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Nutritional variability in 42 cultivars of spineless cactus pear cladodes for crop improvement

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In addition to their use in traditional medicine, cactus pear (*Opuntia* species) are a source of vegetal nutrients in many countries. The aims of this study were to determine variability in the nutritional value of 42 spineless cactus pear cultivars, identify correlations between nutritional traits, and determine superior cultivars based on cladode nutritional value. Cladodes of 42 spineless cactus pear cultivars grown under the same glasshouse conditions were collected from the Roodeplaat research farm of the Agricultural Research Council, South Africa. The sliced, dried and powdered cladodes were subjected to quantitative analysis of their vitamin C, β-carotene and mineral element contents. Significant variations in nutritional values were observed among the different cultivars. Vitamin C content ranged from 8.95 mg/100 g in cultivar Ofer to 124 mg/100 g in cultivar Malta, whilst β-carotene content ranged from 3.9 mg/100 g in cultivar Murado to 31.4 mg/100 g in cultivar Cross X. Potassium and calcium were the most abundant mineral elements present, whilst iron was found to be the least present among the mineral elements quantified. A significant positive association was established between calcium (Ca), potassium (K) and zinc (Zn) contents. A direct selection for increased concentration of Ca could be a selection criterion for the development of population with an indirect improvement of K and Zn concentrations. The observed variations highlight the need for a careful cultivar selection for population and product development using spineless cactus pear cladodes.

Introduction

Opuntia species (family: Cactaceae) commonly known as cactus pear, have a long history of use as a source of fruits and vegetables (Bouzoubaa et al., 2014). Within the genus, *Opuntia ficus-indica* (L.) Mill. is the most economical, distributed and consumed species with about 50,000 ha cultivated in Mexico alone, linked to a production of about 300 000 megatons of cultivated cactus pear for fresh produce (Yahia and Mondragon-Jacobo, 2011). The consumption of its fruit and cladodes is becoming common in different parts of the world (Ayadi et al., 2009; De Santiago et al., 2018). The young cladodes are consumed either fresh or cooked, as green vegetables in salads, soups and as commercial products, such as beverages and sauces (Méndez et al., 2015; Trivedi and Raval, 2017; De Santiago et al., 2018). *Opuntia ficus-indica* is a climate-smart crop due to its high water use efficiency and its ability to grow under harsh conditions. The importance of this plant is particularly evident in arid and semi-arid environments where accessibility to most vegetables is relatively poor (Morales et al., 2012). Its cladodes are an excellent alternative food source for humans, and it is also a drought-resilient feed resource for animal consumption especially during drought conditions (Domínguez López, 1995; López-García et al., 2001; Aruwa et al., 2018).

The spineless *Opuntia ficus-indica* and *Opuntia robusta* J.C. Wendl. cultivars such as the Burbank spineless cultivars differ agro-morphologically, bearing cladodes of different shapes and sizes, as well as fruits and flowers of different colour (Wilson, 2007; De Wit et al., 2019; Novoa et al., 2019). The spineless cultivars are relatively non-invasive and legally allowed in South Africa, for example, for cultivation and trading unlike the spiny cultivars, which were declared as category 1 weed by the Conservation of Agricultural Resources Act (CARA) and not allowed for planting and trading (Novoa et al., 2019). Due to their advantages and benefits, including their ability to adapt well in arid and semi-arid environments and their nutritional values, these spineless cultivars are considered as a functional food for the future and as a potential crop in broadening the food base, thus ensuring food security (du Toit et al., 2018). Currently, there are 42 Burbank's spineless cultivars available in South Africa and for which

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extensive research on their use in food broadening projects is ongoing (De Wit et al., 2017; du Toit et al., 2018). The morphological attributes of cladodes from the 42 spineless Burbank cultivars have been previously described (De Wit et al., 2019).

In broadening the food base, extensive research focusing on species/cultivar nutritional traits and constituent bioactive compounds is of vital importance for germplasm characterisation, utilisation and selection in breeding and agro-processing programmes. Studies suggest that the bioactive compound profile and nutritional properties of *Opuntia* cladodes can vary with species/cultivar type, plant age, and the environment (Aruwa et al., 2018). Due to the paucity of information on the available spineless cultivars, the aims of this study were to determine nutritional value variability in cladodes of 42 spineless *Opuntia ficus-indica* and *Opuntia robusta* cultivars, establish correlation between the nutritional traits and identify superior cultivars that can potentially be used in the food industry as functional foods.

Materials and methods

2.1. Plant material collection and preparation

Cladodes were randomly harvested from 42 Burbank's spineless *Opuntia* spp. cultivars imported from the USA into South Africa more than a century ago (du Toit et al., 2018). The cladodes were obtained from plants grown under the same glasshouse conditions in the Agricultural Research Council, Roodeplaat research farm, South Africa. The cultivars Robusta and Montery belong to *Opuntia robusta* while the remaining 40 cultivars are from *Opuntia ficus-indica*. The cladodes (about one-year-old) were sliced into small pieces and oven-dried at 50 °C in the dark. The material was then ground into fine powder using a pulverizing mill.

2.2. Quantification of vitamin C content

Vitamin C content of the cladodes was determined using a method described by Odriozola-Serrano et al. (2007) with slight modifications. Plant material (0.2 g) was extracted using 10 ml of 4.5% metaphosphoric acid. The mixture was sonicated in an ultrasonic bath containing ice-cold water for 30 min before filtration. The prepared samples were then analysed using a Shimadzu HPLC (LC-2030C 3D, Shimadzu Corporation, Kyoto, Japan) equipped with a C_{18} Luna® column (150 × 4.6 mm, 5μ) at 25 °C. Water: acetonitrile: formic acid (99: 0.9: 0.1 v/v/v) was used as the mobile phase at a flow rate of 1 ml/min in isocratic mode, with an injection volume of 20 μl and detection wavelength of 245 nm. Ascorbic acid prepared at different concentrations was used as a standard for the preparation of a calibration curve. The vitamin C content was expressed as mg/100 g dry weight (DW) of sample.

2.3. Quantification of β-carotene content

β-carotene content was determined using a method described by Biehler et al. (2010) with slight modifications. The procedure was carried out in the dark to avoid the effect of light on β-carotene. Methanol (5 ml) was added to 0.2 g sample and vortexed for 10 s, followed by the addition of 15 ml hexane: acetone (1:1 v/v). The mixture was vortexed for 10 s before sonicating in an ultrasonic bath containing ice-cold water for 15 min. Saturated sodium chloride solution (5 ml) was added to the mixture, vortexed for 10 s and centrifuged at 2000 rpm for 2 min. The collected supernatant was filtered through a 0.45 μm syringe filter before analysis using a Shimadzu HPLC (LC-2030C 3D, Shimadzu Corporation, Kyoto, Japan) equipped with a C_{18} Luna® column (150 × 4.6 mm, 5μ) at 35 °C. Acetonitrile: dichloromethane: methanol (70:20:10, v/v/v) was used as a mobile phase at a flow rate of 1 ml/min in isocratic mode, with an injection volume and detection wavelength of 20 μl and 450 nm, respectively. Identification and quantification of β-carotene was achieved by plotting a calibration curve using a β-carotene standard. β-carotene content was expressed as mg/100 g DW of sample.

2.4. Quantification of mineral elements

Quantification of mineral elements was done according to the method described by Ang and Lee (2005) with slight modifications. An amount of 0.5 g of sample was digested with 10 ml of 1:3 (v/v) nitric acid: hydrochloric acid. The mixture was boiled over a hotplate at 95 °C until the sample was dissolved. The mineral elements in the digested samples were quantified using inductively coupled plasma atomic emission spectroscopy (ICPE-9820, Shimadzu Corporation, Kyoto, Japan).

