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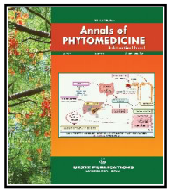

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Phytochemical analysis and antioxidant activity of *Manilkara zapota* L. peel

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Abstract

Some fruit peels are widely used to combat oxidative stress because they are known to be abundant in antioxidants. *Manilkara zapota* L. peel has many nutraceutical phytochemicals, such as polyphenols, carotenoids, sterols, saponins, terpenes, and vitamins, which are what give them their health-promoting qualities. Ascorbic acid, carotenoids, and phenolics are examples of phytochemicals that may directly affect their capacity to scavenge radicals. The current study compares the phytochemicals (total phenols, flavonoids, total sugar, reducing sugar, and non-reducing sugar) and *in vitro* antioxidant activities in *M. zapota* peel extracts prepared with methanol and ethyl acetate.

A variety of phytochemicals are examined in methanol and ethyl acetate extract of peel, including total phenolic content (29.791 ± 0.373 and 9.894 ± 0.326 mg GAE/g), total flavonoids (6.876 ± 0.200 and 3.352 ± 0.260 mg CE/g), total sugars (606.188 ± 0.244 and 139.955 ± 0.159 mg/g), reducing sugars (343.425 ± 0.56 and 88.307 ± 0.290 mg/g), and non-reducing sugars (262.103 ± 0.221, and 50.847 ± 0.178 mg/g). The antioxidant activity of methanol and ethyl acetate extract was analyzed *via* the FRAP assay (10.18 mMFe(II)/ml and 8.27 mM Fe(II)/ml), total antioxidant capacity (0.54 mg AAE/ml and 0.51 mg AAE/ml), DPPH free radical scavenging activity (IC_{50} 93.782 µg/ml and 309.005 µg/ml), and ABTS free radical scavenging activity (IC_{50} 96.52 µg/ml and 305.07 µg/ml). These results find that *M. zapotais* a good source of phytochemicals and minerals with pharmaceutical and good physical condition.

1. Introduction

A wealth of medicinal plants can be found in nature and a variety of natural foods with potentially beneficial bioactive components have long been used as herbal remedies for a wide range of serious illnesses. Because of their potential bioactive compounds, medicinal plants are used to attend to a wide range of illnesses, including cancer, oxidative stress, diabetes mellitus, obesity and hypertension, skin care, neurological complaints, and inflammations (Moond *et al*., 2023). They are also used to control the expression of different genes (Islam *et al*., 2020; Devi *et al*., 2023; Moond *et al*., 2023). Research on phytochemicals aims to find new uses for plants that can be used to make medications. The health benefits of phytonutrients are numerous (Beniwal *et al*., 2023, Dalal *et al*., 2022; Moond *et al*., 2023). Plants are valuable because of their phytochemical components having special physiological properties. Primary components, which include protein, sugars, chlorophyll, amino acids, and so forth, and secondary components, which include terpenoids, alkaloids, essential oils, saponins, tannin, phenolic, and flavonoid compounds, among others, are the two chief classifications of phytochemicals, which are non-essential nutrients in the form of chemical compounds that come from plants (Devi *et al*., 2023; Moond *et al*., 2023).

M. zapota a member of the Sapotaceae family, is locally known as sofeda in Bangladesh and is usually referred to as sapodilla/chikoo/ sapota in India. This is an evergreen, long-living plant that is typically found in Central America, southern Mexico, and India. *M. zapota* leaves, fruit peel, seeds, fruits, and bark have all been the subject of studies on the phytochemical characterization and evaluation of biological activities, such as antititrosinase, antioxidant, antihyperglycemic, antimicrobial, antidiabetic, hypocholesterolemic, and properties (Islam *et al*., 2020). One fruit that can be eaten both with and without a peel is the sapota. Since the sapota peel has no astringent properties, it can be eaten with the peel. Fruit peels have been shown in numerous studies to contain higher levels of ascorbic acid, antioxidant activity, and other antioxidant enzymes than fruit pulp (Bala and Kumar, 2017). The bark is a tonic, and a decoction is administered for preludes, diarrhea, and dysentery. Additionally, *M. zapota* bark has long been used to treat inflammatory conditions, fever, pain, and gastrointestinal disorders.Traditional medicine uses the leaves of the plant to treat coughs, diarrhoea, and colds. The leaves also possess hypocholesterolemic, antihyperglycemic, and analgesic properties (Gomathy *et al*., 2013).

Figure 1: *M. zapota* plant.

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The *M. zapota* fruit and peel are rich in saponin, a substance with astringent qualities similar to those of tannin. A good amount of quercetin and kaempferol are present in sapota peel. Known for its anti-inflammatory, antihypertensive, vasodilator, antiobesity, antihypercholesterolemic, and antiatherosclerotic properties, quercetin is a significant bioflavonoid (David *et al*., 2016). In order to prevent the deposition of amyloid fibrils (such as Aβ, tau, and α-synuclein), inhibit microglia activation, reduce the release of inflammatory factors, restore the mitochondrial membrane to prevent oxidative stress, protect the blood-brain barrier, and inhibit specific enzyme activities (such as cholinesterase), kaempferol and its derivatives play a major role in neuroprotection (Jin *et al*., 2023). Caffeic acid is present in leaves of *M. zapota* and although caffeic acid's structure which includes an aromatic core, a conjugated double bond, and hydroxyl groups allows it to act as an antioxidant, its benefits go far beyond that. The data that have been published demonstrate effects on cancers of different kinds, diabetes, obesity, and neurodegenerative illnesses like Parkinson's and Alzheimer's (Pavlíková, 2022).

