Research Article

Estimation of Heavy Metal Contamination in Canned Fish Products Sold in Ernakulam District of Kerala State

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Abstract

Background : Fish is an important source of food which is rich in protein and omega-3 fatty acids. Canned fish is preferred by many due to its convenient nature of use, less cooking time, affordable price etc. Canned fish get contaminated with naturally occurring metals during the process of storage, manufacture and transportation. Among the heavy metals lead, arsenic and mercury are classified as carcinogenic agents to humans by the International Agency for Research on Cancer (IARC) & are limited for human consumption by various International Organizations.

Methods: This is a cross-sectional study of the estimation of lead levels in selected canned fish products sold in the Ernakulam district of Kerala State. Quantitative estimation of selected heavy metals was done in canned fish samples using Inductively Coupled Plasma-Mass Spectrometry(ICP-MS) and the values were compared with the permissible limits for human consumption.

Results: Out of 13 canned fish samples screened for lead, arsenic and mercury. Arsenic was detected in all samples (100%). Lead and mercury were detected in 12 samples (92.3%). The mean level of lead obtained was within a range of 0.305 ± 0.267 mg/kg and in two samples (15.4%) lead levels crossed the permissible limit for human consumption. Arsenic and Mercury detected in the samples analysed did not cross the permissible limits for human consumption.

Conclusions: From this study, we conclude that even though heavy metals such as lead, arsenic and mercury were detected in more than 90% of samples, two samples were contaminated with lead levels more than the permissible limit for human consumption by FSSAI. Frequent monitoring should be conducted for food items crossing the permissible limit set by various international agencies.

Keywords: Heavy Metal; Canned Fish Products; Kerala

Introduction

Fish is an important source of food rich in protein and omega-3 fatty acids all over the world. Both Indian fisheries and aquaculture play a role in providing nutritional security and provide a major mainstay of income generation among the public. In the field of agricultural exports India has become a superpower with a turnover of Rs 33,000 crores and with more than ten lakh tonnes in quantity. India holds 2nd position in aquaculture and 3rd in fisheries all around the world.[1]

Kerala, a southern state of India, stands second in the overall fish folk population across the country.[2] There is at least a minimum of a single meal daily with fish in most of the houses in Kerala and people living in rural areas consume higher quantities of fish when compared with that of urban areas.[3,4] People of Kerala consume fish four times the national average. Kerala stands second in relation to the shares of total marine exports of India.[5,6] Kerala is considered to be a leading state in fish consumption and the average per capita consumption of fish is estimated to be 30 kg.[7]

In recent times, an upward and forward trend is seen in fish consumption due to the wide publicity and increased awareness among the public regarding the health benefits of the consumption of fish.[8] According to dietary guidelines of the American Heart Association (AHA), a minimum of two servings of fish should be included in the diet per week. To reduce adverse effects, it is recommended to consume different fish species.[9] Omega-3 PUFA improves cognitive development in children[10] and slows cognitive decline in the elderly.[11]

Canned food is mainly eaten by consumers who work or stay at home as it is simple to cook and less time-consuming. Due to the convenient nature of its use and also affordable prices, canned fish is a better alternative for working-class families around the globe. Fish gets contaminated with toxins during the stage of growth of fish, storage of fish and transportation. Handling during the production process and canning also cause contamination.[12] For the manufacture of cans, solders are used, which also acts as a source of contamination with lead during the process of canning.[13] Heavy metals have relatively long biological half-lives and can accumulate in living organisms over a period of time. Heavy metals are mutagenic, carcinogenic and cytotoxic in nature, and urban populations who are dependent on the vegetables and crops are grown in the peri-urban areas are at risk of getting exposed to heavy metals.[14]

Heavy metals are metallic elements with a high atomic mass and density measuring five times more than that of water. There are more than 20 heavy metals, among which, the leading cause of concern for humans are lead, arsenic and mercury.[15] Heavy metal is a term that does not have a precise definition; however, toxicologists use the term "heavy metals" to cover all metallic poisons regardless of atomic number.[16] Most heavy metals are not essential in the daily diet, even in traces. Essential elements like copper, zinc, iron, manganese, etc. play a key role in certain metabolic pathways, as well as are essential in the activation of certain enzymes.[17] On the other hand, metals like lead, arsenic, cadmium, antimony and mercury are non-essential metals and can be toxic even in trace amounts. Even the essential metals can be toxic in nature if they exceed the recommended level.[18]

