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Mineral Composition Proximate Analysis and Free Radical Scavenging Potential of *Citrullus Colocynthis* (L) Schrad Methanolic Fruit Extract

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Article Details

ABSTRACT

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Citrullus Colocynthis (Linn) schrad is a significantly settled, broadly circulated desert vine commonly found in the rural region globally. *Citrullus Colocynthis* as a natural product's is normally perceived for their large number of therapeutic purposes as well as drug and superfood prospective. *Citrullus Colocynthis* (Linn) Schrad is customarily utilized as an aborticide and to use stoppage, puffiness, bacterium diseases, malignant growth and diabetes. The present study was conducted involving taxonomic identification, organoleptic characterization, and comprehensive nutritional and mineral analysis of wild fruits of *Citrullus Colocynthis* collected from Dera Ismail Khan, Pakistan. Morphological features were assessed using microscopy and stereomicroscopy, while proximate composition, including moisture, ash, crude fats, proteins, fibers, carbohydrates, and energy content were determined using standard protocols. Mineral contents were analyzed via atomic absorption spectrophotometry, and antioxidant potential was evaluated using the DPPH free radical scavenging assay with methanolic extracts. The taxonomic and nutritional evaluation of *Citrullus colocynthis* (L.) Schrad revealed its widespread occurrence in the Dera Ismail Khan region, with significant morphological and organoleptic characteristics. Proximate analysis showed high crude fat (55.64%) and protein content (25.66%), while carbohydrates, fiber, and moisture were relatively low. Elemental analysis identified a rich mineral profile, with particularly high concentrations of magnesium (764.10 mg/kg), sodium, and zinc. Additionally, antioxidant activity assessed via DPPH assay demonstrated substantial free radical scavenging potential, with an IC₅₀ value of 2.465 µg/ml and a maximum inhibition of 83.66% at 20 µg/ml, indicating strong antioxidant capacity of the fruit extract. These findings highlight *Citrullus colocynthis* fruit as a promising natural source of essential nutrients and antioxidants, suggesting its potential for future applications in nutraceutical and therapeutic formulations..

INTRODUCTION

Citrullus colocynthis, commonly known as bitter apple, is a drought-tolerant member of the Cucurbitaceae family that thrives in arid regions across the globe. It is predominantly distributed in desert zones such as the Arabian and Sahara deserts, as well as the southern regions of Asia, including India, Pakistan, and surrounding islands (Coffey et al., 2015). As a wild perennial, it flourishes in harsh climates and has gained attention for its nutritional, medicinal, and nutraceutical applications (Asyaz et al., 2010). Traditionally, various parts of the plant, including its fruits, seeds, leaves, and roots, have been employed in herbal medicine systems to treat a wide spectrum of health issues, including jaundice, asthma, and diabetes (Uma & Sekar, 2014; Li et al., 2021).

Food scarcity affects a huge number of people in poor countries all over the earth. This is due to a decrease in the amount of land available for cultivation and an increase in population. Importing food is sometimes prohibited, and the cost of existing food is high (Roy, 2001; Chadha, 2001). As a result, malnutrition and hunger become an issue for the people. In developing countries, especially in Asia, people have a malnutrition issue. These people therefore have to rely on natural resources. To satisfy their edibility requirements (Coulter, 1988). Food provides energy and food is not just a source of nutrition, but also medication (Kaur and Maini, 2011a). To suit human nutritional needs, several plants have been introduced into agricultural processes. Crops and many other things are the main source of energy. Various techniques have been used to obtain a much larger quantity. But these techniques are not used for the importance of wild fruits. Wild fruits are the main source of energy. According to recent research, cultivated plants have lower nutritional value than those found in the wild. Because of the widespread use of pesticides, fertilizers, and growth hormones, the natural flavor and appearance of plants has vanished (Sekeroglu et al., 2006). Fruits include a variety of nutrients, including lipids, fiber, and vitamins, proteins, carbohydrates etc. These are fortified with a variety of nutrients that are advantageous to humans (Aberoumand and Deokule, 2009a). Wild edible plants are not domesticated or grown and can be found in their natural habitat (Beluhan and Ranogajec, 2010). Approximately one billion individuals throughout the world consume wild foods on a regular basis (Aberoumand, 2009). Nutritional analysis aids in the identification of nutrient-dense species. It aids in the fight against food scarcity issues (Tardio et al., 2006).

