

# Portable X-ray Fluorescence as a Rapid Determination Tool to Detect Parts per Million Levels of Ni, Zn, As, Se, and Pb in Human Toenails: A South India Case Study

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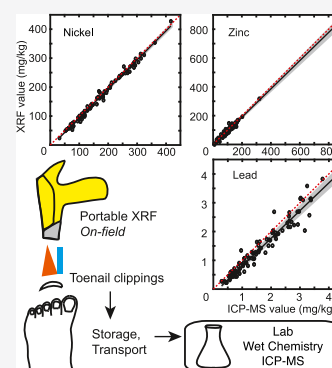
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Supporting Information

**ABSTRACT:** Chronic exposure to inorganic pollutants adversely affects human health. Inductively coupled plasma mass spectrometry (ICP–MS) is the most common method used for trace metal(loid) analysis of human biomarkers. However, it leads to sample destruction, generation of secondary waste, and significant recurring costs. Portable X-ray fluorescence (XRF) instruments can rapidly and nondestructively determine low concentrations of metal(loid)s. In this work, we evaluated the applicability of portable XRF as a rapid method for analyzing trace metal(loid)s in toenail samples from three populations ( $n = 97$ ) near the city of Chennai, India. A Passing–Bablok regression analysis of results from both methods revealed that there was no proportional bias among the two methods for nickel (measurement range  $\sim 25$  to  $420$  mg/kg), zinc ( $10$  to  $890$  mg/kg), and lead ( $0.29$  to  $4.47$  mg/kg). There was a small absolute bias between the two methods. There was a strong proportional bias (slope =  $0.253$ , 95% CI:  $0.027$ ,  $0.614$ ) between the two methods for arsenic (below detection to  $3.8$  mg/kg) and for selenium when the concentrations were lower than  $2$  mg/kg. Limits of agreement between the two methods using Bland–Altman analysis were derived for nickel, zinc, and lead. Overall, a suitably calibrated and evaluated portable XRF shows promise in making high-throughput assessments at population scales.

**KEYWORDS:** portable XRF, ICP–MS, toenails, high correlation



## 1. INTRODUCTION

Industrial pollution has increased contamination of the environment by toxic heavy metals.<sup>1,2</sup> Exposure pathways for inorganic pollutants include inhalation,<sup>3</sup> dermal, and dietary uptake.<sup>1</sup> Chronic exposure to heavy metals has been associated with neurodevelopmental, hormonal, reproductive, and gastrointestinal health effects in humans.<sup>4,5</sup> Here, we evaluate a method for rapid detection of multiple heavy metals and metalloids in human nails that can facilitate population-wide assessments in remote regions and countries with minimal experimental costs.

Blood, urine, hair, and nails are the most commonly used biomarkers for studying human metal(loid) exposure. Several studies have reported the utility of using toenails as a preferred biomarker.<sup>6–8</sup> Toenails provide a long-term integrated record of exposure (2–12 months) and can be collected non-invasively, leading to greater acceptability by volunteers.<sup>9–11</sup> Nails also often have higher concentrations of metal(loid)s compared to body fluids and tissues and can therefore be detected more easily.<sup>12</sup> Toenails are less prone to exogenous contamination such as bleaching, dyeing, and other cosmetic exposures that could potentially influence elemental exposure assessment.<sup>6,7</sup>

Metal(loid)s are conventionally quantified by inductively coupled plasma mass spectrometry (ICP–MS). ICP–MS is a robust and sensitive method but has a few limitations. The samples must be transported back to a laboratory and require acid digestion, which leads to sample destruction and production of secondary hazardous waste. The instrument requires meticulous operation to avoid contamination and issues with drift and interferences in order to obtain accurate and precise concentration results.

By contrast, portable X-ray fluorescence (XRF) instruments are mobile, do not require sample processing, and are nondestructive. They have potential to provide time-efficient, high-throughput, and cost-effective measurements.<sup>13,14</sup> In recent years, XRF has been extensively used as a rapid screening tool in geological explorations, locating hotspots of contamination, and for analysis of biological samples.<sup>15–19</sup>

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XRF has been shown to accurately determine concentrations of heavy metals and metalloids in model human nails/phantom nails, made of polyester or other commercial resins and solidifying agents dosed with known quantities of metal(loid)s, for zinc, selenium, arsenic, mercury, and chromium.<sup>20–24</sup> Validation with real nail clippings and comparison with ICP–MS has been performed for manganese and lead.<sup>17</sup> The results show a high correlation coefficient for manganese ( $r = 0.91$ ) but a much lower value for lead ( $r = 0.3$ ). This may have occurred because most of the sample concentrations were below the detection limit of the XRF for Pb (0.6 mg/kg).

The goal of this work was to evaluate the usability of XRF, in particular portable XRF, for reliably measuring multiple elements from human nails. We evaluated the capability of portable XRF to reliably quantify multiple elements (nickel, zinc, arsenic, selenium, and lead) in toenail samples by assessing the concentrations of these elements in 97 toenail samples determined by both portable XRF and ICP–MS.

