Nutrient content of eight African leafy vegetables and their potential contribution to dietary reference intakes

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ABSTRACT

Nutrient content and potential contribution of one average portion towards nutritional requirements (Recommended Dietary Allowance; RDA) of eight African leafy vegetables (ALVs) was determined. Compared to dark-green leafy vegetables (DGLVs) as sub-group, calcium and magnesium content were similar or considerably higher, vitamin C content was considerably lower, while pigweed had higher potassium content and spider flower similar folate content. All ALVs, except Chinese cabbage, had higher iron content. Black nightshade, pigweed, cowpea and spider flower leaves had higher β-carotene content than DGLVs. For children, pigweed and cowpea leaves emerged as good sources of vitamin A (>75% RDA). followed by spider flower, black nightshade, tsamma melon, Jew's mallow and pumpkin leaves (50 - 75% RDA). For iron, pumpkin leaves provided 50 - 75% RDA. Black nightshade, tsamma melon, pigweed and cowpea leaves contributed 25 – 50% RDA, with Jew's mallow, spider flower and Chinese cabbage providing < 25% RDA. The ALVs were not a good source of zinc. Most ALVs were nutritionally similar to DGLVs. For most nutrients Chinese cabbage had considerably lower values than the other ALVs. Most of the ALVs can considerably contribute to requirements of vitamin A and, to a lesser extent, iron, both critical nutrients for developing countries.

Keywords: Traditional foods; leafy vegetables; underutilized species; South Africa; food analysis; food composition; nutrient content; vitamin A; Iron; Recommended dietary allowance

1. Introduction

Worldwide, a low intake of vegetables and fruit is among the top ten risk factors contributing to mortality (Ezzati et al., 2002). In developing countries, the diets of the poor are predominantly cereal-based and nutrient-poor, with few foods of animal origin, vegetables and fruit (Hotz and Gibson, 2007). In South Africa in 2000, an estimated 11.1 million males and 12.5 million females over 15 years of age had a low intake of vegetables and fruit (Schneider et al., 2007). The World Health Organization (WHO) recommends a daily intake of more than 400 g of fruits and vegetables per person to protect against diet-related non-communicable diseases (Lock at al., 2005; WHO, 2003), roughly double the amount consumed by the average South African (Rose et al., 2002). Vitamin A-rich vegetables and fruit, together with eggs and legumes, are the least consumed foods by South African adults (Labadarios et al., 2011). The South African diet typically lacks variety (Labadarios et al., 2011) and micronutrient deficiencies (vitamin A, iron and zinc) are widespread (Labadarios, 2007).

To address the above, increased consumption of vegetables and fruit is promoted. However, affordability, availability and seasonality affect intake (Faber et al., 2013; Love et al., 2001). Under-utilised natural resources such as African leafy vegetables may contribute to improving food and nutrition security (Toledo and Burlingame, 2006). Reportedly, African leafy vegetables may grow on soils of low fertility; are relatively drought tolerant; provide good ground cover; and can be harvested within a short period of time (Shiundu, 2002).

Green leafy vegetables (locally called *morogo* and *imifino*) are frequently consumed in some rural areas of South Africa by children and adults (Faber et al., 2010; Labadarios et al., 2000;

Nesamvuni et al., 2001; Steyn et al., 2001a,b). The leaves are typically harvested from the wild, but also from cultivated crops. In the present work African leafy vegetables refer to the collective of traditional leafy vegetables that form part of the culinary repertoire of contemporary African communities, whilst leafy vegetable refers to plant species of which the leafy parts, which may include young succulent stems, flowers and young fruit, are used as a vegetable (Jansen van Rensburg et al., 2007). A recent review on the nutrient content of African leafy vegetables consumed in Sub-Saharan Africa reported large variation in nutrient content and concluded that available information is limited and often incomplete (Uusiku et al., 2010). Most data focus on wild growing leaves, yet there is an increasing interest in the potential of cultivating African leafy vegetables.

Thus, the primary aim of this study was to determine the nutrient content of eight cultivated African leafy vegetables. The eight species were non-heading Chinese cabbage (*Brassica rapa* L. subsp. *chinensis*), black nightshade (*Solanum retroflexum* Dun.), pigweed (*Amaranthus cruentus* L.), Jew's mallow (*Corchorus olitorius* L.), spider flower (*Cleome gynandra* L.), cowpea (*Vigna unguiculata* L. Walp.), pumpkin (*Cucurbita maxima* Duchesne) and tsamma melon (*Citrullus lanatus* (Thunb.)). Secondary objectives were to explore the nutritional potential of these leaves by comparing their composition to dark-green leafy vegetables as a sub-group, and by estimating their potential contribution to dietary requirements.

2. Materials and Methods

2.1 Production and collection of the plants for analysis

Two of the eight African leafy vegetables (black nightshade and Chinese cabbage) are grown commercially and for this study were cultivated at an irrigation scheme in the Limpopo Province. The other six African leafy vegetables are usually grown without addition of much fertiliser and were cultivated at the research institute in the Gauteng Province in South Africa.

