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ORIGINAL ARTICLE

Chemical Composition and Antioxidant Activity of Seeds of Various Halophytic Grasses

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Abstract Increasing population has resulted in overexploitation of conventional seeds. The limited supply of water and salinization of agricultural lands are threats to crop production. This creates food insecurity and results in ever-increasing prices of crops and edible oils. Halophytes that produce high-quality seeds can serve as sources of oil and edible products. We analyzed the chemical composition and antioxidant activity of seeds from 5 halophytic grasses, *i.e.*, *Aeluropus lagopoides*, *Eragrostis ciliaris*, *Eragrostis pilosa*, *Panicum antidotale*, and *Sporobolus ioclados.* These seeds contained crude protein (10–29%), carbohydrates (32–55%), crude fiber (4–21%), minerals (3.8–9.2%), and oil (4–11%), indicating their nutritional potential. Oils of these seeds had suitable fatty-acid composition with 62–82% unsaturation and only 17–24% saturation. Out of this, 91–94% of the total oil constituted by linoleic, oleic, and palmitic acids. High contents of total phenols (2.8–4.2 mg gallic acid equivalent [GAE] g^{-1}), flavonoids (0.5–1.3 mg Quercetin equivalent [QE] g^{-1}), and tannins (0.3–1.3 mg catechin equivalent [CE] g^{-1}) supported their high antioxidant activity (1,1-Diphenyl-2-picryl-hydrazyl (DPPH) activity in terms of half maximal inhibitory concentration-IC_{50} 1.1–5.86 mg mL^{-1}; 2,2′-azino-bis3-ethylbenzothiazoline-6-sulphonic acid (ABTS) 18.8–72.8 mmol Trolox g^{-1}; ferric-reducing antioxidant power 2.0–4.4 mmol Fe^{+2} g^{-1}). The reverse phase-high-pressure liquid chromatography analysis identified the presence of bioactive phenolic antioxidants (mainly gallic acid, chlorogenic acid, coumaric acid, ferulic acid, kaempferol, and quercetin). Due to these characteristic composition and salt tolerability, these plants can serve as potential sources of industrial raw materials for food, edible oil, phytochemicals, and oliochemicals.

Keywords Edible oil · Fatty acid · Food · Nutritive value · Phenolic compounds · Salinity

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Introduction

The progressive gap between growing population and limited food supply is a threat to human survival. Currently, almost 900 million out of 7 billion people are suffering from chronic undernourishment, which is the largest figure in history (Dawson, Perryman, & Osborne, 2016; Gualtieri, 2013). In addition, particularly in developing countries, limited supply of fresh water and salinization of agricultural lands are threats to crop production (Sadik, 1991). To cope with this situation, scientists are exploring ways to utilize nonconventional plants (Marfo, Oke, & Afolabi, 1988). However, these efforts have extensively focused on leaves or fruits, while seeds are mostly unexplored. Seeds of halophytes, especially from the plants that occupy the spare coastal areas, are perhaps some of the viable options to supplement food crises. They can serve as a good dietary source of protein, carbohydrate, fiber, minerals, and vitamins, and seed oils have a high concentration of polyunsaturated fats (Abbasian, Asgarpanah, & Ziarati, 2015; Moser, Seliskar, & Gallagher, 2016).

Increasing awareness about the quality, nutritional composition, and health promotion are the other motives, besides increasing global demand for seeking potential in

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nonconventional plants. The basic criterion for selecting edible oils among new crops is the fatty-acid composition specifically the ratio of the saturated and unsaturated content (Bressani, Gonzales, Elias, & Melgar, 1987; Cherif, Ben-Miled, Marzouk, Smaoui, & Zarrouk, 1992). Oils with high saturation are less desirable as they increased the risk of blood clotting and associated heart problems (Chaudhry, Hameed, Ahamd, & Hussain, 2001), while unsaturated fats protect from heart diseases and stroke (Willett, 2012). Halophyte seeds provide a good yield of oil with suitable fatty-acid composition (Glenn, O'Leary, Watson, Thompson, & Kuehl, 1991; Weber et al., 2001). Seed oil of *Salicornia bigelovii* has an unsaturation level higher than conventional oils (Glenn et al., 1991). Weber et al. (2001) have highlighted the halophyte seed oils with 85–90% unsaturation among which highly salt-tolerant species *Suaeda moquinii* had the best oil quality. Seed oils from other halophytes including *Crithmum maritimum*, *Chenopodium glaucum*, *Descurainaia sophia*, *Nitraria sibiria*, *Suaeda salsa*, and *Zygophyllum album* are also suitable for nutritional, pharmaceutical, cosmetic, and biodiesel purposes (Yajun, Xiaojing, & Weiqiang, 2003; Zarrouk et al., 2003). Yajun et al. (2003) reported that salt-treated *D. sophia* produced seed oil with up to 53.7% linolenic acid (53.7%) compared to only 36% from the same plant grown under nonsaline conditions. Linolenic acid is an essential fatty acid that helps in blood circulation (by vasodilatation), alleviates gastric ulcer, and helps in managing asthma and other diseases (Ascherio & Willett, 1997). The quality of halophytic seed oils is not only comparable to conventional oils but also has several putative health benefits (DeClercq, Daun, & Tipples, 1998).

The scientific investigation of the biochemical and nutritional characteristics is necessary for the future utilization of halophytes as nonconventional crops. To bridge the knowledge gap, researchers from all over the world are exploring halophytes as new alternatives for sustainable industrial materials (Gupta, 2014). Such evaluations will be helpful to identify potential plants, which can grow on saline resources and produce biomass for multiple industrial uses. In this context, we analyzed the nutritive value, fatty-acid profile, mineral content, antioxidant capacity, and phenolic composition of seeds from five halophytic grasses, *i.e.*, *Aeluropus lagopoides*, *Eragrostis ciliaris*, *Eragrostis pilosa*, *Panicum antidotale*, and *Sporobolus ioclados.*

Materials and Methods

Seed Materials

Seeds of *A. lagopoides*, *E. ciliaris*, *E. pilosa*, *P. antidotale*, and *S. ioclados* were collected from coastal areas of Sindh

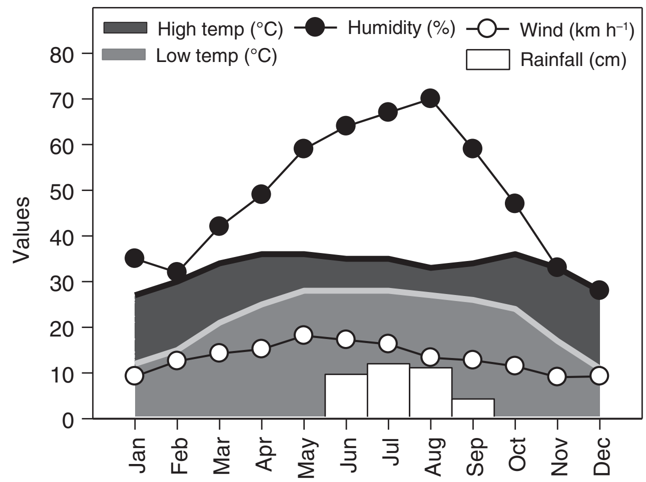

Fig. 1 Environmental data of study area comprising mean annual temperature (low and high), rainfall, wind velocity, and humidity (Pakistan Meteorological Department)

and Baluchistan provinces of Pakistan, during 2010. The study area represents an arid to semiarid climate with a high average temperature (~30 °C), and low rainfall (<250 mm) (Fig. 1). Seeds were manually separated, cleaned, and stored at 4 °C and were analyzed within 6 months after collection. Seed material was ground to fine powder using an oscillating mill (MM-400 Retsch GmbH, Haan, Germany) and grinding was done separately to avoid temperature rise.