2.5. Data analysis

Data were subjected to analysis of variance using Statsoft (Statistica 8) software. The mean values were further separated by Duncan's multiple range test based on $P = 0.05$. Data were reported as mean ± standard error of triplicate analyses. In order to determine the degree

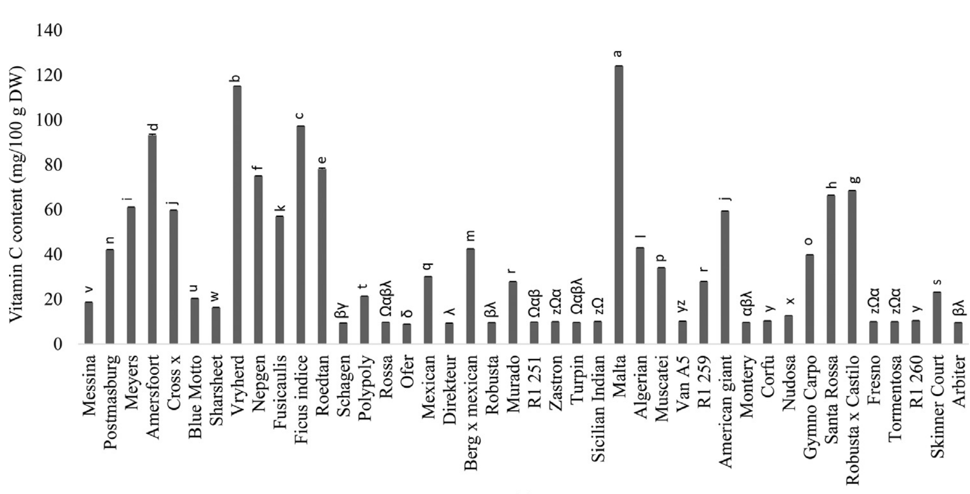

Fig. 1. Vitamin C content of cladodes obtained from 42 spineless cactus pear cultivars. Results are presented as mean values with bars containing different letters indicating significant difference ($P = 0.05$) according to Duncan's multiple range test. DW = Dry weight.

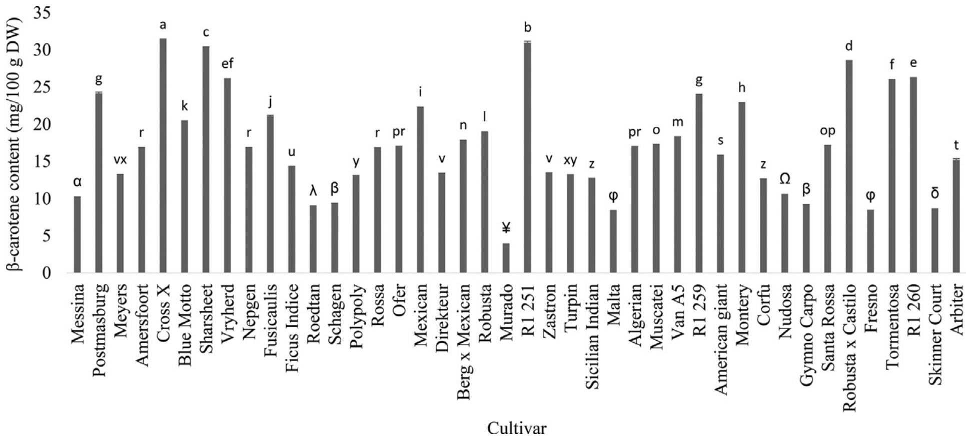

Fig. 2. β-carotene content of cladodes obtained from 42 spineless cactus pear cultivars. Results are presented as mean values with bars containing different letters indicating significant difference ($P = 0.05$) according to Duncan's multiple range test. DW = Dry weight.

of association between the nutritional traits as well as the pattern of the associations and differences in the different cultivars, correlation coefficients and principal component analysis (PCA) were computed using GenStat for Windows 17th edition (VSN, International Hempstead, UK) (Payne et al., 2016) statistical software. The method described by Chatfield and Collis (1980) was used to extract principal components (PCs) from the mean data set for the tested cultivars.

Table 1
Mineral element content (mg/100 g DW) of cladodes obtained from 42 spineless cactus pear cultivars.

