

Tire wear particles co-exposure with Nickel inhibits mung bean yield by reducing nutrient uptake

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Figure S6: Homeostasis of inorganic nutrients in soil (A) and mungbean grain (B) Under the application of NiSO₄ and TWPs. The numbers show the fold change of elemental concentration as compared to control. The negative value highlights the concentration was decreased while the positive value shows the concentration was increased. * Indicates a significant difference at $p < 0.05$ ($n = 3$) compared with the control evaluated by one-way ANOVA analysis.

Figure S7: Correlations of plant physiological parameters, yield parameter, nutritional and antioxidant enzymes with TWPs and Ni. Principal component analysis (A and B) of plant physiological parameters, yield parameters, nutritional and antioxidant enzymes under different TWPs (0.01, 0.1 and 0.25% w/w) and Ni (50 and 100 mg kg⁻¹) are presented.

Figure S8: Fourier transform infrared spectroscopy of Tire wear particles.

Table.

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Text S1. Hydroponic experiment.

The exposure of TWPs and Ni experiments were conducted in an incubator at China Agriculture University Beijing. The *Vigna radiata* seeds were purchased from the Chinese academy of Agriculture Science. The mung bean seeds were sterilized with 3% sodium hypochlorite for ten minutes and subsequently rinsed three times with distilled water before sowing. The seeds were grown in petri dish for germination. After 3 days of seed was transferred to filter paper and put in the beaker for further growth and distal water was added. Two leaves of the plant were transferred to 250 ml of bottle with Hoagland solution. After 30 day the water uptake data was recorded over three days of intervals up to 60 days and average data were presented.

Text S2. Correlation based principal component analysis

The correlation based principal component analysis (PCA) was performed to determine the response pattern of mungbean yield, nutritional balance and antioxidant enzymes to various TWPs and Ni concentrations. The loading plots of PCA are given in Fig. S8A and S8B. Among all the components of PCA, PC1 and PC2 showed maximum contribution and exhibited 73.8% accounted by PC1 and 16.2% by PC2 accordingly. The distribution of the variables in PCA indicated that various TWPs and Ni significantly affected the mungbean yield, nutritional balance antioxidant enzymes. The larger distance of PCA antioxidant enzyme variables between TWPs and Ni exhibited that mungbean were highly responsive to various TWPs and Ni concentrations. The group of variables in PC1 that correlates positively includes antioxidant enzymes, Ni contents in plant while a significant negative correlation of variables in PC1 was found with variables aligned in PC2.

PCA and Pearson correlation matrix indicates strongly correlating variables across treatments (Fig. S8). In our experiment, out of the 88 % of total variance represented by the two components, 80.1% variance participated by PC1 and 17.7% by PC2. The PCA analysis showed that most of the antioxidant and Ni contents were strongly positively correlated with Ni 100 mg kg⁻¹ alone and co-exposure with TWPs concentrations and contributed to the total variations explained by PC1. Furthermore, SOD, POD, CAT, and MDA significantly indicated a strong correlation in Ni 100 mg kg⁻¹. However, most of the TWPs (0.01%) and Ni 50 mg kg⁻¹ related parameters contributed to PC2 which was mainly associated with growth and yield parameters of mungbean. Two exposure levels of TWPs and one exposure level of Ni inducing the toxicity to

the mungbean were identified, showing 80% of the original data structure (Table S8C and S8D). Values in PC1 (orange color) correspond for each observation to the factor that may control the major toxicity mechanism. Pearson correlation matrix indicated some of the strongly correlating variables in the mungbean. A strong and positive correlation was observed between SOD ($R^2 = 0.335$), POD ($R^2 = 0.3783$), CAT ($R^2 = 0.4452$), MDA ($R^2 = 0.4809$), soil Ni ($R^2 = 0.3389$) Root Ni ($R^2 = 0.2765$), Shoot Ni ($R^2 = 0.1715$), Grain Ni ($R^2 = 0.2026$).

