DOI: https://doi.org/10.54393/df.v6i1.164

DIET FACTOR
Journal of Nutritional & Food Sciences
https://www.dietfactor.com.pk/index.php/df
ISSN (E): 2789-8105, (P): 2789-8091
Volume 6, Issue 1 (Jan-Mar 2025)

Original Article

OPEN ACCESS

Whole Orange Powder as A Rich Source of Polyphenols, Flavonoids and Antioxidants

Muhammad Khalid Saeed[1*], Naseem Zahra[1], Asma Saeed[1], Salman Saeed[1], Khurram Shehzad[1], Shaista Nawaz[1], Abdul Ahad Rasheed[1], Syed Hussain Imam Abidi[1] and Qurat Ul Ain Syed[1]

[1]Food and Biotechnology Research Centre, Pakistan Council of Scientific and Industrial Research Laboratories Complex, Lahore, Pakistan

ARTICLE INFO

Keywords:
Whole Orange Powder, Polyphenols, Flavonoids, Antioxidants

How to Cite:
Saeed, M. K., Zahra, N., Saeed, A., Saeed, S., Shehzad, K., Nawaz, S., Rasheed, A. A., Abidi, S. H. I., & Syed, Q. U. A. (2025). Whole Orange Powder as A Rich Source of Polyphenols, Flavonoids and Antioxidants: Orange Powder: Source of Polyphenols, Flavonoids and Antioxidants. DIET FACTOR (Journal of Nutritional and Food Sciences), 6(1), 24-28. https://doi.org/10.54393/df.v6i1.164

***Corresponding Author:**
Muhammad Khalid Saeed
Food and Biotechnology Research Centre, Pakistan Council of Scientific and Industrial Research Laboratories Complex, Lahore, Pakistan
rosefbrc1@gmail.com

Received Date: 25th January, 2025
Revised Date: 10th March, 2025
Acceptance Date: 18th March, 2025
Published Date: 31st March, 2025

ABSTRACT

On a global scale, consumers' faith in dietary therapy for illness remediation has been bolstered by health claims for phytochemical-containing foods, including fruits and vegetables. Thanks to their antioxidant capabilities and chemical variety, polyphenols gave dietary supplements and nutraceuticals new life. **Objectives:** To investigate the antioxidant activity and phytochemicals (TPC and TFC) of the entire orange powder while extracted in water, ethanol and methanol. **Methods:** Each extract was tested for its total flavonoid composition using the aluminium chloride technique and its total polyphenolic content using the Folin reagent. The DPPH test was used to measure the antioxidant activity. **Results:** Results demonstrated that whole orange powder water extract had the lowest total phenolic content values (167.2 ± 3.3 mg GAE/g), flavonoids (35.8 ± 0.2 mg QE/g) whereas methanol extracts displayed the highest values (350.8 ± 6.3mg GAE/g; 72.5 ± 2.2 mg QE/g) and ethanol extracts showed the moderate values (283.4 ± 5.2 mg GAE/g; 57.4 ± 1.8mg QE/g) respectively. At a concentration of 20-100µg/ml, the methanol extract had the greatest antioxidant% inhibition value, 38.50 ± 1.3-87.67 ± 2.4%, followed by the ethanol extract (28.70 ± 1.1-65.40 ± 2.1%) whereas the water extract had the lowest antioxidant% inhibition value, 17.95±0.3-52.25 ± 1.6% and it showed a statistically significant difference values ($p<0.05$) among the extracts. **Conclusions:** It was concluded that the antioxidant levels, polyphenols and flavonoids in whole orange powder were strongly affected by the solvent type employed for extraction, with methanol being the solvent of choice.