Cultivar	Ca	Fe	K	Mg	Mn	Na	P	Zn
Messina	1182.00 ± 2.31$^{@}$	8.31 ± 0.09^{f}	2206.67 ± 6.67^{v}	649.33 ± 0.67^{c}	56.73 ± 0.07$^{\Omega}$	99.00 ± 0.20^{b}	190.67 ± 1.15^{qrs}	11.83 ± 0.05^{rs}
Postmasburg	1860.67 ± 5.46^{y}	4.94 ± 0.05^{kl}	1711.33 ± 5.21^{z}	623.33 ± 3.33^{e}	121.13 ± 0.35^{ij}	65.07 ± 0.37^{w}	169.60 ± 3.33^{t}	18.79 ± 0.08^{g}
Meyers	1608.67 ± 4.37λ	1.71 ± 0.07^{u}	2146.67 ± 17.64^{w}	586.67 ± 6.67^{jkl}	120.47 ± 0.18^{j}	76.60 ± 0.20^{n}	216.00 ± 4.67^{op}	11.57 ± 0.06st
Amersfoort	1408.67 ± 4.67$^{£}$	1.39 ± 0.03^{v}	2240.00 ± 11.55^{uv}	562.00 ± 1.15^{n}	56.60 ± 0.12$^{\Omega}$	82.13 ± 0.59^{hi}	186.80 ± 4.00^{rs}	11.45 ± 0.16^{tu}
Cross X	1576.67 ± 4.67$^{\varphi}$	2.85 ± 0.10^{p}	1840.00 ± 23.86^{y}	596.67 ± 1.76^{hij}	92.73 ± 0.64^{p}	97.47 ± 1.33^{c}	262.00 ± 0.67^{ij}	12.50 ± 0.09^{p}
Blue Motto	2186.67 ± 6.67^{s}	1.33 ± 0.07^{v}	2966.67 ± 33.33^{p}	498.00 ± 0.00^{r}	160.33 ± 0.64^{f}	54.73 ± 0.35^{x}	258.00 ± 5.26^{j}	17.87 ± 0.04^{h}
Sharsheet	2560.00 ± 11.55^{n}	0.20 ± 0.06^{y}	2980.00 ± 11.55^{p}	597.33 ± 1.76^{ghij}	176.40 ± 0.12^{d}	76.00 ± 0.37^{no}	335.33 ± 1.76^{c}	16.71 ± 0.11^{j}
Vryherd	2140.00 ± 11.55^{t}	2.87 ± 0.08^{p}	2686.67 ± 6.67^{r}	600.00 ± 1.15^{gh}	120.67 ± 0.52^{j}	73.33 ± 0.73^{qrs}	226.67 ± 1.76^{n}	13.19 ± 0.12^{o}
Nepgen	2633.33 ± 6.67^{m}	37.20 ± 0.20^{b}	3700.00 ± 11.55^{i}	**670.67 ± 0.67^{ab}**	162.60 ± 0.61^{e}	83.20 ± 0.40^{gh}	269.33 ± 5.03^{hi}	14.19 ± 0.19^{n}
Fusicaulis	2706.67 ± 6.67^{k}	0.91 ± 0.05^{w}	3993.33 ± 17.64^{de}	582.00 ± 1.15^{l}	**220.00 ± 0.00^{a}**	74.27 ±0.29^{pqr}	**402.00 ± 2.91^{a}**	27.67 ± 0.13^{b}
Ficus Indice	1820.00 ± 2.31^{z}	6.49 ± 0.09^{i}	4226.67 ± 24.04^{b}	600.67 ± 0.67^{gh}	75.20 ± 0.40^{u}	**113.40 ± 0.81^{a}**	248.00 ± 0.12^{kl}	17.37 ± 0.11^{i}
Roedtan	1322.00 ± 0.00$^{¥}$	1.42 ± 0.02^{v}	2240.00 ± 11.55^{uv}	518.00 ± 0.00^{p}	40.67 ± 0.13$^{\lambda}$	72.80 ± 0.12^{rs}	244.00 ± 1.03^{lm}	10.56 ± 0.07^{y}
Schagen	3520.00 ± 0.00^{d}	2.37 ± 0.10^{rs}	4040.00 ± 23.09^{d}	601.33 ± 1.33^{gh}	58.80 ± 0.31^{z}	69.47 ± 0.59^{tu}	269.33 ± 5.81^{hi}	18.72 ± 0.20^{g}
Polypoly	1504.00 ± 1.15$^{\sigma}$	2.65 ± 0.09^{pq}	3760.00 ± 20.00^{h}	508.00 ± 4.16^{q}	111.33 ± 0.29^{l}	79.53 ± 0.27^{kl}	196.73 ± 4.37^{q}	14.19 ± 0.07^{n}
Rossa	1780.67 ± 1.33$^{\Omega}$	0.59 ± 0.02^{x}	2373.33 ± 13.33^{t}	600.00 ± 1.15^{gh}	87.27 ± 0.13^{r}	75.13 ± 0.81^{op}	275.33 ± 3.33^{gh}	18.48 ± 0.15^{g}
Ofer	2286.67 ± 6.67^{q}	0.74 ± 0.01^{wx}	3060.00 ± 11.55^{o}	524.67 ± 4.67^{op}	66.53 ± 0.24^{y}	67.27 ± 0.35^{v}	359.33 ± 0.12^{b}	16.25 ± 0.12^{k}
Mexican	2106.67 ± 6.67^{u}	2.75 ± 0.09^{pq}	3520.00 ± 11.55^{k}	662.00 ± 1.15^{b}	156.00 ± 0.64^{g}	64.73 ± 0.44^{w}	235.33 ± 2.45^{m}	14.76 ± 0.14^{m}
Direkteur	3840.00 ± 11.55^{b}	1.56 ± 0.06^{uv}	**4980.00 ± 30.55^{a}**	572.00 ± 5.29^{m}	183.47 ± 0.44^{c}	72.80 ± 0.23^{s}	255.33 ± 0.48^{jk}	20.60 ± 0.18^{e}
Berg x Mexican	1655.33 ± 8.97β	2.29 ± 0.05^{rst}	2886.67 ± 6.67^{q}	608.00 ± 3.06^{fg}	50.60 ± 0.12$^{\alpha}$	68.53 ± 0.27^{uv}	157.13 ± 1.60^{u}	10.84 ± 0.18^{wxy}
Robusta	1260.00 ± 5.03$^{\$}$	2.92 ± 0.04^{p}	2286.67 ± 6.67^{u}	623.33 ± 5.46^{e}	159.67 ± 0.48^{f}	78.67 ± 0.75^{l}	212.67 ± 1.05^{p}	15.81 ± 0.13^{l}
Murado	2206.67 ± 6.67^{s}	2.49 ± 0.08^{qr}	3686.67 ± 6.67^{i}	592.67 ± 4.67^{hijk}	44.07 ± 0.29$^{\beta}$	77.07 ± 0.35^{mn}	184.40 ± 3.53^{s}	10.71 ± 0.08^{xy}
R1251	1344.00 ± 2.31$^{¥}$	**54.67 ± 0.27^{a}**	1858.67 ± 5.21^{y}	**669.33 ± 1.76^{ab}**	67.00 ± 0.23^{y}	69.73 ± 0.24^{tu}	157.80 ± 1.15^{u}	9.38 ±0.05^{z}
Zastron	2773.33 ± 6.67^{j}	2.17 ± 0.10st	3600.00 ± 34.64^{j}	619.33 ± 3.53^{e}	86.20 ± 0.23^{s}	78.40 ± 0.50^{lm}	158.33 ± 0.67^{u}	11.37 ± 0.24^{tu}
Turpin	3460.00 ± 11.55^{e}	4.93 ± 0.02^{kl}	4133.33 ± 17.64^{c}	**674.67 ± 4.37^{a}**	67.53 ±0.07^{y}	77.27 ± 0.35^{mn}	195.13 ± 1.15^{qr}	11.89 ± 0.09^{rs}
Sicilian Indian	2580.00 ± 0.00^{n}	2.85 ± 0.02^{p}	3680.00 ± 23.09^{i}	558.67 ± 2.40^{n}	109.80 ± 0.42^{m}	73.87 ± 0.29^{pqrs}	286.67 ± 2.91^{f}	14.86 ± 0.04^{m}
Malta	2073.33 ± 6.67^{v}	5.04 ± 0.04^{k}	3213.33 ± 17.64^{m}	532.00 ± 6.11^{o}	28.60 ± 0.12$^{\sigma}$	81.07 ± 0.13^{ij}	354.00 ± 2.31^{b}	18.57 ± 0.09^{g}
Algerian	3066.67 ± 13.33^{h}	4.70 ± 0.01^{l}	3866.67 ± 17.64^{f}	**667.33 ± 4.06^{ab}**	55.80 ± 0.12$^{\Omega}$	85.87 ± 0.13^{f}	225.33 ± 3.71^{n}	18.81 ± 0.07^{g}
Muscatei	**4146.67 ± 17.64^{a}**	5.01 ± 0.08^{k}	3833.33 ± 6.67^{fg}	588.67 ± 4.37^{ijkl}	44.27 ± 0.13$^{\beta}$	83.87 ± 0.52^{g}	144.00 ± 4.16^{v}	11.10 ± 0.04^{uvw}
Van A5	2246.67 ± 13.33^{r}	7.65 ± 0.13^{g}	2860.00 ± 11.55^{q}	614.00 ± 1.15^{ef}	71.67 ± 0.41^{w}	72.80 ± 0.42^{s}	329.33 ± 0.67^{c}	11.01 ± 0.14^{vwx}
R1259	3626.67 ± 17.64^{c}	11.13 ± 0.11^{d}	3986.67 ± 6.67^{e}	516.67 ± 1.33^{pq}	69.13 ± 0.13^{x}	74.53 ± 0.37^{pq}	282.00 ± 1.15^{fg}	19.96 ± 0.05^{f}
American giant	2660.00 ± 0.00^{l}	5.99 ± 0.13^{j}	2253.33 ± 6.67^{uv}	601.33 ± 1.76^{gh}	90.47 ± 0.24^{q}	65.27 ± 0.13^{w}	320.67 ± 0.24^{d}	12.06 ± 0.00^{qr}
Montery	2706.67 ± 6.67^{k}	1.31 ± 0.05^{v}	2433.33 ± 13.33^{s}	600.00 ± 1.15^{gh}	194.47 ± 0.84^{b}	83.93 ± 0.24^{g}	303.33 ± 0.58^{e}	**28.40 ± 0.05^{a}**
Corfu	2560.00 ± 11.55^{n}	9.57 ± 0.06^{e}	3953.33 ± 17.64^{e}	**670.67 ± 0.67^{ab}**	115.73 ± 0.13^{k}	85.73 ± 0.48^{f}	272.67 ± 0.12^{h}	17.88 ± 0.01^{h}
Nudosa	3140.00 ± 0.00^{g}	3.57 ± 0.07^{o}	3793.33 ± 17.64^{gh}	634.00 ± 5.03^{d}	121.87 ± 0.18^{i}	76.53 ± 0.18^{n}	290.67 ± 0.67^{f}	21.27 ± 0.09^{d}
Gymno Carpo	3173.33 ± 6.67^{f}	4.45 ± 0.08^{m}	3406.67 ± 6.67^{l}	638.00 ± 1.15^{d}	30.07 ± 0.24$^{\varphi}$	86.67 ± 0.64^{f}	135.87 ± 4.90^{v}	8.68 ± 0.08$^{\Omega}$
Santa Rossa	1614.67 ± 0.67$^{\lambda}$	0.84 ± 0.02^{wx}	1943.33 ± 11.62^{x}	594.67 ± 2.91^{hijk}	92.73 ± 0.13^{p}	64.00 ± 0.23^{w}	142.27 ± 0.64^{v}	8.57 ± 0.09$^{\Omega}$
Robusta x Castilo	2873.33 ± 6.67^{i}	4.05 ± 0.07^{n}	4240.00 ± 11.55^{b}	**677.33 ± 1.76^{a}**	100.67 ± 0.48^{o}	90.40 ± 0.50^{e}	301.33 ± 2.91^{e}	20.53 ± 0.04^{e}
Fresno	1716.67 ± 7.69$^{\alpha}$	4.17 ± 0.14^{n}	3073.33 ± 26.67^{no}	558.67 ± 9.96^{n}	79.20 ± 0.35^{t}	80.47 ± 0.58^{jk}	222.67 ± 1.76^{no}	12.26 ± 0.17^{pq}
Tormentosa	2360.00 ± 11.55^{p}	2.07 ± 0.05^{t}	3113.33 ± 17.64^{n}	598.67 ± 1.33^{ghi}	102.40 ± 0.35^{n}	76.73 ± 0.18^{n}	300.00 ± 6.36^{e}	23.73 ± 0.18^{c}
R1260	1484.00 ± 4.62$^{\sigma}$	12.41 ± 0.04^{c}	1303.33 ± 11.62$^{\Omega}$	592.67 ± 0.67^{hijk}	56.47 ± 0.13$^{\Omega}$	98.33 ± 0.33^{bc}	157.80 ± 6.00^{u}	11.31 ± 0.14^{tuv}
Skinner Court	3833.33 ± 13.33^{b}	7.15 ± 0.09^{h}	3413.33 ± 13.33^{l}	584.67 ± 1.33^{kl}	126.07 ± 0.29^{h}	95.07 ± 0.53^{d}	242.67 ± 2.40^{lm}	23.87 ± 0.09^{c}
Arbiter	2420.00 ± 11.55^{o}	0.20 ± 0.04^{y}	3620.00 ± 30.55^{j}	580.67 ± 0.67^{lm}	73.27 ± 0.24^{v}	70.67 ± 0.13^{t}	362.67 ± 2.91^{b}	17.89 ± 0.11^{h}

