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Original Article 1

Bioactive potential of some fruit by-products: antioxidant, antimicrobial, and anti-inflammatory properties

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Background
Agro-byproducts are substances that are produced by the bioprocessing sector that can offer functionality and bioactivity when included in food products. Currently, there is a growing interest in using by-products obtained from fruits and vegetables.
Objective
In the current study, We investigated the bioactive potential of some fruit by-products, including olive meal, Barbary fig peel, date seed, avocado peel and seed, mango peel and kernel, and apricot (kernel).
Materials and methods
Various organic solvents were utilized to extract plant parts (70% ethanol, 70% acetone, and hexane). Standard methods were used to measure the composite index for total antioxidant potency, (which was determined based on total phenols, total flavonoids, 1,1-Diphenyl-2-picrylhydrazyl (DPPH), ferric reducing antioxidant power **(**FRAP), and 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid (ABTS)) antimicrobial activity, *In vitro* anti-inflammatory activity by human red blood cell membrane (HRBC), heat-induced hemolysis, and cytotoxicity assay with brine shrimp larvae (*Artemia salina*).
Results and conclusion
Acetone and ethanol extracts of mango kernel had the highest levels of phenolic compounds, while hexane extract of apricot kernel had the lowest amount. The inhibitory activity against DPPH radical was highest in the acetone extract of date seed, 68.12±1.16 µg/mL. On the other hand, the inhibitory activity against ABTS radical was highest in the acetone extract of mango kernel, 110.85±1.7 µg/mL. This trend was followed by the fruit by-product extracts' antioxidant potency composite index (ACI). The acetone extract of the mango kernel had the highest ACI, followed by its ethanolic extract due to the higher total phenolic content, that was found in these extracts. Avocado peel and seed had antibacterial effects against most of the pathogenic bacteria with different solvents. Anti-inflammatory HRBC stabilization was used to assess the *in vitro* approach. In this method, apricot kernel showed high anti-inflammatory properties with different solvents.
Based on the obtained results, mango kernels, date seeds, and avocado peels can be recommended as potential natural sources of antioxidants that are suitable for use in the pharmaceutical field.

Keywords: fruit by-products, olive, barbary fig, date, avocado, mango, apricot.

Introduction

Plants are beneficial in both their crude and refined forms, providing a pure source of pharmaceuticals. For the ability of plants to produce numerous secondary metabolites, many of the plants have pharmacological and biological activities, which serve as the starting point in the development of modern medicines [1–4]. According to the World Health Organization, 88% of all countries use traditional medicine, such as indigenous therapies, herbal medicines, acupuncture, yoga, etc. Over 40% of pharmaceutical formulations are based on landmark drugs and natural products, including aspirin and artemisinin, which originated from traditional medicine *[5]*.

The food processing industry, which includes the processing of vegetables and fruits, is the second-largest source of environmental pollution after home sewage treatment systems. Agriculture remains a major source of income in developing

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countries, and approximately 1.3 billion tonnes of food grains are wasted or annually destroyed, reducing energy consumption while reducing pro-inflammatory states, stress, oxidative, and physical disorders [6].

Oxidative stress is responsible for the development of several prevalent illnesses, including cancer, obesity, coronary heart disease, type 2 diabetes, cataracts, and hypertension [7–11]. Oxidation is a chemical process that can generate free radicals, which occurs due to the existence of reactive oxygen species (ROS). The predominant reactive oxygen species (ROS) are hydrogen peroxide (H_2O_2), superoxide anion (O_2-), and hydroxyl radicals (OH-). These ROS have been linked to the development and progression of several human illnesses, including inflammation, viral infections, autoimmune disorders, and ulcers. ROS are able to easily interact with and oxidize a wide range of biomolecules, such as carbohydrates, proteins, DNA, and lipids [12,13]. Lately, there has been a growing fascination with discovering natural antioxidants derived from plant components as substitutes for synthetic ones. [14–16].

Phenolic compounds, including phenolic acids and flavonoids, are natural antioxidants found in plants. These compounds can stop the harmful effects of free radicals and protect the human body from various diseases. They have been proven to possess antioxidant activity [17,18]. This activity is due to different mechanisms, such as hydrogen or electron donation, metal ion chelation, free-radical scavenging, singlet oxygen quenching, and scavenging hydroxyl, peroxide, and superoxide radicals [19]. By limiting the onset or propagation of oxidative chain reactions, the oxidation of molecules can be prevented or delayed. [20]. For this, the antioxidant assay is the most extensively used method to detect the biological activity in plant extracts. The diversity of phytochemical sources provides a one-of-a-kind and renewable resource for identifying novel biological functions [21,22].

For many years, the food sector has been utilizing various chemicals to impede the proliferation of bacteria that initiate food spoilage. In order to minimize the utilization of these preservation and processing techniques, bio-preservatives have been developed. Bio-preservatives are naturally derived substances taken from microbes, animals, and plants that extend the shelf life of food items [23]. These substances minimize or eradicate harmful organisms in food and enhance the functioning and quality of food. Many of these chemicals function as both antimicrobials and antioxidants, breaking down cell membranes and disrupting the processes of biosynthetic bacteria [24].

Inflammation, which serves as the initial defensive mechanism against pathogens, can have a role in all stages of carcinogenesis, including the development, promotion, and spread of tumors. Tumor growth heavily relies on the presence of inflammation. A significant number of malignancies originate from areas of infection, persistent irritation, and inflammation [25–27]. The primary mechanism of action of anti-inflammatory drugs is the suppression of cyclooxygenase enzymes, which play a crucial role in converting arachidonic acid into prostaglandins [28].

The cytotoxicity tests are frequently laborious and costly, and there is a need for a convenient and efficient sample screening process. The Brine Shrimp Lethality Assay (BSLA) is currently being used as an alternative method to assess the toxicity of different plant extracts [29]. The BSLA is highly valuable due to its ability to identify a wide range of bioactivity, encompassing cytotoxic, pesticidal, and anticancer characteristics [30]. The median lethal concentration [LC_{50}] values of herbal extracts are usually measured using Meyer's or Clarkson's toxicity index. Meyer's toxicity index classifies extracts as dangerous if LC_{50} is <1000 µg/mL and non-toxic if it exceeds 1000 µg/mL [31]. Clarkson's toxicity criterion ranks plant extracts in order: non-toxic extracts have LC_{50} over 1000 µg/mL, low toxicity is between 500 and 1000 µg/mL, medium toxicity is between 100 and 500 µg/mL, and very toxic is 0 to 100 µg/mL [32].

The present study was an attempt to utilize some fruit by-products [olive meals, date seeds, Barbary fig peels, avocado (peels and seeds), mango (peels and kernel), and apricot kernel] as natural sources of antioxidant, antimicrobial, anti-inflammatory, and cytotoxicity effects.

Materials and methods

Chemicals and reagents

All the used chemicals were of analytical grade. Ethanol, acetone, hexane, hydrochloric acid (HCL), Dimethyl sulfoxide (DMSO), 1,1-Diphenyl-2-picrylhydrazyl (DPPH), ascorbic acid, gallic acid (GA), rutin (RU), anhydrous sodium carbonate (Na_2CO_3), ferric chloride hexahydrate ($FeCl_3.6H_2O$), Folin–Ciocalteu reagent. All chemicals were of reagent grade and employed without further purification.

Plant collection

Different fresh parts of fruit samples [olive meal (*Olea europaea*), Barbary fig peel (*Opuntia ficus-indica*), date seed (Phoenix *dactylifera*), avocado (*Persea Americana*) peel and seed, mango (*Mangifera indica*) (zebdeya) peel and kernel and apricot (*Prunus armeniaca*) (amar) kernel] were collected from the local market. Samples were dried in the oven at 40°C. then powdered by milling

with an electric blender [33]. The powder was stored at -18 ℃ in an airtight container until used.

Extraction of samples

The extraction process was conducted with several adjustments, as [33,34] stated. Each powdered sample, weighing 50 g, was individually soaked in 250 mL of three different solvents (ethanol 70%, acetone 70% and hexane) for 48 h at room temperature. The soaking process involved occasional shaking, and an ultrasonic treatment was applied for 45 minutes at the beginning of the soaking period and for 15 minutes at the end. The extracts were filtered using filter paper,Whatman No.1, and the resulting liquid was concentrated by removing the solvent under decreased pressure using a rotary evaporator at 40°C. Subsequently, the extracts were preserved at a temperature of 4℃ until further analysis. The extract's yield was calculated.

Phytochemical screening

Preliminary tests of phytosterol, terpenoids, coumarins, tannins, glycosides, phenolic compounds, alkaloids, flavonoid contents, and saponins were carried out on each extract as per standard methods described by [35].

Phytochemical constituents

Determination of total phenolic compounds

The total phenolic content (TPC) was measured by the Folin-Ciocalteu reagent, as described by [36]. Therefore, a volume of 10 µL of adequately diluted extract in DMSO was combined with 100 µL of freshly diluted Folin-Ciocalteu reagent, which was diluted 1/10 with distilled water. After a duration of five minutes, the solution was combined with 100 µL of Na_2CO_3 at a concentration of 7.5%. After waiting for 60 min, the absorbance at 650 nm was measured using an ELISA reader (ELx800 Absorbance Reader., Bio Tek Instruments, Inc., USA). The total phenolic components were quantified and reported as (mg GA/g DE).

Determination of total flavonoids

The total flavonoid content (TFC) was conducted using the colorimetric technique derived from [37], employing aluminum chloride reagent. 50 µL samples of each extract's accessions were added to a 96-well plate together with 130 µL of 96% ethanol, 10 µL of 10% aluminum chloride, and 10 µL of 1 M sodium acetate. The mixtures were left to incubate for 40 min at room temperature in the absence of light. The absorbance at a wavelength of 415 nm was measured using an ELISA reader (ELx800 Absorbance Reader, Bio Tek Instruments, Inc., USA). The total flavonoid content (TFC) was quantified in [mg RU/g DE extract] using a calibration curve with rutin.