## 2. MATERIALS AND METHODS

**2.1. Toenail Sample Collection and Preparation.** The overall study and its experimental protocol were approved by the Institute Ethics Committee of the IIT Hyderabad (IEC protocol no: IITH/IEC/2019/05/13). Toenails were collected from three communities, Sepakkam (13°16'37"N, 80°18'19"E), Athipattu Pudu Nagar (13°14'58"N, 80°17'54"E), and Avurivakkam (13°25'40"N, 80°16'19"E) situated 20–50 km from Chennai, Tamil Nadu, India. Sepakkam and Athipattu Pudu Nagar are situated in the Ennore suburb of Chennai. There are three power plants operating in the area: North Chennai Thermal Power Station (NCTPS, Capacity: 3 × 210 MW; 2 × 600 MW), Vallur thermal power plant (Capacity: 3 × 500 MW), and Ennore thermal power plant (Capacity: 2 × 60 MW; 3 × 110 MW). The two sites are located within 10 km of the power plants. Avurivakkam is located 37 km from Sepakkam and Athipattu Pudu Nagar. There was no industry in the vicinity but the community was located next to the Pulicat Lake, which is a brackish lagoon of area 759 km<sup>2</sup>. The volunteers filled a consent form and a survey was conducted to collect the sociodemographic information including details on age, gender, income, education, rice and fish consumption per week, duration of residence, profession and water source of the three communities (Table S1). Analysis of metal concentrations in nails and their relation to the potential sources at each of the three sites will be addressed in another work and is beyond the scope of this work.

Ninety-seven toenail samples were collected from three communities, 36 each from Sepakkam and Athipattu Pudu Nagar and 25 from Avurivakkam. The toenails were clipped from all the toes. The collected samples were stored in metal-free double zip lock bags and analyzed in a metal-free lab environment.

A nonionic detergent (Tween 20, Croda International PLC) was used to clean toenail samples prior to metal(loid) analysis using a 1% solution for 30 min in a sonicator. Tween 20 is polyoxyethylene sorbitan ester and is a solubilizing and emulsifying agent.<sup>25</sup> Samples with nail polish were cleaned with acetone before washing. The mixture was vortexed and the aqueous solution was removed. The samples were then soaked in 1% Tween 20 solution for a period of 2 days. After soaking, the aqueous solution was replaced with fresh 1% Tween 20 and sonicated for 30 min, followed by washing with

type 1 water (ultrapure water with resistivity >18 MΩ cm). The nails were then dried, weighed, and ready for portable XRF and ICP–MS analysis.<sup>17</sup>

**2.2. Portable XRF Analysis.** The portable XRF device used in this study was a customized ThermoNiton XL3t GOLDD+ (Thermo Fisher Scientific, Billerica, MA) handheld XRF instrument. The device operates at a 2 W power output, and we used settings of 50 kV and 40 μA with a silver filter. The toenail samples were measured by placing all clippings over the X-ray aperture for 3 min. The calibration and analysis followed methods used in previous studies and account for variability in toenail mass and thickness.<sup>17,24</sup> In brief, we utilized a competing process through Compton scattering in order to normalize the total size of clippings and any discrepancy in thickness of the toenails that may be influencing the measurements. The Compton scattering peak is primarily composed of characteristic X-rays generated from the X-ray anode and interacting with the sample before being collected at a slightly lower energy by the device's Si radiation detector. We fit the Compton scattering peak (~20 keV) and elemental peaks (Ni, Zn, As, Se, and Pb) using a Gaussian function with exponential background components to obtain net counts. Ni (7.48 and 8.26 keV), Zn (8.64 and 9.57 keV), As (10.54 and 11.73 keV), and Se (11.22 and 12.50 keV) were fitted using the K-alpha and K-beta lines, respectively. Pb was quantified using the fitting from the L-beta line of 12.61 keV. The L-alpha line at the 10.55 keV line was still fitted to remove the competing counts from the As 10.54 keV peak but not used in the quantification as this reduced the overall certainty of findings. Over all fittings, the average chi square goodness of fit parameter was  $1.2 \pm 2.6$  (494 degrees of freedom). The net elemental counts were then divided by the Compton scattering counts to normalize the results. Following our previous work,<sup>17,24</sup> we derived sample calibrations from lab-made epoxy resin toenail samples doped with concentrations from 0 to 50 mg/kg of Ni, Zn, As, Se, and Pb. These samples were made with compositions such that they mimicked nail properties for the purposes of X-ray absorption. Standards included differing thicknesses and clipping masses to accommodate variations in natural sample collection and allow for our calibration to include the potential uncertainty introduced during the normalization process for mass and thickness, as outlined in our previous work.<sup>17,24</sup>

