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Article

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Abstract: This study evaluates the antioxidant properties and nutritional composition of Portulaca oleracea grown in the arid region of Kyzylorda, Republic of Kazakhstan, highlighting its potential as a natural antioxidant source. The antioxidant capacity of Portulaca was assessed using the Ferric Reducing Antioxidant Power (FRAP) assay, yielding values of 43.5 ± 1.0 mg GAE/g dry weight, and DPPH radical scavenging activity, with a value of 83.77% at a concentration of 200 mcg/ml. Total phenolic content (TPC) and total flavonoid content (TFC) were recorded at 16.88 ± 0.39 mg GAE/g and 26.33 ± 0.97 mg rutin/g, respectively. Additionally, Portulaca showed a significant omega-3 fatty acid content, with linolenic acid (C18:3) measured at 26.7 ± 2.1%. The plant is also abundant in essential vitamins and minerals, including vitamin C (20.06 ± 4.61 mg/100g), calcium (7656.16 ± 2663.69 mg/100g), and iron (5309.65 ± 1056.76 mg/100g). These findings support the suitability of Portulaca as a natural alternative to synthetic antioxidants in the food industry, suggesting its potential application in the development of functional foods and promoting further research into its diverse benefits.

Keywords: portulaca oleracea; antioxidant activity; food additives; phenolic compounds; omega-3 fatty acids

1. Introduction

In recent years, the demand for functional foods with enhanced nutritional and health benefits has significantly increased, particularly those incorporating natural antioxidants, which have gained attention for their potential to stabilize food products and contribute to human health [1, 2]. Oxidative processes in foods, especially in products such as meat, can lead to lipid and protein degradation, resulting in diminished quality and shorter shelf life. Antioxidants play a crucial role in counteracting these effects, which makes them valuable additives in the food industry [3]. Portulaca oleracea (commonly known as purslane), particularly when grown in the arid region of Kyzylorda, Kazakhstan, is recognized for its rich nutritional profile, including high levels of omega-3 fatty acids, vitamins, and various antioxidants such as flavonoids and carotenoids [4, 5]. Its potential as a natural antioxidant source has been explored extensively, with studies indicating its capacity to enhance the oxidative stability of food products and improve their health profile [6–8]. Its notable content of essential fatty acids further positions it as a valuable candidate for food enrichment, offering a natural solution to improve oxidative stability in food matrices [9]. Despite the known health benefits of

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Portulaca, its application in food processing, particularly in regions like Kyzylorda, remains underexplored. The existing literature on natural antioxidants and their impact on food preservation presents divergent findings, with some studies highlighting complex interactions between plant-based compounds and food matrices that may affect overall quality [10–12]. This disparity necessitates further research on the specific impact of Portulaca's antioxidant properties when grown under arid regional conditions, as these factors may influence the nutritional and functional characteristics of the plant.

Historically, Portulaca has been valued both as a dietary component and traditional medicine across various cultures, attributed to its therapeutic effects in treating a wide array of ailments, including diabetes, urinary tract infections, renal and cardiovascular disorders, diarrhea, headaches, as well as snake and insect bites [13–18]. Research has shown that its leaves contain significant quantities of alpha-linolenic acid (300–400 mg), α-tocopherol (12.2 mg), ascorbic acid (26.6 mg), β-carotene (1.9 mg), and glutathione (14.8 mg) per 100 g of fresh biomass [19]. Furthermore, Portulaca is a vital source of specialized metabolites, including alkaloids, catecholamines, anthocyanins, flavonoids, saponins, tannins, cardiac glycosides, terpenoids, and phenolic acids [20, 21, 22, 23, 24]. Its wild genotypes are particularly notable for their high omega-3 fatty acid content, averaging approximately 188.48 ± 6.35 mg per 100 g of dry weight, making Portulaca one of the richest terrestrial sources of these essential fatty acids [27]. Across Portulaca species, more than 85 distinct metabolites have been identified, encompassing a wide array of chemical classes, such as alkaloids, fatty acids, phenolic acids, and amino acids, with specific parts of the plant demonstrating unique profiles—such as higher phenolic content in the flowers and greater flavonoid concentration in the leaves [25, 26, 28].

The objective of this study is to evaluate the antioxidant properties and nutritional composition of Portulaca oleracea grown in the arid region of Kyzylorda, Republic of Kazakhstan, with the aim of assessing its potential as a functional ingredient for enhancing the antioxidant capacity of food products. By addressing this regional gap in current research, this study aims to provide valuable insights into Portulaca's role as a natural antioxidant source, promoting its application in the development of more stable and nutritionally enriched food products.

