

EVALUATING NUTRITIONAL CHANGES AND HEAVY METAL CONCENTRATIONS IN SHRIMPS: THE EFFECTS OF DIFFERENT CULINARY METHODS

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Abstract

This study analyzed moisture, protein, carbohydrate, lipid, and metal concentrations (Fe, Zn, Cu, Li, Cr, Pb) in *Penaeus monodon* and *Penaeus indicus* harvested in Pakistan, comparing raw, pan-fried, and sushi preparations. Frying reduced moisture but increased protein content, with fried shrimp showing higher protein levels by adding gram flour coating in traditional methods than raw and sushi. The highest Fe and Li concentrations were found in fried *P. monodon*, while Cu was most concentrated in its raw form. In *P. indicus*, Cu was either very low or below detection limits, and Li was undetectable in most preparations except sushi. Cr and Pb were not detected in any preparation method of both *P. monodon* and *P. indicus* throughout the analysis, indicating their concentrations were below the detectable limits of the applied analytical technique. Overall, all heavy metal concentrations were well below the WHO permissible limits, indicating the shrimp samples are safe for human consumption. The study also highlighted higher fat and energy content in fried shrimp, while sushi had lower protein and energy levels. These findings provide insights into the nutritional impacts of different cooking methods and update knowledge on seafood quality and metal concentrations during culinary processes.

Keywords: Proximate composition, culinary methods, *Penaeus monodon*, *Penaeus indicus*, health index, Karachi

Introduction

Global seafood demand is an ongoing process. Among the various seafood items, shellfish like shrimp have always been popular among consumers and have consistently held high market value. The nutritional importance of different types of

shrimp can be determined by analyzing their proximate biochemical structure, which includes protein, fat, moisture, and ash content. This information is crucial for both industrial and marketable processing of shrimp (Zafar *et al.*, 2019a). The shrimp substance is likewise a respectable reason for mineral deposits such as calcium (Oksuz *et al.*, 2009). Lots of studies have been done on shrimp and fishery products, therefore considering the various health risk and nutritional benefits associated with shrimp consumption (Ndomou *et al.*, 2022). In addition to providing important nutrients like vitamins, essential amino acids, ω 3 polyunsaturated fatty acids, trace elements, and other essential elements, fish and seafood also accumulate significant levels of heavy metals, which could be harmful to human health (Catsiki and Stroglyoudi, 1999; Leblanc *et al.*, 2000; Llobet *et al.*, 2003). Investigations into the impact of cooking methods on heavy metal levels in fish through seafood consumption have been undertaken. Despite the common consumption of cooked fish and shellfish, existing studies primarily examine trace elements using data from uncooked/raw products, as highlighted by Domingo (2010). Analyzing heavy metals in raw samples poses a challenge in determining their intake. Previous studies explore concentration of heavy metals in various fish, mollusks, and crustacea species (Bat and Arici, 2018). However, there is a lack of research on how cooking methods affect the content of trace metals in small marine fish and shellfish. In our current investigation, we aimed to evaluate the impact of diverse culinary practices on the levels of Cr, Cu, Fe, Ni, and Zn in samples. We compared heavy metals concentration in cooked samples to those in raw ones and estimate the dietary intake of essential metals, discussing potential risks associated with Cd, Hg and Pb from fish and sea food consumption (Kalogeropoulos *et al.*, 2012).

Due to the swift mercantile enhancement and trade progress, heavy metals that are lethal, stubborn and bio-accumulative have become more severe and now it is a major environmental problem worldwide (Balkas *et al.*, 1982; Rajeshkumar and Li, 2018; Bat *et al.*, 2018). Fish, shrimps, crabs, and other aquatic organisms consume toxic metals in the water column, depositing them in remains and expending it (Rahman and Singh, 2020). The presence or accumulation of non-essential heavy metals above a secure level can lead to haphazard interactions with essential elements and proteins, the following substances have the potential to disrupt the metabolic rate of cells (Rahman and Singh, 2020).