**2.3. Inductively Coupled Plasma Mass Spectrometry.** An Agilent 7900 ICP–MS instrument was used for the analysis of toenail samples. It has a robust plasma and ultrahigh matrix introduction (UHMI) technology that enables the measurement of rare-earth metals and metalloids with high sensitivity. Dried toenails were digested with concentrated nitric acid (0.01%) at 100 °C for 1 h.<sup>6</sup> The digested solution was cooled at room temperature and diluted to 10 mL with type 1 water. The aqueous solution was then analyzed using ICP–MS.<sup>6</sup>

Matrix blanks were prepared by replacing the sample with type 1 water. The method detection limit (MDL) was determined as follows

$$\text{MDL} = t_c \times \text{SD} \quad (1)$$

where  $t_c$  is 3.143 [the critical value obtained from a  $t$ -test at a confidence level of 0.01 and a degree of freedom of 6 (i.e.,  $n - 1$ , where  $n$  is the sample size)]. One matrix blank was run 7 times, and the standard deviation (SD) was calculated. The reported sample analyte concentrations were a minimum of 2.5 times higher than the MDL.<sup>26</sup> The multielemental standard

used for analysis was Elemental Scientific (ESI) control standard solution #26. Stock solutions were prepared with concentrations ranging from 10 to 200  $\mu\text{g/L}$  for all metal(loid)s.

**2.4. Statistical Analysis.** Two analyses were used to compare the results obtained from XRF and ICP–MS: Passing–Bablok regression<sup>27–29</sup> and Bland–Altman.<sup>30–32</sup> Passing–Bablok regression is a nonparametric method that does not require normality of data obtained from the two methods and is robust in the presence of outliers. The method assumes that there is a linear relationship between the two sets of values. This was checked using the CUSUM test. The method fits the intercept and slope of a linear regression,  $y = a + bx$ , and determines the respective 95% confidence intervals (CIs).<sup>29</sup> If zero is not in the CI for  $a$ , there is a systematic difference (bias) between the two methods; if unity (one) is not in the CI for  $b$ , there is a proportional difference (bias) between the two methods. A reasonable sample size for Passing–Bablok analysis has been suggested to be 50.<sup>33</sup>

Bland–Altman analysis is also used to compare two different methods. It calculates the mean difference (bias) and a 95% CI between which the differences of the two methods would fall, thereby constructing the limits of the agreement. If  $y_1$  is a particular measurement value from the first method and  $y_2$  is the corresponding measurement value from the second method, a mean value of the two is calculated as  $(y_1 + y_2)/2$  and the difference as  $(y_1 - y_2)$ . Then, differences in all sets of measurement values are plotted against all the corresponding mean values. The mean bias between the two methods was calculated as

$$\bar{y} = \frac{1}{n} \sum_{k=1}^n (y_1 - y_2)_k \quad (2)$$

95% limits of agreement are calculated as mean bias plus or minus 1.96 times the SD of the differences. If in a future analysis it is decided that the expected differences in measurement values between the two methods fall within the 95% limits of agreement, then the methods can be used interchangeably. This analysis assumes that the differences of values obtained from the two methods are normally distributed.

Pearson's correlation coefficient ( $r$ ) was calculated only as a means to assess the linearity between the concentrations of each metal(loid) measured by XRF and ICP–MS and the deviation from the expected ideal 1:1 relation using IBM SPSS statistics 20. It may be noted that the correlation coefficient does not represent an agreement of the methods.<sup>32</sup> Calculation of  $r$  assumes that the values are normally distributed and do not contain outliers. The linear regression model was used to estimate beta, intercept, and variance at the 95% CI. All obtained values were retained for all the statistical analyses.<sup>34</sup>

### 3. RESULTS AND DISCUSSION

**3.1. Calibration and Quantification.** The calibration results of ICP–MS for target metal(loid) ions, nickel, zinc, arsenic, selenium, and lead are given in Figure 1. The calibration curve has a regression coefficient of more than 0.99 for the metal(loid)s selected. The MDLs are given in Table 1. The residual standard deviation (RSD) obtained from multiple analyses ( $n = 3$ ) of each of the standard solution concentrations used for calibration for each metal(loid) was less than 4% for all metal(loid)s for all concentrations.