Proximate Analysis

Weight of fresh seeds was measured and subsequently dry weight was recorded after air drying of the samples. Crude protein (by Kjeldahl apparatus), soluble sugars, oil (by Soxhlet apparatus), and fiber (by digestion method) contents were determined in ground samples using official methods of Association of Official Agricultural Chemists (AOAC) (Horwitz & Latimer, 2005). Total carbohydrates were analyzed using the Anthron reagent method (Yemm & Willis, 1954) and total energy was calculated by multiplying the amount of protein, carbohydrate, and fat by the factors of 4, 4, and 9 kcal, respectively, according to the European Economic Community (EEC) council directive on nutrition labeling for foodstuffs (European Economic Community (EEC), 1990). All determinations were performed in five replicates.

Ash and Mineral Contents

Ash and mineral contents were determined using the standard methods of AOAC (Horwitz & Latimer, 2005). To remove carbon, 0.5 g of the ground seed samples were ignited and incinerated in the muffle furnace at 550 °C for 12 hours. Ash was dissolved in HNO_3 and mineral constituents (Na^+, K^+, Ca^{2+}, and Mg^{2+}) were determined directly from the extract or diluted samples using an atomic absorption spectrometer (AA-700; PerkinElmer Inc., Shelton, CT, USA). All determinations were performed in five replicates.

Fatty-Acid Analyses using Gas Chromatography

Oil extraction, derivatization, and fatty-acid profiling were performed using standard methods of AOAC (Horwitz & Latimer, 2005) with little modifications. Briefly, ground seed material was extracted in hexane using a Soxhlet apparatus (Extraction System B-811; Buchi, Labortechnik, Flawil, Switzerland) at continuous mode for 8 hours. After extraction, the remaining solvent was evaporated and fatty-acid composition of the seed oil samples was analyzed using gas chromatography–flame-ionization detector (GC–FID) after derivatization to fatty-acid methyl esters (FAME) using sulfuric acid (1%) in methanol. A GC instrument (Agilent model, 7890A; H.P. Co., Amsterdam, The Netherlands) equipped with a DB-WAX capillary column (10 m × 0.100 mm × 0.1 μm film thickness microbore) coupled with FID and mass spectrometer (MS) (Agilent model, 5975C) was used. The column was heated from 50 to 220 °C at a rate of 5 °C min^{-1} with an initial and a final hold time of 4 and 12 min, respectively. Injector and detector temperatures were maintained at 220 and 250 °C, respectively. Helium was used as a carrier gas at a flow rate of 0.5 mL min^{-1} with a split injector (split ratio 1:50) and nitrogen was used as a makeup gas. Identification and quantification of FAME were done on GC–FID by comparing the retention times and the peak spectra with those of reference standards (Product #47885-U) purchased from Supelco, Sigma-Aldrich, Saint-Quentin Fallavier, France. The results were further confirmed by GC–MS using NIST library and expressed as percentage of individual fatty acid in an extracted portion. All determinations were performed in triplicate.

Antioxidant Capacity

Ground seed material was extracted in 80% methanol and the supernatant was separated and used quickly for antioxidant analyses. Antioxidant capacity of the seed samples was determined using 1,1-Diphenyl-2-picryl-hydrazyl (DPPH) (Brand-Williams, Cuvelier, & Berset, 1995) and 2,2′-azino-bis3-ethylbenzothiazoline-6-sulphonic acid (ABTS) (Re et al., 1999) radical-scavenging tests. The ferric-reducing antioxidant power (FRAP) assay was also carried out to determine the reducing potential of the seed samples (Benzie & Strain, 1996). All determinations were performed in five replicates.

Total Phenols, Flavonoids, and Tannins

Methanol (80%) extracts were also used to determine the total phenol content (TPC) and total flavonoid content (TFC); however, for total tannins content (TTC), seed material was extracted in acidified water. TPC was determined using the Folin–Ciocalteu colorimetric method (Singleton & Rossi, 1965). Colorimetric methods were also used to quantify TFC (Chang, Yang, Wen, & Chern, 2002) and TTC (Pearson, 1976). All determinations were performed in five replicates.

Phenolic Profiling using High-Pressure Liquid Chromatography

Ground defatted materials of the halophyte seeds were extracted according to the method described by Proestos, Sereli, and Komaitis (2006). Briefly, 0.25 g seed material was extracted in 62.5% methanol (20 mL). In each extract, nitrogen was bubbled after adding 6 M HCl (3 mL). Extracts were then sonicated and refluxed in a boiling water bath for 2 hours to make up to 50 mL with methanol. Samples were double filtered using a 0.45 μm filter (Micropore, San Diego, CA, USA) and analyzed using a high-pressure liquid chromatography (HPLC) system (LC-20AT, Shimadzu, Tokyo, Japan) equipped with an analytical column, Nucleosil C18, 5 μm 100 Å (250 × 4.60 mm, Phenomenex [Torrance, CA, USA]) coupled with a precolumn (KJO-4282; Phenomenex), an autosampler (SIL-20A, Shimadzu, Tokyo, Japan), a column oven (CTO-20A, Shimadzu, Tokyo, Japan), a diode array detector (SPD-M20A, Shimadzu, Tokyo, Japan), and lab solution software. The mobile phase composed of (A) sodium phosphate buffer (50 mM; pH 3.3) in methanol (10%) and (B) 70% methanol. The gradient program by Qasim et al. (2017) was used with a flow rate of 1 mL min^{-1}. Quantification of the phenolic compounds was performed in triplicates against the reference standards purchased from Sigma–Aldrich, Saint Quentin, Fallavier, France.

Results and Discussion

Halophyte seeds could be used for multifarious purposes such as food, fodder, medicine, and seed oil. In this study, seeds of five halophytic grasses, traditionally used for edible, medicinal, and fodder purposes (Qasim et al., 2017; Qasim, Gulzar, Shinwari, Aziz, & Khan, 2010) were analyzed to reveal their potential as industrial feedstock. Seeds

Table 1 Taxonomic information, seed characters, and salinity tolerance ranges of five halophytic grasses

Species	Flowering period	Seed availability	Salinity tolerance (mole NaCl)	Seed weight[a]	Seed color[b]	
					Color	RGB value
Perennial						
Aeluropus lagopoides	January	April–May	0.5–1.0	11.45 (1.16)	Beige brown	121-085-061
Panicum antidotale	April–June	August–October	0.3–0.4	15.64 (0.13)	Ivory	225-204-079
Sporobolus ioclados	July–August	February–April	0.5–6.0	12.86 (0.91)	Olive brown	111-079-040
Annual						
Eragrostis ciliaris	May–July	September–October	0.15–2.0	2.37 (0.3)	Pearl ruby red	114-020-034
Eragrostis pilosa	July–August	September–October	0.15–2.0	38.85 (0.68)	Pearl copper	118-060-040

[a] mg/100 seeds, mean (±SD) values are the results of five determinations.

