



Risk assessment of heavy metals in rooibos (*Aspalathus linearis*) tea consumed in South Africa

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Abstract

A total of 80 rooibos tea samples from a range of brands were purchased from various registered retail shops in South Africa. The samples were bought during 2019 winter (40) and summer (40) period which are classified as 6 natural rooibos, 18 herbal rooibos samples, and 16 flavor rooibos samples and subjected for heavy metal analysis such as chromium (Cr), iron (Fe), arsenic (As), cadmium (Cd), and lead (Pb) using inductively coupled plasma mass spectrometer (ICP-MS). Human health risks were determined by estimating the daily intake non-cancer hazard quotient (THQ) and hazard index (HI) via oral exposure to toxic elements based on daily tea consumption. The concentration range of the determined heavy metals in rooibos tea samples were as follows: Cr (0.17–11.98 mg/kg), Fe (31–450 mg/kg), As (ND–0.51 mg/kg), Cd (0.09–0.17 mg/kg), and Pb (0.06–2.73 mg/kg). Cr was found in higher amount when compared to the World Health Organization (WHO) permissive limit (1.3 mg/kg). The concentrations of all studied heavy metals during winter and summer period were compared using two-way Anova, and no significant differences ($p = 0.832$) were observed for the two seasons. Both the target risk quotient (THQ) and the hazard index (HI) levels in all analyzed tea were well below 1, implying that intake of rooibos tea with analyzed heavy metals should not cause a threat to human health. On the other hand, the continuous intake due to the high concentrations of trace metals such as Cr may pose a serious chronic health risk due to accumulation in body tissues over time. The study, therefore, suggests constant monitoring of these heavy metals in teas in order to limit the risk of exceeding the permissive limits.

Keywords Health risk assessment · Heavy metals · ICP-MS · Rooibos tea · South Africa

Introduction

Over the past few years, tea has become the world's most popular and consumed functional beverage around the world because of its smell, aroma, taste, and diversity and also because of its several stimulating health effects (Ipeaiyeda and Dawodu 2011; Salahinejad and Aflaki 2010).

The Food and Agriculture (FAO) reported that about 4.8 million tons of tea in 2013 were consumed globally (Chang and Atici 2015); meanwhile, many medicinal plants are consumed as herbal teas in South Africa for decades (Bhat and Moskovitz 2009; McGaw and Eloff 2008; van Wyk and

Gericke 2000), and estimates of the WHO indicate that about 75 per cent of world's population consume medicinal herbs as substitute for modern medicine (Owolabi et al. 2016). One of the most consumed herbal tea as reported in the literature is rooibos (Iswaldi et al. 2011; Jaganyi and Ndlovu 2001). Rooibos is an indigenous herbal plant usually rooted in the Cederberg mountainous region of South Africa's Western Cape Province according to the South African Rooibos Council (Iswaldi et al. 2011; Stander et al. 2019). Rooibos tea has many characteristics which make it intensively unique among other teas in South Africa. These unique characteristics include its medicinal properties against infantile colic, dermatological problems, allergies, asthma, and other gastrointestinal disorders such as heartburn and nausea (Joubert et al. 2008; van Wyk et al. 1997). Also, the protective effects against diseases associated with oxidative stress are partly due to the antioxidant components it contains.

For ages, contamination of beverages like any food material with toxic chemicals has been a major concern, and their consumption has been noted as one of the major routes of