Assessment of antioxidant activity

DPPH radical scavenging activity, the test was conducted according to the methodology outlined by [38]. The experiment involved running appropriate blank samples containing DMSO at a concentration of 2% and standard samples containing Trolox and ascorbic acid solutions in DMSO at concentrations of 5, 10, 50, and 100 µg/mL. These samples were run at the same time. Various concentrations of extracts (50, 100, 250, 500, 1000, and 2000 µg/mL) were produced. 20 µL of sample was combined with 180 µL of DPPH solution in each well in a 96-well plate, then incubated in the dark for 30 min. The percentage inhibition of DPPH radical scavenging activity was determined using the formula: % inhibition = {(A blank – A sample) / A blank} × 100. Here, A blank represents the absorbance of the DPPH solution without extract at 490 nm, measured using an ELISA reader (ELx800 Absorbance Reader, Bio Tek Instruments, Inc., USA), and A sample represents the absorbance of the sample with DPPH solution.

ABTS radical scavenging activity, the test was performed using the methodology outlined by [39], with certain modifications. The ABTS cation radical was generated via the interaction of 7 mM ABTS in water with 2.45 mM potassium persulfate in a 1:1 ratio. The solution was kept under light-restricted conditions at ambient temperature for 12 to 16 hours before use. Subsequently, the solution was mixed with distilled water until an absorbance of 0.7 ± 0.005 was achieved. A volume of 67 µL of the sample and 133 µL of ABTS solution were added to each well of a 96-well microplate. The microplate was then incubated in the dark for 15 min. The sample's absorbance was quantified at a wavelength of 630 nm using an ELISA reader (ELx800 Absorbance Reader, Bio Tek Instruments, Inc., USA). The ABTS scavenging activity is calculated as a percentage using the formula: {(A blank – A sample)\A blank]} ×100. Where: A_blank represents the absorbance without extracts, and A_sample represents the absorbance of the extract with ABTS solution.

Ferric reducing antioxidant power (FRAP) assay, the technique was conducted following the methodology outlined by [38], with minor adjustments. For each well in a 96-well plate, 20 µL of each extract (at a concentration of 1000 µg/mL) was suitably diluted in DMSO. The diluted extracts were then combined with 180 µL of FRAP reagent and allowed to sit for 6 minutes. The absorbance of the mixture was thereafter measured at a wavelength of 630 nm using an ELISA reader (ELx800 Absorbance Reader, Bio Tek Instruments, Inc., USA). The FRAP values are expressed as micromoles of ferrous equivalents (FE) per mg of the sample extract using the calibration curve constructed for different concentrations of $FeSO_4$ (between 30-1000 µg/ml).

Antioxidant potency composite index [ACI]
The composite index for total antioxidant potency was determined based on the methodology described by [40]. This involved assigning equal importance to several assays, including total phenols, total flavonoids, DPPH, FRAP, and ABTS. Each test was given a score out of 100, with the highest score representing the best performance. An index score for all remaining samples in the test was calculated: Antioxidant Index Score = Sample Score Best Score ×100.

Evaluation of antimicrobial activity
To measure the antimicrobial activity of extracts. Gram-negative bacteria (*Escherichia coli ATCC10536, Salmonella typhi ATCC14028*), Gram-positive bacteria (*Staphylococcus aureus ATCC 9027*, *Bacillus cereus ATCC14579*), yeast (*Candida albicans MTCC183*), and fungi (*Aspergillus niger NRRL595*) were used as test organisms. The antibacterial activity was determined using a well diffusion experiment on a nutrient agar medium. Bacterial plates were incubated at 37^{o}C for 24 hours. Mould and yeast were incubated on potato dextrose agar (PDA) at 25^{o}C for 72 hours. Following incubation, the inhibition zones were measured in millimeters according to [41].
Minimal inhibitory concentration (MIC) was evaluated by the disk diffusion assay[42] with extracts that have antimicrobial activity, the concentrations used were (50, 25, 12.5, 6.25, 3.12 and 1.56 mg/mL).

In vitro anti-inflammatory activity
Human red blood cells (HRBC) were combined with an equivalent amount of Alsevers solution, which consists of 0.42% NaCl, 0.5% citric acid, 0.8% sodium citrate, and 2% dextrose. The mixture was then centrifuged at 3000 rpm for 10 min. The concentrated cells were rinsed with isosaline and a 10% suspension was prepared. one mL of each extract (25mg/mL) was combined with 0.5 mL of HRBC solution, 2 mL of hyposaline, and 1 mL of 2mM phosphate buffer. The reaction mixtures were placed in a water bath set at a temperature of 56 °C for a duration of 30 min. Subsequently, the reactions were cooled and centrifuged at 2500 rpm for 5 minutes. The supernatants were analyzed for absorbance at a wavelength of 560 nm. Experiment using pure water instead of hyposaline to achieve complete hemolysis. [43,44]. The percentage of hemolysis inhibition =100 ×{($A1-A2$)/$A1$ }. A1 = Absorbance of control solution , A2 = Absorbance of sample or standard.

Cytotoxicity assay with brine shrimp larvae (Artemia salina)
The hatching process of brine shrimp eggs (Artemia salina) involved preparing artificial seawater by dissolving 40 g of sodium chloride in 1 L of distilled water. The water was then oxygenated using an aquarium pump and illuminated with a light source. The brine shrimp were subjected to incubation at a temperature range of 22-29°C for a duration of 48 h until the larvae (nauplii) emerged from their eggs. The experiment was conducted on brine shrimp larvae [45], incorporating certain modifications. Each well of the 96-well microtiter plates was filled with 200 µL of extract at various concentrations (50, 100, 250, 500, 1000 and 2000 µg/mL). Subsequently, 10 nauplii were introduced into each well. The control wells contained 10 nauplii and artificial seawater with a 0.5% concentration of DMSO. The plate was placed in an incubator for 24h. The plates were analyzed using a binocular microscope (magnification x 12.5) to determine the quantity of deceased (non-mobile) nauplii in each well. The larval mortality % was determined using the calculation provided by(46): The mortality rate = (number of dead larvae/number of tested larvae) x100.

Statistical analysis
The statistical analysis presented as the mean ± standard error (SE) of three separate determinations according to [46]. The significance levels for comparing differences were calculated using a one-way Analysis of variance (ANOVA) at a significance level of $P < 0.05$, using the Costat software. The Pearson correlation test was utilized to ascertain the correlation coefficients between bioactive compounds and various antioxidant assays. Principal Component Analysis (PCA) by Minitab® 19.2020.1 program was employed to enhance the visualization of the distinct data sets and the variability of the antioxidant activity of the different extracts utilized.

Results

Extraction yields
The extraction yield results of the different extracts are displayed in (Table 1). The ethanolic and acetone extracts of mango peel had extraction yields of 40.38 % and 33.71 %, respectively. Similarly, Barbary fig peel's ethanolic and acetone extracts had 35.42% and 22.92% extraction yields, respectively. The ethanol extract of date seeds had a yield of 6.51%, whereas the acetone extract had a yield of 3.81%. The hexane extract of Barbary fig peel yielded the lowest amount (1.03%). In addition, the ethanolic extract yielded a higher amount than other solvents used for the same sample, except for the hexane extract of the apricot kernel, which yielded 14.301%.

Table 1 The extraction yield for the different extracts and abbreviations.

Number	Plant sample	yield % ethanole 70%	abrev.	acetone70%	abrev.	hexane	abrev.
1	Olive meal	11	1E	7.23	1A	6.64	1H
2	Barbary fig peel	35.42	2E	22.92	2A	1.03	2H
3	Date seed	6.51	3E	3.81	3A	4.87	3H
4	Avocado peel	14.28	4E	11.2	4A	2.5	4H
5	Avocado seed	18.34	5E	13.59	5A	1.81	5H
6	Mango peel	40.38	6E	33.71	6A	2.06	6H
7	Mango kernel	22.05	7E	21.871	7A	4.9	7H
8	Apricot kernel	11.02	8E	6.08	8A	14.301	8H

Phytochemical screening

Preliminary chemical reactions were carried out to detect and identify the presence or absence of bioactive compounds. The results in Table 2 reveal the presence of many classes of compounds that might indicate different bioactive compounds (phytosterol, terpenoids, coumarin, flavonoids, tannins, glycosides, phenolic compounds, alkaloids, and saponins) in most ethanol and acetone extracts. The ethanol and acetone extracts of date seeds were the richest of the tested bioactive compounds, followed by the ethanol extract of avocado seeds, while all hexane extracts were few in the bioactive compounds. However, glycoside was detected in all extracts, while terpenoids detected in few extracts, the highest phenolic was found in the acetone extract of mango kernel, while the highest flavonoids were found in mango kernel in both the ethanol and acetone extracts as well as the acetone extract of date seed and barbary fig peel.

Table 2 The phytochemical screening test for the different extracts.