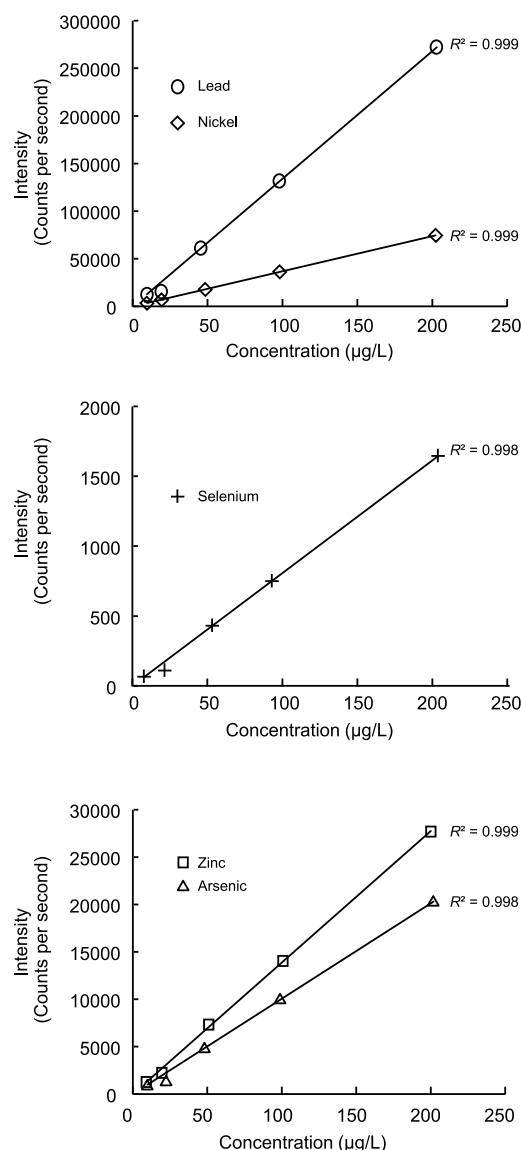


Figure 1. Calibration curves for ICP–MS.

Table 1. QA/QC and Method Detection Limits for ICP–MS for Target Metals

metal ions	method blanks ( $n = 7$ )		method detection limit (ppb, $\mu\text{g/kg}$ ) <sup>a</sup>	standard recovery (%) ( $n = 7$ )	$R^2$
	mean	SD			
Ni	1.62	0.923	2.90	97.5 $\pm$ 2.23	0.999
Zn	3.20	3.21	10.1	98.6 $\pm$ 4.25	0.999
As	0.248	0.058	0.181	99.02 $\pm$ 2.14	0.997
Se	0.837	0.316	0.994	97.8 $\pm$ 11.3	0.997
Pb	1.21	0.213	0.670	96.2 $\pm$ 3.85	0.998

<sup>a</sup>Method detection limit calculated as 3 times the SD.

XRF analysis was calibrated using nail phantoms as described by Specht et al.,<sup>17</sup> and the calibration curves for each metal(loid) are given in Figure 2. A sample spectrum is presented in Supporting Information Figure S1. The Ni calibration has a y-intercept due to inherent Ni in the XRF device collimation, but this should be accounted for in our quantification. The MDLs are given in Table 2. The detection

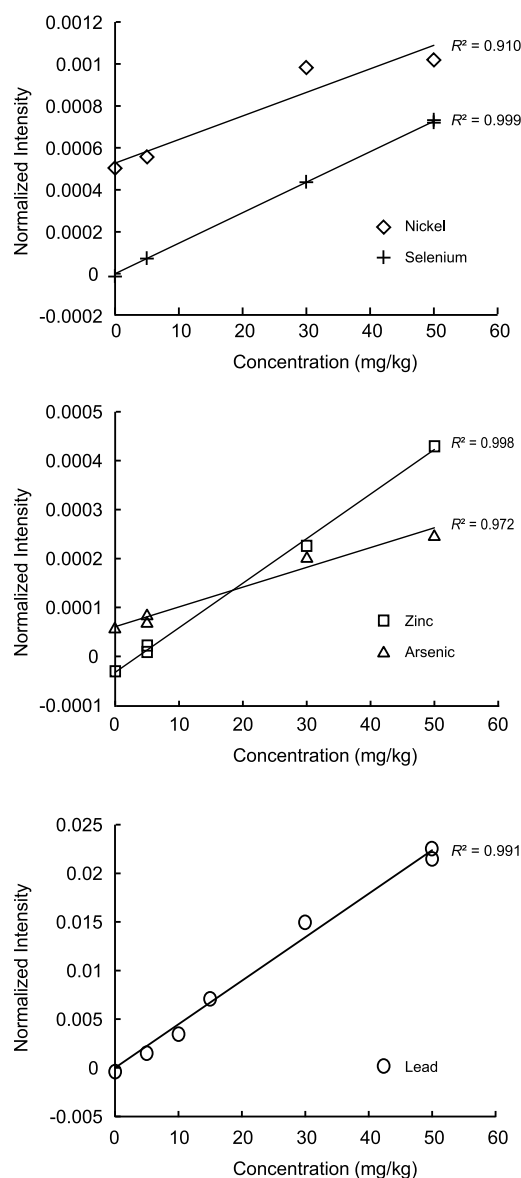


Figure 2. Calibration curves for XRF.

limit of ICP–MS is much lower than that of XRF and should be considered prior to exposure assessment.

