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Health Risk of Consuming Heavy Metals in Farmed Tilapia in Central Taiwan

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Abstract Taiwan is a small island nation, and many aquaculture sites are located near industrial parks. Anthropogenic activities may contaminate fish ponds. Therefore, we investigated the concentrations of eight metal elements in tilapia tissues (muscle and scale) at two fisheries. We compared the difference in metal content in tilapia at these two fisheries with that in non-contaminated fish at the Fisheries Research Institute. Probabilistic risk analysis was carried out to assess the health risk for people who eat contaminated tilapia. The predicted 95th percentiles of the hazard quotient and excess lifetime cancer risk for residents consuming contaminated tilapia were found to be in the range of 3.1–9.2 and $1.03 \times 10^{-5} \sim 1.85 \times 10^{-5}$, respectively.

Keywords Heavy metals · Human health risk · Tilapia · Monte Carlo simulation

Heavy metals occur in the environment both as a result of natural processes and as pollutants from human activities (Karadede-Akin and Unlu 2007). Anthropogenic activities, such as mining and industrial processing, are the main sources of heavy metal contamination in the environment. Under certain conditions, these heavy metals may accumulate to toxic concentration levels, which can lead to ecological damage (Wang et al. 2005). Human exposure to heavy metals can occur through a variety of routes, such as inhalation of air pollutants or contaminated soil particles and consumption of contaminated foods. Dietary intake is the main route of exposure for most people, although inhalation can play an important role in very contaminated sites. Thus, information about heavy metal concentrations in food products and their dietary intake is very important for assessing their risks to human health (Li et al. 2007). Bioaccumulation can potentially threaten the health of many species at the top of the food chain, especially fish and humans (Adeniyi et al. 2008). Heavy metals can accumulate in the tissues of aquatic animals, as such, tissue concentrations of heavy metals can be of public concern (Farombi et al. 2007).

Tilapia represents more than 30% of the total species of commercially raised fish in Taiwan (Liu 2004). In 2007, the annual production of tilapia was about 76,097 tons and the output value was about 7,750 US dollars (TFA 2007). Taiwan is a small island nation, and many aquaculture sites are located within a short distance from industrial parks. It is highly possible that anthropogenic activities contaminate the fish ponds. People can be exposed to toxic chemicals that have accumulated in contaminated fish when they are consumed (Chien et al. 2002). Therefore, it is important to assess the potential health risk of commercially farmed tilapia in Taiwan. Excess lifetime cancer risk (ELCR) and hazard quotients (HQ) equations are commonly applied to calculate the carcinogenic and noncarcinogenic risk associated with a variety of chemical exposures. In addition, Monte Carlo simulations are often applied to ELCRs and HQs equations to help determine the overall impact of

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parameter variability and uncertainty on risk assessment predictions (Clewell et al. 1999). The objective of this study was to (1) determine the heavy metal concentrations in tissues (dorsal muscle, ventral muscle and scale) of tilapia, (2) investigate the heavy metal concentrations in fish ponds located near highly industrialized areas, and (3) estimate the health risk of heavy metals to humans via consumption of tilapia muscle. The findings are expected to assist the relevant government agencies in ensuring a good surface water ecological status.

Materials and Methods

Two sites were selected as our sampling sites for biomonitoring, Long-Jing (LJ) and Sheng-Keng (SK). The LJ sampling site is situated ~3 km southeast of a coal-fired power plant (CFPP) and 5 km south of an iron and steel manufacturing plant (ISMP). The SK sampling site is situated ~3 km south of a CFPP and 7 km south of an ISMP. Therefore, these sites are representative of aquaculture projects located in industrialized areas. For comparison purposes, tilapia were also collected from the Fisheries Research Institute at Chu-Pei (CP). The CP is situated ~100 km north of a CFPP and an ISMP. The CP was considered to be unaffected by anthropogenic emissions and thereby suitable as a reference site (Fig. 1).

