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Mineral uptake in *Solanum nigrum* L. cultivated on fertiliser amended soils of the Eastern Cape, South Africa

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Summary

A considerable interest has been manifested in the cultivation of wild vegetables to combat the ever increasing hunger and micronutrient deficiencies especially in children from the developing world. *Solanum nigrum*, one of the popular wild vegetables consumed in the Eastern Cape was cultivated on sandy loam soils to determine the effect of fertilisers on Cu, Fe, Mn and Zn uptake in relation to its growth stages. Five treatments (control; 100 kg N/ha; 8.13 t manure/ha; 100 kg N/ha + 8.13 t manure/ha and 50 kg N/ha + 4.07 t manure/ha) were arranged in plots in a Randomised Complete Block Design with five replicates. Cu (mg/kg) remained variably high throughout the trial, ranging between 7.20-23.50 on the field and 5.30-21.40 in the glasshouse. Fe (mg/kg) also remained variably high, ranging between 213-766 on the field and 178-523 in the glasshouse. Mn (mg/kg) increased with increasing plant maturity and ranged between 85-222 on the field and 64-215 in the glasshouse, but Zn (mg/kg) decreased with plant maturity and ranged between 33-78 on the field and 16-74 in the glasshouse. The results indicate that *Solanum nigrum* has the potential to supply the recommended daily micronutrient intake values throughout all its growth stages.

Introduction

There is no doubt that one of the causative factors to low nutritional status of food plants is poor soil fertility. Plants obtain their nutrients from the soil on which they are cultivated. The replenishment of nutrients to low yielding soils through the addition of fertilisers often leads to better performance of food plants.

Nutrient uptake and composition of food plants has for a long time been a subject of interest among researchers. Food plants play a critical role in human life by supplying the much needed nutritional elements as well as boosting the human immune system. However, most food plants are currently based on a limited number of crops and these comprise about 103 plant species which contribute 90 % of national per capita supplies of food plants (Prescott-Allen and Prescott-Allen, 1990). Although wild vegetable plants were an important part of traditional agricultural systems, their consumption has over the years declined in South Africa due to their association with poverty and poverty foods, degree of urbanisation, modernisation of agriculture, distance to fresh produce markets and season of the year, among other factors (Flyman and Afolayan, 2007; Jansen van Rensberg et al., 2007). According to Modi et al., (2006), the poor utilisation of wild vegetables may be associated with a lack of knowledge about how to access quantities that can satisfy daily human food requirements. Nevertheless, in many parts of the world, including South Africa, the use of wild vegetables is not negligible (Misra et al., 2008).

One of the major health problems affecting children in South Africa is micronutrient deficiency and these have been well documented in South Africa particularly for Fe, Zn and Cu (Faber and Wenhold, 2007; Voster, 2010). The rich nutritional value of wild vegetables has also been documented by many authors (Edmonds and Chweya, 1997; Flyman and Afolayan, 2007; Odhav et al., 2007; Lewu and Mavengahama, 2010). Faber and Wenhold (2007) suggested the cultivation of wild vegetables as one of the strategies that can be adopted to address micronutrient malnutrition.

Wild vegetables usually grow as volunteer plants alongside conventional crops in agricultural fields during the planting season and are mostly viewed as weeds that must be removed usually by mechanical or chemical means. Although they are viewed as weeds, the cultivation of wild vegetables has been documented in South Africa for some species, for example, *Amaranthus* and *Brassica* (Jansen van Rensberg et al., 2007; Husselman and Sizane, 2006). However, agronomic data required to cultivate wild vegetable plants is scanty. A preliminary survey of wild vegetables consumed in the Eastern Cape Province of South Africa, where the current study was conducted indicated that *S. nigrum* is one of the popular wild vegetables gathered from the wild for food and consumed by the province's rural populace (Bvenura and Afolayan, 2014). This wild vegetable was therefore cultivated in the glasshouse and on the field under different concentrations of organic and/or inorganic fertilisers with a view to determine the wild vegetable's response to these fertilisers and also determine the best fertiliser option for its cultivation as well as the best time to harvest the leaves for extraction of the micronutrients; Zn, Fe, Cu and Mn.

