

Carotenoids, tocopherols, minerals and phytate in fresh and carotenoids and tocopherol contents in sun-dried dark green leafy vegetable varieties from the Lindi region, Tanzania

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Abstract

The present study explores the nutritional value of different dark green leafy vegetable (DGLV) varieties from Tanzania. Carotenoids, tocopherols, minerals, and phytates were analyzed in unprocessed and differently sun-dried DGLV. HPLC analyses revealed highest concentrations of lutein-zeaxanthin (20.3 mg/100 g estimated fresh weight (FW) in *Manihot esculenta*), β -carotene (6.89 mg/100 g FW in *Cleome gynandra*) and α -tocopherol (12.5 mg/100 g FW in *Manihot esculenta*); thus 100 g of a DGLV could provide up to 116% of the recommended daily intake (RDI) for vitamin A and 168% of the RDI for vitamin E. High concentrations (per 100 g calculated FW) of iron (up to 11.1 mg in *Cleome gynandra*), calcium (up to 469 mg in *Amaranthus viridis*) and magnesium (up to 371 mg in *Amaranthus graecizans*) were found; thus 100 g could contribute up to 80%, 47% and 169% of the RDI for iron, calcium, and magnesium, respectively. HPLC analyses revealed pleasantly low phytate (1.66–13.1 mg/100g FW); this resulted in low phytate-to-mineral molar ratios not exceeding the critical values of bioavailability. The differently sun-dried DGLV had similarly high concentrations of carotenoids and tocopherols compared to the unprocessed fresh DGLV (mg/ 100 g dry weight), with essentially no marked differences between simple open sun-dried and direct and indirect solar drying techniques. In conclusion, results indicate that both, fresh and sundried DGLV could make a substantial contribution to micronutrient supply, e.g. pro-vitamin A and iron in the diets of the Tanzanian population.

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Introduction

Vegetables are an important source of micronutrients and phytochemicals such as tocopherol, provitamin A carotenoids, iron, zinc, calcium, and magnesium^[1]. Green leafy vegetables contain these micronutrients and therefore have the potential to address daily requirements, especially of individuals with poor nutritional status^[2,3]. Indigenized species, within the context of Africa, are the ones that originated in other continents^[4,5], but have become part and parcel of traditional African food culture and agriculture production systems^[6]. Dark green leafy vegetables (DGLVs) sometimes indigenous to the locals have been shown to provide carotenoids and iron in diets consumed in Tanzania^[5]. Weinberger & Msuya^[7] reported that in Northern Tanzania traditional leafy vegetables are good sources of micronutrients such as iron, zinc, and β -carotene. In this study, DGLVs will refer to crop species or varieties genuinely native to a region, or crops introduced to the region over time, although the species may not be native. Despite their merits, the consumption of vegetables in Tanzania has been 130 g per day, which remains below the recommended amount of 400 g per

person per day by the World Health Organization^[8]. Micronutrient malnutrition especially among children is a major concern in Tanzania, where 30% of children under 5 years of age are stunted and 59% of them are anemic while in the case of Lindi stunting and anemia were 21% and 71% respectively^[9,10]. Gowele et al.^[11] reported that more than 42% of children aged 5–10 years in the Kilosa and Chamwino districts of Tanzania were anaemic and in the Lindi region prevalence of anemia was 76.8% of children and women aged 15–49^[12]. Male farmers (35%) in the Lindi region were also reported to be anaemic^[12]. This study is part of the 'Vegi-Leg' project which focuses on the development of DGLVs processing technologies and corresponding products. In the context of DGLVs, simple solar drying technologies were developed to produce dried DGLVs with improved shelf-life for consumption during the dry season. Different preservation and storage techniques such as sun drying, solar drying, pickling and fermentation have been used in Africa in an attempt to make vegetables available during the lean season^[13–16]. Traditional leafy vegetables production is dominated by female farmers to sustain household food security^[17]. Solar dried leafy vegetables in the Lindi region have

been shown to be safe and of good quality^[18]. Limited studies have been conducted in the Lindi region about processed DGLVs. In addition, concentrations of different phytochemicals and bioactive compounds and micronutrients such as carotenoids, vitamins, and minerals in the different DGLVs species found there, are less documented. Therefore, this study aimed to determine carotenoids, tocopherols, and minerals (e.g. iron, zinc) of 14 different DGLVs from the Nachingwea and Ruangwa districts in the Lindi region (screening trial). The phytate concentration as an important naturally occurring anti-nutrient that may reduce the bioavailability of minerals was also determined. In addition, a drying assessment was conducted to analyze concentrations of carotenoids and tocopherols to compare three drying techniques (open sun drying (traditional), direct and indirect solar drying) and corresponding changes in micronutrient and secondary plant metabolite contents.

Materials and methods

Description of the study area

The study was conducted in the Lindi region of Tanzania, involving Nachingwea and Ruangwa districts (Fig. 1). Two villages, Mitumbati in Nachingwea district and Mibure in Ruangwa district were selected for this study^[19]. The two districts experience erratic, but adequate, rainfall between December and March.

Sample collection and preparation

Fresh vegetables and their sources (screening trial 2018)

Fourteen different species of dark green leafy vegetables (DGLVs) (Table 1) were collected from both study villages

during the rainy season in May 2019 (fresh vegetables). The vegetables were selected based on a preliminary survey and focus group discussions with the village leadership and farmers to identify available and commonly consumed DGLVs. Identification of the vegetable samples was done by a botanist from the Department of Crop Science and Horticulture at Sokoine University of Agriculture (SUA). For the fresh vegetables, the edible parts of the vegetables were separated, washed with potable (bottled) drinking water, and air-dried at room temperature at the study sites. Then they were placed in black polyethylene bags, put in a cool box containing ice, transported to the laboratory at SUA, and stored in the freezer at -30°C . Fresh DGLVs samples were finally packed with dry ice and transported (Turkish airline cargo) to the University of Hohenheim, Stuttgart (Germany), stored at -80°C , then freeze-dried (2014 model LyoQuest, Telstar, Spain) and again stored at -80°C until being analyzed for the individual measurements including carotenoids and tocopherols, minerals, and phytates.

All individual 'fresh' samples were weighed before and after freeze-drying to estimate weight lost by freeze-drying; the resulting conversion factor was used to convert the results per analyzed freeze-dried weight (mg/100 g DW) to estimate fresh weight (mg/100 g FW) as follows:

Values per fresh weight (mg/100g FW) = Values per dried weight (mg/100 g DW)/Conversion factor

Collection and drying of vegetables (drying experiment 2021)

The ten most consumed varieties of 14 dark green leafy vegetables (DGLVs) were collected from each study village in March 2021 and dried on-site using three drying methods;

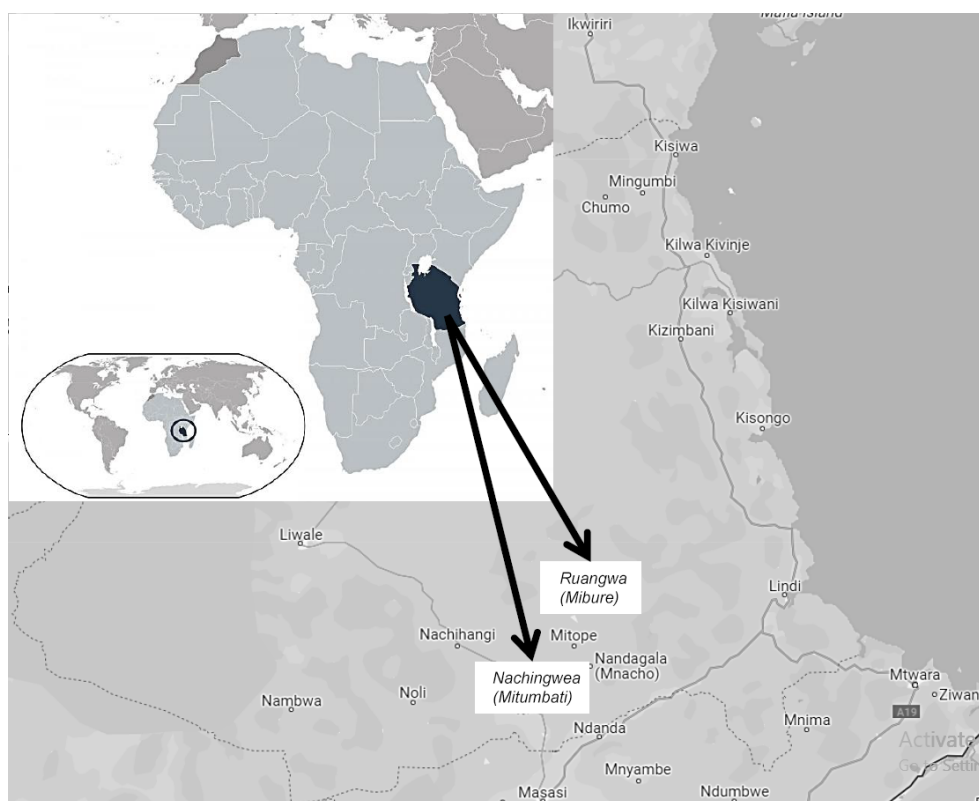


Fig. 1 Study areas, Nachingwea and Ruangwa districts.

Micronutrients in dark green leafy vegetables

Table 1. Fresh vegetable samples (collected 2018) names and origin.

