frontiers in Sustainable Food Systems

TYPE Original Research
PUBLISHED 08 April 2026
DOI 10.3389/fsufs.2026.1762690

OPEN ACCESS

EDITED BY
Pankaj Singh,
Dr. Rammanohar Lohia Avadh University, India

REVIEWED BY
Bulu Mohanta,
Seemanta Institute of Pharmaceutical Sciences (SIPS), India
Roque Ribeiro Da Silva Junior,
University of the State of Rio Grande do Norte, Brazil

*CORRESPONDENCE
Rokayya Sami
✉ rokayya.d@tu.edu.sa

RECEIVED 07 December 2025
REVISED 13 March 2026
ACCEPTED 23 March 2026
PUBLISHED 08 April 2026

CITATION
Jalloun RA, Alharbi MH, Alfarteesh HA, Alqahtani NK, Felemban AH, Jawharji MT, Hafiz HA, Sami R, Wazzan H, Hijazi M, Altuwairki D, Alharbi HF, Algheshairy RM, Altalhi R and Alharbi NHJ (2026) Investigation of sustainable strawberry parts against quercetin on pro-inflammatory responses and oxidative stress in LPS-activated RAW 264.7 macrophages. *Front. Sustain. Food Syst.* 10:1762690. doi: 10.3389/fsufs.2026.1762690

COPYRIGHT
© 2026 Jalloun, Alharbi, Alfarteesh, Alqahtani, Felemban, Jawharji, Hafiz, Sami, Wazzan, Hijazi, Altuwairki, Alharbi, Algheshairy, Altalhi and Alharbi. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.

Investigation of sustainable strawberry parts against quercetin on pro-inflammatory responses and oxidative stress in LPS-activated RAW 264.7 macrophages

Rola A. Jalloun[1], Mudi H. Alharbi[1], Hajer A. Alfarteesh[2], Nashi K. Alqahtani[3,4], Alaa H. Felemban[5], Monya T. Jawharji[5], Halah A. Hafiz[5], Rokayya Sami[6]*, Huda Wazzan[7], Maha Hijazi[7], Duaa Altuwairki[7], Hend F. Alharbi[8], Reham M. Algheshairy[8], Rawan Altalhi[9] and Nora H. J. Alharbi[10]

[1]Department of Clinical Nutrition, Taibah University, Medina, Saudi Arabia, [2]Department of Biology, College of Science, King Khalid University, Abha, Saudi Arabia, [3]Department of Food and Nutrition Sciences, College of Agricultural and Food Sciences, King Faisal University, Al-Ahsa, Saudi Arabia, [4]Date Palm Research Center of Excellence, King Faisal University, Al-Ahsa, Saudi Arabia, [5]Department of Clinical Nutrition, Faculty of Applied Medical Sciences, Umm Al-Qura University, Makkah, Saudi Arabia, [6]Department of Food Science and Nutrition, College of Sciences, Taif University, Taif, Saudi Arabia, [7]Department of Food and Nutrition, Faculty of Human Sciences and Design, King Abdulaziz University, Jeddah, Saudi Arabia, [8]Department of Food Science and Human Nutrition, College of Agriculture and Food, Qassim University, Buraydah, Saudi Arabia, [9]Department of Biological Sciences, College of Science, University of Jeddah, Jeddah, Saudi Arabia, [10]Department of Pharmacology and Toxicology, College of Pharmacy, University of Ha'il, Ha'il, Saudi Arabia

A large amount of by-products, mostly peels and seeds, are produced during the industrial processing of strawberries and are currently underutilized and thrown away as environmental trash. By valuing these particular residues as functional agents, this study closes this sustainability gap. We examined the anti-inflammatory and antioxidant properties of several strawberry fractions in comparison to the common flavonoid, quercetin, using a lipopolysaccharide (LPS)-induced RAW 264.7 macrophage paradigm. Our results showed that whereas LPS and strawberry extracts preserved high cell viability (above 84%), the extracts considerably reduced the oxidative and inflammatory stress caused by LPS, supporting the conversion of industrial waste into valuable nutraceuticals. Strawberry parts decreased both nitric oxide (NO) production (6.25: 44.15 μM) and reactive oxygen species (ROS) (0.11: 0.51 μM). The peel group reduced ROS levels to 0.17 μM, demonstrating the strongest antioxidant activity; quercetin reduced levels closest to the control sample (11.78; 0.15 μM), respectively. Strawberry peel reduced the inflammatory markers to reach tumor necrosis factor-alpha (TNF-α) 117.04 pg./mL, Interleukin 6 (IL-6) 160.47 pg./mL, and Interleukin-1 beta (IL-1β) 88.2 pg./mL. Antioxidant activity was highest in the peel (•OH 65.96%, ABTS 156.05 μmol Trolox/g), followed by the whole fruit (•OH 48.29%, ABTS 98.87 μmol Trolox/g, FRAP 89.48 μmol Fe^{2+}/g, DPPH IC_{50} 32.28 μg/mL), and the seed (•OH 56.08%, ABTS 112.44 μmol Trolox/g, FRAP 102.16 μmol Fe^{2+}/g, DPPH IC_{50} 46.17 μg/mL). The pulp (•OH 33.14%, ABTS 71.25 μmol Trolox/g, FRAP 58.30 μmol Fe^{2+}/g, DPPH IC_{50} 88.15 μg/mL). Peel had the highest ascorbic acid (71.22 mg/100 g) and folate (36.41 μg/100 g), while seed had the lowest (7.68 mg/100 g; 12.12 μg/100 g), respectively. The largest concentrations of total phenol (159.16 mg GAE/100 g),

flavonoid (44.16 mg QE/100 g), anthocyanin (33.08 mg/100 g), catechin (27.16 mg/100 g), and carotenoid (5.28 mg/100 g) were found in peel; proanthocyanidin (28.92 mg/100 g) were found in seed samples. Peel showed the highest POD (3.11 U/mL) and PPO (3.99 U/mL) activity; pulp had the lowest PPO (1.71 U/mL) and seed the lowest POD (1.89 U/mL); the whole fruit was intermediate. The peel part had the greatest antioxidant activities, anti-inflammatory markers, and bioactive attributes, whereas the pulp and seed showed lower beneficial effects, and it can be used in manufacture instead of being dumped as waste material.

KEYWORDS

anti-inflammatory markers, bioactive compounds, oxidation, RAW 264.7 cell line, strawberry parts, sustainable use

Introduction

An immunological reaction to tissue damage carried on by infection or even adverse stimuli as microbes and viruses, is inflammation. It is crucial for preserving immunological homeostasis and acts as a physiological reaction in the body (Justine et al., 2024). Macrophages, which are found in the blood and tissues as monocytes, are one type of immune cell that controls the inflammatory response. They are essential for the rapid induction and amplification of inflammation and immunity (Bahuguna et al., 2020). Gram-negative bacterial cells produce pro-inflammatory cytokines, while their outer membrane contains a pro-inflammatory substance called lipopolysaccharide (LPS) that activates macrophages (Seke et al., 2023). However, it's crucial to identify anti-inflammatory medications to treat autoimmune disorders, including cancer, multiple sclerosis, pneumonia, and rheumatoid arthritis, because excessive inflammatory reactions can cause those conditions (El-Feky and El-Rashedy, 2023). This has led to the current method of using research on the control of inflammatory reactions to LPS-stimulated macrophages to examine the effectiveness of novel natural materials (Lin et al., 2020). The strawberry (*Fragaria ananassa Duch.*), belongs to the Rosaceae family, is recognized for its nutritional components, gorgeous red color, scent, and distinctive balance of sweet and acidic flavors (Urün et al., 2021). The flavor of strawberries is formed by sugars, fiber, micronutrients, organic acids, and other volatile organic compounds (VOCs). VOCs significantly affect the fruit's aroma through ester, alcohol, aldehyde, ketone, acid, and lactone, including ethyl hexanoate, linalool, and γ-decalactone, even though they presents $\leq$0.01% of the fruit's fresh weight. Different cultivars, ripening phases, and environmental factors all affect their concentration, which results in different flavor profiles. While organic acids add flavor and antioxidant qualities, the sugar-to-acid ratio is equally important in influencing sweetness and overall taste (Liu et al., 2025). Strawberries are typically eaten as whole fruits or in processed forms such asjuice, ice cream, jam, and jelly. Therefore, the fruits are consumed; however, the stem, leaf, root, seed, and calyx are not used and considered as bio-waste (González-Domínguez et al., 2020). Anthocyanins, quercetins, flavonoids, polyphenols, ellagitannins, catechins, ascorbic acids, and folic acids are among the phytochemical components found in strawberries (Villamil-Galindo et al., 2020).

As the primary innate immune cells, macrophages are essential in both infection and inflammation, performing vital regulatory roles in pathological inflammation (Xu et al., 2024). A variety of biological characteristics, including antioxidants, anti-inflammatory, antiviral, and neuroprotective actions, are possessed by the flavonoid compound quercetin (Newerli-Guz et al., 2023).

Quercetin inhibited LPS-induced macrophage activation by targeting spleen tyrosine kinase (Syk)/ proto-oncogene tyrosine-protein kinase src (Src)/ interleukin-1 receptor–associated kinase 1 (IRAK-1). It also prevented LPS-induced oxidative stress and inflammation through pathways NADPH oxidase 2 (NOX2)/ Reactive oxygen species (ROS)/Nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB) (Sul and Ra, 2021). Figure 1 presents the molecular structure of the quercetin as a strong antioxidant components.

The present study aimed to examine the ethanolic extracts of sustainable strawberry parts against against quercetin on pro-inflammatory responses, antioxidant activities, phytochemical compounds, vitamins, enzymes, and oxidative stress in LPS-activated RAW 264.7 macrophages.