Materials and methods

The Experimental site

The experiment was conducted in the glasshouse and on the field at the University of Fort Hare, Alice campus, South Africa between September and December 2012. The study area falls under 32° 47' S and 26° 50' E and 535 m a.s.l. and is within a semi arid ecological zone with an average annual rainfall of approximately 575 mm in summer; mean daily temperatures of 22.5 °C during the day and 18.8 °C at night while during the winter the temperature is about 13.6 °C during the day and less than 10.3 °C at night (Marais and Brutsch, 1994). According to the South African system of soil classification, the soils are deep alluvial; of the Oakleaf form (Oa) and belong to the Jozini series and are texturally sandy loam (Soil Classification Working Group, 1991). According to the soil map of the world, the soils are Eutric fluvisols (Fle) (FAO-UNESCO-ISRIC, 1988). The properties of the soil, used for this experiment are shown in Tab. 1.

Agronomic practices

Ripe and mature *S. nigrum* berries were harvested between the 3rd and 26th of April 2012 from the wild in Alice. The seeds were separated from the pulp, washed in distilled water and dried at room temperature on the laboratory bench for 2 h and kept in sealed bottles until further use. The extracted seeds were planted in cavity trays in the glasshouse and later transplanted to the field when they were

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about 6 weeks old. For the glasshouse trial, the seedlings were transplanted into prepared polythene bags containing 5 kg of soil. The soil used in the glasshouse was obtained from the field where the field trial was conducted to ensure consistency in soil properties from the two trials. The organic fertiliser (goat manure) used in this experiment was obtained from the University of Fort Hare animal farm while the inorganic fertilisers (NPK [2:3:4] and Limestone Ammonium Nitrate [LAN]) were purchased from a local fertiliser dealer. The properties of the organic fertiliser used for the experiment are shown in Tab. 1.

Tab. 1: The chemical properties of the experimental soil (Upper 0-30 cm depth) and organic fertiliser

	Soil	Organic fertiliser
pH(KCI)	6.54	7.17
Bulk density (g cm^{-3})	1.20	-
EC (μS/cm)	162.05	10.75
CEC_{sum} (meq/ 100g)	12.10	-
Available P (mg kg^{-1})	71	8 500
Exchangeable K (mg kg^{-1})	406	26 000
Exchangeable Ca (mg kg^{-1})	1653	29 700
Exchangeable Mg (mg kg^{-1})	335	9 900
Exchangeable acidity (cmol/L)	0.06	-
Total cations (cmol/L)	12.10	-
Saturated acid (%)	0	-
Zn (mg kg^{-1})	10.2	172
Mn (mg kg^{-1})	17	582
Cu (mg kg^{-1})	5.7	54
Organic C (mg kg^{-1})	10000	-
N (mg kg^{-1})	1400	24 800
Clay (%)	17	-
Na (mg kg^{-1})	-	1 564
Fe (mg kg^{-1})	-	12 439
Al (mg kg^{-1})	-	5 335

Experimental design

The experiments were laid out in a Randomised Complete Block Design (RCBD) with five treatments and five replicates. The treatments were: Control (T1); 100 kg N/ha (T2); 8.13 t manure/ha (T3); 100 kg N/ha + 8.13 t manure/ ha (T4) and 50 kg N/ha + 4.07 t manure/ ha (T5). Nitrogen was supplied in the form of NPK and LAN fertilisers. The organic fertiliser (goat manure) and NPK were applied at transplanting and LAN fertiliser applied 4 weeks after transplanting. These fertilisers were applied in the top 5-7 cm of soil depth by mixing with a spade in plots measuring 3 m × 2 m on the field. In the glasshouse, the experiment was laid out as described in the field except that each treatment had 5 replicates but each replicate had 10 experimental units to ensure a sufficient number of plant samples for the duration of the trial. Each replicate consisted of one *S. nigrum* plant in a polythene bag containing 5 kg of soil.

Data collection

The third youngest fully expanded leaves (JONES et al., 1971) were collected from the shoots by uprooting the whole plant; washed in distilled water to remove sediments and other impurities before drying the samples in a dust free, forced-draft oven at 40 ºC to a constant weight. The samples were then ground to a powder using a mortar and pestle and passed through a 2 mm sieve. The samples were kept in plastic val containers and stored in a refrigerator at 4 ºC till when needed. The first data were collected on the day of transplanting followed by 3 weeks after transplanting after which data were collected on a weekly basis. The experiment was terminated in the 9th week for the glasshouse experiment and 12th week for the field experiment when all the berries on the plant were mature and ripe.