Botanical name	Local/English name	Origin		Conversion factor
		Mitumbati	Mibure	
<i>Amaranthus graecizans</i>	Tindi pori/Amaranthus	√	–	5.27
<i>Amaranthus maadira</i>	Mchicha bangi/Amaranthus	√	–	4.87
<i>Amaranthus spinosus</i>	Mchicha pori/Amaranthus	√	√	5.78
<i>Amaranthus</i> spp.	Mchicha wa nyumbani/Amaranthus	√	√	6.33
<i>Amaranthus viridis</i>	Tindi ya nyumbani/Amaranthus	√	√	4.49
<i>Cleome gynandra</i>	Mgagani/African spider plant	√	–	4.60
<i>Corchorus olitorius</i>	LilendeJute mallow)	√	√	6.06
<i>Curcubita maxima</i>	Majani ya maboga/Pumpkin leaves	√	√	4.51
<i>Dioscorea</i> spp.	Nandelele	√	–	5.49
<i>Gisekia pharnaceoides</i> L.	Mtimba mwisi	√	–	5.96
<i>Ipomea aquatica</i>	Mkokobwado	√	√	7.48
<i>Ipomea batatas</i> L.	Tembele/Sweet potato leaves	√	√	5.67
<i>Manihot esculenta</i>	Kisamvu/Cassava leaves	√	√	2.96
<i>Vigna unguiculata</i> L.	Majani ya kunde/Cowpea leaves	√	√	4.90

√ means the vegetable is from the respective district while – means that the vegetable is not present in the district. Conversion factor = FW/DW, fresh weight/freeze-dried weight.

open sun drying (traditional drying), direct solar drying, and indirect solar drying as shown in Fig. 2.

For open sun drying, edible leafy parts of the vegetables were collected, washed, and left to air dry. The leaves were then traditionally processed by boiling them for 2 min at 100 °C, putting them in a winnowing basket and left to dry under the open sun. For direct and indirect solar drying (Fig. 2), edible leafy parts of the vegetables were collected, washed, weighed and then blanched at 80 °C for 1 min then taken directly to the dryers and left to dry. Times, ambient, and solar dryer (internal) temperatures were also recorded. Once the vegetables were dried to a crispy texture, they were put immediately in airtight opaque polyethylene bags transported to the SUA laboratory and stored in a dry clean place away from sunlight. The samples were transported by Turkish Airline Cargo and DHL to the University of Hohenheim, Stuttgart in Germany in May 2022 and stored at –80 °C for one week. Afterwards, the individual DGLVs samples were freeze-dried for 48 h (to constant weight),

using a Telstar freeze drier (2014 model LyoQuest; Telstar, Spain) and protected from light (using aluminum foil). The samples were then ground in an electric mortar to a fine homogenous powder and stored again at –80 °C in airtight containers protected from light; aliquots of the (freeze-dried) powder were finally weighed as samples for analyses on carotenoids and tocopherols.

Temperature of dryers

The mean temperatures of ambient (open sun), and internal direct and indirect solar dryers were determined using food-grade thermometers (Brewferm, BROWLAND) from 900 to 1,500 h.

Determination of micronutrients, secondary plant metabolites and phytate

Determination of carotenoids and tocopherols

The measurement of carotenoids (lutein, zeaxanthin, α -/ β -carotene) and tocols (tocopherols and tocotrienols) were



Fig. 2 (a) Open sun drying (traditional drying), (b) direct solar drying, and (c) indirect solar drying.

performed using HPLC as previously described^[20–22]. Freeze-dried DGLV samples (100 mg) were saponified with 500 μ L of KOH (50% w/w) and 1 mL ethanol containing 1 μ L/mL of β -apo-8'-carotenal methyloxime (internal standard) at 70 °C for 30 min. After saponification, 2 mL of saline solution (15% w/v) were added and the samples were neutralized with 500 L of glacial acetic acid. The fat-soluble components were extracted with hexane (2 \times 1 mL), and combined fractions were evaporated in a rotational vacuum evaporator (RVC 2-33 IR, Christ, Osterode am Harz, Germany). The residue was re-dissolved in 70 μ L ethanol (> 96%) and 210 μ L acetonitrile for analysis of carotenoids using RP-HPLC, UV-vis at 450 nm and fluorescence detection excitation set at 298 nm and emission set at 328 nm for α - γ -tocopherol analysis. All reagents and solvents were of analytical and (ultra) gradient HPLC grade. N-hexane, 1,4-dioxane, potassium hydroxide solution (50%), acetic acid (100%), and ethanol were from Carl Roth GmbH + Co. KG (Karlsruhe, Germany), while methanol and acetonitrile were from J.T. Baker (Deventer, The Netherlands).

Determination of minerals

Iron (Fe), zinc (Zn), calcium (Ca), and magnesium (Mg) were determined by inductively coupled plasma optical emission spectrometry (ICP-OES, Vista Pro Radial (Varian Inc.) after microwave-heated nitric acid digestion (Milestone Ultra Clave II; Leutkirch, Germany), as previously described by Stuetz et al.^[23]. The percentage contribution of each DGLV variety (per 100 g FW) in the respective mineral recommendation according to WHO/FAO^[24] was calculated taking into account the bio-availability and corresponding values for male and female adults (19–65 years): Fe (10% bio-availability) – 14 mg (males) and 29 mg (females), Zn (low bio-availability) – 14 mg (males) and 9.8 mg (females), Ca – 1,000 mg (males) and 1,300 mg (females), Mg – 260 mg (males) and 220 mg (females).

Determination of phytate

Phytate (inositol hexakisphosphate or IP6), was quantitatively measured by HPLC as previously described^[25]. For the analysis of InsP isomers (IP6, IP5 and IP4) in digested samples, 1.0 g of the sample was extracted for 30 min with 10 mL of a solution containing 0.2 M-EDTA and 0.1 M-sodium fluoride (pH = 10) as phytase inhibitor using a rotary shaker. The samples were centrifuged at 12,000 g for 15 min and the supernatant fraction was removed and preserved on ice. The residue was re-suspended in 5 mL of the EDTA–sodium fluoride solution and extracted again for 30 min. Next, 1 mL of the pooled supernatant fraction (of the two extraction steps) was centrifuged at 14,000 g for 15 min and 0.5 mL of the resulting supernatant fraction, filtered through a 0.2 μ m cellulose acetate filter (VWR) into a Microcon filter (cut-off 30 kDa) device (Millipore) and centrifuged again at 14,000 g for 30 min. Throughout the whole extraction procedure, the samples were kept below 6 °C.

Filtrates were analyzed by high-performance ion chromatography and UV detection at 290 nm after post-column derivatization using an ICS-3000 system (Dionex). InsP with different degrees of phosphorylation (InsP3–6) and their positional isomers were separated, without enantiomer differentiation, on a Carbo Pac PA 200 column and corresponding guard column. Fe (NO₃)₃ solution (1 g/L, Fe (NO₃)₃·9H₂O, product no. 103883; Merck KGaA) in HClO₄ (20 g/L, product no. 100518; Merck KGaA) was used as a reagent for derivatization according to

Phillippy & Bland^[26]. InsP5 isomer standards were purchased from Sirius Fine Chemicals. Seven out of nine myo-inositol tetrakisphosphate (InsP4) and nine out of 12 myo-inositol trisphosphate (InsP3) isomer standards were available from Santa Cruz Biotechnology. However, only IP6 was detected in DGLV samples. Critical values of molar ratios of phytate:iron > 1, phytate:zinc > 15, phytate:calcium > 0.24, were used to predict the inhibitory effect of phytate on Fe, Zn, and Ca bioavailability^[24].

Statistical analysis

Concentrations of carotenoids, tocopherols, and minerals in the 14 different 'estimated' fresh leafy vegetables, and carotenoids and tocopherols in the 10 different dried leafy vegetables are described as medians and 25th and 75th percentiles while phytates are presented as mean with SD. Median values for carotenoids and tocopherols were computed from eight determinations while for minerals from six and phytate from two or four determinations, as appropriate. The concentrations of carotenoids, tocopherols, and minerals between the 14 fresh DGLVs were compared using the Kruskal-Wallis pairwise multiple comparison test while the phytate content of the 14 fresh DGLVs were compared using One-Way ANOVA and Tukey's post-hoc test. The individual carotenoid (Lutein-zeaxanthin, α - β -carotene) and tocopherol (α - γ -tocopherol) concentrations of the 10 dried DGLVs were compared between open-, direct- and indirect sun (or solar) drying technique using Friedman's multiple pairwise comparison test. All statistical analyses were carried out using IBM SPSS software (Version 20, IBM Corp., Armonk, NY, USA); a *p*-value < 0.05 was considered statistically significant.

