CAFEi2012-232

HEALTH RISK ASSESMENT OF TRACE ELEMENTS UPTAKE BY LEAFY VEGETABLE GROWTH ON SECONDARY TREATED WASTEWATER-IRRIGATED SOIL AND SOILLESS CULTURE

Ali, M.F.[1], Shakrani, S.A.[1]

[1] Division of Water Resources and Environmental System, Faculty of Civil Engineering, Universiti Teknologi MARA, 40450, Shah Alam, Selangor Darul Ehsan, Malaysia.
Email: shahrulazwan.shakrani@gmail.com

ABSTRACT

In the era of there is no guarantee of water resources, reuse of wastewater for agricultural irrigation provides an alternative solution of fresh water dwindling especially in most countries with dry and arid climate. In general, wastewater contains beneficial features such nutrients required for plants growth and development. However, the application of wastewater in irrigation may result in the presence of harmful trace metals to the plants which contribute to the detrimental effects to the people. This paper discusses the health risk associated with the effects of secondary treated wastewater irrigation on *Brassica Campestris* Sp. *Parachinensis* vegetable growth with soil and soilless culture. The study involves the assessment of trace elements such as Cd, Fe, Ni, Pb, and Zn in wastewater, soil and plants. The results revealed the significant concentrations of trace elements in soil and plants irrigated with secondary treated wastewater. Besides, trace element concentrations were found higher in plant growth under soilless culture as compared to soil culture. However, the analysis indicated that the health risk index (HRI) values were less than 1. Thus, the consumption of plants irrigated with secondary treated wastewater by adults and children was no consequences and assumed to be safe in general.

Keywords: *secondary treated wastewater, soil culture, soilless culture, trace elements, health risk index (HRI).*

INTRODUCTION

The population growth, urbanization, indiscrimination use of water and climate change has led to the era of water scarcity problems worldwide. Therefore, most of researchers have shown a great interest in treatment, recycling and reuse of wastewater. The agriculture sector as a biggest freshwater user desperately needs a solution, and wastewater reuse has been seen as a potential alternative to fresh water dwindling especially in most countries with dry or arid climate. In Malaysia, wastewater irrigation is not a common practice but some has shown an interest in agricultural and landscape irrigation.

The wastewater irrigation provides the beneficial aspects by adding valuable plant nutrients and organic matter to soil besides providing a convenient method of waste product disposal [1]. Previously, numerous studies founded that wastewater rich with valuable sources such as organic matter, macronutrient and micronutrient that required by the plant for fertility and productivity of soil [2]. Other researchers found wastewater used for irrigation supply nitrogen N, phosphorus P and potassium K with heavy metal to plants and soil, thus increasing yields and quality [3,4]

However, along with beneficial to horticultural crops, wastewater and sewage effluents contain a significant amount of trace elements and other substances that may be harmed and toxic to people [5]. Besides, sewage wastewater also contains high concentrations of trace elements, salts, bacteria, viruses depending upon its sources and treatment applied [6]. Trace elements and heavy metals such as Zn,Cd,Pb, Fe, Mn and Mo found in wastewater and sewage effluents may be phytotoxic and if accumulated in the crops and fruit will impose a health risk to humans [7,8,9].

The effects of wastewater irrigation on soils have been widely documented, especially on trace elements concentrations and toxicity [10,11]. With continous wastewater applications trace elements can accumulate in the soil thus toxic the plant growth [12]. Besides, human is subjected to a direct impact on health by contamination of soils due the facts that soils are easily contacted and transferred to them [13,14,15]. Soils act as toxic chemical filters may absorb and retain trace elements from wastewater. However, due to continous loading of pollutants and changes in pH, the capacity of soils to retain toxic elements being reduced thus soil may release trace elements into groundwater or available for plant uptake [16].

The main objective of this present work aimed to assess the health risk associated with the effects of secondary treated wastewater irrigation on *Brassica Campestris Sp. Parachinensis* vegetable growth with soil and soilless culture. The specific objectives of this study were to study the characteristics of soil and plant irrigated with secondary treated wastewater along risk associated with the human health with determination of daily intake of metals (DIM) and health risk index (HRI).

MATERIALS AND METHODS

Experimental Details

The experiment was conducted from October 4, 2011 to November 29, 2011 at two different place located at Universiti Teknologi MARA (UiTM) Shah Alam, Malaysia. This equatorial region is affluent with the annual precipitation of 2000-3000 mm and relative humidity of 70-80 % together with an air temperature of 29 – 31 °C days and 20 – 24 °C nights. The final secondary treated wastewater (FSTW) and biological secondary treated wastewater (BSTW) used for irrigation were collected from the Mawar Wastewater Treatment Plant located at UiTM. This wastewater treatment plant consists of treatment process such as screening, grit removal, primary sedimentation and conventional activated sludge treatment with 11 000 population equivalent (PE).

In soil cultivation, the crops were planted on the experimental plot located at the Faculty of Plantation and Agrotechnology, UiTM whilst polybag soil located at the Mawar Wastewater Treatment Plant, UiTM. The 7.5m x 7.5m plot area was divided into six sections for both FSTW and BSTW effluent. Each section was planted with *Brassica campestris Sp. parachinensis* at 1.2m x 7.5m x 0.3m area size. Seeds were planted within 2 cm of the top soil, 20 cm between plot and 10 cm between another. Five polybags with a top diameter of 35mm, bottom diameter of 35mm and the height of 40mm contained mainly freshly mixed soil were obtained from an experimental plot for each FSTW and BSTW effluent . Five seeds were planted within the top of 2cm of soil in the each of polybag. Then, surface irrigation was performed by manually hand watering. Water with approximately the same amount was directly applied to the soil surface using a plastic watering can for both experimental plot and polybag soil cases. *Brassica campestris Sp. parachinensis* were watered seven days per week.