**3.2. Correlation between XRF and ICP–MS Data.** The measured metal(loid) concentrations from the two methods are given in Table 3 (overlapping histograms in Supporting Information Figure S2 and in Supporting Information section S1 for individual sites), the parameters obtained from the Passing–Bablok regression are presented in Table 4 and Figure

Table 2. Method Detection Limits for XRF

metals	XRF method detection limit (ppm, mg/kg) <sup>a</sup>	$R^2$
Ni	3.53	0.910
Zn	2.87	0.998
As	1.27	0.972
Se	4.60	1.00
Pb	0.58	0.991

<sup>a</sup>Calculated from the calibration line of the XRF. This limit calculated from calibration parameters can be dependent on sample properties such as mass and composition, which vary from sample to sample.

Table 3. Mean, SD, Maximum, Minimum, and Median Concentrations (mg/kg) Corresponding to Each Metal Measured in Toenails

concentrations	nickel			zinc			arsenic			selenium			lead		
	ICP–MS (mg/kg)	XRF (mg/kg)		ICP–MS (mg/kg)	XRF (mg/kg)		ICP–MS (mg/kg)	XRF (mg/kg)		ICP–MS (mg/kg)	XRF (mg/kg)		ICP–MS (mg/kg)	XRF (mg/kg)	
mean	163	156		113	105		1.19	0.702		6.54	5.62		1.64	1.44	
S.D.	83.9	84.5		90.8	90.7		0.83	0.636		16.6	15.6		0.998	0.971	
median	139	133		102	95.6		1.11	1.04		0.775	0.369		1.40	1.23	
maximum	417	427		893	887		3.80	1.56		92.9	90.2		4.47	3.97	
minimum	31.8	23.9		9.35	BDL <sup>a</sup>		0.006	BDL		0.019	BDL		0.285	0.225	

<sup>a</sup>BDL = below detection limit.

**Table 4. Statistical Parameters from the Passing–Bablok Analysis of XRF and ICP–MS Measurements<sup>a</sup>**

element	<i>a</i> median (95% CI)	<i>b</i> median (95% CI)
Ni	−6.51 (−4.23, −9.56)	1.00 (0.985, 1.02)
Zn	−5.89 (−0.943, −9.22)	0.980 (0.930, 1.02)
Se	−0.029 (−0.045, 0.00)	0.928 (0.878, 0.945)
As	−0.057 (−0.227, 0.00)	0.253 (0.037, 0.614)
Pb	−0.160 (−0.092, −0.216)	0.962 (0.919, 1.00)

<sup>a</sup>Results are the parameters of the equation  $y = a + bx$ , where  $x$  is the ICP–MS measurement and  $y$  is the XRF measurement expressed as median and 95% CI. <sup>b</sup>For concentrations less than 2 mg/kg ( $n = 80$ ):  $a = -0.003$  (−0.029, 0.013),  $b = 0.835$  (0.694, 0.916). For concentrations greater than 2 mg/kg ( $n = 17$ ):  $a = -0.682$  (−1.73, 0.151),  $b = 0.980$  (0.933, 1.02).

3, the Bland–Altman analysis in Figure 4, and Pearson's correlation in Supporting Information (section S2). The values obtained from ICP–MS and XRF were normally distributed for nickel and lead. For zinc, once we removed the highest measured value (880 mg/kg, compared to a median of <105 mg/kg), the distribution became normal for both ICP–MS and XRF-measured values. For arsenic, the distribution was lognormal only for ICP–MS data (Supporting Information section S3, Table S6). The differences between the measurements taken from ICP–MS and XRF were normal for nickel, zinc, and lead. Therefore, Passing–Bablok regression analysis is applicable for all elements (the relationship between ICP–MS and XRF measurements was linear), Bland–Altman analysis for nickel, zinc, and lead, and the classic Pearson regression also for nickel, zinc, and lead.

The average nickel concentrations quantified by XRF and ICP–MS were 156 and 163 mg/kg, respectively. The 95% CI of slope  $b$  of the Passing–Bablok regression spans across 1 (median value of 1.00 with a 95% CI of 0.985 to 1.02), suggesting no proportional difference between the two methods across the entire measurement range. The 95% CI for  $a$  is between −4.23 and −9.56 with a median value of −6.51, suggesting a slight absolute bias in values obtained by the two methods. The regression results for individual sites presented in Supporting Information Tables S7–S9 are largely consistent with the overall results in Table 4. There is a larger range of 95% CI in bias and the 95% CI crossed zero for one of the sites, Athipattu Pudu Nagar, meaning that the XRF and ICP–MS analysis could be used interchangeably; however, the sample size was less than 50<sup>33</sup> and this interchangeability inference may be an artifact of a somewhat smaller sample size. The results were closer for other sites, and there was no indication of site-specific performance. Bland–Altman analysis shows that the mean estimated bias is −7.04 mg/kg, consistent with the absolute bias determined from the Passing–Bablok analysis, and the 95% limits of agreement were 8.58 and −22.6 mg/kg (Figure 4). Linear regression analysis also showed a high correlation (Pearson's correlation coefficient  $r = 0.996$ ,  $R^2 = 0.991$ , slope  $\approx 1$ ; Supporting Information Table S5 and Figure S2).