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human exposure to these toxic chemicals (Coelho et al. 2016). Together with other pollutants, heavy metals are released into the atmosphere through industrial activities, automotive exhaust, heavy-duty power generators, and agricultural pesticides. Essential metals, including iron (Fe), copper (Cu), and zinc (Zn) (Ghuniem et al. 2019b), play a significant role in the body when present in trace elements to maintain a healthy body. However, other heavy metals, such as arsenic (As), cadmium (Cd), and lead (Pb), may pose serious health risk to human health if present in high quantity (Ghuniem et al. 2019a; Thirulogachandar et al. 2014). During the production and manufacturing process, teas may be prone to heavy metal contamination. This contamination may subsequently pose a severe threat to human lives owing to heavy metal increase in the body (Al-Oud 2003). Because of their ability to accumulate in vital human organs over an extended period, heavy metals may, thus, pose a significant risk to health. Despite the high level of toxic and essential elements in commercial teas reported in many studies, copper (Cu), nickel (Ni), and chromium (Cr) levels were investigated in cultivated black tea in north of Iran. In their studies, Ni was found higher with the concentration 13.72 mg/kg (Mohagheghian et al. 2015). Also, another study in Egypt investigate the levels of Cu, Fe, and Pb in several commercially available brands of tea (*Camellia sinensis*); among the investigated metals, Fe was found the highest with the concentration of 193.82 µg/g (Soliman 2016); no data is available to the best of our knowledge, and as at time writing, this report on metal composition of rooibos teas sold in South Africa. Also, there is a lack of quantitative statistics to equate the health risk of heavy metal levels in tea infusions. Therefore, this study aims to determine and quantify the level of heavy metals in rooibos tea in South Africa using the ICP-MS with a view to establishing their health risk.

Materials and methods

Sample collection

A total number of 80 different brands of rooibos tea samples from South Africa were purchased during the 2019 summer (40) and winter (40) seasons from various registered retail shops including 12 natural rooibos, 36 herbal rooibos, and 12 flavor rooibos samples. The tea samples in zip lock bags were stored at room temperature, until analysis. Sample preparation and analysis were done at the Department of Biochemistry of the University of Johannesburg, South Africa.

Instruments and reagents

ICP-MS NexION 300Q from PerkinElmer, USA, microwave digestion system CEM 5 (CEM Corporation, NC, USA),

ultra-pure de-ionized water (18.2 MΩ cm) from a Milli-Q analytical grade water purification system (Millipore), 65% ultra-pure nitric acid (HNO₃), acetone, and hydrogen peroxide (H₂O₂) were used throughout the investigation. Argon, 99 % pure from Afrox, South Africa, was used as purging gas and protective.

Sample preparation

Microwave sample digestion

A microwave acid-assisted system was used to digest the tea materials. Tea samples (0.5 ± 0.001g) were weighed into Teflon vessels and 10 mL 65% HNO₃ added. The vessels were heated up to 1200 W in a microwave system, followed by a microwave digestion program (Table 1) as per the manufacturer's instructions. After complete digestion, the clear solutions were filtered through Whatman No.1 filter paper (Whatman Ltd., England), transferred into 50-mL volumetric flasks and made up to 50 mL using ultra-pure de-ionized water. The content of Fe, Cd, As, Cr, and Pb were determined via ICP-MS.

Quality assurance and quality control

Reference standard solutions purchased from Merck, Darmstadt, Germany, were prepared daily from heavy metal stock standards (1000 mg/l) and were diluted to the corresponding heavy metal solution. Samples, intermediate standard solutions, calibration standard solutions, and the blank solutions were prepared by diluting in 65 % Suprapur® HNO₃ (obtained from Merck, Darmstadt, Germany). To obtain the sensitivity factors for the different elements, a calibration solution was prepared from the ICP-MS multi-element stock standard solution (Merck) with internal standard (Scandium). Certified reference materials (CRM) INCT-1 Tea Leaves, obtained from the Institute of Nuclear Chemistry and Technology, Warsaw, Poland, was used to validate the analytical procedure.

The method followed in this study was validated by determining the parameters, i.e., linearity, limits of detection and quantification (LOQ), as well as recovery. The standard solution of mixed heavy metals at equal concentrations was diluted into 0, 10, 50, 100, 500, 800, and 1000 µg/L of 1% nitric

Table 1 Microwave oven program

Step no	Status	Time (min)	Temperature (°C)
1	Ramp	20	200
2	Hold	10	200
3	Cooling	20	80

acid solution, and 50 µg/L of the internal standard (Sc) was added to each standard solution. The quality of the analytical procedures was checked using Polish Certified Reference Material Tea Leaves (INCT-TL-1) from the Food and Drugs Control Center, Poland, and treated in the same manner as for the analyzed tea samples.