In soilless cultivation, an active hydroponics system called nutrient film technique (NFT) and passive hydroponics system named wick system were applied located at the Mawar Wastewater Treatment Plant, UiTM. In NFT system, two systems were manually constructed for FSTW and BSTW effluent. The ditch of the NFT system used consisted of UPVC pipes of 10 cm wide, 10 cm deep and 2 m long. This hydroponics system comprises of four conduits and each conduit able to cater six growing basket. The submersible pump was applied to the system as the medium to allow the movement of wastewater throughout the system. The plastic cup with holes was used to germinate the seeds. In order to sprout the *Brassica campestris Sp. parachinensis,* two to three numbers of seed were sown on the lumps of wet thread to allow germination. In wick system (directly obtained from KAZZ Marketing Sdn Bhd, Malacca) three systems were planted hydroponically with *Brassica campestris Sp. parachinensis* for FSTW and BSTW effluent. Wet threads were used as support media to grow a *Brassica Sp.* seeds in growing basket. In each basket, two or three of the seeds was placed to be germinated.
The detailed of all the experiments were presented in Table 1 as adopted from Ali & Shakrani [17, 18, 19, 20].

Table 1: Methods conducted in the present study

	Types of Planting Media			
	Soilless (Hydroponic) Cultivation		Soil Cultivation	
Types of Wastewater				
Biological Secondary Treated Wastewater (BSTW)	Wick System (HWS)	NFT System (HNS)	Polybag Soil (SPS)	Experimental Plot (SEPS)
Final Secondary Treated Wastewater (FSTW)	Wick System (HWS)	NFT System (HNS)	Polybag Soil (SPS)	Experimental Plot (SEPS)

e-ISBN 978-967-960-322-4

Wastewater Sampling and Analysis

Before wastewater was applied for irrigation, the FSTW and BSTW effluent samples were taken six times periodically along overall study. Thus, the wastewater sampling was conducted in accordance with APHA [21], BS EN ISO 5667 Part 1 [22] and BS 6068 Part 6 Section 6.10 [23]. For wastewater sampling, about 1 litre of samples was taken for each case and polypropylene sampling bottles were used as a medium for transferable of wastewater from site to laboratory. The wastewater was preserved with concentrated HNO_3 (pH<2) to prevent microbial degradation of heavy metals and stored in a refrigerator at about 4^oC to prevent any possible change in volume due to evaporation. The collected samples later were analyzed for trace element's characteristics such as Cd, Fe, Ni, Pb, and Zn by means of Thermo Scientific iCAP 6000 Series, Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES) (Thermo Fisher Scientific, Cambridge, UK) as prescribed in APHA [21].

Soil Sampling

The soil sampling was conducted in accordance with BS ISO 10381 Part 1 [24], BS ISO 10381 Part 2 [25] and ASTM D5633 [26]. Six samples of top soil at 15cm depth were manually collected from centre of experimental plot and polybag soil with clean stainless steel trowel of 8 cm diameter. At least 500g of soil were collected and transported into polyethylene containers. Samples were later transferred to laboratory together with labelled and sealed.

Vegetable Sampling

Vegetable sampling was conducted in accordance to ISO 874 [27].The process took place after 2 months growth and performed in the late afternoon or early morning to avoid moisture losses. Fresh mustard greens with the same variety and degree of maturity were collected by cutting off the whole edible portion with plastic scissors. Afterward, the samples were washed under tab water and double rinsed with deionized water to eradicate soil particle and to remove suspended pollutant. Finally, samples with labelled were wrapped and sealed in sterile plastic bags and later were transported as quickly as possible to the laboratory for further procedures.

Soil and Vegetable Sample Preparation

After oven-dried at 70^oC for 48 h to remove all moisture, plant tissue and soil samples were ground to the fine powder by using a mortar and pestle. The samples were later sieved and passed through 2-mm mesh sieve.

Acid Wet Digestion

The wet digestion of vegetable and soil samples was conducted using HACH Digesdahl Digestion Apparatus [28]. This method was introduced by Jones and Case [29] and later prescribed as Piranha Clean Method (also known as Vigreux Method) where a mixture of sulphuric acid and hydrogen peroxide were initiated [30,31, 32].This procedure was performed after preparation of samples. After oven-dried, 0.25-0.5g of each soil and vegetable samples were accurately weighted by analytical weighing balance (A & D Company Limited, Tokyo Japan) into a 100-mL digesdahl digestion flask (HACH, CO USA). Then, 4 to 6mL of concentrated sulphuric acid (Mallinckordt Chemicals, USA) (spec. gravity 1.84), H_2SO_4 was added to the digestion flask. The sample was then heated and boiled at 440^oC (825^oF) for 4 minutes. Afterwards, 10 to 20 mL of 50% hydrogen peroxide (Merck Schuchardt OHG, Hohenbrunn Germany) (H_2O_2) (spec. gravity 1.11) was added to the flask via the funnel. The sample was then continued heated for one more minute. After that, the digested sample was diluted with approximately 70mL with 18Ω deionized water (ELGA Labwater, UK). After cooling, the sample was filtered through a Whatman No.41 filter paper into a 100 mL volumetric flask pre-washed with 10% concentrated HNO_3. Later, samples were further filtered through a 0.5 μm PTFE membrane (Whatman Ltd, UK).

Soil and Vegetable Analysis

The concentration of trace elements such as Cd, Fe, Ni, Pb, and Zn were determined by Thermo Scientific iCAP 6000 Series, Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES) (Thermo Fisher Scientific, Cambridge, UK) (USEPA Method 7000B). Analyzed were carried out as described in Standard Methods for Examination of Water and Wastewater [21]. Multi elements standard solution were used for calibration and were prepared at a concentration of 0.25, 0.5. 1, 3 and 5 ppm. Blank preparation used for calibration was distilled water. Each sample was replicated at three times to obtain the accurate values.