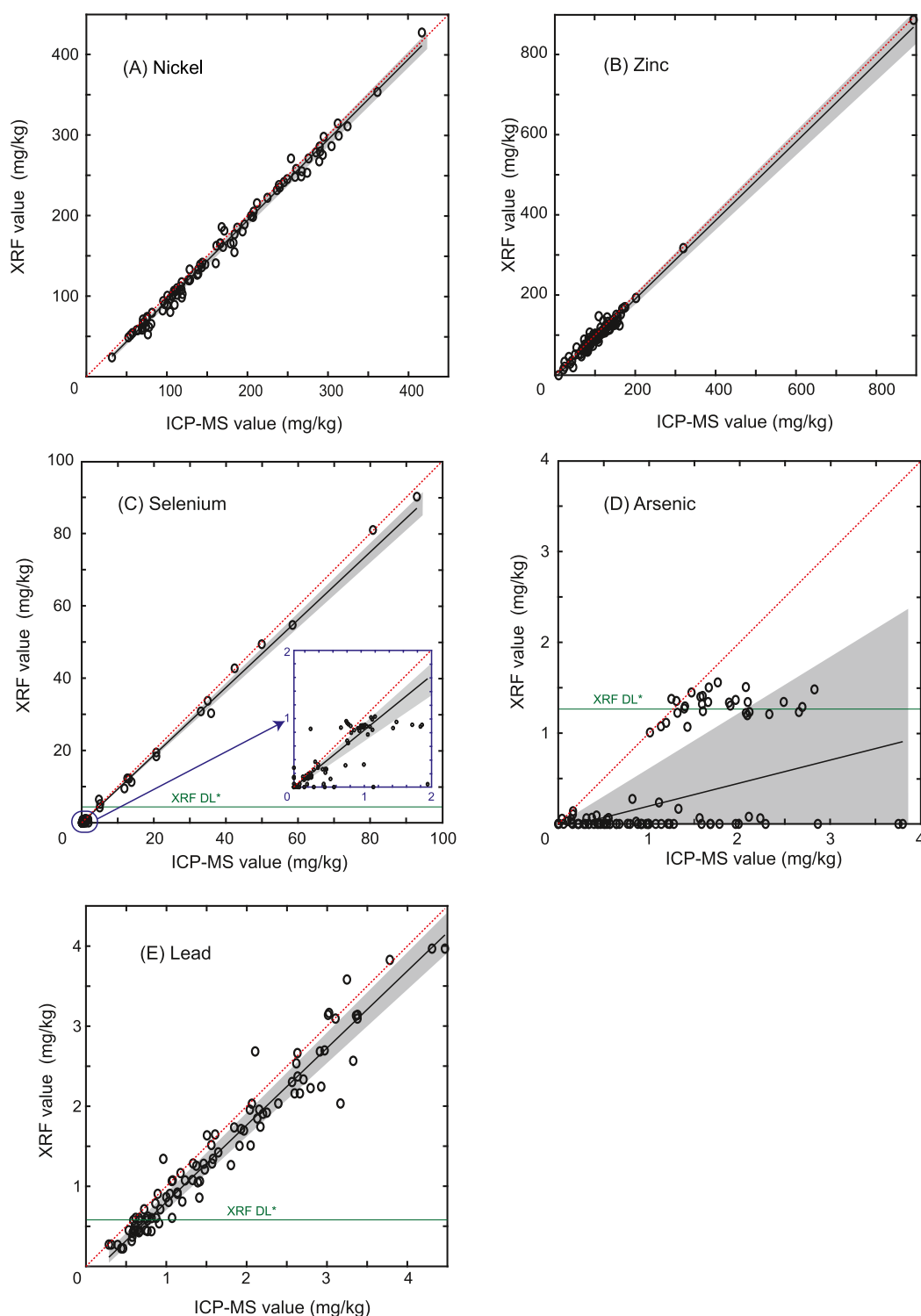
The widest concentration range was detected for zinc (XRF = 0–887 mg/kg and ICP–MS = 9.35–893 mg/kg). Most zinc concentrations were below 200 mg/kg. Passing–Bablok analysis revealed no proportional difference [95% CI for slope  $b$  spanned across (1)] between the two methods, but there was a small systematic difference (Table 4). The results from individual sites (Supporting Information Table S7–S9)

were similar but with a larger 95% CI, and XRF and ICP–MS were in agreement for Athipattu Pudu Nagar and Sepakkam, and there was a small absolute bias for Avurivakkam. The overall bias estimated from Bland–Altman analysis was −7.29 mg/kg with a 95% limit of agreement of −28.4 mg/kg and 13.8 mg/kg (Figure 4). The concentrations determined by XRF and ICP–MS were also highly correlated in the linear regression analysis ( $r = 0.993$ ,  $R^2 = 0.986$ , and slope = 0.992).