Materials and methods

Materials and cell culture

Sigma company (St. Louis, MO, USA) supplied quercetin (Q4951), lipopolysaccharide (LPS), which derived from *Escherichia coli* (0111:B4), and dimethyl sulfoxide (DMSO). The RAW264.7 mouse macrophage cell line was supplied from the Shanghai Institute of Biological Sciences in China. The cells were cultivated at 37 °C in a fully humidified incubator with 5% CO_2 in Dulbecco's Modified Eagle's Medium (DMEM) of 2 mM glutamine, 1 mM pyruvate, 4.5 g/L glucose, which was supplemented with 10% heat-inactivated fetal bovine serum (FBS) (Xue et al., 2017). The cells were frequently

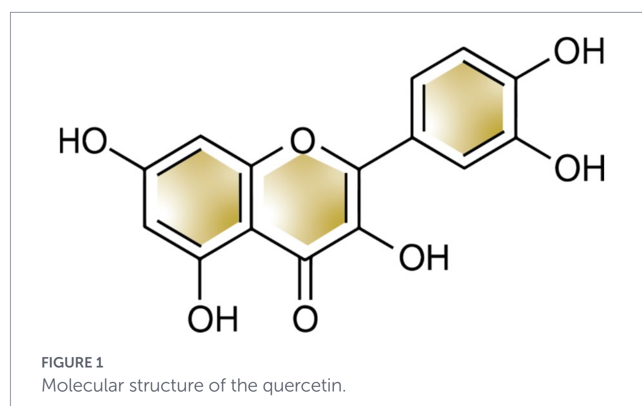

FIGURE 1
Molecular structure of the quercetin.

transferred to a third generation after being cultivated as a dense monolayer.

Sample extraction

Fresh strawberries (*n* = 60 fruit) were grown under controlled conditions of agronomic and environmental factors and propagated by stolon cuttings in Taif, Saudi Arabia. The fruits were in good condition when they were hand-picked in the early morning after the ripening stage in November, 2024. Fresh strawberries were arranged in plastic trays at random, while three separated sections were manually separated from the strawberry samples. The whole fruit presented the fourth group. The pulp was obtained by chopping off the crimson interior part of the fruit, leaving out the peel and seeds. The tiny seeds were removed from the skin using mechanical vacuum separation (MVS-S300, Zhucheng, China). Samples were milled into a fine powder to improve the surface area for extraction after being freeze-dried (FD-1000 L, Ningbo, China) to retain phenolic chemicals and anthocyanins. In order to get a pure peel fraction, the peel, including portions containing seeds, was collected separately. The samples were ground into a consistent powder (MF-10, Jingxin Co., Ltd., China). The samples are placed in dark tubes and combined at a 1:10 (w/v) ratio with 70% that contained 0.1% HCl. The mixture was shaken (LS-200, Changzhou, China) at 125 rpm for 24 h at room temperature before the precipitate and supernatant were separated. This process was repeated twice. After the extraction process was applied, the supernatant was filtered before being concentrated under low pressure using a rotating vacuum evaporator (YRE 2000E, Gongyi, China) and the extract was lyophilized and stored at −20 °C in the dark (Kim et al., 2024).

Study design

Seven treatment groups were utilized in the experiment: Control (untreated cells), LPS, (LPS + Pulp), (LPS + Peel), (LPS + Seed), (LPS + Fruit), and (LPS + 1 μM Quercetin). Initially, the effects of the biological substance were assessed using RAW264.7 cells. While the LPS group received 1 μg/mL of LPS to cause inflammation, the control group did not receive any form of treatment (Monmai et al., 2022).

Cell viability assay

The MTT assay was used to determine cell viability (Zhong et al., 2022). A 96-well plate was seeded with a density of 1×10^4 RAW 264.7 cells each well and allowed to adhere overnight. Following the preparation and addition of extracts at a concentration of 100 μg/mL based on the crude dry weight, the cells were stabilized at 37 °C in an incubator with 5% CO_2. Each well was then injected with 40 μL of MTT reagent (2.5 mg/mL) and allowed to react for 3 hours. Following the incubation period, 100 μL of DMSO was added to dissolve the formazan crystals, agitated for 30 min at room temperature, and the absorbance (490 nm) was measured using a microplate reader (RT 2100C, Shenzhen, China). Cell viability measures were represented as a percentage (%).

Oxidative stress

NO production inhibitory activity

Nitrite (NO^{2-}) accumulation was measured to identify the NO concentrations in cell culture supernatants. In 24-well plates, Raw 264.7 cells were propagated at a density of 1×10^5 before being given the appropriate treatment according to the experimental groups. Following 24 h of incubation at 37 °C in an incubator with 5% CO_2, the cell supernatant was collected, and the NO expression level was assessed using the Griess method with a nitric oxide (NO) kit (MAK002, Sigma Aldrich, USA). In 96-well flat-bottomed microplates, 100 μL of Griess reagent was combined with an equal volume of the culture supernatant of different groups, shaken for 30 min, and incubated for 10 min at room temperature in dark conditions. A microtiter plate reader was used to measure the absorbance at 540 nm (Cui et al., 2019). A standard curve created by serially diluting $NaNO_2$ was used to calculate nitrite concentrations and expressed in μM.

Cellular ROS production

Cells seeded (2×10^5 cells/well) on the confocal dish were cultivated for 24 h at 37 °C. After washing the confocal plate with 100 μL of PBS, the growing media were taken out. After adding various extracts of different groups for 30 min, cells were exposed to DCFH-DA kit (35845, Sigma-Aldrich). DMEM (1:2,000) was used to dilute DCFH-DA reagent (10 mM) to 5 μM in the dark (Zhong et al., 2022). After discarding the cells, 1 mL of diluted DCFH-DA solution was added, and the mixture was incubated for 20 min at 37 °C, followed by two times washes by DMEM to eliminate DCFH-DA from cell surfaces. Cells were gathered into 1.5 mL tubes and suspended with 200 μL PBS to measure the cellular ROS production. Cellular esterases degraded DCFH-DA to DCFH carboxylate anion, which was further oxidized by ROS to produce extremely fluorescent dichlorofluorescein (DCF). The fluorescence intensity, which correlates to intracellular ROS levels, was measured using a microplate reader at an excitation wavelength of 485 nm and an emission wavelength of 535 nm (Peng et al., 2023).

Inflammatory markers

In a 24-well plate, RAW 264.7cells were seeded at a density of 3×10^5 cells/well and allowed to stabilize overnight. After 2 hours of treatment with the LPS (1 μg/mL), extracts were added for 12 h to cause inflammation (Kim et al., 2024). After centrifuging the culture at 13,500 rpm for 20 min, the supernatant were detected for the amounts of cytokine levels. The experiment was carried out in accordance with the instructions supplied by the ELISA kit manufacturer, and the supernatant was added to the corresponding ELISA kits for TNF-α, IL-6, and IL-1β (RAB0308, RAB0477, and RAB0304, Sigma-Aldrich, USA). Absorbance was determined at 450 nm using an ELISA microplate reader (BK EL10A, Shandong, China) and reported as pg./mL (Zhong et al., 2022).

Antioxidant activity assays

A selection of *in vitro* procedures was used to measure antioxidant activities. The (DPPH) assay was used to measure the samples' ability to scavenge free radicals utilizing a spectrophotometric technique at 517 nm in accordance with the manufacturer's instructions Kit (D9132, Sigma-Aldrich, USA). DPPH reacts with an antioxidant that can scavenge free radicals to form DPPHH. When coupled with a source of hydrogen atoms, DPPH creates diphenylpicrylhydrazine, which appears purple; however, the resultant DPPH has a reduced hydrogen content and is decolorized, appearing yellow (Kim et al., 2024).

The FRAP assay was used to measure the ferric reducing antioxidant power at a wavelength of 700 nm. The conversion of Fe^{3+} to Fe^{2+} was used to examine this reducing power, in which, depending on the sample's reduction power, the test solution's yellow color turns to different hues of green and blue (Liu et al., 2025).

The results were reported as μmol Fe^{2+} per gram of sample according to the described protocol by Koraqi et al. (2023). The (ABTS) assay is based on the theory that ferrylmyoglobin radicals are generated when ABTS is oxidized. Antioxidant reactions then bleach these radicals from blue to colorless (Kim et al., 2024). When ABTS is oxidized, ferrylmyoglobin radicals and ABTS cationic radicals are generated. Antioxidant reactions bleach radicals from blue to colorless (Kim et al., 2024). The manufacturer's instructions were followed while applying the ABTS assay at 750 nm. Kit (CS0790, Sigma-Aldrich, USA), which evaluates antioxidant activity in μmol Trolox per gram, was used to examine radical scavenging capability (Olennikov et al., 2022). Additionally, the hydroxyl radical scavenging activity was assessed, and the percentage of hydroxyl radical (•OH) inhibition was calculated using the relative drop in absorbance. The ability of strawberry extract to neutralize highly reactive •OH radicals is measured using the Fenton reaction-based •OH scavenging assay. By interacting with hydrogen peroxide (H_2O_2) with ferrous ions (Fe^{2+}), this mechanism produces hydroxyl radicals. These radicals and salicylic acid usually interact to form a colorful material that absorbs light at a wavelength of 510 nm (Chang et al., 2008). These tests provided a comprehensive assessment of the antioxidant properties of strawberries.

Determination of vitamins

Vitamin C

According to the manufacturer's instructions, a spectrophotometric assay at 534 nm was used to determine the ascorbic acid (Vitamin C) concentration (MAK505, Sigma-Aldrich, USA). After homogenizing 0.5 g of fresh strawberries in 1 mL of cooled extraction buffer, the samples were centrifuged at 10,000 rpm for 10 min at 4 °C (Duan et al., 2025). 280 μL of reagent (1), 100 μL of ethanol, 50 μL of agent (2B) (prepared by diluting 0.047 mL of reagent (2A) in 9.953 mL of ethanol), 100 μL of reagent (3) (dissolved in 13 mL of ethanol), and 50 μL of agent (4) were mixed with 20 μL of the sample. The results were given in mg/100 g.

Vitamin B9

Ethanol was used for the folate (Vitamin B9) determination with the homogenized fresh strawberry tissue at a 1:10 (w/v) ratio to help in the solubilization of folate. 0.1 M sodium phosphate and 1.0% (w/v) L-ascorbic acid were added to decrease oxidative loss of the vitamin and support folate stability during extraction. The mixtures were centrifuged (4,500 rpm, 4 °C, 30 min) and filtered by a 0.45 μm filter pore size (Qaderi et al., 2023). The resulting supernatant was spectrophotometrically analyzed at 280 nm, while findings were represented in μg/100 g.