Mineral analysis

The AGRILASA (2008) method of determining mineral elements in plant samples was followed. About 0.5 g of finely ground vegetable samples was placed in dry, clean digestion tubes and 5 ml of the digestion mixture comprising 1 part $HClO_4$ + 2 parts HNO_3 added. This mixture was digested at 230 ºC on a digestion block for 70 min, allowed to cool down and made up to 100 ml volume with distilled water. The concentration of Cu, Fe, Mn and Zn was then determined using the Inductively Coupled Plasma - Optical Emission Spectrometer (ICP OES).

Statistical analysis

Data of the nutrient concentrations of various treatments were subjected to statistical analysis using MNITAB Release 12. A one way analysis of variance was used to compare the means of various nutrient concentrations among the treatments and a two way analysis of variance used to determine the interaction between plant age (weeks after transplanting) and treatment on nutrient accumulation in the plant. Means were segregated using Duncan's multiple range test. The means were treated as significantly different at $p < 0.05$.

Results and discussion

Copper (Cu)

Cu exponentially increased from the time of transplanting to the 3rd week on the field, after which it varied, but was highest in the 11th week in T3 (Tab. 2a). Treatment means significantly differed ($p < 0.05$) and ranged between 7.20 and 21.60 mg/kg in the week of transplanting and the 4th week respectively. The means for the duration of the trial were highest in T1 (16.2 mg/kg) and lowest in T4 (15.5 %). Statistical analysis showed an interaction between plant age and the fertiliser treatment on Cu uptake. Regression analysis with Cu as the dependable variable and time (plant age) as the regressor showed a coefficient of determination (R^2) of 25.7 % indicating that plant age had a minimum effect on Cu. In the glasshouse, Cu exponentially increased from the time of transplanting to the 3rd week after which it decreased (Tab. 2b). Treatment means significantly differed ($p < 0.05$) and ranged between 5.30 and 21.40 mg/kg in the 9th and 4th week respectively. The means for the trial period were highest in T3 (13.6 mg/kg) and least in T1 (9.50 %). Statistical analysis showed an interaction between plant age and the fertiliser treatment on Cu. Regression analysis with Cu as the dependable variable and time (plant age) as the regressor showed a coefficient of determination (R^2) of 12.7 % indicating that plant age had a minimum effect on Cu uptake.

The field experiment showed higher concentrations of Cu than the glasshouse experiment. According to SHORROCKS and ALLOWAY (1988), the availability of Cu for uptake by plants is determined by soil pH and organic matter content among other factors. Low pH enhances the absorption of Cu from the soil. The higher concentration of Cu on the field may be due to high organic matter concentrations

Tab. 2a: Effect of organic and inorganic fertilisers on Cu (mg/kg) of *Solanum nigrum* L. cultivated in the field

	Plant age (Weeks after transplanting)										
	0	3	4	5	6	7	8	9	10	11	12
T1	7.20±0.18	17.20±0.18^{a}	15.70±0.18^{a}	18.20±0.18^{a}	16.90±0.36	12.40±0.18^{a}	16.80±0.36^{a}	19.80±0.18^{a}	17.40±0.18^{a}	18.70±0.18^{a}	18.00±0.18^{a}
T2	7.20±0.18	16.60±0.18^{b}	16.10±0.09^{b}	17.80±0.18^{b}	14.50±0.18^{a}	14.30±0.27^{b}	12.70±0.18^{b}	17.40±0.36^{b}	19.50±0.18^{b}	16.40±0.18^{b}	21.50±0.18^{b}
T3	7.20±0.18	16.70±0.09^{b}	21.60±0.27^{c}	15.60±0.27^{c}	16.60±.027	16.60±0.27^{c}	12.70±0.18^{b}	15.60±0.27^{c}	20.60±0.27^{c}	23.50±0.18^{c}	8.10±0.18^{c}
T4	7.20±0.18	18.80±0.36^{c}	17.90±0.09^{d}	17.10±0.09^{d}	16.90±0.09	15.40±0.18^{d}	14.40±0.18^{c}	19.00±0.09^{d}	17.70±0.18^{a}	15.00±0.09^{d}	10.93±0.23^{d}
T5	7.20±0.18	17.00±0.18^{ab}	17.60±0.27^{d}	16.10±0.09^{e}	16.80±0.18	15.00±0.09^{e}	16.50±0.45^{a}	17.70±0.27^{e}	19.50±0.09^{b}	15.60±0.27^{e}	14.10±0.09^{e}