Black nightshade and Chinese cabbage were planted from seed during the winter of July 2007 and June 2008, respectively, on a deep, red, well-drained soil at Dzindi Irrigation Scheme, Itsani, Thulamela Municipality, Vhembe District, Limpopo Province on plots that were 1000 m² in size. The method used to grow Chinese cabbage was explained in detail elsewhere and involved band-placed application of 40 kg N ha⁻¹, 60 kg P ha⁻¹ and 80 kg K ha⁻¹ at planting using the fertiliser mixture 2:3:4 (32), followed by two band-placed applications of 50 kg N ha⁻¹ in the form of limestone ammonium nitrate (28% N) four weeks and six weeks after planting, respectively (Van Averbeke and Netshithuthuni, 2010). The irrigation strip from which the sample was taken had a homogeneous soil (based on the soil survey done). Sampling of the plants for analysis occurred when the first plants in the stand showed peduncle elongation, at which stage all plants in the stand had passed the tenth leaf stage. Only plants that had not reached the stage of peduncle elongation were sampled. The rate of application of fertilisers used for black nightshade at planting was the same as for Chinese cabbage. Sampling of the black nightshade plants occurred as soon as the first flowers appeared. Harvesting of black nightshade usually involves three shoot cuts (Van Averbeke et al., 2007) but only the first cut was used when sampling plants for analysis.

Pigweed, Jew's mallow, cowpea and tsamma melon were grown during the summer of 2007/2008 and pumpkin and spider flower during the summer of 2009/2010 at the Agricultural Research Council – Vegetable and Plant Institute, Roodeplaat in the Gauteng

Province. In both years, seeds of the African leafy vegetables were sown directly in the same field, which was irrigated three times per week (about 15 mm per irrigation). In 2007/2008 the crops relied on residual fertiliser for nutrient acquisition. In 2009/10, compost was incorporated into the soil before sowing.

All eight of the African leafy vegetables were harvested between two to three months after sowing early in the morning, before 10:00 am. For each African leafy vegetable, three separate lots were harvested at random on the same day from different positions across the same field, so as to obtain representative sample batches for each species. Each lot of Chinese cabbage consisted of 10 whole mature plants that were cut at the base just above soil level keeping the leafy head intact. Individual lots of black nightshade and Jew's mallow comprised 40 plants each; those of pigweed and cowpea consisted of 50 plants each; individual lots of spider flower weighed approximately 3500 g each comprising plant stems with healthy edible leaves; pumpkin leaves (lamina with petiole) were cut from vines and each lot weighed approximately 1550 g; individual lots of tsamma melon leaves weighed approximately 1300 g.

Pumpkin and tsamma melon leaves were transported in labelled brown paper bags to the sensory analysis laboratory of the Meat Industry Centre of the ARC – LBD:API and processed the same day. All the other plants were sprinkled with water and each lot of plants loosely packed in a marked black plastic bag that was pierced to avoid "sweating" of the leaves and to allow some airflow. The bags were transported on the same day to the laboratory where they were kept in a cold storage room at +4 °C overnight for processing the next day. The individual lots for pigweed, Jew's Mallow and cowpea were transferred to buckets half-filled with water for overnight cold storage.

2.2 Sampling and processing of leaves for analysis

Each of the three collected lots per species was processed separately. For each lot of plants healthy edible leaves were removed from the base/stem and combined to constitute a composite sample. The number of leaves taken per plant varied between five and 15 for the different African leafy vegetables. In all cases only the lamina (leaf blade) that was removed from the petiole was taken for analysis except for Jew's mallow, pumpkin and spider flower leaves for which the lamina attached to the petiole was taken. The weights of the individual composite samples for each African leafy vegetable were as follows: Chinese cabbage, 980 g; pigweed, 550 g; spider flower, 870 g; pumpkin, 850 g; tsamma melon, 450 g; black nightshade, Jew's mallow and cowpea, 460 g.

The leaves were soaked and thoroughly washed with tap water (three to four times) to remove soil debris, followed by washing with distilled water. The leaves were air-dried on absorbent paper at room temperature for two hours. The leaves were cut into smaller pieces and homogenised using a household food processor; from this, a portion of adequate amount that was needed for all the nutrient analyses was further homogenised to a finer texture using a hand-held stick blender. Portions were transferred to marked plastic containers with screw caps. The containers were frozen at -20 °C until required for analysis. The portions for β -carotene analysis were dispatched to the Medical Research Council's (MRC) Laboratory in Cape Town. All nutrient analyses for each lot per species were therefore done on portions obtained from the same homogenized sample.

Key nutrients, important in the diet of the nutritionally vulnerable, were determined for the various African leafy vegetables. The analyses were done on a double blind basis in a South African National Accreditation System (SANAS) accredited laboratory (with the exception of β-carotene). Reference samples form part of the daily routine in these laboratories to assure the quality of results. Accepted standardised techniques were used for nutrient analysis. After initial standardisation of techniques during a pilot study, the eight vegetables were treated identically for each test, throughout the study.

