



## Metal Contaminants in *Litopenaeus Vannamei* and *Penaeus Silasi* at Various Supply Chain Stages in Johor Bahru, Malaysia and Their Health Risk Assessment

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### Abstract

This present research evaluated the concentrations of As and total Hg as prescribed by the Malaysian Food Act (1983) and Regulations (2021) in *P. silasi* and *L. vannamei* shrimps sampled from the different supply chain stages (the landing site Pontian, a major market and grocers) in Johor Bahru, Johor, Malaysia at three sampling intervals. Graphite furnace atomic absorption spectrometer analysis revealed the median of As concentrations during the three sampling intervals in Johor, ranging between 0.77 – 36.11 mg/kg. Flow injection mercury/hydride system analysis revealed the total Hg concentrations ranged between 0.03-0.88 mg/kg. The ranges of As (all sampling sites: 0.16 - 49.99 mg/kg) and total Hg (landing site Pontian: 0.17- 1.20 mg/kg) concentrations exceeded the maximum permitted proportions prescribed by the Malaysian law. Identified as *P. silasi*, shrimps from landing site Pontian had significantly higher As and total Hg concentrations than those of *L. vannamei* from major market A and its surrounding grocers ( $P < 0.05$ ). Alarmingly, Hazard Index for *P. silasi* from landing site Pontian as well as *L. vannamei* from major market and grocers exceeded 1.00. Hence, formulating suitable intervention programs and continuous monitoring programs are paramount for benefiting the public at large.

**Keywords:** *Litopenaeus vannamei*; *Penaeus silasi*; Metal contaminants; Health risk assessment.

### Introduction

White shrimp (*Litopenaeus vannamei*) and false white shrimp (*Penaeus silasi*) are popularly consumed aquaculture products in Malaysia [1, 2]. The escalating demand for these shrimps has been attributable to its comparatively affordable price and favorable nutritional values [3], as well as the rapidly growing population [4].

Pollution by metal contaminants in the aquatic ecosystem is one of the most alarming, environmental issues because of its inherent toxicity, non-degradability and persistency in water [5]. The situation would cause toxic accumulations in aquatic organisms [6]. Industrial, agricultural and mining activities are among the major sources of the anthropogenic contamination of the aquatic ecosystem for metal contaminants [7].

Specifically for Malaysia, its Food Act 1983 (Act 281) and Regulations (2021) [8] outlines the Maximum Permitted Proportions (MPPs) (wet weight) for arsenic (As) as 1.0 mg/kg and with slightly lower MPP of 0.5 mg/kg for total mercury (total Hg) in crustaceans/shrimps. Since contamination by metal contaminants in shrimps has been indicated [9, 10], efforts to monitor such contaminants in important aquaculture commodities like *L. vannamei* and *P. silasi* consumed by the consumers appears imperative.

Review of literature revealed no study that investigated the amount of metal contaminants (As and total Hg) in the same matrix sold at various supply chain stages (seafood landing site, major market and retail grocers) in Malaysia. While it is important to ensure that the concentrations of metal contaminants (viz. As and total Hg) in *L. vannamei* and *P. silasi* comply fully with the prescribed MPPs, it is also paramount to perform its health risk assessment (target hazard quotient (THQ) and hazard index (HI)). Such evaluations are important because the long-term consumption of metal contaminants (even at low concentrations) can cause detrimental effects on human [11].

Hence, this study was designed to determine the amounts of As and total Hg and in *L. vannamei* and *P. silasi* sampled from various supply chain stages (viz. landing site Pontian, a major market and retail grocers) in Johor Bahru, Malaysia. Moreover, this research investigated its health risk assessment which may prove beneficial for the relevant health authorities for implementing appropriate intervention strategies for the community.

### Materials and methods

All chemicals and reagents as well as standards (As in 2% HNO<sub>3</sub> and total Hg in 10% HNO<sub>3</sub>) were purchased from Merck (Germany). All standards were purchased in the liquid form ( $\geq 99.5\%$ ) of 1000 mg/L for each standard.

Each sample (300 g of pooled shrimps) was purchased from three different stages of supply chain in Johor Bahru, Johor, Malaysia involving landing site Pontian, major market A and selected retail grocers over a period of three sampling intervals (September, October and December 2020) The grocers were situated within 2 km radius from the major market A (4 grocers for each surrounding area). Morphological taxonomic identification of the shrimps was performed by a fishery expert from Universiti Malaysia Terengganu.