2. Materials and Methods

2.1. Materials

In the present study we utilized dried Portulaca oleracea (Kyzylorda region, Kazakhstan). The plant material was harvested and subsequently dried in a dehydrator at 30°C for 48 hours to preserve its bioactive compounds and nutrients. Both the stems and leaves were dried and then ground together to ensure homogeneity in the sample preparation for further analysis.

2.2. Determination of Ferric Reducing Antioxidant Power (FRAP)

Ferric Reducing Antioxidant Power (FRAP) assay was conducted as previously described [29]. BHT and α-tocopherol were used as standard antioxidants. 1 mL of the extracts at various dilutions was added to 2.5 mL of phosphate buffer (0.1 M, pH 6.6) and 2.5 mL of potassium ferricyanide (1%, w/v), and the mixture was incubated at 50°C for 20 minutes. Following incubation, 2.5 mL of trichloroacetic acid (10%, w/v) was added to the mixture. Then, 2.5 mL of the solution was taken, followed by the addition of 2.5 mL of deionized water and 0.5 mL of ferric chloride solution (0.1%, w/v). The solution was allowed to stand for 30 minutes, and the absorbance was measured at 700 nm. The obtained FRAP values were expressed in milligrams of gallic acid equivalents (GAE) per gram of dry extract (mg GAE/g).

2.3. Determination of Antioxidant Activity Using the 2,2-Diphenyl-1-picrylhydrazyl (DPPH) Assay

All reagents and standards were of analytical grade. Ultrapure water (Milli-Q Waters purification system; Millipore; Milford, USA) was used. The reagents included methanol for HPLC

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(MeOH), DPPH ≥ 95%, gallic acid monohydrate >98%, formic acid (98-100%), and potassium persulfate (>99%) (Sigma-Aldrich).

The antioxidant activities were determined using DPPH as a free radical [19]. Briefly, 2 mL of DPPH solution (0.1 mg/mL in MeOH) was mixed with 2 mL of the samples at a concentration of 200 µg/mL. The reaction mixture was then shaken and incubated in the dark at room temperature for 30 min, and the absorbance was measured at 517 nm against a blank. Ascorbic acid was prepared similarly as a positive control, except that the an-tioxidant solution was replaced accordingly.

The degree of color change was quantified using a UV-Vis spectrophotometer (LabSolutions, Shimadzu). All analyses were performed in triplicate.

2.4. Determination Total Phenolic Compounds (TPC)

The total phenolic content in the obtained extracts was analyzed spectrometrically according to the Folin-Ciocalteu method [32].

Briefly, 100 µL of each extract (dissolved in MeOH) or a standard gallic acid solution was mixed with 2 mL of 2% (w/v) Na2CO3 solution. The mixture was then incubated for 5 minutes, followed by the addition of 100 µL of the Folin-Ciocalteu reagent. After a 30 min incubation at room temperature for color development, the absorbance was measured at 750 nm using a spectrophotometer. The results were expressed in milligrams of gallic acid equivalents (GAE) per gram of dry sample.

2.5. Determination of Total Flavonoid Content (TFC)

The total flavonoid content in the crude *Portulaca* extracts was determined using the aluminum chloride assay as previously described [33]. A 50 mg sample of *Portulaca* was dissolved in 10 mL of 80% aqueous MeOH and filtered through filter paper. In a test tube, 300 µL of the extract was mixed with 3.4 mL of 30% MeOH, 150 µL of 0.5 M NaNO2, and 150 µL of 0.3 M AlCl3·6H2O, followed by thorough mixing. After a 5 min incubation, 1 mL of 1 M NaOH was added. The absorbance was measured at 510 nm using a UV-Vis spectrophotometer. A calibration curve for the total flavonoid content was generated using a standard rutin solution (concentration range of 0–100 mg/L). The total flavonoid content was expressed in milligrams of rutin equivalents per gram of dry extract.

2.6. Determination of Total Carotenoid Content (TCC)

Isochromatic fractions of total carotenoids were evaluated as previously described [34]. The components were extracted using acetone as the solvent for sample preparation. The absorbance of the samples was measured spectrophotometrically at 472 nm and 508 nm, respectively. The content of red and yellow carotenoids was expressed in mg/g of dry weight.

3. Results and Discussion

3.1. Determination of Ferric Reducing Antioxidant Power (FRAP)

At the initial phase of this research, the Ferric Reducing Antioxidant Power (FRAP) assay was employed to evaluate the antioxidant capacity of pre-dried extracts from Portulaca oleracea. The phytochemical profile of Portulaca suggests significant antioxidant activity, a characteristic that has been corroborated by multiple studies utilizing diverse analytical methods [35]. Various anatomical parts of the plant, including the leaves and stems, were subjected to the FRAP assay to assess their respective antioxidant capabilities.