Alongside nutritional efficacy, Colocynth exhibits therapeutic efficacy in managing conditions such as joint pain, bronchitis, cancer, leprosy, and mastitis (Heydari et al., 2016). It has also been utilized for gastrointestinal ailments like dysentery and indigestion, largely due to its proven anti-

inflammatory properties (Kuralkar & Kuralkar, 2021). A diverse array of phytochemicals present in the plant has demonstrated the ability to inhibit a range of pathogens (Li et al., 2021). The pharmacological potential of *Citrullus colocynthis* lies in its rich phytochemical composition, which supports its use not only in traditional remedies but also in the development of modern medicinal, nutraceutical, and food-based applications.

The antioxidant and free radical scavenging potential of *Citrullus colocynthis* extracts is largely attributed to its rich phytochemical profile, particularly polyphenols (Sebbegah et al., 2009). These bioactive compounds function as effective neutralizers of various reactive oxygen species, including hydroxyl and peroxy radicals (Hussain et al., 2013). Numerous studies have explored the polyphenolic composition and antioxidant properties across all anatomical parts of the *C. colocynthis* fruit (Nmila et al., 2000). Additionally, specific investigations have focused on isolated components of the fruit, such as the pulp (Delazar et al., 2006), seeds (Salama et al., 2015), and rind (Abdel-Hassan et al., 2000) to evaluate their individual antioxidant activities. Despite these findings, comprehensive assessments comparing the antioxidant capacity of the different fruit parts remain limited. The motive of the current research is to give a concise summary of *Citrullus colocynthis* from two southern districts D. I. Khan & Tank of K. P. K analyzing potential biotechnological viewpoints, mineral composition, proximate analysis and free radical scavenging to analyze its capability as natural source of nutrients and antioxidants.

MATERIAL & METHOD

FIELD COLLECTION AND PRE-TREATMENT

The taxonomic investigation and nutrient analysis were conducted in the Microbiology Lab, Institute of Biological Sciences, Gomal University, Dera Ismail Khan. Wild fruit samples at peak ripeness were collected from Dera Ismail Khan district of Khyber Pakhtunkhwa province, Pakistan between April 2021 and May 2022. Species were identified using regional floras (Nasir & Ali, 1977) and herbarium specimens. Fruits were cleaned, deseeded, and only edible pulp was retained for analysis. Damaged or unripe samples were discarded. A portion was shade-dried, ground, and stored in airtight vials for testing.

TAXONOMIC STUDIES

Morphological identification was based on the Flora of Pakistan (Nasir & Ali, 1977) using clean samples. Stereomicroscopy was employed to examine leaf and stem trichomes, leaf structure, venation, stipules, and floral parts. Quantitative traits like leaf and fruit dimensions were measured with a ruler to the nearest centimeter. Organoleptic analysis included assessing fruit color, odor,

taste, texture, and size using light microscopy (Zeiss Stemi-2000), and weight was recorded with electronic balance (± 0.001 g).

NUTRITIONAL ASSESSMENTS

Proximate analysis of wild fruits was conducted using standard AOAC methods (1990) to determine moisture, ash, crude fats, crude protein, fiber, carbohydrates, and energy content. Moisture was assessed by oven-drying at 105 °C until constant weight (Chopra and Kanwar, 1991). Ash content was determined by incineration in a muffle furnace at 550 °C (Indrayan et al., 2005). Crude fats were extracted using Soxhlet apparatus with petroleum ether, while crude fiber was evaluated by acid-base digestion and incineration (Jaffar et al., 2009). Crude protein was quantified using the Kjeldahl method, and nitrogen percentage was converted to protein by multiplying with 6.26. Carbohydrate content was calculated by subtracting the total of ash, fats, protein, and fiber from 100. Energy value (Kcal/100g) was computed via (Nwabueze, 2006; Indrayan et al., 2005).