Mean values with different letters within a column indicate significant differences ($P = 0.05$) between the cultivars according to Duncan's multiple range test. DW = Dry weight.

Table 2
A selection of the top twenty cultivars with high concentrations for each nutritional trait.

Species	Macro elements					Micro elements					Vitamin C	Beta carotene	Total frequency
	K	Ca	Mg	P	Frequency	Na	Fe	Mn	Zn	Frequency			
Messina		X	X		2	X	X			2	X	X	6
Postmasburg			X		1		X	X	X	3	X		5
Meyers					0			X		1	X		2
Amersfoort					0	X				1	X	X	3
Cross x				X	1	X		X		2		X	4
Blue Motto				X	1			X	X	2		X	4
Sharsheet				X	1			X	X	2	X	X	5
Vryherd					0			X		1	X		2
Nepgen	X	X	X	X	4	X	X	X		3	X	X	9
Fusicaulis	X			X	2			X	X	2	X		5
Ficus indice	X		X		2	X	X		X	3	X		6
Roedtan					0					0			0
Schagen	X	X	X	X	4				X	1			5
Polypoly	X				1	X		X		2			3
Rossa				X	2			X	X	2		X	5
Ofer		X		X	2				X	1	X	X	5
Mexican	X		X		2			X		1			3
Direkteur	X	X		X	3			X	X	2	X	X	7
Berg x mexican			X		1					0		X	2
Robusta			X		1	X	X	X	X	4	X		6
Murado	X				1	X				1		X	3
R1 251			X		1		X			1			2
Zastron	X	X	X		3	X				1			4
Turpin	X	X	X		3	X	X			2			5
Sicilian Indian fig	X	X		X	3			X		1	X		5
Malta				X	1	X	X		X	3	X	X	6
Algerian	X	X	X		3	X	X		X	3	X	X	8
Muscatei	X	X			2	X	X			2		X	5
Van A5			X	X	2		X			1	X	X	5
R1 259	X	X		X	3		X		X	2	X		6
American giant		X	X	X	3		X	X		2		X	6
Montery		X	X	X	3	X		X	X	3			6
Corfu	X	X	X	X	4	X	X	X	X	4			8
Nudosa	X	X	X	X	4		X	X	X	3	X		8
Gymno Carpo	X	X	X		3	X	X			2	X	X	7
Santa Rossa					0			X		1	X	X	3
Robusta x Castilo	X	X	X	X	4	X	X	X	X	4			8
Fresno					0	X	X			2		X	3
Tormentosa		X		X	2			X	X	2		X	5
R1 260					0	X	X			2			2
Skinner Court	X	X			2	X	X	X	X	4			6
Arbiter	X	X		X	3				X	1			4

Results and discussion

3.1. Vitamin C and β-carotene contents

Vitamins are naturally occurring essential nutrients found in plants and play a significant role in metabolism (Kim et al., 2016). Some of the vitamins reported in cactus pear include vitamins C and E (Kuti, 2004; Aruwa et al., 2018; du Toit et al., 2018). Cactus pear plant also contains natural pigments known as carotenoids with significant antioxidant capacities (El-Kharrassi et al., 2016). Both vitamins A and C are characterized by their high antioxidant properties, and are thus regarded as nutritional or dietary antioxidants (FAO/WHO, 2004; Kim et al., 2016).

The vitamin C and β-carotene content of cladodes from the 42 spineless *Opuntia ficus-indica* and *Opuntia robusta* cultivars studied are shown in Fig. 1 and Fig. 2, respectively. Significant variations in vitamin C and β-carotene contents were observed with the vitamin C content ranging from 8.95 mg/100 g DW in cultivar Ofer to 124 mg/100 g DW in cultivar Malta (Fig. 1). Using similar quantification method, the vitamin C content in 16 cultivars (cvs. Postmasburg, Meyers, Amersfoort, Cross X, Vryherd, Nepgen, Fusicaulis, Ficus indice, Roedtan, Berg x Mexican, Malta, Algerian, American giant, Gymno Carpo, Santa Rossa, and Robusta x Castilo) were at least two-fold of what was recorded in two widely consumed commercial vegetables, *Brassica oleracea* and *Beta vulgaris* (Moyo et al., 2018). About 8 mg/100 g of vitamin C content is adequate to prevent signs of scurvy (associated with vitamin C deficiency) in infants (FAO/WHO, 2004). Thus, the incorporation of small cladode amounts from vitamin C-rich cultivars in food products holds a potential for eliminating vitamin C deficiencies. Although it is believed that vitamin C content in cactus pear fruits is significantly high compared to the cladodes on a fresh weight basis (Feugang et al., 2006), the vegetative parts are considered as an excellent and reliable source of vitamin C, as the supply of vegetables and their preservation can be extended for longer periods than fruits (FAO/WHO, 2004).