2. Materials and Methods

2.1 Collection of plant material

The fully ripe *M. zapota* fruits were harvested for their peel from the orchard of Chaudhary Charan Singh Haryana Agricultural University in Hisar (29º.15' N latitude and 75º.69' E longitude).The collected sample was verified by Dr. Anita, Asstt. Scientist, Department of Botany and Plant Physiology, CCS HAU, by using online platform (Tropicos & IPNI). Plant parts were cleaned, and then dried at room temperature in the shade.

2.2 Preparation of methanol and ethyl acetate extract of *M. zapota* fruit peel for phytochemical analysis

10 g of powdered *M. zapota* peel was collected and placed in a Whatman No. 1 filter paper thimble. This thimble was positioned inside a 500 ml flask with a circular bottom, which works for the majorpart of the standard Borosil Soxhlet apparatus. A total of about 250 ml or more of methanol and ethyl acetate were added in up to 1.5 syphons. As a result, the powdered peel sample was percolated using methanol and ethyl acetate as the solvent in a Soxhlet utensil. The total sugars, total phenolic content, reducing sugars, total flavonoids and non-reducing sugars, and capacity were all measured using this methanol and ethyl acetate extract.

2.3 Quantitative analysis of phytochemicals

2.3.1 Total phenolic content

The total phenolic content of the methanol and ethyl acetate, *M. zapota* peel was measured using the Folin-Ciocalteu method (Singleton and Rossi, 1965), with gallic acid serving as the reference standard. A test tube was filled with 1.0 ml of the Folin-Ciocalteu reagent, 2.0 ml of Na_2CO_3 (20%, w/v), and 1.0 ml of each of the extracts of methanol and ethyl acetate. Following the mixing, water was added to reach a final volume of 10.0 ml. The reaction mixture was then centrifuged for 10 min at 6200 rpm following an incubation period of 10 min. The absorbance of the supernatant solution was measured at 730 nm using a UV-Vis spectrophotometer. The results were contrasted with a blank that was made in the same way, but with the correct solvent used in place of the extract. The total phenolic content of the ethyl acetate and methanol peel extract of *M. zapota* was determined and expressed as mg GAE/g using a regression equation derived from the gallic acid standard curve.

2.3.2 Total flavonoid content

1.0 ml of ethyl acetate and methanol extract and 0.3 ml of 5% $NaNO_2$ were added to a test tube and further add 4.0 ml of distilled water and six min later, 2.0 ml of 1M NaOH and 0.3 ml of 10% $AlCl_3$ were added. Lastly, dilutions were added, containing 10.0 ml of distilled water. Using an $AlCl_3$ colorimetric test, the total quantity of flavonoids was determined using catechin as a reference (Marinova *et al*., 2005). Using a UV-Vis double beam spectrophotometer, the mixture's absorbance at 510 nm was measured in comparison to a blank that was made following the same steps, but with methanol and ethyl acetate in place of the extract. The total amount of flavonoids in the peel extract was calculated using a regression equation created from the catechin standard curve and expressed as mg CE/g.

2.3.3 Total sugar

Utilizing a modified version of Dubois's method (Dubois *et al*., 1956), total sugars were calculated. The *M. zapota* were extracted in methanol and ethyl acetate in a test tube with 1 ml, and 2.0 ml of phenol solution was then added. 5.0 ml of concentrated H_2SO_4 was then added right away to the reaction mixture. After allowing the solution to cool for thirty min, the absorbance of the reaction mixture at 492 nm was measured using a UV-Vis double-beam spectrophotometer. This was done in contrast to a blank that was prepared in the same way, but with methanol and ethyl acetate instead of the extract. Using the regression equation developed from the D-glucose standard curve, the total quantity of sugars in the *M. zapota* peel extract was deliberated and the results were expressed as mg/g.

2.3.4 Reducing sugar

To determine the amount of reducing sugars in peel extract, the Nelson method modified by Somogyi (1952) was employed (Nelson, 1944). In a test tube containing one ml of methanol and ethyl acetate extract of the peel of *M. zapota*, one ml of distilled water was added. After adding the alkaline copper reagent and fully mixing the mixture, it was heated for 20 to 25 min in a hot water bath. Following room temperature and thorough mixing, the arsenomolybdate reagent was poured into the boiling tubes and diluted to a volume of 10.0 ml using distilled water. A UV-Vis double beam spectrophotometer was used to measure the absorbance of the reaction mixture at 520 nm. The absorbance of the blank was prepared in the same way, but 1.0 ml of distilled water was used instead of extract. Using the D-glucose standard curve, the amount of reducing sugars in the methanol and ethyl acetate extract was calculated and expressed as mg/g.

