

Phytochemical screening for antioxidant activity and elemental analysis of the leaves and cracked - barks of *Mangifera indica* (Mango)

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Abstract

Mango leaves and cracked backs have been reported for various medicinal effects like antioxidant, antimicrobial, antihelminthic, antidiabetic and antiallergic activities. The present study focused on antibacterial activities and level of Trace elements in leaves and barks of *Mangifera i.* grown in Yilo Krobo Municipality, Eastern region of Ghana. Preliminary phytochemical screening and antioxidant properties of 75% ethanolic extract of the leaves and barks of the plant was conducted. The antioxidant properties were determined using 2, 2-diphenyl-1-picryl hydrazyl (DPPH) assay. Total Flavonoid and Total Phenolic extracts were performed spectrophotometrically use of Folin-Ciocalteu and aluminium chloride reagents. The result proved that the leaves and cracked barks of Mango plant exhibited potent antioxidant properties therefore they might be rich source of natural antioxidants and can be used in place of synthetic antibiotics because of having similar chemical properties as synthetic antibiotics. Trace elements such as copper (Cu), lead (Pb), calcium (Ca), zinc (Zn), iron (Fe), manganese (Mn), nickel (Ni), magnesium (Mg), sodium (Na) and cobalt (Co) were assessed by atomic absorption spectrophotometry.

Keywords: Antioxidant, mango, phytochemical, flavonoid, phenolic, diphenyl-1-picryl hydrazyl and extract

Introduction

Dodowa in Yilo Krobo Municipal in the Eastern region of Ghana is known for its commercial mango farming. The farmers grow it purposely for the sale of its fruits for exporters fruit juices companies and local consumption. However, the local people sometimes use their leaves and

barks to treat typhoid fever. There are some herbal medicine manufacturing companies which normally procure the leaves, barks and roots of mango plants. Before the advent of modern medicine, herbal preparations have been used to treat various infections. Natural products from plants as herbal remedies are still being employed as alternative medicine to prevent and cure several illnesses and this vary in different communities.¹ These medicinal plants are largely raw source for the production of modern antibiotics. According to WHO, a medicinal plant could be any plant that contains substances which can be obtained from its different parts and can be applied for beneficial purposes or can be precursor for the production of useful drugs.² Unfortunately, knowledge on various plants acquired by through experience is rapidly eroding due to the lack of research and documentation of this indigenous knowledge.³

Currently, about thousands of drugs commonly prescribed are derived from plants sources.^{4,5} Phytochemical exhibit various important biological, pharmacological and antioxidant activities.⁶⁻⁸ The effectiveness of phytochemical

Citation: Asare Ebenezer Aquisman, Charles Kofi Klutse, Eric Kwabena Droepenu *Phytochemical screening for antioxidant activity and elemental analysis of the leaves and cracked - barks of *Mangifera indica* (2019) Archives of Pharmacology and Pharmaceutical Sciences (APPS) ReDelve RD-PHA-10002*

Received date: 01 June 2019 ; **Accepted date:** 08 June 2019
Published date: 09 July 2019

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in the treatment of various diseases may lie in their antioxidant effects because oxidative stress is associated with pathogenic mechanisms. Antioxidants can inhibit the oxidation by scavenging free radicals and diminishing oxidative damage.⁹

Some research has been conducted into edible plants like pawpaw and banana with respect to their antioxidant and antimicrobial activity. According to,^{10,11} unripe banana peels used in their study showed a high potential of natural antioxidant than the ripe and leaky ones. The prospective applications of banana peels depend on its chemical composition, as different fruit parts contain different antioxidant and antimicrobial components. Unripe banana peels contain some elements such as copper, zinc, sodium, potassium, calcium, phosphorus, and iron.,¹² notwithstanding its rich nutrients.¹³

Mangifera indica L, commonly called mango and locally as Amangu (Akan Language) is a plant belonging to the family Anacardiaceae which consists of about sixty genera and six hundred species.¹⁴ It is one of the most popular tropical fruit bearing trees in the world.¹⁵ Fruits are highly nutritive and sweet in taste. Not only fruits but also the leaves, seeds and barks of this plant are also used for medicinal purposes.¹⁶ The availability of *Mangifera indica* in Ghana, its large production volume and the medicinal value has necessitated for this study to analyse its antioxidant activity of some parts of the plant and also screen for its elemental and phytochemical constituents.

Materials and methods

Plant materials

The leaves and cracked barks of *Mangifera indica* samples were collected from the vicinity of Dodowa in the Yilo Krobo Municipal. The samples collected were identified in Biotechnology and Nuclear Agriculture Research Institute (BNARI), Ghana Atomic Energy Commission. The samples were air dried and crushed into powder and stored in polythene bags for further analysis.