Statistical Analysis

The data were statistically analyzed using the SPSS version 17 software with a significance level of $P < 0.05$. All collected data carried out were subjected to independent t-Test and one way analysis of variance (1 Way - ANOVA).

Daily Intake of Metals

The daily intake metals (DIM) as the following equation [33,34]:

$$DIM = (C_{metal} \times C_{factor} \times D_{food\ intake}) / (B_{average\ weight}) \tag{1}$$

Where C_{metal} = heavy metal concentrations in plants (mg/kg), C_{factor} = conversion factor; take 0.085 [35], $D_{food\ intake}$ = daily intake of vegetables; take 0.345 and 0.232 kg/person/day for adults and children [36]. B average weight = average body weight ; take 55.9 and 32.7 kg for adults and children [36].

Health Risk Index

The risk index (HRI) as the following equation [33]:

$$HRI = (DIM)/(RfD) \tag{2}$$

Where RfD = reference oral dose for each metal as described in Table 2
Human are considered to be safe if HRI < 1.

Table 2: RfD for each trace elements

Trace elements	RfD (mg/kg/day)	References
Cadmium, Cd	0.001	IRIS, USEPA [37]
Iron, Fe	0.7	PPRTV, USEPA [37]
Nickel, Ni	0.02	IRIS,USEPA [37]
Lead, Pb	0.0035	JECFA[38], [39]
Zinc, Zn	0.3	IRIS,USEPA [37]

IRIS: Integrated Risk Information System, PPRTV=Provisional Peer Reviewed Toxicity Values, HEAST=Health Effects Assessment Summary Tables

RESULTS AND DISCUSSION

Wastewater Characteristics

Trace element's characteristics of biological (BSTW) and final (FSTW) secondary treated wastewater effluents measured during a two month period of irrigation are presented in Table 3. The independent t-Test performed at 0.05% probability indicate all of the P-value to determine if there is any significant difference between the means of trace element's characteristics among of both secondary treated wastewater effluents studied. Most of parameters demonstrated the p-value less than 0.05 which rejected the null hypothesis of the study. Thus, the quality of both secondary treated wastewater effluents was found to be relatively significant in most trace element parameters for instance iron, nickel, lead and zinc. However, cadmium was not found in both secondary treated wastewater effluents. Besides, the values of these macronutrients and trace element parameters were significantly higher in final secondary treated wastewater (FSTW) as compared to biological secondary treated wastewater (BSTW). This is driven by the fact that the wastewater treatment plant studied have been seen not to be able to maintain and work properly. The secondary treatment breakdown has been discovered during experimental work with trickling filter dysfunctional. Thus, this is suggested that the proper monitoring needs to be made in order to ensure the sustainable wastewater treatment processes. Besides, a study was conducted during monsoon season and resulted in a slow rate of an organism metabolism process which contributed to the ineffective secondary treatment.

Table 3: Characteristics of secondary treated wastewater

	BSTW	FSTW	t-Test		Allowable Standards			
			P Value	Significance Level ns=P_{Value}>0.05 * = P_{Value}<0.05	Malaysian EQA [40]		WHO [41]/ FAO [42]	USEPA [43]
					A	B		
Cadmium, Cd (mg/L)	ND	ND	NT	NT	0.01	0.02	0.01	0.01-0.05
Iron, Fe (mg/L)	0.465±0.031	0.260±0.047	0.000	*	1.0	5.0	5.00	5.00-20.00
Nickel, Ni (mg/L)	0.006±0.001	0.007±0.001	0.000	*	0.20	1.0	0.20	0.20-2.00
Lead, Pb (mg/L)	0.011±0.003	0.019±0.005	0.000	*	0.10	0.5	5.00	5.00-10.00
Zinc, Zn (mg/L)	1.122±0.058	1.029±0.134	0.000	*	2.0	2.0	2.00	2.00-10.00

Each data represents the mean of 6 values ± the standard deviation, *=statistically significance, ns= statistically not significance, ND= none detected, NT=none tested.

In general, most of the trace element's characteristics of secondary treated wastewater were found within the allowable limits and standards regulated by Malaysian Environmental Quality Act 2009 (EQA), World Health Organization (WHO), Food and Agriculture Organization of the United Nations (FAO) and United States Environmental Protection Agency (USEPA). None detection of Cadmium (Cd) in secondary treated wastewater resulted from an impurity in galvanized pipes and fitting along with free contaminant used of phosphate in detergent and washing powder. Meanwhile, Iron (Fe) concentrations in secondary treated wastewater were found in ranges of 0.26 mg/L to 0.465 mg/L. Iron pipes contributed to the high sources of Iron (Fe) in wastewater. Besides, cleaning and personal products include toothpaste also contributed to the high level of Iron (Fe) in wastewater. On the other hands, Nickel (Ni) concentrations in secondary treated wastewater however was found lower ranging from 0.006 to 0.007 mg/L in the present secondary treated wastewater study. Low concentrations of Nickel (Ni) were resulted from rechargeable batteries, metal coated and galvanized pipes and fittings. Besides, nickel concentrations found in wastewater were due to the used of deodorants, toothpaste, conditioners, hand wash, body wash and shampoo. Similarly, Lead (Pb) concentrations were found lower to vary from 0.011 to 0.019 mg/L. The low level of Lead in secondary treated wastewater were resulted from the uses of toilet paper, toilet refreshers, dishwashing tablets, personal care products for instance mouthwash, hair conditioner and toothpaste, detergent as well as laundry. Finally, Zinc (Zn) concentrations were being detected at ranges of 0.2 to 1.18 mg/L. The highest concentrations of Zinc (Zn) in wastewater were resulted from galvanized iron pipe and fitting, personal and body care product such as deodorant, shampoo as well as cosmetic.