[b] Source: http://www.ralcolor.com

were collected from the coastal areas of Sindh and Baluchistan. Most of the grasses are perennial and highly salt-tolerant (0.3–1.0 M NaCl), and can produce enormous amounts of seeds (Table 1).

Proximate Analyses

The nutritive composition of five halophytic grass seeds is shown in Table 2. Seeds had a preferred range of moisture content (4–6%), ash (4–9%), dry weight (94–96%), and organic matter (86–92%). Seeds had a low moisture content, ranging from 3.7% (*P. antidotale*) to 5.7% (*E. pilosa*), which is important for maintaining the quality and shelf life. The lower moisture content decreases the onset of microbial growth, premature seed germination, unwarranted fermentation, and many other undesirable biochemical changes. The moisture content of halophytic seeds is compatible to sesame (5–6%) and lower than cereal grains (11–13%), beans (9–11%), and lentils (8–9%) (Hefnawy, 2011; Koehler & Wieser, 2013; Nzikou et al., 2009). A higher amount of dry matter (94–96%) and organic fraction (86–92%) in these seeds indicated a good food reserve and more energy compared to conventional grains such as corn, oat, rice, and sorghum (Onyeike & Acheru, 2002).

A wide variation in the contents of crude protein (10–29%) was observed in halophytic seeds. Crude protein was higher in *P. antidotale* (29%) followed by *A. lagopoides* (22%), *E. pilosa* (21%), and *S. ioclados* (20%), while the *E. ciliaris* showed lowest value (11%). These are favorable protein values reported for some commercial beans, chickpeas, lentils, legumes, cereals, and oilseeds (Hefnawy, 2011; Koehler & Wieser, 2013; McKevith, 2009; Wang, Hatcher, Tyler, Toews, & Gawalko, 2010). Soluble sugars ranged between 6% (*A. lagopoides*) and 29% (*S. ioclados*), which are similar to or even higher than common edible peas, beans, legumes, lentils, and oilseeds (Blair & Reichert,

Table 2 Nutritive analyses of seeds of five halophytic grasses

Parameters	*A. lagopoides*	*E. ciliaris*	*E. pilosa*	*P. antidotale*	*S. ioclados*
Moisture (%)	4.3 (1.64)	4.9 (0.04)	5.7 (0.02)	3.7 (0.05)	4.0 (0.06)
Ash (%)	7.0 (0.05)	9.2 (1.88)	6.2 (0.50)	6.4 (0.49)	3.8 (0.07)
Na^+ (%)	1.8 (0.13)	0.1 (0.15)	0.2 (0.06)	0.2 (0.04)	0.7 (0.08)
K^+ (%)	2.3 (0.10)	3.1 (0.18)	1.2 (0.14)	1.4 (0.12)	1.1 (0.11)
Ca^{++} (%)	1.3 (0.11)	2.1 (0.16)	0.9 (0.11)	1.1 (0.15)	0.8 (0.14)
Mg^{++} (%)	0.9 (0.08)	1.2 (0.12)	1.6 (0.06)	0.5 (0.03)	0.9 (0.04)
Dry matter (%)	95.7 (2.34)	95.1 (3.12)	94.3 (5.63)	96.3 (3.46)	96.0 (5.73)
Organic matter (%)	88.6 (0.05)	85.9 (1.88)	88.1 (0.50)	89.9 (0.49)	92.2 (0.07)
Crude protein (%)	22.4 (1.16)	10.7 (0.30)	21.0 (0.68)	29.0 (0.13)	19.7 (0.91)
Oil (%)	4.1 (0.13)	11.0 (0.09)	8.1 (0.08)	9.0 (0.06)	7.6 (0.09)
Crude fiber (%)	7.9 (1.63)	13.0 (0.08)	4.1 (0.26)	20.9 (0.50)	16.9 (0.98)
Soluble sugars (%)	6.2 (0.01)	9.5 (0.94)	12.1 (0.14)	8.0 (0.31)	29.0 (0.49)
Carbohydrates (%)	54.2 (2.11)	51.9 (1.96)	54.9 (3.41)	32.2 (1.14)	47.9 (2.35)
Energy (kcal 100 g^{-1})	343.3 (11.3)	346.9 (15.5)	376.3 (15.1)	323.2 (18.4)	338.7 (10.1)

Mean (±standard error) values are the results of five determinations.

Table 3 Fatty-acid composition of seed oil from five halophytic grasses

Fatty-acid composition (%)	*A. lagopoides*	*E. ciliaris*	*E. pilosa*	*P. antidotale*	*Sporobolus ioclados*
Palmitic acid (16:0)	15.4 (0.53)	20.1 (0.64)	20.3 (0.94)	13.7 (0.56)	18.5 (0.84)
Palmitoleic acid (16:1)	0.7 (0.01)	nd	nd	nd	nd
Stearic acid (18:0)	2.8 (0.08)	2.8 (0.05)	2.8 (0.02)	2.5 (0.01)	2.3 (0.01)
Oleic acid (18:1)	47.2 (1.54)	23.8 (0.98)	26.6 (1.13)	26.6 (0.94)	31.5 (0.78)
Linoleic acid (18:2)	30.4 (1.13)	49.9 (2.14)	44.1 (1.87)	53.2 (3.41)	29.8 (1.18)
Linolenic acid (18:3)	1.1 (0.03)	1.4 (0.02)	3.3 (0.04)	2.0 (0.01)	0.8 (0.03)
Arachidic acid (20:0)	0.8 (0.02)	0.7 (0.01)	1.2 (0.01)	0.8 (0.02)	0.9 (0.01)
Gondoic acid (20:1)	1.6 (0.03)	nd	0.4 (0.01)	nd	0.4 (0.01)
Erucic acid (22:1)	nd	nd	0.5 (0.01)	nd	0.9 (0.02)
Docosadienoic acid (22:2)	nd	nd	nd	nd	2.4 (0.01)
Others	0	1.2 (0.01)	0.7 (0.02)	2.4 (0.01)	14.1 (0.35)
SFA	19.0	23.7	24.3	16.9	21.7
USFA	81.0	75.1	74.5	81.8	62.4
MUFA	49.5	23.8	27.0	26.6	31.9
PUFA	31.5	51.3	47.4	55.2	30.6
USFA/SFA	4.3	3.2	3.1	4.8	2.9
PUFA/SFA	1.6	2.2	1.9	3.3	1.4
MUFA/SFA	2.6	1.0	1.1	1.6	1.5
Ω6/Ω3	28.7	36.2	13.7	26.7	36.8

Mean (±standard error) values are the results of triplicate determinations. nd, not detected.

1984; Vidal-Valverde, Frias, & Valverde, 1993), whereas total carbohydrates varied from 32% (*P. antidotale*) to 55% (*E. pilosa*). Proximate composition of studied grass seeds was comparable to the chemical and nutritional composition of several other species such as *Canava liaensiformis*, *Canava gladiata*, and *Canava maritime* (Bressani et al., 1987).