Significant variations in β-carotene content ranging from 3.9 mg/100 g DW in cultivar Murado to 31.4 mg/100 g DW in cultivar Cross X were recorded (Figure 2). In particular, 13 cultivars (cvs. Postmasburg, Cross X, Blue Motto, Sharsheet, Vryherd, Fusicaulis, Mexican, R1 251, R1 259, Montery, Robusta x Castilo, Tormentosa, and R1 260) had a significantly high β-carotene content (above 20 mg/100 g DW) compared to other cultivars. Their β-carotene content was more than 11 times greater than that of cabbage (*Brassica oleracea* var. *capitata*), which was quantified using the same method (Moyo et al., 2018). Stintzing and Carle (2005) indicated an average of 11.3 – 53.5 μg/100 g β-carotene content in fresh cladodes. As expected, the dry

Table 3
Amount of cladodes needed to satisfy daily nutrient requirements in human nutrition based on adult (19–65 years old) group.

Nutritional trait	Daily recommended intake (mg/day)		§Cladode mass needed to satisfy the daily requirement (g DW)				Reference
	Male	Female	Male		Female		
			Range	Average	Range	Average	
Vitamin A*	0.27	0.30	1.72 – 13.81	3.14	1.91 – 15.35	3.49	FAO/WHO (2004)
Vitamin C	45	45	36.26 – 502.2	125.10	36.26 – 502.2	125.10	FAO/WHO (2004)
Calcium	800	800	19.29 – 67.68	33.93	19.29 – 67.68	33.93	FAO/WHO (2004)
Magnesium	220	260	32.48 – 44.18	36.74	38.39 – 52.21	43.42	FAO/WHO (2004)
Zinc#	7	4.9	24.65 – 81.68	44.73	17.25 – 57.18	31.31	FAO/WHO (2004)
Iron#	11.4	24.5	20.85 – 5700	197.92	44.81 – 12250	425.35	FAO/WHO (2004)
Potassium	3500	3500	70.28 – 268.54	112.12	70.28 – 268.54	112.12	World Health Organisation (2012)(World Health Organisation 2012)
Manganese	2.3	1.8	0.34 – 8.04	2.35	0.27 – 6.29	1.84	Institute of Medicine (US) Panel on Micronutrients (2001)Institute of Medicine (US) Panel on Micronutrients 2001
Phosphorus	700	700	174.13 – 515.20	284.36	174.13 – 515.20	284.36	Institute of Medicine, Food and Nutrition Board (1997)(Institute of Medicine, Food and Nutrition Board 1997)

*Vitamin A expressed as retinol equivalent. #Daily recommended intake for zinc and iron based on moderate and 12% bioavailability, respectively. §Cladode mass expressed as a range (using the highest and lowest cultivar nutrient content) or average (using the average nutrient value of all the cultivars). DW = Dry weight.

cladodes in the current study had more β-carotene content compared to fresh cladodes due to the concentrating effect of an effective drying process (du Toit et al., 2018).

3.2. Quantification of mineral elements

Nutritional elements are essential for human health and metabolic activities (Abdel-Hameed et al., 2014). Unlike the cladodes of spineless cactus grown in northeastern Brazil where cultivar had no effect on mineral element composition (Batista et al., 2009), significant differences in the macro- (potassium, calcium, magnesium and phosphorus) and micro-elements (sodium, iron, manganese and zinc) were observed amongst the cultivars (Table 1). Potassium and calcium were the most abundant elements present with the highest levels at 4980 mg/100 g DW in cultivar Direkteur and 4146.67 mg/100 g DW in cultivar Muscatei, respectively. These potassium and calcium concentrations were comparable (Mayer and Cushman, 2019) or higher than what was previously reported in cactus pear cladodes (Shoop et al., 1977; Teles, 1977; Nobel, 1983, Ben Salem et al., 2005; Stintzing and Carle, 2005; Osuna et al., 2014, Méndez et al., 2015). Intake of potassium has been linked with lowering of blood pressure while calcium intake is associated with promotion of bone health, neuromuscular function, blood clotting and reduction of osteoporosis risks (Kim et al., 2016; World Health Organization, 2009). Micronutrient deficiencies, especially zinc and iron deficiency, remain a major global concern particularly in the developing countries of the world (Hambidge, 2000; Hallberg, 2001). Although generally low in most vegetables, zinc plays a crucial role in cellular, immune and antioxidant functions, whilst iron is associated with haemoglobin formation and transportation of oxygen from lungs to body tissues (Kim et al. 2016). The highest iron content (54.67 mg/100 g DW) was recorded in cultivar R1251, whilst the highest zinc content (28.40 mg/100 g DW) was recorded in cultivar Montery. These values were higher than the iron and zinc contents previously recorded in some cladodes (Shoop et al., 1977; Nobel, 1983; Osuna et al., 2014; Mayer and Cushman, 2019). In comparison to cactus pear fruits (Feugang et al, 2006; Kunyanga et al., 2014), *Opuntia* species cladodes used in this study have higher macro- (K, Ca, Mg, P) and micronutrient (Zn, Fe, Mn, Na) contents.

Table 2 provides a summary of the top twenty cultivars with high concentration for each nutritional trait. Overall, cultivar Nepgen ranked consistently very high, appearing in the top 20 cultivars with significantly high concentrations for all the nutritional traits except zinc. Other cultivars among the top 20 with significantly high concentrations of multiple nutritional traits (eight out of ten traits) are Algerian, Corfu, Nudosa and Robusta x Castilo. The cultivars Nepgen, Schagen, Corfu, Nudosa and Robusta x Castilo showed significantly high concentrations for all the macro-elements (K, Ca, Mg and P), while cultivars Robusta, Corfu, Robusta x Castilo, and Skinner Court demonstrated the same for all the micro-elements (Na, Fe, Mn and Zn). Therefore, these cultivars can be selected for multiple traits in the development of population for the traits of interest in the cactus-breeding programme in South Africa. The cultivar Robusta (*O. robusta*) contained high concentrations of all the micro-elements

Table 4
Pearson correlation coefficient analysis in cladodes of spineless cactus pear.

Nutritional traits	Ca	Fe	K	Mg	Mn	Na	P	Zn	Vitamin C
Ca	1.00								
Fe	−0.10	1.00							
K	**0.71**	−0.14	1.00						
Mg	0.11	**0.38**	0.06	1.00					
Mn	0.12	−0.06	0.12	0.05	1.00				
Na	−0.02	0.08	0.07	0.24	−0.19	1.00			
P	0.20	−0.19	0.27	−0.22	**0.36**	−0.13	1.00		
Zn	**0.40**	−0.21	**0.37**	−0.08	**0.57**	0.05	**0.63**	1.00	
Vitamin C	−0.22	−0.02	−0.11	−0.10	−0.14	0.18	0.02	−0.13	1.00
Beta Carotene	−0.16	0.25	**−0.34**	0.17	**0.33**	−0.06	0.10	0.14	0.02

Ca = Calcium; Fe = Iron; K = Potassium; Mg = Magnesium; Mn = Manganese; Na = Sodium; P = Phosphorus; Zn = Zinc. Values in bold are different from zero with a significant level α=0.05.

while cultivar Montery (*O. robusta*) had the highest zinc content. Nevertheless, both *O. robusta* cultivars had low potassium and β-carotene content. du Toit et al. (2018) similarly observed low carotene content in *O. robusta* cv. *robusta* when compared to some *O. ficus-indica* cultivars. In general, high nutritional trait contents were observed in *O. ficus-indica* cultivars.