Results

Carotenoids, tocopherols, minerals and phytates in (fresh) dark green leafy vegetables (DGLVs)

Carotenoids and tocopherols contents

Provitamin A carotenoid and tocopherol contents of different fresh DGLVs species including corresponding calculated retinol equivalents (RE) and tocopherol equivalents (TE) with percentages of contribution regarding the recommended nutrient intakes (RNI) are presented in Table 2. In contrast to other analysed DGLVs, *Manihot esculenta* had the highest content of lutein-zeaxanthin (20.28 mg/100 g FW) but was not significantly (*p* > 0.05) different from *Vigna unguiculata* L., *Curcubita maxima*, *Cleome gynandra*, *Corchorus olitorius*, *Amaranthus graecizans* and *Amaranthus viridis*. *Amaranthus spinosus* had the lowest lutein-zeaxanthin concentration (4.90 mg/100 g FW). The range of β -carotene levels was 1.01 to 6.89 mg/100 g FW with *Cleome gynandra* having the highest content of 6.89 mg/100 g FW compared to other DGLVs, but significantly different from *Dioscorea* spp., *Amaranthus spinosus*, and *Amaranthus maadira*. α -carotene was highest in *Amaranthus spinosus* (0.96 mg/100 g). *Cleome gynandra*, *Amaranthus graecizans* and *Manihot esculenta* could provide \geq 92% and 111% of the retinol RNI for males and females, respectively. Generally, α -tocopherol was the predominant tocopherol. The highest concentration of α -tocopherol were detected in *Manihot esculenta* (12.50 mg/100 g FW), *Corchorus olitorius* (3.20 mg/100 g) and *Curcubita maxima* (2.78 mg/100 g FW). In the case of γ -tocopherol, the highest contents were

Table 2. Lutein-zeaxanthin, α - and β -carotene and γ -tocopherol in fresh DGLVs (mg/100 g fresh weight).

Botanical name	lutein-zeaxanthin	α -carotene	β -carotene	RE	M	F	α -tocopherol	γ -tocopherol	α -TE	M	F
<i>Amaranthus maadira</i>	5.14 (4.80, 5.50) ^c	0.07 (0.05, 0.07) ^{ab}	1.08 (0.8, 1.26) ^b	93	15	19	1.39 (1.30, 1.40) ^{ab}	0.01 (0.01, 0.01) ^b	1.40	14	19
<i>Amaranthus graecizans</i>	10.6 (10.4, 11.1) ^{ac}	0.50 (0.40, 0.60) ^a	6.38 (4.90, 7.40) ^{ab}	553	92	111	2.07 (1.80, 2.20) ^{ab}	0.60 (0.50, 0.60) ^{ab}	2.16	22	29
<i>Amaranthus spinosus</i>	4.90 (4.90, 5.10) ^c	0.09 (0.09, 0.10) ^{ab}	1.47 (1.30, 1.50) ^b	127	27	33	0.99 (0.90, 1.10) ^b	0.01 (0.01, 0.01) ^b	1.00	10	13
<i>Amaranthus spp</i>	6.67 (6.12, 6.94) ^{bc}	0.38 (0.20, 0.40) ^a	4.66 (3.05, 5.50) ^{ab}	404	67	81	1.21 (1.10, 1.40) ^{ab}	0.05 (0.03, 0.05) ^b	1.21	12	16
<i>Amaranthus viridis</i>	8.01 (6.53, 9.29) ^{ac}	0.38 (0.20, 0.80) ^a	2.75 (2.20, 4.70) ^{ab}	245	41	49	1.27 (1.20, 1.40) ^{ab}	0.04 (0.03, 0.05) ^b	1.30	13	17
<i>Cleome gynandra</i>	11.9 (11.5, 12.0) ^{ac}	0.12 (0.10, 0.13) ^{ab}	6.89 (5.50, 9.70) ^a	579	97	116	1.06 (1.00, 1.10) ^{ab}	0.19 (0.10, 0.20) ^{ab}	1.12	11	15
<i>Corchorus olerius</i>	8.73 (8.20, 9.26) ^{ac}	0.03 (0.02, 0.05) ^b	2.82 (2.20, 5.20) ^{ab}	236	39	47	3.20 (1.50, 5.20) ^{ab}	0.35 (0.2, 0.5) ^{ab}	3.23	32	43
<i>Curcubita maxima</i>	12.0 (9.58, 14.8) ^a	0.21 (0.08, 0.29) ^{ab}	2.84 (2.20, 3.50) ^{ab}	245	41	49	2.78 (2.10, 3.80) ^{ab}	1.23 (0.90, 1.50) ^a	2.92	29	39
<i>Dioscorea spp</i>	5.02 (4.74, 5.20) ^c	0.01 (0.01, 0.02) ^b	1.01 (0.90, 1.40) ^b	85	14	17	0.59 (0.50, 0.70) ^b	2.30 (2.10, 2.50) ^a	0.83	8	11
<i>Gisekia pharmaceoides L.</i>	5.92 (5.70, 6.34) ^{bc}	0.02 (0.01, 0.02) ^b	1.53 (1.40, 2.10) ^{ab}	128	21	26	0.43 (0.40, 0.50) ^b	0.15 (0.14, 0.16) ^{ab}	0.44	4.4	6
<i>Ipomea aquatica</i>	5.40 (5.00, 6.00) ^c	0.10 (0.08, 0.10) ^{ab}	2.80 (2.40, 4.40) ^{ab}	238	40	48	0.76 (0.70, 0.80) ^b	0.10 (0.07, 0.10) ^b	0.77	8	10
<i>Ipomea batatas L.</i>	7.91 (7.74, 8.07) ^{bc}	0.29 (0.24, 0.30) ^a	4.50 (2.80, 6.90) ^{ab}	387	65	77	0.74 (0.60, 0.80) ^b	0.08 (0.08, 0.10) ^b	0.75	7	10
<i>Manihot esculenta</i>	20.3 (17.8, 21.4) ^a	0.07 (0.05, 0.10) ^{ab}	6.74 (4.40, 7.70) ^a	565	94	113	12.5 (8.20, 16.6) ^a	1.03 (0.70, 1.10) ^a	12.6	126	168
<i>Vigna unguiculata L.</i>	12.9 (10.9, 15.0) ^{ab}	0.10 (0.05, 0.30) ^{ab}	4.28 (3.10, 10.9) ^{ab}	362	60	72	1.60 (1.10, 2.00) ^{ab}	0.27 (0.20, 0.30) ^{ab}	1.63	16	22

Values are expressed as medians of four or eight determinations (extraction/analysis) for carotenoids and tocopherols; values in parenthesis represent minimum and maximum values; values within a column not sharing a common superscript letter (a,b,c) are significantly different (p -value < 0.05); M = Males and F = Females; RE, retinol equivalents (sum of RE), for which 1 RE = 24 μ g α -carotene or 12 μ g β -carotene. RNI, recommended nutrient intakes for vitamin A by WHO/FAO^[24], values (microgram) for male (600 μ g RE/d) and female (500 μ g RE/d) adults (19–65 years). α -TEs, α -tocopherol equivalents (1 mg α -TE = 1 mg α -tocopherol or 10 mg γ -tocopherol. RNI, recommended nutrient intakes for vitamin E by WHO/FAO; values (mg) for male (10 mg/d) and female (7.5 mg/d) adults (19–65 years).

Table 3. Mineral contents (mg/100 g 'estimated' fresh weight) of DGLV and their contribution (%) to the daily recommended nutrient intake by gender (M, F).

Botanical name	Iron		Zinc		Calcium		Magnesium		M	F
	M	F	M	F	M	F	M	F		
<i>Amaranthus maadira</i>	2.55 (2.52, 2.56) ^{ab}	18	9	0.41 (0.41, 0.40) ^b	3	4	381 (376, 393) ^{ab}	309 (309, 314) ^{ab}	119	141
<i>Amaranthus graecizans</i>	6.28 (6.19, 6.32) ^{ab}	45	21	1.15 (1.14, 1.17) ^{ab}	8	12	393 (379, 403) ^{ab}	371 (371, 379) ^a	143	169
<i>Amaranthus spinosus</i>	3.50 (3.45, 3.56) ^{ab}	25	12	0.39 (0.39, 0.39) ^b	3	4	435 (408, 456) ^a	165 (165, 166) ^{ab}	63	75
<i>Amaranthus spp</i>	1.70 (1.71, 1.81) ^b	12	6	0.55 (0.54, 0.57) ^{ab}	4	6	295 (283, 314) ^{ab}	115 (105, 117) ^{ab}	44	52
<i>Amaranthus viridis</i>	5.31 (5.24, 5.38) ^{ab}	38	18	0.72 (0.71, 0.75) ^{ab}	5	7	469 (405, 547) ^a	217 (214, 218) ^{ab}	84	99
<i>Cleome gynandra</i>	11.1 (10.6, 11.7) ^a	80	38	0.65 (0.65, 0.66) ^{ab}	5	7	237 (236, 237) ^{ab}	54.0 (53.9, 54.2) ^{ab}	21	25
<i>Corchorus olerius</i>	4.69 (1.71, 7.70) ^{ab}	34	16	0.50 (0.49, 0.50) ^{ab}	4	5	235 (151, 321) ^{ab}	55.0 (44.5, 64.8) ^b	21	25
<i>Curcubita maxima</i>	4.20 (3.10, 5.31) ^{ab}	30	14	0.71 (0.70, 0.71) ^{ab}	5	7	232 (216, 249) ^{ab}	115 (106, 126) ^{ab}	44	53
<i>Dioscorea spp</i>	8.74 (8.66, 8.79) ^a	62	30	0.44 (0.44, 0.44) ^{ab}	3	4	235 (235, 238) ^{ab}	278 (276, 279) ^{ab}	107	126
<i>Gisekia pharmaceoides L.</i>	5.14 (5.08, 5.25) ^{ab}	37	18	0.51 (0.51, 0.52) ^{ab}	4	5	76.9 (75.3, 78.0) ^b	61.2 (59.6, 62.0) ^{ab}	24	28
<i>Ipomea aquatica</i>	3.64 (3.13, 4.21) ^{ab}	26	12	0.39 (0.237, 0.547) ^b	2	3	115 (105, 130) ^b	37.4 (33.9, 41.6) ^b	14	17
<i>Ipomea batatas L.</i>	3.79 (3.55, 4.02) ^{ab}	27	13	0.34 (0.29, 0.40) ^b	2	3	127 (119, 136) ^b	68.0 (61.1, 73.5) ^{ab}	26	31
<i>Manihot esculenta</i>	2.21 (2.11, 2.34) ^b	16	8	1.41 (1.37, 1.43) ^a	10	14	212 (190, 237) ^{ab}	94.5 (94.0, 94.9) ^{ab}	36	43
<i>Vigna unguiculata L.</i>	2.93 (2.23, 3.87) ^{ab}	21	10	0.58 (0.54, 0.63) ^{ab}	4	6	208 (159, 258) ^{ab}	58.0 (56.8, 59.5) ^b	22	26

Values are medians (minimum, maximum) of three or six determinations (extraction/analysis) or percentages (contributions for M, F); values within a column not sharing a common superscript letter (a,b) are significantly different (Kruskal-Wallis multiple pairwise comparison; Adj. p < 0.05). M = Males and F = Females; RNI, recommended nutrient intakes by WHO/FAO^[24], values all in [mg/d] for adults by gender (19–65 years): iron (10% bioavailability) – 14 (M) and 29 (F); zinc (low availability) – 14 (M) and 9.8 (F); calcium 1,000 (M) and 1,300 (F); magnesium – 260 (M) and 220 (F).