Fish samples were collected in September 2007 from fish ponds at SK, LJ, and CP. At each site, 4 samples of tilapia of the same age and similar size were collected with nets. Tilapia were dissected after asphyxiation. Dorsal muscle, ventral muscle, and scales were extracted

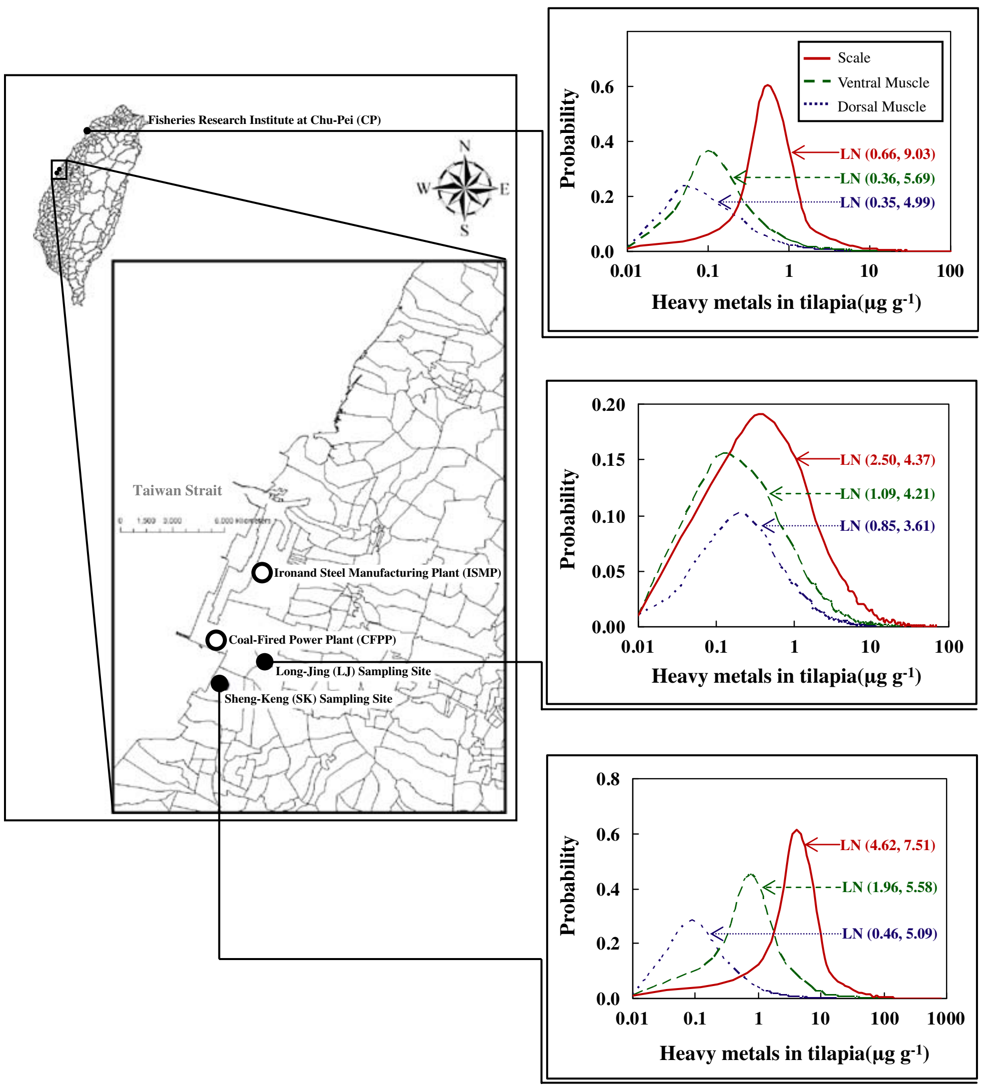

Fig. 1 Map of fishery ponds for this study and rose histogram of wind direction (1 h average data) for the period 2001–2005 recorded at the Long-Jing sampling site. Overall display of probabilistic distributions of heavy metal concentrations at two selected fishery ponds (Long-Jing and Sheng-Keng) and Fisheries Research Institute (Chu-Pei)