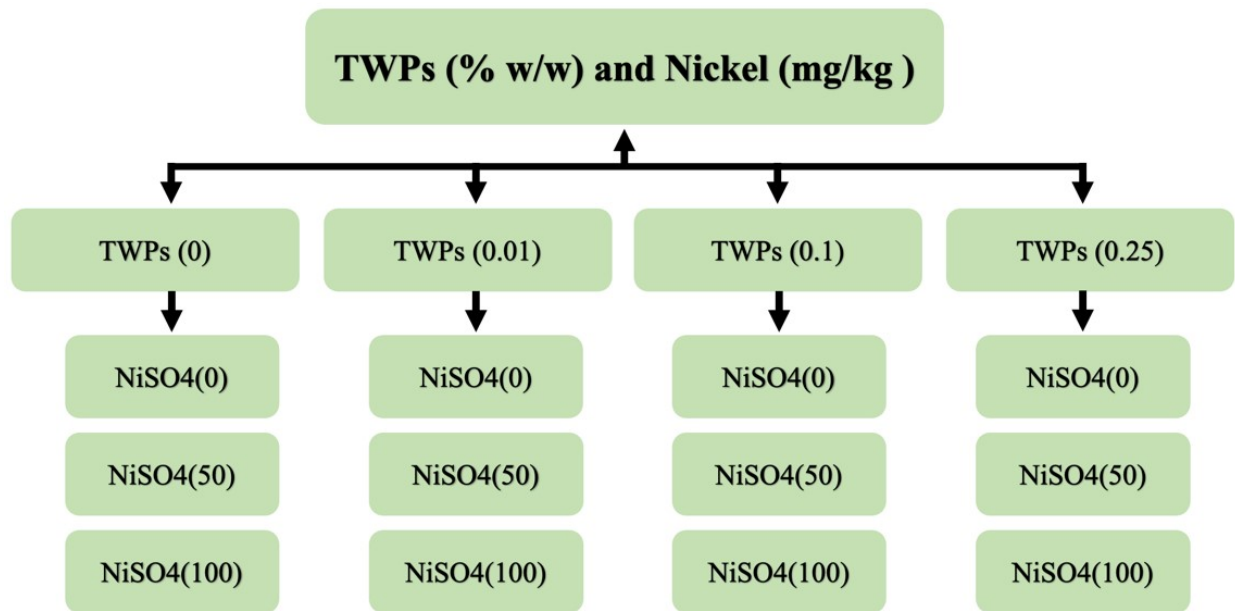
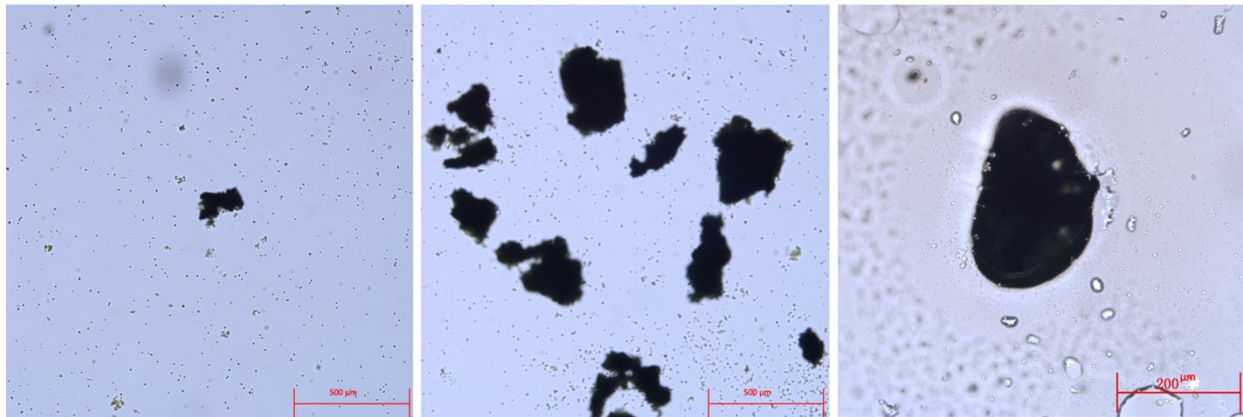


Figure S1: Exposure conditions involving the treatment combination of TWPs and Ni, both individually and in co-exposure.