The average concentrations of selenium determined by XRF and ICP–MS were 5.62 and 6.54 mg/kg, respectively. Passing–Bablok analysis showed a regression slope of 0.928 (95% CI: 0.878 to 0.945) and no absolute bias −0.029 mg/kg (95% CI: −0.045 to 0.00) when the entire range of values was considered. However, the majority of samples ( $n = 80$ ) had values below 2 mg/kg. When we removed all values greater than 2 mg/kg, the regression slope decreased to 0.835 (95% CI: 0.694 to 0.916) and the intercept to −0.003 (95% CI: −0.029 to 0.013 mg/kg). This suggests a proportional difference in measurements made by the two methods and a no absolute bias at low concentrations. When we only considered values above 2 mg/kg ( $n = 17$ ), the 95% CI for the regression slope and intercepts contained unity and zero, respectively. The proportional difference was more pronounced for Sepakkam (Table S7, slope of 0.628 with a 95% CI of 0.484 to 0.881) probably because the lower concentrations of selenium detected in its population by the XRF, and the 95% CI of slope and bias were close to one and zero for Avurivakkam, likely in part due to the higher concentrations measured by the XRF (Supporting Information Table S4 and Figure S5). Spearman's correlation, when used just to assess the deviation from a 1:1 line, was 0.999 ( $R^2 > 0.998$  and slope = 0.970) when the entire range of values was considered but fell to  $r = 0.744$  ( $R^2 = 0.554$  and slope  $\approx 0.577$ ) when we consider values less than 2 mg/kg.

Arsenic concentrations varied between the below detection limit (BDL) and 3.80 mg/kg with an average concentrations of 0.702 mg/kg and 1.19 mg/kg measured by XRF and ICP–MS, respectively. The Passing–Bablok slope was 0.253 (95% CI: 0.037, 0.614), and the intercept was −0.057 (95% CI: −0.227, 0.00), suggesting no absolute bias but a large proportional bias between the two methods. Both slope and intercept parameters were zero for Avurivakkam because arsenic concentration in all except 3 of the 25 toenail samples was recorded as zero by XRF (see Supporting Information Figure S6). The performance difference seems to be related to the ability of XRF to measure low values rather than the actual difference in concentrations among the sites. This is likely compounded by the difficulty in fitting algorithms to address the difference between lead and arsenic peaks, which in our case has raised the detection limit for arsenic and potentially made measured concentrations in some of these nails lower than the limit of detection for the device. Spearman's correlation, when used just to assess the deviation from a 1:1 line, was weak ( $r = 0.491$ ,  $R^2 = 0.241$ , and slope = 0.354).

Contrary to what was observed for selenium and arsenic, measurements of lead from the two methods showed no proportional difference ( $b = 0.963$ , 95% CI: 0.919 to 1.001) and a slight absolute bias of −0.160 (−0.092 and −0.216) (Table 4), even though the values were always lower than 4.5 mg/kg with a mean concentration of 1.44 mg/kg. The parameter values for individual sites were largely consistent with the parameters in Table 4 for all sites combined. The mean absolute bias from the Bland–Altman analysis was

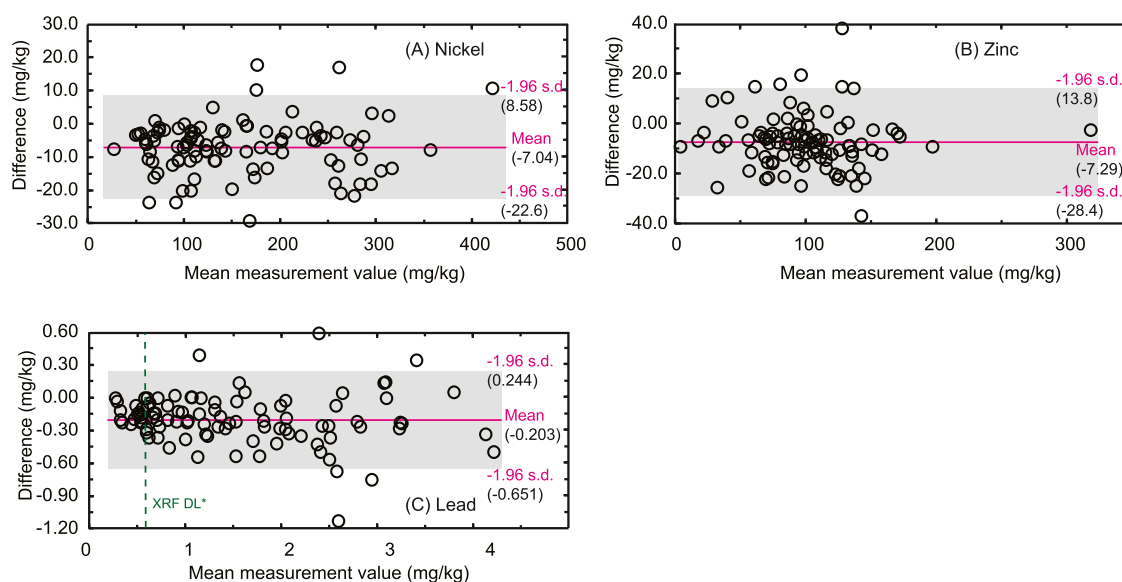


**Figure 3.** Passing–Bablok regression curve. Dotted red line is the 1:1 line. Shaded band represents the 95% CI of the slope. The solid green line denoting XRF DL\* represents the detection limit of XRF calculated from its calibration parameters.