Chemicals and preparation of 75% ethanolic extract

Folin-Ciocalteu's phenol reagent, gallic acid, quercetin, 2,2-diphenyl-1-picrylhydrazyl (DPPH) and others were purchased from sigma chemical company, USA. Distilled water was used in the preparation of all the solutions. Chemicals and reagents used were of analytical grade. Extraction of the Mango leaves and cracked barks parts were

done with ethanol and distilled water (75:25). Exactly 40.0 g of each powdered mango leaves and cracked backs were mixed with 400 ml of ethanol solvent. Then, the mixtures were inserted into an orbital shaker and agitated for 2 days. After that, the mixtures were filtered using Whatman Filter paper no. 1. The filtrates were evaporated under reduced pressure by using rotatory evaporator at the temperature ranging between 35 °C to 50 °C in order to obtain the crude extract of the plant materials. Lastly, the crude extract of both mango leaves and cracked barks were kept in the refrigerator until required for analysis.

Preliminary phytochemical screening

The extracts were used for the preliminary phytochemical analysis to identify the various chemical compositions. All the tests were performed in triplicate mode and standard procedures were taken for the analysis.^{17,18}

Spot test (volatile oil)

About 4.0mL extract solution each of Mango leaves and cracked bark was evaporated to get residue and it was mixed with 1mL methanol. The solution was shaken vigorously and filtered for about 5 minutes. Few drops of filtrate were poured on a filter paper by means of a capillary tube. A yellow spot on a filter paper that persists after evaporation indicates the presence of volatile oil.

Flavonoid

Crude extract of each was mixed with 4.0mL of 4% solution of NaOH. An intense yellow colour formed which turned colourless on addition of few drops of diluted HCl indicated the presence of flavonoid.

Keller-kilani test

A combination of Acetic acid glacial (3.0mL) with 3 drops of 3% FeCl₃ solution was added to the each of the plant extract and drops of concentrated H₂SO₄ in addition. An observation of brown ring at the interface indicated the presence of cardiac glycosides.

Alkaloids: mayer's reagent

About 2.0mL each of the plant extract, 4.0mL of Mayer's reagent (potassium mercuric iodide solution) was added. Appearance of dull white precipitate indicated the presence of alkaloids.

Salkowski's test (triterpenoids)

About 5.0mL of each plant extract was mixed in 2.0mL of chloroform and 3mL of concentrated H_2SO_4 and it was carefully added to form a layer. An observation of reddish brown precipitate at interface indicated the presence of triterpenoids.

Phenols and tannins:

Each of plant extract was mixed with 2.0mL of 2% solution of $FeCl_3$. An observation of blue -green or black coloration indicated the presence of phenols and tannins.

Lieberman-burchard's test (steroids)

The crude extract was dissolved in 3mL of chloroform to which 15 drops of acetic acid and 8 drops of concentrated H_2SO_4 were added and mixed. The change of red colour through blue to green indicated the presence of steroids.

Reducing compounds (Fehling's test)

About 0.5ml alcoholic extract was added with 1.0mL water and 0.5ml Fehling's solution (A+B) then warm or gently boiled. A reddish brick precipitation shows the presence of reducing compounds.

Saponins

About 3.0mL crude extract of each was mixed with 7.0mL of distilled water in a test tube and it was shaken vigorously. An observation of stable foam was an indication for the presence of saponins.

Determination of total phenolic content and total flavonoid content

a) Preparation of standard for phenolic content and flavonoid content

The total phenolic content of extracts were determined by Folin - Ciocalteu reagent (FCR) method as described by.¹⁹ Stock solution of gallic acid was prepared by dissolving 2mg gallic acid in 2mL of methanol (1mg/mL). Different concentrations of gallic acid such as 25 μ g/mL, 50 μ g/mL, 75 μ g/mL, 100 μ g/mL, 150 μ g/mL and 200 μ g/mL were prepared by serial dilution of stock solution. An aliquot of 1mL gallic acid of each concentration in methanol was added to 20mL test tube. To that 5.0mL of Folin - Ciocalteu reagent (10%) and 4.0mL of 7% Na_2CO_3 were added to get a total of 10mL. The blue coloured mixture was shaken well and incubated for 30 minutes at 40°C in a water bath. Then

the absorbance was measured at 760nm against blank. The absorbance values obtained at different concentrations of gallic acid were used to plot the calibration curve.

Similarly, the total flavonoid content was quantified by aluminium chloride colorimetric assay.²⁰ Different concentration of standard quercetin such as 25 μ g/mL, 50 μ g/mL, 75 μ g/mL, 100 μ g/mL, 150 μ g/mL and 200 μ g/mL were prepared by serial dilution of stock solution of concentration of 4mg/mL. An aliquot of 1mL quercetin of each concentration in methanol was added to 10mL volumetric flask containing 4mL of double distilled water. At the zero time, 0.3mL, 5% sodium nitrite was added to the flask. After 5 min, 0.3mL of 10% $AlCl_3$ was added to the flask. At 6 min, 2ml of 1M sodium hydroxide was added to the mixture. Immediately, the total volume of the mixture was made up to 10mL by the addition of 2.4mL double distilled water and mixed thoroughly. Absorbance of the pink colored mixture was determined at 470nm versus a blank containing all reagents except quercetin. Absorbance values obtained at different concentrations of quercetin were used to plot the calibration curve.