0 indicates readings taken at the time of transplanting
Values shown are mean ± S.D.
Means with different letters down the same column represent significant differences at $p < 0.05$.

Tab. 2b: Effect of organic and inorganic fertilisers on Cu (mg/kg) of *Solanum nigrum* L. cultivated in the glasshouse

	Plant age (Weeks after transplanting)							
	0	3	4	5	6	7	8	9
T1	7.20±0.18	17.60±0.27^{a}	13.70±0.18^{a}	12.80±0.18^{a}	6.90±0.09^{a}	5.80±0.18^{a}	6.10±0.18^{a}	5.60±0.27^{a}
T2	7.20±0.18	16.20±0.18^{b}	17.90±0.18^{b}	12.50±0.18^{a}	9.00±0.18^{b}	5.50±0.18^{a}	6.20±0.18^{a}	5.50±0.18^{a}
T3	7.20±0.18	20.40±0.36^{c}	15.10±0.09^{c}	15.50±0.18^{b}	15.40±0.18^{c}	12.70±0.18^{b}	10.90±0.09^{b}	11.50±0.27^{b}
T4	7.20±0.18	18.20±0.18^{d}	14.00±0.18^{a}	11.50±0.45^{c}	7.60±0.18^{a}	6.20±0.18^{a}	7.00±0.18^{c}	5.30±0.27^{a}
T5	7.20±0.18	19.70±0.18^{e}	21.40±0.18^{d}	17.00±0.27^{d}	13.10±0.09^{d}	11.30±0.27^{c}	7.30±0.18^{c}	9.20±0.18^{c}

0 indicates readings taken at the time of transplanting
Values shown are mean ± S.D.
Means with different letters down the same column represent significant differences at $p < 0.05$.

and therefore slightly lower soil pH as compared to the glasshouse. In another species, Atta et al. (2010) observed that Cu was highest during the first stages or the final stages of *H. sabdariffa* growth depending on the ecotype. However, the concentration of Cu in their study was at least 600 % higher than in the current study. On the other hand, Morillo et al. (1997) reported lower concentrations of Cu as compared to the current study but in *A. gayanus*. These results indicate that *S. nigrum* has the potential to provide more Cu than the needed daily recommended values according to New Zealand and Australian standards (NHMRC, 2005) at any growth phase of the plant if children and adults respectively consume about 150 g and 300 g of the vegetable. Under field conditions, *S. nigrum* may best be harvested in the final stages of growth while under glasshouse conditions, the early growth stages would be ideal as the micronutrient is at its peak during these stages of growth.

Iron (Fe)

Fe uptake differed significantly ($p < 0.05$) and ranged between 213 and 766 mg/kg at the time of transplanting and the 5th week respectively (Tab. 3a). The means for the trial period were highest in T3 (528 mg/kg) and lowest in T5 (440 mg/kg). Statistical analysis showed an interaction between plant age and the fertiliser treatment on Fe. Regression analysis with Fe as the dependable variable and time (plant age) as the regressor showed a coefficient of determination (R^2) of 37.7 % indicating that plant age had a minimum effect on Fe uptake. Results of the glasshouse experiment showed lower means which ranged between 178 and 523 mg/kg in the 4th and 5th week respectively except in T1 (Tab. 3b). The means for the duration of the trial were highest in T1 (526 mg/kg) and lowest in T5 (239 mg/kg). Statistical analysis showed an interaction between plant age and the fertiliser treatment on Fe. Regression analysis with Fe as the dependable variable and time (plant age) as the regressor showed a coefficient of determination (R^2) of 0.9 % indicating that plant age had a very minimum effect on Fe uptake.