2.3.5 Non-reducing sugar

The difference between the concentration of total sugars and that of reducing sugars was used to calculate the non-reducing sugars:

Nonreducing sugars = Total sugar – Reducing sugar

2.4 Evaluation of antioxidant activity

2.4.1 DPPH free radical scavenging activity

The Hatano *et al*. (1988) methodology was utilised to evaluate the DPPH free radical scavenging activity. 2.0 ml of 2, 2'-diphenyl-1-picrylhydrazyl radical (0.1 mM) and 1 ml of methanol and ethyl acetate extracts (at varying concentrations) were combined, and the

mixture was actively stirred for 2 min to measure the antioxidant activity. Using a UV-VIS spectrophotometer, the absorbance of the sample and control was determined at 518 nm following half an hour dark incubation period at room temperature. This formula used to determine the percentage of DPPH scavenged was:

$$\% \text{ DPPH free radical scavenging activity} = \left[Ac - \frac{As}{Ac} \right] \times 100$$

where,

$A_{control}$ = Absorbance of control ; A_{sample} = Absorbance of sample

2.4.2 ABTS assay

Firstly ABTS radical cation was made by mixing 5 ml of ammonium sulphate (2.45 mM) with 5 ml of ABTS (7 mM) and keeping the solution incubated for 16-18 h. ABTS solution was diluted to get an absorbance of nearly 0.7 ± 0.02 at 734 nm. Various concentrations (0.2 ml) of each sample (methanol and ethyl acetate extract) were combined with ABTS radical solution and incubated at the ambient temperature for 4 min. A UV-VIS spectrophotometer was used to estimate the absorbance of the sample and a control at 734 nm and BHA were used as standard.

The formula used to determine the percentage of ABTS scavenged was:

$$\% \text{ ABTS free radical scavenging activity} = \left[Ac - \frac{As}{Ac} \right] \times 100$$

where,

$A_{control}$ = Absorbance of control; A_{sample} = Absorbance of sample

2.4.3 FRAP assay

The FRAP reagent was prepared by mixing 300 mM of acetate buffer (pH 3.6), 20 mM $FeCl_3.6H_2O$, and 10 mM TPTZ in 40 mMHCl in a 10:1:1 ratio. This FRAP reagent (3 ml) was combined with different concentrations of each sample (0.1 ml), and it was then incubated at 37ºC for 5 min. Using a UV-VIS spectrophotometer and $FeSO_4.7H_2O$ as the standard, the absorbance of the control and sample was measured at 593 nm.

2.4.4 Phosphomolybdnum assay

Prieto *et al*. (1999) employed the modified phosphomolybdenum method to ascertain the overall antioxidant capacity of *M. zapota* peel extracts. Glass vials holding 1 ml of each peel extract were filled with 3 ml of phosphomolybdenum reagent. The mixture was then well combined and sealed and for 90 min, they were incubated at 95°C. The contents of the vial were then measured at 695 nm using a Shimadzu UV-VIS Double Beam Spectrophotometer.

2.5 Statistical analysis

For statistical analysis, the samples were collected in threes. The statistical software SPSS (Statistical Package for Social Sciences) version 23 was used to present the data regarding proximal composition and phytochemicals as mean standard error (SE). Using Origin 2018, the IC_{50} values for antioxidant activity were subjected to a regression analysis.

3. Results

3.1 Phytochemical analysis

A quantitative analysis was conducted on the methanol and ethyl acetate peel extract of *M. zapota* to quantify the amounts of different phytochemicals, including total sugars, reducing sugars, total flavonoids, and total phenolic content. Table 1 displays the results.

Table 1: Phytochemicals in methanol and ethyl acetate extract of the *M. zapota* peel

S. No.	Phytochemicals	Methanol extract	Ethyl acetate extract
1.	Total phenolic content (mg GAE/g)	29.791 ± 0.373	9.894 ± 0.326
2.	Total flavonoid (mgCE/g)	6.876 ± 0.200	3.352 ± 0.260
3.	Total sugar (mg/g)	606.188 ± 0.244	139.955 ± 0.159
4.	Reducing sugar (mg/g)	343.425 ± 0.56	88.307 ± 0.290
5.	Non-reducing sugar (mg/g)	262.103 ± 0.221	50.847 ± 0.178

3.1.1 Total phenolic content

Gallic acid was used as the reference in a standard curve to estimate the total phenolic content of *M. zapota* peel. The regression equation demonstrated a linear relationship between the absorbance and gallic acid concentration (Figure 2). The total phenolic content of the *M. zapota* methanol and ethyl acetate extract was calculated using regression analysis (y = –0.0153 + 0.0103x, R^2 = 0.9954) and it was determined to be 29.791 ± 0.373 and 9.894 ± 0.326 mg GAE/g.

3.1.2 Total flavonoid content

The total amount of flavonoids contained in the ethyl acetate and methanol extract of *M. zapota* peel was calculated using a standard curve that used catechin as the reference compound (Figure 3). The outcomes of the regression equation indicate that there is a linear relationship between absorbance and catechin concentration. The ethyl acetate and methanol extract of the peel contained 3.352 ± 0.260 and 6.876 ± 0.200 mgCE/g of total flavonoids. Regression analysis was carried out using the equation (y = 0.0337 + 0.0034x, R^2 = 0.9927).

3.1.3 Total sugar content

A standard curve and D-glucose were used as the basis for comparison in order to estimate the total amount of sugar present in the ethyl acetate and methanol extract of the *M. zapota* peel (Figure 4). The regression equation demonstrated a linear relationship between absorbance and D-glucose concentration. The total sugar content of the peel's methanol and ethyl acetate extract was determined using the regression equation (y = 0.000013 + 0.0011x, R^2 = 0.9935). The sugar content was 606.188 ± 0.244 and 139.955 ± 0.159 mg/g, according to the results.