The analytical methods commonly used for the determination of heavy metals include Inductively Coupled Plasma-Atomic Emission Spectrophotometry, Inductively Coupled Plasma Mass Spectrometry, Flame Atomic Absorption Spectroscopy, Graphite Furnace Atomic Absorption Spectroscopy and Microwave Induced Plasma Atomic Absorption Spectrometry. Inductively Coupled Plasma Mass Spectrometry is an advanced type of mass spectrometry and is a technique which is capable of detecting metals and several non-metals at concentrations as low as parts per billion on a non-interfered low background. Spectroscopy has applications in analytical toxicology, involving the interaction between matter and light. Spectroscopists carry out processes for determining the concentration and content of the molecular species which are present in the chemical system.[19]

The Food Safety and Standards Authority of India or FSSAI was established in the year 2006. FSSAI was set up to develop food safety in the country based on science-based standards for food and its related articles. It helps in regulating the manufacture and also storage, as well as in distribution and support sale, along with ensuring the availability of safe and wholesome food which is used for human consumption. As per Food Safety and Standards Contaminants, Toxins and Residues Regulations, 2011, the accepted levels of lead and arsenic in fish are 0.5mg/kg and 76mg/kg respectively. And the level of mercury in fish is 1mg/kg, while that of methyl mercury in any food items is 0.5mg/kg.[20]

As per global market reports, the canned seafood business size is expected to reach 42 billion US Dollars by the year 2028. There are multiple factors contributing to this increase in Compound Annual Growth Rate. Changing lifestyles combined with an increase in the affordability of canned seafood, and the lockdown following the COVID-19 pandemic led to a temporary closure of numerous manufacturing units and companies. Also due to travel restrictions enforced by various Governments, there was a major disruption in the worldwide supply chain of goods and services including canned seafood.[21]

Methods

The present study was undertaken in the Department of Forensic Medicine and Toxicology, Amrita Institute of Medical Sciences and Research Centre, Kerala state. This is a cross-sectional study of commonly consumed canned fish sold in the Ernakulam district of Kerala. The concentrations of selected heavy metals (lead, arsenic and mercury) in the canned fish samples were estimated with the help of an Inductively Coupled Plasma-Mass Spectrophotometer (ICP-MS). The dissertation review committee of this institute had given scientific, ethical and financial clearance to conduct the study.

Selection and Description of Participants: This study was a pilot study and the sample size after statistical analysis with a 20% allowable error and 95% confidence interval, was determined to be 13. Canned fish samples were commonly used and processed in the State and sold at different supermarkets around the District of Ernakulam. The selected canned fish species included tuna species including Yellowfin tuna (Thunnus albacares) and longfin tuna (Thunnus alalunga), Indian oil sardine (Sardinella longiceps) and Indian mackerel (Rastrelliger kanagurta). The exclusion criteria included all other canned fish.

Technical Information

The primary objective of the study was to estimate the extent of selected heavy metal contamination (lead, arsenic and mercury) of canned fish such as Canned Tuna, Canned Sardine, and Canned Mackerel. The secondary objectives were quantitative estimation of selected heavy metals (lead, arsenic, and mercury) in canned fish products sold in Ernakulam District of Kerala State, India and to check whether the concentration of selected heavy metals lies within permissible limits for human consumption.

Selected metals were analysed using ICP-MS which is a relatively new but powerful technique for the determination of metals in an aqueous solution. It is emission spectroscopy that uses the inductively coupled plasma to produce excited atoms and ions that emit electromagnetic radiation at wavelengths characteristic of a particular element and their detection by mass spectrometry. Standard calibration curves were made for the metals by using several solutions of known concentrations (20–100 ppb). All readings were taken in triplicate. The glassware for the sample digestion was cleaned by immersing them in 5% v/v nitric acid for 24 hours and then rinsing them with Millipore water for a minimum of six times. The glass wares were then dried in a hot air oven at 110°C. Fish tissue for the metal estimation was placed on a small piece of clean white paper and weighed using an electronic balance. It was then dried in an incubator at 65°C for 4 hours to constant weight. Later the paper with dry fish tissue was accurately weighed, and the fish tissue was scraped with a small knife, placed into a conical flask and sealed. We reweighed the paper to obtain the dry weight of the fish tissue. One fish sample with more than or around 1 mg dry weight was used for metals measurement.

