyses of heavy metals in the sand fraction of Nigerian oil sands for human and ecological risk assessment. Hum Ecol Risk Assess Int J 23(8):2046–2068
24. Peng Q, Nunes LM, Greenfield BK, Dang F, Zhong H (2016) Are Chinese consumers at risk due to exposure to metals in crayfish? A bioaccessibility-adjusted probabilistic risk assessment. Environ Int 88:261–268
25. Saha N, Zaman MR (2012) Evaluation of possible health risks of heavy metals by consumption of foodstuffs available in the central market of Rajshahi City, Bangladesh. Environ Monit Assess 185(5):3867–3878. https://doi.org/10.1007/s10661-012-2835-2
26. Saha N, Mollah M, Alam M, Rahman MS (2016) Seasonal investigation of heavy metals in marine fishes captured from the bay of Bengal and the implications for human health risk assessment. Food Control 70:110–118
27. Sallam KI (2007) Chemical, sensory and shelf life evaluation of sliced salmon treated with salts of organic acids. Food Chem 101:592–600
28. Shaheen N, Irfan NM, Khan IN, Islam S, Islam MS, Ahmed MK (2016) Presence of heavy metals in fruits and vegetables: health risk implications in Bangladesh. Chemosphere 152:431–438
29. Tidwell JH, Allan GL (2001) Fish as food: aquaculture's contribution. Ecological and economic impacts and contributions of fish farming and capture fisheries. Eur Mol Biol Org 21:958–963

30. Turan H, Kaya Y, Erdem ME, Sönmez G, Kodalak N, Erkoyuncu İ (2006) Effect of different def-rost conditions on quality of freeze salmons. Univ Istanbul J Fish 20:21–32
31. Turkdogan MK, Kilicel F, Kara K, Tuncer I, Uygan I (2003) Heavy metals in soils, vegetables and fruits in the endemic upper gastrointestinal cancer region of Turkey. Environ Toxicol Pharmacol 13:175–179
32. USEPA (United States Environmental Protection Agency) (2015**)** Regional screening level (RSL) summary table, November 2015
33. USEPA (1989) Risk assessment guidance for superfund, human health evaluation manual part A, interim final, vol I. United States Environmental Protection Agency, Washington DC
34. USEPA (2000) Agency. U.S.E.P, Supplementary guidance for conducting health risk assessment of chemical mixtures
35. Venugopal V (2002) Biosensors in fish production and quality control. Biosens Bioelectron 17:147–157
36. Venugopal V, Doke SN, Thomas P (1997) Thermostable water dispersion of shark meat and its application to prepare protein powder. J Aquat Food Prod Technol 6:53–67
37. Ward AK, Classen HL, Buchanan FC (2009) Fishy-egg tainting is recessively inherited when brown-shelled layers are fed canola meal. Poult Sci 88(4):714–721. https://doi.org/10.3382/ps.2008-00430
38. Womack RM (2010) The anthropology of health and healing. Rowman and littlefield. P243. ISBN 0759110441
39. Yu Y, Wang X, Yang D, Lei B, Zhang X, Zhang X (2014) Evaluation of human health risks posed by carcinogenic and non-carcinogenic multiple contaminants associated with consumption of fish from Taihu Lake. Food and Chemical Toxicology, China 69:86–93