Proximate analysis: Total fat content was determined using a 2 g freeze-dried portion and applying the AOAC method 960.39 (AOAC, 2005). The Tecator Soxtec System 1034 extraction unit with reagent petroleum ether (boiling point range of 40 to 60 °C) was used for the extraction. Moisture content was determined by measuring the weight loss of a 5 g portion of fresh material using the AOAC method 950.46 (AOAC, 2005). Total ash (inorganic matter) content was determined using the AOAC method 920.153 (AOAC, 2005). The organic matter of the samples was removed by heating at 550 °C overnight; the remaining residue is inorganic matter (ash). Protein content was determined in duplicate using the Dumas Combustion method, AOAC 992.15 (AOAC, 2005). The sample was combusted at approximately 1100 to 1350 °C, and 10 cm³ of the sample gas was analysed. A thermal conductivity cell detects the difference in thermal conductivity caused by the presence of nitrogen. A conversion factor of 6.25 was used in the calculation of the protein content. Dietary fibre content was determined using the Official Method 985.29, (AOAC, 1995). Total carbohydrate content was calculated "by difference". It includes all the noncarbohydrate material not analysed in the other proximate analyses and the cumulative errors

from the other measurements (Greenfield and Southgate, 2003). The total carbohydrate content obtained in this manner was used in the calculation of energy content. The energy content (kJoule/100g) was calculated using the percentage (g/100g) protein multiplied by 17, plus the percentage total carbohydrates multiplied by 17, plus the percentage fat multiplied by 37 (Greenfield and Southgate, 2003).

Minerals: Sodium, potassium, calcium, manganese, magnesium, copper, phosphorus, iron and zinc content were determined using Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES) at a sub-contracted laboratory, ARC-Institute for Soil, Climate and Water. A 0.5 g portion of the freeze-dried samples were digested with nitric-perchloric acid at temperatures up to a maximum of 200 °C (Zasoski and Burau, 1977). The Varian Liberty 200 sequential ICP-OES with Varian SPS-5 autosampler (Agilent Technologies, Inc., Santa Clara, CA, USA) unlike the simultaneous instruments, measures each wavelength one at a time immediately after each other, using a diffraction grating monochromator with a single photomultiplier tube detector. It uses a V-groove nebuliser with a Sturman-Masters spray chamber with the torch in a radial orientation. The instrument was calibrated against a series of standard solutions containing all the elements of interest in the proportions found in typical leaf samples. Each element was measured at an appropriate emission wavelength chosen for high sensitivity and lack of spectral interferences: sodium, 589.592 nm; potassium, 769.896 nm; calcium, 422.673 and 317.933 nm; manganese, 257.61 nm; magnesium, 383.826 nm; copper, 324.754 nm; phosphorus, 213.618 nm; iron, 259.940 nm; and zinc, 213.856 nm. The moisture content values were used to convert the "freeze-dried basis" values back to the "raw basis" values.

Water soluble vitamins: Thiamin and riboflavin content were determined according to a modified method described by Fellman et al. (1992) using High-Performance Liquid Chromatography (HPLC; Thermo Separation Products, Fremont, CA, USA; P400 quaternary pump; Rheodyne Model 7125 sample injector; 100 μL sample loop) and fluorescence detection (Jasco 821-FP Intelligent Fluoremeter, Tokyo, Japan). Chromatography was performed with a Luna 10 μ m C₁₈₍₂₎ 100 Å reverse phase (4.6 x 250 mm) column (Phenomenex, Torrance, CA, USA); mobile phase consisted of methanol:0.005 M ammonium acetate (pH 5) buffer (40:60); flow rate was 1.7 mL/min. For riboflavin the detector's excitation and emission wavelengths were set at 450 nm and 530 nm, respectively; for thiamin (thiochrome) the excitation and emission wavelengths were 370 nm and 435 nm, respectively. The method for folate analysis is based on the observation that specific bacteria, yeasts and fungi are able to grow and form metabolic products only in the presence of vitamins of the B-group. When the vitamin, or substrate containing this vitamin, is added, growth takes place which is dependent on the amount of vitamin. Therefore, the amount of vitamin present can be detected by measuring the turbidity resulting from growth. A pure vitamin preparation of known activity is used in a parallel test and serves as the reference standard (Barton-Wright, 1952). Vitamin C content was determined by HPLC and fluorescence detection (Dodson et al., 1992) using the same system described for thiamin and riboflavin. Chromatography was performed with a LiChrosorb RP-18 5 μm (4.6 x 250 mm) column with guard column (Phenomenex, Torrance, CA, USA); mobile phase consisted of 50% methanol; flow rate was 1.0 mL/min; excitation and emission wavelengths were 350 nm and 430 nm, respectively.