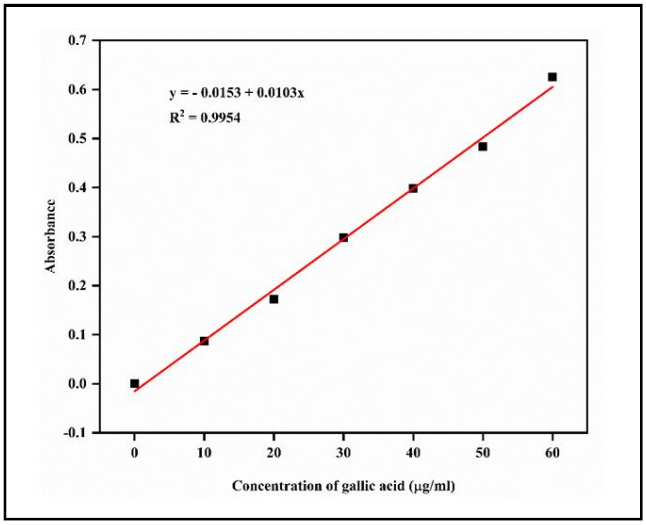

Figure 2: Gallic acid is used as the standard in the total phenol standard curve.

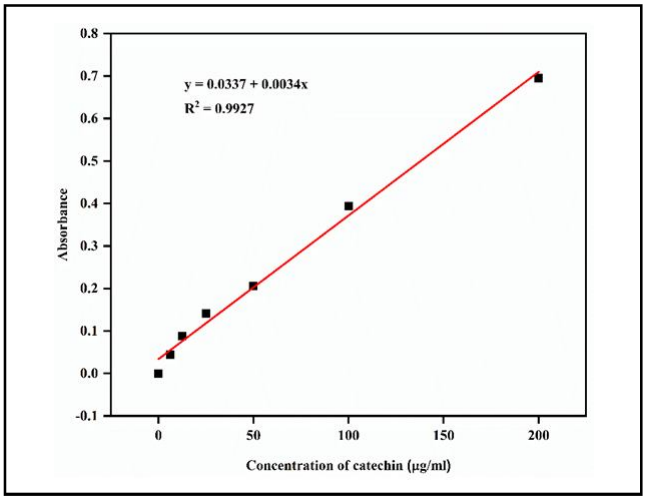

Figure 3: Total flavonoid standard curve with catechin as the reference.

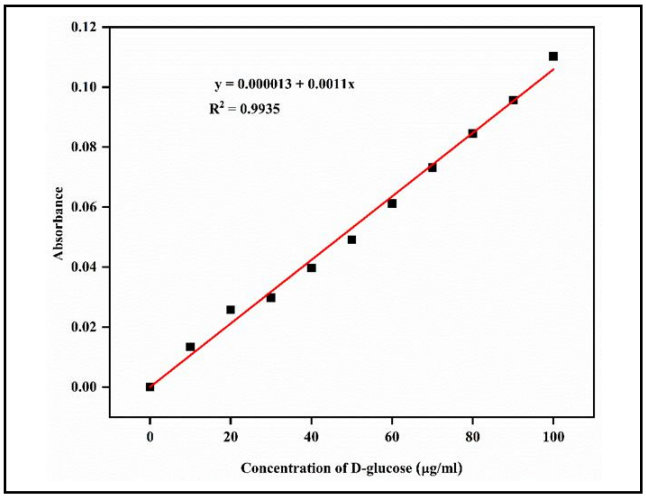

Figure 4: Total sugar standard curve with glucose as the reference.

3.1.4 Reducing sugar

The amount of reducing sugars in the methanol and ethyl acetate extract of the peel of *M. zapota* was determined by creating a standard curve with D-glucose as the reference (Figure 5).

The regression analysis's findings indicated that absorbance and D-glucose concentration had a linear relationship. The total sugar content of a methanol and ethyl acetate extract of peel was calculated using regression analysis (y = – 0.0119 + 0.00184x, R^2 = 0.9858). It was found to be 343.425 ± 0.56 and 88.307 ± 0.290 mg/g.

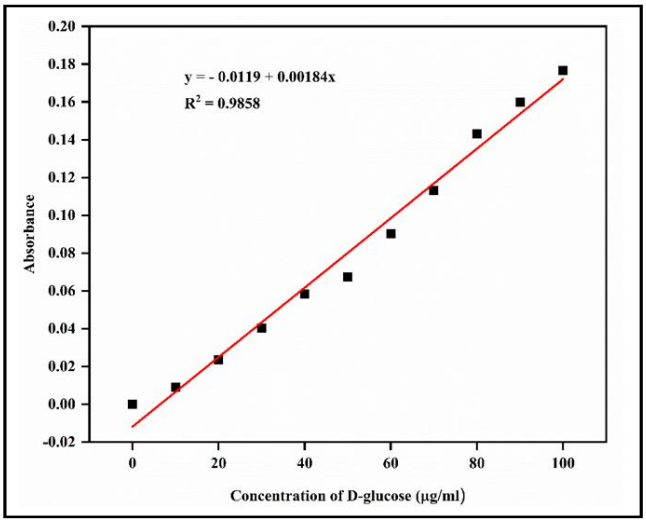

Figure 5: Standard curve for reducing sugarsusing glucose as the reference.