Studies on substantial metals in sea food have been a main ecological concentration mainly in previous years (Kalvinš *et al.*, 2000; Grosheva *et al.*, 2000; Bat and Arici, 2018). Several edible shrimp species are sold abroad from distinct countries to get foreign exchange (Meshram *et al.*, 2014). The enormous tiger shrimp, *Penaeus monodon* Fabricius, 1798, is a highly prized seafood species due to its marketability. A short time ago, rapid industrialization and urbanization may have contributed to major marine pollution, which in turn may have contributed to the buildup of heavy metals in the tissues of tiger shrimp (Meshram *et al.*, 2014). Human health may be adversely affected by eating these shrimps thus it is necessary to determine the levels of heavy metals in the shrimp. Metals and metalloids ions are classified into two groups that are essential or non- essential elements. The non-essential fundamentals

consist of such as mercury (Hg), cadmium (Cd), lead (Pb), nickel (Ni), manganese (Mn), chromium (Cr), arsenic (As), as that might not any resolute role in the body but, essential elements such as copper (Cu), zinc (Zn), iron (Fe), and selenium (Se), which are part of numerous biochemical manner in the human body and have level of poisonousness and determination capability processing probable for bio amplification, bioaccumulation and assimilation addicted to the food series after reaching a certain limit in the oceanic surroundings (Zhang and Shu, 2010; Ahmed *et al.*, 2015; Kibria *et al.*, 2016; Odobašić *et al.*, 2019). Heavy metals pollution harms surroundings protection and effect destruction to community fitness and the state of the worldwide (CDC, 2022; Chris *et al.*, 2023). In additional term, the manifestation of harmful biochemical contaminants such as substantial metals in aquatic environs have mutilation biodiversity ecology and affected concern everywhere the biosphere (Stanković *et al.*, 2012). Pb depletion effects kidney or liver failure cancer and elevated body fluid stress and Cd special effects high blood pressure, skeletal faintness and cancer-causing consequences. (Al-Busaidi *et al.*, 2011). Substantial metals are specific metals and metallic complex that can affect human fitness (Martin and Griswold, 2009). Non-essential substantial metals be able to distract biological method organizing and vital danger for the fitness of human beings (Mudgal *et al.*, 2010). Shrimp is one of the extreme prevalent constituents in each state as it is an exceptional basis of protein, nutrient, vitamins, antioxidant, animated amino acid, and monounsaturated full of fat, and truncated in calories (Dayal *et al.*, 2014, Bernard, 2016; Đorđević and Buchtová, 2020).

This study was aimed at ascertaining qualitative analysis and energy transformation through different culinary methods. Besides determining metal contamination during pre and post cooking.

Materials and methods

Sample collection

The most usually consumed and exported shrimp species in Karachi were take-in in this study i.e. *P. monodon* and *P. indicus* which are commonly known as “giant tiger prawn” or “Asian tiger shrimp” and “Indian white shrimp” respectively. These shrimp species have high export demand in the local and international sea food industry. Each species is highly preferred due to its large size, delicious taste and versatile culinary application. The size of the samples must be carefully sorted out during collection, it means the size of shrimps which are commonly used for selling in local markets were taken. The five-kilogram samples of shrimps were purchased from the Karachi fish harbor and local fish markets of Karachi, Pakistan. After sampling shrimp samples were divided into three groups to obtain the means of their length and weight. The common, scientific names and average size and weight are presented in Table 1.

Table 1. Common, scientific names, and biometric measurements of shrimp studies.