INTRODUCTION

Every year, more than 100 million tons of citrus fruits are produced throughout the world, marking a remarkable increase in output. The fruit juice extraction process is the lifeblood of this booming sector, but it also produces copious amounts of waste items, including seeds, pulp, and peels. Half of the fruit is not fit for human consumption, leading to an annual waste of 60 million tons [1, 2]. The peels of citrus fruits, which make up more than half of the fruit itself, are often wasted and ignored, which has serious consequences for the environment, the economy, and our nutrition. These problems are made worse in emerging nations like India due to their inadequate infrastructure for managing this massive amount of biomass. To encourage environmentally friendly production and consumption, the European Union and the United Nations have made efforts in the last decade to redistribute and valorize food waste under a hierarchical system. One of the biggest producers of citrus (Oranges),

DOI: https://doi.org/10.54393/df.v6i1.164

which is aptly referred to as the king of all simple peeler kinds and surpasses the greatest varieties worldwide, is Pakistan. Furthermore, in Pakistan's overall fruit culture, citrus is the country's main crop in terms of both area and production. With a production level of 2.29 million tons, these fruits are cultivated on 210.47 thousand hectares in the Punjab province [3]. There is a lost chance to extract useful bioactive chemicals from citrus peels and entire trash oranges, which are now thrown away. The wide variety of chemicals that make up these compounds has made them famous for the positive effects they may have on health [4]. Citrus peel extracts have not been well investigated. There are several nutritional advantages of eating citrus fruits, which are common in the subcontinental diet. For example, oranges are a good source of flavonoids like narirutin 4'-glucoside, eriocitrin, narirutin, a compound called isosakuranetin, sinensetin, quercetagetin/gossypetin, a medication called no, 3,5,6,7,8,3',4'-heptamethoxyflavone, tangeritin/5-hydroxy-3,7,8,3',4'-pentamethoxy-flavone, Chrysoeriol, limocitrol, limocitrol, and limocitrol, among others, which are most commonly found in orange juice [5]. In addition to a wealth of nutrients, such as vitamins C, A, and B, micronutrients (calcium, phosphorus, potassium), and polyphenols (carotenoids, amino acids, which are triterpenes, phenolic acids, and flavonoids) found in orange peel powder, it is also an outstanding source of dietary fiber [6]. The presence of a group of flavonoids, including narirutin 4'-glucoside (1), eriocitrin (2), narirutin (3), hesperidin (4), isosakuranetin (5), sinensetin (6), quercetagetin/gossypetin (7), nobiletin (8), 3,5,6,7,8,3',4'-heptamethoxyflavone (9), tangeritin/5-hydroxy-3,7,8,3',4'-pentamethoxyflavone (10), Chrysoeriol (11), limocitrin (12), limocitrol (13) (Figure 1) [7].

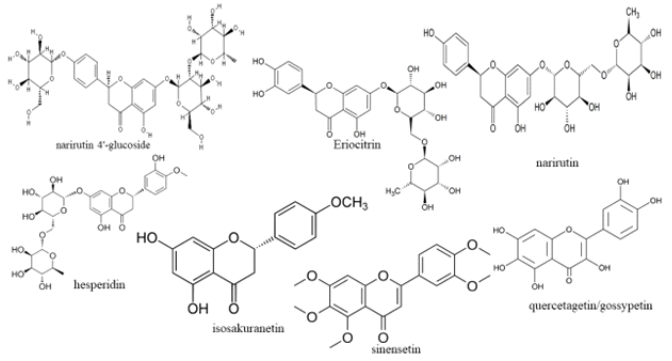

Figure 1: The Presence of a Group of Flavonoids

Using HPLC-DAD, a research by researcher [8] identified nine peaks of polyphenolic compounds, including catechin, caffeic acid, naringin, epicatechin, rutin, quercitrin, quercetin, kaempferol, and luteolin. According to [9], these phenolic compounds can be incorporated into food products or, upon extraction, may be used as natural protectors to shield certain foods from oxidation. Scientific research has shown that orange by-products, which contain a high concentration of bioactive compounds, may have unique and boosted therapeutic effects against oxidative stress in cancer cells, Type 2 diabetes, and cardiovascular diseases [10]. Catechin (peak 1), caffeic acid (peak 2), naringin (peak 3), epicatechin (peak 4), rutin (peak 5), quercitrin (peak 6), quercetin (peak 7) kaempferol (peak 8) and luteolin (peak 9) (Figure 2) [8].