immediately after collection from the fish ponds. The conventional pressure bomb digestion method was used to determine the concentrations of arsenic (As), chromium (Cr), copper (Cu), manganese (Mn), nickel (Ni), lead (Pb), selenium (Se), and zinc (Zn) in tilapia muscle and scales. Conventional high-pressure digestion with acid mixture was performed in a pressure bomb system (Berghof, Enningen, Germany) using PTFE digestion vessels. The fish from each site ($\sim$0.2 g) were weighed in the PTFE digestion vessels prior to the addition of a predetermined amount of 3 mL of nitric acid. Because of the thermal properties of PTFE, the maximum temperature should be below 180°C during the digestion process in the aluminum heating block. In this study, a heating period of 4 h was employed to ensure the achievement of total decomposition. After the decomposition, the samples were diluted to 25 mL with purified water, and preserved at 5°C until analyzed. The concentrations of heavy metals (As, Cr, Cu, Mn, Ni, Pb, Se, and Zn) were measured with an inductively coupled plasma-mass spectrometer (PerkinElmer ELAN DRC II ICP-MS). In this study, we assumed that inorganic As accounts for 7.4% of the total As in tilapia muscle, as suggested by Huang et al. (2003).

The methodology for estimation of ELCRs and HQs was provided in the USEPA Region III risk-based concentration table (USEPA 2006). The ELCR associated with the carcinogenic risk of As was expressed as the excess probability of developing cancer over a lifetime of 70 years. The HQ was used to indicate non-carcinogenic risk associated with As, Cr, Cu, Mn, Ni, Pb, Se, and Zn over an average time of 30 years. The acceptable risk distribution was assigned by constraint on percentiles. The lower end of the range of acceptable risk distribution is defined by a single constraint on the 95th percentile of risk distribution that must be equal to or lower than 1×10^{-6} for ELCR and equal to or lower than 1 for HQ (Burmaster and Hull 1997).

The ELCR in adults is defined as:

$$\mathrm{ELCR} = \frac{C_{\mathrm{f}} \times \left(\mathrm{CSF_o} \times \left(\frac{\mathrm{BW_a}}{70\,\mathrm{kg}}\right)^{\frac{1}{3}}\right) \times \mathrm{IR} \times \mathrm{EF_r} \times \mathrm{ED_{tot}}}{\mathrm{BW_a} \times \mathrm{AT_c} \times 10^3} \tag{1}$$

where ELCR is excess lifetime cancer risk (dimensionless); C_{f} is heavy metal concentration in tilapia (μg g^{-1}); $\mathrm{CSF_o}$ is the oral carcinogenic slope factor from the IRIS (Integrated Risk Information System, provided by USEPA) database (mg kg^{-1} $\mathrm{day}^{-1})^{-1}$; $\mathrm{BW_a}$ is the adult body weight (kg); IR is the tilapia ingestion rate (g day^{-1}); $\mathrm{EF_r}$ is the exposure frequency (350 days year^{-1}); $\mathrm{ED_{tot}}$ is the exposure duration (30 years); $\mathrm{AT_c}$ is the average human lifespan of 70 years (i.e., $\mathrm{AT_c}$ = 365 days year^{-1} × 70 years = 25,550 days); and 10^3 is the unit conversion factor.

The noncancer risk was estimated using the HQ approach in adults, defined as:

$$\mathrm{HQ} = \frac{C \times \mathrm{IR} \times \mathrm{EF_r} \times \mathrm{ED_{tot}}}{\left(\mathrm{RfD_o} \times \left(\frac{\mathrm{BW_a}}{70\,\mathrm{kg}}\right)^{\frac{1}{3}}\right) \times \mathrm{BW_a} \times \mathrm{AT_{nc}} \times 10^3} \tag{2}$$

where HQ is the toxicity hazard quotient (dimensionless); $\mathrm{RfD_o}$ is the oral reference dose from the IRIS database (mg kg^{-1} day^{-1}); $\mathrm{AT_{nc}}$ is the average exposure duration of 30 years (i.e., $\mathrm{AT_{nc}}$ = 365 days year^{-1} × 30 years = 10,950 days). We treated C_{f}, $\mathrm{BW_a}$, and IR in Eqs. (1) and (2) probabilistically.