Common name	Scientific name	Mean $\pm$ SD (min.-max.) length (cm)	Mean $\pm$ SD whole weight (g)	Mean $\pm$ SD muscle weight (g)
Indian tiger shrimps	<i>Penaeus monodon</i>	12.8 $\pm$ 0.37 (12-13)	16.6 $\pm$ 2.56	8.38 $\pm$ 1.45
Indian white shrimps	<i>Penaeus indicus</i>	13.4 $\pm$ 1.03 (13-14)	17.8 $\pm$ 2.35	7.22 $\pm$ 1.38

The shrimps were all taken off the coast of Karachi and landed at the fish harbor several hours after the catch. On board they were kept covered with ice and were subsequently shipped in a refrigerator truck to the central market. The species of shrimps were bought randomly during August 2023. About five kilograms of whole shrimp were attained, kept in icebox and immediately brought to the laboratory without delay, where the identification and measurement of sample were taken. For making of culinary procedure, sunflower cooking oil, spices, plain boiled long grained basmati rice (for coating of sushi shrimp fillet) and Gram flour (for coating of fried shrimp samples) were purchased from local supermarket.

Sample preparation and cooking

The samples of *P. monodon* and *P. indicus* were washed with distilled water to remove any dirt or scum. After that measurement of the length and weight of all individual samples were taken with the help of electrical balance Acura tech Taiwan plus and measuring scale. Then according to their length and weight all samples sort out into three groups. The samples were cleaned as per usual cooking practice (Beheading, removing the notochord and exoskeleton.). After washing and cleaning the only edible muscles parts of both shrimp species are divided into three batches, the first one is uncooked (raw sample) and the other two groups were used for culinary practices. Two different culinary (frying and sushi) methods were applied on shrimp's species which are traditionally and internationally used on domestic and commercial level worldwide. The frying of the marinated species of shrimp was performed in a domestic non-stick pan (2 liter capacity) at medium flame approximately 15 minutes then kept at kitchen towel to remove excesses oil. For sushi preparation, the shrimp was first cooked with hot water (2 minutes) in high flame and then transferred into iced chilled water for 3 minutes and after that samples were coated with cooked boiled rice (Table 2).

Table 2. List of ingredients and traditional methods of preparation of shrimp species commonly consumed in Karachi, Pakistan.

Species	Process	Sample weight (g)	Sunflower Oil used (ml)	Ingredients	Methods of preparation	Cooking time (min)
<i>Penaeus monodon</i>	Fried	130	200	*	***	5-7
	Sushi	130	--	**	*****	10-15
<i>Penaeus indicus</i>	Fried	100	200	*	***	5-7
	Sushi	100	--	**	*****	10-15

* Spices used for fried samples primarily included salt, red chili powder, turmeric powder, fresh garlic paste, and gram flour.

** Spices used for sushi samples primarily included soy sauce, vinegar, and long-grained basmati rice.

*** Shrimps were washed with a salt and vinegar solution and then rinsed with distilled water. They were coated with spices and a gram flour batter, left in the refrigerator for 15 minutes to enhance adhesion, and then fried in moderately hot sunflower oil until golden brown.

***** Only the shrimp heads were removed before washing with a salt and vinegar solution, followed by rinsing with tap water. A skewer was inserted beneath the shell to straighten the shrimp, which were then blanched in hot water for 3 minutes and transferred to chilled water for another 3 minutes. After removing the skewer and peeling the shell, the shrimp were coated with boiled rice and soy sauce.

After cooking, samples cooled and kept below 4°C till analyzed for heavy metals concentration in fisheries laboratories, CEMB University of Karachi and Department of Geology, University of Karachi, while proximate composition analysis were taken in PCSIR laboratories of Karachi, Pakistan.

Heavy metal analysis

The same technique used by Zafar *et al.* (2019b) was performed in this study. For the analysis of heavy metals, a total of 30 raw samples of same size and weight group of shrimps were selected and then dried in oven at 80°C for 3-4 hrs. The completely dried and the cooked shrimps of 30 samples of individual were homogenized with the help of pestle and mortar. For testing of Fe, Zn, Mn, Pb, Cr, Cu taken 5 g of homogenized samples of each group were taken in volumetric flask then accurately 10ml of HNO₃ were added to each flask and the samples were digested on the plate between 80-100 degree until the solution become cleared (El-Moselhy, 2014). The contents were filtered through filter paper in 25 ml volumetric flasks, and the volume was made with deionized distilled water. Hitachi model Z-5000 Atomic Absorption Spectrophotometer (AAS) equipped with Zeeman Background Correction and equipped with graphite was used to determine heavy metals in the samples. The recovery of the spiked metals was close to (97- 99%) for all tested metals by proposed method.