For Open digestion, accurately weighed dried fish tissue was placed into the pre-cleaned nitric acid-washed conical flask. 6 ml of concentrated Nitric acid ER was added. The conical flask with the funnel on the rim was placed in the sand bath at 60°C. Profuse fumes evolved. After 48 hours of digestion, 8 ml Perchloric acid ER was added. Perchloric acid fumes were allowed to evolve for about 2-4 hours and then the digestion was stopped. For sample dilution, the cooled digested sample was filtered using Whatman No 1 filter paper and the solution was made up to 50 ml by adding Millipore water drop by drop. And the final sample solution was mixed after capping by hand-shaking.

Statistical Analysis

Statistical analysis was done by using the software - IBM SPSS version 20.0. Categorical variables were expressed using frequency and percentage. Numerical variables were presented using mean and standard deviation. Descriptive statistical methods were used to represent the heavy metal levels in canned fish.

Results

A total of 13 canned fish samples, each weighing 180 gm were taken, a few samples of which were canned in brine, tomato sauce, and sunflower oil which were used and processed in the State and sold at different supermarkets around the District of Ernakulam. Lead was detected in 12 samples (92.3%), arsenic was detected in all 13 samples (100%) and mercury was detected in 12 samples (92.3%) **(Table 1)**. The mean concentration of lead, arsenic and mercury in canned fish species were 0.3054 ± 0.2662mg/kg, 0.3639 ± 0.2446mg/kg and 0.0210 ± 0.0382mg/kg respectively (**Table 2)**.As per FSSAI, the permissible level of lead in canned fish is 0.5mg/kg. Two samples (15.4%) of lead levels obtained in the current study were above the permissible level and eleven samples (84.6%) were below the permissible levels of the Food Safety and Standards Authority of India (**Chart 1)**.

Discussion

In the present study, lead was detected in 12 samples (92.3%) and was below the detectable level in the remaining 1 sample (7.7%). Lead levels obtained were in the range of 0.020-0.905 mg/kg, with a mean level of 0.305 ± 0.266 mg/kg. As per FSSAI, the permissible level of lead in canned fish is 0.5 mg/kg. Lead levels obtained from 2 samples are above the permissible limits, while 11 samples are within the permissible limits. As per Codex Alimentarius, the permissible level of lead in fish is 1.5 mg/kg. No samples were above permissible levels. As per Commission of European Communities, the permissible level of lead is 0.1 mg/kg. Four samples were below the permissible limits and 9 samples were above the permissible limits.

Arsenic in the present study was above the detectable level in 13 samples, and the mean level of arsenic is 0.363 ± 0.245. As per FSSAI, the permissible level of arsenic in fish and crustaceans is 76 mg/kg. As per Codex Alimentarius, the permissible level of arsenic in marine fish, shellfish and crustaceans is 50 mg/kg. As per the Commission of European Communities, the permissible level of arsenic is 0.1mg/kg.

Mercury was detected in 12 samples (92.3%), while it was below the detectable level in the remaining 1 sample (7.7%). Mercury was not detected in a sample of canned white meat tuna, and the mean level obtained was 0.021 ± 0.038 mg/kg. As per FSSAI, the permissible level of mercury in predatory fish is 1.0 mg/kg, and mercury levels obtained from all the samples are within the permissible level. As per Codex Alimentarius and Commission of European Communities, the permissible level of mercury is 0.1 mg/kg. The mercury level obtained in the present study was just below the permissible level of Codex Alimentarius and Commission of European Communities.

Mansouri et al [22] in Iran conducted a study on canned tuna and the lead levels obtained in that study were in the range of 0.042 - 0.112mg/kg. Arsenic was only in the below detectable limit, mercury levels obtained from canned tuna were 0.02-0.04 mg/kg. The method used for analysis in this study was Inductively Coupled Plasma Mass Spectrometry (ICP-MS). Morshdy et al [23] in Egypt conducted a study on canned tuna and the lead levels obtained were in the range of 1.17 ± 0.28mg/kg. Arsenic level obtained was in the range of 4.23-7.79 mg/kg and mercury levels were in the range of 0.03 ± 0.01mg/kg. The analytical method used in this study was High-Pressure Liquid Chromatography (HPLC). Ormaza et al [24] in Ecuador, conducted a study on canned tuna and the lead levels obtained from this study were in the range of 0.23 ± 0.14mg/kg and arsenic levels obtained were in the range of 0.14-0.30 mg/kg. Mercury levels obtained were in the range of 0.043 ± 0.004mg/kg. The analytical method used in this study was Atomic Absorption Spectroscopy.