Soil Characteristics

The trace element's characteristics of the soil before and after irrigated with both final (FSTW) and biological (BSTW) secondary treated wastewater effluents are presented in Table 4. In general, trace the element's characteristics of soil irrigated with FSTW and BSTW are found to be relatively higher in the soil polybag system (SPS) as compared to the soil experimental plot system (SEPS). Since SEPS covered a larger cross sectional area as compared to the SPS, the irrigated wastewater tends to distribute along the sectional area and later infiltrated in the groundwater. Thus, the accumulation of trace elements were slightly higher on the top and reduce as it goes deeper. On the other hand, irrigated wastewater tends to converge in the smaller cross sectional area of SPS during irrigation process. As a result, the SPS is much more likely to accumulate a greater level of trace element's characteristics in soil. Trace element's characteristics for instance Cd in both SPS and SEPS are increased after irrigated with both FSTW and BSTW. However, trace elements such Fe, Ni, Pb, and Zn in soil are decreased after irrigated with FSTW and FSTW.

e-ISBN 978-967-960-322-4

Table 4 : Characteristics of soil before and after irrigated with secondary treated wastewater

		Cadmium, Cd (mg/L)	Iron, Fe (mg/L)	Nickel, Ni (mg/L)	Lead, Pb (mg/L)	Zinc, Zn (mg/L)
Present Study	Before Irrigation					
	SOIL-BEFORE	0.003±0.002	105.840±14.354	0.031±0.007	0.080±0.007	0.582±0.306
	After Irrigation					
	FSTW-SEPS	0.021±0.000	0.548±0.020	0.009±0.000	ND	0.149±0.003
	BSTW-SEPS	0.025±0.000	0.557±0.013	0.009±0.000	ND	0.088±0.004
	FSTW-SPS	0.026±0.002	44.820±16.343	0.019±0.003	0.008±0.010	0.258±0.110
	BSTW-SPS	0.032±0.002	1.877±0.822	0.012±0.001	ND	0.059±0.017
Analysis of Variances (ANOVA)	P-Value	0.000	0.000	0.000	0.000	0.000
	Significance Level ns=P_{Value}>0.05 *=P_{Value}<0.05	*	*	*	*	*

Each data represents the mean of 6 values ± the standard deviation, ND= none detected

Meanwhile, the analysis of variance (ANOVA) was carried out on trace elements characteristics of the soil before and after irrigated with FSTW and BSTW. The analysis performed at 0.05% probability indicate all of the F-value and P-value to determine if there is any significant difference between the means of trace element's characteristics of both soil polybag system (SPS) and soil experimental plot system (SEPS) after irrigated with secondary treated wastewater effluents . Most of parameters demonstrated the p-value less than 0.05 which rejected the null hypothesis of the study. Thus, both BSTW and FSTW are believed to be relatively significance of the contribution of the availability of trace elements in soil after irrigation. The concentrations of cadmium in soil increased from 0.003 mg/L to 0.032 mg/L. The excessive level of cadmium in soil resulted in soil toxicity. Nevertheless, the regulatory limit for cadmium in soil is 100 mg/L [44]. However, the results indicated that there are reduction in most trace elements' characteristics of soil after irrigated with both FSTW and BSTW for instance Fe, Ni, and Zn which also acts as micronutrients. The reduction of most micronutrients concentrations in soil is believed to be due to the absorbed of elements by the plants for physiological functions as an important in growth and development for plant whole life cycle completion [45]. The lower level of nickel is believed to be due to the absorbed by clays, iron, manganese oxides and organic matter while lead is reacted with clays, phosphates, sulfates, carbonates, hydroxides and organic matters in soils [46] As nickel, zinc is absorbed by clay minerals, carbonates and hydrous oxides thus present in soil at lower levels.

Vegetable Characteristics

The concentrations of trace elements in plant after irrigated with final (FSTW) and biological (BSTW) secondary treated wastewater is presented in Table 5. The results indicated the concentrations of trace elements were found higher in plants grown under hydroponic cultivation as compared to soil cultivation. The analysis of variance (ANOVA) performed at 0.05% probability indicates all of the F-value and P-value to determine if there is any significant difference between the means of trace element concentrations among of plant grown under soil and hydroponic cultivation. The analysis demonstrated the p-value less than 0.05 which rejected the null hypothesis of the study. As a result, there are significant differences between means of trace element concentrations in plants grown with soil and hydroponic system during eight weeks of irrigation. Thus, it is believed the secondary treated wastewater irrigation significantly improved the trace element concentrations in plants.

Table 5: Plants characteristics after irrigated with secondary treated wastewater

		Cadmium, Cd (mg/L)	Iron, Fe (mg/L)	Nickel, Ni (mg/L)	Lead, Pb (mg/L)	Zinc, Zn (mg/L)
Present Study	FSTW-SEPS	ND	0.005±0.001	0.004±0.000	ND	0.034±0.002
	BSTW-SEPS	ND	0.005±0.002	0.004±0.001	ND	0.031±0.005
	FSTW-SPS	ND	0.004±0.001	0.004±0.000	ND	0.031±0.007
	BSTW-SPS	ND	ND	0.004±0.000	ND	0.039±0.001
	FSTW-HNS	ND	0.014±0.001	0.004±0.000	ND	0.041±0.002
	BSTW-HNS	ND	0.004±0.001	0.004±0.001	ND	0.040±0.001
	FSTW-HWS	ND	0.020±0.002	0.004±0.000	ND	0.076±0.014
	BSTW-HWS	ND	0.001±0.001	0.004±0.001	ND	0.043±0.001
Analysis of Variances (ANOVA)	F Value	NT	14958.111	1366.504	NT	24.074
	P Value	NT	0.000	0.000	NT	0.000
	Significance Level ns=P_{Value}>0.05 *=P_{Value}<0.05	NT	*	*	NT	*