Extract		Test Phytoster	Terpenoi	Coumari	Flavonoi	Tannin	Glycosid	Phenolic	Alkaloid	Saponin
Olive meal	Ethanol	+	+	-	+	++	+		+	+
Barbary fig	Ethanol	-	-	+	+	+	+	+	-	-
Date seed	Ethanol	++	++	-	++	+++	+	+	++	-
Avocado peel	Ethanol	+	-	-	++	++	++	+	++	+
Avocado seed	Ethanol	+	+	-	++	+++	++	+	+	++
Mango peel	Ethanol	-	-	+	+	+	+	+	-	-
Mango kernel	Ethanol	+	-	+	+++	+	+	++	-	-
Apricot kernel	Ethanol	-	-	+	-	-	+	-	-	-
Olive meal	Acetone	++	++	-	+	+	+	+	+	+
Barbary fig	Acetone	-	-	+	+	-	+	+	-	-
Date seed	Acetone	+	++	-	+++	+	++	++	+	+
Avocado peel	Acetone	-	-	-	+++	++	+++	++	++	+
Avocado seed	Acetone	-	-	-	++	+++	++	+	++	++
Mango peel	Acetone	-	-	++	+	+	+	+	-	-
Mango kernel	Acetone	+	+	-	+++	+	+	+++	+	-
Apricot kernal	Acetone	-	-	+	-	-	+	-	+	-
Olive meal	Hexane	-	-	-	-	-	+	-	+	-
Barbary	Hexane	-	-	+	-	-	+	-	+	-
Date seed	Hexane	-	-	-	-	-	+	-	-	-
Avocado peel	Hexane	-	-	+	-	-	+	-	+	-
Avocado seed	Hexane	-	-	-	-	-	+	-	++	+
Mango peel	Hexane	-	-	+	-	-	+	-	+	-
Mango kernel	Hexane	-	-	-	-	-	+	-	-	-
Apricot kernal	Hexane	-	-	-	-	-	+	-	-	-

[-] not detected, [+] low, [++] moderate, [+++] high.

Phytochemical constituents

The content of phenolic compounds and flavonoids of the extract samples was determined from the standard curve (R^2=0.9995) and (R^2=0.9988), respectively. Table 3 shows that highest contents of phenolic compounds and flavonoids were found in the acetone extract of mango kernel (390.54±0.03 mg GA/g DE and 76.96± 2.7 mg RU/g DE) respectively, followed by avocado peel extract (279.15±1.9 mg GA/g DE and 75.39±2.5 mg RU/g DE) respectively.

The ethanolic extract of mango kernel had the highest phenolic compounds (332.26±2.5 mg GA/g dry DE) and flavonoids (60.55±1.85)**,** followed by date seed extract which contains phenolic compounds (176±2.9 mg GA/g DE) and flavonoids (41.6±0.88 mg RU/g DE). While the lowest values were observed in apricot kernel in the both extracts of acetone and ethanol (11.13±0.8 and 14.4±0.8) respectively. The result indicates that the mango kernel is significantly increases in phenolic and flavonoid compounds content in the both extracts (acetone and ethanol) compared with other extracted samples.

Table 3 Phenolic compounds and Flavonoid concentration for different sample extracts.

Extracts		Phenolic conc. Eq mg GA /g dry extract	Flavonoids conc. Eq mg RU /g dry extract
Olive meal	Ethanol	37±0.38 [f]	23.88±1.22 [d]
Barbary fig peel		30.77±0.29 [f]	11.75±0.77 [f]
Date seed		176.56±2.9 [b]	41.6±0.88 [b]
Avocado peel		131.66±2.9 [d]	42.69±1.37 [b]
Avocado seed		143.77±2.8 [c]	36.82±1.45 [c]
Mango peel		73.66±1.5 [e]	16.69±1.44 [e]
Mango kernel		332.26±2.5 [a]	60.55±1.85 [a]
Apricot kernal		14.4±0.8 [g]	3.55±0.9 [g]
Olive meal	Acetone	42.11±0.9 [f]	22.39±1.5 [c]
Barbary figpeel		29.57±0.9 [g]	16.42±1.5 [d]
Date seed		246.77±2.8 [c]	47.74±1.2 [b]
Avocado peel		279.15±1.9 [b]	75.39±2.5 [a]
Avocado seed		122.55±1.5 [e]	24.43±0.9 [c]
Mango peel		129.43±2.01 [d]	9.56±0.7 [e]
Mango kernel		390.54±2.9 [a]	76.96±2.7 [a]
Apricot kernal		11.13±0.8 [h]	3.17±0.9 [f]
Olive meal	Hexane	0.95±0.02 [f]	0.36±0.03 [f]
Barbary figpeel		1.92±0.06 [d]	0.54±0.02 [e]
Date seed		1.47±0.12 [e]	0.71±0.03 [c]
Avocado peel		2.63±0.06 [c]	1.05±0.03 [b]
Avocado seed		4.92±0.06 [a]	2.06±0.05 [a]
Mango peel		2.85±0.12 [c]	0.61±0.02 [d]
Mango kernel		4.17±0.1 [b]	0.23±0.02 [g]
Apricot kernel		0.52±0.2 [g]	0.06±0.01 [h]

Antioxidant activity

In the DPPH assay, different sample extracts showed variability in their inhibitory activity against the DPPH radical. A noticeable effect on radical scavenging activity as shown in Fig. 1 and Table 4. The antioxidant activity is inversely related to IC_{50}, so the lower IC_{50} value had the higher antioxidant activity. Amongst different extracts, the highest radical scavenging activity was detected in the acetone extract for date seeds, mango kernel, and avocado peels, The IC_{50} values of date seeds and mango kernel were 68.12±1.16 µg/mL, 73.41±2.33 µg/mL respectively which were significantly lower than the IC_{50} of ascorbic acid (79.9±1.23 µg/mL) and higher than the IC_{50} of Trolox (59.72±1.54 µg/mL), while the IC_{50} of avocado peels was (85.66±0.88 µg/mL) which is significantly higher than the IC_{50} of ascorbic acid and Trolox. In ethanol extract, the IC_{50} for mango kernel was (80.3±0.8 µg/mL)**,** nearest to the IC_{50} of ascorbic acid. On the other hand, hexane extracts from different plants had the highest IC_{50} values.

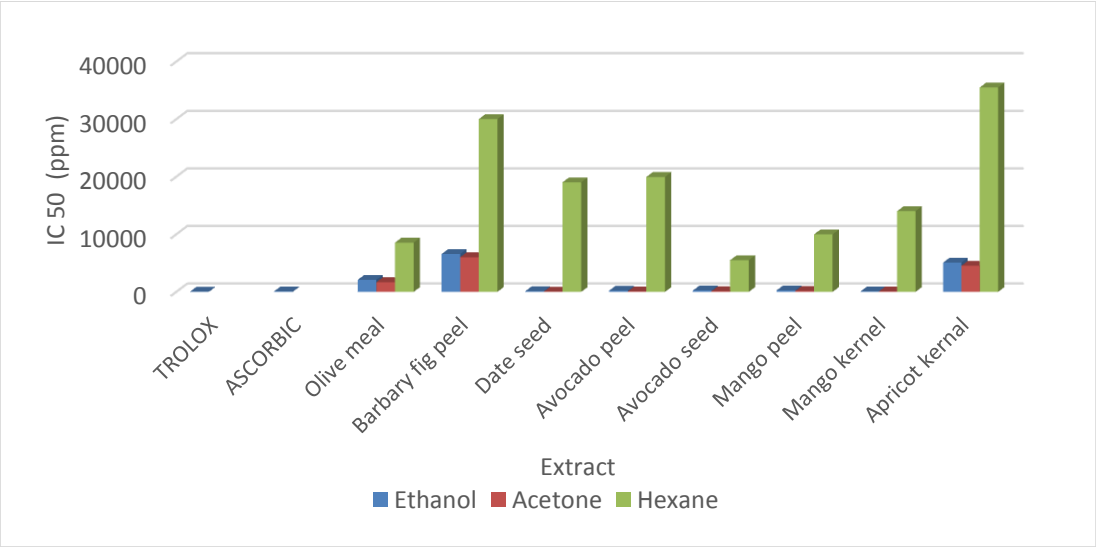

Fig. 1 The DPPH IC_{50} radical scavenging for different extracts.

Table 4 Antioxidant assays for different sample extracts.

Sample		DPPH IC_{50}	ABTS IC_{50}	FRAP Conc. eq $FeSO_4$
TROLOX		59.72±1.54 ^{i}	151.65±2.56 ^{h}	
ASCORBIC		79.91±1.24 ^{h}	199.71±1.9 ^{g}	
Olive meal		2101.12±5.4 ^{c}	1583.16±5.18 ^{c}	21.96±1.22 ^{g}
Barbary fig peel		6572.94±3.65 ^{a}	1757.95±8.41 ^{b}	50.56±0.63 ^{f}
Date seed	Ethanol	124.75±2.04 ^{g}	284.54±4.79 ^{f}	539.86±1.82 ^{b}
Avocado peel		210.94±2.51 ^{f}	488.35±2.94 ^{d}	478.46±1.82 ^{d}
Avocado seed		247.7±1.49 ^{e}	430.91±1.41 ^{e}	500.04±1.5 ^{c}
Mango peel		271.06±1.56 ^{d}	496.12±2.64 ^{d}	377.93±1.27 ^{e}
Mango kernel		80.3±0.88 ^{h}	134.35±2.47 ^{i}	572.21±2.01 ^{a}
Apricot kernel		5084.88±4.18 ^{b}	12245.39±3.03 ^{a}	18.81±0.35 ^{g}
TROLOX		59.72±1.54 ^{i}	151.65±2.56 ^{h}	
ASCORBIC		79.91±1.23 ^{fg}	199.71±1.91 ^{g}	
Olive meal		1694.41±3.23 ^{c}	1438.13±3.51 ^{c}	164.77±2.01 ^{f}
Barbary fig peel		6022.85±3.81 ^{a}	2041.193.12 ^{b}	63.54±1.07 ^{g}
Date seed	Acetone	68.12±1.16 ^{hi}	222.71±4.63 ^{f}	552.14±2.02 ^{b}
Avocado peel		85.66±0.88 ^{f}	150.44±0.79 ^{h}	547.05±1.4 ^{c}
Avocado seed		128.95±2.33 ^{e}	454.94±4.62 ^{d}	431.26±2 ^{e}
Mango peel		170.36±4.98 ^{d}	306.722.52 ^{e}	510.56±2.02 ^{d}
Mango kernel		73.41±2.33 ^{gh}	110.85±1.7 ^{i}	580.91±1.9 ^{a}
Apricot kernal		4532.88±4.12 ^{b}	5254.12±9.07 ^{a}	43.017±0.76 ^{h}
TROLOX		59.72±1.54 ^{j}	151.65±2.56 ^{j}	
ASCORBIC		79.91±1.23 ^{i}	199.71±1.91 b ^{i}	
Olive meal		8563.67±3.33 ^{g}	50222.39±5.38 ^{b}	6.2±0.5 ^{d}
Barbary fig peel		30021.86±3.83 ^{b}	22045.06±6.7 ^{e}	17.75±0.47 ^{b}
Date seed	Hexane	19049.09±2.48 ^{d}	20087.34±2.12 ^{f}	5.12±0.7 ^{d}
Avocado peel		19970.96±4.65 ^{c}	26233.05±4.2 ^{c}	11.43±0.35 ^{c}
Avocado seed		5514.48±3.87 ^{h}	14653.85±8.5 ^{h}	46.7±1.9 ^{a}
Mango peel		9995.46±1.98 ^{f}	22346.38±7.2 ^{d}	11.26±0.41 ^{c}
Mango kernel		14026.74±4.84 ^{e}	18481.87±8.07 ^{g}	20.56±0.87 ^{b}
Apricot kernal		35502.35±1.85 ^{a}	63317.29±9.02 ^{a}	6.88±0.63 ^{d}