Mehouel et al [25] in Algeria, conducted a study on canned sardine and the lead levels obtained were in the range of 2.13 ± 0.69 mg/kg and the mercury levels obtained from canned sardine were in the range of 0.052 – 0.076mg/kg. The analytical method used in this study was Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-AES). González et al [26] in Spain conducted a study on canned mackerel and the level of lead obtained was 0.045 – 0.124 mg/kg and the arsenic level obtained from canned white meat tuna fish in brine was 3.592-5.047mg/kg, mercury level obtained from canned white meat tuna fish in sunflower oil was 0.152 – 0.278mg/kg. The study was conducted using Inductively Coupled Plasma Mass Spectrometry (ICPMS). Rodriguez et al [27] conducted a study in Mexico on canned tuna using Graphite Furnace Atomic Absorption Spectroscopy (GFAAS). And the lead level obtained was in the range of 1.040-1.116mg/kg and the mercury levels obtained from canned tuna were 0.545 – 0.724mg/kg and the analytical method used was Cold Vapor Atomic Absorption Spectrometry (CVAAS).

Núñez et al [28] in Spain conducted a study on canned yellowfin tuna using Inductively Coupled Plasma Mass Spectrometry (ICPMS), lead and arsenic levels were in the range of 0.006 ± 0.017mg/kg, 3.78 ± 2.24mg/kg respectively. Popov et al [29] in Serbia conducted a study on canned offal fish and the lead and mercury levels obtained were in the range of 1.13 ± 0.49mg/kg and 0.068 -0.082mg/kg using ICP-MS. Arsenic levels obtained using mass spectrometry from canned sardine were 1.70-3.18mg/kg.Makedonski et al [30] in Bulgaria conducted a study on canned fish gills and lead levels obtained were in the range of 0.06 - 0.08mg/kg. and arsenic levels obtained figures of 0.73 – 1.26mg/kg Analytical method used in this study was Atomic Absorption Spectroscopy. Mercury values ranged from 0.05 – 0.16mg/kg. The analytical method used for this study was a direct Mercury Analyzer.

Shutyuk et al [31] at the National University of Food Technologies, Ukraine, conducted a study and obtained lead levels from canned white meat tuna and these were in the range of 0.086-0.928mg/kg. The method used in this analytical study was Atomic Absorption Spectrometry. Andayesh et al [32] conducted a study in Iran and obtained arsenic and mercury levels from canned white meat tuna fish in oil in the range of 0.64 ± 0.26mg/kg, 0.11 ± 0.09mg/kg. The analytical method used in this study was Atomic Absorption Spectroscopy. Okyere et al [33] obtained mercury levels from canned fish in the range of 0.02 ± 0.02 mg/kg. The analytical method used in this study was a flame Atomic Absorption Spectrophotometer.

As far as India is concerned, in a study conducted by Giri and Singh [34] lead and arsenic levels obtained from canned shrimp were in the range of 0.03–0.41 mg/kg and 0.027–1.52 mg/kg respectively. Inductively Coupled Plasma Mass Spectrometry (ICPMS) was used for analysis for this study.

Strength and Limitations

The quantitative estimation of heavy metals including lead, arsenic and mercury in canned fish samples was performed using Inductively Coupled Plasma Mass Spectrometry (ICP-MS), which is a sophisticated method with a high degree of precision and specificity.

Different batches and manufacturing units of the same brand were not considered, as the cost of sample analysis is expensive. Hence inter-batch comparison was not possible. The source of contamination of canned fish products could not be determined as raw fish used for processing was not analysed for heavy metal contamination.