The chemical analyses were conducted in a certified (MS ISO/IEC 17025:2017) commercial laboratory at Universiti Teknologi Malaysia, Johor Bahru. Analysis of As and total Hg was performed using the laboratory in-house methods (TM/UTM/CHEM/SF), adapted from the AOAC 999.10 standard methods, Association of Official Analytical Collaboration, AOAC. Prior to chemical analysis using graphite furnace-atomic absorption spectrometer (GF-AAS) for As and flow injection mercury/hydride system (FI-MHS) for total Hg, the sample was digested using a microwave digester (Titan-MPS, Perkin Elmer).

The data were analyzed using the UTM licensed IBM Statistical Package for Social Sciences (SPSS) version 27.0. Since the data violated the assumption of normality, inferential statistic was done using the non-parametric Kruskal-Wallis H and Mann-Whitney U tests. Level of significance of 0.05 ( $\alpha < 0.05$ ) was utilized for determining the significant differences among groups [12].

The THQ and HI were used to evaluate the health risk assessment for consuming the shrimps for human using equation, as suggested by Mahat et al. [13].

### Results and discussion

The taxonomic and molecular identifications of the shrimps sampled from all the sampling sites revealed the presence of two different species namely *P. silasi* (false white shrimp) and *L. vannamei* (white shrimp). *P. silasi* was the only species of 'white shrimps' available during sampling at Pontian landing sites. On the other hand, *L. vannamei* was the one found in major markets A as well as its surrounding grocers. As such, it can be construed that the shrimps harvested from the ocean nearby Pontian may not be the one contributing to the supply chain of shrimps sold in major market A and its surrounding grocers in Johor Bahru, as investigated during the course of three different sampling intervals of this present research.

Table 1 represents the comparison of As and total Hg concentrations in shrimps between Pontian landing site (*P. silasi*) and major market A (*L. vannamei*) as well as its surrounding grocers in Johor Bahru during the three sampling intervals. Since the data violated the assumption of normality, they are presented as median (range) and tested using the non-parametric Mann-Whitney U test.

**Table 1.** Comparison of metal contaminant concentrations in shrimps between Pontian landing sites (*P. silasi*) and major market A (*L. vannamei*) as well as its surrounding grocers in Johor Bahru during the three sampling intervals.

Metal contaminants [Maximum Permitted Proportion]	Sampling sites					
	Landing site-Pontian	Major market A	Grocer A1	Grocer A2	Grocer A3	Grocer A4
As [1.00 mg/kg]	<b>36.11<sup>S</sup></b> (1.61 - 49.99)	1.50 (0.16 - 3.55)	2.24 (1.5 - 2.67)	0.77 (0.31 - 2.54)	2.95 (0.20 - 3.59)	1.57 (0.54 - 1.93)
Total Hg [0.50 mg/kg]	<b>0.88<sup>S</sup></b> (0.17 - 1.20)	0.03 (0.02 - 0.07)	0.05 (0.03 - 0.05)	0.06 (0.03 - 0.09)	0.03 (0.02 - 0.10)	0.03 (0.02 - 0.05)

The data are presented as median (range), and analyzed using the Mann-Whitney U test for comparing the concentrations of metal contaminants in shrimps sampled between landing sites Pontian and major markets A in Johor Bahru. Significantly higher concentrations of As and total Hg were observed in shrimps sampled from the landing site Pontian when compared with nearby major market A (S). As for comparing the major market A and its surrounding grocers, significant differences in the concentrations of metal contaminants in shrimps were not observed. Level of significance of 0.05 was used for assigning significant differences

Significantly higher median concentrations of As (36.11 mg/kg) and total Hg (0.88 mg/kg) were observed for shrimp samples from the landing site of Pontian when compared with major market A (As: 1.50 mg/kg; total Hg: 0.03 mg/kg) ( $P < 0.05$ ). Since no significant difference in the concentrations of metal contaminants in shrimps between the major market A and its surrounding four grocers was observed (Table 1,  $P > 0.05$ ), the proposition that the shrimps available in the surrounding grocers may originate from their respective nearby major market, appears to be supported.