ICP-MS working parameters.

The operational parameters of ICP-MS were set as specified by the manufacturer, using the normal detection mode and the output status of the instrument and controlled by inhaling liquid mass spectrometry. Double charge, oxide, sensitivity, and device resolution satisfied the measuring criteria. The exact parameters were ICP RF power of 1050 W, plasma gas flow of 16 L/min, acquisition mode: Jump peak, Atomizer flow of 0.89 L/min, integral time of 36 s, auxiliary gas flow rate of 1.2 L/min, scanning time of 20 s, repetition 3 times, detector being in a double mode, number of readings of 1, atomizer that was a concentric Nebulizer, and an automatic detection method followed.

Statistical analysis

The collected data were analyzed using Minitab (16 version) statistical software packages. A two-way ANOVA was used to establish significant differences between concentrations of heavy metals in samples obtained in the two seasons.

Health risk assessment of heavy metals

Health risk assessment is usually performed in research related with the well-being of humans (Aigberua et al. 2018). Risk assessment of heavy metal intake via tea consumption among humans was evaluated by establishing estimated daily intake (EDI), hazard index (HI), and target hazard quotient (THQ) (Fu et al. 2015). Estimated daily intake was calculated to evaluate the everyday exposure of individual via consumption of rooibos teas using Eq. 1 according to Chary et al. (Chary et al. 2008).

$$EDI = \frac{C * ED * EF * FiR}{W_{AB} * TA * 1000} \tag{1}$$

Where:

- C Concentration of metals
- E_D Exposure time (70 years) (Fu et al. 2015)
- E_F Exposure frequency (365 days/year) (Fu et al. 2015)
- FiR Amount of daily intake (8 g/person/day) (Cao et al. 2010)
- W_{AB} Average body weight (60 kg was adopted for South African adults) (Walpole et al. 2012)
- T_A E_D × E_F (25,550)

Target hazard quotient

THQ indicates the possibility that a regular tea consumption might potentiate some carcinogenic health risk. The hazard quotient was determined following Eq. 2.

$$\text{Target Hazard Quotient} = \frac{\text{Estimated daily intake}}{\text{Oral Reference dose (RfD)}} \tag{2}$$

RfD for each element according to USEPA (USEPA 2013) are As (0.0002), Cd (0.0036), Cr (1.5), Fe (0.7), and Pb (0.004).

Hazard index

Hazard index is the total risk for a combination of heavy metals and was determined using Eq. 3.

$$\text{Hazard Index} = \sum THQ(\text{USEPA 2013}) \tag{3}$$

Result and discussion

The results of the five heavy metals under the optimized instrument conditions had linear regression above 0.999; however, the linearity of the calibration curve was considered satisfactory when R² > 0.995. The LOD and LOQ were determined using the blank values of five target heavy metals measured by ICP-MS, and a good linearity for the calibration curves was obtained for all the elements analyzed in this study. When comparing the certified values for the heavy metals against the present determined values using ICP-MS, recovery of each element was in the range of 87 and 100% as presented in Table 2.

The relatively very little difference between the determined values and those of certified reference tea showed the reliability of the analytical method as presented in Table 2. The quantitative methods used in this work are suitable for determining the levels of heavy metals in tea. Table 3 shows the levels of heavy metals recovered from rooibos tea in the present study in comparison with those regulated in some countries.

From the present study, the concentrations of As in summer samples were all below the LOD, whereas in winter samples, concentrations ranged from <LOD to 0.51 mg/kg (see Tables 4 and 5).

When comparing these results as shown in Tables 4 and 5 and others reported in the literature (Shekoohiyan et al. 2012; Zhao and Zhao 2019), those recovered and reported in the present study were below the permissible levels stipulated by the WHO (WHO 2006). Long-term contact to arsenic from food and drinking water may lead to cancer and skin lesions (Martinez et al. 2011).