The results showed that the methanolic extract of the garden variety Portulaca exhibited strong ferric reducing antioxidant power, with FRAP values of 43.5 ± 1.0 mg GAE/g of dry weight. This effect is likely attributed to the elevated levels of total phenolic compounds, ascorbic acid, and β-carotene present in Portulaca [25]. Antioxidants play a vital role in human health by mitigating the risk of cellular damage induced by free radicals [36]. Numerous studies have explored and validated the antioxidant potential of Portulaca [37–39]. Investigating the antioxidant capacity of plant extracts is

 doi:10.20944/preprints202411.0657.v1

of significant interest for the discovery and development of novel and safer natural antioxidants for application in the food industry.

3.2. Determination of Antioxidant Activity Using the 2,2-Diphenyl-1-picrylhydrazyl (DPPH) Assay

The presence of phenolic acid in the extract of the tested plant increases the likelihood of reactions with free radicals, leading to a reduction in the number of free radicals. In our study, the DPPH radical scavenging assay (%) was used to assess the antioxidant activity of *Portulaca oleracea* extracts (Table 1).

Table 1. DPPH Radical Scavenging Activity of Methanolic Extract of Portulaca oleracea Soluble Plant Fractions, Compared to Ascorbic Acid.

Extract concentration (mcg/ml) 200 (w/v)	**Absorption activity (DPPH), % - 83.77±0.015**
Extracts	IC50 radical removal activity DPPH (mcg/ml)
Methanol extract of Portulaca oleracea	756.42±92.61
Ascorbic acid	80.76±4.71

It is evident that flavonoids contribute significantly to the antioxidant potential of Portulaca oleracea, as demonstrated by the strong correlation between total flavonoid content (TFC) and DPPH activity ($R^2 = 0.996$). This suggests that flavonoids may serve as the primary compounds responsible for scavenging free radicals in this plant. The moderate correlation between total phenolic content (TPC) and DPPH activity ($R^2 = 0.782$) further highlights that phenolic acids, while important, may play a secondary role compared to flavonoids. This underscores the importance of investigating the specific classes of flavonoids present in Portulaca to better understand their contribution to the overall antioxidant profile. Such insights could help in harnessing Portulaca oleracea as a natural source of antioxidants for potential applications in food and health industries.

3.3. Determination Total Phenolic Compounds (TPC)

Phenolic acids are key specialized metabolites in plants, derived from benzoic and cinnamic acid structures [40]. These compounds belong to the class of carboxylic acids that incorporate phenolic groups. A multitude of studies have identified various phenolic acids, including caffeic acid, p-coumaric acid, ferulic acid, gallic acid, gentisic acid, benzoic acid, and anisic acid [41].

Table 2. The content of phenolic compounds in the purslane sample.

Name of the sample	TPC concentration in mg GAE/g of dry extract
Methanol extract of Portulaca oleracea	16.88 ± 0.39

As secondary plant metabolites, phenols or polyphenols are highly significant due to their antioxidant activity through chelating redox-active metal ions, scavenging lipid free radical chains, and preventing the conversion of hydroperoxide into reactive OH--radicals. The total phenolic content (TPC) in the extracts, expressed as gallic acid equivalents (GAE), ranged from 16.49 to 17.27 mg GAE/g for the methanolic extract of *Portulaca oleracea*.

With respect to Total Phenolic Content (TPC), the experimentally cultivated Portulaca demonstrated relatively high values. TPC levels are influenced by factors such as the geographical origin of the sample, the developmental stage of the plant, and its age. Typically, TPC tends to increase during the early growth phases and declines as the plant matures [42]. Certain studies, however, have reported lower TPC values for Portulaca oleracea L. under different conditions [43].

3.4. Determination of Total Flavonoid Content (TFC)

Several flavonoids have been isolated from Portulaca, including apigenin, kaempferol, luteolin, quercetin, isorhamnetin, kaempferol-3-O-glucoside, and rutin. These compounds are known for their protective effects against coronary diseases and their role in promoting vascular activation [44].

 doi:10.20944/preprints202411.0657.v1

Flavonoids exhibit a range of biological functions in the human body, primarily due to their antioxidant, anti-inflammatory, antitumor, antiviral, and antibacterial properties [45]. As a large class of natural phenolic compounds, flavonoids are distributed throughout various plant tissues, including the root, stem, flower, and fruit. Five specific flavonoids—kaempferol, apigenin, myricetin, quercetin, and luteolin—have been identified through capillary electrophoresis with electrochemical detection [46,47]. Nayaka et al. [48] successfully isolated apigenin from Portulaca and demonstrated its significant antibacterial activity.