Data on C_{f} were obtained in September 2007 at the SK, LJ, and CP sites. Probabilistic distributions of C_{f} were fitted to the field observations obtained from each tilapia farm. The selected lognormal distributions had an acceptable χ^2 fit and Kolmogorov–Smirnov (K–S) fit (Fig. 1). Distributions of the estimated $\mathrm{BW_a}$ of Taiwanese adults were fitted to the data obtained from the Bureau of Health Promotion, Department of Health (2008). Herein, Taiwanese adults were defined as individuals ranging in age from 18 to 85 years. The estimated $\mathrm{BW_a}$ of Taiwanese adults was LN(61.62 kg, 1.12), and the selected lognormal distributions had optimal χ^2 and K–S goodness-of-fit. The estimated IR of N(65.76 g day^{-1}, 22.63) was adopted from the Bureau of Health Promotion, Department of Health (2008), and the selected normal distributions had optimal χ^2 and K–S goodness-of-fit.

In risk assessment, there are several sources of uncertainty. Due to inherent natural variability, equation variables can be defined in terms of a probability density function derived from a limited number of observations. The software program Crystal Ball® (Version 7.3, Decisioneering, Inc., Denver, CO, USA) was used to analyze data and to estimate distribution parameters. The selected distribution type was based on statistical criteria. To explicitly account for this uncertainty/variablity and its impact on the estimation of ELCRs and HQs, a Monte Carlo simulation was adopted. To test the convergence and the stability of the numerical output, we performed independent runs at 1, 4, 5, and 10 thousand iterations with each parameter sampled independently from the appropriate distribution at the start of each replicate. Inputs were assumed to be independent, largely because of limitations in the data used to derive model parameters. The result showed that 10,000 iterations were sufficient to ensure the stability of results.

Results and Discussion

Since one of the purposes of our study is to compare the heavy metal concentrations in fish farmed in highly industrialized areas with fish farmed at a reference site, we

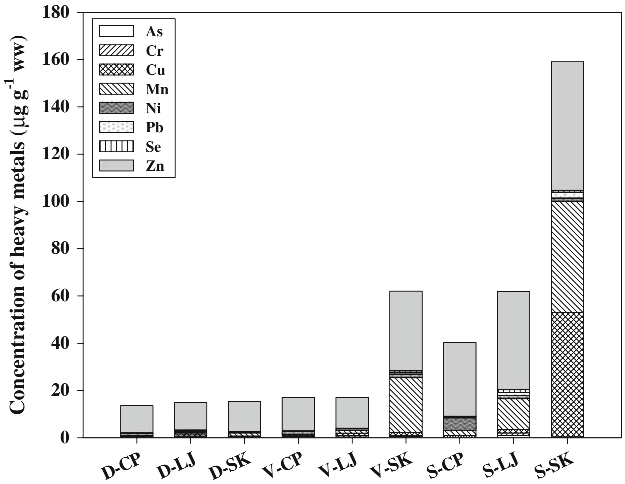

Fig. 2 The mean concentrations of heavy metal in tilapia dorsal muscle, ventral muscle and scale at Chu-Pei, Long-Jing and Shen-Kang (*D*: Dorsal muscle; *V*: Ventral muscle; *S*: Scale; *CP*: Chu-Pei; *LJ*: Long-Jing; and *SK*: Shen-Kang)