β-Carotene: Carotenoids were extracted in duplicate from 2 to 3 g homogenised portions with tetrahydrofuran:methanol (1:1, v/v) and the total and trans-β-carotene content

determined with HPLC as previously described (Kimura and Rodriguez-Amaya, 2002; Low and van Jaarsveld, 2008). Each sample was extracted twice ensuring that the extractionfiltration solvent was colourless after the second extraction. Quantification was performed by external standardization. The purity of a purchased β-carotene standard (CaroteNature GmbH, Lupsingen, Switzerland) was verified by HPLC (97.1%) and the concentration of the standard solution was corrected accordingly. A five-point standard curve, ranging from 2 to 15.98 µg/mL and bracketing the concentration present in the samples, was constructed in triplicate. The linear curve passed through the origin and had a coefficient of correlation of 0.9996. On each day of analysis, quantification was done by one-point calibration verifying any change in the HPLC detector's response. The liquid chromatograph (Thermo Separation Products, Fremont, CA, USA) comprised the following SpectraSERIES subunits: P100 isocratic pump; AS100 fixed-loop autosampler (operated with a standard full-loop injection technique; 250 µL syringe; 10 µL sample loop); UV150 detector (variable wavelength UV/Visible absorbance detector; UV range 190–365 nm with deuterium lamp, or visible range 366–800 nm with tungsten lamp). Chromatography was performed with a monomeric C_{18} column (Waters Spherisorb ODS 2, 3 µm, 4.6×150 mm). The mobile phase consisted of acetonitrile (containing 0.05% triethylamine):methanol:ethyl acetate (80:10:10), with a flow rate of 0.7 mL/min. The detector wavelength was set at 450 nm for the detection of βcarotene. Integration of the chromatograms was performed by the DELTA Chromatography Data System for windows software, version 5.5 (Data worX Pty. Ltd., Kangaroo Point, QLD, Australia). Results were expressed as μg β-carotene per 100 g raw edible portion. The vitamin A value in µg Retinol Activity Equivalents (RAE) was calculated as the sum of (alltrans-β-carotene (μg) + cis-β-carotene (μg) \times 0.5) divided by 12 assuming RAE to be 12 μg β-carotene: 1μg retinol (Otten et al., 2006).

For each nutrient, the mean and standard deviation from the three individually analysed sample lots were calculated and rounded off to the number of decimal places given for the specific nutrient in the SAFOODs database (Wolmarans et al., 2010). In order to account for nutrient losses during cooking, nutrient retention factors given for "veg, greens, boiled, little water, drain" (USDA Table of nutrient retention factors, 2007) were used for the individual nutrients. The following nutrient retention factors (USDA Table of nutrient retention factors, 2007) were used: calcium, magnesium, sodium, copper, iron, zinc, riboflavin and β -carotene = 0.95; phosphorus and potassium = 0.90; thiamin = 0.85; folate = 0.65; vitamin C = 0.60. The USDA Table of nutrient retention factors (2007) does not provide retention factors for "veg, greens, boiled, little water, drain" for protein, energy and manganese; we therefore, for this calculation exercise, assumed no loss during cooking and used a retention factor of 1 for protein, energy and manganese. Should reliable retention factors become available it can be used in conjunction with the nutrient values provided in Table 1.

An average portion size was set at 130 g boiled leaves for adult females and 90 g for young children (Faber et al., 2007). A raw to cooked yield factor of 1.3 was applied, based on cooking experiments at the laboratory of the Nutritional Intervention Research Unit of the MRC (unpublished data). The nutrient contribution of an average portion of cooked leaves to nutrient intake recommendations of individuals was calculated and expressed as a percentage of the Dietary Reference Intakes (DRI) (recommended dietary allowance [RDA], estimated energy requirements [EER], adequate intakes [AI]) for 4-8-year-old children and 19-30-year-old non-pregnant, non-breastfeeding females (Otten et al., 2006; Ross et al., 2011).

3. Results and Discussion

The nutrient composition of the African leafy vegetables is given in Table 1. The energy and fat content was low. The leaves contained between 2.2 and 10.8 g fibre per 100 g raw edible portion.

The micronutrient values given for certain green leafy vegetables in the SAFOODS database (Wolmarans et al., 2010) differ from those observed in the present study, with significant notable difference in β-carotene (and vitamin A; μg RAE) and vitamin C content. The mean zinc content in the raw African leafy vegetables of the present study is similar to that of a single collection and a limited amount of material of similar leaf species analysed by Schönfeldt and Pretorius (2011). However, the mean iron content in some leaf species of the present study was considerably lower (3.2- to 6.8-fold), whilst total β-carotene content was higher (2.5- to 4.5-fold) than reported (Schönfeldt and Pretorius, 2011). Schönfeldt and Pretorius (2011) argued that the high mineral (iron) content of the leaf species in their study could have resulted from the chicken and cattle manure that were used as soil fertilisers. A review by Uusiku et al. (2010) showed large variation in iron content in leaves which could have been influenced by various factors. Large variation and high iron content of leafy vegetables should in general be interpreted with caution, particularly for ground-level growing leaves, such as pumpkin -, cowpea - and spider flower leaves, because soil contamination may occur. In the present study, the edible leave parts were meticulously soaked and washed with several changes of water before sample homogenasion in order to avoid contamination. The mean total β-carotene content in three leaf species of the present study were considerably higher than three similar raw leaf species reported by Schönfeldt and Pretorius (2011). It is uncertain whether the difference in β -carotene content in leaves

between the two studies could be ascribed to the differences in analytical sample preparation and the form of the sample subjected to analysis. Portions of homogenised fresh raw leaves were analysed for β -carotene content in the present study, whereas the analytical sample subjected to analysis for β -carotene content in raw leaves was of a freeze-dried powder form in the Schönfeldt and Pretorius (2011) study. Nutrient content of raw plant foods vary widely and is affected by factors such as variety or cultivar; part of the plant consumed; stage of maturity; geographic site of production or climate; harvesting and post-harvest handling conditions; and storage. Comparing nutrient content of leaves from different data sources is therefore challenging. Common errors introduced during sampling procedures, sample preparation and analytical methods can also affect the reported nutrient values.