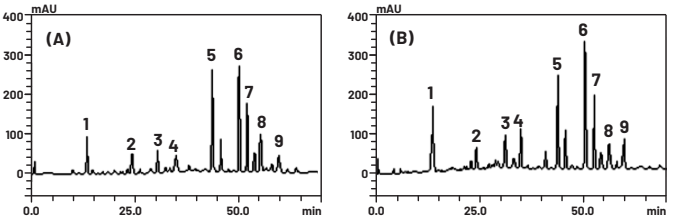

Figure 2: Representative High Performance Liquid Chromatography Profile of Unripe and Ripe Orange Peel Powder

Internally produced reactive oxygen species (ROS) like hydrogen peroxide, superoxide anion radicals, and hydroxyl radicals can cause oxidative stress, which in turn can lead to several diseases, such as cancer, cardiovascular disease, ageing, and neurodegenerative disorders [11, 12]. Exogenous antioxidants from polyphenol-rich foods, such as fresh produce, play a key role in regulating reactive oxygen species (ROS), which in turn has a good effect on human health [13].

This study aims to determine the whole orange powder's polyphenols, flavonoids, its antioxidant activity, and the phytochemical bioactive components. This research has the potential to reveal these byproducts' untapped potential and confirm their status as an affordable, all-natural antioxidant source.

METHODS

After being thoroughly cleaned and chopped, the waste, approximately 40 kg of oranges 500 (Citrus aurantium L.) from the citrus orchard in Sargodha, Pakistan, were dried in a hot air oven dryer for six to eight hours, and the samples were kept in airtight containers [14]. For effective extraction, 20 grams of powdered material was extracted in 200 milliliters of distilled water, methanol, and ethanol, then left at room temperature for 24 hours. After passing the extract through Whatman filter paper No.1, it was kept at 4°C until needed. Following the recommendation of Singleton [15], the total polyphenolic content was determined. To summarize, 2.5 mL of 10% Folin-Ciocalteau's reagent (v/v) was used to oxidize the proper dilutions of the extracts and 2.0 mL of 7.5% sodium carbonate was used to neutralize the reaction. In a spectrophotometer (UV-700 Shimadzu Japan), the intensity of absorption was detected at 765 nm after 40 minutes of incubation at 45°C. After that, the gallic acid

DOI: https://doi.org/10.54393/df.v6i1.164

equivalent was used to determine the total phenol level. As reported earlier, the colorimetric approach was modified to estimate the total flavonoid makeup of both extracts. For this experiment, we mixed half a milliliter of the extract solution with half a milliliter of an ethanol solution containing 20 milligrams per milliliter of $AlCl_3$. The amount of flavonoids was determined as mg QE/g of the extract mixture using the absorbance at 420 nm reading from a wavelength analyzer (UV-1700, Shimadzu Japan) after an hour of incubation at 25°C. The DPPH, which (2,2-diphenyl-1-picrylhydrazyl) scavenging technique, was used to assess the anti-radical activity of the extracts and was slightly modified. A purple DPPH solution (20 mg/L in methanol) was added to the extracts (50 µg/mL at the finish throughout the well), and the mixture was left to incubate at a comfortable temperature for 15 minutes. With the use of an ultraviolet (UV)-visible spectrophotometer (UV-1700, Shimadzu Japan), the DPPH radical's decolonization was seen at 517 nm. The result was reported as DPPH antiradical activity as a percentage compared to the control group that received just DPPH and solvent, without the extracts. The mean ± standard deviation (SD) was used to express the results. Analysis of variance unidirectional (ANOVA) was used to identify significant differences ($p<0.05$), and the Tukey test for multiple comparisons was then used.

RESULTS

Results demonstrated that whole orange water extracts had the lowest total phenolic content values (167.2 ± 3.3 mg GAE/g), flavonoids (35.8 ± 1.2 mg QE/g) whereas methanol extracts displayed the highest values (350.8 ± 6.3mg GAE/g; 72.5 ± 2.2 mg QE/g) and ethanol extracts showed the moderate values (283.4 ± 5.2 mg GAE/g; 57.4 ± 1.8mg QE/g) respectively. At a concentration of 20-100µg/ml, the methanol extract had the greatest antioxidant% inhibition value, 38.50 ± 0.3-87.67 ± 2.4%, followed by the ethanol extract (28.70 ± 1.1-65.40 ± 2.1%), and the water extract had the lowest, 17.95 ± 0.3-52.25 ± 1.6% (Table 1).