In the ABTS assay, the highest sample results of ABTS radical scavenging activity are presented in Fig. 2. The IC_{50} as Trolox and ascorbic were (151±2.56 and 199.7±1.9 µg/mL) respectively**,** while the best plant extract was acetone extract, which had the highest antioxidant activity in mango kernel and avocado peels (110.85±1.75 and 150.44±0.79 µg/mL respectively). Similar to the result of the DPPH assay, mango kernel was the best extract in ethanol extracts (134.357±2.47 µg/mL).The IC_{50} of acetone extract for mango kernel and avocado peels and the ethanolic extract of mango kernel were lower than the the IC_{50} of Trolox and ascorbic acid. While the lower value was found in all hexane extracts Table 4.

In the FRAP assay, the results are presented in Fig. 3, Table 4. The standard solution were used to obtain standard curves, which were $FeSO_4.7H_2O$. The acetone extract exhibited the highest antioxidant activity among the extracts from mango kernel, date seed, avocado peel, and mango peel which were 580.91±1.9, 552.14±2.02, 547.05±1.4 and 510.56±2.02 µg/mL respectively wherein the ethanol extract showed the highest value in mango kernel (572.21±2.01 µg/mL), as well as the results of the DPPH and ABTS assays.

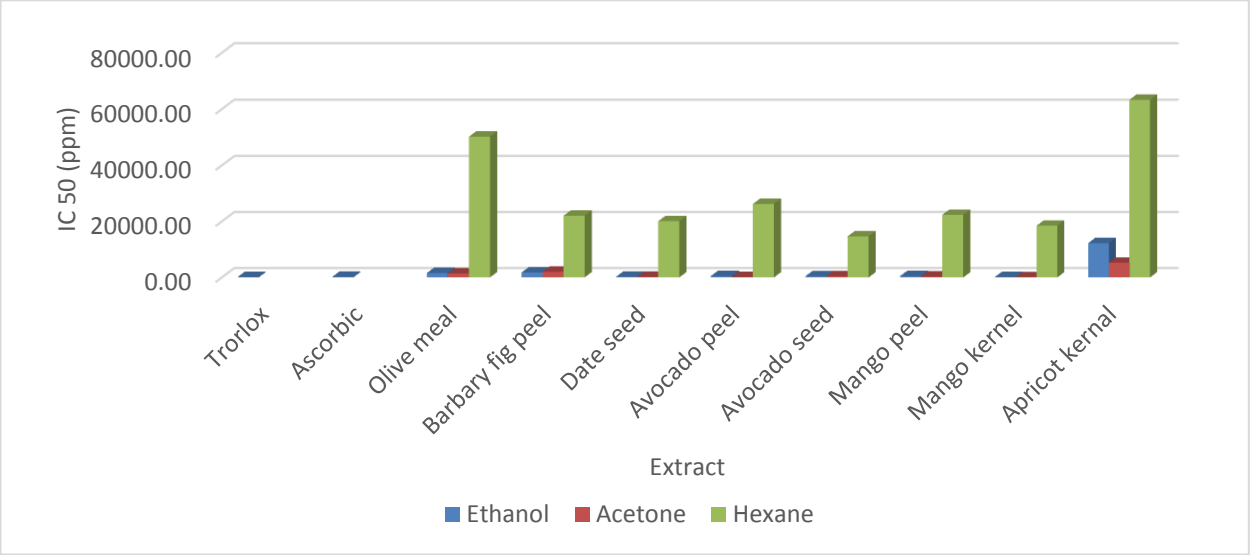

Fig. 2 $ABTS^{++}$ radical scavenging activity (IC_{50} ppm) of different extracts.

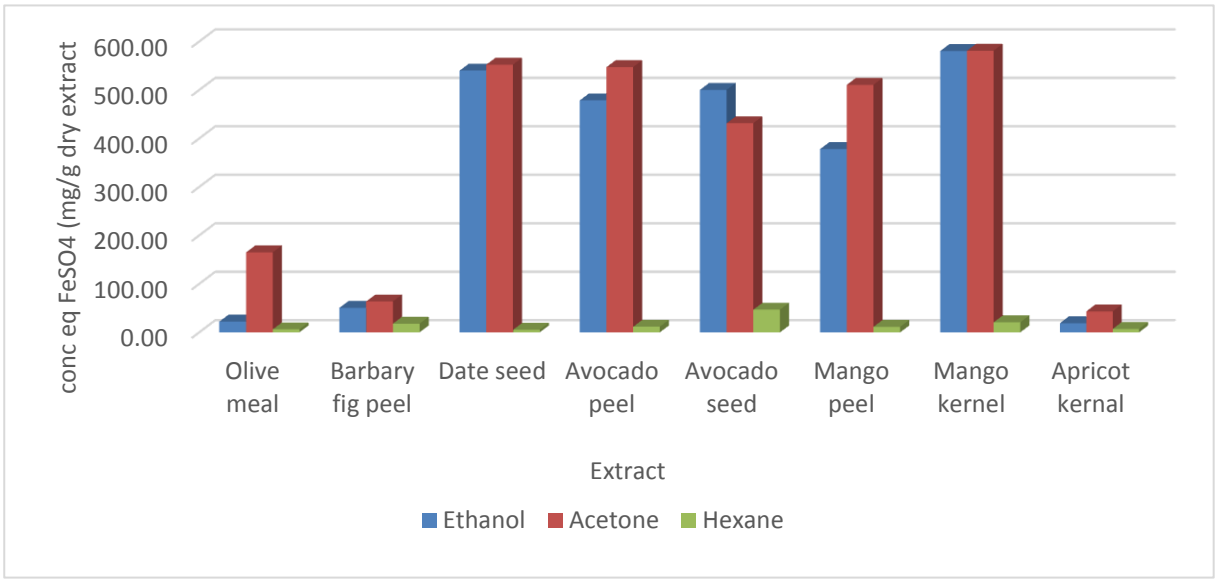

Fig. 3 Ferric reducing antioxidant power (FRAP) of different extracts.

Antioxidant potency composite index (ACI)

Different mechanisms of action used by the DPPH, ABTS, and FRAP assays to measure plant extract antioxidant capabilities resulted in varying rankings for the same extract. The antioxidant potency composite index (ACI) was computed to assign equal significance to all these methodologies. The ACI values are shown in Fig. 4. The highest values were obtained with acetone and ethanol extracts for mango kernel (98.55 and 86.19 %, respectively),

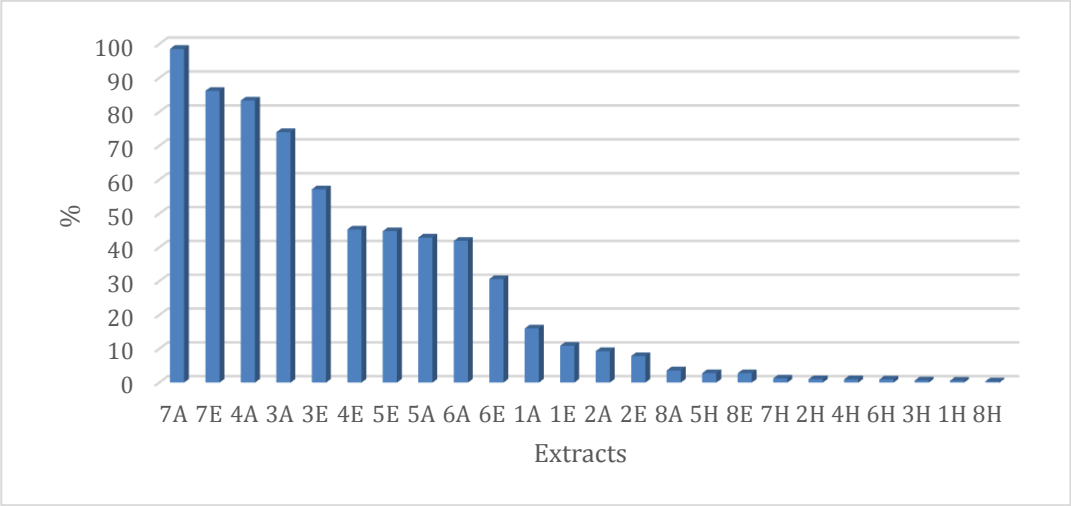

Fig. 4 Antioxidant potency composite index of different extracts, A acetone E ethanol H hexane 1 olive meal, 2 Barbary fig peel, 3 date seed, 4 avocado peel, 5 avocado seed, 6 mango peel, 7 mango kernel, and 8 apricot kernel

The absorbance dataset was further analyzed using Principal Component Analysis (PCA) with Minitab software. Fig. 5 shows the score plot of PCA of 24 extracts of plant samples. The highest values were recorded for the ethanol and acetone extracts of mango kernel followed by acetone extract of avocado peels.