3.1.5 Non-reducing sugar

The difference in total sugar concentration and reducing sugar concentration was used to determine the amount of non-reducing sugars in the methanol and ethyl acetate extract of the peel of *M. zapota*. There were two found: 262.103 ± 0.221 and 50.847 ± 0.178 mg/g.

3.2 Antioxidant activity

3.2.1 DPPH free radical scavenging activity

Ascorbic acid to scavenge to DPPH free radical was 88.29% at concentrations of 120 µg/ml, 83.56, 70.98, 61.56, 48.38 and 28.45% at concentrationsof 100, 80, 60, 40, 20 µg/ml, respectively. With a concentration of 160 µg/ml, methanol peel extract exhibited the highest DPPH free radical scavenging activity 77.86, followed by concentrations of 71.05, 63.62, 52.59, 43.49, 32.817, 15.94, and 7.27%, in that order.

The ethyl acetate extract exhibited the highest DPPH free radical scavenging activity of 80.245 per cent at 700 µg/ml. This was followed by 76.89, 70.57, 61.29, 48.89, 31.39, and 18.23%, in that order. The IC_{50} of the methanol extract was 93.782 µg/ml, which was higher than the ethyl acetate extract's 309.005 µg/ml. Figure 6 displays the quadratic regression equation for the DPPH free radical scavenging activity's IC_{50} (µg/ml) value.

3.2.2 ABTS assay

BHA to scavenge to ABTS free radical was 92.58% at concentrations of 24 µg/ml, 84.36, 72.41, 59.79, 46.53, and 24.87% at concentrations of 20, 16, 12, 8, 4 µg/ml, respectively. At 160 µg/ml, methanol peel

extract exhibited the highest ABTS free radical scavenging activity, which was followed by concentrations of 77.37, 69.73, 57.82, 47.23, 35.41, 19.01, and 9.37%, respectively, were the next highest (Table 2). The maximum ABTS free radical scavenging activity of the ethyl acetate extract was 79.29% at 700 µg/ml, and the lowest values were 73.20, 64.81, 58.32, 46.29, 26.32, and 16.73%, in that order. The IC_{50} of the methanol extract was 96.52 µg/ml, which was greater than the ethyl acetate extract's 305.07 µg/ml. Figure 7 displays the quadratic regression equation for the ABTS free radical scavenging activity's IC_{50} (µg/ml) value.

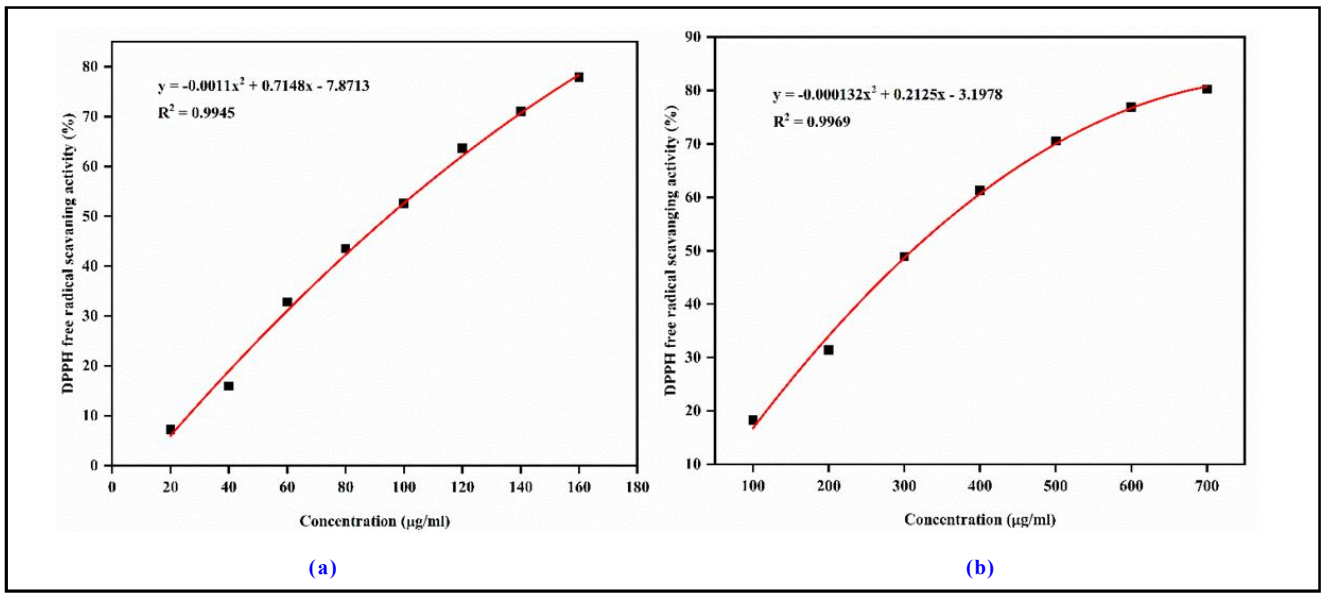

Figure 6: Quadratic regression equation for IC_{50} (µg/ml) of (a) methanol, (b) ethyl acetate peel extract DPPH free radical scavenging activity.