Phytochemicals compositions

Total phenolics

The total phenolic contents were calculated using the Folin–Ciocalteu colorimetric technique (El-Hawary et al., 2021). Each extract (100 mL) was combined with 0.5 mL of Folin–Ciocalteu reagent and 7 mL of deionized water, then the mixture was incubated for 3 minutes at room temperature. After 1 h of reaction in the darkness, 1.5 mL of 20% Na_2CO_3 was combined in an equal ratio with water, and the absorbance (724 nm) was measured after incubation for 1 h. A standard material (gallic acid) was used to plot a standard curve, and the concentration was given as mg gallic acid equivalents (GAE) per 100 g.

Total flavonoids

Total flavonoids were measured by the aluminum chloride colorimetric assay in accordance with the manufacturer's instructions Kit KTB1530-96, Star Tech., Egypt. Fresh samples (0.1 g) were homogenized in 1.5 mL of 70% ethanol, extracted with shaking at 60 °C for 2 hours, and centrifuged at 10000 rpm for 10 minutes. 50 μL of the supernatant was combined with 15 μL of reagent (1) ($NaNO_2$ 5 g/100 mL) and incubated for 6 min at 25 °C. 30 μL of reagent (2) (Al (NO_3)3 10 g/100 mL) and 105 μL of reagent (3) (4 g/100 mL NaOH) were added. Total flavonoids were measured by using a spectrophotometric assay at 510 nm and reported as mg quercetin equivalents (QE) per 100 g (Chen et al., 2024).

Total anthocyanins

The pH differential approach was used to determine total anthocyanins (Duan et al., 2025). Fresh samples (0.05 g) were centrifuged (10,000 rpm, 10 min) after being homogenized in 1 mL extraction buffer and heated to 75 °C for 25 min while shaking. After combining 300 μL of pH 1.0 buffer reagent (1) or pH 4.5 buffer reagent (2) with the supernatant (100 μL), the mixture was incubated in the dark for 60 min at 25 °C. At 530 nm and 700 nm, absorbance was observed. The main red-pigmented, total anthocyanins were identified using spectrophotometry and reported as mg per 100 g.

Total catechins

A colorimetric technique was used for determining the total catechin concentration. One gram of fresh tissues was extracted using 10 mL of 70% ethanol while being constantly stirred at room temperature for 2 hours. After centrifuging the extract for 10 minutes at 6,000 rpm, the supernatant was recovered. 2.5 mL of concentrated hydrochloric acid and 2.5 mL of 0.1% vanillin in methanol were combined with an aliquot of 0.5 mL of the extract. A spectrophotometer was used at 500 nm after the reaction mixture was incubated at 30 °C for 20 min (Sun et al., 1998). A standard curve made with catechin was used to calculate the total catechin content. The extract's principal flavan-3-ols, total catechins, were measured and expressed on a mg per 100 g.

Total proanthocyanidins

The vanillin–HCl colorimetric technique was used to determine the total proanthocyanidin concentration. One gram of fresh tissues was extracted using 10 mL of 70% ethanol for 2 hours at room temperature while being constantly stirred. After centrifuging the extract for 10 minutes at 7,000 rpm, the supernatant was recovered. 0.5 mL of the extract was combined with 1.5 mL of concentrated hydrochloric acid and 3 mL of 4% vanillin in methanol. A spectrophotometer was used to measure the absorbance at 500 nm after the reaction mixture

was incubated for 20 min at 30 °C against catechin as a blank (Sun et al., 1998). Total proanthocyanidins, expressed as mg per 100 g, were used for evaluating polymerized flavonoids.

Total carotenoids

Five hundred milligrams of the fresh tissue was placed in a 50 mL centrifuge tube and mixed with 10 mL of ethanol for 1 minute using a vortex mixer. The supernatants were measured by adding 95% ethanol, and the final volume of the carotenoid extract was adjusted to 75 mL. The carotenoid extract's absorbance value at 450 nm was calculated (Piedad and Natividad, 2024). Total carotenoids, which were represented as mg per 100 g, were used to measure lipophilic pigments.

Enzyme activities

In order to measure the polyphenoloxidase (PPO) enzyme, around 1 g of fresh samples were weighed, homogenized in 101 mL of sodium phosphate buffer (100 mM, pH 7.0), centrifuged at 10,000 rpm, filtered, and stored at −18 °C. The extracts' ionically bound enzymes were detected at 410 nm after 0.5 mL of 0.175 mol/L catechol solution and 2.2 mol/L sodium phosphate buffer were added (Kou et al., 2019). Peroxidase (POD) enzyme was evaluated by combining 1 g of fresh samples with 3 mL of 0.1 mol/L pyrocatechol solution, centrifuging at 10,000 rpm, and filtering with a detector at 470 nm (Veljovic-Jovanovic et al., 2006).

Statistical analysis

Every biological replicate had measurements from technical triplicates, and the mean value was used for statistical analysis. Data is presented as mean ± standard deviation (SD). One-way analysis of variance (ANOVA) and Tukey's Honest Significant Difference (HSD) post-hoc test were used to compare the values from each group. SAS (version 8.02) was employed for statistical analysis. To analyze the data, GraphPad Prism was used. A p-value of less than 0.05 was considered to be significant.

Results and discussion

Strawberries are mostly eaten as a fruit or as part of processed meals, including juices, yogurts, jellies, and jams (Villamil-Galindo et al., 2020; Abdussameea et al., 2023). Leaves, roots, seeds, and calyces are reported to be produced in large quantities during this industrial processing; these byproducts are usually disposed of as biomass wastes (Ronie et al., 2024). These byproducts have significant concentrations of phenolic substances, anti-inflammatory, anti-oxidant, and can be exploited to reduce ultimate disposal expenses as well as get raw materials at a reasonable cost (Shahbaz et al., 2025).

Cell viability

The effects of quercetin and strawberry components on the viability of LPS-activated RAW 264.7 macrophages were examined. The MTT assay, based on the dehydrogenases in the mitochondria of living cells, convert tetrazolium salts to insoluble formazan (in color purple), is a widely used technique to measure cell viability. After dissolving the reduced formazan in organic solvents, the absorbance can be quantified (Abdussameea et al., 2023). LPS treatment alone decreased cell viability to 89.64%, indicating an insignificant cytotoxic effect. The viability of LPS-stimulated cells was reduced when strawberry components were co-treated. The viabilities of cells treated with LPS + pulp, LPS + peel, LPS + seed, and LPS + whole fruit were 87.54%, 84.67%, 86.02%, and 85.17%, respectively. In comparison to the LPS group, the peel caused the biggest reduction in viability. Quercetin treatment tended to maintain cell viability; the LPS + quercetin group demonstrated 94.88% viability, suggesting a protective effect against the LPS-induced cytotoxicity (Figure 2).

LPS exposure decreased the survival of RAW 264.7 macrophages, which is in line with its established cytotoxic and pro-inflammatory effects on immune cells. Macrophages are crucial for innate immunity, and tissue healing and infection control depend on their recruitment to infection or damage sites. On the other hand, uncontrolled monocyte accumulation may result in the development of malignancy or even persistent inflammation (Cui et al., 2019). A study for measuring anti-inflammatory and antioxidant approaches was provided by the slight reduction observed, which indicated that the selected LPS concentration successfully stimulated macrophages without generating significant cytotoxicity (Peng et al., 2023).

Despite the effects of LPS alone, treatments with strawberry parts or whole fruit extracts marginally reduced cell viability (Kim et al., 2024). These slight decreases indicated that some phytochemical components in strawberry parts might have extract-specific or dose-dependent impacts on metabolic function due to its larger concentration of phenolics or other bioactive substances that alter mitochondrial activity, the peel produced the largest drop. However, viability maintained above 80%, suggesting that none of the strawberry samples showed overt cytotoxicity.

When compared to LPS alone, quercetin showed the best results by increasing cell viability. This protective effect was consistent with earlier research demonstrating that quercetin improves cellular metabolic function and reduces oxidative stress caused by LPS (Cui et al., 2019). Quercetin may have maintained mitochondrial function and minimized the harmful consequences of inflammatory activation, based on the increased viability shown in the treated cells (Peng et al.,

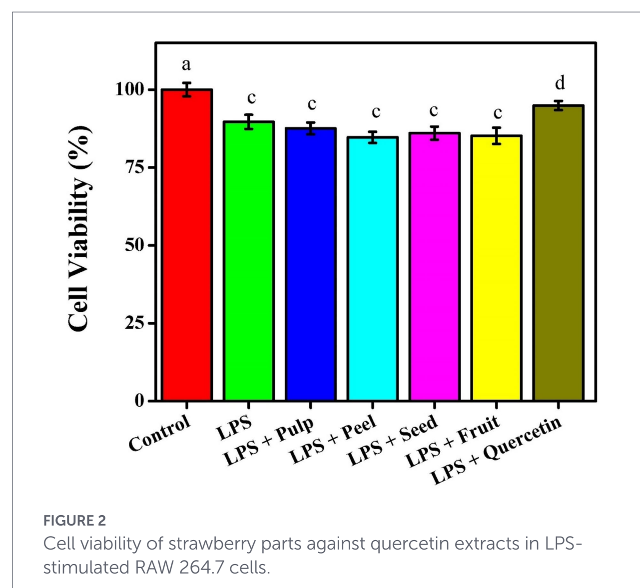

FIGURE 2
Cell viability of strawberry parts against quercetin extracts in LPS-stimulated RAW 264.7 cells.

2023). Across a number of pathways, including NF-κB, signal transducer and activator of transcription 1 (STAT-1), and Syk/Src/IRAK-1, quercetin inhibits the LPS-induced inflammatory response (Yang et al., 2014; Nga et al., 2020). The cell viability was ≥ 84% for all groups, representing that the detected antioxidant and anti-inflammatory properties were only not because of non-specific cytotoxicity nevertheless slightly to the biological activities of the strawberry fruit extracts.