Fe concentration was observed to be constant on the field but slightly decreased as the plant aged in the glasshouse experiment. The observation here is different from the report of Morillo et al. (1997) in *A. gayanus*. In the same way, Flyman and Afolayan (2008) observed variations in Fe concentration on *M. balsamina* and *V. unguiculata*. According to Graham and Stangoulis (2003), solubility of Fe is very low particularly in the presence of moderate oxygen and acidic conditions. The high organic matter content on the field and the continuos release of minerals to the soil may be attributed to the differences reported in comparison with the glasshouse in the current study. The variations in concentration between the field and the glasshouse concentration of Fe in *S. nigrum* remained more than sufficient to supply the required human daily average intake across all age groups and gender according to the New Zealand and Australian standards (NHMRC, 2005). However, harvesting the plant for maximum Fe content would be best during the middle stages of the plant's growth cycle when Fe will be at its peak. Furthermore, the concentration range reported in the current study is higher than what Odhav et al. (2007) reported in wild uncultivated *S. nigrum* leaves. A portion of about 150 g and 300 g of cultivated and cooked *S. nigrum* therefore has the potential to supplement a starch rich diet of poor rural communities at all stages of growth in children and adults respectively in view of the fact that Fe is one of the most prevalent forms of micronutrient malnutrition in the world (FAO, 2004).

Tab. 3a: Effect of organic and inorganic fertilisers on Fe (mg/kg) of *Solanum nigrum* L. cultivated in the field

	Plant age (Weeks after transplanting)										
	0	3	4	5	6	7	8	9	10	11	12
T1	213±2.68	426±2.68^{a}	348±3.58^{a}	591±0.89^{a}	562±1.79^{a}	475±4.47^{a}	439±2.25^{a}	629±1.79^{a}	408±3.58^{a}	570±1.79^{a}	435±4.47^{a}
T2	213±2.68	624±3.58^{b}	246±1.79^{b}	766±2.68^{b}	403±2.68^{b}	480±1.79^{a}	374±1.79^{b}	521±0.89^{b}	483±2.68^{b}	540±1.79^{b}	750±1.79^{b}
T3	213±2.68	514±1.79^{c}	300±1.79^{c}	587±1.79^{a}	678±1.79^{c}	669±3.58^{b}	548±1.79^{c}	556±2.68^{c}	667±1.79^{c}	677±1.79^{c}	403±2.68^{a}
T4	213±2.68	371±0.89^{d}	406±2.68^{d}	617±1.79^{c}	395±4.47^{b}	575±4.47^{c}	671±0.89^{d}	396±2.68^{d}	567±1.79^{d}	578±1.79^{a}	375±4.47^{c}
T5	213±2.68	305±4.47^{e}	499±1.37^{e}	655±4.47^{d}	366±2.68^{d}	499±0.89^{d}	414±1.79^{e}	501±0.89^{b}	466±2.68^{b}	567±1.37^{a}	356±3.58^{c}

0 indicates readings taken at the time of transplanting
Values shown are mean ± S.D.
Means with different letters down the same column represent significant differences at $p < 0.05$.

Tab. 3b: Effect of organic and inorganic fertilisers on Fe (mg/kg) of *Solanum nigrum* L. cultivated in the glasshouse

	Plant age (Weeks after transplanting)							
	0	3	4	5	6	7	8	9
T1	213±2.68	430±1.79^{a}	299±4.03^{a}	337±6.26^{a}	246±1.79^{a}	325±4.47^{a}	327±1.79^{a}	433±2.68^{a}
T2	213±2.68	320±1.79^{b}	323±2.68^{b}	212±5.37^{b}	255±4.47^{b}	254±1.79^{b}	302±1.79^{b}	256±2.68^{b}
T3	213±2.68	447±1.79^{a}	340±1.79^{b}	265±4.47^{c}	296±2.68^{c}	363±2.68^{c}	269±4.03^{c}	304±3.58^{c}
T4	213±2.68	260±1.79^{c}	523±2.68^{c}	178±3.58^{d}	195±4.47^{d}	239±1.79^{d}	224±1.79^{d}	258±7.16^{b}
T5	213±2.68	217±1.79^{d}	432±1.79^{d}	191±0.89^{d}	179±3.14^{e}	230±3.58^{d}	209±8.05^{e}	239±0.89^{b}

0 indicates readings taken at the time of transplanting
Values shown are mean ± S.D.
Means with different letters down the same column represent significant differences at $p < 0.05$.