A)



B)

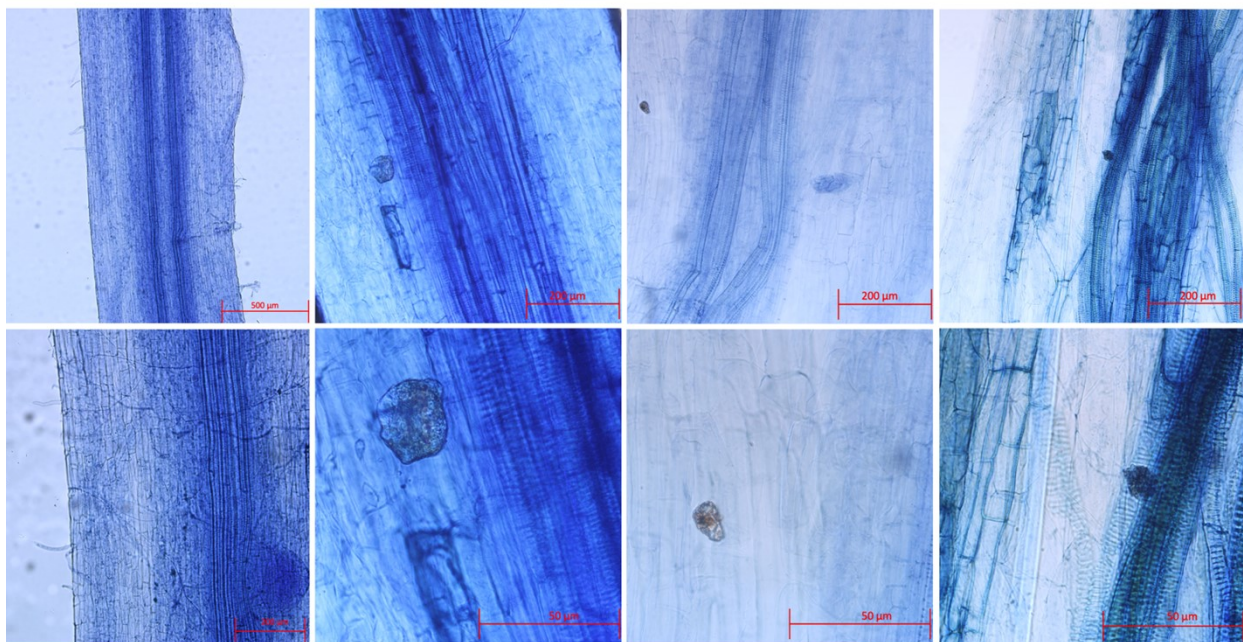


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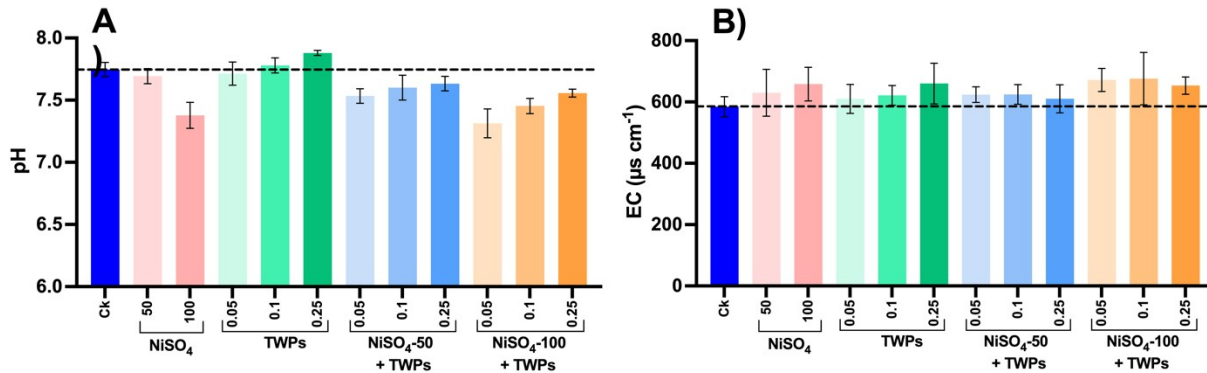


Figure S3. Effects of Ni and TWP on pH and EC over 105 days of exposure. (A) represents the pH and (B) represents EC. Error bars represent the standard deviation of triplicate replication in each treatment.