Each data represents the mean of 6 values ± the standard deviation, *=statistically significance, ns= statistically not significance, ND= none detected, NT=none tested

The concentrations of iron observed in plants for both soil and hydroponic cultivation range from 0.004 mg/L to 0.020 mg/L. In general, iron is an important for protein and metabolism activities [47] along photosynthesis and cellular activities [48]. Usually, plants exposed to iron deficiency at 50 to 150 mg/kg [49] while toxicity at level of 500mg/kg [50].The toxicity of iron resulted in yield and photosynthesis reduction [51] along cellular damages [52,53].The concentrations of nickel measured in plants for both soil and hydroponic cultivation ranges around 0.004 mg/L Previously, most of researchers agreed nickel is required for growth and development of plants but in small quantities varies from 0.01 to 5 µg/g [54,55,56]. The nickel is usually tolerance of 0.05 to 5.0µg/g [57] whilst subjected to toxicity at range 10 µg/g to 50 µg/g [58]. The concentrations of zinc measured in plants for both soil and hydroponic cultivation ranges from 0.031 mg/L to 0.076 mg/L. Usually, zinc is responsible for growth and metabolism of plants [59].However, plants subjected to zinc deficiency at ranges of 15 to 20 µg/g [49] and resulted in growth reduction [60].Meanwhile, plants exposed to zinc toxicity under concentrations ranges from 100 to 300 µg/g [61] which resulted in reduction in growth and development of plants [62] as well as root elongation restrained [63].However, cadmium and lead concentrations were not detected in plants for both soil and hydroponic cultivation.

Daily Intake of Metals (DIM) and Health Risk Index (HRI)

The risk associated to trace element accumulation in plants to the human is performed throughout determination of daily intake of metals (DIM) and health risk index (HRI). The DIM and HRI for adults and children through the consumption of plants irrigated with final (FSTW) and biological (BSTW) secondary treated wastewater was presented in Table 6.The highest DIM results in both adults and children measured in zinc which were 3.987E-05 and 4.583E-05. Besides, DIM results are higher in plants grown with hydroponic cultivation as compared to which found in soil cultivation.

The DIM of Fe varied from 5.246E-07 to 7.344E-06, Ni 2.098E-06 and Zn 1.626E-05 to 3.987E-05 respectively for adults. Meanwhile, the DIM of Fe varied from 6.030E-07 to 8.443E-06,Ni 2.412E-06 and Zn 1.869E-05 to 4.583E-05 respectively for children. However, none DIM was detected for Cd and Pb. In addition, children posed to higher health risks than adults since the daily intake metals via consumption of plants found to be significantly higher for children as compared to adults. Furthermore, the HRI of Fe varied from, 7.494E-07 to 1.049E-05, Ni 1.049E-04 and Zn 5.421E-05 to 1.329E-04 respectively for adults. In the meantime, the HRI of Fe varied from Fe 8.615E-07 to 1.206E-05, Ni 1.206E-04 and Zn 5.427E-5 to 1.528E-04 respectively for children. On the other hand, none DIM was detected for Cd and Pb. However, the analysis indicated that the HRI values were < 1. Thus, the risk associated to trace element contamination through consumption of plants irrigated with secondary treated wastewater by adults and children was no consequences and assumed to be safe in general.

Table 6: Daily Intake of Metals (DIM) and Health Risk Index (HRI) for adults and children

	C metal (mean)	DIM (adults)	DIM (children)	HRI (adults)	HRI (children)	Remarks
			Cadmium, Cd (RfD=0.001 mg/kg/day)			
FSTW-SEPS	ND	NT	NT	NT	NT	NT
BSTW-SEPS	ND	NT	NT	NT	NT	NT
FSTW-SPS	ND	NT	NT	NT	NT	NT
BSTW-SPS	ND	NT	NT	NT	NT	NT
FSTW-HNS	ND	NT	NT	NT	NT	NT
BSTW-HNS	ND	NT	NT	NT	NT	NT
FSTW-HWS	ND	NT	NT	NT	NT	NT
BSTW-HWS	ND	NT	NT	NT	NT	NT
			Iron, Fe (RfD=0.7 mg/kg/day)			
FSTW-SEPS	0.005	2.623E-06	3.015E-06	3.747E-06	4.307E-06	Safe
BSTW-SEPS	0.005	2.623E-06	3.015E-06	3.747E-06	4.307E-06	Safe
FSTW-SPS	0.004	2.098E-06	2.412E-06	2.998E-06	3.446E-06	Safe
BSTW-SPS	ND	NT	NT	NT	NT	NT
FSTW-HNS	0.014	7.344E-06	8.443E-06	1.049E-05	1.206E-05	Safe
BSTW-HNS	0.004	2.098E-06	2.412E-06	2.998E-06	3.446E-06	Safe
FSTW-HWS	0.02	1.049E-05	1.206E-05	1.499E-05	1.723E-05	Safe
BSTW-HWS	0.001	5.246E-07	6.030E-07	7.494E-07	8.615E-07	Safe

Table 6: continue.