The oil content of the studied grass seeds ranged from 8% to 11%, except *A. lagopoides* (4%). The seed oil content of most of the species compared well with other halophytes, *e.g.*, 8% oil in *N. sibiria* (Yajun, 2003). Differences in plant species, cultivation time, climate, ripening stage, harvesting stage, and/or extraction method could cause variations in oil yields. Considerable genetic variability exists among halophytic genotypes, which may lead them to perform better under suboptimal conditions (Llanes, Bonercarrere, Capdevielle, Vidal, & Luna, 2011).

The crude fiber content was considered a useful measure for estimating the energy values of edible plants. A fiber-rich diet is usually larger in volume and rich in micronutrients, which takes time to digest and brings a feel of satiety (Pereira & Ludwig, 2001). A daily intake of 20–35 g of dietary fiber is suggested for healthy adults (Dhingra, Michael, Rajput, & Patil, 2012). Considering the dietary importance, a large market of fiber-rich products has been developing in recent years and to search for new and safe sources of dietary fiber is becoming a focus of the scientific community (Chau & Huang, 2003). In general, most of the studied seeds contained a high fiber content (8–21%), except *E. pilosa* (4%). These values are in line with crude fiber of most of the cereals/grains (0.7–17%), legumes/pulses (2–18%), vegetables (0.6–17%), fruits (0.5–3%), and nuts/seeds (6–22%) (Dhingra et al., 2012).

Mineral Content

Halophytic seeds could have a total mineral content as high as 35% (Khan & Unger, 1996). In this study, the seed ash content (Table 2) ranged from 4% (*S. ioclados*) to 9% (*E. ciliaris*). Generally, halophytes accumulated a higher amount of ash (Chau & Huang, 2003) and their seeds could be used as a mineral source. Many essential minerals including potassium (K^+), calcium (Ca^{++}), sodium (Na^+), and magnesium (Mg^{++}) were found in the studied seeds (Table 2). K^+ and Ca^{++} were higher in *E. ciliaris* (3.1% and 2.1%, respectively), while higher amounts of Na^+ and Mg^{++} were detected in *A. lagopoides* (1.8%) and *E. pilosa* (1.61%), respectively. The availability of essential minerals in edible plants is important, especially in those areas where the incidence of mineral deficiency among the local population is high (Barrett, 2010). The considerable amount of essential minerals in halophyte seeds presents a rich source of minerals for edible as well as cattle-feed purposes, because pressed cake left behind oil extraction that could be used for animals.

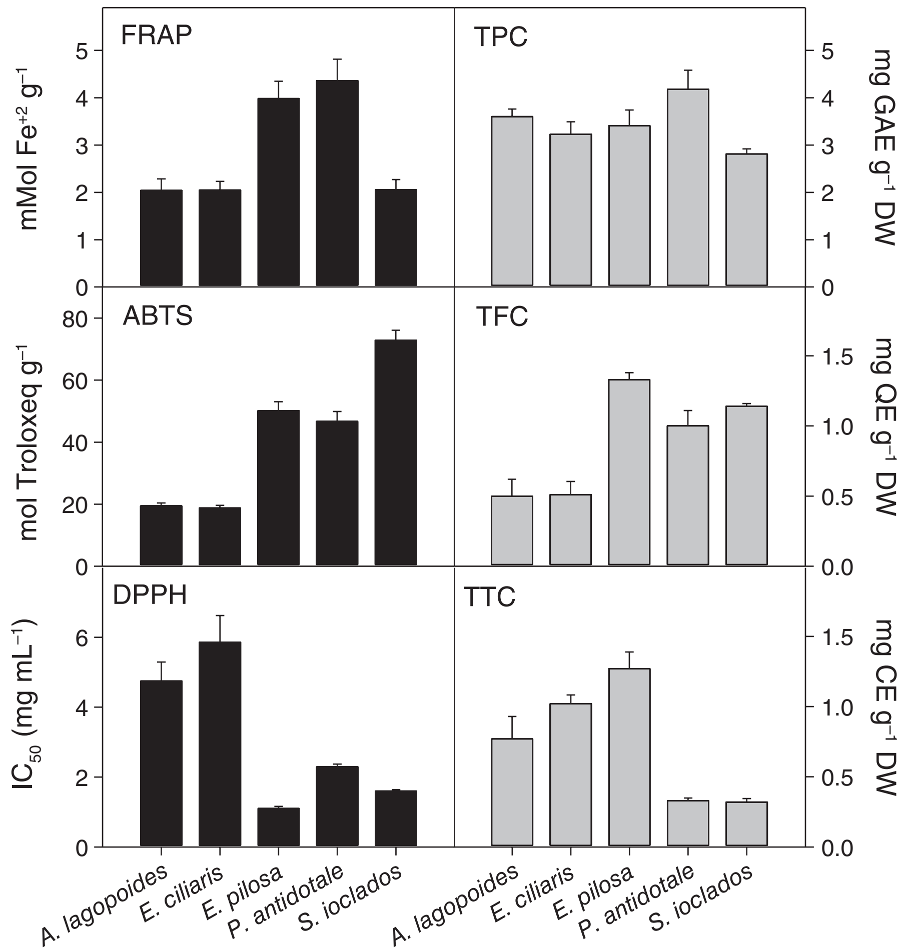

Fig. 2 Seed antioxidant capacity (DPPH, ABTS, and FRAP), total phenols (TPC), flavonoids (TFC), and tannins (TTC) of seeds of five halophytic grasses. All determinations were performed in five replicates

Fatty-Acid Composition

Fatty-acid composition of the studied halophytic seeds is presented in Table 3. Ten different fatty acids were identified. The predominant fatty acids were linoleic, oleic, and palmitic acids, which constituted 91–94% of the total oil (except *S. ioclados*, 79%). Linoleic acid is an indispensable and essential polyunsaturated fatty acid (PUFA) (Bruckert, 2001). It is the predominant fatty acid of *P. antidotale* (53%), *E. ciliaris* (50%), *E. pilosa* (44%), and *S. ioclados* (30%) seed oils; however, *A. lagopoides* oil contained a higher amount of oleic acid (47%). Oleic acid was the second dominant fatty acid of seeds of halophytic grasses (24–32%). The amount of linoleic acid in these species is comparable with edible oil crops like sunflower (Karleskind & Wolff, 1996), corn, and cotton (Smaoui & Cherif, 1992). The presence of oleic acid determined the oil quality and its high content is usually favored because it helps in constructing the nervous system, lowering blood cholesterol, and decreasing the risk of heart diseases (Mensink & Katan, 1989). Palmitic acid ranged from 14% (*P. antidotale*) to 20% (*E. pilosa* and *E. ciliaris*). Palmitoleic and docosadienoic acids were only detected in *A. lagopoides* (0.7%) and *S. ioclados* (2.45%), whereas gondoic acid was determined in *A. lagopoides* (1.57%), *E. pilosa* (0.42%), and *S. ioclados* (0.39%). A high level (>2%) of erucic acid (22:1) is considered unhealthy (Food Standards Australia New Zealand, 2003). Seed oils of the tested species were either free from any such undesirable fractions or they were found in traces (0.55% in *E. pilosa* and 0.93% in *S. ioclados*). In total, the fatty-acid composition of studied seed oils was as good as other commercial oils like sunflower (Karleskind & Wolff, 1996) corn, cotton (Smaoui & Cherif, 1992), canola (DeClercq et al., 1998), and olive (Zarrouk et al., 2003).