The concentrations of Cd ranged from 0.10 to 0.17 mg/kg for summer and 0.09 to 0.17 mg/kg during winter are

Table 2 Certified reference materials for element determination (mg/kg), expressed as mean value \pm standard deviation

Element	Certified value (mg/kg)	Determined value (mg/kg)	LOD	LOQ	Recovery %
As	0.106 \pm 0.021	0.110 \pm 0.21	0.32	0.45	96
Cd	0.030 \pm 0.004	0.029 \pm 0.01	0.09	0.19	97
Cr	1.91 \pm 0.22	1.91 \pm 0.01	0.05	0.13	100
Fe	432 \pm 0.00	423 \pm 10.27	1.01	1.12	97
Pb	1.78 \pm 0.24	1.54 \pm 0.24	0.04	0.05	87

LOD limit of detection, LOQ limit of quantification

presented in Tables 4 and 5. These values are within the (WHO 2006) permissible limit. In Uganda, lower concentrations of Cd were reported in tea with the metal classified by one of the most toxic heavy metals and a human carcinogen (Bamuwamyé et al. 2017). Chronic exposure to Cd at the lower levels may result in the skeletal system, kidney and respiratory system (Jaishankar et al. 2014). The main sources that link to the accumulation of Cd in soil and plants are combustion of fossil fuels, non-ferrous smelters, phosphate fertilizers, zinc mines, and sewage sludge application (Roberts 2014).

In the case of Cr, the concentrations ranged from 0.17 to 6.71 mg/kg for the summer period and 0.10 to 11.98 mg/kg for the winter period. The highest Cr concentration was recorded in winter sample MM18; this may be because during the rainy season, there is a lot of soil erosion, caused by underground drainage and seepage water that is deposited in lower-lying tea fields. The concentrations recorded for Cr were higher than the permissible limit as shown in Table 5. Bamuwamyé et al. (Bamuwamyé et al. 2017), Nazir et al. (Nazir et al. 2015), Olowoyo et al. (Olowoyo et al. 2012), and Patrick-Iwuanyanwu and Udowelle (Patrick-Iwuanyanwu and Udowelle 2017) also reported higher concentrations of Cr in their study and similar to our study. Cr is present in all plants but is a non-essential element for plants; there is no absorption pathway for its uptake but relies on Cr speciation (Singh et al. 2013). This metal is not

absorbed directly by plants but is accumulated by carrier ions such as sulfates or ferrous ions. Its toxicity alters plant germination and compromises growth by affecting photosynthesis, other metabolic processes, and total dry matter production (Shanker et al. 2005). Hence, the constant intake of such teas in search of health benefits may exceed limits that are attributed to their toxicity. Chromium is generally considered as an indigenous contaminant and is ascribed mostly to contamination from the CTC rollers during the production of tea, due to the gum metals in the CTC rollers which have only trace level of Cr content (Seenivasan et al. 2008). Cr has a wide use in the industries and is considered as a serious environmental pollutant. Cr mining are among the major business within the mining industry in South Africa, and its presence could be trace to various industrial activities and plating of cars, among others (Iloms et al. 2020). However, there is still the need for a further work on speciation of Cr in rooibos tea consumed in South Africa.

Iron is among the essential elements in all living organisms due to its major role in metabolic processes such as O₂ transport and DNA synthesis (Dordas 2008). Iron in nature is present in two states of oxidation, i.e., a divalent (ferrous ion, Fe²⁺) and trivalent (ferric ion, Fe³⁺). Generally, it is an important trace factor for humans, animals, and plants (Dabanović et al. 2016); however, it may be toxic in excess of concentrations due to its pro-oxidant activity, and its deficiency cause a

Table 3 Comparison of levels (mg/kg) of heavy metals in analyzed teas relative to those of some African countries

Country	As	Cd	Cr	Fe	Pb	Reference
This study	ND–0.51	0.09–0.17	0.10–11.98	31–450	0.06–2.73	
Kenya	-	-	0.4–1.8	5.1–5.4	0.1–0.2	(Karak and Bhagat 2010)
Uganda	-	ND–0.2	ND–7.73	1.10–39.89	0.35–8.39	(Karak et al. 2017)
Nigeria	-	-	0.39–7.61	141–453	0.005–0.8	(Karimzadeh et al. 2013)
Ghana	1.40–2.00	0.10–1.50	-	1.05–7.45	0.10–0.40	(Liu et al. 2013)
Egypt	-	-	-	97.9–488.49	0.29–3.2	(Ghuniem et al. 2019a)
Ghana	-	-	<0.001–0.9	27.3–302.95	<0.001–2.05	(Martinez et al. 2011)