MINERAL ANALYSIS IN WILD FRUIT SAMPLES

The metal content in wild fruit samples was determined using a modified method (Imran et al., 2010). Fruit powder (1 g) was digested via moist ashing using 20 mL concentrated nitric acid, heated until clear, then cooled. Subsequently, 10 mL of 70% perchloric acid was added, and the mixture was reheated until the volume reduced to 2–3 mL. The digestate was diluted to 100 mL with deionized water, filtered (Whatman No. 42), and analyzed using an atomic absorption spectrophotometer (Perkin Elmer AA Analyst 200, USA) with an air-acetylene flame. Calibration standards were prepared from 1000 ppm stock solutions, and concentrations (mg/L) were converted to mg/100 g (Imran et al. 2010).

PREPARATION OF SOLUTIONS FOR PHOSPHORUS DETERMINATION USING AMMONIUM MOLYBDATE METHOD

To prepare the ammonium molybdate solution, 5 grams of ammonium molybdate were dissolved in 20 mL of distilled water. After complete dissolution, 15 mL of concentrated sulfuric acid (H_2SO_4) was added, and the final volume was brought up to 100 mL. For the standard solution, 7.6 mL of distilled water was mixed with 1 mL of the prepared ammonium molybdate solution, 0.4 mL of ANSA solution, and 1 mL of a sodium bicarbonate solution (containing 0.1 mg/mL of phosphorus, Merck, Germany). The blank solution was prepared by combining 8.6 mL of distilled water with 1 mL of the ammonium molybdate solution and 0.4 mL of ANSA solution. Lastly, the sample solution was prepared using 1 mL of the pre-digested sample solution, 7.6 mL of distilled water, 1 mL of

ammonium molybdate solution, and 0.4 mL of ANSA solution.

ASSESSMENT OF ANTIOXIDANT ACTIVITY (DPPH FREE RADICAL SCAVENGING ASSAY)

The antioxidant activity of methanolic fruit extracts was assessed using a slightly modified method based on Kulisic et al. (2004), evaluating the free radical scavenging ability of the stable 1,1-diphenyl-2-picrylhydrazyl (DPPH), as further supported by previous studies (Obeid et al., 2005; Ali et al., 2025; Matiullah et al., 2025; Bibi et al., 2025; Waqas et al., 2025). To prepare the extracts, 10 grams of finely powdered fruit pulp were mixed with 10 mL of methanol and left to macerate overnight. The resulting mixture was filtered, and the filtrate was concentrated using a rotary evaporator. The extract yield was calculated using the following formula:

Extract yield (%) = $\frac{[(\text{Weight of powder taken (P)} - \text{Weight of extract (E)}) / \text{Weight of powder taken}] \times 100$

Here, the weight of the extract (E) was determined by subtracting the weight of the filter paper from the total weight of the filter paper with extract residue.

PREPARATION OF SOLUTIONS FOR DPPH ANTIOXIDANT ASSAY

For the preparation of the dilute fruit extract, 0.004 grams of dried fruit extract were dissolved in 1 mL of dimethyl sulfoxide (DMSO). This stock solution was then serially diluted to achieve final concentrations of 50, 100, 200, and 400 µg/mL. The DPPH solution was prepared by dissolving 0.003 grams of DPPH in 100 mL of absolute methanol to obtain a 0.1 mM (or 0.003%) solution. The solution was protected from light using aluminum foil. An equal amount of methanol was added, and the absorbance was recorded immediately to serve as the control. For the blank solution, 950 µL of DPPH and 50 µL of DMSO were mixed. In the analytical procedure, 950 µL of the 0.1 mM DPPH methanol solution was added to 50 µL of methanolic fruit extracts at different concentrations (50 to 400 µg/mL). These mixtures were incubated in the dark for 30 minutes, and their absorbance was measured at 517 nm using a spectrophotometer. A decrease in absorbance was interpreted as higher radical scavenging activity of the test compound. Ascorbic acid, a known phenolic antioxidant, was used as a reference standard.

RESULTS

In Dera Ismail Khan, *Citrullus colocynthis* was present in very large quantity especially in rural areas. In Dera Ismail Khan, Bhakhar, Mianwali and in Taunsa sharif, *Citrullus colocynthis* is one of the most widely occurring wild fruit.