Manganese (Mn)

Mn concentration increased as the plant matured and ranged between 86 and 159 mg/kg in the 4th and 10th week respectively (Tab. 4a). The means for the trial period were highest in T2 (123 mg/kg) and lowest in T1 (104 mg/kg). Statistical analysis showed an interaction between plant age and the fertiliser treatment on Mn. Regression analysis with Mn as the dependable variable and time (plant age) as the regressor showed a coefficient of determination (R^2) of 71.1 % indicating that plant age had a significant effect on Mn. Similarly, the glasshouse means also differed significantly ($p < 0.05$) and ranged between 64 and 215 mg/kg in the 6th and 9th weeks of respectively (Tab. 4b). The means for the trial were highest in T5 (144 mg/kg) and lowest in T1 (83 mg/kg) and this was lower than what was reported on the field except for T5 which was higher in the glasshouse. Statistical analysis showed an interaction between plant age and the fertiliser treatment on Mn. Regression analysis with Mn as the dependable variable and time (plant age) as the regressor showed a coefficient of determination (R^2) of 65.8 % indicating that plant age had a significant effect on Mn.

Statistical analysis shows that there is a relationship between the age of the plant and Mn concentration. As the plant matured in age, the concentration of Mn also increased. This observation is in line with the report of Flyman and Afolayan (2008) and Atta et al. (2010) but at variance with that of Morillo et al. (1997). The continuous increase of Mn in *S. nigrum* leaves in the present study is an indication of the continuous release of the mineral from the soil for uptake by the plant. According to McGrath et al. (1994), during the summer season, the relatively high decomposition rate of organic matter releases Mn in the soil solution for possible uptake by plants and this is a possible reason why concentrations were higher on the field than in the glasshouse where organic matter content is expectedly low. Once taken up into plant tissues, Mn becomes immobile and this is possibly why the mineral continued to increase in the leaves (McGrath et al., 1994). Instead of being reassigned to the reproductive parts of the plant, the element remained immobile in the leaves of *S. nigrum*. The results of this work further indicate the ability of *S. nigrum* to provide Mn needed to supply the required human daily intake according to New Zealand and Australian standards (NHMRC, 2005) at all stages of the plant's growth if children and adults respectively consume 150 and 300 g of the cooked leaves. In addition, results of this study indicate that in order to harness the maximum amount of the mineral, leaves of mature plants should be harvested.

Zinc (Zn)

Zn exponentially increased on the field between the time of transplanting and the 3rd week and decreased until week 12 (Tab. 5a). The treatment means significantly differed ($p < 0.05$) and ranged between 33 and 78 mg/kg in the 12th and 3rd week respectively. The means for the duration of the trial were highest in T5 (62 mg/kg) and least in T1 (59 mg/kg). Statistical analysis showed an interaction between plant age and the fertiliser treatment on Zn. Regression analysis with Zn as the dependable variable and time (plant age) as the regressor showed a coefficient of determination (R^2) of 59.4 % indicating that plant age had a fairly significant effect on Zn. In the glasshouse, Zn also increased between the time of transplanting and the 4th week after which it decreased (Tab. 5b). The treatment means significantly differed and ranged between 19 and 74 mg/kg in the 9th and 4th week respectively. The means for the trial were highest

Tab. 4a: Effect of organic and inorganic fertilisers on Mn (mg/kg) of *Solanum nigrum* L. cultivated in the field