Table 4. Phytate content (mg/100 g fresh weight) and its ratios to individual minerals in DGLVs.

Botanical name	Total phytate	Phy:Fe	Phy:Zn	Phy:Ca	Phy:Mg
<i>Amaranthus maadira</i>	6.08 ± 0.57 ^b	0.20	1.47	0.00	0.00
<i>Amaranthus graecizans</i>	5.64 ± 0.53 ^b	0.08	0.49	0.00	0.00
<i>Amaranthus spinosus</i>	5.14 ± 0.48 ^b	0.12	1.31	0.00	0.00
<i>Amaranthus</i> spp.	4.69 ± 0.44 ^b	0.23	0.84	0.00	0.00
<i>Amaranthus viridis</i>	6.26 ± 0.63 ^b	0.10	0.86	0.00	0.00
<i>Cleome gynandra</i>	23.0 ± 1.90 ^a	0.17	3.50	0.01	0.02
<i>Corchorus olitorius</i>	13.1 ± 9.54 ^{a,b}	0.23	2.52	0.00	0.01
<i>Curcubita maxima</i>	9.50 ± 3.24 ^b	0.18	1.25	0.00	0.00
<i>Dioscorea</i> spp.	1.82 ± 0.16 ^b	0.02	0.41	0.00	0.00
<i>Gisekia pharnaceoides</i> L.	1.66 ± 0.15 ^b	0.03	0.32	0.00	0.00
<i>Ipomea aquatica</i>	10.1 ± 1.87 ^b	0.25	3.50	0.01	0.01
<i>Ipomea batatas</i> L.	4.88 ± 0.54 ^b	0.11	1.42	0.00	0.00
<i>Manihot esculenta</i>	9.59 ± 0.88 ^b	0.37	0.67	0.00	0.00
<i>Vigna unguiculata</i> L.	6.44 ± 0.66 ^b	0.19	1.10	0.00	0.00

Values are means and standard deviation of two or four determinations (extraction/analysis) for phytate; values within a column not sharing a common superscript letter (a,b) are significantly different (p -value < 0.05). Phy = phytate, Fe = iron, Zn = zinc, Ca = calcium and Mg = magnesium; Molar masses (μ /mol) for phytate: 660.04, Fe: 55.84, Zn: 65.38, Ca: 40.07 and Mg: 24.30; Critical values of molar ratios predicting the inhibitory effect of phytate on Fe, Zn, and Ca: phytate:iron > 1, phytate:zinc > 15, phytate:calcium > 0.24.

found in *Dioscorea* spp. (2.30 mg/100 g FW) followed by *Curcubita maxima* (1.23 mg/100 g FW). The concentration of γ -tocopherol of the DGLVs ranged from 0.01 to 2.03 mg/100 g FW with *Dioscorea* spp having the highest content. *Amaranthus maadira*, *Amaranthus spinosus*, *Amaranthus* spp, *Amaranthus viridis*, *Ipomea aquatica*, and *Ipomea batatas* L. differed significantly (p > 0.05) from other DGLVs in the γ -tocopherol concentration.

Mineral contents of DGLVs

The median mineral concentrations of the fresh vegetables are presented in Table 3. Substantial amounts of Fe (1.70–11.13 mg/100 g FW), Zn (0.30–1.40 mg/100 g FW), Ca (76.88–469.15 mg/100 g FW), and Mg (37.43–370.71 mg/100 g FW) were found in the analyzed DGLVs species. The DGLVs with the highest Fe content was *Cleome gynandra* (11.13 mg/100 g FW) which was significantly different from *Manihot esculenta*, and *Amaranthus* spp. *Manihot esculenta* contained the highest amount of Zn (1.15 mg/100 g FW) and was significantly different from *Ipomea aquatica*, *Ipomea batatas*, *Amaranthus maadira*, and *Amaranthus spinosus*. Calcium contents ranged from 115.3 mg/100 g FW in *Ipomea aquatica* to 370.76 mg/100 g FW in *Amaranthus viridis*, while magnesium concentrations ranged from 37.43 mg/100 g FW in *Ipomea aquatica* to 370.71 mg/100 g FW in *Amaranthus graecizans*. The concentration of iron in 100 g FW of *Cleome gynandra* could contribute to more than 80% of RNI for males and 270% for females, respectively, while *Manihot esculenta* could contribute more than 10% of RNI for zinc in both, males and females. *Amaranthus viridis* (100 g FW) could provide more than 47% and 36% of RNI of calcium for males and females, respectively. *Amaranthus viridis*, *Amaranthus graecizans*, *Amaranthus maadira*, and *Dioscorea* spp (100 g FM) could provide more than 80% of RNI for magnesium for both sexes.

Phytate content of DGLVs

Total phytate using IP6 contents of DGLVs and phytate ratios to Fe, Zn, Ca, and Mg are presented in Table 4. Molar ratios

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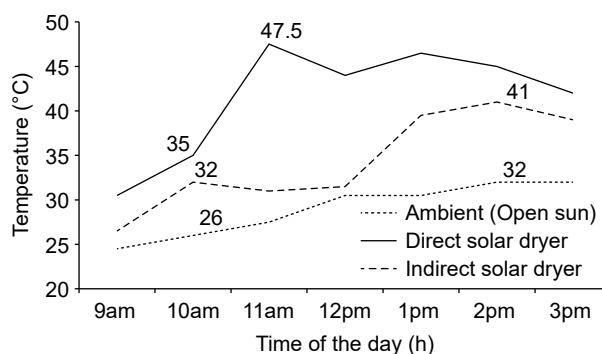


Fig. 3 Variation of the average ambient and drying temperatures (°C) inside the dryers over time (d) in both villages for 7 d.

along with the suggested critical values for estimating the effect of phytate on their bioavailability are also presented. *Cleome gynandra* (22.98 mg/100 g FW) had the highest phytate contents followed by *Corchorus olitorius* (12.73 mg/100 g FW) which were not significantly (p > 0.05) different. *Gisekia pharnaceoides* L. and *Dioscorea* spp. had the lowest phytate content and molar ratios of phytate to minerals. All DGLVs did not exceed the critical values of the inhibitory effect of phytate on mineral bioavailability.

Effect of different drying methods on carotenoids and tocopherols contents

Temperature of dryers used for drying DGLVs (drying experiment 2021)

The mean ambient and internal direct and indirect solar dryers' temperatures are presented in Fig. 3. The values changed over time with direct solar drying attaining higher temperatures (47.5 °C; 11 am) as compared to the indirect solar dryer temperature (41 °C; 2 pm) and ambient (open sun drying) temperature (32 °C; 2 pm).

Effect of drying methods on lutein- zeaxanthin, α and β -carotene contents of DGLVs

The effect of different drying methods on lutein-zeaxanthin, α and β carotene contents of DGLV species are presented in Table 5. For *Manihot esculenta*, the highest lutein-zeaxanthin contents (43.08 mg/100 g DW) were found in open sun-dried samples, followed by indirect (41.55 mg/100 g DM) and direct solar drying (33.95 mg/100 g DM). On the other hand, *Corchorus olitorius* (43.72 mg/100g) and *Manihot esculenta* (41.55 mg/100 g) had higher contents from direct solar and indirect solar drying technologies, respectively, compared to open sun drying. There was no significant difference in drying methods for *Manihot esculenta*, *Curcubita maxima*, and *Ipomea batatas* L. There was no significant difference in lutein-zeaxanthin concentration of open sun drying and direct solar drying except for *Amaranthus viridis*.

Open sun-dried *Amaranthus viridis* had the highest α -carotene content (1.90 mg/100 g DW) followed by *Amaranthus graecizans* (1.04 mg/100 g DW). On the other hand, direct solar-dried *Vigna unguiculata* L. (0.86 mg/100 g DW) and indirect solar dried *Amaranthus graecizans* (1.12 mg/100 g DW) had higher α -carotene contents compared to other vegetables. Direct solar drying retained higher levels of α -carotene concentration when compared to the other drying method for most vegetables. On the other hand, direct solar-dried *Corchorus olitorius* (27.36 mg/100 g DW), and indirect solar-dried *Manihot*

Table 5. Lutein- zeaxanthin, α and β -carotene contents (mg/100 g dry weight) of DGLVs from different drying methods.