1. Effect of TMP and NiSO₄ on germination, pod length and chlorophyll contents

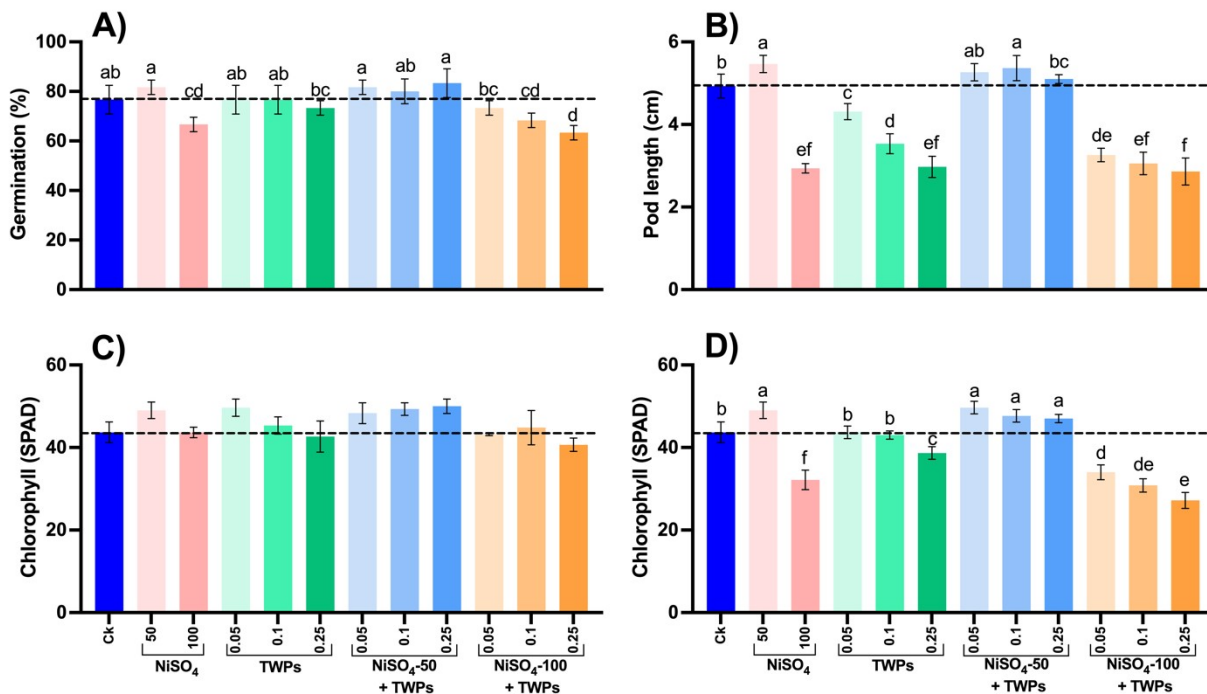


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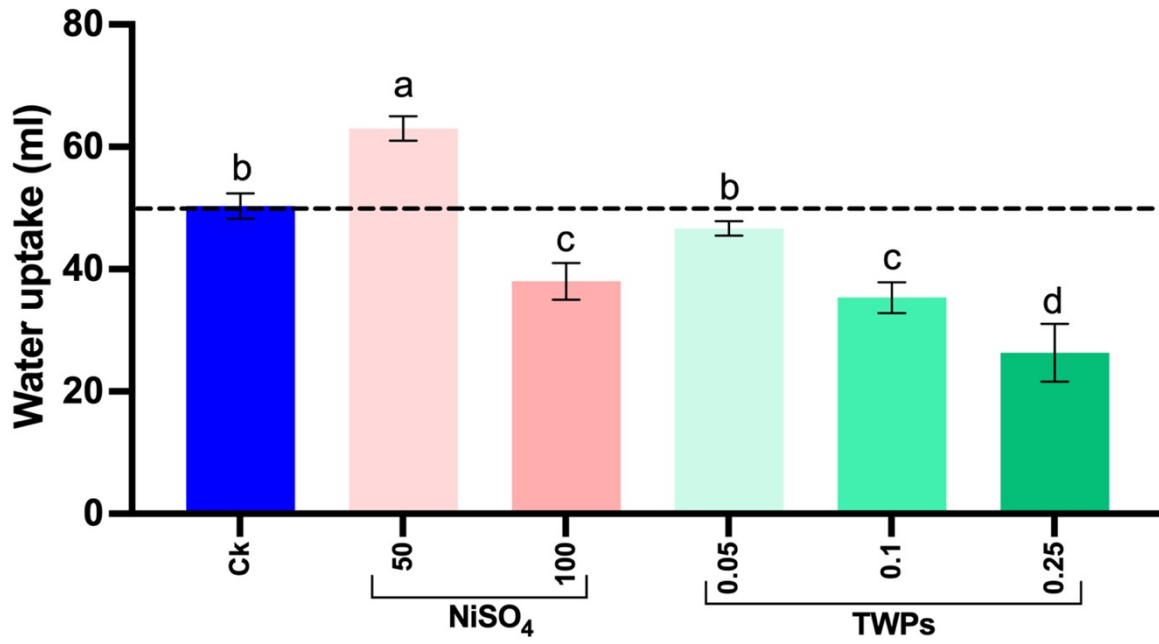