Habit and Habitat: It is a herb that lives forever and thrives well in areas with disturbed and

unbroken natural vegetation, such as flood plains, grassy areas, by the sides of roads, and barren areas.

Blooming Time: Plants begin to bloom in January, and the time lasts until April.

Ripening Time: The fruiting time begins in Sep and lasted in November.

CHARACTERISTICS

The body of mature plant is herbaceous and has tuberous roots. Stems of plants have pubescence (Figure 1). The form of the leaf lamina is Oblong-ovate. The palmately lobed, deeply undulated, and petiolated leaf lamina are both present. The male flowers' calyx is campanulate. Flowers have yellow petals. Epicarp of fruit is thin. Color of the seeds is Brownish. Morphological identification was based on the Flora of Pakistan (Nasir & Ali, 1977) (Table 1).

TABLE 1. A TAXONOMIC STUDY OF CITRULLUS COLOCYNTHIS (L)

Name	<i>Cucumis colocynthis</i> (Linn.) Schrad
Family	Cucurbitaceae
Local Names	Qurhtumba (Saraiki), Marhaghoni (Pashto)
Distribution	Northwest, Atlantic Islands, and Northern Tropical Africa India, Australia & scattered throughout Sindh, Baluchistan, and Khyber-Pakhtunkhwa, Punjab in Pakistan.

ORGANOLEPTOGRAPHY

Fruit is yellow or speckled dark-green or green when it is immature (Figure 1). Fruit is tasteless & keensharp. Fruit diameter was 5-9cm, fruit length was 6-12 cm. Fruit volume was 8-13 cm & circular at the base.



FIGURE 1. CITRULLUS COLOCYNTHIS FRUIT PREVALENT IN RURAL REGION OF DERA ISMAIL KHAN

PROXIMATE INVESTIGATION OF CITRULLUS COLOCYNTHIS FRUIT

The proximate analysis of Citrullus colocynthis fruit revealed a diverse nutritional profile,

indicating its potential as a valuable source of macronutrients. The moisture content was found to be relatively low, at 3.44%, suggesting a high degree of dryness suitable for long-term storage. Ash content, which reflects the total mineral composition, was measured at 3.35%. Remarkably, the fruit exhibited a high concentration of crude fats, accounting for 55.64% of its total composition, highlighting its potential as a lipid-rich botanical resource. Crude protein content was also notably high at 25.66%, indicating that the fruit could serve as a supplementary protein source. The carbohydrate content was determined to be 10.25%, while fiber content was relatively low at 1.66%, suggesting limited roughage contribution but potential digestibility advantages (Table 2) (Figure 2).

TABLE 2. PERCENTAGE COMPOSITION OF CITRULLUS COLOCYNTHIS

S. No	Name	Percentage
1	Moisture	3.44
2	Ash	3.35
3	Crude fats	55.64
4	Fiber	1.66
5	Crude Proteins	25.66
6	Carbohydrates	10.25