Botanical names	Lutein- zeaxanthin			α -carotene			β -carotene		
	Open sun	Direct solar	Indirect solar	Open sun	Direct solar	Indirect solar	Open sun	Direct solar	Indirect solar
<i>Amaranthus graecizans</i>	33.0 ^{ab} (31.2, 38.2)	22.58 ^b (22.4, 22.8)	38.9 ^a (37.0, 42.0)	1.04 ^{ab} (0.89, 1.17)	0.09 ^b (0.09, 0.19)	1.12 ^a (0.95, 1.37)	20.4 ^d (13.2, 23.7)	6.96 ^b (6.89, 7.32)	17.4 ^a (16.0, 22.9)
<i>Amaranthus</i> spp.	18.8 ^{ab} (18.3, 20.9)	21.1 ^a (20.4, 22.4)	11.4 ^b (11.1, 11.8)	1.01 ^a (0.90, 1.10)	0.38 ^{ab} (0.35, 0.54)	0.21 ^b (0.18, 0.24)	12.0 ^a (11.6, 12.6)	11.2 ^{ab} (10.6, 11.4)	4.44 ^b (4.32, 4.48)
<i>Amaranthus viridis</i>	28.1 ^a (24.8, 30.2)	16.0 ^b (14.1, 17.3)	21.0 ^{ab} (17.4, 23.6)	1.90 ^a (1.76, 2.37)	0.81 ^b (0.75, 0.86)	1.20 ^{ab} (1.11, 1.32)	13.9 ^a (12.4, 19.4)	7.58 ^b (6.92, 8.23)	8.73 ^{ab} (8.26, 9.18)
<i>Corchorus olitorius</i>	33.3 ^{ab} (28.7, 34.8)	43.7 (40.6, 45.6)	29.1 ^b (28.7, 29.3)	0.22 ^{ab} (0.10, 0.31)	0.49 ^a (0.29, 0.60)	0.14 ^b (0.10, 0.17)	8.81 ^b (8.04, 9.07)	27.4 ^a (20.4, 28.7)	9.83 ^{ab} (9.53, 10.1)
<i>Curcubita maxima</i>	20.2 ^a (19.0, 20.9)	24.2 ^a (20.2, 25.3)	23.0 ^a (15.9, 30.5)	0.44 ^a (0.39, 0.54)	0.76 ^a (0.52, 1.24)	0.54 ^a (0.27, 0.64)	6.04 ^a (5.78, 6.59)	7.31 ^a (6.56, 7.66)	6.93 ^a (4.81, 9.14)
<i>Gisekia pharnaceoides</i> L.	18.1 ^{ab} (16.8, 19.0)	27.2 ^a (25.3, 28.5)	16.3 ^b (14.9, 17.7)	0.63 ^a (0.18, 0.68)	0.23 ^{ab} (0.18, 0.30)	0.12 ^b (0.4, 0.15)	6.55 ^a (6.24, 7.31)	13.2 ^b (9.26, 14.7)	6.30 ^a (5.12, 6.99)
<i>Ipomea aquatica</i>	21.1 ^{ab} (20.5, 22.0)	24.3 ^a (23.7, 24.6)	17.2 ^b (16.9, 17.4)	0.39 ^{ab} (0.33, 0.50)	0.29 ^b (0.23, 0.33)	0.52 ^a (0.37, 0.61)	7.62 ^{ab} (7.21, 8.05)	9.11 ^a (8.84, 9.43)	5.20 ^b (5.07, 5.21)
<i>Ipomea batatas</i> L.	28.8 ^a (20.5, 35.5)	32.1 ^a (22.8, 35.4)	26.9 ^a (24.1, 28.4)	0.74 ^a (0.26, 1.23)	0.78 ^a (0.63, 1.07)	0.59 ^a (0.48, 0.74)	14.0 ^a (10.7, 17.5)	15.6 ^a (11.1, 20.3)	11.8 ^a (10.5, 14.1)
<i>Manihot esculenta</i>	43.1 ^{as} (40.0, 40.7)	34.0 ^a (27.5, 47.2)	41.6 ^a (40.1, 50.4)	0.25 ^a (0.18, 0.32)	0.34 ^a (0.13, 0.66)	0.29 ^a (0.22, 0.41) ^a	17.7 ^a (12.6, 23.4)	13.5 ^a (7.6, 24.3)	20.5 ^a (14.2, 23.8)
<i>Vigna unguiculata</i> L.	35.3 ^{ab} (23.6, 40.9)	39.3 ^a (36.1, 41.4)	31.1 ^b (30.4, 31.8)	0.62 ^a (0.41, 0.99)	0.86 ^a (0.54, 1.10)	0.67 ^a (0.48, 0.90) ^a	12.1 ^a (1.1, 14.3)	12.0 ^a (10.2, 15.8)	9.77 ^a (9.13, 12.5)

Values are medians of four determinations (extraction /analysis) for lutein-zeaxanthin, α and β -carotene (n = 4). Values in parenthesis represent minimum and maximum values; values within a row not sharing a common superscript letter are significantly different at p < 0.05 (Friedman 2 way ANOVA, multiple pairwise comparison).

esculenta (20.53 mg/100 g DW) had higher α -carotene content compared to other vegetables. Open sun dried, direct, and indirect solar-dried α -carotene concentrations of *Ipomea batatas* L., *Vigna unguiculata* L., *Curcubita maxima* and *Manihot esculenta* were not significantly different.

The β -carotene concentrations of *Ipomea batatas* L., *Vigna unguiculata* L., *Curcubita maxima*, and *Manihot esculenta* were not significantly different. Open sun-dried *Amaranthus graecizans*, *Amaranthus viridis*, *Corchorus olitorius*, and *Gisekia pharnaceoides* L. were not significantly different from indirect solar drying in their β -carotene concentrations. For some of the vegetables such as *Corchorus olitorius*, *Gisekia pharnaceoides* L., *Ipomea aquatica*, and *Ipomea batatas* L. showed to have more β -carotene when compared to the other drying methods.

Effect of drying methods on tocopherol contents of DGLVs

The α and γ -tocopherol contents of different DGLV species subjected to different drying methods are presented in Table 6. *Ipomea batatas* L., *Vigna unguiculata* L., *Curcubita maxima*, and *Manihot esculenta* showed no significant difference (p < 0.05) in their γ -tocopherol concentrations from the different drying methods. Open sun-dried *Amaranthus spp.*, *Amaranthus viridis* and *Corchorus olitorius* were not significantly different (p < 0.05) from indirect drying in γ -tocopherol concentrations but different from direct-solar drying. *Amaranthus graecizans* showed to have the highest γ -tocopherol (6.96 mg/100 g DW) and α -tocopherol (40.83 mg/100 g DW) contents when indirectly solar-dried compared to the other drying methods. Open sun-dried *Ipomea batatas* L., and *Curcubita maxima* were significantly different (p < 0.05) from direct and indirect forms.

The α -tocopherol concentration among DGLV varieties differs among drying methods. The concentration of α -tocopherol in *Vigna unguiculata* L., *Curcubita maxima* and *Manihot esculenta* was not significantly different (p < 0.05) among the drying methods. However, there was also no significant difference in α -tocopherol concentration between direct and indirect solar drying methods for *Amaranthus spp.*, *Amaranthus viridis*, *Corchorus olitorius*, *Ipomea aquatica*, and *Ipomea batatas*. The concentrations of α -tocopherol in direct and indirect solar dried *Ipomea batatas* L. were higher (> 6 mg/100 g DW) and were significantly different (p < 0.05) from its open sun-dried form.

Discussion

Lutein-zeaxanthin, carotene, and tocopherols contents of fresh DGLVs

The concentrations of lutein-zeaxanthin, α and β -carotene were highest in *Manihot esculenta*, *Amaranthus graecizans* and *Cleome gynandra*. The α -carotene (0.38 mg/100 g FW) and β -carotene (4.66 g/100 g FW) concentrations of *Amaranthus spp* from this study were higher for α - and lower for β -carotene with

Table 6. α and γ -tocopherol contents (mg/100 g dry weight) of DGLVs by different drying methods.