Table 1 also gives the mean nutrient composition of nine dark-green leafy vegetables as a sub-group (beet greens, collards, kale, mustard greens, parsley, romaine, spinach, Swiss chard and turnip greens; Pennington and Fisher, 2010). The particular nutritional importance of dark-green leafy vegetables as a vegetable and fruit sub-group is the contribution to intakes of vitamin C and K (615 μ g/100 g) (where 100 g contributes to more than 50% of the DRI, recommended dietary intake per day, of non-pregnant, non-breastfeeding adults), folate (more than 25% of the mentioned DRI), and potassium, calcium, magnesium, iron, copper, manganese and vitamin B6 (more than 10% of mentioned DRI) (Pennington and Fisher, 2010). The vitamin K content of the African leafy vegetables was not determined. Six of the eight African leafy vegetables investigated in the present study had higher dietary fibre content than dark-green leafy vegetables as a sub-group. Calcium and magnesium content of the African leafy vegetables were similar (Chinese cabbage) or considerably higher. Chinese cabbage did not reach the group average of dark-green leafy vegetables for copper, zinc, iron, manganese, folate, vitamin C and β -carotene. Only pigweed had potassium contents that

exceeded the group mean for dark-green leafy vegetables and only spider flower contained folate comparable to the group mean. Interestingly, none of the investigated African leafy vegetables had vitamin C content approaching that of dark-green leafy vegetables as subgroup. Conversely, apart from Chinese cabbage, they all had higher iron content, but the fact that the vitamin C content was very low and the fibre content very high, may affect the bioavailability of the iron unfavourably. For β -carotene content, black nightshade, pigweed, cowpea and spider flower leaves had higher values than the dark-green leafy vegetables group mean.

The calculated nutrients retained for 100 g raw leaves after cooking are given in Table 2. The retention factors used to calculate the amount of nutrients retained are estimates, as cooking method, -temperature and -time all effect the retention (Greenfield and Southgate, 2003). In rural areas, women may "overcook" the leaves or "discard and replace" the cooking water several times to get rid of the bitter taste (Jansen van Rensburg et al., 2007; Van Averbeke et al., 2007), which may affect the nutrient retention of particularly the water soluble nutrients.

In Table 3 the estimated amount of nutrients retained after preparation and taking the yield factor into account are expressed as a percentage of the RDA for two age groups, namely 4-8-year-old children and 19-30-year-old non-pregnant, non-breastfeeding females. We used these two age groups because young children and women of child-bearing age are nutritionally most vulnerable. The RDA is the average daily dietary intake level that is sufficient to meet the nutrient requirement for nearly all (97 to 98%) healthy individuals in a particular life stage and gender group (Otten et al., 2006; Ross et al., 2011).

When comparing the estimated nutrient contribution of an average cooked portion size of African leafy vegetables (130 g) to the DRI's (EER, RDA and AI) of 19-30-year-old women, the following emerges (Table 3): The African leafy vegetables were low in energy (<5% of the estimated requirement). Together with the high fibre and low fat content, these characteristics of ALVs are potentially important elements of a dietary pattern to treat and prevent obesity. More than 50% of South African adult females are either overweight or obese (Department of Health, 2004), and often maternal obesity and child micronutrient malnutrition co-exist within the same community, and often the same household (Faber et al., 2001).

The calculated estimates showed that African leafy vegetables provide 10% or less of the RDA for 19-30-year-old females for protein (except for spider flower), zinc (except for spider flower), thiamin, riboflavin (except for black nightshade and spider flower) and vitamin C. Phosphorous, copper and folate content ranged from 4 to 26% of the RDA. Calcium and magnesium contents for all the African leafy vegetables were above 10% of the requirement, with the two best sources for calcium being cowpea and pigweed, and the best source for magnesium being pigweed. Iron content showed a large variation, with Chinese cabbage having the lowest content (<10% of the RDA), black nightshade and tsamma melon both providing at least a third of the RDA, and pumpkin leaves providing almost 50% of the RDA. Chinese cabbage had the lowest vitamin A content (36% of the RDA), whereas leaves of black nightshade, pigweed, cowpea, tsamma melon and spider flower provided more than 50% of the RDA.

A similar trend was observed for children. The calculated estimates showed that an average cooked portion size of African leafy vegetables (90 g) provide less than 5% of the estimated

requirement for energy. Chinese cabbage and black nightshade provide less than 10% of the RDA for protein. Generally, an average cooked portion size of African leafy vegetable provide a greater percentage of the DRI requirement for many of the minerals and vitamins as compared to the adult females, albeit a smaller portion size and lower DRIs. From a public health perspective and mineral and vitamin deficiencies, dietary sources of iron, zinc and vitamin A are of particular importance. Three of the eight African leafy vegetables provide 10% or more of the RDA for 4-8-year-old children for zinc, five provide more than 25% of the RDA for iron and six provide more than 50% of the RDA for vitamin A with Chinese cabbage providing the least vitamin A.