Since the ranges of As (all sampling sites: 0.16 - 49.99 mg/kg) and total Hg (landing site Pontian: 0.17- 1.20 mg/kg) concentrations expanded beyond the prescribed MPPs (As: 1.00 mg/kg, total Hg: 0.5 mg/kg), therefore, investigating its possible detrimental health implications for consuming the shrimps is a matter of public health significance. A similar indication has also been made by previous researchers whom investigated the parallel issue following the ingestion of *Perna viridis* mussels in Johor Bahru, Johor, Malaysia [13].

**Table 2.** Target hazard quotients (THQs) of metal contaminants as well as Hazard index (HI) by consuming *L. vannamei* and *P. silasi* shrimps sampled during three month of sampling interval. Average consumer (AC); high consumer (HC).

Sampling sites	Target Hazard Quotient				Hazard index
	As		Hg		
	AC	HC	AC	HC	
Landing site-Pontian	<b>0.30-9.08</b>	<b>0.90-27.23</b>	0.01-0.06	0.03-0.19	<b>0.93-27.43</b>
Major market A	0.03-0.62	<b>0.09-1.85</b>	0.00-0.00	0.00-0.01	<b>0.10-1.86</b>
Grocer A1	0.28-0.45	<b>0.83-1.35</b>	0.00-0.00	0.01-0.01	<b>0.83-1.36</b>
Grocer A2	0.06-0.45	<b>0.19-1.36</b>	0.00-0.01	0.01-0.02	<b>0.19-1.37</b>
Grocer A3	0.04-0.63	<b>0.12-1.88</b>	0.00-0.00	0.00-0.01	<b>0.12-1.89</b>
Grocer A4	0.11-0.35	<b>0.32-1.05</b>	0.00-0.00	0.00-0.01	<b>0.33-1.05</b>

Values in **bold** represent the values of THQ and/or HI that exceeded 1.00.

THQ > 1: may 'experience obvious deleterious effects', the probability of having greater hazard risk

HI > 1: Occurrence of non- cancer human health impact is expected.

Table 2 represents the THQ and HI values of As and total Hg by consuming the shrimps sampled during three sampling intervals. The calculated THQs for As for HC exceeded 1.00 for all the sampling sites. The highest ranges of THQs for HC were observed in shrimps from the landing sites Pontian (0.90-27.23), followed by the remaining sampling sites of major markets and their surrounding grocers. On the other hand, the ranges of THQs for AC exceeded 1.00 only for shrimps from the landing sites Pontian (0.30-9.08) alone (Table 2). The fact that the ranges of THQ (especially for those of HC) were higher than 1.00, the possibility of human population exposed to such contaminated shrimp may 'experience deleterious effects' cannot be excluded.

Moreover, assessment of the HI value is important to consider the combinatory influence of multiple metal contaminants that occur in aquaculture products (like shrimps). Results revealed that the ranges of HI exceeded 1.00 in all the samples, and expectedly, the highest HI ranges were evident in shrimps sampled from the landing site Pontian (0.94-27.43) (Table 2). Since the ranges of HI reported here exceeded the prescribed cut-off value of 1.00, the fact that populations exposed to such contaminated shrimps may be at risk for developing non-cancer health impacts is an alarming matter of public health consideration.

### Conclusion

Significantly higher concentrations of As and total Hg were observed in *P. silasi* sampled from the landing site Pontian when compared with that of *L. vannamei* from major markets A and its surrounding grocers ( $P < 0.05$ ). For As and total Hg, their concentrations appear to exceed the MPPs prescribed by the Malaysian law. The fact that the THQ and HI for HC exceeded the cut-off values of 1.00, the shrimps may not be safe for prolonged human consumption. This is a matter of importance that requires immediate remedial intervention, especially for developing suitable removal technologies prior to consumption. Considering the different species of 'white shrimps' observed at Pontian landing sites (*P. silasi*) as well as their varying concentrations of metal contaminants when compared with the major market and their surrounding grocers (*L. vannamei*), it is apparent that the shrimps sold at major market and grocers in Johor Bahru are originating from the different sources than that of Pontian landing sites. The fact that in this present research the sampling was done only in three-month sampling interval, further medium to long-term studies focusing on this aspect merit consideration. Nonetheless, this traceability study for shrimps at various stages of supply chain in Johor is deemed useful for strengthening the overall safety and security of our seafood industry.

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