Botanical names	γ -tocopherol			α -tocopherol		
	Open sun	Direct solar	Indirect solar	Open sun	Direct solar	Indirect solar
<i>Amaranthus graecizans</i>	5.79 ^{ab} (5.43, 5.85)	1.29 ^b (1.24, 1.34)	6.96 ^a (6.81, 7.21)	31.8 ^a (30.3, 33.1)	2.34 ^b (2.28, 2.46)	40.4 ^a (39.3, 40.7)
<i>Amaranthus spp.</i>	0.96 ^a (0.90, 1.01)	0.26 ^b (0.23, 0.31)	0.56 ^{ab} (0.38, 0.58)	12.5 ^a (12.0, 13.3)	6.24 ^{ab} (5.35, 6.92)	4.76 ^b (4.15, 5.01)
<i>Amaranthus viridis</i>	1.43 ^a (1.35, 1.47)	0.28 ^b (0.25, 0.39)	1.07 ^{ab} (0.94, 1.19)	25.9 ^a (25.1, 27.7)	6.14 ^b (5.41, 7.04)	19.8 ^{ab} (17.2, 22.7)
<i>Corchorus olitorius</i>	3.79 ^a (3.49, 4.01)	2.40 ^b (2.20, 2.59)	3.51 ^{ab} (3.35, 3.64)	50.6 ^a (47.7, 52.9)	22.2 ^b (20.2, 24.3)	40.8 ^{ab} (38.6, 43.2)
<i>Curcubita maxima</i>	3.49 ^a (2.61, 4.25)	3.73 ^a (1.40, 6.56)	4.99 ^a (2.26, 7.65)	14.6 ^a (12.2, 16.4)	16.3 ^a (8.07, 24.9)	18.5 ^a (6.36, 31.1)
<i>Gisekia pharnaceoides</i> L.	4.71 ^a (4.67, 4.80)	4.07 ^{ab} (3.95, 4.51)	3.88 ^b (3.63, 4.12)	6.68 ^b (6.15, 7.18)	28.2 ^a (27.4, 32.1)	8.47 ^{ab} (7.55, 9.32)
<i>Ipomea aquatica</i>	1.15 ^a (1.10, 1.25)	0.53 ^b (0.52, 0.58)	0.64 ^{ab} (0.63, 0.66)	15.8 ^a (14.5, 18.4)	1.55 ^b (1.42, 1.61)	3.96 ^{ab} (3.88, 4.09)
<i>Ipomea batatas</i> L.	1.25 ^a (1.02, 1.53)	0.90 ^a (0.36, 1.35)	0.74 ^a (0.52, 1.07)	24.1 ^a (16.0, 33.8)	6.73 ^b (1.46, 12.5)	6.49 ^b (4.81, 9.16)
<i>Manihot esculenta</i>	2.57 ^a (1.69, 3.45)	2.78 ^a (0.93, 4.91)	2.35 ^a (1.62, 3.08)	25.9 ^a (22.2, 29.2)	34.5 ^a (18.0, 59.6)	32.0 ^a (22.5, 37.5)
<i>Vigna unguiculata</i> L.	1.67 ^a (1.21, 2.74)	5.03 ^a (0.72, 10.3)	1.24 ^a (1.16, 6.41)	10.8 ^a (10.4, 14.4)	19.9 ^a (7.51, 34.9)	12.0 ^a (8.60, 15.6)

Values are medians of four determinations (extraction/analysis) for α and γ -tocopherol; values in parenthesis represent minimum and maximum values; values within a row not sharing a common superscript letter are significantly different (p -value < 0.05).

those reported by Gowele et al.^[20] (α -carotene: 0.32 mg/100 g FW and β -carotene: 4.82 g/100 g FW) from a study conducted in Tanzania. A study in Tanzania as reported by Gowele et al.^[20] showed *Vigna unguiculata* L. to have lower levels of β -carotene of 2.96 mg/100 g FW indicating a lower concentration as compared to this study. Gowele et al.^[20] reported for *Amaranthus spinosus*, *Amaranthus spp.*, *Cleome gynandra*, and *Vigna unguiculata* α -carotene contents ranging from 0.05–0.32 mg/100 g FW in comparison to our findings where; *Amaranthus spinosus* (0.96 mg/ 100 FWg), *Amaranthus spp* (0.38 mg/100 g FW), *Cleome gynandra* (0.12 mg/100 g FW) and *Vigna unguiculata* (0.13 mg/100 g FW), overall had higher levels of α -carotene contents. Results in this study show that *Vigna unguiculata* L., *Ipomea batatas* L., *Amaranthus spp.*, *Amaranthus graecizans* and *Cleome gynandra* could be good sources of carotenoids as consumption of 100 g FW of these vegetables can provide more than 60% of the RNI for retinol equivalents (RE) for females and males. These results are consistent with those reported by Gowele et al.^[20]. Except for this study, data on lutein-zeaxanthin contents in leafy vegetables are currently not available from Tanzania. A study conducted in South Africa of different *Ipomea batatas* L. species showed lutein-zeaxanthin contents ranging from 0.05 to 0.72 mg/100 g dry weight (DW)^[27], in comparison to ours which had 48.87 mg/100g DW meaning a higher content. Neugart et al.^[28] reported β -carotene concentration in some species was high for amaranth (up to 10.17 mg/100 g DW) and *Cleome gynandra* (up to 6.47 mg/100 g DW). The current study showed higher concentrations of β -carotene in *Cleome gynandra* (up to 31.74 mg/100 g DW) and *Amaranthus graecizans* (up to 6.47 mg/100g DW). These differences may be due to soil and agricultural practices or the type of analysis done using HPLC as the current study.

Nevertheless, the current study cannot be compared directly to previously reported concentrations, due to limited data on freshly frozen and later gently freeze-dried samples (on freeze-dried varieties). Moreover, the applied method using HPLC to

analyze carotenoids in the present study differs, with the exception of Gowele et al.^[20], from previous publications on DGLV, which frequently analyzed DGLV by photometric methods only, thus showing the tendency of higher carotenoid contents. The highest levels of α -tocopherol were detected in *Manihot esculenta* (38.40 mg/100 g FW), *Corchorus olitorius* (14.88 mg/100 g FW) and *Curcubita maxima* (12.33 mg/100 g FW). The highest level of α -tocopherol equivalents (α -TE) was detected in *Manihot esculenta*. A 100 g portion of fresh *Amaranthus spp*, *Corchorus olitorius*, *Ipomea batatas* L., and *Manihot esculenta* could provide more than 100% of the RNI for vitamin E for males and females aged 19–65 years (Table 2). To date, limited data are available on the tocopherol contents of *Amaranthus spp*, *Corchorus olitorius*, *Ipomea batatas* L. and *Manihot esculenta*. A previous study showed *Vigna unguiculata* L. to contain 0.02 mg/100g of α -TE contributing to 20% of RNI of female adults aged 19–50 years^[20]. The α -TE of *Vigna unguiculata* L. in the present study showed concentrations of 1.63 mg/100 g FW, which could contribute to 22% of RNI of female adults aged 19–65 years. Findings from this study suggest that *Amaranthus spp*, *Corchorus olitorius*, *Ipomea batatas* L., and *Manihot esculenta* could contribute to improving dietary TE intake.

Mineral and phytate contents

The calcium contents of *Amaranthus viridis* (469 mg/100 g FW) and *Corchorus olitorius* (235 mg/100 g FW) were similar to those reported in Nigeria^[29]. Gowele et al.^[20] reported higher concentrations in *Amaranthus spinosus* (289.1 mg/100 g FW), *Cleome gynandra* (206.1 mg/100 g FW), *Amaranthus spp* (853.7 mg/100 g FW); *Vigna unguiculata* L. (274.2 mg/100 g FW). Low concentrations of iron (6.62 mg/100 g DW), calcium (3.73 mg/100 g DW), zinc (5.03 mg/100 g DW) and magnesium (6.63 mg/100 g DW) in fresh cowpea leaves were reported by Bighaghire et al.^[13] using AAS analyzing techniques. The higher contents of the same minerals of cowpea leaves in the present

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study in mg/100 g when converted to dry weight basis (Fe: 11.05; Zn: 2.71; Ca: 948.92 and Mg: 267.8) may be due to the use of analyzing ICP-OES technique. The phytate levels in all DGLVs analyzed are low compared to a previous study conducted in Tanzania^[29].

Gowele et al.^[20] reported very high phytate levels of 739 mg/100 g FW (*Amaranthus spp*) and 334 mg/100 g FW (*Amaranthus spinosus*) compared to our results of 4.69 mg/100 g FW and 5.14 mg/100 g FW for *Amaranthus spp* and 334 mg/100 g FW *Amaranthus spinosus*, respectively. The calculated phytate:iron molar ratios for all DGLVs were below the suggested critical level (> 1) indicating good bioavailability of iron. The same was for all DGLVs for the phytate: zinc molar ratios being below the suggested critical level of 15, above which bioavailability of zinc is decreased^[30].

The phytate:calcium molar ratios in all DGLVs samples ranged between 0.01–0.06, which is below the critical level of 0.24 where calcium bioavailability is reduced^[30–32]. This indicates that the phytate levels of the analyzed DGLVs will not have a significant reducing effect on calcium bioavailability in humans and thus DGLVs can be an important and excellent source of minerals in the human diet. Gowele et al.^[20] reported similar phytate:calcium molar ratios in leafy vegetables from Tanzania. Total phytate values in the present study (all in the form of IP6) using the HPLC technique were lower compared to previously reported phytate content in DGLVs by Gowele et al.^[20] using an indirect measurement *via* reaction with Wade reagent (0.06% FeCl₃·6H₂O + 0.6% sulfosalicylic acid) and following photometric detection only. Previous studies reported that traditional food preparation and cooking methods such as heating and fermentation can reduce the levels of phytate in vegetables^[17,33,34]. Fermentation and traditional cooking methods can increase the contribution of DGLVs as important sources of Fe, Zn, Ca, and Mg^[21,35,26]. The HPLC techniques in the present study thus gave lower phytate values as compared to previous studies which overestimate the phytate content due to unspecific photometric methods. The low phytate thus makes the DGLVs as good sources of iron, zinc, calcium, and magnesium.