Table 1: Total Polyphenols and Flavonoids of Various Extracts of Whole Orange Powder

Extracts	TPC (mg GAE/g)	TFC (mg QE/g)
Methanol (Me-OH)	350.8 ± 6.3	72.5 ± 0.1
Ethanol (Et-OH)	283.4 ± 5.2	57.4 ± 0.4
Water (H_2O)	167.2 ± 3.3	35.8 ± 1.2

Data are represented as mean ± SD

The DPPH radical was used to assess the extracts' free radical scavenging capabilities. At the concentration 20-100 µg/ml, the methanol extract exhibited the highest antioxidant DPPH (% inhibiting) value, ranging from 38.50 ± 1.3-87.67 ± 2.4%, while the ethanol extract came in second with a value ranging from 28.70 ± 1.1-65.40 ± 2.1%. The water extraction procedure of whole dried orange peel had the lowest value, ranging from 17.95 ± 0.3-52.25 ± 1.6%, and it showed statistically significant differences in values ($p<0.05$) among the extracts. The availability of several bioactive components, including the flavonoids that greatly determines the antioxidant potential of whole orange powder peel extracts. Preventing cellular damage and chronic illnesses relies on these chemicals' ability to scavenge free radicals and alleviate oxidative stress. Because these bioactive chemicals have variable distributions and solubility, the antioxidant capabilities of the various plant sections and solvents used in this investigation varied significantly (Figure 3).

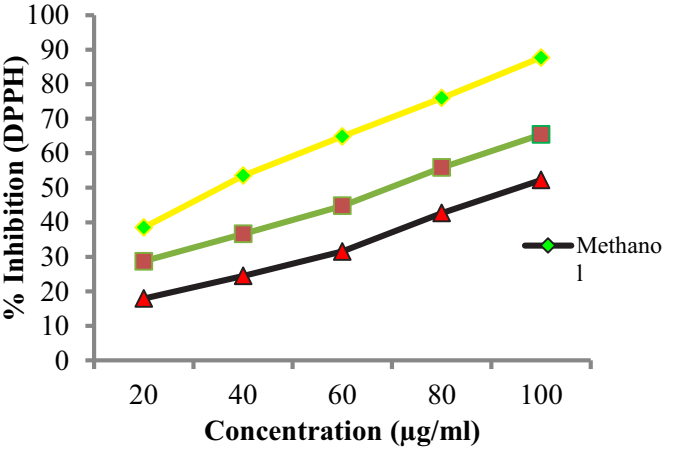

Figure 3: % Inhibition (DPPH) Various Extracts of Whole Orange Peel Powder

DISCUSSION

It was necessary to identify phenolics in food due to the health claims linked with them. The Folin-Ciocalteu technique, which is based on reducing phosphor-tungstic acid to phosphor-tungstic blue, was used for phenolic analysis. This led to a rise in absorbance because there were more aromatic phenolics [16]. If the blue hue becomes more intense, it means that there are more antioxidants in the sample. The orange peel extracts in methanol had the highest concentrations of phenol (350.8 ± 6.3 mg GAE/100 g), and the ethanol extract had TPC (283.4 ± 5.2 mg GAE/g), while the water extract from the whole orange powder showed the lowest concentration of TPC (167.0 ± 3.3 mg GAE/g). The anti-inflammatory, flavor-enhancing, colour-enhancing and phenolic-rich β-carotene found in abundance in orange peels has been linked to the alleviation of several ailments [17]. Carotenoids present in orange peel powder are phytonutrients that are soluble in fat and have effects that prevent cancer, oxidative stress and mutations [18]. The unique citrus scent that has a profound effect on human existence is believed to be mostly produced by flavonoids, which are secondary metabolic product components often present in sweet orange peels [19]. Anticancer,

DOI: https://doi.org/10.54393/df.v6i1.164

antibacterial, antioxidant, anti-inflammatory and anti-allergic are only a few of the many biological effects shown by polyphenols, terpenes and terpenoids [20]. The antioxidant activity of the extracts in this investigation was excellent, with methanol showing the greatest results. Additionally, compared to the ethanol as well as water extracts, the methanol extracts exhibited somewhat greater antioxidant activity. This could be because different solvents extract antioxidant molecules to different degrees. Research has shown that orange peel extracts have strong antioxidant properties; our findings are in line with the literature cited [21].