Oxidative stress

NO is a type of free radical that controls physical processes, however an excessive amount of NO produced by Inducible nitric oxide synthase (iNOS) can cause inflammation, cytotoxicity, and autoimmune diseases (Zhong et al., 2022). The Griess assay in this work suggested that when LPS was triggered, the overproduction of NO was linked to the overexpression of iNOS (Malayil et al., 2022), which converts the amino acid L-arginine into NO (Kim et al., 2024). Tumor development and inflammation can result from prolonged NO generation. Numerous cell types, including primary aortic smooth muscle and epithelial cells, are stimulated to migrate by NO release (Cui et al., 2019). Inflammatory infections were significantly influenced by oxidative stress, with ROS levels acting as a key mediator. The human body's oxidant/antioxidant balance is upset by excessive ROS generation, which damages DNA, lipids, and proteins (Wang et al., 2021).

When compared to the control group, LPS stimulation significantly raised NO levels. LPS-treated cells showed a significant increase to 44.15 μM, indicating effective inflammatory activation, while control cells produced 6.25 μM. Treatment with strawberry parts considerably decreased NO production; the peel group caused the greatest reduction, lowering NO down to 18.23 μM. This was followed by the pulp (28.47 μM), the seed (23.8 μM), and the whole fruit (19.07 μM). As a positive control, quercetin significantly reduced NO release to 11.78 μM, which is close to baseline values (Figure 3).

Similar to the results of NO production, ROS levels improved significantly following LPS stimulation, from 0.11 μM in control cells to 0.51 μM in the LPS group. ROS accumulation was successfully reduced by the treatment with strawberry parts. While the whole fruit (0.21 μM) and seed (0.25 μM) showed similar reductions, the peel group once again caused one of the largest reductions, lowering ROS down to 0.17 μM. The pulp reduced ROS to 0.33 μM, which was a milder effect. As an active antioxidant reference, quercetin reduced ROS levels to 0.15 μM (Figure 4).

Both NO and ROS levels were significantly increased by LPS stimulation, which is in line with the activation of macrophage inflammatory pathways. Treatment with strawberry samples, however, considerably reduced these reactions, suggesting that sustainable strawberry by-products could be useful sources of bioactive substances. Flavonols inhibited NO production, which was very consistent with iNOS protein down-regulation (Zhang et al., 2020).

The peel continuously demonstrated the best inhibitory properties on the production of both NO and ROS among the studied groups. This finding was consistent with earlier research indicating that, in comparison to edible pulp tissues, fruit peels frequently have higher quantities of phenolics, flavonoids, and other antioxidant metabolites. Significant decreases were observed as well in the whole fruit and seed groups, indicating that the strawberry's many parts contribute bioactive chemicals that can alter oxidative pathways. Results were in arrangement with the investigation on LPS-stimulated RAW264.7cells, strawberries have an anti-inflammatory impact by lowering NO generation and blocking inflammatory mediators and cytokines (Gu et al., 2020). Strawberry extract demonstrated anti-inflammatory properties, lowering intracellular generation of NO and ROS as well as inflammatory indicators by activating the nuclear factor erythroid 2–2-related factor 2 (Nrf2) and NF-κB signaling pathways after *in vitro* LPS stimulation (Gasparrini et al., 2017). Quercetin suppressed NO and ROS productions. However, the activity of quercetin was approached by the peel and whole-fruit extracts, indicating that natural extracts of strawberry phytochemicals might have synergistic effects. Quercetin may prevent LPS-stimulated macrophages from migrating by inhibiting NO release (Cui et al., 2019).

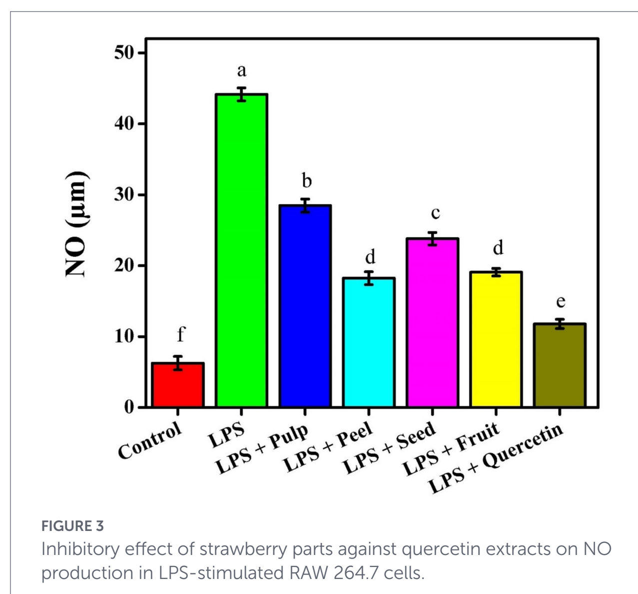

FIGURE 3
Inhibitory effect of strawberry parts against quercetin extracts on NO production in LPS-stimulated RAW 264.7 cells.

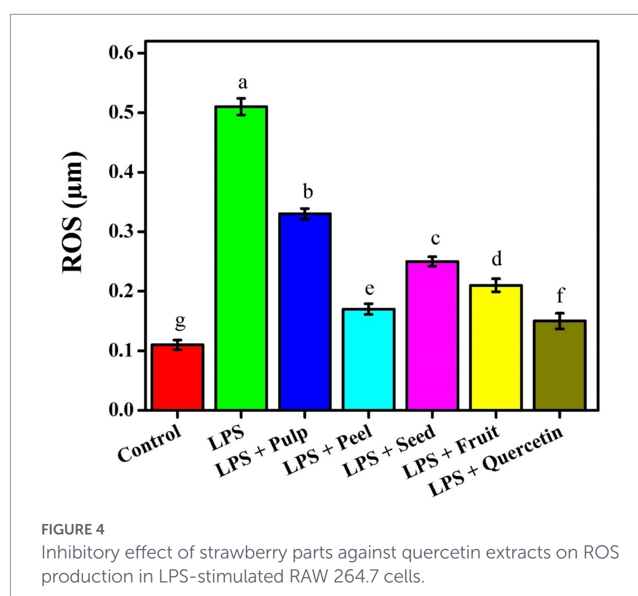

FIGURE 4
Inhibitory effect of strawberry parts against quercetin extracts on ROS production in LPS-stimulated RAW 264.7 cells.

Pro-inflammatory cytokine production

A physiological defensive mechanism, inflammation is triggered by an external stimulus as LPS. Inflammatory proteins are overexpressed and a variety of immune defense cells, including macrophages. Uncontrolled and overexpressed pro-inflammatory cytokines lead to

inflammation, which can result in a number of illnesses, including tissue damage and chronic disease degeneration (Kim et al., 2024).

LPS stimulation significantly increased the release of inflammatory cytokines. After LPS exposure, TNF-α levels raised from 28.25 pg./mL in control cells to 277.68 pg./mL. This rise was considerably reduced by treating with strawberry samples. The peel group caused the most suppression, lowering TNF-α to 117.04 pg./mL, followed by the pulp (191.21 pg./mL), whole fruit (122.09 pg./mL), and seed (144.61 pg./mL). TNF-α was lowered to 102.85 pg./mL using quercetin as a positive anti-inflammatory reference (Figure 5).

For IL-6, a similar pattern was observed. In control cells, LPS treatment raised IL-6 from 24.55 pg./mL to 478.05 pg./mL. IL-6 was reduced to 267.31 pg./mL (pulp), 160.47 pg./mL (peel), 213.48 pg./mL (seed), and 182.3 pg./mL (fruit). The greatest decrease was caused by quercetin, which reduced IL-6 to 122.11 pg./mL.

The same pattern was observed in IL-1β, which increased with LPS from 14.59 pg./mL in control macrophages to 198.67 pg./mL. The whole fruit (83.14 pg./mL), pulp (111.58 pg./mL), and seed (93.17 pg./mL) all markedly reduced IL-1β production, however the peel demonstrated the greatest inhibition among strawberry samples, lowering IL-1β to 88.2 pg./mL. Quercetin demonstrated a strong anti-inflammatory impact by lowering IL-1β to 55.7 pg./mL. These cytokines were successfully decreased by treatment with various strawberry anatomical portions, suggesting that the fruit's by-products had potent medicinal properties. The peel consistently showed the highest inhibitory effects among the studied groups, with the whole fruit and seed samples coming in second and third. Strawberry peels might have higher levels of bioactive substances that can reduce inflammation, including flavonoids, phenolic acids, and anthocyanins. These substances are known to prevent LPS from activating the NF-κB pathway, which controls the inflammatory markers. Therefore, suppression of NF-κB signaling and downstream inflammatory gene expression was probably the cause of the observed reductions in cytokine levels. The substantial activity of the whole fruit group supports the anti-inflammatory efficacy of the pulp, peel, and seed samples. The pulp exhibited the least amount of suppression since edible parts often contain lower levels of defense-related phytochemicals. Berries' ellagitannins have been established to have antibacterial and anti-inflammatory properties. According to both *in-vivo* and *in-vitro* research, strawberries have anticancer, anti-inflammatory, neuroprotective, cardiovascular, and antioxidant effects (Martinelli et al., 2025). In the clinical investigation, strawberries also dramatically decreased the biomarkers linked to inflammation. The anti-inflammatory action was observed in serum obtained from animals fed diets supplemented with strawberries (Schell et al., 2017). Strawberry-derived pelargonidin-3-O-glucoside, ellagic acids, and phenolic extracts similarly reduced the expression of inflammatory markers and the synthesis of NO, TNF-α, and IL-6 (Gu et al., 2020; Duarte et al., 2018). It was discovered that the calyx portion's 70% ethanol extract had inhibitory effects on NF-κB p65 subunit (p-p65) and phosphorylated IκBα (p-IκB), demonstrating its influence on the NF-κB signaling pathway (Kim et al., 2024; Zhong et al., 2022). As the positive control, quercetin induced the strongest reduction of all three cytokines, which is consistent with its known ability to inhibit NF-κB and lower the formation of NO and ROS (Xue et al., 2017; Peng et al., 2023). The strawberry peel and whole-fruit samples showed possibilities as sustainable, natural anti-inflammatory resources by coming within range of quercetin's efficacy. These findings provided evidence for the potential value-adding of strawberry waste products, particularly the peel as useful component for oxidative stress and inflammation management. The strawberry peel extract had a targeted anti-inflammatory impact rather than a non-specific cytotoxic reaction by the strong suppression of pro-inflammatory cytokines, which greatly exceeded the marginal 15% loss in cell viability.