The degree of unsaturation is another important aspect for the determination of oil quality. Oil with high levels of unsaturation is considered good for human health (Williams,

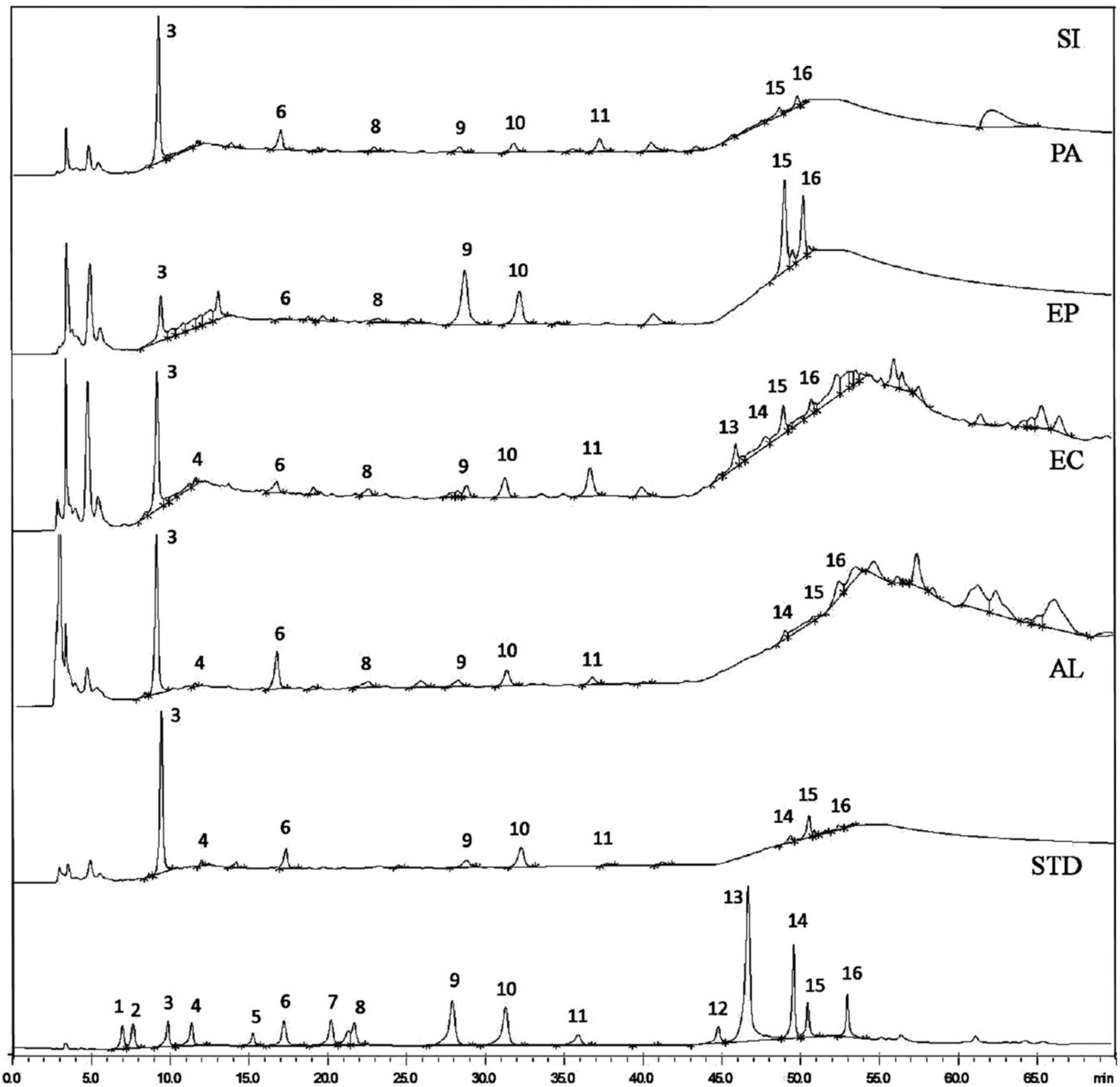

Fig. 3 RP-HPLC chromatograms showing phenolic metabolites of standard compounds (STD) and seed extracts of *Aeluropus lagopoides* (AL), *Eragrostis ciliaris* (EC), *Eragrostis pilosa* (EP), *Panicum antidotale* (PA) and *Sporobolus ioclados* (SI). Phenolic compounds includes 1) pyrogallol 2) hydroquinone 3) gallic acid 4) resorcinol 5) vanillic acid 6) chlorogenic acid 7) syringic acid 8) caffeic acid 9) coumaric acid 10) ferulic acid 11) coumarin 12) naringenin 13) transcinnamic acid 14) rutin 15) quercetin and 16) kaempferol. All determinations were performed in triplicates

2000). Fatty acids identified in this study were composed of approximately 17–24% saturated fatty acid (SFA), 62–82% unsaturated fatty acid (USFA), 24–50% monounsaturated fatty acid (MUFA), and 30–55% PUFA (Table 3). A higher percent of USFA in the studied seed oils is in accordance with other halophyte seeds, *i.e.*, *Atriplex rosea*, *Atriplex heterosperma*, *Allenrolfea occidentialis*, *Suaeda fruticosa*, *Kochia scoparia*, *Halogeton glomeratus*, *Sarcobatus vermiculatus*, and *Kosteletzkya virginica* (78–89% USFA) (Weber et al., 2001). The PUFA found in studied oils were dominated by Omega-6 (ω-6) and Omega-3 (ω-3). Grass seed oils contained higher PUFA than MUFA and the predominant fatty acids were also from PUFA (Table 3). Higher ratios of USFA/SFA and PUFA/SFA also show the suitability of these oils for edible purpose with health-promoting effects (Binkowski, Joachimiak, & Liang, 2005).

Table 4 Phenol profiling (mg g^{-1}) of seeds of five halophytic grasses

Phenolic compounds	Retention time	*Aeluropus lagapoides*	*E. ciliaris*	*E. pilosa*	*P. antidotale*	*S. ioclados*
Gallic acid	9.6	1.43 (0.021)	0.81 (0.013)	1.10 (0.011)	0.19 (0.011)	0.04 (0.001)
Resorcinol	11.2	0.06 (0.013)	0.04 (0.011)	0.04 (0.002)	nd	nd
Chlorogenic acid	17.1	0.27 (0.014)	0.53 (0.021)	0.10 (0.008)	0.22 (0.015)	0.24 (0.008)
Caffeic acid	21.5	nd	0.20 (0.011)	0.08 (0.001)	0.04 (0.001)	0.05 (0.001)
p-Coumaric acid	21.8	0.07 (0.012)	0.02 (0.012)	0.30 (0.004)	0.69 (0.003)	0.06 (0.001)
Ferulic acid	27.9	0.19 (0.017)	0.06 (0.012)	0.45 (0.006)	0.52 (0.002)	0.03 (0.001)
Coumarin	31.1	nd	0.14 (0.011)	0.28 (0.004)	nd	0.07 (0.001)
Transcinnamic acid	46.5	nd	nd	0.09 (0.001)	nd	nd
Rutin	49.6	0.01 (0.001)	0.11 (0.010)	0.02 (0.001)	nd	nd
Quercetin	50.5	0.03 (0.001)	0.01 (0.001)	0.05 (0.001)	0.19 (0.001)	0.02 (0.001)
Kaempferol	52.9	0.03 (0.001)	0.28 (0.023)	0.31 (0.004)	0.76 (0.007)	0.01 (0.001)
Total		2.09	2.21	2.83	2.62	0.52

Mean (±standard error) values are the results of triplicate determinations. nd, not detected.