The values in bold form, shows the present study

WHO World Health Organization (McGaw and Eloff 2008) (As, 1.0; Cd, 0.3; Cr, 1.3; Fe, 450; and Pb, 10); ND not detected

Table 4 Concentrations (mg/kg) of heavy metals from rooibos tea samples marketed in South Africa (summer)

CODE	As	Cd	Cr	Fe	Pb
MM1	ND	< LOD	3.96 ± 0.09	286 ± 10.54	0.67 ± 0.01
MM2	ND	< LOD	6.71 ± 0.23	275 ± 9.59	0.29 ± 0.01
MM3	ND	N.D	4.65 ± 0.25	126 ± 3.31	0.12 ± 0.00
MM4	<LOD	0.17 ± 0.14	3.95 ± 0.16	253 ± 8.05	0.56 ± 0.00
MM5	ND	<LOD	2.35 ± 0.11	81 ± 2.99	0.07 ± 0.00
MM6	ND	<LOD	2.07 ± 0.14	47 ± 1.74	< LOD
MM7	ND	0.15 ± 0.20	3.81 ± 0.11	239 ± 8.31	0.36 ± 0.01
MM8	ND	<LOD	2.29 ± 0.11	130 ± 4.85	0.32 ± 0.00
MM9	ND	ND	4.90 ± 0.21	208 ± 7.07	0.17 ± 0.00
MM10	<LOD	<LOD	4.07 ± 0.12	302 ± 10.91	0.85 ± 0.01
MM11	ND	<LOD	2.47 ± 0.13	127 ± 5.34	0.18 ± 0.01
MM12	ND	<LOD	3.43 ± 0.25	158 ± 5.91	2.73 ± 0.02
MM13	ND	< LOD	2.13 ± 0.04	103 ± 2.99	0.09 ± 0.00
MM14	ND	< LOD	2.56 ± 0.05	115 ± 3.50	0.12 ± 0.00
MM15	ND	<LOD	2.10 ± 0.12	163 ± 4.81	0.19 ± 0.00
MM16	ND	<LOD	2.85 ± 0.04	184 ± 6.17	0.29 ± 0.01
MM17	ND	< LOD	2.29 ± 0.09	123 ± 4.22	0.20 ± 0.01
MM18	ND	< LOD	2.16 ± 0.05	104 ± 3.42	0.09 ± 0.00
MM19	ND	< LOD	4.04 ± 0.16	271 ± 10.06	0.40 ± 0.01
MM20	ND	<LOD	2.27 ± 0.09	106 ± 3.68	0.14 ± 0.00
MM21	ND	< LOD	2.11 ± 0.07	90 ± 2.82	0.08 ± 0.00
MM22	ND	<LOD	2.17 ± 0.12	132 ± 3.68	0.17 ± 0.00
MM23	ND	<LOD	2.65 ± 0.09	141 ± 5.02	0.40 ± 0.01
MM24	ND	0.12 ± 0.15	3.07 ± 0.16	176 ± 5.69	0.31 ± 0.00
MM25	ND	<LOD	5.04 ± 0.19	450 ± 12.55	0.43 ± 0.00
MM26	ND	<LOD	3.95 ± 0.15	392 ± 11.94	0.48 ± 0.00
MM27	ND	<LOD	4.19 ± 0.28	300 ± 10.73	0.83 ± 0.01
MM28	ND	<LOD	2.08 ± 0.11	113 ± 3.63	0.12 ± 0.00
MM29	ND	< LOD	2.28 ± 0.08	144 ± 4.69	0.13 ± 0.00
MM30	ND	<LOD	2.11 ± 0.12	128 ± 4.57	0.13 ± 0.00
MM31	ND	<LOD	1.91 ± 0.06	108 ± 3.39	0.12 ± 0.01
MM32	ND	< LOD	2.06 ± 0.05	128 ± 3.42	0.13 ± 0.00
MM33	ND	<LOD	2.01 ± 0.06	93 ± 2.94	0.09 ± 0.00
MM34	ND	< LOD	3.22 ± 0.09	241 ± 8.14	0.36 ± 0.00
MM35	ND	ND	2.07 ± 0.09	103 ± 3.61	0.13 ± 0.00
MM36	ND	ND	0.17 ± 0.02	109 ± 4.22	0.13 ± 0.00
MM37	ND	ND	1.42 ± 0.06	434 ± 13.92	0.32 ± 0.00
MM38	ND	ND	0.91 ± 0.06	210 ± 5.86	0.25 ± 0.00
MM39	ND	0.10 ± 0.11	3.29 ± 0.09	155 ± 4.39	2.59 ± 0.03
MM40	ND	ND	3.82 ± 0.15	104 ± 3.71	0.07 ± 0.00