collected samples of fish of approximately the same age (9–12 months) and similar size (20 ± 2 cm) to minimize the variation from endogenous factors. Thompson et al. (2000) indicated that the lognormal distribution is often considered the default in environmental analysis. Distributions were fit to heavy metal contents in tilapia dorsal muscle, ventral muscle, and scales and the selected lognormal distributions had acceptable χ^2 fit and K–S fit in that optimizations using either statistics yielded geometric mean (gm) and geometric standard deviation (gsd) expressed as LN (gm, gsd) (Fig. 1). Figure 2 shows the mean concentrations of heavy metals of eight analyzed trace elements in tilapia dorsal muscle, ventral muscle, and scales collected in SK, LJ, and CP. The concentrations of almost all of the toxic elements analyzed in this study were found to be higher in the SK and LJ samples than in CP samples. The result shows that tilapia farmed sites in polluted areas have higher concentrations of heavy metals in their body tissue than tilapia farmed at sites in relatively clean areas. In Fig. 1, the wind rose map shows that the dominant wind direction in LJ and SK counties was from north to south (27%) during 2001–2005. This supports our finding that the fisheries located south of the CFPP and ISMP where the prevailing winds were directed had the highest level of heavy metal concentrations. Figures 1 and 2 also indicate that the heavy metal concentrations in tilapia scales are higher than those in tilapia muscle.

A comparison with other studies worldwide is summarized in Table 1. The concentrations of heavy metal in tilapia in our study were significantly higher than those tilapia analyzed in Egypt (Rashed 2001) and Vietnam (Wagner and Boman 2003). This indicates that fish ponds located in polluted areas can result in higher concentrations of metal in fish tissue. Concentrations of As, Cu, Ni, and Se in tilapia muscle were found to be about the same as those observed in Lin's study (Lin et al. 2005b); however, concentrations of Mn, Ni, Pb, and Zn in fish muscle in our study were significantly or relatively higher than those in their study. Aunela-Tapola et al. (1998) investigated trace metal emissions from a coal-fired power plant. Their results showed remarkably high concentrations of toxic metals in the flue gases (e.g., Pb, Zn, Mn, and As) and clear accumulation of Pb, Cd, Zn, Tl, and As on the fly ash. In our study, the concentration of these elements in tilapia muscle were also high. These findings indicate that CFPP or ISMP can be a source of contamination. In Table 1, we found an interesting phenomenon. In our study, the concentrations of all the heavy metal elements were higher in ventral muscle than in dorsal muscle. This indicates that when conducting a risk assessment for consumption of contaminated fish, the variation can be significantly different from the true risk, if the part of the fish muscle in which concentrations will be analyzed are not specified.

A box-and-whisker plot represents the uncertainty in comparing ELCRs and HQs. Figures 3a and b depict the box plots of interquartile and 50th percentile prediction-associated whisker plots indicating 5th percentile and 95th percentile predictions of ELCRs and HQs for consumption of tilapia muscle among residents who live in the vicinity of industrial parks. The *x*-axis represents fish farms in LJ and SK, respectively, whereas the *y*-axis shows ELCRs and HQs resulting from different fish farms. Under most regulatory programs, an ELCR exceeding 1×10^{-6} and an HQ exceeding 1 indicate potential risk. The predicted 95th percentiles of ELCR were 1.85×10^{-5} for SK and 1.03×10^{-5} for LJ, whereas those of HQ were estimated as 9.2 for SK and 3.1 for LJ. Figures 3a and b show that all 95% probability ELCRs or HQs are larger than 1×10^{-6} or larger than 1, indicating high potential health risks. The 95th percentile ELCR or HQ for SK is higher than that for LJ. Figure 3c shows the stacked column chart compositions of heavy metal contributions of HQ in LJ and SK. The predicted 95th percentiles of the ELCRs and HQs for residents consuming contaminated tilapia located in the vicinity of industrial parks were found to be higher than those for residents consuming tilapia farmed at the non-contaminated fishery.