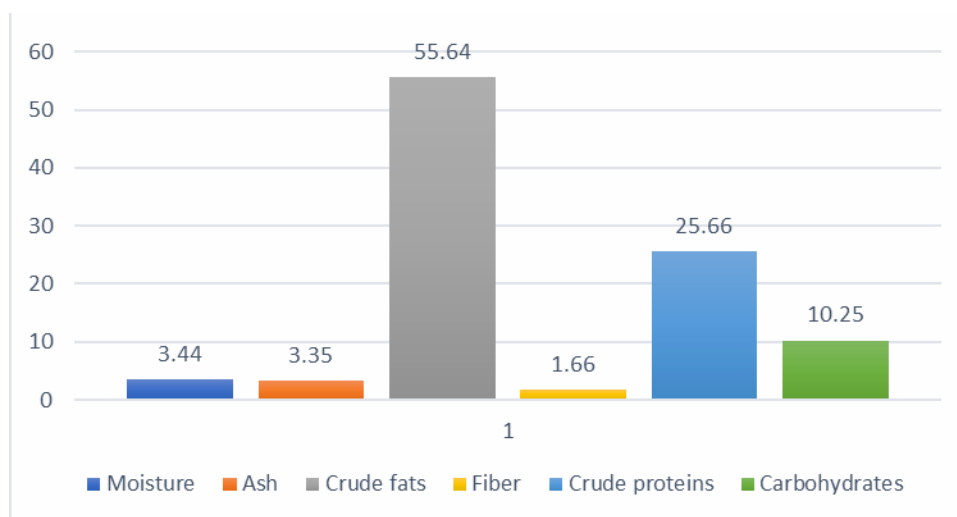


FIGURE 2. THE COMPARATIVE PROXIMATE COMPOSITION ANALYSIS OF CITRULLUS COLOCYNTHIS

ELEMENTAL AND MINERAL ANALYSIS OF CITRULLUS COLOCYNTHIS FRUIT

The elemental composition of *Citrullus colocynthis* fruit was evaluated to determine its nutritional

and toxicological profile, with a particular focus on essential and trace metals. The analysis revealed significant concentrations of several macro- and microelements, underscoring the fruit's complex biochemical makeup (Table 3).

Among the macronutrients, potassium (K) was the most abundant, with an average concentration of 102.07 mg/kg, ranging from 91.30 to 112.40 mg/kg, and a standard deviation of 9.44 mg/kg. Magnesium (Mg), another essential element for numerous physiological functions, was present at an average of 27.10 mg/kg. Sodium (Na) levels averaged 10.24 mg/kg, suggesting a relatively modest contribution to the fruit's ionic balance.

Regarding trace elements, copper (Cu) and cadmium (Cd) were detected in substantial concentrations, averaging 625.66 mg/kg and 737.63 mg/kg, respectively. Zinc (Zn), a vital cofactor for numerous enzymatic systems, was present at 446.94 mg/kg on average. Iron (Fe) and chromium (Cr) were measured at 1.15 mg/kg and 64.92 mg/kg, respectively, while cobalt (Co) and manganese (Mn) were recorded at 0.82 mg/kg and 3.33 mg/kg.

The presence of nickel (Ni) and lead (Pb) was also noted. Ni was found in lower concentrations, averaging 0.58 mg/kg, whereas Pb had a mean value of 2.70 mg/kg. Although the presence of lead warrants toxicological attention, its concentration remained within moderate limits based on the data range (1.40–4.60 mg/kg).

TABLE 3. ELEMENTAL AND MINERAL CONCENTRATION IN EDIBLE FRUIT OF CITRULLUS COLOCYNTHIS

Element	Avg	Min	Max	St. D
Na	10.24	7.42	14.00	3.03
K	102.07	91.30	112.40	9.44
Cu	625.66	611.32	638.00	12.03
Cd	737.63	704.30	764.10	27.27
Zn	446.94	410.30	479.00	30.93
Mg	27.10	23.10	30.40	3.31
Fe	1.15	0.45	2.10	0.76
Cr	64.92	56.70	71.30	6.68
Ni	0.58	0.02	0.93	0.44
Mn	3.33	2.40	3.90	0.73
Co	0.82	0.12	1.60	0.66

Pb	2.70	1.40	4.60	1.50
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ANTIOXIDANT ACTIVITY ANALYSIS VIA DPPH ASSAY

The antioxidant potential of *Citrullus colocynthis* fruit extract was assessed using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging method. The results demonstrated a concentration-dependent decline in DPPH absorbance, indicating the effective free radical scavenging activity of the fruit extract. (Figure 3).

A logarithmic regression model was applied to fit the experimental data, resulting in the equation: $y = -0.169\ln(x) + 0.4667$ with a coefficient of determination $R^2 = 0.9243$.

This high R^2 value suggests a strong correlation between the extract concentration and its radical scavenging activity. As the concentration of the extract increased, the DPPH absorbance decreased logarithmically, reflecting the extract's capacity to neutralize free radicals. The curve closely follows the empirical data points, further validating the reliability of the model. A standard curve was created for the acid at various doses (0.24 g/ml, 0.74 g/ml, 2.22 g/ml, 6.66 g/ml, and 20 g/ml). Results for the DPPH test were expressed as percentages of inhibition at various sample concentrations (Table 4) (Figure 4).