Antioxidant Activity

Oilseeds also contain antioxidants but attracted less attention because they are less often directly consumed, except nuts. However, antioxidant activity and natural antioxidants have been reported in the seeds of canola (Naczk, Amarowicz, Sullivan, & Shahidi, 1998) and sunflower (Kubicka, Jędrychowski, & Amarowicz, 1999). Studies on antioxidant activity of halophyte seeds are not well documented. Therefore, this study was extended to analyze the antioxidant activity of the seed samples using most common and reliable methods based on radical scavenging (DPPH and ABTS) and reducing power (FRAP) abilities. Most of the seeds showed considerable antioxidant activity (Fig. 2). The highest DPPH activity was found in *E. pilosa* (1.1 IC_{50} mg mL^{-1}) followed by *S. ioclados* (1.6 IC_{50} mg mL^{-1}) and *P. antidotale* (2.3 IC_{50} mg mL^{-1}). The ABTS values were higher in *S. ioclados* (72.8 mmol Trolox g^{-1}), *E. pilosa* (50.1 mmol Trolox g^{-1}), and *P. antidotale* (46.7 mmol Trolox g^{-1}). In the case of FRAP, *P. antidotale* (4.4 mmol Fe^{+2} g^{-1}) and *E. pilosa* (4.0 mmol Fe^{+2} g^{-1}) both showed a higher activity. These results suggest that halophyte seeds can also be served as dietary sources of antioxidants and may be used as natural food additives to improve the quality, stability, and safety of the edible products.

Total Phenols, Flavonoids, and Tannins

Seed phenolics inhibit the growth of pathogenic bacteria and help in the oxidative stability, improve the shelf life, and maintain the quality of seed and seed oil for longer periods (Baldioli, Servili, Perretti, & Montedoro, 1996). Halophytic grasses contained phenols (2.8–4.2 mg gallic acid equivalent [GAE] g^{-1}), flavonoids (0.5–1.3 mg QE g^{-1}), and tannins (0.3–1.3 mg catechin equivalent [CE] g^{-1}) (Fig. 2). Total phenols and flavonoids of the tested samples were comparable to wheat (TPC 4.2 mg g^{-1} and TFC 1.5 mg g^{-1}) and higher than rice (TPC 0.99 mg g^{-1} and TFC 0.17 mg g^{-1}) and sorghum (TPC 0.76 mg g^{-1} and TFC 0.3 mg g^{-1}) (Chan, Khong, Iqbal, Mansor, & Ismail, 2013). In addition, seeds that showed a higher antioxidant activity also had higher polyphenolic contents (Fig. 2). Seeds of *P. antidotale* and *E. pilosa* contained high phenols (4.2 and 3.4 mg GAE g^{-1}), flavonoids (1.0 and 1.3 mg QE g^{-1}), and tannins (0.3 and 1.3 mg CE g^{-1}), respectively. From these results, it can be predicted that phenolic compounds are the major antioxidants in these seeds, as reported for other plants (Abideen et al., 2015; Ahmed, Fatima, Qasim, Gul, & Haq, 2017; Qasim, Aziz, Rasheed, Gul, & Khan, 2016; Zahra et al., 2017).

Phenolic Profile using HPLC

HPLC analyses showed the presence of 11 phenolic compounds (gallic acid, resorcinol, chlorogenic acid, caffeic acid, coumaric acid, ferulic acid, coumarin, transcinnamic acid, rutin, quercetin hydrate, and kaempferol) in all seed samples (Fig. 3). The dominant phenolic compounds were gallic, chlorogenic, coumaric, and ferulic acids, while kaempferol and quercetin were major flavonoids (Table 4). Several studies showed the bioactive properties of these compounds such as gallic acid, which is known for antimalarial, antimicrobial, antitumor, antiinflammatory, and neuroprotective properties (Choubey, Varughese, Kumar, & Beniwal, 2015). Chlorogenic acid exhibits antiviral, anticancer, and hepatoprotective effects (Niggeweg, Michael, & Martin, 2004). Coumaric acid bears antioxidant, antiinflammatory, anticancer, antigenotoxic, and antiviral activities (Ferguson, Zhu, & Harris, 2005). Besides strong antioxidant activity, ferulic acid is also good against certain types of cancers, microbial infections, and skin diseases (Kampa et al., 2003). Some flavonoids detected in this study have several biological activities. Quercetin is used to treat hardening of arteries, cardiovascular problems, diabetes, cataracts, peptic ulcer, asthma, and prostate infections (Hirpara, Aggarwal, Mukherjee, Joshi, & Burman, 2009). Kaempferol serves as a strong antimicrobial, antitumor, anti-inflammatory, antioxidant, and anticancer agent (Calderón-Montaño, Burgos-Morón, Pérez-Guerrero, & López-Lázaro, 2011). In addition, most of these compounds and their derivatives are also used as food additives. In this capacity, these compounds help to improve the shelf life by protecting essential nutrients from oxidation and microbial deterioration (Sanches-Silva et al., 2014).

Conclusions

Seeds of five halophytic grasses were analyzed for their nutritional composition, fatty-acid profile, essential minerals, phenolic compounds, and antioxidant activity. These oil seeds presented suitable edible characteristics and fatty-acid composition, which are comparable to or even better than most of the conventional oil seeds. These seeds also have a considerable amount of bioactive compounds, like phenolic metabolites, with a high antioxidant activity. Due to the distinctive chemical composition and the ability to withstand salt stress, these plants can serve as potential sources of industrial raw materials for food, edible oils, phytochemicals, and oliochemicals. Furthermore, these plants can be grown on saline lands, which have no agronomic relevance and can produce biomass of commercial and economic importance. In the line of the suggested results, further investigations are needed to convert scientific knowledge into economically feasible outcome.

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Conflict of Interest The authors declare that they have no conflicts of interest.

References

Abbasian, K., Asgarpanah, J., & Ziarati, P. (2015) Chemical composition profile of *Acacia nilotica* seed growing wild in south of Iran. *Oriental Journal of Chemistry*, **31**:1027–1033.

Abideen, Z., Qasim, M., Rasheed, A., Yousuf Adnan, M., Gul, B., & Ajmal Khan, M. (2015) Antioxidant activity and polyphenolic content of *Phragmites karka* under saline conditions. *Pakistan Journal of Botany*, **47**:813–818.