The calculated potential contribution of the African leafy vegetables to reference intakes of the key nutrients zinc, iron and vitamin A showed that these leaves are not a good source of zinc, but, with the exception of Chinese cabbage, provide considerable amounts of iron and vitamin A. Pumpkin leaves had the highest iron content, followed by black nightshade and tsamma melon, and thus appear to be a very good source of non-haeme iron compared to the other African leafy vegetables analysed. The presence of oxalates, phytates and polyphenols in dark green leafy vegetables (Gupta et al., 2005) may, however, inhibit the absorption of non-haeme iron (Zimmermann et al., 2005). Since the bioavailability of the iron in plant foods is low, the potential contribution of plant foods towards controlling iron deficiency in developing countries has been questioned (De Pee et al., 1996). Agricultural interventions to increase the supply and intake of iron from plant foods are not popular. Instead, the production and consumption of animal foods are usually encouraged because of the high bioavailability of haeme iron from animal foods (Ruel, 2001).

The African leafy vegetables provide a significant proportion of the RDA for vitamin A. Chinese cabbage provides the least amount of vitamin A, while pigweed and cowpea provide the largest amount of vitamin A. Globally 190 million children under five years are vitamin A deficient (WHO, 2009), mostly because of an inadequate dietary intake of vitamin A. These children have a higher risk for diseases, such as diarrhoea (Fawzi et al., 1995), as vitamin A deficiency reduces the child's ability to fight infections (Ross, 1996). Consumption of cooked and pureed green leafy vegetables (containing 750 – 850 µg RE, which is equal to 375 – 425 µg RAE, per unknown portion size) was shown to have a beneficial effect on improving vitamin A status (Haskell et al., 2004, 2005; Takyi, 1999). Table 2 shows that after cooking 100 g leaves (based on calculations using nutrient retention factors), African leafy vegetables exceeding 375 µg RAE are pigweed, cowpea, black nightshade and spider flower leaves; pigweed and cowpea exceed 425 µg RAE. Using fat, such as cooking oil, during preparation of African leafy vegetables will have a beneficial effect on vitamin A status because fat enhances carotenoid absorption for bioconversion to vitamin A (Jayarajan et al., 1980).

4. Conclusion

The results of this study showed that the eight African leafy vegetables that were investigated were nutritionally diverse. On the whole, they contained concentrations of indicator nutrients that were more or less similar to those reported by Pennington and Fisher (2010) for darkgreen leafy vegetables as a sub-group, but zinc content was persistently lower, whilst iron content tended to be higher. Among the eight African leafy vegetables, Chinese cabbage was nutritionally the poorest. Pigweed and cowpeas are the best sources of vitamin A, while pumpkin leaves are the best source for iron. Most of the eight African leafy vegetables can

potentially make a considerable contribution towards the requirements for nutrients, particularly vitamin A and iron (provided that these nutrients are bioavaible), which are micronutrients of public health significance in South Africa. For this reason, their cultivation and consumption should be promoted. Further research on preparation and processing aimed at optimising retention and bioavailability of nutrients in African leafy vegetables is recommended.

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Table 1Nutrient content per 100 g edible portion of leaves of raw African leafy vegetables

	Unit	Chinese cabbage	Black nightshade	Pigweed	Jew's mallow	Cowpea	Pumpkin	Tsamma melon	Spider flower	DGLV as a sub-group ^a	
PROXIMATE COMPOSITION:											
Moisture	g	92.2	89.5	82.0	79.6	82.4	85.6	81.3	87.5	NA ^b	
		(0.2)	(3.5)	(0.8)	(1.8)	(1.0)	(1.2)	(0.5)	(0.7)		
Protein	g	2.5	0.5	4.2	3.2	4.7	2.9	3.5	5.0	NA	
		(0.2)	(0.2)	(0.0)	(0.2)	(0.1)	(0.2)	(0.1)	(0.2)		
Ash	g	1.00	1.32	2.38	1.81	1.76	1.51	1.66	1.46	NA	
		(0.02)	(0.41)	(0.06)	(0.14)	(0.07)	(0.14)	(0.05)	(0.09)		
Fat	g	0.2	0.4	0.3	0.1	0.6	0.2	0.4	0.3	NA	
		(0.1)	(0.1)	(0.0)	(0.0)	(0.0)	(0.0)	(0.1)	(0.0)		
Energy	kJ	120	162	272	319	280	222	296	195	NA	
		(5)	(53)	(13)	(28)	(17)	(18)	(10)	(10)		
Dietary fibre	g	2.2	2.5	6.7	10.8	5.8	3.0	3.8	3.1	2.8	
		(0.1)	(0.9)	(0.2)	(1.1)	(0.0)	(1.3)	(0.2)	(0.2)	(0.8)	
Carbohydrates	g	4.1	8.2	11.2	15.3	10.5	9.8	13.1	5.7	NA	
		(0.1)	(2.9)	(0.8)	(1.4)	(0.9)	(1.0)	(0.5)	(0.3)		
MINERALS:											
Calcium	mg	152	199	443	310	398	177	212	232	112	
	-	(5)	(62)	(25)	(31)	(33)	(11)	(20)	(10)	(48)	
Magnesium	mg	42	92	242	87	62	67	59	76	44	
<u>U</u>	0	(1)	(27)	(19)	(8)	(4)	(5)	(2)	(3)	(27)	
Phosphorus	mg	32	36	81	118	51	102	119	138	NA	
i nospiioi us	1115	(5)	(11)	(5)	(10)	(1)	(8)	(2)	(4)	11/1	