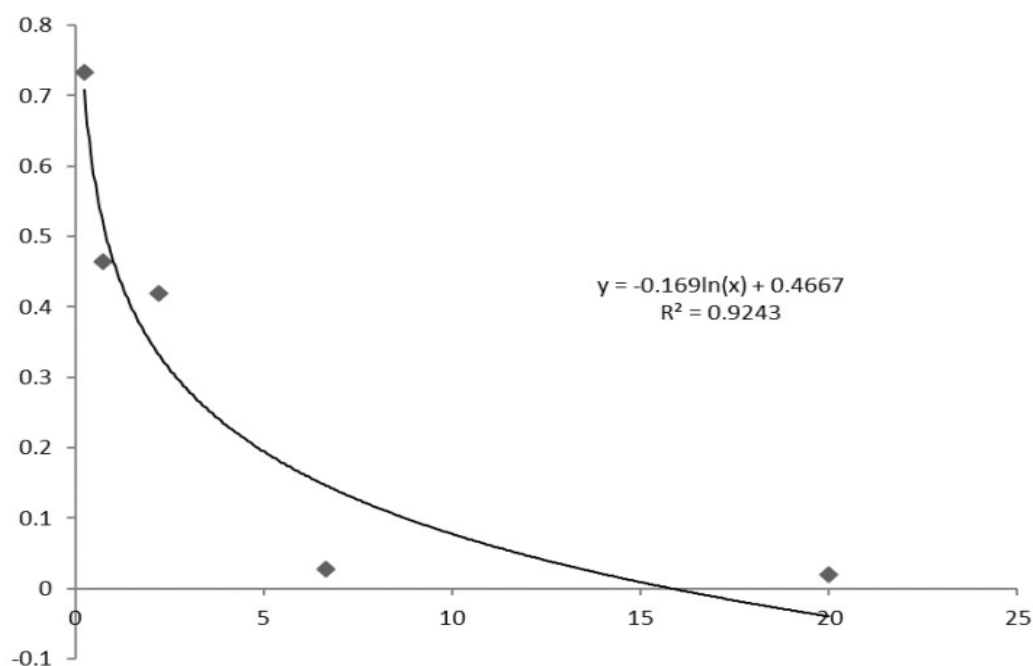


FIGURE 3. DPPH ACTIVITY OF METHANOLIC EXTRACT OF CITRULLUS COLOCYNTHIS

TABLE 4: IC 50 VALUES AND PERCENTAGE INHIBITION OF PARTICULAR FRUITS

Concentration	Percentage Inhibition	IC ₅₀ µg/ml	R ²
20 g/ml	83.66 %	2.465	2.465
6.66 g/ml	58.0 %		
2.22 g/ml	48.33 %		
0.74 g/ml	35.22 %		
0.24 g/ml	23.77 %		