	C metal (mean)	DIM (adults)	DIM (children)	HRI (adults)	HRI (children)	Remarks
			Nickel, Ni (RfD=0.02 mg/kg/day)			
FSTW-SEPS	0.004	2.098E-06	2.412E-06	1.049E-04	1.206E-04	Safe
BSTW-SEPS	0.004	2.098E-06	2.412E-06	1.049E-04	1.206E-04	Safe
FSTW-SPS	0.004	2.098E-06	2.412E-06	1.049E-04	1.206E-04	Safe
BSTW-SPS	0.004	2.098E-06	2.412E-06	1.049E-04	1.206E-04	Safe
FSTW-HNS	0.004	2.098E-06	2.412E-06	1.049E-04	1.206E-04	Safe
BSTW-HNS	0.004	2.098E-06	2.412E-06	1.049E-04	1.206E-04	Safe
FSTW-HWS	0.004	2.098E-06	2.412E-06	1.049E-04	1.206E-04	Safe
BSTW-HWS	0.004	2.098E-06	2.412E-06	1.049E-04	1.206E-04	Safe
			Lead, Pb (RfD=0.0035 mg/kg/day)			
FSTW-SEPS	ND	NT	NT	NT	NT	NT
BSTW-SEPS	ND	NT	NT	NT	NT	NT
FSTW-SPS	ND	NT	NT	NT	NT	NT
BSTW-SPS	ND	NT	NT	NT	NT	NT
FSTW-HNS	ND	NT	NT	NT	NT	NT
BSTW-HNS	ND	NT	NT	NT	NT	NT
FSTW-HWS	ND	NT	NT	NT	NT	NT
BSTW-HWS	ND	NT	NT	NT	NT	NT
			Zinc, Zn (RfD=0.3 mg/kg/day)			
FSTW-SEPS	0.034	1.784E-05	2.050E-05	5.945E-05	6.835E-05	Safe
BSTW-SEPS	0.031	1.626E-05	1.869E-05	5.421E-05	6.232E-05	Safe
FSTW-SPS	0.031	1.626E-05	1.869E-05	5.421E-05	6.232E-05	Safe
BSTW-SPS	0.039	2.046E-05	2.352E-05	6.820E-05	7.840E-05	Safe
FSTW-HNS	0.041	2.151E-05	2.472E-05	7.170E-05	8.242E-05	Safe
BSTW-HNS	0.04	2.098E-05	2.412E-05	6.995E-05	8.041E-05	Safe
FSTW-HWS	0.076	3.987E-05	4.583E-05	1.329E-04	1.528E-04	Safe
BSTW-HWS	0.043	2.256E-05	2.593E-05	7.519E-05	8.644E-05	Safe

ND=None detected, NT= None tested, Human are considered to be safe if HRI<1

CONCLUSIONS

The present of trace elements in both types of secondary treated wastewater had believed to provide an agronomic benefit. However, both FSTW and BSTW demonstrated the significance increase in term of trace elements concentrations in irrigated soil and plants which may contribute to detrimental effects to people. Meanwhile, trace element concentrations were found higher in plant growth under soilless culture as compared to soil culture. Therefore, the application of wastewater irrigation with soil culture provides a minimal risk for accumulation and translocation of trace elements in plants as compared to soilless culture. However, the analysis indicated that the HRI values were < 1. Thus, the risk associated to trace elements contamination through consumption of plants irrigated with secondary treated wastewater by adults and children was no consequences and assumed to be safe in general. Yet, proper monitoring and continuous assessments are required in order to prevent any long term risk associated to wastewater irrigation application.

ACKNOWLEDGEMENT

The authors would like to address special thanks to Faculty of Civil Engineering, Faculty of Plantation and Agrotechnology, Faculty of Chemical Engineering and Facility Office Management, Universiti Teknologi MARA, Shah Alam for providing facilities.

REFERENCES

[1] Horswell, J., Speir, T. W., and van Schaik, A. P. (2003). Bio-indicators to assess impacts of heavy metals in land-applied sewage sludge. *Soil Biology and Biochemistry* , 35, pp.1501-1505.

[2] Kiziloglu, F. M., Turan, M., Sahin, U., Kuslu, Y., and Dursun, A. (2008). Effects of untreated and treated wastewater irrigation on some chemical properties of cauliflower (Brassica olerecea L. var. botrytis) and red cabbage (Brassica olerecea L. var. rubra) grown on calcareous soil in Turkey. *Agricultural Water Management* , 95, pp.716-724.

[3] Mohammad Rusan, M. J., Hinnawi, S., and Rousan, L. (2007). Long term effect of wastewater irigation of forage crops on soil and plant quality parameters. *Desalination* , 215, pp.143-152.

[4] Emongor, V. E., and Ramolemana, G. M. (2004). Treated sewage effluent (water) potential to be used for horticultural production in Botswana. *Physics and Chemistry of the Earth* , 29, pp.1101-1108.

[5] Sanderson, K. C. (1986). Introduction to the workshop on wastewater utilization in horticulture. *HortScience* , 21, pp.23-24.

[6] Pedrero, F., and Alarcon, J. J. (2009). Effects of treated wastewater irrigation on lemon trees. *Desalination* , 246, pp.631-639.

[7] Omran, M. S., Waly, T. M., Abd Elnaim, E. M., and El Nashar, B. B. (1988). Effect of sewage irrigation on yield tree components and heavy metal accumulation in Navel orange trees . *Biological Wastes* , 23, pp.17-24.

[8] Yadav, R. K., Goyal, B., Sharma, R. K., Dubey, S. K., and Minhas, P. S. (2002). Post-irrigation impact of domestic sewage effluent on composition of soils, crops and ground water- A case study. *Environment International* , 28, pp.481-486.

[9] Banin, A. J., Naverot, Y. N., and Yoles, D. (1981**).** Accumulation of heavy metal in arid zone soils irrigated with treated sewage effluent and their uptake by Rhodes grass. *Journal of Environmental Quality* , 10 (4), pp.536-540.

[10] Dominguez-Mariana, E., Carrillo-Chvez, A., and Ortega, A. (2004). Wastewater reuse in valsequillo agricultural area,Mexico:environmental impact on groundwater. *Water, Air and Soil Pollution* , 155, pp.251-267.

[11] Yang, H. X. (2002). Effects of irrigation with wastewater on crops in Datong, Shanxi China. *Agro-Environment and Development* , 4, pp.18-19.