Table 3 presents the amount of cladodes needed to satisfy daily recommended vitamin and mineral requirements in human nutrition based on adult (19–65 years old) group. The amount of cladode required to meet recommended daily nutrient requirements varied widely, depending on the cultivars used. The highest variation was observed with iron, ranging from 20.85 g to 12250 g in terms of the amount required to meet daily recommended amount. This further amplifies the need for selecting the appropriate cultivar in order to realize the intended nutritional benefit. On the other hand, the ranges of cladode needed to meet the daily recommended amount for vitamin A and manganese were comparatively low (1.72 to 15.35 g and 0.27 to 8.04 g for vitamin A and manganese, respectively). With the use of appropriate cultivars, cactus pear cladodes has potential nutritional benefit in food security, especially in combating micronutrient deficiency.

3.3. Estimation of correlation for nutritional traits

The estimates of genetic correlation matrix between the nutritional components are presented in Table 4. The results showed significant ($P \leq 0.05$) and positive association between Ca and K, and between Ca and Zn (Table 4). Due to the significant and positive association observed between Ca, K and Zn, direct selection for increased concentration of Ca could be considered as a selection criterion for development of population with an indirect improvement of K and Zn concentrations, and vice-versa in the spineless cactus pear nutritional quality breeding programme. Furthermore, the level of the degree of association ($r = 0.71$) for Ca and K was higher compared to the rest of the nutritional traits, followed by P and Zn ($r = 0.63$). Similarly, K was significantly and positively associated with Zn but negatively associated with β-carotene. Mn was also positively and significantly associated with P, Zn and β-carotene. The concentration of Fe is significantly ($P \leq 0.05$) and positively correlated with the concentration of Mg indicating that the improvement of Fe will most likely improve the concentration of Mg simultaneously. Karim et al. (1997) reported significant and positive association among N, P, K, Ca and Mg in both the fruits and cladodes of *Opuntia* spp. Significant and positive correlations among nutritional traits allow for simultaneous and direct trait selection towards improving the nutritional quality in a breeding programme within a short period of time using suitable parental lines (Gerrano et al., 2021). Therefore, the nutritional traits with positive and significant correlations would be preferred as selection criteria for high nutrient dense element for the simultaneous improvement of nutritional traits in cactus pear breeding programme.

3.4. Principal component analysis of nutritional traits

The principal component analysis (PCA) is a multivariate analysis that assists in differentiating significant associations between the traits studied and explains the positive associations between a large dataset of variables in terms of a small number of independent components (Saleem et al., 2016). The PCA results showed that the first four principal components (PCs) were significantly important and explained 71.43% of the total variation among the cactus pear cultivars (Table 5). The first two principal components (PC1 and PC2) accounted for 43.71% of the entire genetic variation among the nutritional traits studied. The first principal component (PC1) had an eigenvalue of 2.65 and accounted for 26.49% of total variation. This PC was significantly

Table 5
Principal component analysis for nutritional compositions in spineless cactus pear cladodes revealing eigenvalue, total variance and eigenvectors and their contributions to total variation explained by the first four PC axes.

PC	Eigen value	Total variance (%)		Eigen vectors (loading) for									
		Variability	Cumulative	Ca	Fe	K	Mg	Mn	Na	P	Zn	Vitamin C	Beta Carotene
1	2.65	26.49	26.49	0.41 (16.67)*	−0.21 (4.50)	0.41 (17.01)	−0.08 (0.72)	0.36 (12.79)	−0.08 (0.70)	0.43 (18.54)	0.52 (26.74)	−0.15 (2.30)	−0.02 (0.04)
2	1.72	17.22	43.71	−0.32 (10.26)	0.19 (3.64)	−0.41 (16.83)	0.04 (0.19)	0.44 (19.55)	−0.19 (3.62)	0.20 (4.08)	0.19 (3.46)	0.00 (0.00)	0.62 (38.36)
3	1.59	15.90	59.61	0.28 (7.76)	0.49 (23.96)	0.21 (4.30)	0.66 (44.14)	0.07 (0.46)	0.30 (9.00)	−0.19 (3.42)	0.03 (0.10)	−0.19 (3.66)	0.18 (3.22)
4	1.18	11.82	71.43	−0.12 (1.43)	−0.05 (0.30)	0.03 (0.10)	0.02 (0.05)	−0.04 (0.17)	0.65 (41.76)	0.22 (4.80)	0.20 (4.19)	0.68 (46.73)	0.07 (0.46)

*Contribution of each nutritional trait (%) given in parenthesis

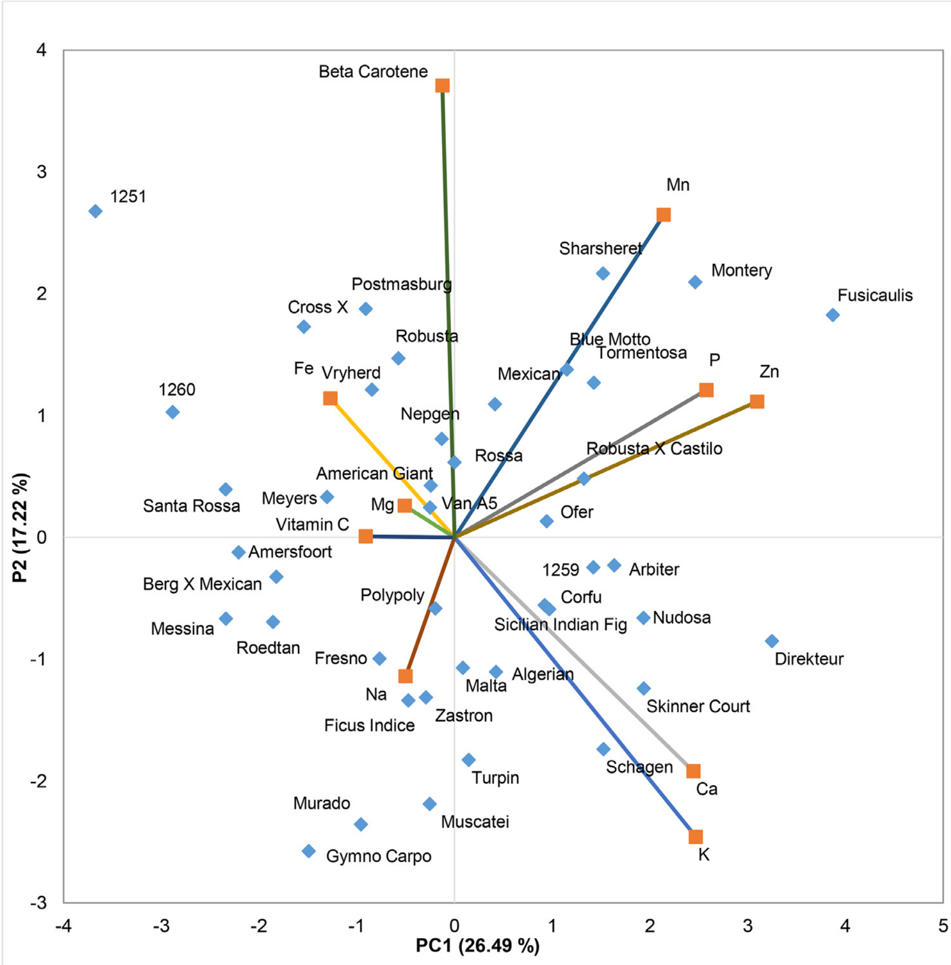

Fig. 3. The principal component analysis score plot of first and second principal components showing the overall genetic variation in nutritional composition in the spineless cactus pear cladodes.