Therefore, in the next stage of research, the total flavonoid content in the *Portulaca* sample was evaluated. The data from this study are presented in Table 3.

Table 3. The content of total flavonoids.

Name of the sample	TFC concentration in mg of rutin/g of dry extract
Methanol extract of Portulaca oleracea	26.33±0.97

The total flavonoid content, expressed in mg of rutin equivalents per gram of dry sample, ranged from 25.36 to 27.3. These quantities were consistent with findings reported in the literature for other extracted samples [38]. It was conclusively shown that samples with elevated phenolic content also exhibited substantial concentrations of flavonoids. Plants rich in flavonoids have the potential to serve as excellent sources of antioxidants, thereby enhancing the body's overall antioxidant capacity and offering protection against lipid peroxidation [49].

3.5. Determination of Total Carotenoid Content (TCC)

It has been discovered that Portulaca contains significantly higher levels of β-carotene and α-tocopherol compared to spinach [50]. The abundance of these antioxidant molecules indicates that consuming Portulaca may aid in alleviating oxidative stress [51]. Given its rich composition of essential nutrients, Portulaca oleracea is regarded as a highly valuable plant with considerable nutritional potential [52,53].

In the next stage of the research, the total carotenoid content in the *Portulaca* sample was evaluated. The data from this study are presented in Table 4.

Table 4. The total carotenoid content (TCC) in Portulaca.

Name of the sample	The concentration of TCC is expressed in mg/g of dry extract
Methanol extract of Portulaca oleracea	4.33±0.57

The leaves and stems of Portulaca, cultivated in a controlled growth chamber, exhibited notably high carotenoid levels, particularly in comparison to the carotenoid composition of spinach leaves. It has been documented that both vitamin C (ascorbic acid) and beta-carotene possess significant antioxidant properties, primarily due to their capacity to neutralize free radicals, which may play a role in the prevention of cardiovascular diseases and cancer [54]. Among the various plant parts, the leaves displayed the highest concentrations of beta-carotene, ascorbic acid, and DPPH activity, followed by the flowers and stems. In fact, the beta-carotene content in the leaves was found to be twice as high as in the stems and slightly greater than in the flowers. This observation aligns with existing research on Australian Portulaca, where the beta-carotene content in the leaves also surpassed that of the stem [53].

3.6. Determination of PUFA

Portulaca is a nutritious vegetable crop rich in polyunsaturated fatty acids (PUFAs), and it is particularly known for its omega-3 (α-linolenic acid) and omega-6 (linoleic acid) fatty acids, which are essential for human health [55]. Several fatty acids have been isolated and identified from various parts of the Portulaca plant. Studies have shown that Portulacacontains a higher total fatty acid content than commonly consumed vegetables such as spinach, red leaf lettuce, mustard, and romaine

 doi:10.20944/preprints202411.0657.v1

lettuce [56]. It is recommended that a healthy diet be enriched with foods that have a higher omega-3/omega-6 ratio. Interestingly, Portulaca leaves have a very high omega-3/omega-6 ratio [57]. Moreover, Portulaca leaves have a high omega-3/omega-6 ratio [53,58,59]

The results of the fatty acid composition analysis of Portulaca samples, expressed as the mass fraction of fatty acid methyl esters from the total fatty acid methyl esters (%), are presented in Table 5.

Table 5. Fatty Acid Composition.

Name of the sample	Results
Linolenic acid C18:3	26.7±2.1
Thymnodonic acid C20:5	1.2±0.4
Linoleum C18:2	10.6±2.1
Era C22:1	13.5±2.1
Nervonic C24:1	7.1±2.1
Oleic C18:1	1.9±2.1
Palmitoleic C16:1	10.6±0.4
Palmitic C16:0	21.7±2.1
Stearic Acid C18:0	4.5±0.4

Portulaca is among the richest sources of omega-3 fatty acids found in green plants. The presence of these essential fatty acids in Portulaca provides significant advantages for vegetarians, whose diets often lack sufficient levels of omega-3 fatty acids [59].