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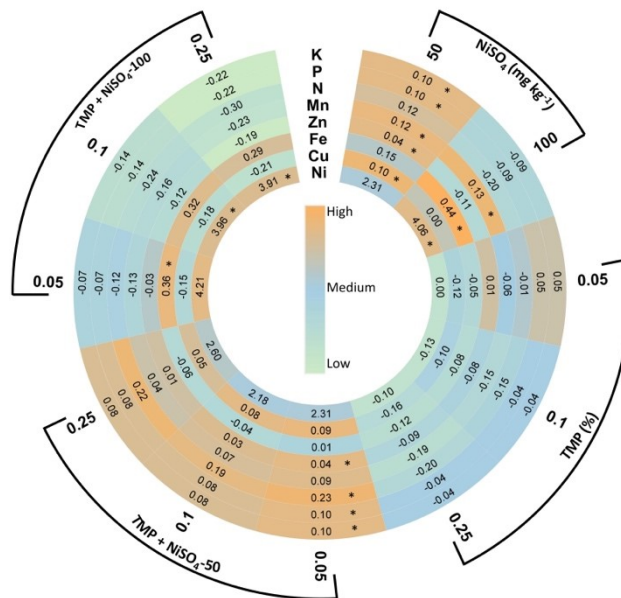


Figure S6: Homeostasis of inorganic nutrients in soil (A) and mungbean grain (B) Under the application of NiSO₄ and TWPs. The numbers show the fold change of elemental concentration as compared to control. The negative value highlights the concentration was decreased while the

positive value shows the concentration was increased. * Indicates a significant difference at $p < 0.05$ ($n = 3$) compared with the control evaluated by one-way ANOVA analysis.

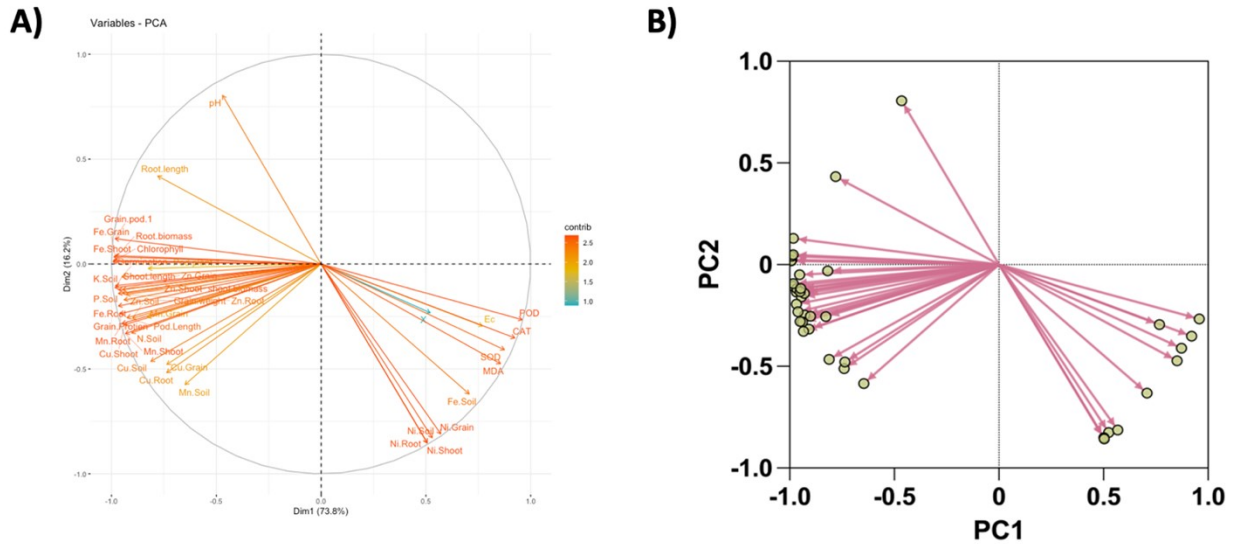


Figure S7: Correlations of plant phycological parameters, yield parameter, nutritional and antioxidant enzymes with TWPs and Ni. Principal component analysis (A and B) of plant phycological parameters, yield parameters, nutritional and antioxidant enzymes under different TWPs (0.01, 0.1 and 0.25% w/w) and Ni (50 and 100 mg kg⁻¹) are presented.