	Unit	Chinese cabbage	Black nightshade	Pigweed	Jew's mallow	Cowpea	Pumpkin	Tsamma melon	Spider flower	DGLV as a sub-group ^a
Sodium	mg	29	8	10	11	10	12	9	15	NA
		(8)	(3)	(1)	(1)	(0)	(1)	(0)	(1)	
Potassium	mg	165	257	459	407	238	351	260	374	418
		(29)	(77)	(47)	(46)	(27)	(43)	(12)	(34)	(183)
Copper	mg	0.07	0.16	0.17	0.19	0.14	0.21	0.20	0.25	0.17
		(0.01)	(0.05)	(0.02)	(0.03)	(0.01)	(0.03)	(0.01)	(0.01)	(0.10)
Zinc	mg	0.30	0.56	0.70	0.57	0.42	0.75	0.74	1.04	0.39
		(0.06)	(0.17)	(0.05)	(0.04)	(0.03)	(0.06)	(0.04)	(0.05)	(0.29)
Iron	mg	1.4	7.2	5.1	3.6	4.7	9.2	6.4	2.1	2.08
		(0.3)	(3.1)	(0.7)	(0.6)	(0.7)	(1.6)	(1.4)	(0.1)	(1.73)
Manganese	μg	350	2080	2340	790	2690	540	760	580	440
		(40)	(520)	(240)	(100)	(30)	(50)	(60)	(40)	(250)
VITAMINS:										
Thiamin	mg	0.04	0.08	0.04	0.02	0.07	0.04	0.01	0.06	NA
		(0.0)	2) (0.02	(0.00)	(0.00)	(0.01)	(0.01)	(0.01)	(0.01)	
Riboflavin	mg	0.06	0.17	0.05	0.03	0.08	0.10	0.10	0.21	NA
		(0.0)	0) (0.03	(0.00)	(0.00)	(0.01)	(0.01)	(0.01)	(0.01)	
Folate	μg	92	56	75	45	105	47	68	121	121
		(16)	(15)	(13)	(14)	(6)	(12)	(7)	(9)	(79)
Vitamin C	mg	8	5	2	1	9	2	10	2	59
		(2)	(3)	(0)	(0)	(0)	(1)	(0)	(0)	(41)
Total β-	μg	3593	3 5566	7138	4307	7031	4247	4956	5936	5325
Carotene		(47)	(32)	(492)	(120)	(186)	(190)	(182)	(300)	(1928)
All-trans-β-	μg	2823	3 4568	5757	3578	5857	3547	4043	4477	NA

	Unit	cabbage	Black nightshade	Pigweed	Jew's mallow	Cowpea	Pumpkin	Tsamma melon	Spider flower	DGLV as a sub-group ^a
Carotene		(34)	(36)	(406)	(115)	(168)	(178)	(166)	(204)	
Vitamin A	μg RAE	267	422	537	329	537	325	375	434	444 ^d
		(3)	(3)	(37)	(10)	(15)	(15)	(14)	(21)	(161)

Values are mean and (standard deviation) of three sample lots analysed individually. Single analysis was done for proximates, minerals and vitamins except protein and β -carotene which were analysed in duplicate

^a DGLV: Dark-green leafy vegetables as a sub-group include beet greens, collards, kale, mustard greens, parsley, romaine, spinach, Swiss chard and turnip greens (Pennington and Fisher, 2010)

^bNA: not available

^c Retinol activity equivalents (RAE): sum of (all-*trans*-β-carotene (μ g) + *cis*-β-carotene [total β-carotene – all-*trans*-β-carotene] (μ g) × 0.5) divided by 12 assuming RAE to be 12 μ g β-carotene:1 μ g retinol (Otten et al., 2006)

 $^{^{}d}$ Calculated from the β -carotene content of dark-green leafy vegetables as a group (Pennington and Fisher, 2010)

Table 2Estimated^a amount of nutrients retained after cooking 100 g leaves of raw African leafy vegetables