Ahmed, M., Fatima, H., Qasim, M., Gul, B., & Haq, I. U. (2017) Polarity directed optimization of phytochemical and in vitro biological potential of an indigenous folklore: *Quercus dilatata* Lindl. ex Royle. *BMC Complementary and Alternative Medicine*, **17**: 386–401.

Ascherio, A., & Willett, W. C. (1997) Health effects of trans fatty acids. *The American Journal of Clinical Nutrition*, **66**: 1006S–1010S.

Baldioli, M., Servili, M., Perretti, G., & Montedoro, G. (1996) Antioxidant activity of tocopherols and phenolic compounds of virgin olive oil. *Journal of the American Oil Chemists' Society*, **73**: 1589–1593.

Barrett, C. B. (2010) Measuring food insecurity. *Science*, **327**: 825–828.

Benzie, F. F., & Strain, J. J. (1996) The ferric reducing ability of plasma (FRAP) as a measure of "antioxidant power": The FRAP assay. *Analytical Biochemistry*, **239**:70–76.

Binkowski, T. A., Joachimiak, A., & Liang, J. (2005) Protein surface analysis for function annotation in high-throughput structural genomics pipeline. *Protein Science*, **14**:2972–2981.

Blair, R., & Reichert, R. D. (1984) Carbohydrate and phenolic constituents in a comprehensive range of rapeseed and canola fractions: Nutritional significance for animals. *Journal of the Science of Food and Agriculture*, **35**:29–35.

Brand-Williams, W., Cuvelier, M. E., & Berset, C. (1995) Use of a free radical method to evaluate antioxidant activity. *LWT—Food Science and Technology*, **28**:25–30.

Bressani, R., Gonzales, J. M., Elias, L. G., & Melgar, M. (1987) Effect of fertilizer application on the yield, protein and fat content, and protein quality of raw and cooked grain of three amaranth species. *Plant Foods for Human Nutrition*, **37**:59–67.

Bruckert, E. (2001) Functionality of lipids in the context of a diet-health phytosterols, square relationship in the management of hyperlipidemic patients. *Oilseeds and Fats, Crops and Lipids*, **8**: 312–316.

Calderón-Montaño, J. M., Burgos-Morón, E., Pérez-Guerrero, C., & López-Lázaro, M. A. (2011) Review on the dietary flavonoid kaempferol. *Mini-Reviews in Medicinal Chemistry*, **11**:298–344.

Chan, K. W., Khong, N. M., Iqbal, S., Mansor, S. M., & Ismail, M. (2013) Defatted kenaf seed meal (DKSM): Prospective edible flour from agricultural waste with high antioxidant activity. *LWT—Food Science and Technology*, **53**:308–313.

Chang, C. C., Yang, M. H., Wen, H. M., & Chern, J. C. (2002) Estimation of total flavonoid content in propolis by two complementary colorimetric methods. *Journal of Food and Drug Analysis*, **10**: 178–182.

Chau, C. F., & Huang, Y. L. (2003) Comparison of the chemical composition and physicochemical properties of different fibers prepared from the peel of *Citrus sinensis* L. Cv. Liucheng. *Journal of Agricultural and Food Chemistry*, **51**:2615–2618.

Chaudhry, A. A., Hameed, M., Ahamd, R., & Hussain, A. (2001) Phyto-sociological studies in Chhumbisurla wildlife sanctuary, Chakwal, Pakistan. I. Species diversity. *International Journal of Agriculture and Biology*, **3**:363–368.

Cherif, A., Ben-Miled, D., Marzouk, B., Smaoui, A., & Zarrouk, M. (1992) *Metabolism, structure and utilization of plant lipids*. Tunis: CNP.

Choubey, S., Varughese, L. R., Kumar, V., & Beniwal, V. (2015) Medicinal importance of gallic acid and its ester derivatives: A patent review. *Pharmaceutical Patent Analyst*, **4**:305–315.

Dawson, T. P., Perryman, A. H., & Osborne, T. M. (2016) Modelling impacts of climate change on global food security. *Climatic Change*, **134**:429–440.

DeClercq, D. R., Daun, J. K., & Tipples, K. (1998) *Quality of western Canadian canola*. Winnipeg, Canada: Grain Research Laboratory, Canadian Grain Commission.

Dhingra, D., Michael, M., Rajput, H., & Patil, R. T. (2012) Dietary fibre in foods: A review. *Journal of Food Science and Technology*, **49**:255–266.

European Economic Community (EEC) (1990) Council directive on nutrition labeling for foodstuffs (90/496). *Official Journal of European Communities*, **L276**:40–44.

Ferguson, L. R., Zhu, S. T., & Harris, P. J. (2005) Antioxidant and antigenotoxic effects of plant cell wall hydroxycinnamic acids in cultured HT-29 cells. *Molecular Nutrition & Food Research*, **49**: 585–593.

Food Standards Australia New Zealand. (2003) *Erucic acid in food: A toxicological review and risk assessment.* Technical Report Series No. 21. Retrieved from http://www.foodstandards.gov.au

Glenn, E. P., O'Leary, J. W., Watson, M. C., Thompson, T. L., & Kuehl, R. O. (1991) *Salicornia bigelovii* Torr.: An oilseed halophyte for seawater irrigation. *Science*, **251**:1065–1067.

Gualtieri, D. (2013) Right to food, food security and food aid under international law, or the limits of a right-based approach. *Future of Food: Journal on Food, Agriculture and Society*, **1**:18–28.

Gupta, V. K. (Ed.) (2014) *Natural products: Research review* (Vol. **2**). New Delhi: Daya Publishing House.

Hefnawy, T. H. (2011) Effect of processing methods on nutritional composition and anti-nutritional factors in lentils (*Lens culinaris*). *Annals of Agricultural Science*, **56**:57–61.

Hirpara, K. V., Aggarwal, P., Mukherjee, A. J., Joshi, N., & Burman, A. C. (2009) Quercetin and its derivatives: Synthesis, pharmacological uses with special emphasis on anti-tumor properties and prodrug with enhanced bio-availability. *Anti-Cancer Agents in Medicinal Chemistry*, **9**:138–161.

Horwitz, W., & Latimer, G. W. (2005) *Official methods of analysis* (18th ed.). Gaitherburg: AOAC International.

Kampa, M., Alexaki, V. I., Notas, G., Nifli, A. P., Nistikaki, A., Hatzoglou, A., … Gravanis, A. (2003) Antiproliferative and apoptotic effects of selective phenolic acids on T47D human breast cancer cells: Potential mechanisms of action. *Breast Cancer Research*, **6**:63–74.

Karleskind, A., & Wolff, J. P. (1996) *Oils and fats manual: A comprehensive treatise: Properties, production, applications*. Paris: Tec & Doc-Lavoisier.

Khan, M. A., & Unger, I. A. (1996) Influence of salinity and temperature on the germination of *Haloxylon recurvum* Bunge ex. Boiss. *Annals of Botany*, **78**:547–551.

Koehler, P., & Wieser, H. (2013) Chemistry of cereal grains. In M. Gobbetti & M. Ganzle (Eds.), *Handbook on sourdough biotechnology* (pp. 11–45). New York, NY: Springer.