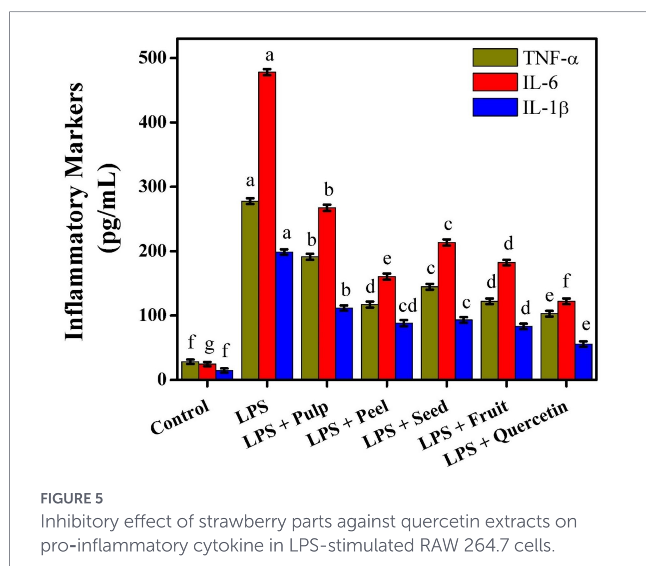

FIGURE 5
Inhibitory effect of strawberry parts against quercetin extracts on pro-inflammatory cytokine in LPS-stimulated RAW 264.7 cells.

Antioxidant activities

The antioxidant capacities of the strawberry pulp, peel, seed, and whole-fruit extracts were assessed using DPPH, FRAP, ABTS, and hydroxyl (•OH) radical scavenging techniques. The fruit's anatomical components affected the oxidation levels (Duan et al., 2025).

The strongest antioxidant effectiveness was shown by the peel extract. It showed the lowest DPPH IC_{50} value (31.2 μg/mL), indicating strong free-radical scavenging action, and the greatest FRAP value (133.18 μmol Fe^{2+}/g), indicating superior ferric-reducing capability. Furthermore, the peel consistently gave the highest percentage of hydroxyl radical scavenging (65.96%) and the best ABTS scavenging ability (156.05 μmol Trolox/g) (Figure 6). The seed extract showed the second-highest antioxidant effectiveness, with a DPPH IC_{50} of 46.17 μg/mL, FRAP of 102.16 μmol Fe^{2+}/g, ABTS of 112.44 μmol Trolox/g, and •OH scavenging of 56.08%. The whole-fruit extract also showed modest antioxidant activity, with an ABTS of 98.87 μmol Trolox/g, FRAP of 89.48 μmol Fe^{2+}/g, DPPH IC_{50} of 32.28 μg/mL, and •OH scavenging of 48.29%. The aqueous extract of strawberry fruits had FRAP values of 72.40 EqmM $FeSO_4$/100 g, according to Ganhão et al. (2019); our results were the best in this antioxidant test. The pulp extract consistently showed the weakest antioxidant capacity, with the greatest DPPH IC_{50} value (88.15 μg/mL), lowest FRAP value (58.3 μmol Fe^{2+}/g), lowest ABTS activity (71.25 μmol Trolox/g), and lowest hydroxyl radical scavenging activity (33.14%). The investigation of strawberry fruit extract's capacity to scavenge DPPH radicals revealed a similar pattern (Zitouni et al., 2020). Results for the whole fruit by •OH scavenging was higher than those by Milosavljević et al. (2022) estimated the extracts by using the spin-trapping assay in conjunction with electron paramagnetic resonance (EPR) spectroscopy.

The bioactivity of the different strawberry parts varied significantly, according to the antioxidant activities, the peel and seed parts' strong antioxidant potential indicated that these by-products might be crucial in controlling oxidative stress in biological systems. The

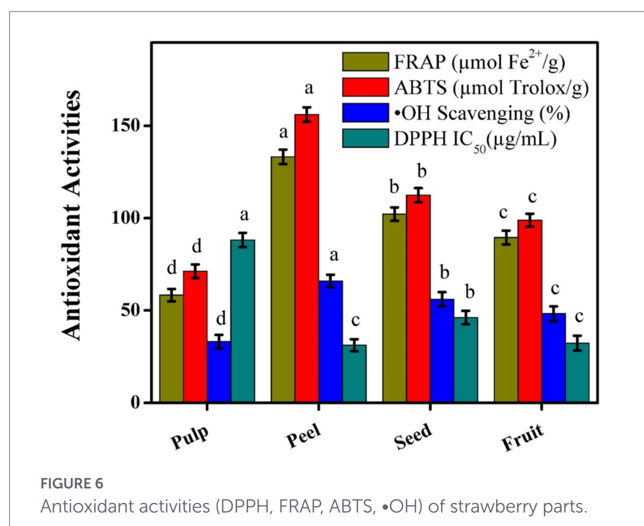

FIGURE 6
Antioxidant activities (DPPH, FRAP, ABTS, •OH) of strawberry parts.

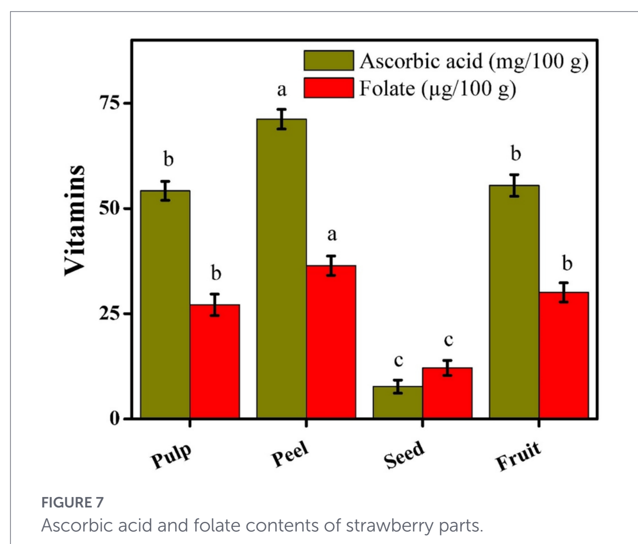

FIGURE 7
Ascorbic acid and folate contents of strawberry parts.

oxidative-stress findings from LPS-activated RAW 264.7 macrophages, which showed that the peel and whole-fruit samples produced the most significant decreases in NO and ROS levels. The samples' protective benefits during inflammatory activation were probably influenced by their antioxidant qualities. The pulp extract, showed minimal antioxidant activity, which is in line with other research (Abbasi et al., 2015), demonstrating that edible fruit parts frequently have lower amounts of beneficial phytochemicals than the peel and seed. When compared to the pulp, the whole-fruit extract's moderate activity indicated synergistic interactions between the various fruit tissues, which might improve the total bioactivity. The possibility of sustainable strawberry by-products, especially the peel and seed, as important sources of natural antioxidants, was generally validated by the antioxidant studies. These characteristics may aid in reducing oxidative and inflammatory stress, providing a mechanistic understanding of how these extracts enhanced redox balance in LPS-stimulated macrophages and inhibited cytokine production.

Vitamins

The strawberry is regarded as a functional fruit and one of the most significant sources of vitamins (Milosavljević et al., 2022; Giampieri et al., 2015). The various strawberry parts had significantly differing vitamin contents. The peel extract has the highest ascorbic acid content (71.22 mg/100 g), followed by the pulp (54.2 mg/100 g) and the whole fruit (55.47 mg/100 g). Ascorbic acid concentration in the seed were the lowest, at 7.68 mg/100 g (Figure 7). Results for the whole fruit by vitamin C was in agreement with those by Olennikov et al. (2022), which ranged from 25 to 112 mg/100 g for strawberry extracts.

For folate content, a similar pattern was noted. The peel extract (36.41 μg/100 g) showed higher folate contents than the whole fruit (30.06 μg/100 g) and pulp (27.11 μg/100 g). The lowest folate content (12.12 μg/100 g) was reported in the seed sample.

The peel consistently contained the highest quantities of these vitamins when compared to the other portions of the strawberry, while the seed had far lower concentrations. These results were consistent with earlier studies showing that fruit peels, which protect the fruit from oxidative and environmental stress, typically have higher concentrations of vitamins than edible tissues (Abbasi et al., 2015). The peel's improved antioxidant efficacy in DPPH, FRAP, ABTS, and hydroxyl radical tests was further enhanced by its increased vitamin content. While folate has a role in redox equilibrium and cellular metabolism, ascorbic acid is a powerful free-radical scavenger that frequently adds to total antioxidant capacity (Qaderi et al., 2023). Thus, the decreases in ROS and NO levels observed during LPS-induced macrophage activation were probably caused by the peel's enhanced vitamin profile. Concentration of vitamin C has been demonstrated to be an efficient promoter of the oxidative regulation of the strawberry sample (Duan et al., 2025). The seed sample showed the lowest amounts of both vitamins, which was consistent with its comparatively lower antioxidant activity as compared to the peel. The fruit's activity may have been increased by the synergistic interactions between its constituent tissues, despite the fruit's overall mild vitamin concentration. In this regard, fluctuations in the vitamin C concentration of strawberries may have an impact on the folate value since a higher vitamin C value may result in greater folate stability. The same parameters affected the retention of ascorbic acid and folate, and a high ascorbic acid level may offer protection against folate breakdown (Strålsjö et al., 2003). Ringling and Rychlik (2017), who conducted *in vivo* experiments to mimic dietary folate digestion. Vitamin C stabilizes folate, especially 5-CH3-H4 folate, throughout the digestion. The stability of different types of folate was enhanced by adding vitamin C at physiological levels, depending on the dietary matrix. The findings showed that the peel sample contained the highest levels of folate and vitamin C, indicating that this by-product is a more nutritious source than the pulp, seed, or whole-fruit extracts. The vitamin analysis generally corroborated the significance of strawberry by-products, particularly the peel, as nutrient-dense and bioactive components that can enhance anti-inflammatory and antioxidant effects in macrophages.