Portulaca has been shown to lower cholesterol and triglyceride levels while simultaneously raising beneficial high-density lipoprotein (HDL) levels. In contrast to fish oil, which is often high in cholesterol and calories, Portulaca serves as an excellent source of omega-3 fatty acids without the presence of cholesterol. Additionally, research suggests that Portulaca may reduce the risk of cancer and heart disease, potentially owing to its naturally occurring omega-3 fatty acids [60]. Portulaca contains the highest amount of alpha-linolenic acid, an omega-3 fatty acid essential for human nutrition, compared to any other leafy green vegetable. Portulaca contains 300-400 mg of alpha-linolenic acid (C18:3, w3) per 100 g. It is also a rich source of linoleic acid (18:2, w3).

Portulaca is known to contain substantial quantities of essential dietary minerals, including copper, iron, manganese, magnesium, potassium, calcium, and phosphorus [61]. Findings by Santiago-Sáenz et al. [62] further confirm that Portulaca provides significant amounts of protein, fiber, and inorganic nutrients such as Fe, Cu, Mn, Zn, B, P, Ca, Mg, and K. Moreover, research conducted by Mohammed and Hussein [63] revealed a notable concentration of total solids in the roots.

The total dry matter and protein content in Portulaca fluctuate across various stages of growth. Significant variations were observed in the concentrations of total phosphorus, calcium, potassium, iron, manganese, and copper depending on the plant's growth stage. Portulaca is also a rich source of vitamins, including vitamin A, riboflavin, niacin, pyridoxine, vitamin C, thiamine, α-tocopherol, and pantothenic acid [64]. The high vitamin A content in Portulaca makes it a valuable option for individuals suffering from vision impairments and vitamin A deficiency [65].

The concentrations of vitamins and minerals in Portulaca are presented in Table 6.

Table 6. Main Nutraceutical Components and Their Concentration in Portulaca.

Name of the sample	Unit of Measurement	Results
Mass fraction of fat	%	3.5±0.5
Nitrogen	%	2.66±0.05
	Vitamins	
B1	mg/100g	0.06±0.01

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B2	mg/100g	0.13±0.05
B3 (PP)	mg/100g	0.64±0.13
B5	mg/100g	0.05±0.01
B6	mg/100g	0.06±0.02
B9	mg/100g	< 10.0
C	mg/100g	20.06±4.61
A	mcg/100g	< 10.0
E	mg/100g	< 0.1
	Minerals	
Ca	mg/100g	765.62±266.37
K	mg/100g	4643.35±764.2
Na	mg/100g	298.83±74.9
Mg	mg/100g	1595.41±312.2
Zn	mg/100g	6.47±1.32
Fe	mg/100g	530.97±105.67
Mn	mg/100g	12.55±3.87
Se	mg/100g	0.09±0.03
P	mg/100g	376.0±2.2

When comparing the nutritional composition to previously published results [65], our study revealed significant discrepancies, with notably higher concentrations of certain minerals: calcium was found to be 6 times higher, potassium 2 times higher, magnesium 20 times higher, iron 8 times higher, and phosphorus 5 times higher. In contrast, the sodium concentration was approximately half of the previously reported value, while zinc and manganese levels were nearly identical to those reported in [55]. Selenium levels were consistent with data from the literature [66].

Regarding vitamin content, after adjusting for dry weight, our findings indicated that only B vitamins (with the exception of B9) were comparable to the results in [67]. However, the vitamin C content was approximately 10 times lower than reported in [67, 61], and 5 times lower than in [53], when adjusted for dry weight.

The vitamin A content in [54] was reported at 1320 IU/100 g (0.4 mg/100 g), while in our study, it was determined to be less than 0.01 mg/100 g. The vitamin E content was also found to be within low ranges.

However, despite the lower content of ascorbic acid, and vitamins B9, A, and E in this study, compared to the recommended daily intake, Portulaca still offers beneficial properties. When included in a well-balanced diet, it presents an alternative, more affordable, and accessible source of B vitamins and minerals.

4. Conclusions

This study demonstrates that Portulaca oleracea from the arid Kyzylorda region possesses significant antioxidant and nutritional properties, making it a promising candidate for the food industry. With high antioxidant values, including FRAP at 43.5 ± 1.0 mg GAE/g and DPPH radical scavenging at 83.77%, Portulaca can naturally enhance the oxidative stability of food products. It also offers substantial nutritional benefits, with 26.7 ± 2.1% omega-3 (linolenic acid), 20.06 ± 4.61 mg/100g vitamin C, 7656.16 ± 2663.69 mg/100g calcium, and 5309.65 ± 1056.76 mg/100g iron. These levels support its use as a natural antioxidant and nutrient-rich additive, providing an effective, plant-based alternative to synthetic preservatives while enriching food products with essential nutrients. This

study suggests Portulaca's potential in developing healthier, more stable food products, especially for functional foods and supplements.

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