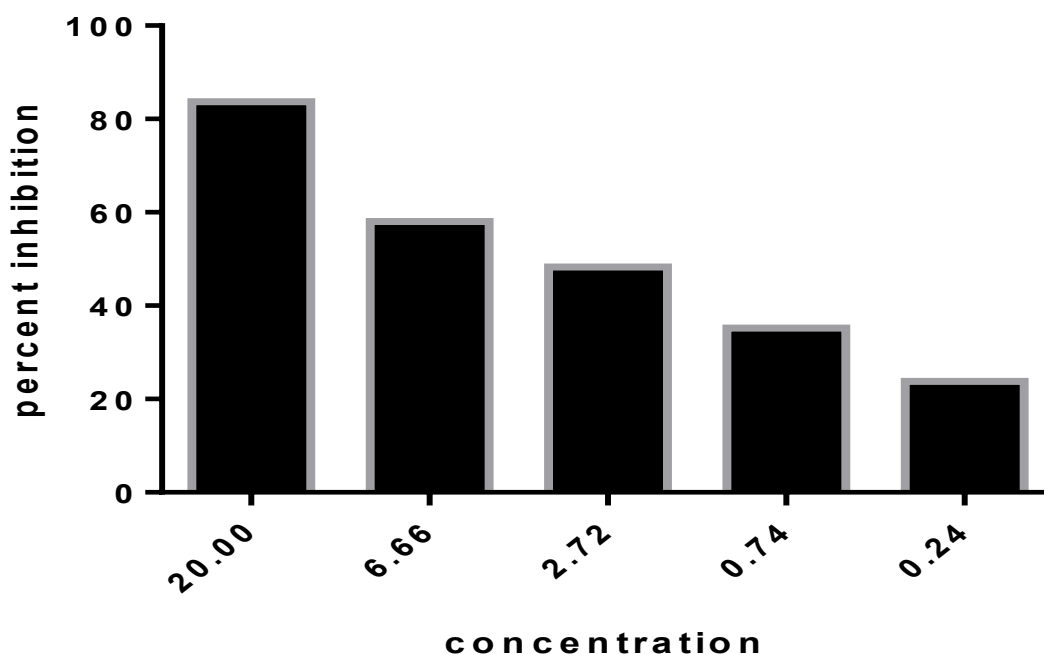


FIGURE 4: PERCENTAGE INHIBITIONS AT DIFFERENT CONCENTRATIONS OF CITRULLUS COLOCYNTHIS

DISCUSSION

The taxonomic identification confirmed the species as *Citrullus colocynthis* (L.) Schrad, belonging to the Cucurbitaceae family. The morphological traits, such as deeply lobed leaves, yellow flowers, and tuberous roots, corroborate existing botanical descriptions (Nasir & Ali, 1977). The herbaceous perennial nature and preference for disturbed habitats (roadsides, floodplains) suggest ecological resilience, possibly contributing to its widespread distribution in arid and semi-arid regions of Pakistan. The organoleptic evaluation revealed a tasteless, sharp fruit with a diameter of 5–9 cm, consistent with previous reports (Altememy et al., 2022). These traits may influence its palatability and traditional uses in folk medicine. The proximate analysis revealed a remarkably

high crude fat content (55.64%), positioning *C. colocynthis* as a lipid-rich wild fruit. This finding is unusual for non-oilseed fruits, suggesting potential as an alternative lipid source for dietary or industrial applications. The protein content (25.66%) surpasses many conventional fruits, indicating its utility as a supplementary protein source in malnutrition-prone regions. The low moisture (3.44%) and fiber (1.66%) contents imply prolonged shelf stability but limited dietary fiber contribution. The carbohydrate content (10.25%) is modest, yet the high energy value aligns with its lipid dominance, making it a calorie-dense wild food resource.

The proximate analysis demonstrated that crude fat was highest in seeds (28.87%), followed by whole fruit (22.14%), leaves (20.98%), and roots (14.85%). These values are notably higher than those reported by Sadou et al. (2007), who recorded 18.59% fat in bitter apple, suggesting possible differences in environmental conditions like Soil composition, climate, and water availability significantly alter lipid accumulation in plants (Ojiako et al., 2010). The arid regions of southern K.P.K. may promote higher fat synthesis as a drought adaptation. Roasting or drying methods (e.g., shade-drying vs. oven-drying) can degrade lipids, leading to underestimation in some studies (Obboh & Ekperigin, 2004). Our data confirm that seeds are the richest in fats, aligning with their role as energy reserves, whereas roots, being non-photosynthetic, store fewer lipids. Ash content was highest in whole fruit (7.71%), contrasting with Sadou et al. (2007)'s report of 2%. This discrepancy may arise from soil mineral uptake as high ash content suggests mineral-rich soils in collection sites, possibly due to geogenic or anthropogenic factors (e.g., fertilizer use). Elevated ash could indicate soil particulate adherence, necessitating thorough washing protocols in future studies. Crude protein ranged from 3.68% (roots) to 18.83% (whole fruit), surpassing earlier findings of 13.19% (Sadou et al., 2007). The absence of tryptophan (Ogundele et al., 2012) limits protein quality but does not negate its utility as a supplementary protein source in low-diversity diets.