associated and influenced by the mineral elements Ca, K, Mn, P and Zn, which were important in contributing to the variability among the tested cultivars in such a way that these mineral elements explained the component loadings greater than ±0.3 that need to be considered (Hair et al., 1998), as they are meaningful in defining the variances compared to the rest of the mineral elements in this PC. Ca and K had same eigenvectors contributing equally to the genetic variation in PC1 (Table 5). Zinc contributed more than 26% of the variation in PC1 compared to the rest of the nutritional traits (Table 5). The PC2 had an eigenvalue of 1.72, which contributed 17.22% of genetic variation. The nutritional traits Mn and β-carotene are the main contributing factor, compared to the rest of the traits and contributed variation of 19.55 and 38.36%, respectively in this PC. Moreover, Ca and K contributed 10.26 and 16.83% variations, respectively in this PC. PC3 had eigenvalues of 1.59 contributing 15.90% of genetic variation, with Fe and Mg being the major contributing traits to the variation. PC4 had an eigenvalue of 1.18, which contributed 11.82% of the variation. In this PC, Na and vitamin C played a major role in contributing the widest variation with 41.76% and 46.73%, respectively. In general, the PC1 (26.49%) and PC2 (17.22%) explained most of the variation among the cactus pear cultivars tested based on the degree of the concentration of nutritional traits.

The biplot revealed the interrelationship of the tested cultivars and nutritional traits (Fig. 3). The evaluated cactus pear cultivars located at the top right side of the biplot were positively associated with high concentration of Mn, P, and Zn (Table 4 and Fig. 3). These mineral elements are also positively and significantly associated (Table 4). The evaluated cultivars that are found at the top left quadrant are positively associated with the nutritional traits β-carotene, Mg, vitamin C and Fe with high concentration values. Similarly, the cultivars that are located at the bottom left quadrant are associated with high concentration of Na. The cultivars located at the bottom right quadrant are positively and significantly associated with Ca and K, and these elements are positively and significantly associated with each other (Table 4 and Fig. 3). The test genotypes concentrated around the origin had relatively similar genetic characteristics for the performance of the nutritional traits evaluated. The PCA grouped different cactus pear cultivars into different clusters based on the concentration of mineral elements, vitamin C and β-carotene (Fig. 3), which showed that there was a wide genetic difference among the tested cultivars. The diverse cactus pear cultivars belonging to different groups could be deployed in the cactus pear hybridization program with the cultivars belonging to other groups for the improvement of cactus pear cultivars for the traits of interest. Furthermore, the biplot revealed that the cactus pear cultivars such as R1251, R1260, Gymno carpo, Direkteur, and Fusicaulis were the most divergent cultivars among the rest of the tested cultivars that could be used as potential/candidate cultivars for population development due to their peculiar alleles that differentiated them from others. Moreover, these cultivars could also be selected for use as functional foods in the food industries. Hence, the genotype-trait association biplot allows the identification of candidate cactus pear genotypes with high concentration of multiple traits for breeding.

Conclusion

The findings indicate that the cladodes of spineless cactus pear cultivars are important sources of nutrients and can be considered as functional foods with a potential to be used in the battle against malnutrition or micronutrient deficiency including vitamin C, zinc and iron deficiency. The spineless cultivars are excellent sources of macronutrients, particularly potassium and calcium. The cultivars Nepgen, Algerian, Corfu, Nudosa and Robusta x Castilo had significantly high concentrations of multiple nutritional traits. As a result of significant and positive association established between Ca, K and Zn, direct selection for increased Ca concentration could serve as a selection criterion for developing population with an indirect improvement of K and Zn concentrations. The study also shows that variation among the cactus pear cultivars differ with the degree of the concentration of nutritional traits. Overall, these spineless *Opuntia* species cultivars show great variability in nutritional traits, a valuable tool for crop genetic improvement. This variability underscores the need for a careful cultivar selection for population and product development using spineless cactus pear cladodes.

Declaration of Competing Interest

None.

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References

Abdel-Hameed, E.S., Nagaty, M.A., Salman, M.S., Bazaid, S.A., 2014. Phytochemicals, nutritionals and antioxidant properties of two prickly pear cactus cultivars (Opuntia ficus-indica Mill.) growing in Taif, KSA. Food Chem. 160, 31–38.

Ang, H.H., Lee, K.L., 2005. Analysis of mercury in Malaysian herbal preparations. J. Med. Biomed. Res. 4, 31–36.

Aruwa, C.E., Amoo, S.O., Kudanga, T., 2018. *Opuntia* (Cactaceae) plant compounds, biological activities and prospects − a comprehensive review. Food Res. Int. 112, 328–344.

Ayadi, M.A., Abdelmaksoud, W., Ennouri, M., Attia, H., 2009. Cladodes from *Opuntia ficus-indica* as a source of dietary fiber: Effect on dough characteristics and cake making. Ind. Crops Prod. 30, 40–47.

Batista, Â.M.V., Ribeironeto, A.C., Lucena, R.B., Santos, D.C., Dubeux, J., Mustafa, A.F., 2009. Chemical composition and ruminal degradability of spineless cactus grown in northeastern Brazil. Rangel. Ecol. Manag. 62, 297–301.

Ben Salem, H., Abdouli, H., Nefzaoui, A., El-Mastouri, A., Ben Salem, L., 2005. Nutritive value, behaviour, and growth of Barbarine lambs fed on oldman saltbush (*Atriplex nummularia* L.) and supplemented or not with barley grains or spineless cactus (*Opuntia ficus-indica* f. *inermis*) pads. Small Rumin. Res. 59, 229–237.

Biehler, E., Mayer, F., Hoffmann, L., Krause, E., Bohn, T., 2010. Comparison of three spectrophotometric methods for carotenoid determination in frequently consumed fruits and vegetables. J. Food Sci. 75, 55–61.

Bouzoubaa, Z., Essoukrati, Y., Tahrouch, S., Hatimi, A., Gharby, S., Harhar, H., 2014. Phytochemical study of prickly pear from southern Morocco. J. Saudi Soc. Agricul. Sci. 15, 155–161.

Chatfield, C., Collis, A., 1980. Introduction to Multivariate Analysis. CRC Press, Boca Raton.

De Santiago, E., Domínguez-Fernández, M., Cid, C., De Peña, M., 2018. Impact of cooking process on nutritional composition and antioxidants of cactus cladodes (*Opuntia ficus-indica*). Food Chem. 240, 1055–1062.

De Wit, M., Du Toit, A., Fouche, H.J., Hugo, A., Venter, S.L., 2019. Screening of cladodes from 42 South African spineless cactus pear cultivars for morphology, mucilage yield and mucilage viscosity. Acta Horticult. 1247, 47–56.

De Wit, M., Hugo, A., Shongwe, N., 2017. South African cactus pear seed oil: a comprehensive study on 42 spineless Burbank *Opuntia ficus-indica* and *Opuntia robusta* cultivars. Eur. J. Lipid Sci. Technol. 120, 1700343.

Domínguez López, A., 1995. Use of the fruits and stems of the prickly pear cactus (*Opuntia* spp.) into human food. Food Sci. Technol. Int. J. 1, 65–69.

du Toit, A., de Wit, M., Osthoff, G., Hugo, A., 2018. Antioxidant properties of fresh and processed cactus pear cladodes from selected *Opuntia ficus-indica* and *O. robusta* cultivars. South Afr. J. Bot. 118, 44–51.