Proximate composition

Moisture, total ash, crude protein and crude fat content in shrimps' samples were determined using accepted analysis methods of the Association of Official Analytical Chemists (AOAC, 2012). Moisture content was determined by drying the sample at 105°C according to the AOAC method 925.10. Ash content was determined by incineration at 600°C according to the AOAC method 942.05. Total

nitrogen (N) was determined by the Kjeldahl method, AOAC method 65.17, and protein was calculated as total N $\times$ 6.25. Fat content was determined using the Soxhlet apparatus with hexane, following AOAC 963.15 method. The carbohydrate content (%) was calculated by difference: 100 – (Moisture + Crude protein + Crude fat + Total ash). Gross energy value (calories) was used for standard factor of 3.75, 9 and 4 Kcal/g for protein, lipid and carbohydrate, respectively. The energy contents were summed to give total or gross energy of the sample (AOAC, 2012).

Health risk assessment

The health risk of consuming heavy metal contaminated shrimps was determined by calculating Estimated Daily Intake (EDI), Hazard Quotient (HQ) and Hazard Index (HI). These were calculated for each detected heavy metals in the shrimps' samples. If HI is greater than 1 it shows significant risk associated with the consumptions of *P. monodon* and *P. indicus*, while HI is lower than 1 indicates no potential health risk.

EDI (mg/kg body wt. per day) values of heavy metals were calculated and compared to their reference dose (Rf. D.) established by Risk Assessment Information System (RAIS, 2023). EDI was calculated by using formula 1.

$$EDI = \frac{MC_s \times IR_s}{Body\ wt.} \quad (1)$$

Where MCs is the heavy metals concentrations in shrimps (mg/kg wet wt.); IRs is the ingestion rate of shrimps (g/day). The IRs values were 27.4 and 41 g/day for children, and adults, respectively (UNSCEAR, 2010). The body wt. is the average body weight of the persons; 30 and 70 kg for children, and adults, respectively (UNSCEAR, 2010).

The HQs were calculated by formula 2.

$$HQ = \frac{EDI}{Rf.D.} \quad (2)$$

The HI estimated the cumulative risk associated with exposure to multiple heavy metals and was calculated by formula 3.

$$HI = \sum HQ \quad (3)$$

Statistical analysis

To determine significant differences among the samples obtained from different shrimp species (*P. monodon* and *P. indicus*) and cooking methods (raw, fried, and sushi), one-way analysis of variance (ANOVA) was performed. When significant differences were observed ($p < 0.05$), post hoc comparisons were conducted using Tukey's HSD test to identify pairwise differences among the groups. All statistical analyses were carried out using IBM SPSS Statistics version 27, and results were reported as mean $\pm$ standard deviation.

Results and discussion

P. monodon and *P. indicus* have great commercial value in Pakistan. These species are known as Jaira and Safeed Jhenga in local parlance

Biochemical Composition

Biochemical composition of *P. monodon* and *P. indicus* was made to determine nutritive quality in (pre and post cooking) raw, fried and sushi which is a common cooking practice of Asian countries. The results of biochemical composition analysis and statistical differences among samples obtained from different shrimp species with different cooking methods were presented in Table 3. The results indicated that moisture concentration in raw samples was the highest as compared to fried and sushi. It is assumed that fried cooking practice reduce moisture content of the food.