Kubicka, E., Jędrychowski, L., & Amarowicz, R. (1999) Effect of phenolic compounds extracted from sunflower seeds on native lipoxygenase activity. *Grasas y aceites (Fats and Oils)*, **50**: 127–130.

Llanes, A., Bonercarrere, V., Capdevielle, F., Vidal, S., & Luna, V. (2011) Genetic diversity in a natural population of the halophytic legume *Prosopis strombulifera* revealed by AFLP fingerprinting. *Boletín de la Sociedad Argentina de Botánica*, **46**:305–312.

Marfo, E. K., Oke, O. L., & Afolabi, O. A. (1988) Nutritional evaluation of pawpaw (*Carica papaya*) and flamboyant (*Denolix regia*) seed oils. *Nutrition Reports International*, **37**:303–310.

McKevith, B. (2009) Nutritional aspects of oilseeds. *Nutrition Bulletin*, **30**:13–26.

Mensink, R. P., & Katan, M. B. (1989) Effect of a diet enriched with monounsaturated or polyunsaturated fatty acids on levels of low-density and high-density lipoprotein cholesterol in healthy women and men. *New England Journal of Medicine*, **321**:436–441.

Moser, B. R., Seliskar, D. M., & Gallagher, J. L. (2016) Fatty acid composition of fourteen seashore mallow (*Kosteletzkya pentacarpos*) seed oil accessions collected from the Atlantic and Gulf coasts of the United States. *Industrial Crops and Products*, **7**:20–26.

Naczk, M., Amarowicz, R., Sullivan, A., & Shahidi, F. (1998) Current research developments on polyphenolics of rapeseed/canola: A review. *Food Chemistry*, **62**:489–502.

Niggeweg, R., Michael, A. J., & Martin, C. (2004) Engineering plants with increased levels of the antioxidant chlorogenic acid. *Nature Biotechnology*, **22**:746–754.

Nzikou, J., Matos, L., Bouanga-Kalou, G., Ndangui, C., Pambou-Tobi, N., Kimbonguila, A., … Desobry, S. (2009) Chemical composition on the seeds and oil of sesame (*Sesamum indicum* L.) grown in Congo-Brazzaville. *Advance Journal of Food Science and Technology*, **1**:6–11.

Onyeike, E. N., & Acheru, G. N. (2002) Chemical composition of selected Nigerian oil seeds and physicochemical properties of the oil extracts. *Food Chemistry*, **77**:431–437.

Pearson, D. A. (1976) *Chemical analysis of foods* (7th ed.). Edinburgh: Churchill Livingstone.

Pereira, M. A., & Ludwig, D. S. (2001) Dietary fiber and body-weight regulation: Observations and mechanisms. *Pediatric Clinics of North America*, **48**:969–980.

Proestos, C., Sereli, D., & Komaitis, M. (2006) Determination of phenolic compounds in aromatic plants by RP-HPLC and GC–MS. *Food Chemistry*, **95**:44–52.

Qasim, M., Abideen, Z., Adnan, M. Y., Gulzar, S., Gul, B., Rasheed, M., & Khan, M. A. (2017) Antioxidant properties, phenolic composition, bioactive compounds and nutritive value of medicinal halophytes commonly used as herbal teas. *South African Journal of Botany*, **110**:240–250.

Qasim, M., Aziz, I., Rasheed, M., Gul, B., & Khan, M. A. (2016) Effect of extraction solvents on polyphenols and antioxidant activity of medicinal halophytes. *Pakistan Journal of Botany*, **48**:621–627.

Qasim, M., Gulzar, S., Shinwari, Z. K., Aziz, I., & Khan, M. A. (2010) Traditional ethnobotanical uses of halophytes from Hub, Balochistan. *Pakistan Journal of Botany*, **42**:1543–1551.

Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., & Rice-Evans, C. (1999) Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biology & Medicine*, **26**:1231–1237.

Sadik, N. (1991) Population policies and programmes: Lessons learned from two decades of experience. New York University Press.

Sanches-Silva, A., Costa, D., Albuquerque, T. G., Buonocore, G. G., Ramos, F., Castilho, M. C., … Costa, H. S. (2014) Trends in the use of natural antioxidants in active food packaging: A review. *Food Additives and Contaminants: Part A*, **31**:374–395.

Singleton, V., & Rossi, J. A. (1965) Colorimetry of total phenolics with phosphomolybdic–phosphotungstic acid reagents. *American Journal of Enology and Viticulture*, **16**:144–158.

Smaoui, A., & Cherif, A. (1992) Glycerolipids in salt stressed cotton seeds. In I. A. Ungar (Ed.), *Ecophysiology of vascular halophytes*. Boca Raton, FL: CRC Press.

Vidal-Valverde, C., Frias, J., & Valverde, S. (1993) Changes in the carbohydrate composition of legumes after soaking and cooking. *Journal of the American Dietetic Association*, **93**:547–550.

Wang, N., Hatcher, D. W., Tyler, R. T., Toews, R., & Gawalko, E. J. (2010) Effect of cooking on the composition of beans (*Phaseolus vulgaris* L.) and chickpeas (*Cicer arietinum* L.). *Food Research International*, **43**:589–594.

Weber, D., Gul, B., Khan, M., Williams, T., Wayman, P., & Warner, S. (2001) Composition of vegetable oil from seeds of native halophytic shrubs. In E. D. McArthur & D. J. Fairbanks (Eds.), *Shrub land and ecosystem genetics and biodiversity* (pp. 287–290). Ogden, UT: Department of Agriculture, Forest Service, Rocky Mountain Research Station.

Willett, W. C. (2012) Dietary fats and coronary heart disease. *Journal of Internal Medicine*, **272**:13–24.

Williams, C. M. (2000) Dietary fatty acids and human health. *Annales de Zootechnie*, **49**:165–180.

Yajun, B., Xiaojing, L., & Weiqiang, L. (2003) Primary analysis of four salt tolerant plants growing in Hai-He plain, China. In H. Lieth & M. Mochtchenko (Eds.), *Cash crop halophytes: Recent studies. Task for Vegetation Science* (Vol. **38**, pp. 135–138). Dordrecht, Netherlands: Springer.

Yemm, E., & Willis, A. (1954) The estimation of carbohydrates in plant extracts by anthrone. *The Biochemical Journal*, **57**:508–514.

Zahra, S. S., Ahmed, M., Qasim, M., Gul, B., Zia, M., Mirza, B., & Haq, I. U. (2017) Polarity based characterization of biologically active extracts of *Ajuga bracteosa* Wall. ex Benth. and RP-HPLC analysis. *BMC Complementary and Alternative Medicine*, **17**: 443–458.

Zarrouk, M., El Almi, H., Youssef, N. B., Sleimi, N., Smaoui, A., Miled, D. B., & Abdelly, C. (2003) Lipid composition of seeds of local halophytes: *Cakile maritima*, *Zygophyllum album* and *Crithmum maritimum*. In H. Lieth & M. Mochtchenko (Eds.), *Cash crop halophytes: Recent studies. Task for Vegetation Science* (Vol. **38**, pp. 121–124). Dordrecht, Netherlands: Springer.