	Plant age (Weeks after transplanting)										
	0	3	4	5	6	7	8	9	10	11	12
T1	88.67±3.14	101±0.89^{a}	89±3.14^{a}	90±3.58^{a}	95±2.68^{a}	108±7.16^{a}	102±1.79^{a}	85±4.47^{a}	110±4.47^{a}	137±2.68^{a}	143±2.68^{a}
T2	88.67±3.14	116±4.47^{b}	87±3.58^{a}	121±2.68^{b}	110±2.68^{b}	139±4.03^{b}	153±2.68^{b}	114±6.26^{b}	156±2.68^{b}	129±4.03^{b}	134±1.79^{b}
T3	88.67±3.14	109±4.93^{c}	86±2.68^{a}	90±4.47^{a}	95±1.79^{a}	124±1.79^{c}	89±4.03^{c}	86±2.68^{a}	114±6.26^{a}	132±1.79^{b}	222±0.89^{c}
T4	88.67±3.14	103±1.37^{d}	104±3.58^{b}	104±0.52^{c}	101±0.89^{c}	126±2.68^{c}	132±1.79^{d}	127±6.26^{c}	159±4.03^{b}	121±0.89^{c}	154±3.58^{d}
T5	88.67±3.14	106±1.79^{cd}	112±5.37^{c}	108±2.68^{c}	92±1.79^{a}	131±0.89^{d}	136±5.37^{d}	111±1.79^{b}	127±1.79^{c}	149±4.03^{d}	154±2.68^{d}

0 indicates readings taken at the time of transplanting
Values shown are mean ± S.D.
Means with different letters down the same column represent significant differences at $p < 0.05$.

Tab. 4b: Effect of organic and inorganic fertilisers on Mn (mg/kg) of *Solanum nigrum* L. cultivated in the glasshouse

	Plant age (Weeks after transplanting)							
	0	3	4	5	6	7	8	9
T1	89±3.14	88±3.58b^{c}	78±3.58^{a}	73±0.89^{a}	64±1.79^{a}	79±4.03^{a}	79±4.47^{a}	114±6.26^{a}
T2	89±3.14	83±2.68^{c}	108±3.58^{b}	88±1.79^{b}	103±2.25^{b}	101±0.89^{b}	128±3.58^{bc}	148±1.79^{b}
T3	89±3.14	94±1.79^{b}	79±4.03^{a}	90±4.47^{b}	118±2.68^{c}	125±1.79^{c}	125±4.47^{c}	167±6.26^{c}
T4	89±3.14	86±2.68^{bc}	98±3.58^{c}	78±3.58^{a}	90±4.47^{d}	102±1.79^{b}	138±5.37^{b}	147±1.79^{b}
T5	89±3.14	117±13.42^{a}	153±2.68^{d}	119±4.47^{c}	160±4.47^{e}	164±3.58^{d}	134±3.58^{b}	215±1.79^{d}

0 indicates readings taken at the time of transplanting
Values shown are mean ± S.D.
Means with different letters down the same column represent significant differences at $p < 0.05$.

in T2 (49 mg/kg) and least in T5 (37 mg/kg) and these values were lower than those reported on the field. Statistical analysis showed an interaction between plant age and the fertiliser treatment on Zn. Regression analysis with Zn as the dependable variable and time (plant age) as the regressor showed a coefficient of determination (R^2) of 81.7 % indicating that plant age had a very significant effect on Zn.

Zn was found to be gradually decreasing as the plant matured. This observation is in line with the report by Afolayan and Flyman (2008). However, Atta et al. (2010) reported variations in Zn concentration in *H. Sabdariffa* as it grew older while Morillo et al. (1997) observed no change in Zn concentration in *A. gayanus*. According to Kabata-Pendias (2001), about 75 % of total Zn taken up by plants is stored in the shoots of young plants whereas about 20-30 % occurs in the shoots of old plants. This phenomenon is a possible explanation to the decline in Zn concentration with advancing plant maturity in the present study. Furthermore, Kabata-Pendias (2001) proposed that the roots often contain more Zn than the shoots. The results of the current study indicate that the minimum value of Zn reported in the present study (19 mg/kg in the glasshouse) can potentially supply only 51.8 % of the daily recommended human intake values of the mineral in women according to the New Zealand and Australian standards (NHMRC, 2005), however, the maximum value reported (78 mg/kg in the field) can potentially supply about 213 % of the recommended daily human intake in women. Although the values reported in this trial declined with increasing plant maturity, the concentrations have the potential to supply the daily recommended human intake of the mineral across all age groups and gender up to the 12th week on the field while in the glasshouse, the supply is adequate across all age groups and gender up to the 5th week and begins to vary. The optimum time for harvesting *S. nigrum* leaves for Zn would therefore be during early stages of its growth.