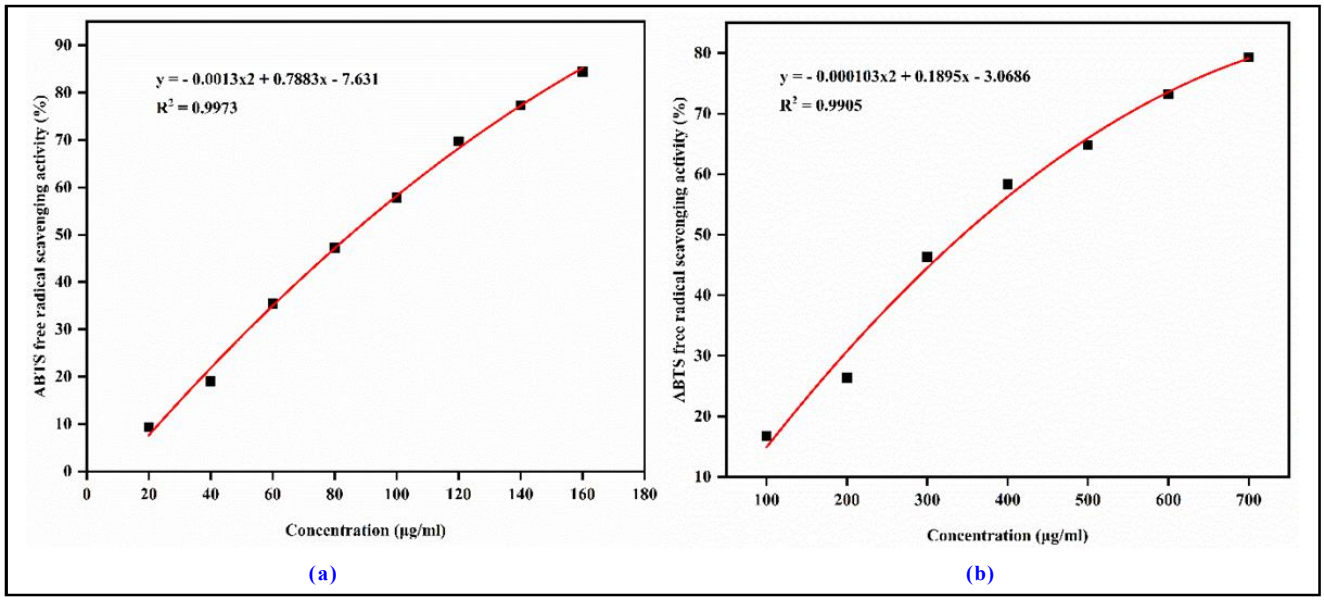

Figure 7: Quadratic regression equation for IC_{50} (µg/ml) of (a) methanol, (b) ethyl acetate peel extract ABTS free radical scavenging activity.

3.2.3 FRAP assay

FRAP tests of the different amounts of methanol peel extract (20, 40, 60, 80, 100, 120, 140, and 160 µg/ml) and ethyl acetate peel extract (100, 200, 300, 400, 500, 600 and 700 µg/ml) were estimateded by their reduction of a ferric 2,4,6-tripyridyl-s-triazine complex (Fe–TPTZ) to the ferrous form (Fe–TPTZ) of intense blue colour. Figure 8 illustrates the direct relationship between the amount of antioxidants and the reduction in maximal absorption. Based on the standard curve of $FeSO_4.7H_2O$, the ferric reducing capacity of methanol and ethyl acetate increased with concentration. The findings were displayed as mMFe(II)/ml for the corresponding concentration. Table 2 illustrates that methanol peel extract (10.18 mM Fe(II)/ml) had a greater capacity to reduce ferric iron than ethyl acetate peel extract (8.27 mM Fe(II)/ml).