Phytochemical composition

Strawberry components have a wide range of phytochemical compositions, with non-edible tissues as the peel and seed typically containing larger concentrations of advantageous compounds such as phenolics, flavonoids, and anthocyanins than the pulp. These compounds increase its potential medical use in reducing oxidative stress and inflammation (Villamil-Galindo et al., 2021).

The pulp, peel, seed, and whole-fruit extracts had considerably varied phytochemical profiles. Total phenolics (159.16 mg GAE/100 g)

and total flavonoids (44.16 mg QE/100 g) were highest in the peel. The seed (88.17 mg GAE/100 g phenolics; 20.03 mg QE/100 g flavonoids) and the whole fruit (102.25 mg GAE/100 g phenolics; 25.77 mg QE/100 g flavonoids) followed the second and the third order. Phenolic (51.2 mg GAE/100 g) and flavonoid (14.25 mg QE/100 g) values were lowest in the pulp. The highest concentration of anthocyanins was found in the peel (33.08 mg/100 g), which was followed by the pulp (9.15 mg/100 g) and the whole fruit (11.13 mg/100 g). The lowest concentration was found in the seed extract (2.88 mg/100 g). The seed sample has the greatest proanthocyanidins (28.92 mg/100 g), followed by the pulp (2.47 mg/100 g), peel (4.59 mg/100 g), and the whole fruit (16.27 mg/100 g). The peel has the highest amount of catechins (27.16 mg/100 g) and carotenoids (5.28 mg/100 g). The pulp and seed extracts had lower levels of carotenoids (3.77 mg/100 g) and catechins (14.35 mg/100 g) than the whole fruit (Figures 8–10).

Significant variations of bioactive compounds among the strawberry parts were identified by the phytochemical investigations. The peel sample showed the highest abundance of phenolic, flavonoid, anthocyanin, carotenoid, and catechin components, demonstrating that non-edible fruit tissues are rich repositories of secondary metabolites (Abbasi et al., 2015). Taxifolin 3-O-a-Larabinofuranoside, polyphenolic acids, fupenzic acids, and sericic acids were found in strawberry leaves (El-Hawary et al., 2021); therefore, the potential use of leaves as a source of nutrition and a component of functional food items (Raudoniūtė et al., 2011). The peel contains the largest concentrations of anthocyanins and catechins, which are known to modulate signaling pathways including NF-κB and Nrf2. Their presence in the peel may explain its better capacity to reduce the inflammatory markers (Kim et al., 2024). The peel's bioactivity was further supported by its high phenolic and flavonoid contents, which are known to neutralize reactive species and suppress inflammatory mediators. Additionally, the seed samples showed significant phytochemical quantity, especially in proanthocyanidins. The seed's strong proanthocyanidin content may have contributed to its moderate suppression of NO, ROS, and cytokines, even though it had lower quantities of anthocyanins and carotenoids than the peel. The majority of phytochemicals showed intermediate quantities in the whole-fruit extract, indicating a synergistic or even cumulative action of the parts. This could allow for the fruit's comparatively high bioactivity even though it contains fewer specific phytochemicals than the peel (Duan et al., 2025; Qaderi et al., 2023). The pulp had the lowest concentrations of almost phytochemical categories, which is in line with its lower antioxidant activity and diminished capacity to influence inflammatory markers in RAW 264.7 macrophages.

Enzyme activities

PPO and POD enzymes are essential for the 'enzymatic tanning' for fruits, that lead to the quick breakdown of nutritious and morphological structure and severely reduces the shelf-life and acceptance by customers (Wesche-Ebeling and Montgomery, 1990).

The activities were evaluated in the pulp, peel, seed, and the whole fruit extracts of strawberry. POD activity varies among the anatomical fruit parts. The peel demonstrated the highest POD activity (3.11 U/mL), followed by the pulp (2.66 U/mL) and whole fruit (2.64 U/mL), while the seed showed the lowest activity (1.89 U/mL) (Figure 11).

PPO activity differed between the strawberry parts. The peel displayed the highest PPO activity (3.99 U/mL), followed by the whole fruit (3.95 U/mL) and seed (3.54 U/mL). The pulp has the lowest PPO activity (1.71 U/mL).

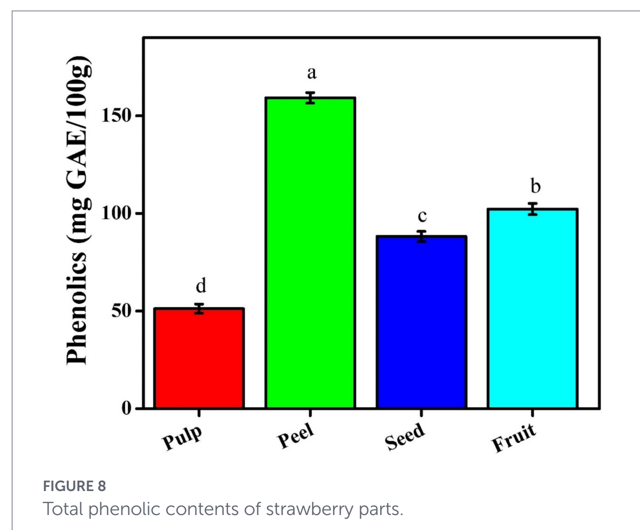

FIGURE 8
Total phenolic contents of strawberry parts.

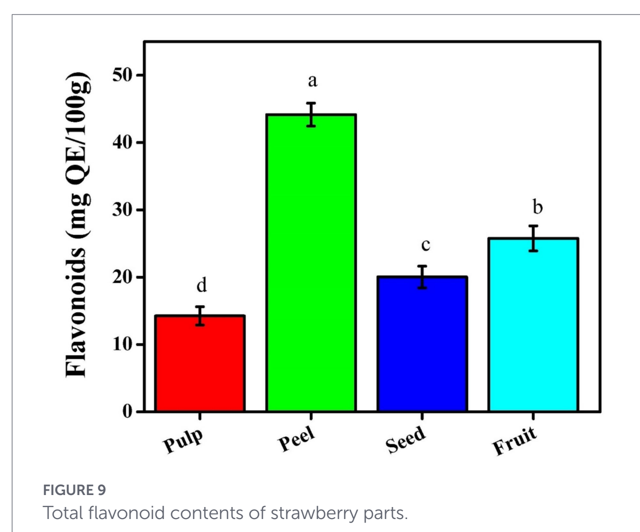

FIGURE 9
Total flavonoid contents of strawberry parts.

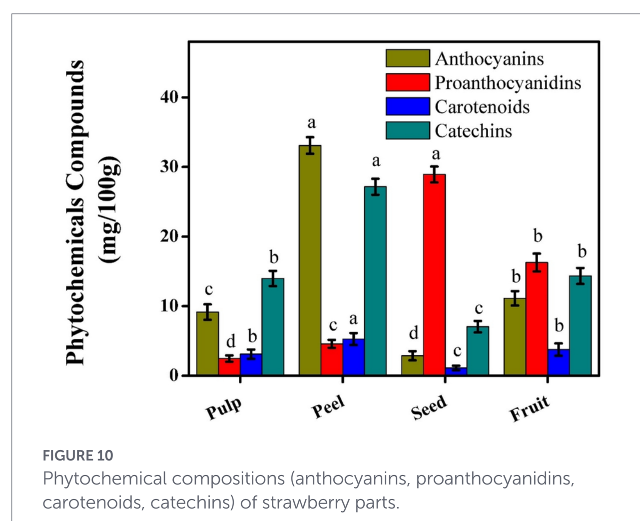

FIGURE 10
Phytochemical compositions (anthocyanins, proanthocyanidins, carotenoids, catechins) of strawberry parts.

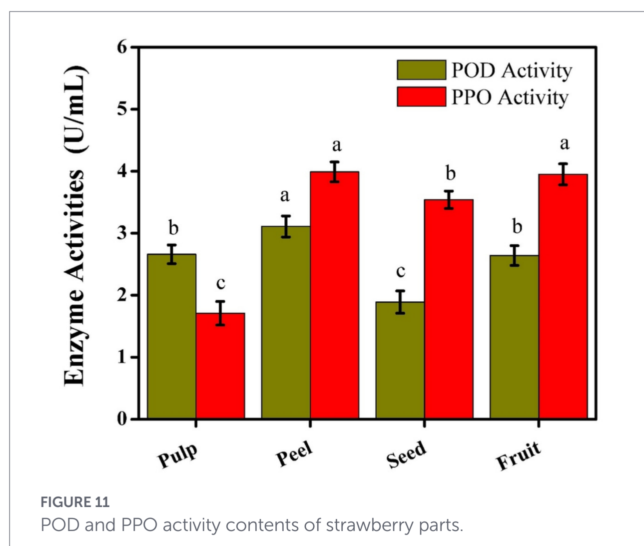

FIGURE 11
POD and PPO activity contents of strawberry parts.

According to these findings, the strawberry peel was the most effective enzymatic source for both POD and PPO activities among the portions under investigation. The enzyme activities found in the peel were consistent with the high metabolic and protective roles of strawberry samples, which typically retain larger levels of phenolic compounds and oxidative enzymes (El-Hawary et al., 2021). The higher enzyme levels in the peel may suggest the tissue's resistance against environmental stressors because these enzymes are involved in phenolic metabolism and redox regulation. The pulp and seed displayed comparatively lower activity, which suggested that these tissues had less potential for oxidative enzymes. Specifically, the pulp's low PPO activity showed a decreased potential for phenolic oxidation, which could be linked to its softer physiology and less sensitivity to oxidative stress (Kim et al., 2024). Perhaps as a result of the peel's substantial involvement, the whole fruit had an integrated enzymatic profile with values closer to the peel. It's possible that the peel's elevated POD and PPO activities affected the oxidative responses of macrophages or enhanced the redox-modulating effects of quercetin. The discovery of strawberry peel showed a tissue that metabolized with potentially improved functional capabilities connected with oxidative stress regulation was corroborated by the patterns of enzyme activity (Milosavljević et al., 2022).