Impact of drying methods on carotenoid and tocopherol contents of DGLVs

The impacts of solar drying on the micronutrient quality of DGLVs among African countries are generally very low^[36]. The observed differences among different drying methods are likely going to affect the drying duration and product quality. The internal air temperatures of the direct and indirect solar dryers (Fig. 1) of this study are comparable to those reported in Africa and India^[37]. An investigation carried out in Tanzania revealed that when cowpea pea leaves were exposed to open sunlight, their β -carotene content was 87.56 mg/100 g DW^[38] which is greater than the results of the current study (12.1 mg/100 g DW). Consistent with our results, in Kenya, it was found that preserved and fresh cowpea leaves were rich in β -carotene with ranges of 0.25–36.55 mg/100 g DW^[39]. Data on open sun-dried *Ipomea batatas* L., *Vigna unguiculata* L., *Curcubita maxima* and *Manihot esculenta* were significantly different from direct and indirect drying methods. This may be due to different temperatures and drying times including the nature of DGLVs species. β -carotene was shown to be less destructed when using lower drying temperature as reported by Bishnoi et al.^[40] in fenugreek leaves. It was shown that the boiling and

sun-drying method significantly reduced provitamin A content in *Vigna unguiculata* samples, with a 50% reduction observed^[13]. The present study showed the β -carotene of open sun-dried *Curcubita maxima* to have 12.48 mg/100 g DW while its direct solar dried form had 7.3 mg/100 g DW and α -carotene of 0.92 mg/100 g DW (open sun-dried) and 0.76 mg/100 g DW (direct solar-dried). 100 g/DW *Curcubita maxima* could provide more than 100% of RE for vitamin A for both females and males. A study on *Vigna unguiculata* L. showed open unblanched cowpea leaves the β -carotene was reduced by 50% when blanched and sun dried^[41]. A study conducted by Nambafu et al.^[42], showed that *Curcubita maxima* and *Vigna unguiculata* L. subjected to direct solar drying showed significantly lower β -carotene contents ranging from 54–65 mg/100 g DW as compared to fresh blanched forms. Lee et al.^[43] reported that high-temperature cooking of carrots can alter their physical condition, with blanching allowing cellular lipids to solubilize carotenoids. Dripping during cooking may cause variations in beta-carotene retention in cooked vegetables. A study indicated a loss of β -carotene in *Cleome gynandra* and *Vigna unguiculata* when blanched then sundried^[44]. β -carotene also being sensitive to light its concentration may be affected^[45]. Although carotenoids are heat-sensitive, UV light may be less important than temperature. According to a study by Sonntag et al.^[46], temperature had an impact on the amount of carotenoids in amaranth leaves; they started to decrease at 20 °C but then dramatically increased 10–14 d after UV treatment. Unlike in the case of carotenoids, γ -tocopherol contents in the vegetables dried using different methods did not show a clear pattern or trend in γ -tocopherol reduction in dried samples. Limited data on γ - and α -tocopherol in DGLVs have been reported in Tanzania. Open sun-dried (4.71 mg/100 g DW) *Gisekea pharnaceoides* L. was the only DGLVs that showed to have a significant difference ($p < 0.05$) in γ -tocopherol concentration from indirect drying (3.88 mg/100 g DW) compared to other DGLVs. A study showed low contents of α -tocopherol observed in *Amaranthus spp* (0.67 mg/100 g DW) and *Vigna unguiculata* L. (0.02 mg/100 g DW)^[29]. Direct solar-dried pumpkin, amaranth, and cowpea leaves were shown to have α -tocopherol contents ranging from 51–65 mg/100 g DW in Kenya^[42]. The α -tocopherol concentration of *Curcubita maxima*, *Manihot esculenta*, and *Vigna unguiculata* among the three drying methods did not differ significantly ($p < 0.05$). The high α and γ -tocopherol contents in blanched open sun and indirect solar dried vegetables could be due to heat treatment (blanching and ambient/solar temperatures). This may soften plant tissue, releasing vitamin E from lipids, and eliminating tocopherol oxidase activity, which is present in all parts of the plant^[43]. Heat treatment could deactivate endogenous oxidative enzymes in vegetables making vitamin E more pronounced in results after extraction^[47]. There were also no significant differences in α and β -carotene and α and γ -tocopherol concentration in *Curcubita maxima*, *Ipomea batatas* L., *Manihot esculenta*, and *Vigna unguiculata* among the different drying methods. This could be due to the similarity of plant matrices, color, and the water content of these DGLVs. Since matrices are part of food structures, they are also subject to some major changes during processing, particularly in their physical state (e.g., due to phase and state transitions), chemical condition (e.g. due to thermal reactions and solubilization). Sun drying and solar drying is applied as a processing method to prolong the shelf

life of vegetables in villages but in turn also adds value to diets from a nutrition perspective. The drying process involves, mass and momentum transfer which may associate with changes in the physical, chemical, biochemical, microbiological, organoleptic, and nutritional properties of foods and severe heating may have deleterious effects in terms of nutrient loss^[41,45,48,49]. Differences in drying techniques and models of dryers used as well as freeze-drying handling of samples and environmental factors might be responsible for different carotenoids and tocopherols concentrations. Findings from this study suggests that open sun drying and solar drying techniques are good means of drying vegetables for food security despite their effect on the concentration of different carotenoids and tocopherols. More research may be done on the role of solar drying temperatures, UV, and matrices of *Amaranthus graciezans*, *Manihot esculenta*, *Corchorus olitorius*, and *Curcubita maxima* play in providing tocopherols for human diets.

Conclusions

Provitamin A carotenoids, lutein-zeaxanthin, tocopherols, and minerals can be found in the DGLVs in Lindi, Tanzania. The phytate concentrations were very low; which means DGLV are excellent sources of minerals, as the concentrations are much lower than those in previous reports in the literature. However, if all the analyzed DGLVs would be consumed in combination with local diets at all times during the year adequate micronutrient supply of carotenoids, tocopherols, and minerals could be achieved especially for *Amaranthus* and *Manihot esculenta*. Preserving DGLVs using affordable drying methods such as direct sun-drying can be used as a strategy for achieving all year around availability of micronutrient-dense foods despite reduction of some nutritional properties of individual varieties. The study has demonstrated that improved solar drying methods such as using direct solar dryers can lead to nutrient retention and provide a cheap but also more time-efficient way of drying vegetables in the village setting. The remarkable content of iron and carotenoids found in the DGLVs can play an important role in meeting daily recommended micronutrient requirements of the farmers in the study area and therefore have the potential to improve food and nutrition security. Promoting consumption of the DGLVs in Lindi supports the Tanzania Nutrition Policy whose goals are to eliminate malnutrition, to improve nutrition status particularly women and children and to ensure food security.

Author contributions

The authors confirm contribution to the paper as follows: study conception and design, data collection, HPLC analysis, statistical analysis, draft manuscript preparation: Tiisekwa AB; freezing dried the DGLVs samples, HPLC analysis, chromatograms cleanup and statistical analysis: Stuetz W; reviewing and refining the manuscript: Chove BE, Mongi R, Rybak C, Stuetz W. All authors reviewed the results and approved the final version of the manuscript.

Data availability

All data generated or analyzed during this study are included in this published article, and are available from the corresponding author on reasonable request.

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Conflict of interest

The authors declare that they have no conflict of interest.

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References

1. Kumar D, Kumar S, Shekhar C. 2020. Nutritional components in green leafy vegetables: a review. *Journal of Pharmacognosy and Phytochemistry* 9:2498–502
2. IRIS. 2017. *Overcoming the challenges of undernutrition in Tanzania through 2021 - Reflecting on trends, questions, and future scenarios for urgent nutrition action*. (Accessed 8 Dec. 2021). www.iris-france.org/wp-content/uploads/2017/03/Tanzania-Final-report.pdf
3. Stuetz W, Goweke V, Kinabo J, Bundala N, Mbwana et al. 2019. Consumption of dark green leafy vegetables predicts vitamin A and iron intake and status among female small-scale farmers in Tanzania. *Nutrients* 11:1025
4. Durst P, Bayasgalanbat N. 2014. Promotion of underutilized indigenous food resources for food security and nutrition in Asia and the Pacific. FAO, National regional office for Asia and the Pacific, Bangkok. 212 pp. www.fao.org/fileadmin/templates/rap/files/meetings/2012/120531-report.pdf
5. Schippers RR. 2006. Traditional vegetables in Africa. *Proc. International Symposium on the Nutritional value and Water Use of Indigenous Crops for Improved Livelihoods*, University of Pretoria. 2016. Pretoria: The Centre for Nutrition, University of Pretoria.
6. Dongus S. 2000. *Vegetable production on open spaces in Dar es Salaam - spatial changes from 1992 to 1999*. (Accessed 5 May 2019). www.cityfarmer.org/daressalaam.html
7. Weinberger K, Msuya J. 2004. Indigenous vegetables in Tanzania: significance and prospects. *Technical Bulletin No. 31*. AVRDC—The World Vegetable Center, AVRDC Publication 04-600, Shanhua, Taiwan. 70 pp. www.zef.de/fileadmin/user_upload/9a22_Weinberger_Msuya_2004.pdf
8. Hall CM, Rasmussen LV, Powell B, Dyngeland C, Jung S, et al. 2022. Deforestation reduces fruit and vegetable consumption in rural Tanzania. *Proceedings of the National Academy of Sciences of the United States of America* 119:e2112063119
9. Chirande L, Charwe D, Mbwana H, Victor R, Kimboka S et al. 2015. Determinants of stunting and severe stunting among under-fives in Tanzania: evidence from the 2010 cross-sectional household survey. *BMC Pediatrics* 15:165
10. TDHS-MIS. 2022. *Tanzania demographic and health survey and malaria indicator survey 2022*. (Accessed 1 May 2024). <https://dhsprogram.com/pubs/pdf/FR382/FR382.pdf>