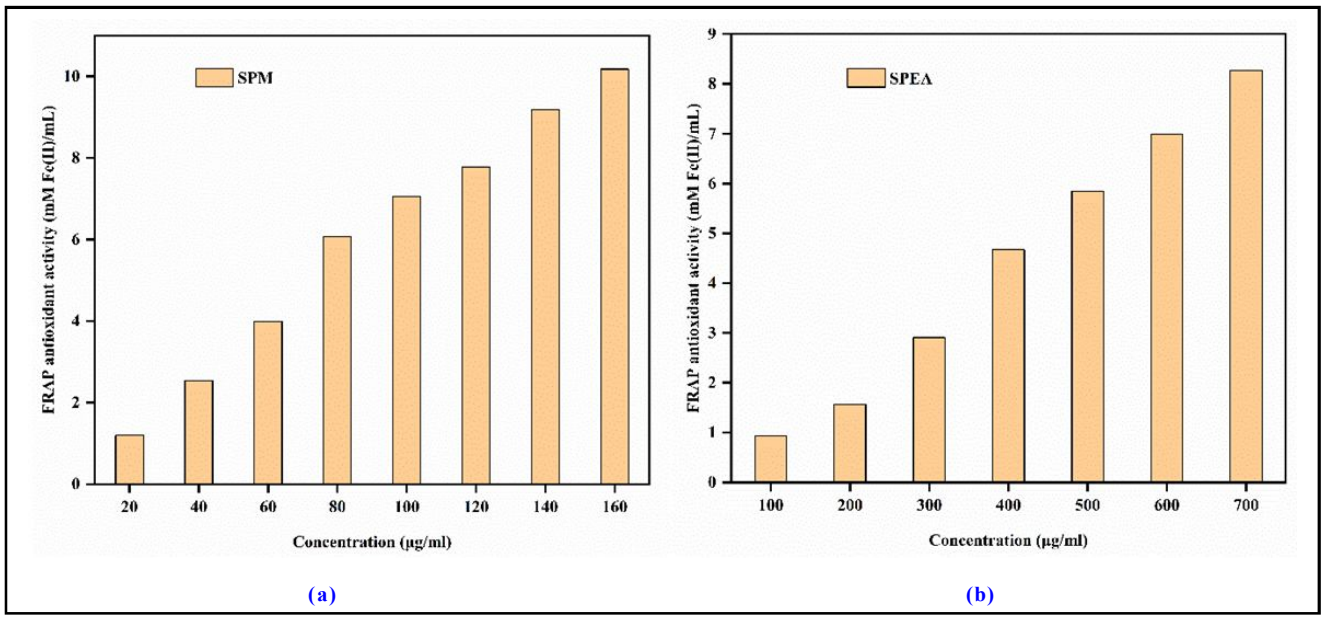

Figure 8: FRAP assay (a) methanol, (b) ethyl acetate peel extract.

3.2.4 Phosphomolybdnum assay

Antioxidants lead to the reduction of Mo (VI) to Mo (V). Phosphomolybdenum complex, which is green in colour, is created when Mo (V) combines with the phosphate group of sodium phosphate in an acidic solution.The standard curve for the total antioxidant capacity using ascorbic acid as the standard.Table 2 presented the results as mgAAE/ml of the corresponding concentration. Methanol peel extract (0.54 mgAAE/ml) were found to be dose dependent and to have higher total antioxidant activity thanethyl acetate peel extract (0.51mg AAE/ml) (Figure 9).

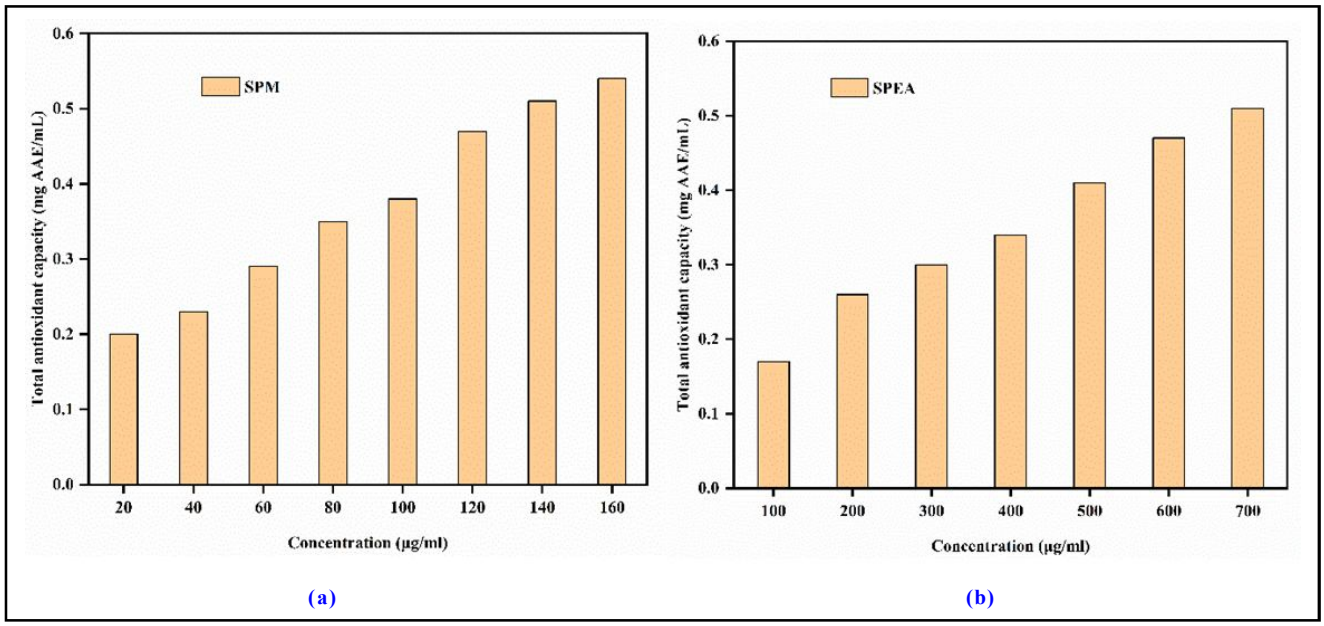

Figure 9: Phosphomolybdnum assay (a) methanol, (b) ethyl acetate peel extract.

Table 2: Antioxidant activity of methanol and ethyl acetate extract of *M. zapota* peel

S. No.	Plant extract	DPPH (IC_{50}) (µg/ml)	ABTS (IC_{50}) (µg/ml)	FRAP (mM Fe(II)/ml)	TAC (mg AAE/ml)
1	SPM	93.782	96.52	10.18	0.54
2	SPEA	309.005	305.07	8.27	0.51

4. Discussion

The main motive of this study was to identify the phytochemical constituents of an extract of dried *M. zapota* peel used to make methanol and ethyl acetate. This study found that the methanol extract fruit peel of *M. zapota* was a remarkable source of total sugars, total phenolics, reducing sugars, total flavonoids, and non-reducing sugars as compared to ethyl acetate extract. Due to the existence of a good amount of phytochemicals (flavonoids and phenolic) in methanol extract, shows adequate antioxidant capacity using ABTS, DPPH free radical scavenging activity, FRAP and phosphomolybdnum assay.