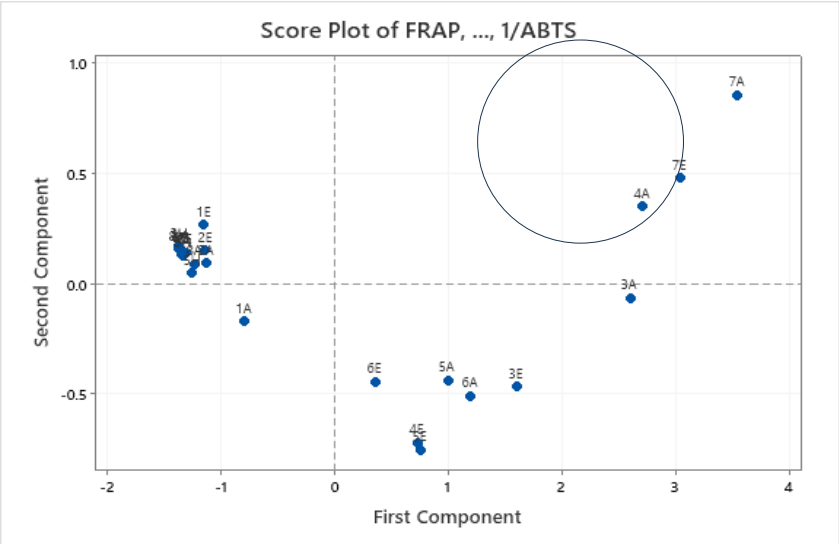

Fig. 5 Principal Component Analysis (PCA) of different extracts, A acetone E ethanol H hexane 1 olive meal, 2 Barbary fig peel, 3 date seed, 4 avocado peel, 5 avocado seed, 6 mango peel, 7 mango kernel, and 8 apricot kernel

Antimicrobial activity

The findings of the current investigation demonstrated a broad spectrum of inhibition zones, ranging from 6 mm to 22 mm (Table 5). The acetone extract of mango kernel showed an inhibitory effect against all the tested microorganisms. The highest inhibition zone were recorded with *Candida* and *Aspergillus niger* as (22±0.94 and 20.5±0.94 mm, respectively). Avocado peels and seeds had an inhibitory effect against most pathogenic bacteria with all solvents range from (7.33±0.33 -15±0.57 mm).Meanwhile, date seed and mango peel showed zones of

inhibition in all tested strains with ethanol and acetone, and no inhibition zones were observed with hexane extracts. Ethanol extract of mango peel showed no significant difference with ampicillin (AMP) antibiotic against *B.cereus* and *E.coli*. The lowest inhibition zones were recorded with *Candida albicans* and *Aspergillus niger* by most of the tested extracts with different concentrations. tetracycline, ampicillin and cefixime are used as control treatments to assess the efficacy of the tested extracts.

Table 5 Antimicrobial activity of the tested extract (inhibition zone mm).

Sample		inhibition zone(mm) Gram [(+ve)		Gram (-ve)		Yeast	Fungi
		B.cereus	*Staphelococcus*	*E.coli*	*Salmonella*	*Candida*	*Aspergillus*
TEC 30		21.67±0.33 ^{a}	16.67±0.33 ^{a}	22.67±0.33 ^{a}	29.67±0.33 ^{a}	-	-
CFM 5		16.33±0.33 ^{b}	16.33±0.33 ^{a}	NA	NA	-	-
AMP10		11.67±0.33 ^{d}	11.33±0.67 ^{b}	18.5±0.29 ^{b}	12.33±0.33 ^{b}	-	-
NYS		-	-	-	-	29±0.7 ^{a}	27±1 ^{a}
Olive meal		NA	NA	9±0.58 ^{f}	NA	NA	NA
Barbary fig peel		NA	NA	12.67±0.33 ^{cd}	NA	NA	NA
Date seed		9.5±0.29 ^{fg}	8±0.57 ^{e}	11.83±0.17 ^{de}	8.67±0.33 ^{d}	NA	12.5±0.33 ^{c}
Avocado peel	Ethanol	10.33±0.33 ^{ef}	8.667±0.33 ^{de}	12.33±0.33 ^{d}	7.33±0.33 ^{e}	NA	13±0.33 ^{c}
Avocado seed		11.33±0.33 ^{de}	7.5±0.28 ^{cd}	11±0.58 ^{e}	10.33±0.33 ^{e}	NA	15±0.33 ^{b}
Mango peel		11.67±0.33 ^{d}	8.67±0.33 ^{de}	19±0.58 ^{b}	7.33±0.33 ^{e}	NA	10±0.33 ^{d}
Mango kernel		9.03±0.61 ^{g}	10.33±0.33 ^{bc}	13.6±0.33 ^{c}	11±0.57 ^{c}	16±0.54 ^{b}	15±0.57 ^{d}
Apricot kernel		14±0.58 ^{c}	0±0 ^{f}	7.67±0.33 ^{g}	NA	NA	7±0.27 ^{e}
TEC 30		21.67±0.33 ^{a}	16.67±0.33 ^{a}	22.6±0.33 ^{a}	29.67±0.33 ^{a}	-	-
CFM 5		16.33±0.33 ^{b}	16.33±0.33 ^{a}	NA	NA	-	-
AMP10		11.67±0.33 ^{d}	11.33±0.67 ^{b}	18.5±0.28 ^{b}	12.33±0.33 ^{bc}	-	-
NYS		-	-	-	-	29±0.7 ^{a}	27±1 ^{a}
Olive meal		NA	NA	6.67±0.33 ^{e}	NA	NA	NA
Barbary fig peel		NA	NA	14±0.58 ^{c}	8.67±0.33 ^{de}	NA	NA
Date seed		12.83±0.17 ^{c}	9.67±0.33 ^{c}	14±0.58 ^{c}	13±0.58 ^{b}	NA	16.5±0.33 ^{c}
Avocado peel	Acetone	12.83±0.17 ^{c}	11±0.58 ^{b}	11.5±0.28 ^{d}	11.67±0.33 ^{bc}	NA	10±0.27 ^{f}
Avocado seed		11.67±0.33 ^{d}	7.5±0.38 ^{d}	12.33±0.33 ^{dc}	9.67±0.88 ^{d}	NA	15±0.57 ^{d}
Mango peel		9.67±0.28 ^{e}	8.33±0.33 ^{d}	11.33±0.88 ^{d}	8±0.58 ^{e}	NA	12.5±0.33 ^{e}
Mango kernel		11.67±0.33 ^{d}	11±0.58 ^{b}	14.67±0.33 ^{c}	11±0.58 ^{c}	22±0.94 ^{b}	20.5±0.94 ^{b}
Apricot kernel		9.5±0.288 ^{e}	NA	8±0.58 ^{e}	NA	NA	7.5±0.33 ^{g}
TEC 30		21.67±0.33 ^{a}	16.67 ±0.33 ^{a}	22.67±0.33 ^{a}	29.67±0.33 ^{a}	-	-
CFM 5		16.33±0.33 ^{b}	16.33±0.33 ^{a}	NA	NA	-	-
AMP10		11.67±0.33 ^{c}	11.33±0.67 ^{b}	18.5±0.28 ^{b}	12.33±0.33 ^{b}	-	-
NYS		-	-	-	-	29±0.7 ^{a}	27±1 ^{a}
Olive meal		NA	NA	8.33±0.33 ^{de}	NA	NA	NA
Barbary fig peel		NA	NA	6.67±0.33 ^{e}	NA	NA	NA
Date seed		NA	NA	6.67±0.33 ^{e}	8.67±0.33 ^{c}	NA	NA
Avocado peel	Hexane	7.33±0.33 ^{e}	11±0.57 ^{b}	11±0.58 ^{c}	9.67±0.88 ^{c}	8±0.27 ^{c}	NA
Avocado seed		9.67±0.33 ^{d}	10.17±0.44 ^{b}	11.67±0.33 ^{c}	10.33±0.88 ^{c}	12.5±0.27 ^{b}	NA
Mango peel		NA	NA	7.67±0.33 ^{de}	NA	8±0.28 ^{c}	NA
Mango kernel		NA	NA	8.33±0.33 ^{de}	NA	NA	NA
Apricot kernel		NA	NA	8.67±0.33 ^{d}	NA	NA	NA

Abbreviations: (-), not tested; NA, no activity, TEC, tetracycline (30µg disc), CFM cefixime (5µg disc), AMP ampicillin (10µg disc), NYS nystatin

Minimum inhibitory concentration (MIC) of extracts against the tested organisms varied between 50 mg/mL and 1.56 mg/mL (Fig. 6). *E.coli*, was the most sensitive strain to the tested extracts with 6.25 to 1.56 mg/mL while *Candida albicans* was the least sensitive strain with most of the tested extracts. The extracts with the best results were made from the mango kernel in both ethanol and acetone extracts, with 1.56 mg /mL for most tested organisms this result is in the same line of antimicrobial activity.