Conclusion

The research focused on the opportunity to use sustainable strawberry by-products for some anti-inflammatory and antioxidant activities in lipopolysaccharide (LPS)-induced RAW 264.7 macrophages against quercetin. Peels are concentrated sources of bioactive phenols and vitamins that have strong anti-inflammatory and antioxidant properties. This makes them a high-value, sustainable substitute for the nutraceutical industry and a practical way to cut down on agro-industrial waste. In terms of antioxidants, anti-inflammatory properties, and bioactive components, strawberry peel outperformed pulp and seed. These results revealed that strawberry peel, instead of being disposed of as wastes, can be considered as a sustainable resource for functional foods, supplements, or other goods with added value.

Limitations

Recognizing the limitations of the current study is crucial. Crude extracts at fixed mass-based concentrations (100 μg/mL) were used in the biological tests. These concentration gradients may have an impact on the observed variations in anti-inflammatory and antioxidant activities since the phenolic and flavonoid density differs greatly between the peel and other fractions. The intrinsic potency of individual chemical ingredients is not isolated by this technology, despite its effectiveness in finding the most potent industrial by-product overall. Deeper understanding of the precise chemical causes of the noted therapeutic effects might be possible with future study concentrating on the standardization of extracts based on particular markers (such as catechin or anthocyanin concentration).

Data availability statement

The original contributions presented in the study are included in the article/supplementary material, further inquiries can be directed to the corresponding author.

Author contributions

RJ: Methodology, Writing – original draft, Software. MA: Investigation, Methodology, Writing – original draft. HAA: Methodology, Software, Writing – review & editing. NKA: Investigation, Methodology, Writing – review & editing. AF: Data curation, Methodology, Writing – original draft. MJ: Data curation, Methodology, Writing – review & editing. HH: Formal analysis, Methodology, Writing – original draft. RS: Methodology, Supervision, Writing – original draft. HW: Methodology, Project administration, Writing – original draft. MH: Methodology, Visualization, Writing – review & editing. DA: Funding acquisition, Methodology, Writing – original draft. HFA: Methodology, Resources, Writing – review & editing. RMA: Methodology, Resources, Writing – review & editing. RA: Methodology, Validation, Writing – original draft. NHJA: Formal analysis, Methodology, Writing – review & editing.

Funding

The author(s) declare that financial support was not received for the research and/or publication of this article.

Acknowledgments

The authors would like to acknowledge the Deanship of Graduate Studies and Scientific Research, Taif University for funding this work.

Conflict of interest

The author(s) declared that this work was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Generative AI statement

The author(s) declared that Generative AI was not used in the creation of this manuscript.

Any alternative text (alt text) provided alongside figures in this article has been generated by Frontiers with the support of artificial intelligence and reasonable efforts have been made to ensure accuracy, including review by the authors wherever possible. If you identify any issues, please contact us.

Publisher's note

All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

References

Abbasi, A. M., Guo, X., Fu, X., Zhou, L., Chen, Y., Zhu, Y., et al. (2015). Comparative assessment of phenolic content and in vitro antioxidant capacity in the pulp and peel of mango cultivars. *Int. J. Mol. Sci.* 16:13507. doi: 10.3390/ijms160613507

Abdussameea, Ibrahim, M. S., Amira, R. M., Hassanb, I., Mudasirali, Zahoor, Z., et al. (2023). A comprehensive review on the health-orientated aspects of strawberries. *Food Sci. Appl. Microbiol. Rep.* 2, 1–7. doi: 10.61363/fsamr.v2i1.57

Bahuguna, A., Bharadwaj, S., Chauhan, A. K., and Kang, S. C. (2020). Inhibitory insights of strawberry (Fragaria × ananassa var. Seolhyang) root extract on tyrosinase activity using computational and in vitro analysis. *Int. J. Biol. Macromol.* 165, 2773–2788. doi: 10.1016/j.ijbiomac.2020.10.135

Chang, C.-Y., Hsieh, Y.-H., Cheng, K.-Y., et al. (2008). Effect of pH on Fenton process using estimation of hydroxyl radical with salicylic acid as trapping reagent. *Water Sci. Technol.* 58, 873–879. doi: 10.2166/wst.2008.429

Chen, S., Wang, X., Cheng, Y., Gao, H., and Chen, X. (2024). Effects of supplemental lighting on flavonoid and anthocyanin biosynthesis in strawberry flesh revealed via metabolome and transcriptome co-analysis. *Plants* 13:1070. doi: 10.3390/plants13081070

Cui, S., Wu, Q., Wang, J., et al. (2019). Quercetin inhibits LPS-induced macrophage migration by suppressing the iNOS/FAK/paxillin pathway and modulating the cytoskeleton. *Cell Adhes. Migr.* 13, 1–12. doi: 10.1080/19336918.2018.1486142

Duan, M., Jiang, T., Wang, X., Song, K., Xiao, X., Fu, X., et al. (2025). LC-MS/MS reveals variation in bioactive flavonoid profiles and MYB regulatory networks in wild vs. cultivated strawberries (white, pink, red). *LWT* 231:118319. doi: 10.1016/j.lwt.2025.118319

Duarte, L. J., Chaves, V. C., Nascimento, M. V. P. S., Calvete, E., Li, M., Ciraolo, E., et al. (2018). Molecular mechanism of action of Pelargonidin-3-O-glucoside, the main anthocyanin responsible for the anti-inflammatory effect of strawberry fruits. *Food Chem.* 247, 56–65. doi: 10.1016/j.foodchem.2017.12.015

El-Feky, A. M., and El-Rashedy, A. A. (2023). Sterols and flavonoids in strawberry calyx with free radical scavenging, anti-inflammatory, and molecular dynamic study. *Beni-Suef Univ. J. Basic Appl. Sci.* 12:108. doi: 10.1186/s43088-023-00445-x

El-Hawary, S. S., Mohammed, R., El-Din, M. E., Hassan, H. M., Ali, Z. Y., Rateb, M. E., et al. (2021). Comparative phytochemical analysis of five Egyptian strawberry cultivars (*Fragaria× ananassa* Duch.) and antidiabetic potential of festival and red Merlin cultivars. *RSC Adv.* 11, 16755–16767. doi: 10.1039/d0ra10748d 35479159

Ganhão, R., Pinheiro, J., Tino, C., Faria, H., and Gil, M. M. (2019). Characterization of nutritional, physicochemical, and phytochemical composition and antioxidant capacity of three strawberry "Fragaria × ananassa Duch." cultivars ("Primoris", "endurance", and "Portola") from Western region of Portugal. *Foods* 8:682. doi: 10.3390/foods8120682

Gasparrini, M., Forbes-Hernandez, T. Y., Giampieri, F., Afrin, S., Alvarez-Suarez, J. M., Mazzoni, L., et al. (2017). Anti-inflammatory effect of strawberry extract against LPS-induced stress in RAW 264.7 macrophages. *Food Chem. Toxicol.* 102, 1–10. doi: 10.1016/j.fct.2017.01.018

Giampieri, F., Forbes-Hernandez, T. Y., Gasparrini, M., Alvarez-Suarez, J. M., Afrin, S., Bompadre, S., et al. (2015). Strawberry as a health promoter: an evidence based review. *Food Funct.* 6, 1386–1398. doi: 10.1039/c5fo00147a

González-Domínguez, R., Sayago, A., Akhatou, I., and Fernández-Recamales, Á. (2020). Volatile profiling of strawberry fruits cultivated in a soilless system to investigate cultivar-dependent chemical descriptors. *Foods* 9:768. doi: 10.3390/foods9060768

Gu, I., Brownmiller, C., Stebbins, N. B., Mauromoustakos, A., Howard, L., and Lee, S.-O. (2020). Berry phenolic and volatile extracts inhibit pro-inflammatory cytokine secretion in LPS-stimulated RAW264.7 cells through suppression of NF-κB signaling pathway. *Antioxidants* 9:871. doi: 10.3390/antiox9090871

Justine, E. E., Jang, S.-W., Lee, S., Lee, H. J., Wang, R. B., and Kim, Y. J. (2024). Comparative anti-inflammatory effect of extract from novel Korean strawberry cultivars (Fragaria × ananassa) on lipopolysaccharide-induced RAW264.7 macrophages and mouse model. *J. Sci. Food Agric.* 104, 2272–2283. doi: 10.1002/jsfa.13115

Kim, H. M., Yoo, D. H., Kang, J. W., Lee, I. C., and Bae, J. S. (2024). Anti-inflammatory effect of extract from Fragaria ananassa Duch. Calyx via MAPK and NF-κB signaling pathway. *J. Microbiol. Biotechnol.* 34, 2662–2674. doi: 10.4014/jmb.2409.09044

Koraqi, H., Petkoska, A. T., Khalid, W., Sehrish, A., Ambreen, S., and Lorenzo, J. M. (2023). Optimization of the extraction conditions of antioxidant phenolic compounds from strawberry fruits (*Fragaria x ananassa* Duch.) using response surface methodology. *Food Anal. Methods* 16, 1030–1042. doi: 10.1007/s12161-023-02469-6

Kou, X., He, Y., Li, Y., Chen, X., Feng, Y., and Xue, Z. (2019). Effect of abscisic acid (ABA) and chitosan/nano-silica/sodium alginate composite film on the color development and quality of postharvest Chinese winter jujube (*Zizyphus jujuba* mill. cv. Dongzao). *Food Chem.* 270, 385–394. doi: 10.1016/j.foodchem.2018.06.151

Lin, D., Ma, Q., Zhang, Y., and Peng, Z. (2020). Phenolic compounds with antioxidant activity from strawberry leaves: a study on microwave-assisted extraction optimization. *Prep. Biochem. Biotechnol.* 50, 874–882. doi: 10.1080/10826068.2020.1762213