Depending on how polar the extraction solvents were, the extract's overall phenolic content could change. The results of this investigation showed that there was a significant amount of activity in the release of secondary metabolites from plant components by methanol extract. This may be the result of the widespread extraction of phenolic compounds using polar solvents such as methanol, ethanol, and aqueous (Choi *et al*., 2007).

It is well known that one of the main sources of free radicals is medicinal plants. Plants can be used to cure illness and make medicine because of the phytochemicals present in them, which have specific biological impacts on human beings.

The results of this investigation matched the estimate of Gomathy *et al*. (2013), who reported that the total flavonoid content and total phenolic content of the peel methanol extract were 5.89 mg/g and 20.85 mg/g, respectively, and also reported DPPH free radical scavenging of methanol peel extract with IC_{50} was 0.45 mg/ml and ABTS free radical scavenging IC_{50} was 0.39 mg/ml. According to their findings of the total antioxidant capacity assay, *M. zapota* peel methanolic extracts exhibit the highest level of antioxidant assay within the range of 63.31 µM/g. When Devatkal *et al*. (2014) assessed the total phenolic content of the *M. zapota* peel of aqueous extract, they discovered that 10 g of TPC peel contained 550 mg of GAE. Aquino *et al*. (2020) assessed the highest concentrations of phenolic compounds (126.0 mg GAE/100 g of residue) and flavonoid compounds (90.0 mg QCE/100 g of residue) found in the peel extract prepared with 40% methanol. The range of the TPC for sapota peel was 34.0 to 126.0 mg GAE/100 g.

To oxidise the Folin-Ciocalteu reagent, phenolic compounds must be identified. After oxidising phenols, phosphotungstic acid and phosphomolybdic acid are reduced to a reagent that contains molybdenum oxides and blue tungsten oxides. The amount of phenolic compound generated in the resultant solution is directly correlated with the brightness of the blue colouring, with a maximum absorption area of approximately 730 nm.

To calculate total flavonoids, one uses the idea that $AlCl_3$ and the C-3, C-5 hydroxyl group, and C-4 keto group of flavones and flavonols form an acid-stable complex. Additionally, acid-labile complexes can be formed when the orthodihydroxyl groups on the flavonoid A or B rings react with $AlCl_3$.

The total sugars were determined by dehydrating glucose in an acidic medium to yield hydroxymethyl furfural, which was subsequently combined with phenol to yield a yellow-brown solution with the highest absorption at 490 nm. Alkaline copper tartrate converts cupric ions into cuprous ions, which results in the production of cuprous oxide, which lowers sugars when heated. When cuprous oxide and arsenomolybdic acid are combined, molybdic acid is converted to molybdenum blue, which an ultraviolet-visible spectrophotometer can detect at 520 nm. When peel extract was added to the purple DPPH solution for the DPPH free radical scavenging activity, the solution turned yellow. This happened as a result of the DPPH molecule being scavenged after obtaining the stabilising hydrogen atom. The concentration of antioxidants determines how much the purple colour fades.

When peel extract was added to the ABTS solution during the free radical scavenging activity, the solution's turquoise colour slightly turned yellow. This happened because the hydrogen atom that was added to stabilise the ABTS molecule was scavenged afterward. The amount that turquoise colour fades depends on the antioxidant concentration.

The ferric-tripyridyltriazine (Fe^{3+}-TPTZ) is reduced to an intense blue ferrous-tripyridyltriazine complex (Fe^{2+}-TPTZ), with a maximum level of absorption at 593 nm, in the FRAP assay, which is employed to determine the power of antioxidants. The assay primarily uses ferrous sulfate as a positive control. The results can be expressed as µm/ml Fe^{2+} calculated from a standard curve or as µm/ml ferrous sulfate equivalents. As the concentration of peel extract increases intensity of the blue color increases due to the formation of more complex (Fe^{2+}-TPTZ).

When evaluating the extract's overall antioxidant capacity, molybdenum (VI) can be changed into the green phosphomolybdate (V) complex by the antioxidants present. Flavonoids and phenolics are responsible for the *M. zapota* peel's antioxidant capacity.

5. Conclusion

According to the outcomes of the current study, phytochemicals found in the methanol and ethyl acetate extract of *M. zapota* peel may be crucial for scavenging species that produce oxidative stress. Quantitative phytochemical research may be useful in understanding the pharmacological effects of *M. zapota* peel. Because the methanol peel of *M. zapota* contains a higher concentration of phytochemicals (TPC 29.791 mg GAE/g and TFC 6.876 mg CE/g) than the ethyl acetate, further research is needed to determine how the peel may be used in the pharmaceutical, medical, and nutritional sectors. To pinpoint, the precise elements of the antioxidant system and create uses for the food and healthcare sectors, more investigation is needed.

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Conflict of interest

The authors declare no conflict of interest relevant to this article.

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