	Unit	Chinese cabbage	Black nightshade	Pigweed	Jew's mallow	Cowpea	Pumpkin	Tsamma melon	Spider Flower
Energy	kJ	120	162	272	319	280	222	296	195
Protein	g	2.5	0.5	4.2	3.2	4.7	2.9	3.5	5.0
Calcium	mg	144	189	421	295	378	168	201	220
Magnesium	mg	40	87	230	82	59	64	56	72
Phosphorus	mg	29	33	73	106	45	92	107	125
Sodium	mg	27	8	9	10	10	11	9	14
Potassium	mg	148	232	413	366	214	316	234	337
Copper	μg	63	152	161	181	129	196	188	238
Zinc	mg	0.28	0.53	0.66	0.54	0.40	0.71	0.70	0.99
Iron	mg	1.4	6.9	4.9	3.4	4.5	8.8	6.1	2.0
Manganese	μg	350	2080	2340	790	2690	540	760	580
Thiamin	mg	0.03	0.07	0.03	0.02	0.06	0.03	0.01	0.05
Riboflavin	mg	0.06	0.16	0.05	0.03	0.08	0.09	0.09	0.20
Folate	μg	60	37	49	29	68	30	44	79
Vitamin C	mg	5	3	1	1	5	1	6	1
Vitamin A	μg RAE ^b	254	401	510	312	510	309	356	412

^a The nutrient content in cooked African leafy vegetables was calculated from the mean nutrient values in raw leaves (Table 1) using the following nutrient retention factors: Protein,

Energy and Manganese = 1 (assumption); Calcium, Magnesium, Sodium, Copper, Iron, Zinc, Riboflavin and β -Carotene = 0.95; Phosphorus and Potassium = 0.90; Thiamin = 0.85; Folate = 0.65; Vitamin C = 0.60 (USDA Table of nutrient retention factors, 2007)

Table 3Estimated nutrient contribution of an average portion size^a of leaves of African leafy vegetables to the recommended dietary allowance (RDA) for children aged 4-8 years and women aged 19-30 years

	Unit	Chinese cabbage	Black nightshade	Pigweed	Jew's mallow	Cowpea	Pumpkin	Tsamma melon	Spider Flower
Energy	% EER 4-8 y ^b	1	2	3	3	3	2	3	2
	% EER 19-30 y	1	2	3	3	3	2	3	2
Protein	% RDA 4-8 y ^c	9	2	15	12	17	11	13	19
	% RDA 19-30 y	5	1	9	7	10	6	8	11
Calcium	% RDA 4-8 y	13	17	38	27	34	15	18	20
	% RDA 19-30 y	19	25	55	38	49	22	26	29
Magnesium	% RDA 4-8 y	22	47	124	44	32	34	30	39
	% RDA 19-30 y	13	28	74	27	19	21	18	23
Phosphorus	% RDA 4-8 y	4	5	10	15	6	13	15	17
	% RDA 19-30 y	4	5	10	15	6	13	15	18
Sodium	% AI 4-8 y ^d	2	0	1	1	1	1	1	1
	% AI 19-30 y	2	1	1	1	1	1	1	1
Potassium	% AI 4-8 y	3	4	8	7	4	6	4	6
	% AI 19-30 y	3	5	9	8	5	7	5	7
Copper	% RDA 4-8 y	10	24	26	29	20	31	30	38
	% RDA 19-30 y	7	17	18	20	14	22	21	26
Zinc	% RDA 4-8 y	4	7	9	8	6	10	10	14
	% RDA 19-30 y	4	7	8	7	5	9	9	12
Iron	% RDA 4-8 y	9	48	34	24	31	61	43	14

	Unit	Chinese cabbage	Black nightshade	Pigweed	Jew's mallow	Cowpea	Pumpkin	Tsamma melon	Spider Flower
	% RDA 19-30 y	8	38	27	19	25	49	34	11
Manganese	% AI 4-8 y	16	97	109	37	125	25	35	27
	% AI 19-30 y	19	115	130	44	149	30	42	32
Thiamin	% RDA 4-8 y	4	8	4	2	7	4	1	6
	% RDA 19-30 y	3	6	3	2	5	3	1	5
Riboflavin	% RDA 4-8 y	7	19	6	3	9	11	11	23
	% RDA 19-30 y	5	15	4	3	7	8	8	18
Folate	% RDA 4-8 y	21	13	17	10	24	11	15	27
	% RDA 19-30 y	15	9	12	7	17	8	11	20
Vitamin C	% RDA 4-8 y	13	8	3	2	15	3	17	4
	% RDA 19-30 y	6	4	1	1	7	1	8	2
Vitamin A	% RDA 4-8 y	44	70	89	55	89	54	62	72
	% RDA 19-30 y	36	57	73	45	73	44	51	59

^a 90 g cooked African leafy vegetables for young children and 130 g cooked African leafy vegetables for females using a yield factor of 1.3 from raw to cooked

Source for nutrient requirements: Otten et al., 2006; Ross et al., 2011

^b EER = estimated energy requirements; EER (kJ) for 4-8 years is the mean of 7316 (EER for boys) and 6896 (EER for girls) = 7106

^c RDA = recommended dietary allowance, which is the average daily dietary intake level that is sufficient to meet the nutrient requirement of nearly all (97 to 98%) healthy individuals in a particular life stage and gender group

^d Adequate Intake (AI) as there is no RDA; the AI is a recommended intake value that is assumed to be adequate