CONCLUSIONS

It was concluded that whole orange (*Citrus aurantium* L.) powder is a great option due to its high phenolic, flavonoid and antioxidant content. Since they contain bioactive compounds or therapeutic candidates that show considerable action against oxidative stress. It is therefore a good choice for either creating new products or treating or aiding in the therapy of various illnesses. Furthermore, the whole orange powder's antioxidant content was significantly impacted by the kind of extraction solvent used.

Authors Contribution

Conceptualization: MKS
Methodology: MKS, NZ, AS, KS
Formal analysis: SS
Writing review and editing: SN, AAR, SHIA, QUAS

All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest

All the authors declare no conflict of interest.

Source of Funding

The author received no financial support for the research, authorship and/or publication of this article.

REFERENCES

[1] Psakis G, Lia F, Valdramidis VP, Gatt R. Exploring Hydrodynamic Cavitation for Citrus Waste Valorization in Malta: From Beverage Enhancement to Potato Sprouting Suppression and Water Remediation. Frontiers in Chemistry. 2024 May; 12: 1411727. doi: 10.3389/fchem.2024.1411727.

[2] Mahato N, Sinha M, Sharma K, Koteswararao R, Cho MH. Modern Extraction and Purification Techniques for Obtaining High-Purity Food-Grade Bioactive Compounds and Value-Added Co-Products from Citrus Wastes. Foods. 2019 Oct; 8(11): 523. doi: 10.3390/foods8110523.

[3] Hayat A, Haque EU, Nasir M, Nawaz R, Mahmood T, Afzaal S. Comparison and Evaluation of Physico-Chemical Properties of Five Orange Varieties Under Agro-Climatic Conditions of Sargodha, Pakistan. Pakistan. Journal of Innovative Sciences. 2022; 8(2): 236-42. doi: 10.17582/journal.jis/2022/8.2.236.242.

[4] Mueed A, Shibli S, Al-Quwaie DA, Ashkan MF, Alharbi M, Alanazi H et al. Extraction, Characterization of Polyphenols from Certain Medicinal Plants and Evaluation of Their Antioxidant, Antitumor, Antidiabetic, Antimicrobial Properties, and Potential Use in Human Nutrition. Frontiers in Nutrition. 2023 Jun; 10: 1125106. doi: 10.3389/fnut.2023.1125106.

[5] Kainat S, Arshad MS, Khalid W, Zubair Khalid M, Koraqi H, Afzal MF *et al.* Sustainable Novel Extraction of Bioactive Compounds from Fruits and Vegetables Waste for Functional Foods: A Review. International Journal of Food Properties. 2022 Dec; 25(1): 2457-76. doi: 10.1080/10942912.2022.2144884.

[6] Rani V, Sangwan V, Malik P. Orange Peel Powder: A Potent Source of Fiber and Antioxidants for Functional Biscuits. International Journal of Current Microbiology and Applied Sciences. 2020; 9(9): 1319-25. doi: 10.20546/ijcmas.2020.909.167.

[7] Mojo T, Sutrisno S, Marfuah S. Chemical Content and Pharmacology of Sweet Orange (Citrus sinensis) Fruit Peel: A Review. In E3S Web of Conferences. 2024; 481: 06002. doi: 10.1051/e3sconf/202448106002.

[8] Omoba OS, Obafaye RO, Salawu SO, Boligon AA, Athayde ML. HPLC-DAD Phenolic Characterization and Antioxidant Activities of Ripe and Unripe Sweet Orange Peels. Antioxidants. 2015 Jul; 4(3): 498-512. doi: 10.3390/antiox4030498.

[9] Caponio GR, Annunziato A, Vacca M, Difonzo G, Celano G, Minervini F et al. Nutritional, Antioxidant and Biological Activity Characterization of Orange Peel Flour to Produce Nutraceutical Gluten-Free Muffins. Food and Function. 2024; 15(16): 8459-76. doi: 10.1039/D4FO01395F.