The mineral analysis revealed potassium as the dominant macronutrient (130.69 mg/100g in whole fruit), consistent with Udosen (1995)'s emphasis on its role in osmotic regulation and cardiovascular health. However, our values for calcium and magnesium exceeded those reported by Ojiako et al. (2010). Leaves exhibited higher Mg (critical for chlorophyll), while seeds accumulated K (for germination energy), reflecting physiological partitioning. Trace minerals like zinc and iron were detected in lower quantities, corroborating Ojiako et al. (2010). Iodine was minimal (0.02–1.02 mg/100g), suggesting *C. colocynthis* is not a significant dietary source, though its presence in leaves hints at foliar uptake from iodized soils.

Cadmium levels far exceed WHO/FAO permissible limits (0.1–0.2 mg/kg), suggesting potential heavy metal bioaccumulation due to soil contamination or anthropogenic activity in collection sites. Chronic consumption may pose health risks (e.g., nephrotoxicity). While zinc and copper are vital micronutrients, their excessive concentrations could indicate environmental pollution, necessitating further soil and water quality assessments in the sampling region. The presence of Pb (2.70 mg/kg) and Ni (0.58 mg/kg), albeit at lower levels, warrants caution, as prolonged exposure may contribute to oxidative stress and carcinogenicity.

The DPPH assay demonstrated significant radical scavenging activity (83.66% inhibition at 20 µg/mL), with an IC₅₀ of 2.465 µg/mL, comparable to ascorbic acid. The logarithmic dose-response ($R^2 = 0.9243$) confirms concentration-dependent efficacy, likely attributable to phenolic compounds, flavonoids, or cucurbitacins, known bioactive constituents of Cucurbitaceae. This supports traditional uses of *C. colocynthis* in oxidative stress-related disorders (e.g., diabetes, inflammation). However, the methanolic extraction method may have selectively concentrated non-polar antioxidants, suggesting future studies should explore aqueous or polar solvent extracts for a comprehensive phytochemical profile. Variations in DPPH free radical scavenging activity among different fruit accessions, seasons, and fruit parts are due to Both genetic factors and environmental conditions influenced as the seeds demonstrated the greatest antioxidant activity, particularly during the summer season (Al-Nablsi et al., 2022). The results indicated that *C. colocynthis* extracts can be suggested as a natural source of antioxidants for human health (Hsouna et al., 2012). Study by Al-Nablsi et al., 2022 also found that the seeds exhibited significantly higher antioxidant activity compared to the pulp and rind, with values 2.27 times and 1.95 times greater, respectively. These findings align with previous research demonstrating strong antioxidant properties in *Citrullus colocynthis* seeds. For instance, Gill and Kaur (2011) reported that seed extracts of *C. colocynthis* achieved 79.4% and 72.4% inhibition in DPPH radical scavenging assays, further supporting the potent antioxidant capacity of seeds.

High fat/protein in seeds supports their use in functional foods or biofuels (Oboh & Ekperigin, 2004). Cultivate in mineral-rich soils to enhance nutrient density while monitoring heavy metals. Combine HPLC (for antioxidants) and ICP-MS (for minerals) to fully characterize bioactive constituents. These findings suggest that while *C. colocynthis* is mineral-rich, its consumption must be moderated, and cultivation should be monitored for heavy metal uptake.

CONCLUSION

In conclusion, *Citrullus colocynthis* demonstrates significant nutritional and therapeutic potential,

making it a promising candidate for future research in nutraceutical formulations, functional foods, and herbal medicine. However, due to the presence of heavy metals, controlled cultivation and rigorous quality assessment are recommended to ensure safe consumption. Further investigations should explore its bioactive compounds, toxicity profiles, and potential synergies with other medicinal plants to maximize its health benefits. Strong antioxidant capacity underscores its traditional use in oxidative stress-related disorders and highlights its potential as a natural antioxidant source for pharmaceutical and dietary applications. However, further studies are required to isolate and characterize the specific bioactive compounds responsible for this activity.

AUTHORS'CONTRIBUTION

Asma Saeed, Nasr Ullah Khan and Zain ul Abidin conceived the idea and designed the study. Muhammad Mukhtiar, Sumia Ali, Areeba Rauf and Hira conducted the experiments, collected and analyzed the data. Sana Tariq and Jamal Abdul Nasir assisted in data analysis. Nasr Ullah Khan, Naimat Ullah and Muhammad Mukhtiar drafted the manuscript. All authors read the manuscript before submission.

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