[12] Chang, A. C., Granato, T. C., and Page, A. L. (1992). The methodology for establishing phytotoxicity criteria for Cr,Cu, Ni and Zn in agricultural land application of municipal sewage sludge . *Journal of Environmental Quality* , 21, pp.521-536.

[13] De Miguel, E., Jimenez de Grado, M., Llamas, J. F., and Martin-Dorado, A. (1998). The overlooked contribution of compost application to the trace element load in the urban soil of Madrid (Spain) . *The Science of the Total Environment* , 215, pp.113-122.

[14] Mielke, H. W., Gonzales, C. R., Smith, M. K., and Mielke, P. W. (1999). The urban environment and children health: soils as an integrator of lead, zinc and cadmium in New Orleans, Louisiana, USA. *Environmental Research* , 81, pp.117-129.

[15] Madrid, L., Diaz-Barrientos, E., and Madrid, F. (2002). Distribution of heavy metals contents of urban soils in parks of Seville. *Chemosphere* , 49, pp.1301-1308.

[16] Mapanda, F., Mangwayana, E. N., Nyamangara, J., and Giller, K. E. (2005). The effect of long-term irrigation using wastewater on heavy metal contents of soils under vegetables in Harare, Zimbabwe. *Agriculture,Ecosystems and Environment* , 107, pp.151-165.

[17] Ali M.F. and Shakrani S.A. (2010).The Effects of Treated Wastewater Effluent on Crops Growth : A Study Using Two Different Planting Media. *Proceedings of Malaysian Science and Technology Congress 2010 – Scaling New Height in S & T for Sustainable National Development*. Crystal Crown Hotel, Petaling Jaya, Malaysia. 9 – 11 November 2010.

[18] Ali, M.F., and Shakrani, S.A. (2011). Soil and Soilless Cultivation Influence on Nutrients and Heavy Metals Availability in Soil and Plant Uptake. *International Journal of Applied Science and Technology,* 1(5), pp.154-160.

[19] Shakrani S.A. and Ali M.F. (2011).Soil and Soilless Cultivation Influence on Nutrients and Heavy Metals in Soil and Plant Uptake. *Proceedings of National Postgraduate Seminar 2011 – Empowering Upcoming Engineers, Builders of the Future.* UiTM Shah Alam, Malaysia. 14 – 15 Jun 2011.

[20] Shakrani S.A. and Ali M.F. (2012). The Effects of Secondary Treated Wastewater on *Brassica Campestris Sp. Parachinensis* Growth Using Soil and Hydroponic Cultivation. *Proceedings of National Postgraduate Seminar 2012 – Challenges and Prospects in Science and Technology for Environmental Sustainability*. UiTM Shah Alam, Malaysia. 16 – 18 July 2012.

[21] American Public Health Association (APHA), American Water Works Association (AWWA)., & Water Environment Federation (WEF). (2005). *Standard methods for the examination of water and wastewater* (21th ed.). Washington D.C: American Public Health Associated.

[22] BS EN ISO 5667-1. (2006). Water quality-Sampling-Part 1: Guidance on the design of sampling programmes and sampling techniques. British Standards Institution.

[23] BS 6068-6.10. (1993). Water quality-Part 6: Sampling-Section 6.10 Guidance on sampling of waste waters. British Standards Institution.

[24] BS ISO 10381-1. (2002). Soil quality – Sampling – Part 1: Guidance on the design of sampling programmes. British Standards Institution.

[25] BS ISO 10381-2. (2002). Soil quality – Sampling – Part 2: Guidance on sampling techniques. British Standards Institution.

[26] ASTM D5633. (2008). Standard practice for sampling with a scoop. American Society for Testing and Materials

[27] ISO 874 . (1980). Fresh fruits and vegetables – Sampling. International Organization For Standardization.

[28] HACH Company. (1999). Models 23130-20 Digesdahl® Digestion Apparatus Instruction Manual. *Hach Company***,** Loveland CO.

[29] Jones, J.B., and Case, V.W.(1990**).** Sampling, handling and analyzing plant tissue samples. In: Westerman, R.L. (Ed.), *Soil Testing and Plant Analysis*. third ed., Soil Science Society of America, Book Series No. 3, Madison, Wisconsin, pp. 389–427.

[30] Brayton, S.V. (1992). Acid digestions using the Hach Digesdahl Apparatus : Sample preparation for protien and elemental analysis. *Technical information series-booklet no.14***,** Hach Company, Loveland, CO.

[31] Tuzen, M. (2003). Determination of heavy metals in soil, mushroom and plant samples by atomic absorption spectrometry**.** *Microchemical Journal***,** 74, pp.289-297.

[32] Hseu, Z.Y., (2004). Evaluating heavy metal contents in nine composts using four digestion methods. *Bioresource Technology*, 95, 53-59.

[33] Khan,S.,Cao,Q.,Zheng,Y.M.,Huang,Y.Z.,and Zhu,Y.G. (2008). Health risks of heavy metals in contaminated soils and food crops irrigated with wastewater in Beijing, China. *Environmental Pollution* , 152, pp.686-692.

[34] Arora, M., Kiran, B., Rani, S., Rani, A., Kaur, B., and Mittal, N. (2008). Heavy metal accumulation in vegetables irrigated with water from different sources. *Food Chemistry* , 111, pp.811-815.

[35] Rattan, R. K., Datta, S. P., Chhonkar, P. K., Suribabu, K., and Singh, A. K. (2005). Long-term impact of irrigation with sewage effluents on heavy metal content in soils, crops and groundwater-a case study. *Agricultural, Ecosystems and Environment* , 109, pp.310-322.

[36] Wang, X., Sato, T., Xing, B.,and Tao, S. (2005). Health risks of heavy metals to the general public in Tianjin, China via consumption of vegetables and fish. Science of the Total Environment, 350, 28–37.