Liu, H., Ji, Z., Pöhnl, T., and Neugart, S. (2025). Light-induced modulation of aroma, sugars, and phenolics in strawberries (Fragaria × ananassa Duchesne) during cold storage. *Food Chem.* 496:146798. doi: 10.1016/j.foodchem.2025.146798

Malayil, D., House, N. C., Puthenparambil, D., et al. (2022). *Borassus flabellifer* haustorium extract prevents pro-oxidant mediated cell death and LPS-induced inflammation. *Drug Chem. Toxicol.* 45, 1716–1722. doi: 10.1080/01480545.2020.1858854

Martinelli, G., Piazza, S., Fumagalli, M., et al. (2025). Investigating ellagitannins from strawberry (Fragaria× ananassa Duch.) as a new strategy to counteract *H. pylori* infection and inflammation. *Food Funct.* 16, 5049–5058. doi: 10.1039/d5fo01022e

Milosavljević, D. M., Maksimović, V. M., Milivojević, J. M., Nakarada, Đ. J., Mojović, M. D., and Dragišić Maksimović, J. J. (2022). Rich in phenolics—strong antioxidant fruit? Comparative study of 25 strawberry cultivars. *Plants* 11:3566. doi: 10.3390/plants11243566

Monmai, C., Choi, J., Rod-In, W., Lee, T. H., and Park, W. J. (2022). Development of fermented rice cake containing strawberry showing anti-inflammatory effect on LPS-stimulated macrophages and paw edema induced mice. *PLoS One* 17:e0276020. doi: 10.1371/journal.pone.0276020

Newerli-Guz, J., Śmiechowska, M., Drzewiecka, A., and Tylingo, R. (2023). Bioactive ingredients with health-promoting properties of strawberry fruit (*Fragaria x ananassa* Duchesne). *Molecules* 28:2711. doi: 10.3390/molecules28062711

Nga, N. T. H., Ngoc, T. T. B., Trinh, N. T. M., Thuoc, T. L., and Thao, D. T. P. (2020). Optimization and application of MTT assay in determining density of suspension cells. *Anal. Biochem.* 610:113937. doi: 10.1016/j.ab.2020.113937

Olennikov, D. N., Khandy, M. T., and Chirikova, N. K. (2022). Oriental strawberry metabolites: LC–MS profiling, antioxidant potential, and postharvest changes of Fragaria orientalis fruits. *Horticulturae* 8:975. doi: 10.3390/horticulturae8100975

Peng, J., Yang, Z., Li, H., Hao, B., Cui, D., Shang, R., et al. (2023). Quercetin reprograms immunometabolism of macrophages via the SIRT1/PGC-1α signaling pathway to ameliorate lipopolysaccharide-induced oxidative damage. *Int. J. Mol. Sci.* 24:5542. doi: 10.3390/ijms24065542

Piedad, M. D., and Natividad, L. (2024). Carotenoid content, phytochemical screening, and antioxidant potential of Kantutay (*Lantana camara* L.), Katuray (*Sesbania grandiflora* L.), and blue ternate (*Clitoria ternatea* L.) flowers in the Philippines. *Int. J. Second. Metab.* 11, 445–461. doi: 10.21448/ijsm.1389365

Qaderi, R., Mezzetti, B., Capocasa, F., and Mazzoni, L. (2023). Stability of strawberry fruit (*Fragaria x ananassa* Duch.) nutritional quality at different storage conditions. *Appl. Sci.* 13, 13:313. doi: 10.3390/app13010313

Raudoniūtė, I., Rovira, J., Venskutonis, P. R., Damašius, J., Rivero-Pérez, M. D., and González-SanJosé, M. L. (2011). Antioxidant properties of garden strawberry leaf extract and its effect on fish oil oxidation. *Int. J. Food Sci. Technol.* 46, 935–943. doi: 10.1111/j.1365-2621.2011.02582.x

Ringling, C., and Rychlik, M. (2017). Simulation of food folate digestion and bioavailability of an oxidation product of 5-methyltetrahydrofolate. *Nutrients* 9:969. doi: 10.3390/nu9090969

Ronie, M. E., Abdul Aziz, A. H., Kobun, R., Pindi, W., Roslan, J., Putra, N. R., et al. (2024). Unveiling the potential applications of plant by-products in food – a review. *Waste Manag. Bull.* 2, 183–203. doi: 10.1016/j.wmb.2024.07.008

Schell, J., Scofield, R. H., Barrett, J. R., Kurien, B. T., Betts, N., Lyons, T. J., et al. (2017). Strawberries improve pain and inflammation in obese adults with radiographic evidence of knee osteoarthritis. *Nutrients* 9:949. doi: 10.3390/nu9090949

Seke, F., Adiamo, O. Q., Sultanbawa, Y., and Sivakumar, D. (2023). In vitro antioxidant activity, bioaccessibility, and thermal stability of encapsulated strawberry fruit (*Fragaria × ananassa*) polyphenols. *Foods* 12:4045. doi: 10.3390/foods12214045

Shahbaz, M., Riaz, M., Momal, U., Rasool, I. F. U., Naeem, H., Raza, N., et al. (2025). Green solvent extraction and eco-friendly novel techniques of bioactive compounds from plant waste: applications, future perspective and circular economy. *Appl. Food Res.* 5:101434. doi: 10.1016/j.afres.2025.101434

Strålsjö, L. M., Witthöft, C. M., Sjöholm, I. M., and Jägerstad, M. I. (2003). Folate content in strawberries (Fragaria × ananassa): effects of cultivar, ripeness, year of harvest, storage, and commercial processing. *J. Agric. Food Chem.* 51, 128–133. doi: 10.1021/jf020699n

Sul, O.-J., and Ra, S. W. (2021). Quercetin prevents LPS-induced oxidative stress and inflammation by modulating NOX2/ROS/NF-kB in lung epithelial cells. *Molecules* 26:6949. doi: 10.3390/molecules26226949

Sun, B., Ricardo-da-Silva, J. M., and Spranger, I. (1998). Critical factors of vanillin assay for catechins and proanthocyanidins. *J. Agric. Food Chem.* 46, 4267–4274. doi: 10.1021/jf980366j

Urün, I., Attar, S. H., Sönmez, D. A., Gündeşli, M. A., Ercişli, S., Kafkas, N. E., et al. (2021). Comparison of polyphenol, sugar, organic acid, volatile compounds, and antioxidant capacity of commercially grown strawberry cultivars in Turkey. *Plants* 10:1654. doi: 10.3390/plants10081654

Veljovic-Jovanovic, S., Kukavica, B., Stevanovic, B., et al. (2006). Senescence-and drought-related changes in peroxidase and superoxide dismutase isoforms in leaves of *Ramonda serbica*. *J. Exp. Bot.* 57, 1759–1768. doi: 10.1093/jxb/erl007

Villamil-Galindo, E., de Van Vel, F., and Piagentini, A. M. (2020). Extracts from strawberry by-products rich in phenolic compounds reduce the activity of apple polyphenol oxidase. *LWT* 133:110097. doi: 10.1016/j.lwt.2020.110097

Villamil-Galindo, E., Van de Velde, F., and Piagentini, A. M. (2021). Strawberry agro-industrial by-products as a source of bioactive compounds: effect of cultivar on the phenolic profile and the antioxidant capacity. *Bioresour. Bioprocess.* 8:61. doi: 10.1186/s40643-021-00416-z

Wang, Z., Liu, X., Bao, Y., Wang, X., Zhai, J., Zhan, X., et al. (2021). Characterization and anti-inflammation of a polysaccharide produced by Chaetomium globosum CGMCC 6882 on LPS-induced RAW 264.7 cells. *Carbohydr. Polym.* 251:117129. doi: 10.1016/j.carbpol.2020.117129

Wesche-Ebeling, P., and Montgomery, M. W. (1990). Strawberry polyphenoloxidase: purification and characterization. *J. Food Sci.* 55, 1315–1319. doi: 10.1111/j.1365-2621.1990.tb03924.x

Xu, J., Li, Y., Yang, X., Li, H., Xiao, X., You, J., et al. (2024). Quercetin inhibited LPS-induced cytokine storm by interacting with the AKT1-FoxO1 and Keap1-Nrf2 signaling pathway in macrophages. *Sci. Rep.* 14:20913. doi: 10.1038/s41598-024-71569-y

Xue, F., Nie, X., Shi, J., Liu, Q., Wang, Z., Li, X., et al. (2017). Quercetin inhibits LPS-induced inflammation and ox-LDL-induced lipid deposition. *Front. Pharmacol.* 8:40. doi: 10.3389/fphar.2017.00040

Yang, W. S., Jeong, D., Yi, Y.-S., et al. (2014). Myrsine seguinii ethanolic extract and its active component quercetin inhibit macrophage activation and peritonitis induced by LPS by targeting to Syk/Src/IRAK-1. *J. Ethnopharmacol.* 151, 1165–1174. doi: 10.1016/j.jep.2013.12.033

Zhang, L., Chen, J., Liao, H., Li, C., and Chen, M. (2020). Anti-inflammatory effect of lipophilic grape seed proanthocyanidin in RAW 264.7 cells and a zebrafish model. *J. Funct. Foods* 75:104217. doi: 10.1016/j.jff.2020.104217

Zhong, R., Miao, L., Zhang, H., Tan, L., Zhao, Y., Tu, Y., et al. (2022). Anti-inflammatory activity of flavonols via inhibiting MAPK and NF-κB signaling pathways in RAW264.7 macrophages. *Curr. Res. Food Sci.* 5, 1176–1184. doi: 10.1016/j.crfs.2022.07.007

Zitouni, H., Hssaini, L., Messaoudi, Z., Ourradi, H., Viuda-Martos, M., Hernández, F., et al. (2020). Phytochemical components and bioactivity assessment among twelve strawberry (*Arbutus unedo* L.) genotypes growing in Morocco using chemometrics. *Foods* 9:1345. doi: 10.3390/foods9101345