ND not detected, LOD limit of detection

problem in metabolism. For example, Fe is a component of the active site of many reductive hydrogenases, most often being linked with sulfur comprising ligands. Fe together with hemoglobin and ferredoxin plays a central role in metabolism. Deficiency of Fe in plants produces chlorosis disease, and the

Table 5 Concentrations (mg/kg) of heavy metals from rooibos tea samples marketed in South Africa (winter)

CODE	As	Cd	Cr	Fe	Pb
MM1	ND	0.17 ± 0.12	1.66±0.05	283±7.63	0.54±0.01
MM2	ND	<LOD	2.12 ± 0.07	138 ± 5.84	0.46 ± 0.00
MM3	0.51 ± 0.06	<LOD	0.84±0.05	75±1.85	0.82±0.01
MM4	ND	ND	0.33±0.02	165±5.33	0.44±0.00
MM5	ND	<LOD	0.15±0.00	84±2.21	0.15±0.00
MM6	<LOD	ND	0.10±0.01	31±1.23	0.07±0.00
MM7	ND	0.13 ± 0.18	1.72±0.10	292±8.445	0.41±0.01
MM8	ND	ND	0.15±0.03	108±1.69	0.10±0.00
MM9	ND	<LOD	1.81 ± 0.06	102 ± 3.29	0.15 ± 0.01
MM10	< LOD	ND	3.21±0.14	208±10.18	0.12±0.00
MM11	ND	< LOD	0.17±0.02	56±1.81	0.08±0.00
MM12	ND	ND	1.42±0.03	118±3.61	0.05±0.00
MM13	ND	ND	3.98±0.15	205±5.94	0.46±0.00
MM14	ND	0.14 ± 0.08	0.73±0.05	220±5.81	0.43±0.01
MM15	ND	<LOD	0.23±0.04	86±2.60	0.12±0.00
MM16	ND	ND	2.83±0.07	170±5.20	0.10±0.00
MM17	ND	ND	2.98±0.09	211±6.40	0.49±0.01
MM18	ND	ND	11.98±0.60	445±15.33	0.26±0.00
MM19	ND	< LOD	1.91±0.14	382±12.21	0.51±0.00
MM20	ND	ND	4.39±0.22	196±6.00	0.46±0.01
MM21	<LOD	0.14 ± 0.01	0.73±0.02	112±3.26	0.58±0.00
MM22	ND	<LOD	0.37±0.03	147±4.42	0.18±0.00
MM22	ND	ND	0.25±0.04	91±2.68	0.09±0.00
MM24	ND	ND	0.27±0.02	110±3.53	0.11±0.00
MM25	ND	< LOD	0.33±0.03	131±4.86	0.87±0.02
MM26	<LOD	<LOD	2.05±0.07	391±10.05	0.55±0.00
MM27	<LOD	0.17 ± 0.22	1.45±0.05	188±6.46	0.51±0.01
MM28	ND	ND	0.75±0.03	156±4.24	0.18±0.00
MM29	ND	<LOD	0.37±0.00	202±7.02	0.21±0.01
MM30	ND	ND	2.14±0.06	136±4.29	0.06±0.00
MM31	N.D	< LOD	0.52±0.03	167±5.41	0.81±0.01
MM32	ND	<LOD	0.44±0.05	102±3.55	0.13±0.00
MM33	ND	ND	1.32±0.10	301±10.36	0.55±0.00
MM34	ND	< LOD	1.91±0.09	284±9.26	0.58±0.01
MM35	ND	< LOD	1.42±0.07	188±5.76	0.40±0.00
MM36	ND	ND	11.56±0.47	405±17.18	0.26±0.00
MM37	ND	<LOD	0.47±0.03	109±3.54	0.35±0.01
MM38	ND	ND	0.37±0.04	161±4.72	0.20±0.01
MM39	ND	0.09 ± 0.10	2.28±0.14	300±9.55	0.60±0.02
MM40	< LOD	<LOD	1.69±0.05	285±9.81	0.52±0.00