In summary, the fisheries located south of the CFPP and ISMP where the prevailing winds were directed had the highest level of heavy metal concentrations. The results indicate that the tilapia farmed sites in polluted areas have higher concentrations of heavy metals in their body tissue than tilapia farmed at sites in relatively clean areas. According to our finding, the concentrations of all the heavy metal elements in tilapia scales are higher than those

Table 1 Heavy metal concentrations (mean ± SD) in fish tissues

Fish species	Location	Tissues	N[a]	As	Cr	Cu	Mn	Ni	Pb	Se	Zn
Tilapia (This study)	CP	Scale (μg g^{-1} ww)	4	0.04 ± 0.02	0.12 ± 0.09	0.78 ± 0.22	2.26 ± 0.24	5.19 ± 4.68	0.56 ± 0.29	0.17 ± 0.09	31.19 ± 6.96
	LJ		4	1.05 ± 0.50	0.98 ± 0.72	1.48 ± 0.58	13.16 ± 12.00	1.12 ± 0.52	1.32 ± 0.77	1.39 ± 0.54	41.43 ± 19.38
	SK		4	0.31 ± 0.13	0.27 ± 0.08	52.72 ± 102.28	47.03 ± 19.65	1.32 ± 0.95	2.48 ± 2.37	0.85 ± 0.04	54.32 ± 28.68
	CP	Dorsal muscle (μg g^{-1} ww)	4	0.08 ± 0.09	0.26 ± 0.08	0.32 ± 0.06	0.27 ± 0.10	0.86 ± 0.47	0.04 ± 0.04	0.17 ± 0.04	11.59 ± 4.80
	LJ		4	0.29 ± 0.20	0.41 ± 0.08	0.92 ± 0.59	0.51 ± 0.29	0.10 ± 0.02	0.51 ± 0.13	0.52 ± 0.11	11.65 ± 5.94
	SK		4	0.23 ± 0.09	0.16 ± 0.05	0.35 ± 0.13	1.26 ± 1.09	0.20 ± 0.16	0.08 ± 0.06	0.30 ± 0.07	12.80 ± 8.25
	CP	Ventral muscle (μg g^{-1} ww)	4	0.10 ± 0.06	0.35 ± 0.29	0.37 ± 0.03	0.55 ± 0.34	1.26 ± 1.20	0.05 ± 0.03	0.19 ± 0.04	14.19 ± 6.33
	LJ		4	0.35 ± 0.13	0.60 ± 0.17	1.04 ± 1.59	0.95 ± 1.21	0.13 ± 0.05	0.67 ± 0.25	0.30 ± 0.21	13.03 ± 10.12
	SK		4	0.64 ± 0.14	0.24 ± 0.03	1.48 ± 0.32	23.14 ± 1.03	1.18 ± 0.28	0.75 ± 0.35	0.86 ± 0.50	33.76 ± 5.55
Tilapia	Egypt[b]	Scale (μg g^{-1} dw)	50	–	0.304 ± 0.054	0.384 ± 0.038	0.314 ± 0.086	0.244 ± 0.066	–	–	1.86 ± 0.59
Tilapia	Egypt[b]	Muscle (μg g^{-1} dw)	50	–	0.078 ± 0.032	0.260 ± 0.084	0.026 ± 0.020	0.062 ± 0.021	–	–	0.630 ± 0.062
Tilapia	Taiwan[c]	Muscle (μg g^{-1} ww)	55	0.319 ± 0.223	0.597 ± 0.384	1.344 ± 1.336	0.450 ± 0.467	0.141 ± 0.084	0.184 ± 0.188	0.423 ± 0.061	10.787 ± 3.354
Freshwater fish	Vietnam[d]	Muscle (μg g^{-1} ww)	10	<0.1	0.3	1.8 ± 0.7	2.6 ± 1.0	0.3	<0.25	1.9 ± 0.7	29 ± 6
			10	<0.1	0.3	1.8 ± 0.3	3.6 ± 1.1	0.3	<0.25	1.5 ± 0.4	31 ± 8
Tilapia	China[e]	Muscle (μg g^{-1} ww)	3	0.65 ± 0.84	0.15 ± 0.06	0.19 ± 0.07	–	0.13 ± 0.11	0.29 ± 1.0	–	8.61 ± 1.14
			3	2.24 ± 0.27	0.27 ± 0.10	2.15 ± 0.36	–	–	2.15 ± 0.36	–	–
			3	0.78 ± 0.33	1.57 ± 0.12	2.42 ± 0.23	–	–	2.42 ± 0.23	–	–
			3	0.42 ± 0.52	2.63 ± 0.41	4.73 ± 0.29	–	–	4.73 ± 0.29	–	–
			3	1.29 ± 0.17	0.10 ± 0.04	7.74 ± 0.39	–	–	7.74 ± 0.39	–	–
			3	1.84 ± 0.98	0.22 ± 0.23	0.55 ± 0.10	–	–	0.55 ± 0.10	–	–