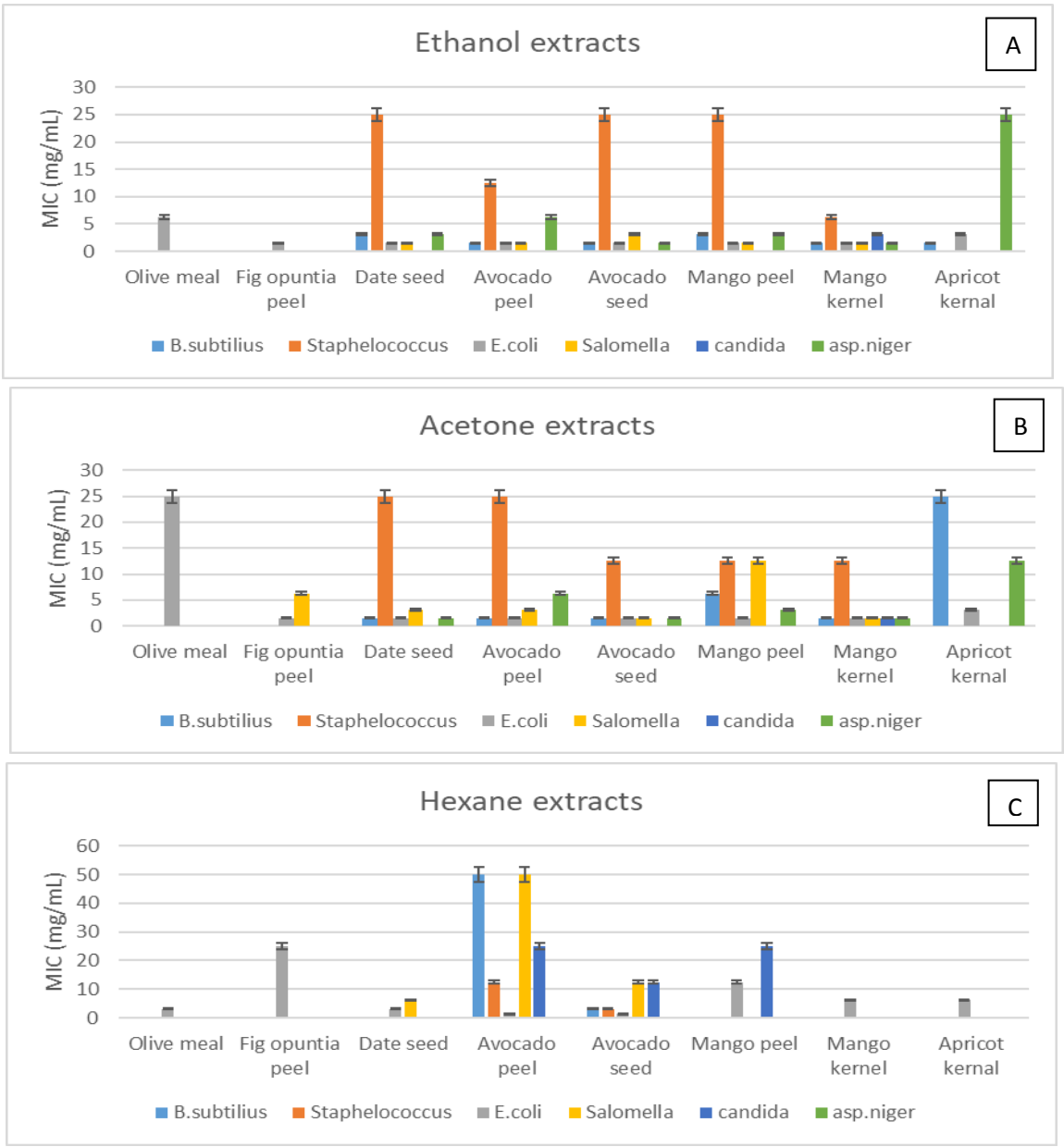

Fig. 6 The minimal inhibitory concentration (MIC) of extracts (a) ethanol extract, (b) acetone extracts, and © hexane extracts.

In vitro anti-inflammatory activity

The percentage of hemolysis inhibition was assessed for both extracts and aspirin at a dosage of 25 mg/mL. The results of our investigation indicated that the tested plant extracts had a significant anti-inflammatory effect, as illustrated in Table 6. The apricot kernel exhibited the most significant increase in hemolysis suppression across all solvents, with values of 69.60±0.31, 87.33±1.2, and 75.27±1, respectively, whereas the lowest significant values of hemolysis inhibition were observed with all extracts of olive meal. The acetone extract of apricot kernel exhibited the most significant suppression of hemolysis, followed by mango kernel, with values of 87.33±1.2 and 83.06±0.3, respectively.

Table 6 Hemolysis Inhibitions (%) of different extracts.

Treatment	Ethanol	Acetone	Hexane
Asprin	90.23±0.28 [a]	90.23±0.28 [a]	90.23±0.28 [a]
Olive meal	10.27±0.29 [i]	15.79±42 [g]	20.4±0.7 [g]
Fig opuntia peel	64.45±0.3 [c]	65.67±1.4 [d]	36.33±0.8 [d]
Date seed	53.64±0.32 [e]	55.67±0.88 [e]	76.32±0.57 [b]
Avocado peel	29.33±1.4 [f]	50.36±0.14 [f]	23.55±0..3 [f]
Avocado seed	26.48±0.3 [g]	51.57±0.31 [f]	24.37±0.38 [ef]
Mango peel	59.37±0.43 [d]	56.4±0.9 [e]	25.14±0.6 [e]
Mango kernel	23.67±0.88 [h]	83.06±0.3 [c]	19.76±0.43 [h]
Apricot kernel	69.60±0.31 [b]	87.33±1.2 [b]	75.27±1 [c]

Toxicity by Artemia salina

The toxicity of herbal extracts, shown by LC_{50} values, was typically assessed concerning either Meyer's or Clarkson's toxicity index. Data in Fig.7 and Table 7. indicate that the most toxic extracts were found in the acetone extracts of avocado seed and mango kernel. On the other hand, the hexane extracts of Barbary fig peels, mango peels, and date seeds showed the highest significant non-toxic effects, which had IC_{50} (2725.85±3.31, 1440.33±2.02 and 1153.39±2.42), respectively. Our findings regarding avocado seed indicated a highly toxic acetone extract at 49.81±0.98 µg/mL, while the ethanol and hexane extracts exhibited medium toxicity levels of 183.4±1.25 and 141.67±1.81 µg/mL, respectively. The hexane and ethanol extracts of mango kernel, as well as the ethanol and acetone extracts of mango peel, were of medium toxicity, while the acetone extract was highly toxic in mango kernel (49.97±0.88) and the hexane extract in mango peel has no toxicity (1440.33±2.02).

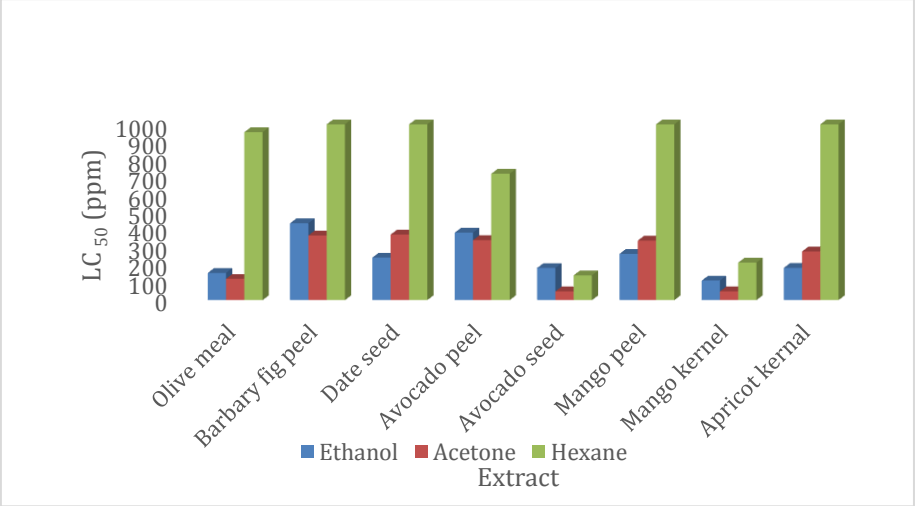

Fig. 7 Brine shrimp toxicity (LC_{50}) of different extracts.

Table 7 Brine shrimp toxicity (LC_{50}) of different extracts µg/mL.

Extracts	Ethanol	Acetone	Hexane
Olive meal	155.14±1.45 [f]	121.24±1.51 [d]	955.51±1.71 [e]
Barbary fig peel	438.78±1.03 [a]	369.06±0.88 [a]	2725.85±3.31 [a]
Date seed	242.66±1.48 [d]	374.04±2.02 [a]	1153.39±2.42 [c]
Avocado peel	384.95±1.23 [b]	342.467±1.41 [b]	720.08±1.98 [f]
Avocado seed	183.4±1.25 [e]	49.81±0.98 [e]	141.67±1.81 [h]
Mango peel	263.63±2.11 [c]	339.6±1.98 [b]	1440.33±2.02 [b]
Mango kernel	111.58±0.95 [g]	49.97±0.88 [e]	214.02±2.03 [g]
Apricot kernel	184.03±1.81 [f]	277.42±2.38 [c]	1000±1.21 [d]

Correlation between the studied variables

Correlation between antioxidant activity, phenol, and flavonoids

Pearson's correlation coefficient was calculated based on the results of each pair of variables, DPPH and ABTS (IC_{50}). It was expressed as the reciprocal of the calculated IC_{50} values, phenol compounds, flavonoid content, and FRAP normal value. the correlation matrix was calculated (Fig 8). As reported in Table 8, There was a highly positive correlation between the total phenolic content, 1/ABTS, 1/DPPH, and flavonoids (r = 0.989, r=0.959, and r=0.946, respectively); as well as between 1/DPPH and 1/ABTS (r= 0.936) and between total flavonoids and 1/ABTS (r=0.923).