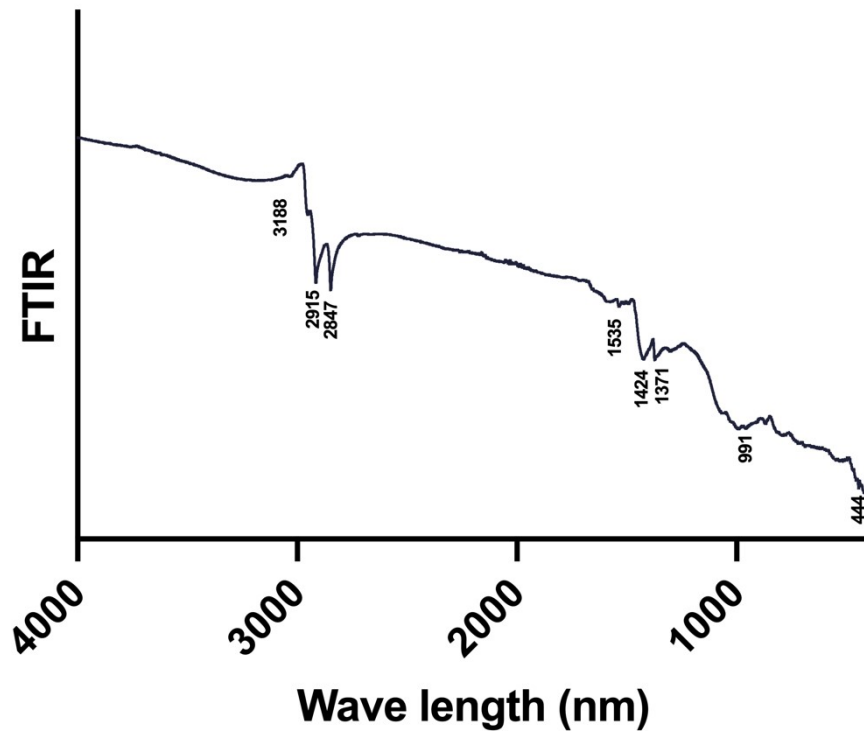


Figure S8: Fourier transform infrared spectroscopy of Tire wear particles.

Table S1. Soil physiochemical properties.

Parameters	value
Soil analysis	
Soil Texture	Loam soil
pH	7.8
EC (1:5) (dSm ⁻¹)	0.27
Organic matter (%)	0.95
Calcium carbonate (%)	15.2
Total Nitrogen (%)	0.09
Available Nitrogen (mg kg ⁻¹)	17
Available phosphorus (mg kg ⁻¹)	4.23
Available potassium (mg kg ⁻¹)	126.5
Nickel (mg kg ⁻¹)	8.21
Copper (mg kg ⁻¹)	6.2
Iron (mg kg ⁻¹)	1385
Zinc (mg kg ⁻¹)	7.64
Manganese (mg kg ⁻¹)	39.47
Tire Wear particle analysis	
Nickel (mg kg ⁻¹)	4.36
Copper (mg kg ⁻¹)	2.51
Iron (mg kg ⁻¹)	8.03
Zinc (mg kg ⁻¹)	7527
Manganese (mg kg ⁻¹)	3.27

Table S2. List of detection, precision and recovery data for AAS for the selected elements. certified reference material (CRM)

Elements	List of detection (µg/L)	Spiking recovery (%)	Recovery from CRM (%)	Correlation coefficient (R ²)	Correlation variance (%)
Ni	0.0120	93	95	0.9716	2.07
Cu	0.1846	91.6	98	0.9906	3.72
Fe	0.0270	100.6	98	0.9909	3.21
Zn	0.0065	91.6	96	0.9906	4.32
Mn	0.0090	86.6	95	0.9989	3.44