Protein increase in fried method is generally due to evaporation of moisture and other contents such as use of gram flour for coating that leaves protein merely in the dish. It should be noted that the cooking practice could affect nutritional composition of the food. It is general observed that fried method of cooking could multiplies calories which is determine in present study. Taking about carbohydrates present it was observed that higher concentration in sushi defines post mixing of rice in the recipe. Further it was noted that shrimps were coated with rice showed higher concentration in sushi method. However, its meagre presence was noted in fried and raw sample.

The concentration of crude fat was higher in fried method whereas in sushi and raw method were lower in concentration. When we fry something in oils, the food absorbs the oil, increasing its fat contents.

Consequently, that deep fried stuff is tasty but hold higher fat concentrations. Consumption of such food can harm human health.

Fried shrimp is likely to have a higher ash content compared to raw shrimps. Ash content represent the mineral contents left after burned, and the frying process can concentrate minerals in the food.

Table 3. Mean $\pm$ SD of proximate composition in raw and cooked shrimp commonly consumed worldwide.

	<i>Penaeus monodon</i>			<i>Penaeus indicus</i>		
	Raw	Fried	Sushi	Raw	Fried	Sushi
Moisture %	72.4 $\pm$ 1.51 ^a	38.1 $\pm$ 1.58 ^b	70.5 $\pm$ 0.55 ^a	77.8 $\pm$ 0.85 ^a	35.6 $\pm$ 0.45 ^b	75.3 $\pm$ 0.62 ^a
Ash %	2.79 $\pm$ 1.5 ^b	2.75 $\pm$ 0.1 ^b	1.88 $\pm$ 0.1 ^c	1.35 $\pm$ 0.2 ^d	3.50 $\pm$ 0.5 ^a	1.91 $\pm$ 0.1 ^c
Crude fat %	3.04 $\pm$ 1.0 ^b	19.31 $\pm$ 0.8 ^a	0.78 $\pm$ 0.1 ^d	0.89 $\pm$ 0.2 ^{c,d}	15.73 $\pm$ 0.5 ^a	1.39 $\pm$ 0.5 ^c
Crude protein	25.37 $\pm$ 1.0 ^b	39.31 $\pm$ 1.0 ^a	15.52 $\pm$ 0.6 ^c	16.16 $\pm$ 0.4 ^c	39.05 $\pm$ 0.4 ^a	13.96 $\pm$ 0.1 ^c
Carbohydrate %	0.07 $\pm$ 0.02 ^e	1.55 $\pm$ 0.05 ^d	12.92 $\pm$ 0.97 ^a	3.77 $\pm$ 0.45 ^c	6.17 $\pm$ 0.21 ^b	7.82 $\pm$ 0.58 ^b
Calories Kcal/100g	121.5 $\pm$ 1.5 ^c	338.79 $\pm$ 1.0 ^a	115.7 $\pm$ 0.8 ^c	87 $\pm$ 5.13 ^e	271 $\pm$ 5.1 ^b	106.2 $\pm$ 0.6 ^d

The different superscript letters (a, b, c, d, e) indicate significant differences ($p < 0.05$) within each row.

Heavy metals analysis

A comparison was made between raw, fried, and sushi to examine the variation and bioaccumulation of Fe, Cu, Zn, Li, Cr, and Pb in the muscle tissues of shrimps. The heavy metal concentrations and statistical differences among samples obtained from different shrimp species with different cooking methods were presented in Table 4. It was further recorded that the concentration of Cr and Pb in all raw, fried, and sushi samples was below the detectable level (BDL). Overall the concentration of Fe and Zn, in raw samples of *P. indicus* were higher as compared to *P. monodon* but after apply the culinary practices the concentrations of Fe, Zn and Li in fried samples of *P. monodon* were high which was 0.42 ± 0.002 , 0.042 ± 0.002 and 0.145 ± 0.002 mg/kg wet wt., respectively as compared to raw and sushi samples. Furthermore, Fe and Zn were 0.213 ± 0.0030 and 0.094 ± 0.001 mg/kg wet wt., respectively in raw samples of *P. indicus* but these metals were decreased in fried and sushi samples. Li was high in concentration 0.145 ± 0.002 mg/kg wet wt. in fried samples of *P. monodon* as compared raw and sushi but in the samples of *P. indicus* the concentration of Li was BDL in both raw and fried samples as compared to sushi sample. Sunflower oil was used while frying shrimps to enhance their texture and taste are the main sources to increase the concentration of these essential heavy metals in case of *P. monodon* but in case of *P. indicus* this pattern was not observed. The concentration of Cu in fried (0.154 ± 0.0016 mg/kg wet wt.) and in sushi (0.033 ± 0.002 mg/kg wet wt.) are low in *P. monodon* as compared to raw (0.263 ± 0.002 mg/kg wet wt.). While the concentration of Cu in sushi of *P. indicus* were below detectable (BDL) as compared to raw and fried samples (0.0089 ± 0.00021 and 0.0085 ± 0.00015 mg/kg wet wt., respectively and so that means the landed specimens of both shrimp species at Karachi fish harbour show positive value of Cu in their muscles tissue except sushi of *P. indicus* which is remarkably affected during selected culinary practice.