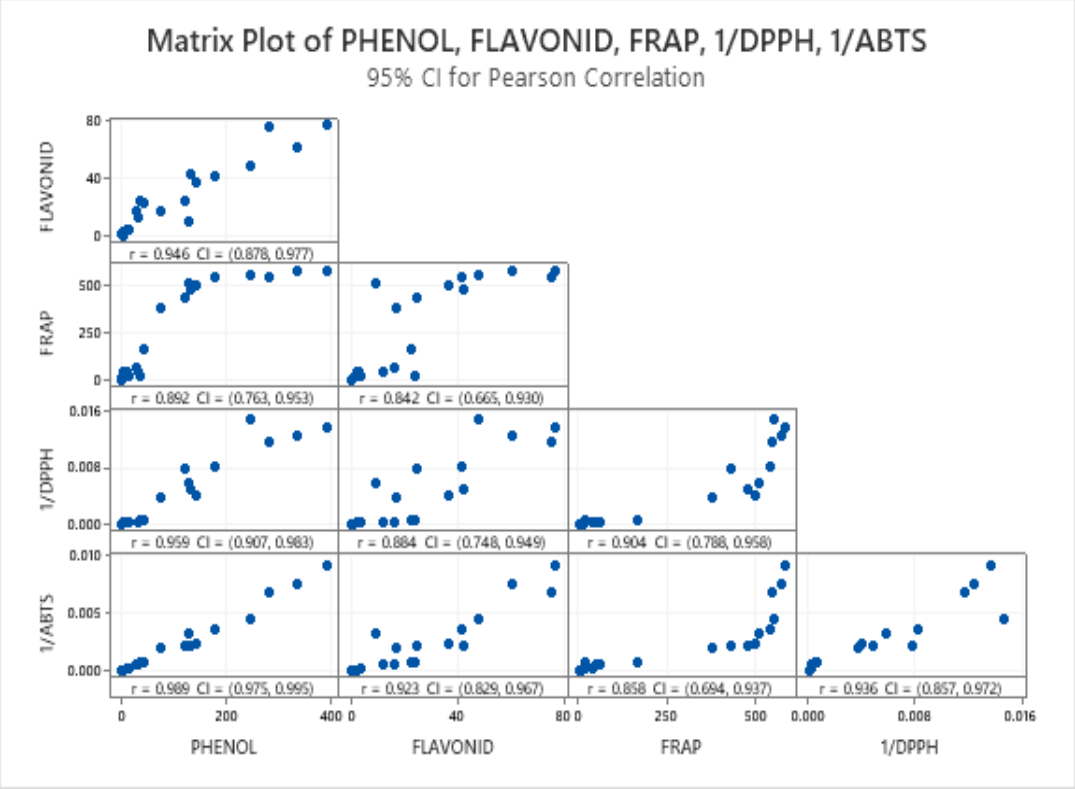

Fig. 8 Correlation between antioxidant, phenolic and flavonoids.

Table 8 Correlation between antioxidants, phenol, and flavonoids.

	PHENOL	FLAVONOID	FRAP	1/DPPH
FLAVONOID	0.946			
FRAP	0.892	0.842		
1/DPPH	0.959	0.884	0.904	
1/ABTS	0.989	0.923	0.858	0.936

Correlation between DPPH scavenging and toxicity by Artemia salina

The correlation matrix between DPPH and toxicity was calculated, as shown in Fig. 9, and a moderate positive correlation between DPPH and toxicity was found (r=0.744).

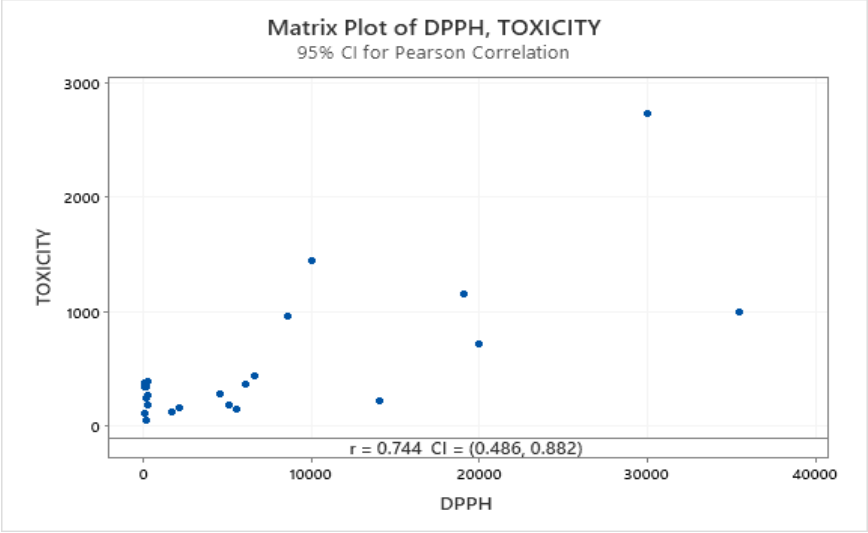

Fig. 9 Correlation between DPPH and toxicity.

Discussion

Fruit by-products are a good source of vitamins, fibers, antioxidants, and other nutrients and bioactive compounds. Therapeutic properties of fruit by-products could be used to make them even more valuable, and they could help avoid diseases like cancer, diabetes, heart disease, and obesity [47] Natural products have significant medicinal characteristics associated with low toxicity and high efficacy. Consequently, the conversion of food industry waste into value-added products promotes the principles of a circular economy [48].

The yield of the obtained extracts using polar solvents might vary from moderate to high, depending on the specific extraction conditions. Apricot kernels contain a substantial quantity of oil, which can be effectively extracted using hexane due to their nonpolar properties and ability to break down lipids efficiently [49].The findings align with the study conducted by [50], which indicated that the extraction yield from mango kernels employing a 96% ethanol solution varied between 11.86% and 22.22%. The study conducted by [51] investigated the impact of solvent type and extraction time on the extraction yield of mango seeds. The results showed that ethanol had the highest yield, measuring 19.86, compared to hexane and petroleum ether, which yielded 9.85 and 8.75, respectively, during the 3-hour Soxhlet extraction process. The study conducted by [52] revealed that the yield of *Opuntia ficus-indica* (Barbary fig) peels extracted using sequential extraction methods ranged from 2.5 to 64.74 µg/100 g. Among the different solvents used, ethanol exhibited the greatest extract yield at 64.74%, followed by methanol extracts at 50.7%. However, the

extraction yield of date seeds using the hexane with ultrasound method was 6.18%, according to [53].

In phytochemical screening, these results coincide with [54], who determined that mango peel extracts' phytochemical chemicals of aqueous and ethanol contain alkaloids, flavonoids, tannins, and saponin compounds. [50] who revealed that 96% ethanol extract of all seven mango cultivars seed extracts contains phenolics, flavonoids, tannins, alkaloids, triterpenoids, steroids, and saponins. [55] Indicated that the phytochemical test results showed that the seed extract of avocado contains phenols, flavonoids, tannins, alkaloids, saponins, and quinones.

In this study the concentration of the total phenolic compound (TP) of mango by-product extracts fell within the range that was reported in a previous study by [56] who found that the ethyl acetate extract on different varieties of peels had higher TPC levels, ranging from 52.67 to 275.61 mg GAE/g DE, followed by the seed kernels with levels ranging from 132.95 to 270.56 mg GAE/g DE. In another study by [57] who investigated the 80% ethanol extract of mango peel for phenol and flavonoid content and revealed that the compounds they found its varied from 55 to 100 mg/g and 26.9±3.76 mg GAE/g of the extract respectively. A study conducted by [58] who used different ratios of ethanol:water to extract the total phenol content of mango peel and found varied concentrations from (70.73 to 127.34 mg GAE/g DE).

The outcomes of our investigation on avocado peel extract closely align with the findings reported by [59] who found that the total phenolic content of the avocado peel extract was greater when acetone-water solvent was used (208.5±19.8 mg GAE/g DE) compared to the extract with ethanol and ethanol-water solvent (183.4 ± 6.0, and 192.6±11.1 mg GAE/g DE) respectively. However, the total phenolic content (TPC) in date seed extracts is within the range reported by [60] who found that the total phenolic content varied from 138.92±1.75 to 381.29±2.31 mg Gallic acid equivalent per gram of dry extract. The concentration of total flavonoid (TF) is similar to these findings, which ranged from 46.74±1.26 to 163.53±1.82 mg quercetin equivalent per gram of dry extract.

These results showed that the high-polarity solvents mean a higher amount of phenolic compounds obtained. These findings may be because the efficiency of polar solvents to extract phenolic compounds, such as aqueous methanol/ethanol was higher than absolute methanol/ethanol [61].

Food's antioxidant activity is determined by its antioxidants' individual actions, which can combine and provide synergistic or inhibitory effects [62]. The concepts of the assay and the testing settings vary among different methodologies. No singular assay sufficiently evaluates all antioxidants within a mixed or complex system due to the frequent involvement of various response characteristics and mechanisms. The utilization of multiple techniques has been shown to enhance confidence in determining the overall antioxidant capacity [63].

[64] Tested the avocado by-product properties and found that hydroalcoholic peel and seed extracts are excellent sources of antioxidants. Nevertheless, the avocado peel demonstrated superior bioactivity compared to the seed, exhibiting exceptional antioxidant properties. While [65] found that the EC_{50} values for the DPPH and ABTS radicals were 72.64 ± 10.70 and 181.68 ± 18.47µg/mL, respectively, when tested with the 80% ethanol extract of avocado peel extract. In the study of [66] who used methanol, acetone, and diethyl ether as solvents, and found that the acetone extract contained the most chlorophylls and polyphenols, as well as the most antioxidant activity as measured by DPPH and ABTS tests.

[67] Found that mango kernels contained more phenolic compounds compared to mango peels, and this is in agreement with our results, However, when comparing the antioxidant activity of mango peel extract with that of mango kernel extract against DPPH, it was found that the former had far higher activity, and this is opposite to our results. In another study by [68] who investigated five types of mango cultivars that had a considerable amount of antioxidants, phenols, and flavonoids, and reported that the aqueous extract showed antioxidant activity (IC_{50} of DPPH ranged from 65.34±0.62 to 169.83±0.89 µg/ml, and IC_{50} of ABTS ranged from 28.29±0.43 to 84.88±0.79 µg/ml), and these are supporting our findings. The present results of the antioxidant assay of date seed showed that acetone extract was better than other extracts; these results are in agreement with those obtained by [69], who found that the highest values of antioxidant activity and antioxidant components of date seeds presented 70% acetone, followed by 80% methanol.