ND not detected, LOD limit of detection

metal facilitates the oxidation of carbohydrates, proteins, and fats to regulate body weight, which is an essential contributor in certain body diseases (diabetes). As established in this study, the concentrations of Fe recovered from tea samples purchased during summer and winter ranged between 47 to 450 mg/kg and 31 to 450 mg/kg, respectively, which were below the WHO permissible limit (Tables 4 and 5). The concentrations reported in the current study are similar to those reported by Nkansah et al. (Nkansah et al. 2016) and Brzezicha-Cirocka et al. (Brzezicha-Cirocka et al. 2017).

Tables 4 and 5 presents data on the concentrations (mean: 0.06 to 2.97 mg/kg) of Pb found in analyzed tea. As found, levels in tea samples collected during winter (range: 0.06 to 2.97 mg/kg) were higher than those recovered from summer (range: 0.07 to 2.73 mg/kg) samples. Some studies have reported low levels of Pb ranging from 0.10 to 0.40 and 0.159 to 0.824 mg/kg in tea samples (Bamuwamye et al. 2017; Karak et al. 2017; Sarfo et al. 2012), which were comparable to those in this study. This metal is extremely toxic and may cause mental disability, especially in children. In general, intake of Pb can affect major organs in the body such as cardiovascular, hematopoietic, renal, and nervous systems (Flora et al. 2012) causing both acute chronic and acute chronic toxicities that could be carcinogenic in nature (Cao et al. 2014). As seen, none of the analyzed samples contained this metal at concentrations that are higher than the WHO permissible limit recommended.

The difference in the concentrations of heavy metals in each tea analyzed generally, can be attributed to the soil characteristics (pH, organic matter, and type) equally regulate the movement of heavy metals in the soil. Long-term tea plantation can induce soil acidification and increased levels of bio-available heavy metals in the soil, thereby increasing the risk of accumulation of heavy metals in tea leaves. Differences in this sample may be due to specific agro-climatic sources and the use of phosphate fertilizers and fungicides in agriculture. Such products also contain large concentrations of radioactive elements such as Pb, Hg, Cd, and Cu (Ferreira et al. 2015; Karak and Bhagat 2010; Soliman 2016). Other conditions responsible for variations in metal activity may be due to differences in farming areas, temperature, and other environmental conditions (Barone et al. 2016; Karimzadeh et al. 2013). Statistically, the two-way analysis of variance (ANOVA) results reveal no statistical differences ($p = 0.832$) as seen in Table 6.

Health risk assessment

Food safety has become a major global issue, as food intake is a major route for human toxic materials. Health risk analysis centered on average daily intake EDI of heavy metal pollutants is one of the critical safety risk management techniques and recognizes the extent and duration of exposure and body weight of exposed individuals (Liu et al. 2013). In light of

Table 6 Two-way ANOVA: response versus season, heavy metals

Source	DF	SS	MS	F	P
Season	1	102	102	0.05	0.832
Heavy metals	4	2102446	525611	231.88	0.000
Interaction	4	700	175	0.08	0.989
Error	390	884043	2267		
Total	399	2987290			

normal consumption habits, it is important to evaluate the health risk level centered on the estimated daily intake EDI of heavy metals associated with tea using health risk tools. It can be accomplished by increasing the average amount of tea ingested per person per day by the level of each item in the tea samples analyzed and comparing it with the overall acceptable daily intake ADI values defined by international health safety organizations.