[10] Lucarini M, Durazzo A, Bernini R, Campo M, Vita C, Souto EB *et al.* Fruit Wastes as A Valuable Source of Value-Added Compounds: A Collaborative Perspective. Molecules. 2021 Oct; 26(21): 6338. doi: 10.3390/molecules26216338.

[11] Sanjaya IP, Chan MT, Sukma D, Buchori A, Putra RP, Nurcholis W. D-Optimal Design Optimization of Solvent Mixture for Flavonoid Extraction from Phalaenopsis Leaves with Antioxidant Activity. Journal of Pharmacy and Bio-Allied Sciences. 2024 Jan; 16(1): 1-7. doi: 10.4103/jpbs.jpbs_372_23.

[12] Rouamba A, Ouedraogo V, Ouédraogo M, Kiendrebeogo M. Antioxidant Effects and Protective Potential of Fruit Extracts of Detarium Microcarpum

DOI: https://doi.org/10.54393/df.v6i1.164

Against Arsenic Trioxide-Induced Human Lymphocytes DNA Oxidative Damages. Avicenna Journal of Phyto-Medicine. 2025 Jan; 15(1).

[13] Ajeena SJ, Al-Zaidi RM, Hadi MT, Ghazal MM. Study of the Antioxidant Activity of Some Active Compounds in Orange Peels. Cellular and Molecular Biology. 2024 Nov; 70(10): 161-6. doi: 10.14715/cmb/2024.70.10.21.

[14] Shanthi PK, Dhanalakshmi B, Pugazhenthi TR, Samuel Masilamoni Ronald B. Characterization and Antioxidant Activity of Orange Peel Extracts. 2019 Jun.

[15] Singleton VL. Lamuela-Raventos: Analysis of Total Phenols and Other Oxidation Substrates and Antioxidants by Means of Folin-Ciocalteu Reagent. Methods in Enzymology. 1999; 299: 152. doi: 10.1016/S0076-6879(99)99017-1.

[16] Ashraf H, Iahtisham-Ul-Haq, Butt MS, Nayik GA, Ramniwas S, Damto T *et al.* Phytochemical and Antioxidant Profile of Citrus Peel Extracts about Different Extraction Parameters. International Journal of Food Properties. 2024 Dec; 27(1): 286-99. doi: 10.1080/10942912.2024.2304274.

[17] Azman NF, Azlan A, Khoo HE, Razman MR. Antioxidant Properties of Fresh and Frozen Peels of Citrus Species. Current Research in Nutrition and Food Science Journal. 2019 Aug; 7(2): 331-9. doi: 10.12944/CRNFSJ.7.2.03.

[18] Razola-Díaz MD, De Montijo-Prieto S, Guerra-Hernández EJ, Jiménez-Valera M, Ruiz-Bravo A, Gómez-Caravaca AM et al. Fermentation of Orange Peels by Lactic Acid Bacteria: Impact on Phenolic Composition and Antioxidant Activity. Foods. 2024 Apr; 13(8): 1212. doi: 10.3390/foods13081212.

[19] Tripoli E, La Guardia M, Giammanco S, Di Majo D, Giammanco M. Citrus Flavonoids: Molecular Structure, Biological Activity and Nutritional Properties: A Review. Food Chemistry. 2007 Jan; 104(2): 466-79. doi: 10.1016/j.foodchem.2006.11.054.

[20] Gutiérrez-del-Río I, Fernández J, Lombó F. Plant Nutraceuticals as Antimicrobial Agents in Food Preservation: Terpenoids, Polyphenols and Thiols. International Journal of Antimicrobial Agents.2018 Sep;52(3):309-15.doi:10.1016/j.ijantimicag.2018.04.024, 04.024

[21] Ogo O, Hembafan N, Amokaha R, Jeremiah O, Inalegwu B. Characterization and Antioxidant Activity of Peel Extracts from Three Varieties of Citrus Sinensis. Heliyon. 2024 Apr; 10(7). doi: 10.1016/j.heliyon.2024.e28456.