Table 4. Mean $\pm$ SD of heavy metal concentrations given mg/kg wet wt. in raw and cooked shrimps with comparison of WHO limitation (BDL: below detectable level).

Species	methods	Heavy metals					
		Fe	Zn	Cu	Li	Cr	Pb
<i>Penaeus monodon</i>	Raw	0.096 ± 0.002^d	0.026 ± 0.001^c	0.263 ± 0.002^a	0.015 ± 0.002^c	BDL	BDL
	Fried	0.42 ± 0.002^a	0.042 ± 0.002^b	0.154 ± 0.001^b	0.145 ± 0.002^a	BDL	BDL
	Sushi	0.134 ± 0.002^c	0.024 ± 0.002^c	0.033 ± 0.002^c	0.053 ± 0.002^b	BDL	BDL
<i>Penaeus indicus</i>	Raw	0.213 ± 0.003^b	0.094 ± 0.001^a	0.009 ± 0.0002^d	BDL	BDL	BDL
	Fried	0.140 ± 0.002^c	0.047 ± 0.005^b	0.0085 ± 0.0002^d	BDL	BDL	BDL
	Sushi	0.087 ± 0.02^d	0.09 ± 0.02^a	BDL	0.051 ± 0.005^b	BDL	BDL
WHO		100	40	30	80	2	--

The different superscript letters (a, b, c, d) indicate significant differences ($p < 0.05$) within each column.

The one-way analysis of variance (ANOVA) revealed significant differences in heavy metal concentrations between the two shrimp species, *P. monodon* and *P. indicus* ($p < 0.05$). The impact of species on metal accumulation was highlighted by

post hoc comparisons using Tukey's HSD test, which showed that the heavy metal levels except Cr, Pb and Li varied significantly between the species. Significant differences in heavy metal concentrations were also found when the same species was cooked using different techniques (raw, fried, and sushi). In particular, the concentrations of heavy metals found in raw shrimp differed significantly from those found in fried and sushi preparations, suggesting that cooking techniques affect shrimp's heavy metal content.

Health risk assessment

The health risk analysis in the present study involved estimating the EDI of metals in shrimps' samples for children and adults based on their seafood consumption rates and average body weights. EDIs are less than unity for all metals (Table 5). All EDI values are lower than the Rf. D., indicating that they will not cause any toxic effects for consumers.

Table 5. EDI of heavy metals through tiger shrimps' consumption by people and their comparison with permissible tolerable daily intake.