Recently, numerous studies have confirmed the effect of heavy metals in tea and leaves on human well-being (Affholder et al. 2013; Sofuoglu and Kavcar 2008; Zhu et al. 2013). According to measured contents of As, Al, Cd, Cu, Pb, Zn, and Hg in Pu-erh tea from the Yunnan Province, China, there are no non-carcinogenic risks reported (Cao et al. 2010). Another study (Shen and Chen 2008) also performed a health risk assessment of heavy metals in tea infusion and recorded no non-carcinogenic risk. The hazard index HI of everyday tea consumption was found to be below acceptable limits. In this study, established daily intake EDI values for all essential and non-essential metals extracted in each rooibos tea analyzed were presented in supplementary material Table S1; iron (Fe) (0.0627) had the highest daily intake of metals, followed by Cr, Pb, Cd, and As as the least metal extracted in this study. The EDI values for all metals in this sample were below their respective RfD values, suggesting that the intake of the tea brand tested might not pose health risk to consumers. Target hazard quotient THQ results in the present study were in accordance with some of the previous studies (Neda Sadat et al. 2020; Nkansah et al. 2016). It should be noted that establishing THQ values does not suggest a quantitative evaluation of the risks of an exposed population facing an ultimate health effects but a step in the process of assessing risk as they fairly provide some estimates of risk associated with metal exposure. In doing so, we found that THQ value for each heavy metal analyzed was below 1, signifying that the daily intake of each metal may not pose a major possible health risk. The variation between the THQ values in this analysis and other research may be due to the parameters affecting THQ (metal content, body weight of the user, per capita intake share of each tea, etc.) and their differences in various geographic areas (Chien et al. 2002). The effect of these heavy metals associated with intake of tea was expressed by establishing total

hazard index (HI) values, which were found to be below 1 (Table S2); the value of HQ or HI <1 designates no significant risk of non-carcinogenic effects; the value of HQ or HI >1 designates significant non carcinogenic effects, which increased with increasing value of HQ or THQ (Fu et al. 2015; Wei et al. 2015). The present study reported herein has made it possible suggesting that there is, in fact, no significant non-carcinogenic health risk to consumers via consumption of rooibos tea from South Africa.

Conclusion

This study showed the level of contamination in selected heavy metals, i.e., arsenic, chromium, cadmium, iron, and lead, in a different brand of rooibos teas sampled during winter and summer in South Africa, and compared the levels established to the maximum permissible limits recommended by the World Health Organization (WHO). All rooibos teas analyzed showed the maximum level of a contaminant in analyzed selected heavy metals with only Cr exceeding the permissible limit for heavy metals in plants. Winter seasons have the highest concentrations compared to summer seasons, which is because there is a lot of soil erosion during the rainy season. It may be caused by runoff and seepage water underground and stored in lower-lying tea fields, resulting in high concentrations. The risk level of heavy metal exposure in South Africa via infusion of rooibos tea may likely not pose any adverse health effects, even at concentrations obtained in this study. However, the degree of contaminants found in beverages together with other edible products such as cereals with notable contaminants such as mycotoxins with similar or related health impacts may be a serious concern, because some contaminants have shown the significant functionalities as additive or synergistic health effects in man. This study establishes some scientific facts indicating that the contaminants are within permissible limits when such commodities are consumed. This study can serve as a reference for future advanced studies.

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Author contribution Oluwaseun Mary Areo: conceptualization and writing—original draft. Patrick Berka Njobeh: supervision, funding acquisition, project administration, and writing—review and editing. All authors read and approved the final manuscript.

Declarations

Ethical approval Not applicable.

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Consent to publish Not applicable.

Conflict of interest The authors declare no competing interests.

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