Conclusion

This study revealed that *Solanum nigrum* is a high micro mineral yielding wild vegetable that can be cultivated and incorporated into the diets of marginalised poor rural communities whose diets are mainly starch based. The best fertiliser to apply and best time to harvest the plant leaves for food may be a challenge as different minerals respond differently to different fertiliser options, therefore it is critical to recommend the best fertiliser and time of harvesting based on specific micronutrient requirements. However, 50 Kg N/ha + 4.07 t manure/ha increased the concentration of Zn on the field and Mn in the glasshouse while 8.13 t/ha goat manure increased the uptake of Mn as well as Fe on the field and Cu in the glasshouse. In addition, 100 Kg N/ha increased the concentration of Ca and Zn in the glasshouse while the field and glasshouse controls increased the concentration of Cu and Fe respectively. Cu and Fe from both the field and glasshouse varied up and down throughout the trail while Mn concentration increased steadily but Zn decreased. In general, the nutrient values recorded indicate the ability of the wild vegetable to supply the molarity of recommended daily mineral intakes of micronutrients at all stages of the plant's growth if children consume about 150 g of the cooked vegetable and adults consume 300 g. However, in order to exploit the maximum potential amounts of the minerals, the results indicate that for Cu and Zn, the leaves are best harvested during the early stages of growth and the middle stages for Fe while the final stages of the plant's life cycle would be ideal for Mn.

Tab. 5a: Effect of organic and inorganic fertilisers on Zn (mg/kg) of *Solanum nigrum* L. cultivated in the field

	Plant age (Weeks after transplanting)										
	0	3	4	5	6	7	8	9	10	11	12
T1	60±3.00	78±1.67	68±8.37	66±5.86	62±2.51^{a}	67±3.35	60±4.67^{ab}	56±3.45	43±3.35^{a}	39±4.18^{ab}	42±6.69
T2	60±3.00	76±1.53	72±4.00	66±2.00	66±3.00^{ab}	69±10.00	59±4.00^{ab}	53±5.00	59±4.00^{b}	37±1.00^{a}	43±3.00
T3	60±3.00	75±5.00	74±2.00	64±4.00	71±1.53^{b}	59±4.00	56±3.00^{ab}	56±2.00	47±2.00^{a}	47±2.00^{b}	33±3.00
T4	60±3.00	78±4.00	71±2.52	65±5.00	70±5.00^{b}	70±6.00	52±4.00^{b}	53±3.00	49±9.00^{ab}	45±6.00^{ab}	37±2.00
T5	60±3.00	77±2.00	73±3.00	67±6.51	73±3.00^{b}	67±5.00	63±3.00^{a}	61±2.00	57±4.00^{b}	41±3.00^{ab}	39±3.51

0 indicates readings taken at the time of transplanting
Values shown are mean ± S.D.
Means with different letters down the same column represent significant differences at $p < 0.05$.

Tab. 5b: Effect of organic and inorganic fertilisers on Zn (mg/kg) of *Solanum nigrum* L. cultivated in the glasshouse Sodium

	Plant age (Weeks after transplanting)							
	0	3	4	5	6	7	8	9
T1	60±2.68	59±7.15^{a}	48±3.58^{a}	46±5.37	26±4.93^{a}	16±5.37^{a}	23±4.00^{a}	19±2.68^{a}
T2	60±2.68	72±1.78^{b}	74±3.58^{b}	46±2.68	46±2.68^{c}	32±3.58^{a}	32±4.00^{ab}	33±2.68^{b}
T3	60±2.68	59±8.05^{a}	38±3.58^{c}	37±3.58^{a}	34±3.58^{b}	21±1.79^{b}	23±3.00^{a}	24±3.58^{c}
T4	60±2.68	68±0.52^{b}	53±2.68^{a}	50±4.47	41±3.14^{bc}	33±2.68^{b}	34±4.00^{b}	34±3.58^{b}
T5	60±2.68	63±2.68^{ab}	60±4.47^{d}	46±2.25	39±4.03^{b}	29±3.57^{b}	32±3.51^{b}	31±1.79^{b}

0 indicates readings taken at the time of transplanting
Values shown are mean ± S.D.
Means with different letters down the same column represent significant differences at $p < 0.05$.

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