Species	Age group	Cooking methods	Fe	Zn	Cu	Li
<i>Penaeus monodon</i>	Children	Raw	8.64×10^{-5}	2.34×10^{-5}	2.37×10^{-4}	1.35×10^{-5}
		Fried	3.78×10^{-4}	3.78×10^{-5}	1.39×10^{-4}	1.31×10^{-4}
		Sushi	1.21×10^{-4}	2.16×10^{-5}	2.97×10^{-5}	4.77×10^{-5}
	Adults	Raw	5.62×10^{-5}	1.52×10^{-5}	1.54×10^{-4}	8.79×10^{-6}
		Fried	2.46×10^{-4}	2.46×10^{-5}	9.02×10^{-5}	8.49×10^{-5}
		Sushi	7.85×10^{-5}	1.41×10^{-5}	1.93×10^{-5}	3.10×10^{-5}
<i>Penaeus Indicus</i>	Children	Raw	1.92×10^{-4}	8.46×10^{-5}	8.01×10^{-6}	BLD
		Fried	1.26×10^{-4}	4.23×10^{-5}	7.65×10^{-6}	BLD
		Sushi	7.83×10^{-5}	8.10×10^{-5}	BLD	4.50×10^{-5}
	Adults	Raw	1.25×10^{-4}	5.51×10^{-5}	5.21×10^{-6}	BLD
		Fried	8.20×10^{-5}	2.75×10^{-5}	4.98×10^{-6}	BLD
		Sushi	5.10×10^{-5}	5.27×10^{-5}	BLD	2.93×10^{-5}
RAIS (mg/day/person)			7.00×10^{-1}	3.00×10^{-1}	4.00×10^{-2}	2.00×10^{-3}

Table 6 presents HQs and HI of heavy metals for children and adults consuming shrimps from Pakistan coasts of the Arabian Sea. HQs are less than unity for all metals and HI values for all metals were found to be less than 1.

Several studies have already been conducted to determine the proximate composition and metal accumulation in seafood. This study aims to provide information on the pre and post cooking effects to understand the quality transformation and losses that occur before the seafood reaches the dining table. According to Ockerman *et al.* (2017) proximate composition varies with species and is influenced by season, water temperature, and spawning cycle. In the present study the protein content seemed to increase in fried sample due to traditionally adding of gram flour as a coating medium for frying, which has high protein content same finding reported by Osibona

(2011). The increase in protein content may be attributed to their high protein dietary intake, which includes algae, diatoms, and crustaceans.

Table 6. THQs and HIs of heavy metals content through tiger shrimps' consumption by people.

Species	Age group	Cooking methods	THQs				HIs
			Fe	Zn	Cu	Li	
<i>Penaeus monodon</i>	Children	Raw	1.2×10^{-4}	7.8×10^{-5}	5.9×10^{-3}	6.8×10^{-3}	1.3×10^{-2}
		Fried	5.4×10^{-4}	1.3×10^{-4}	3.5×10^{-3}	6.5×10^{-2}	6.9×10^{-2}
		Sushi	1.7×10^{-4}	7.2×10^{-5}	7.4×10^{-4}	2.4×10^{-2}	2.5×10^{-2}
	Adults	Raw	8.0×10^{-5}	5.1×10^{-5}	3.9×10^{-3}	4.4×10^{-3}	8.4×10^{-3}
		Fried	3.5×10^{-4}	8.2×10^{-5}	2.3×10^{-3}	4.3×10^{-2}	4.5×10^{-2}
		Sushi	1.1×10^{-4}	4.7×10^{-5}	4.8×10^{-4}	1.6×10^{-2}	1.6×10^{-2}
<i>Penaeus indicus</i>	Children	Raw	2.7×10^{-4}	2.8×10^{-4}	2.0×10^{-4}	BLD	7.6×10^{-4}
		Fried	1.8×10^{-4}	1.4×10^{-4}	1.9×10^{-4}	BLD	5.1×10^{-4}
		Sushi	1.1×10^{-4}	2.7×10^{-4}	BLD	2.3×10^{-2}	2.3×10^{-2}
	Adults	Raw	1.8×10^{-4}	1.8×10^{-4}	1.3×10^{-4}	BLD	4.9×10^{-4}
		Fried	1.2×10^{-4}	9.2×10^{-5}	1.2×10^{-4}	BLD	3.3×10^{-4}
		Sushi	7.3×10^{-5}	1.8×10^{-4}	BLD	1.5×10^{-2}	1.5×10^{-2}