−0.203 mg/kg with 95% limits of agreement of −0.651 and 0.244 mg/kg (Figure 4). Spearman's correlation was high ( $r = 0.974$ ,  $R^2 = 0.949$ , and slope = 0.945).

The portable XRF used in our work was capable of making very low-level lead measurements because lead has the lowest elemental detection limit with the XRF methodology used in this study. XRF calibration plays a significant role in the accuracy and precision of the measured elemental concen-

tration. There are literature reports on various calibration methods used for improving metal(loid) quantification.<sup>20,21</sup> The portable XRF used in the study was calibrated by fitting the Compton scattering peak against the standard nail phantoms doped with known elemental concentrations. Secondary absorption or enhancement of target wavelengths by atoms of other elements is one of the most important sources of bias in XRF measurements.<sup>14</sup> Fluoresced character-



**Figure 4.** Limits of agreement of the two methods obtained using Bland–Altman analysis. Limits are represented as the shaded region (s.d. = standard deviation). The vertical dotted green line denotes the detection limit of XRF determined from the calibration parameters (XRF DL\*).

istic X-ray is absorbed by another atom, and this atom may potentially generate additional X-rays. This may become problematic at low concentrations of the target analyte. The limitations of XRF are thus all focused on the detection capabilities, which can be increased using a higher-powered device with a longer measurement time or both. The detection limits can be further improved by calibration using a higher-power device and increased measurement time.<sup>17,24</sup>

Portable XRF has been widely used for elemental analysis, but limited studies have been conducted on validation of its efficacy with respect to ICP–MS for human sampling. A recent study has reported a high correlation between the portable XRF and ICP–MS values for manganese but less correlation for lead in nails due to lower concentrations.<sup>17</sup> A study on arsenic concentration of dried baby shrimp<sup>35</sup> stated a high correlation between arsenic concentrations quantified by portable XRF and ICP–MS. The lowest concentration reported in that study was 4 mg/kg, but in the present study, the concentrations were much lower than 2 mg/kg. These findings suggest that portable XRF may perform better as a quantification tool for arsenic concentrations higher than 2 mg/kg. A combination of XRF and ICP–MS techniques has been implemented for evaluation of elements accumulated in leaves due to dust (high-resolution ICP–MS) and in various parts of *Couroupita guianensis* required for soil nutrient reclamation, respectively.<sup>36,37</sup> Laser ablation (LA)-ICP–MS and XRF have been used for discriminating the overall composition of document paper.<sup>38</sup> XRF techniques are also widely used for sediment metal(loid) analysis.<sup>39</sup>

It has been previously reported that toenail arsenic concentrations are significantly correlated with urinary arsenic and total-body arsenic levels.<sup>40</sup> Although toenails have not been validated as biomarkers for arsenic, they provide reliable measure of arsenic exposure.<sup>41</sup> Selenium and zinc concentrations in toenails have reproducibility for long-term exposure.<sup>7</sup> Salcedo-Bellido et al.<sup>42</sup> have concluded that 7–12 months of nickel and lead exposure can be detected in the toenails. The purpose of the presented work was to evaluate the usefulness, and possible limitations, of portable XRF in measuring metal(loid) concentrations in human nails. The

results can be interpreted considering information obtained from such aforementioned exposure-biomarker studies.

Overall, we find that portable XRF can be used as an efficient and a high throughput tool to detect exposure of humans to metal(loid)s. Passing–Bablok regression of the measurements of nickel, zinc, and lead suggested that XRF and ICP–MS analyses methods had no proportional bias and a small absolute bias, respectively. The two methods could be used interchangeably for these elements. Limits of agreement from Bland–Altman analysis are presented and may be used to assess what limits will be acceptable in future studies. Future applications of the XRF device for mass screening of humans in potentially high-exposure settings, such as occupational exposure in metal(loid) industries or residential exposure in vicinities of high-metal(loid) emitting industries, would take advantage of the convenient new measurement methodology.

## ■ ASSOCIATED CONTENT

### SI Supporting Information

The Supporting Information is available free of charge at <https://pubs.acs.org/doi/10.1021/acs.est.1c00937>.

Example spectra from XRF, demographic variables from the population, distribution comparisons by the method, tables of summary data by site, detailed correlation results, detailed statistical test results, Bablok regression results, and box plots by site (PDF)

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## Notes

The authors declare no competing financial interest.

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