Conclusions

Two samples (sample containing light meat tuna in water and white meat tuna in virgin oil) obtained in this study relating to lead, have crossed the permissible level of FSSAI limit. Arsenic was present in all the samples analysed, but it did not cross the permissible limit set by various agencies. The permissible limit of FSSAI is much higher in relation with Codex Alimentarius and Commission of European Communities. Mercury level obtained was also below permissible levels set forth by FSSAI, Codex Alimentarius and Commission of European Communities. Heavy metals have relatively long biological half-lives and can accumulate in living organisms over a period of time, hence regular consumption of products contaminated by heavy metals can cause health problems, especially if they are above permissible levels. Further study including different batches and manufacturing units of each brand with a higher sample size has to be conducted to confirm the findings obtained. Raw fish which is used for processing also needs to be analysed for the presence of heavy metals. Long-term retrospective and prospective analysis of health issues in people routinely using canned fish products needs to be done wherever the levels are above permissible limits.

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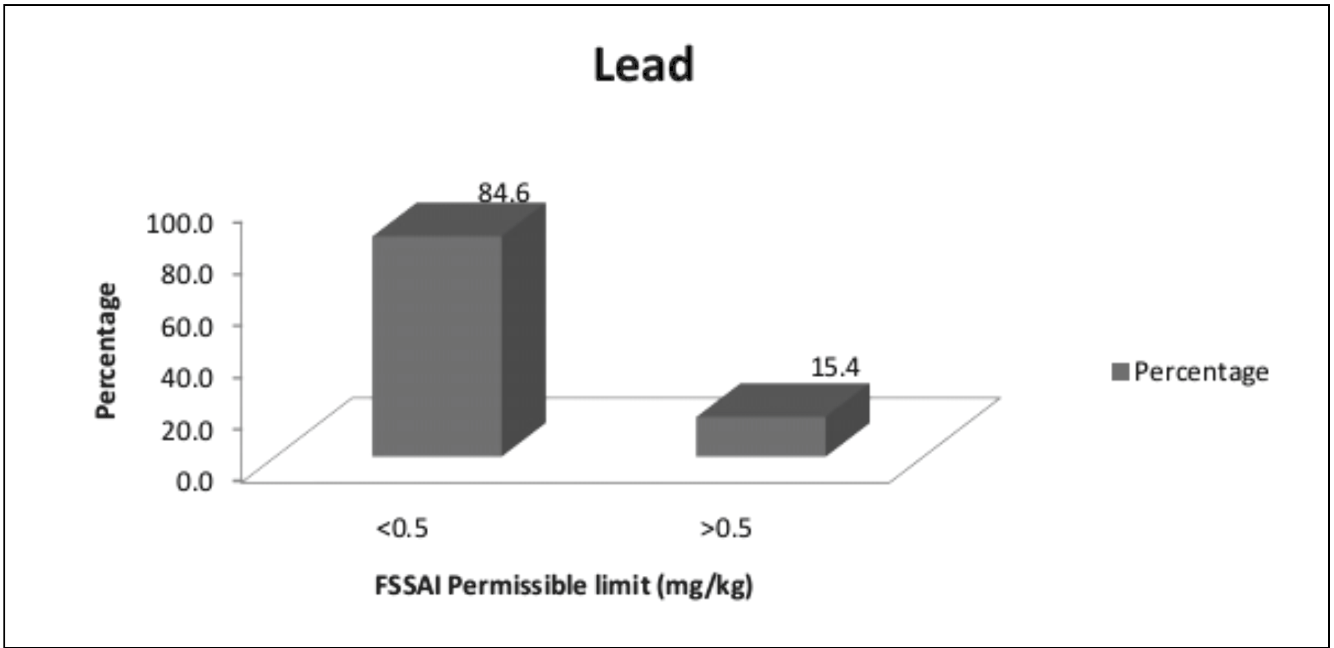
Table 1 : Distribution of Samples in Different Groups of Canned Fish Tested for Selected Metals

Metals	Canned Fish						Total
	Canned Tuna		Canned Sardine		Canned Mackerel		
	Samples (n = 8)	Percentage	Samples (n = 3)	Percentage	Samples (n = 2)	Percentage	
Lead	7	87.5	3	100	2	100	12
Arsenic	8	100	3	100	2	100	13
Mercury	7	87.5	3	100	2	100	12

Table 2 : Mean Level of Heavy Metals in the Samples

Sl No.	Metal	Mean Level (mg/kg)	Standard Deviation (mg/kg)
1	Lead	0.3054	0.2662
2	Arsenic	0.3639	0.2446
3	Mercury	0.0210	0.0382

Chart 1 : Distribution of Permissible Limit of Lead as per FSSAI



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