In the present study, a significantly higher fat content was observed in fried shrimp compared to raw and sushi samples. Saguy and Dana (2003) noted that the increase in fat content of fried seafood is related to oil absorption during the frying process. Additionally, the high amount of fat content can be attributed to the oil penetration after the partially removal of water content during frying. During this study the highest level of carbohydrate was found in sushi sample as compared to raw and fried sample because during the preparation of sushi the coating of rice was used. Same observation was reported by (Đorđević and Buchtová, 2020) in seafood. In the present study, it was found that the sushi samples had the lowest value of ash content compared to the fried samples. This suggests that further research and investigation are needed to understand why the ash content is lower in the sushi samples. It was also found that the moisture content decreased in all cooking methods (Kumar *et al.*, 2012).

Observing calorie content in sushi was very low as compared to raw and fried samples of both shrimp's species. Correspondingly, sushi meal is categorized as very low-calorie meal as data provided by various authors (Hermann *et al.*, 2010; Đorđević and Buchtová, 2020).

The degree of metal augmentation in the cooked samples was mostly caused by the water loss during cooking, which was influenced by the size of the fish. Kalogeropoulos *et al.* (2012) observed that small sized fish and shellfish which were domestically pan-fried and grilled produce higher metal contents than the raw

samples. Grilling produced less water loss at temperatures comparable to pan-frying, which keeps metal concentrations lower.

In present study the non-essential heavy metals (Cr and Pb) were found below detectable levels in all analysed shrimps' samples. It was also observed that the amounts of Pb and Cr in both shrimp species were not accumulated in their muscles tissues and therefore appear to be an acceptable food item (Meshram *et al.*, 2014). Levels of Zn and Cu have been found to be higher and it could be explained that these metals play an important role in enzymatic and respiratory processes in aquatic animals (Çoğun *et al.*, 2005). The highest level of Fe in fried sample of *P. monodon* was noted in this study and the same finding has also been observed in *Fenneropenaeus indicus* by Salam *et al.* (2019). Heavy metals such as Fe, Cu, and Zn exhibit high level of toxicity and durability, capable of persisting in the environment and accumulating in organism, potentially magnifying through the food chain (Hosseini *et al.*, 2012; Zhang and Shu, 2010; Ahmed *et al.* 2015; Kibria *et al.*, 2016).

In the present study EDI and HQs calculated for the edible tissues of shrimps showed HI less than 1 for all metals. This shows that the consumption of these shrimps cannot cause any toxic effects. The calculation of EDI and HI for detected heavy metals

Emphasises the importance of integrating health risk assessment into environmental studies. These findings have direct implications for both fisheries management and public health assessments and interventions.

Conclusions

Edible flesh from tiger shrimps, *P. monodon* and *P. indicus* were used to determine proximate composition and metals concentration in raw, fried and sushi culinary recipes to understand energy loss and metal volatility during different methods. Fried cooking recipe contained high level of fats due to accumulation of oil contents in the flesh. Higher protein level in fried method may be due to gram flour. The carbohydrates in sushi were higher which was due to coating of rice whereas moisture was in elevated form.

The metal concentration of Fe, Zn and Li found to be within the permissible limits of the international organizations like FAO (2009) and WHO (2023) guidelines. Whereas the concentration of Pb and Cr was below detectable. However, overall outcomes of this study suggests that biochemical composition and energy transfer is stable and due to volatility of most of the heavy metals there is no harm to consuming tiger shrimp and Indian white shrimp by these culinary methods in Pakistan.

The present study underscores the importance of monitoring heavy metal contamination in marine environments of Pakistan, Arabian Sea to safeguard public health. The EDI and HI are crucial metrics for evaluating the potential health risks linked to the consumption of seafood containing heavy metals. These indices were calculated for various metals, and the HI values were found to be less than 1 for all metals studied.

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