[a] Number of fishes

[b] Rashed (2001)

[c] Lin et al. (2005a)

[d] Wagner and Boman (2003)

[e] Cheung et al. (2008)

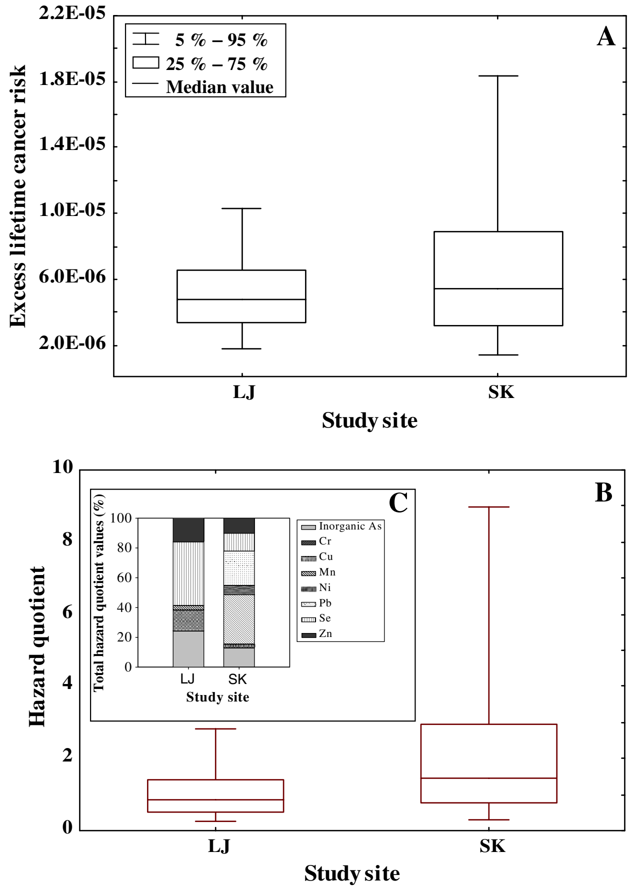

Fig. 3 Box-and-whisker plot representations of inorganic As, Cr, Cu, Mn, Ni, Pb, Se, Zn-induced **a** excess lifetime cancer risk and **b** hazard quotient for residents consumption of tilapia muscle in LJ (Long-Jing) and SK (Shen-Kang). **c** Stacked column chart compositions of heavy metal contributions of hazard quotient in LJ and SK

in tilapia muscle. The heavy metal concentrations were higher in ventral muscle than in dorsal muscle. The finding indicates that when conducting a risk assessment for consumption of contaminated fish, the parts of the fish muscle in which concentrations will be analyzed are specified. If used in a realistic fashion, it can more fully inform the decision-making process for the management of contaminated fish by providing a quantitative expression of the confidence in risk estimates. The ELCRs and HQs for residents consuming contaminated tilapia located in the vicinity of industrial parks were found to be higher than those for residents consuming tilapia farmed at the non-contaminated fishery. In conclusion, tilapia have certain nutrients good for health, however, heavy metals in tilapia muscles may poses significant risk to residents in the vicinity of a industrial complex.

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