The antioxidant potency composite index (ACI) was computed to assign equal significance to all these methodologies. PCA was applied to understand more about variables and variations between extracts based on their phenolic, flavonoid contents, and antioxidant activity [1/ABTS, 1/DPPH, and FRAP]. This technique serves as a valuable tool for understanding much more about explaining results from the research study and the effects of different solvents on them. In PCA the outlier causes are measurement error, wrong labeling, noise, or the most interesting sample [70] Since the outlier in this instance has the greatest ACI value, we propose that it is the most intriguing sample. [71] found that the antioxidant activity was

greater for the avocado peel, followed by seed and pulp, in PCA analysis.

The finding of antimicrobial activity is consistent with the results reported by [72], who tested extracts of mango seeds using methanol and ethanol and found that they were effective against 25 different microorganisms, including acid-fast, Gram-positive, and Gram-negative bacteria. The inhibitory effects of the extracts varied, resulting in different sizes of inhibition zones for each tested organism. Only *Bacillus cereus* and *Rhodococcus equi* exhibit resistance to these extracts. The inhibition zones generated by various extracts were smaller than those produced by the positive control medication. The possible cause for this phenomenon might be attributed to the reduced rate of diffusion of mango extract inside the agarose medium. The antibacterial activity and inhibition zone width may have been influenced by parameters associated with the diffusion of the substance in agars, such as Mueller Hinton's agar. [73] their study discovered that the acetone extract of mango *Mangifera indica Linn* exhibited more activity than methanol against all the Gram-positive bacteria, resulting in zones inhibition ranging from 15 to 16 mm. Additionally, the acetone extract showed a zone of inhibition of 14 mm against the Gram-negative bacterium *Salmonella typhi* at a concentration of 250 mg/mL.

[54] Conducted a study to evaluate the antimicrobial effects of hot and cold aqueous extracts and ethanol extracts of dried mango (*Mangifera indica L.*) peel. The extracts were tested against medically important pathogens isolated from animals and chicken farms, including bacteria such as *Pseudomonas aeruginosa, Bacillus cereus***,** *Staphylococcus aureus, Escherichia coli*, and the fungus *C. albicans*. The antimicrobial efficacy was evaluated by the agar well diffusion technique. The ethanol peel extracts have exhibited substantial antibacterial and antifungal efficacy against all the examined pathogens, whereas the hot aqueous extract had strong antibacterial and antifungal properties.

A recent study by [74] demonstrated that the methanol extract of Ajwa date seed at a dosage of 100 mg/mL displayed inhibitory effect against several bacteria. The inhibition zones for *S. aureus* and *E. coli* were 19 mm, and 15 mm respectively. In another study by [71] who reported the MIC of methanol extracts against *S. aureus* and *E.coli* was 25 mg/mL. They found the best results of MIC were recorded for the ethanol extract of avocado peel followed by seed, which was 0.625 mg/mL against the bacteria *S. aureus* and 5 mg/mL against *B.cereus , Salmonella* and *E.coli.* [75] who studied the MIC of mango seed extracts and found inhibition of many organisms at 1.25 mg/mL for methanol and ethanol extracts of mango seeds against (*S. aureus, Salmonella* and *E.coli*); 3.125 mg/mL against *Aspergillus niger* and 1.785 mg/mL against *Candida albicans***.** As per with [71], various avocado ethanolic peel extracts exhibited superior antioxidant and microbiological action compared to seeds.

Erythrocyte membranes, or human red blood cells (HRBC), have similarities to lysosomal membranes [76,77]. Stabilizing the lysosomal membrane is essential for suppressing the inflammatory response by preventing the release of lysosomal constituents from activated neutrophils, including bacterial proteases. These proteases may induce further tissue inflammation and extracellular release [78,79]. Due to the similarity in components between HRBC membranes and lysosomal membranes, the ability to inhibit HRBC membrane lysis caused by hypotonicity is considered an indicator of the anti-inflammatory action of the studied extracts [80]. Aspirin serves as a positive control since aspirin is an anti-inflammatory drug that can prevent the release of inflammatory mediators, one of which is the inactivation of cyclooxygenase (COX) in prostaglandin synthesis [81].

This investigation demonstrated that most acetone extracts greatly stabilized HRBC membranes and efficiently reduced heat-induced hemolysis, showing the anti-inflammatory properties of the phytoconstituents within the extracts. The apricot kernel had the most significant effect with various solvents, corroborating the findings of [82], who suggested that apricot kernel extracts, both with and without oil, warrant further mechanistic and clinical studies as a supplementary treatment for inflammatory bowel diseases. A further investigation conducted by [83] revealed that the extract derived from apricot seeds had anti-inflammatory properties, which can be linked to its ability to suppress the production of pro-inflammatory cytokines and COX-2 and reduce prostaglandin formation. [84] reported that the amygdalin found in apricot kernels can block TNF-α and IL-1β due to its ability to regulate the transcriptional production of pro-inflammatory cytokines. [74] demonstrated that the methanolic extract of date fruit pulp and seed extract exhibited potent antioxidant capabilities, stabilizing the RBC membrane and inhibiting heat-induced hemolysis by 63.84% and 58.10%, respectively, consistent with the effects of acetone and ethanolic extracts. Our result are in harmony with those reported by (56), who revealed that the ethyl acetate extract from the mango peel had the most potent anti-inflammatory action compared to the kernel and pulp. The peel extracts from the three mango varieties exhibited distinct levels of hemolysis

inhibition, with IC_{50} values ranging from 151 µg/mL to 197 µg/mL.

In a toxicity study by [85] who reported that the LC_{50} value of the ethanol extract of mango (*Mangifera indica Linn*.) was observed to be 41.6869 µg/mL, signifying its toxicity. The LC_{50} value obtained from the ethanol extract of kweni mango rind (*Mangifera odorata Griff*.) was established at 29.4238 µg/mL, signifying a categorization of high toxicity. Alternatively, [86] evaluated the toxicity of *Mangifera indica L*. seed and kernel oil, discovered 50% lethal concentration (LC_{50}) values between 5122.11 ppm and 7663 ppm. The research indicated that the oils were non-toxic. Our results regarding avocado seeds are in contrast with those stated by [87], who assessed the toxicity of avocado seeds on *Artemia salina*, reporting LC_{50} values for hexane and methanol extracts of 2.37 and 24.13 mg mL^{-1}, respectively.

Numerous investigations [88–90], identified a robust positive link between the LC_{50} of brine shrimp and the anticancer efficacy in the formulation of novel phytopharmaceuticals. This relationship has been acknowledged as an efficient prescreening method for cytotoxicity and antitumor assays. [91,92] established a robust correlation between the LC_{50} and LD_{50} of identical plant extracts on brine shrimp larvae, and delivered orally to mice.

The strong positive correlation observed between the total phenolic content and the inverse of `DPPH, ABTS, and FRAP indicates that phenolic compounds make a substantial contribution to the antioxidant activities of these species. Consequently, they may play a crucial role in the beneficial effects of these significant medicinal plants. Multiple studies have discovered that phenolic compounds are the primary elements responsible for the antioxidant properties found in specific plants. Furthermore, there is a clear correlation between the antioxidant activity and the overall phenolic concentration of these compounds. [93,94].

Phenolics have strong antioxidant properties such as free radical scavenging, O_2 scavenging, and Fe^{+2} chelation. Hydroxyl groups in phenolic compounds can donate hydrogen atoms. This is combined with (reactive oxygen species) ROS and (reactive nitrogen species) NOS to end the oxidation reaction, preventing the generation of new radicals. Polyphenols with distinct properties are produced by the reaction of phenolic hydroxyl groups and benzene ring electrons [95]. In addition, phenolics have the ability to form complexes with Fe^{+2} ions, which then interact with the free radicals that are generated. Polyphenolics have a significant affinity for proteins and may readily establish hydrogen bonds, mostly because of the hydrophobic nature of the benzene ring. Phenolics possess features that enable them to function as antioxidant substances and hinder the action of enzymes that generate free radicals, including some xanthine oxidase, cytochrome P450 isoforms, cyclooxygenase, and lipoxygenases [95,96].

Flavonoids possess a notable capacity to directly eliminate reactive oxygen species [ROS] and capture free radicals immediately after their formation by donating hydrogen or transferring a single electron. They can also seize or attach to metal ions in the human body, preventing them from being available for oxidation. Flavonoids are capable of trapping small amounts of metal ions like Fe^{+2} and Cu^{+} and play a crucial role in oxygen metabolism and the creation of free radicals. Additionally, flavonoids can function as antioxidants within cells by inhibiting the activity of enzymes involved in the production of free radicals [97].

Studies have demonstrated different levels of association between antioxidant activity and cytotoxicity. However, the link with brine shrimp cytotoxicity was only minor [98]. This implies that although antioxidants may contribute to cytotoxic effects, other variables also have a substantial impact [99]. Gaining insight into this link facilitates the identification of plant extracts that exhibit both high antioxidant activity and controllable toxicity levels, rendering them viable candidates for future advancement in pharmaceuticals and nutraceuticals [100].

Conclusion

In vitro assays of 24 extracts of by-product fruit were performed. Different determination methods were used [total phenolic content, total flavonoids, FRAP, ABTS and DPPH values]. Mango seed, avocado peel, and date seed proved to have the highest mean values for all methods. The antioxidant potency composite index was calculated and based on the overall antioxidant index. Antimicrobial activity and anti-inflammatory assays showed that the acetone extract from mango seed had the highest antioxidant activity. Therefore mango, avocado, and date by-products, such as peels and seeds, could be considered as important bioactive sources appropriate for utilization in nutritional/pharmaceutical fields. However, further evaluation of their bioactive compounds, and antioxidant activities in living models is required.

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Declaration of competing interest

The authors assert that they possess no identifiable competing financial interests or personal affiliations that may have seemingly influenced the work presented in this study.

Authors' contributions

H. M.: Conceptualization, Methodology, and Writing M. A. M.: Review and Supervision. Gh. I. M.: Review, editing, and Supervision. A. A.: Supervision. M. A.: Data curation, Visualization

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