El-Kharrassi, Y., Mazri, M.A., Benyahia, H., Benaouda, H., Nasser, B., El-Mzouri, M., 2016. Fruit and juice characteristics of 30 accessions of two cactus pear species (*Opuntia ficus indica* and *Opuntia megacantha*) from different regions of Morocco. LWT − Food Sci. Techno. 65, 610–617.

FAO/WHO, 2004. Vitamin and Mineral Requirements in Human Nutrition. FAO/WHO, Geneva. second edition.

Feugang, J.M., Konarski, P., Zou, D., Stintzing, F.C., Zou, C., 2006. Nutritional and medicine use of cactus pear (*Opuntia* spp.) cladodes and fruits. Front. Biosci. 11, 2574–2589.

Gerrano, A.S., Mathew, I., Shayanowako, A.I.T., Amoo, S., Mellem, J.J., Van Rensburg, W.S., Bairu, M.W., Venter, S.L., 2021. Variation in mineral element composition of landrace taro (*Colocasia esculenta*) corms grown under dryland farming system in South Africa. Heliyon 7, e06727.

Hair, J.F., Andrson, J.R., Tatham, R.E., Black, W.C., 1998. Multivariate Data Analysis. Prentice-Hall International, London. fifth edition.

Hallberg, L., 2001. Perspectives on nutritional iron deficiency. Annu. Rev. Nutr. 21, 1–21 .

Hambidge, M., 2000. Human zinc deficiency. The J. Nutr. 130, 1344S–1349S.

Institute of Medicine (US) Panel on Micronutrients, 2001. Dietary Reference Intakes for Vitamin A, Vitamin K, Arsenic, Boron, Chromium, Copper, Iodine, Iron, Manganese, Molybdenum, Nickel, Silicon, Vanadium, and Zinc. National Academies Press, Washington (DC).

Institute of Medicine, Food and Nutrition Board, 1997. Dietary Reference Intakes for Calcium, Phosphorus, Magnesium, Vitamin D, and Fluoride. National Academies Press, Washington (DC).

Karim, M.R., Felker, P., Bingham, R.L., 1997. Correlations between cactus pear (*Opuntia* spp.) cladode nutrient concentrations and fruit yield and quality. Ann. Arid Zone 37, 159–171.

Kim, M.J., Moon, Y., Tou, J.C., Mou, B., Waterland, N.L., 2016. Nutritional value, bioactive compounds and health benefits of lettuce (*Lactuca sativa* L.). J. Food Comp. Anal. 49, 19–34.

Kunyanga, C.N., Vellingiri, V., Imungi, K.J., 2014. Nutritional quality, phytochemical composition and health protective effects of an under-utilized prickly cactus fruit (*Opuntia stricta* Haw.) collected from Kenya. Afr. J. Food, Agric. Nutr. Dev. 14, 9561–9577.

Kuti, J.O., 2004. Antioxidant compounds from four *Opuntia* cactus pear fruit varieties. Food Chem. 85, 527–533.

López-García, J., Fuentes-Rodríguez, J., Rodríguez, R., 2001. Production and use of *Opuntia* as forage in northern Mexico cactus (*Opuntia* spp.) as forage. FAO Plant Prod. Prot. Pap. 29–36.

Mayer, J.A., Cushman, J.C., 2019. Nutritional and mineral content of prickly pear cactus: A highly water-use efficient forage, fodder and food species. J. Agron. Crop Sci. 205, 625–634.

Méndez, L.P., Flores, F.T., Martín, J.D., Rodríguez-Rodríguez, E.M., Romero, C.D., 2015. Physicochemical characterization of cactus pads from *Opuntia dillenii* and *Opuntia ficus-indica*. Food Chem. 188, 393–398.

Morales, P., Ramírez-Moreno, E., Sanchez-Mata, M., Carvalho, A.M., Ferreira, I.C.F.R., 2012. Nutritional and antioxidant properties of pulp and seeds of two xoconostle cultivars (*Opuntia joconostle* F.A.C. Weber ex Diguet and *Opuntia matudae* Scheinvar) of high consumption in Mexico. Food Res. Int. 46, 279–285.

Moyo, M., Amoo, S.O., Aremu, A.O., Gruz, J., Subrtová, M., Jarošová, M., Tarkowski, P., Doležal, K., 2018. F Determination of mineral constituents, phytochemicals and antioxidant qualities of *Cleome gynandra*, compared to *Brassica oleracea* and *Beta vulgaris*. Front. Chem. 5, 128.

Nobel, P.S., 1983. Nutrient levels in cacti-relation to nocturnal acid accumulation and growth. Am. J. Bot. 70, 1244–1253.

Novoa, A., Flepu, V., Boatwright, J.S., 2019. Is spinelessness a stable character in cactus pear cultivars? Implications for invasiveness. J. Arid Environ. 160, 11–16.

Odriozola-Serrano, I., Aguiló-Aquayo, I., Soliva-Fortuny, R., Gimeno-Añó, V., Martín-Belloso, O., 2007. Lycopene, vitamin C, and antioxidant capacity of tomato juice as affected by high-intensity pulsed electric fields critical parameters. J. Agric. Food Chem. 55, 9036–9042.

Osuna, H.T.G., Mendoza, A.B., Morales, C.R., Rubio, E.M., Star, J.V., Ruvalcaba, R.M., 2014. Iodine application increased ascorbic acid content and modified the vascular tissue of *Opuntia ficus-indica* L. Pak. J. Bot. 46, 127–134.

Payne, R.W., Murray, D.A., Harding, S.A., Baird, D.B., Soutar, D.M., 2016. GenStat for Windows, 18th Edition VSN International, Hemel Hempstead.

Saleem, K.N., Al-Faris, S.M., Al-Hinai, S.A., Al-Hinay, A.S., Al-Harthy, A.A., Al-Khamisis, S.A., Al-Bakri, A., 2016. Genetic diversity of 33 forage cactus pear accessions based principal component analysis. J. Prof. Assoc. Cactus Dev. 18, 78–86.

Shoop, M.C., Alford, E.J., Mayland, H.F., 1977. Plains prickly pear is a good forage for cattle. J. Range Manag. 30, 12–17.

Stintzing, F.C., Carle, R., 2005. Cactus stems (*Opuntia* spp.): A review on their chemistry, technology, and uses. Mol. Nutr. Food Res. 49, 175–194.

Teles, F., 1977. Nutrient Analysis of Prickly Pear (*Opuntia ficus indica*Linn). University of Arizona, Tucson, AZ.

Trivedi, S., Raval, P., 2017. A critical review on the wonders of cactus. J. Res. 1, 2456–7752.

Wilson, M.F., 2007. Medicinal Plant Fact Sheet: *Opuntia*: Prickly Pear Cactus. A collaboration of the IUCN Medicinal Plant Specialist Group, PCA-Medicinal Plant Working Group, and North American Pollinator Protection Campaign. Virginia, Arlington.
World Health Organisation, 2009. Global Prevalence of Vitamin A Deficiency in Populations at Risk 1995-2005. WHO Global Database on Vitamin A Deficiency. World Health Organisation, Geneva.
World Health Organisation, 2012. Guideline: Potassium Intake for Adults and Children. World Health Organization (WHO), Geneva.
Yahia, E.M., Mondragon-Jacobo, C., 2011. Nutritional components and anti-oxidant capacity of ten cultivars and lines of cactus pear fruit (*Opuntia* spp.). Food Res. Int. 44, 2311–2318.