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11. Gowele VF, Kinabo J, Jumbe T, Rybak C, Stuetz W. 2021. High prevalence of stunting and anaemia is associated with multiple micronutrient deficiencies in school children of small-scale farmers from Chamwino and Kilosa districts, Tanzania. *Nutrients* 13:1576
12. Eleraky L, Issa R, Maciel S, Mbwana H, Rybak C, et al. 2022. Anthropometrics, haemoglobin status and dietary micronutrient intake among Tanzania and Mozambican pigeon pea farmers. *Nutrients* 14:2914
13. Bighaghire R, Okidi L, Muggaga C, Ongeng D. 2021. Traditional vegetable preservation technologies practiced in Acholi subregion of Uganda improves mineral bioavailability but impacts negatively on the contribution of vegetables to household needs for micronutrients. *Food Science & Nutrition* 9:589–604
14. van 't Hag L, Danthe J, Handschin S, Mutuli GP, Mbugue D, et al. 2020. Drying of African leafy vegetables for their effective preservation: the difference in moisture sorption isotherms explained by their microstructure. *Food & Function* 11:955–64
15. Sangija F, Martin H, Matemu A. 2021. African nightshades (*Solanum nigrum* complex): the potential contribution to human nutrition and livelihoods in sub-Saharan Africa. *Comprehensive Reviews in Food Science and Food Safety* 20:3284–318
16. Udomkun P, Romuli S, Schock S, Mahayothee B, Sartas M, et al. 2020. Review of solar dryers for agricultural products in Asia and Africa: an innovation landscape approach. *Journal of Environmental Management* 268:110730
17. Vorster H, Stevens J, Steyn G. 2008. Production systems of traditional leafy vegetables: challenges for research and extension. *South African Journal of Agricultural Extension* 37:85–96
18. Milandu AA, Chove LM. 2022. Safety of traditional leafy vegetable powders from Lindi in Tanzania. *Tanzania Journal of Agricultural Sciences* 21:22–33
19. Majili ZS, Nyaruhucha C, Kulwa K, Mutabazi K, Rybak C, et al. 2020. Preferences and consumption of pigeon peas among rural households as determinants for developing diversified products for sustainable health. *Sustainability* 12:6130
20. Gowele VF, Kinabo J, Jumbe T, Kirschmann C, Frank J, et al. 2019. Provitamin A carotenoids, tocopherols, ascorbic acid and minerals in indigenous leafy vegetables from Tanzania. *Foods* 8:35
21. Grebenstein N, Frank J. 2012. Rapid baseline-separation of all eight tocopherols and tocotrienols by reversed-phase liquid-chromatography with a solid-core pentafluorophenyl column and their sensitive quantification in plasma and liver. *Journal of Chromatography A* 1243:39–46
22. Lux PE, Freiling M, Stuetz W, von Tucher S, Carle R, et al. 2020. (Poly)phenols, carotenoids and tocopherols in corn (*Zea mays* L.) kernels as affected by phosphate fertilization and sowing time. *Journal of Agricultural Food Chemistry* 68:612–22
23. Stuetz W, Carrara VI, McGready R, Lee SJ, Erhardt JG, et al. 2012. Micronutrient status in lactating mothers before and after introduction of fortified flour: cross-sectional surveys in Maela refugee camp. *European Journal of Nutrition* 51:425–34
24. WHO/FAO. 2004. Vitamin and mineral requirements in human nutrition: report of a joint FAO/WHO expert consultation, Bangkok, Thailand, 21–30 September 1998. WHO: Geneva, Switzerland. 286 pp.
25. Zeller E, Schollenberger M, Kühn I, Rodehutschord M. 2015. Hydrolysis of phytate and formation of inositol phosphate isomers without or with supplemented phytases in different segments of the digestive tract of broilers. *Journal of Nutritional Science* 4:e1
26. Phillippy BQ, Bland JM. 1988. Gradient ion chromatography of inositol phosphates. *Analytical Biochemistry* 175:162–66
27. Phahlane CJ, Laurie SM, Shoko T, Manhivi VE, Sivakumar D. 2021. An evaluation of phenolic compounds, carotenoids, and antioxidant properties in leaves of South African cultivars, Peruvian 199062.1 and USA's Beauregard. *Frontiers in Nutrition* 8:773550
28. Neugart S, Baldermann S, Ngwene B, Wesonga J, Schreiner M. 2017. Indigenous leafy vegetables of Eastern Africa — a source of extraordinary secondary plant metabolites. *Food Research International* 100:411–22
29. Ejoh IS, Wireko-Manu FD, Page D, Catherine M, Renard CMGC. 2021. Traditional green leafy vegetables as underutilised sources of micronutrients in a rural farming community in south-west Nigeria I: estimation of vitamin C, carotenoids and mineral contents. *South African Journal of Clinical Nutrition* 34:40–45
30. Castro-Alba V, Lazarte CE, Bergenstahl B, Granfeldt Y. 2019. Phytate, iron, zinc, and calcium content of common Bolivian foods and their estimated mineral bioavailability. *Food Science & Nutrition* 7:2854–65
31. Ma G, Li Y, Jin Y, Zhai F, Kok FJ, et al. 2007. Phytate intake and molar ratios of phytate to zinc, iron and calcium in the diets of people in China. *European Journal of Clinical Nutrition* 61:368–74
32. Norhaizan ME Jr, Nor Faizadatul Ain AW. 2009. Determination of phytate, iron, zinc, calcium contents and their molar ratios in commonly consumed raw and prepared food in Malaysia. *Malaysian Journal Nutrition* 15:213–22
33. Ijeoma O, Adeyemi OO. 2020. Effect of fermentation on some minerals and antinutrient content of *Telfaira Occidentalis* and *Gnetum Africanum* leaves. *International Journal of Advances in Scientific Research and Engineering* 6:6–14
34. Udousoro II, Ekop RU, Udo EJ. 2013. Effect of thermal processing on antinutrients in common edible green leafy vegetables grown in Ikot Abasi, Nigeria. *Pakistan Journal of Nutrition* 12:162–67
35. Misci C, Taskin E, Dall'Asta M, Fontanella MC, Bandini F, et al. 2021. Fermentation as a tool for increasing food security and nutritional quality of indigenous African leafy vegetables: the case of *Cucurbita* sp. *Food Microbiology* 99:103820
36. Ndukwu MC, Bennamoun L, Abam FI. 2018. Experience of solar drying in Africa: presentation of designs, operations, and models. *Food Engineering Reviews* 10:211–44
37. Natarajan SK, Elangovan E, Elavarasan RM, Balaraman A, Sundaram S. 2022. Review on solar dryers for drying fish, fruits, and vegetables. *Environmental Science and Pollution Research* 29:40478–506
38. Ahmed A, Almas S, Kissaka K, Suleiman R. 2023. Effect of sun-dry on nutritional and sensory acceptability of wilted African leafy vegetables: a case study of Morogoro Region, Tanzania. *Frontiers in Sustainable Food Systems* 7:1161961
39. Owade JO, Abong' GO, Okoth MW, Mwang'ombe AW. 2022. Comparative characterization of trends and patterns of physical and chemical attributes of optimal and traditional processed cowpea leaves. *Journal of Food Quality* 2022:1503221
40. Bishnoi S, Chhikara N, Singhania N, Ray AB. 2020. Effect of cabinet drying on nutritional quality and drying kinetics of fenugreek leaves (*Trigonella foenum-graecum* L.). *Journal of Agriculture and Food Research* 2:100072
41. Aguilera JM. 2019. The food matrix: implications in processing, nutrition and health. *Critical Reviews in Food Science and Nutrition* 59:3612–29
42. Nambafu R, Swaleh S, Nyambaka H. 2021. Determination of β -carotene and α -tocopherol content in selected fresh and dry vegetables in Butula in Busia county. *Advances in Research* 22:12–20
43. Lee S, Choi Y, Jeong HS, Lee J, Sung J. 2017. Effect of different cooking methods on the content of vitamins and true retention in selected vegetables. *Food Science and Biotechnology* 27:333–42
44. Njoroge EW, Matofari JW, Mulwa RMS, Anyango JO. 2015. Effects of blanching time/temperature combination coupled with

- solar-drying on the nutritional and microbial quality of indigenous leafy vegetables in Kenya. *African Journal of Food Science and Technology* 6:209–19
45. Naikwade P. 2015. Effect of drying methods on nutritional value of some vegetables. *Proceeding of the National Conference on Conservation of Natural Resources & Biodiversity for Sustainable Development-2014* 6:72–79
46. Sonntag F, Liu H, Neugart S. 2023. Nutritional and physiological effects of postharvest UV radiation on vegetables: a review. *Journal of Agricultural Food Chemistry* 71:9951–72
47. Wafula EN, Franz CMAP, Rohn S, Huch M, Mathara JM, Trierweiler B, et al. 2016. Fermentation of African indigenous leafy vegetables to lower post-harvest losses, maintain quality and increase product safety. *African Journal Horticultural Sciences* 9:1–13
48. Natumanya P, Twinomuhwezi H, Igwe VS, Maryam S, Awuchi CG. 2021. Effects of drying techniques on nutrient retention and phytochemicals in selected vegetables. *European Journal of Agriculture and Food Sciences* 3:5–14
49. Sobowale SS, Olatidoye OP, Olorode OO, Sokeye OK. 2010. Effect of preservation methods and storage on nutritional quality and sensory properties of leafy vegetables consumed in Nigeria. *Journal of Medical and Applied Biosciences* 2:46–56



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