[37] United States Environmental Protection Agency (USEPA). (2012). http://www.epa.gov/ (Accessed 01 October 2012).

[38] Joint FAO/WHO expert Committee on Food Additives (JECFA).(1993).Evaluation of certain food additives and contaminants: 41st report of the Joint FAO/WHO expert Committee on Food Additives. Geneva: World Health Organization. Technical Reports Series No. 837.

[39] Cui, Y.J., Zhu, Y.G., Zhai, R.H., Chen, D.Y., Huang, Y.Z., and Qui, Y.(2004). Transfer of metals from soil to vegetables in an area near a smelter in Nanning, China. *Environment International* , 30(6), pp.785– 791.

[40] Malaysia Environmental Quality Act .(2009). Environmental Quality (Industrial Effluent) Regulation 2009.

[41] WHO. (2006). WHO guidelines for the safe use of wastewater, excreta and greywater-Volume 2 Wastewater use in agriculture. World Health Organization.

[42] Food and Agriculture Organization of the United Nations (FAO). (1985).Water quality for agriculture. (FAO Irrigation and Drainage Paper 29)

[43] United States Environmental Protection Agency (USEPA). (2004). Guidelines for Water Reuse. EPA 645-R-04-108. United States Environmental Protection Agency, Washington, D.C.

[44] Salt, D. E., Blaylock, M., Kumar, N.P.B.A., Dushenkov, V., Ensley, D., Chet,I., and Raskin,I. (1995).Phytoremediation: a novel strategy for the removal of toxic metals from the environment using plants. *Biotechnology*, 13(5), pp.468–474.

[45] Peralta-Videa, J.R., Lopez, M.L., Narayan, M., Saupe, G., and Gardea-Torresdey, J. (2009).The biochemistry of environmental heavy metal uptake by plant: Implications for the food chain. *The International Journal of Biochemistry & Cell Biology*, 41, pp.1665-1677.

[46] McLean, J. E, & Bledsoe, B. E. (1992). *Behavior of metals in soils.* [Washington, DC: Technology Innovation Office, Office of Solid Waste and Emergency Response .

[47] Nagajyoti, P.C., Lee,K.D., and Sreekanth, T.V.M. (2010). Heavy metals, occurrence and toxicity for plants : a review. *Environmental Chemistry Letters*, 8, pp.199-216.

[48] Hänsch, R., and Mendel, R.R.(2009). Physiological functions of mineral micronutrients (Cu, Zn,Mn,Fe,Ni,Mo,B,Cl).*Current Opinion in Plant Biology*, 12, pp.259-266.

[49] Marschner, M. (2012). *Mineral Nutrition in Higher Plants* (3rd Edition). London:Academic Press.

[50] Yamauchi, M. (1989). Rice bronzing in Nigeria caused by nutrient imbalances and its control by potassium sulfate application. *Plant and Soil* , 117, pp.275–286.

[51] Sinha S, Guptha M, and Chandra P (1997) .Oxidative Stress induced by iron in Hydrilla verticillata (i.f) Royle: response of antioxidants. Ecotoxicology and Environmental Safety, 38 (3), pp.286–291.

[52] Arora, A., Sairam, R.K., and Srivastava, G.C. (2002). Oxidative stress and antioxidative system in plants. *Current Science*, 82 (10), pp.1227–1338.

[53] De Dorlodot, S., Lutts, S., and Bertin, P. (2005). Effects of ferrous iron toxicity on the growth and mineral composition of an inter specific rice. Journal of Plant Nutrition, 28 (1), pp.1–20.

[54] Eskew,D.L., Welch,R.M., and Norvell,W.A. (1984). Nickel in higher plants: further evidence for an essential role. *Plant Physiology,* 76, pp.691-693.

[55] Brown, P.H., Welch, R.M., and Cary, E.E. (1987). Nickel : A micronutrient essential for higher plants. *Plant Physiology*, 85, pp.801-803.

[56] Seregin,I.V., and Kozhevnikova.A.D. (2006).Physiological role of nickel and its toxic effects on higher plants. *Russian Journal of Plant Physiology*, 53(2), pp.257-277.

[57] Welch, R. M. (1981). The biological significance of nickel. *Journal of Plant Nutrition*, 3(1-4), pp. 345–356.

[58] Marschner, H. and Römheld, V. (1983). *In vivo* measurement of root-induced pH changes at the soil-root interface: effect of plant species and nitrogen source. *Zeitschrift für Pflanzenphysiologie.* 111(3), pp.241–251.

[59] Cakmak, I., and Marschner, H.(1993).Effect of zinc nutritional status on superoxide radical and hydrogen peroxide scavenging enzymes in bean leaves. *Plant and Soil*, 155-156 (1), pp.127-130.

[60] Zhang, F., Römheld, V. and Marschner, H. (1991). Release of zinc mobilizing root exudates in different plant species as affected by zinc nutritional status. *Journal of Plant Nutrition*, 14(7), pp.675–686.

[61] Ruano, A., Poschenrieder, Ch. and Barcelo, J. (1988). Growth and biomass partitioning in zinc-toxic bush beans. *Journal of Plant Nutrition*, 11(5), pp.577–588.

[62] Prasad,K.V.S.K., Pardha Saradhi, P., and Sharmila, P. (1999). Concerted action of antioxidant enzyme and curtailed growth under zinc toxicity in Brassica juncea. *Environmental and Experimental Botany*, 42,pp.1–10.

[63] Godbold, D. L., Horst, W. J., Marschner, H., Collins, J. C. and Thurman, D. A. (1983). Root growth and Zn uptake by two ecotypes of *Deschampsia caespitosa* as affected by high Zn concentrations. *Zeitschrift für Pflanzenphysiologie.* 112(4), pp.315–324.