b) Preparation of samples for phenolic content and flavonoids content

To prepare the sample for phenolic content determination, the stock solutions of all the crude extracts were prepared by dissolving 2mg in 2mL of methanol. Serial dilutions were performed to get the concentration of 25 μ g/mL, 50 μ g/mL, 75 μ g/mL, 100 μ g/mL, 150 μ g/mL and 200 μ g/mL. To these diluted solution FCR and Na_2CO_3 were added and incubated for 30 minutes as in the case of standard gallic acid preparation and absorbance was measured at 760nm. Similarly, to prepare the samples of flavonoids content determination, various concentrations of the extracts such as 25 μ g/mL, 50 μ g/mL, 75 μ g/mL, 100 μ g/mL and 200 μ g/mL were prepared. Following the procedure explained above in flavonoid, absorbance for each concentration of extract was recorded. Total Flavonoid Content of the extracts was expressed as mg quercetin equivalents (QE) per gram of extract in dry weight (mg/g).

Estimation for total phenolic and flavonoid content and statistical analysis:

The total phenolic content and flavonoid content was calculated using the formula.²⁰

$$C = cV/m \quad \text{Equation 1.0}$$

C= total contents of compounds in mg/g, in mg GAE/g or total flavonoid content mg QE/g dry extract. c= concentration of gallic acid established from the calibration curve in mg/ml or concentration of quercetin obtain from calibration curve, mg/ml, V= the volume of extract in ml, m= the weight of plant extract in g. Calculation of linear correlation coefficient R^2 and correlation analysis were determined using Microsoft Office Excel 2010. The linear regression equation is given as,

$$y = bx + a \quad \text{Equation 2.0}$$

where y = absorbance of extract, b = slope of the calibration curve, x = concentration of the extract, a = intercept

Assessment of antioxidant activity using 2, 2 - diphenyl-1-picrylhydrazyl free radical (DPPH)

DPPH radical scavenging activity of crude extracts was assessed as suggested by.²¹ DPPH solution (0.1mM) in methanol was prepared by dissolving 3.9mg of DPPH in 100ml methanol and stirred overnight at 4°C. The prepared purple coloured DPPH free radical solution was stored at -20°C for further later use. Three different concentrations (5µg/ml, 10µg/ml and 15µg/ml) of methanolic solutions of each extracts were prepared by the serial dilution of the stock solution of the respective crude extracts. To each 0.5mL extract solution, 2.5mL, 0.1mM methanolic DPPH solution was added. A control was prepared by mixing 0.5mL distilled water and 2.5mL, 0.1mM methanolic DPPH solution. These samples were well shaken and kept in dark for 30min at a room temperature. The absorbance of the mixture was measured spectrophotometrically at 517nm against the blank solution consisting 2.5mL MeOH and 0.5mL distilled water. The radical scavenging activity was expressed as the radical scavenging percentage using the formula:

$$\% \text{ scavenging activity} = (A_c - A_s / A_c) * 100$$

Equation 3.0

Where, A_c = absorbance of control and methanol and A_s = absorbance of sample solution and DPPH radical.

IC₅₀ value:

IC₅₀ value is the concentration of sample required to scavenge 50% of DPPH free radical and was calculated from the plotted graph of radical scavenging activity against concentration of extracts. The antioxidant activity was determined by DPPH assay and the free radical scavenging

activity (IC₅₀) value was calculated. The higher the phenolic content, the lower the IC₅₀ Value and vice versa.

Atomic absorption spectroscopy (AAS)

Elemental composition analysis of *Mangifera indica* was determined using AAS. A crucible containing 6 g of each dried *Mangifera indica* leaf and cracked bark sample were placed in a hot furnace at 550° for 4 hrs. After cooling the sample in a desiccator, 0.5 g of the ashed sample of each was mixed with 5ml each of distilled water, concentrated HNO₃ and perchloric acid (HClO₄) and transferred into the digestion block inside a fume cupboard at 150° for 90min. The temperature was then increased to 230° for 30min and then reduced to 150°, followed by the addition of 1ml of HCl to the tubes immediately. The concentrated digest was not allowed to cool to room temperature to prevent the formation of insoluble precipitate (potassium perchlorate). Water was added to the tube to make up to the mark and the content was mixed and filtered. The resulting solution was used for the elemental analysis using AAS (Analyst 400 Model) at an appropriate wavelength, temperature and lamp-current for the different elements suggested by.²² The elements copper (Cu), lead (Pb), calcium (Ca), zinc (Zn), iron (Fe), manganese (Mn), nickel (Ni), magnesium (Mg), sodium (Na) and cobalt (Co) were assessed.

Statistical analysis

The results are presented as mean and standard deviation obtained from three replicate experiments using Microsoft Office Excel 2010 version.

Results

The powdered leaf and cracked bark of mango plant was extracted by using ethanol solvent and each of the percentage yields was calculated.

Phytochemical analysis of ethanol extract of Mango leaves and cracked-barks were performed to identify the presence of bioactive compounds (flavonoids, phenols, tannins, saponins etc) as illustrated in Table 1-3.

Table 1 Mass of crude extract and percentage yield

| Plant part | Crude mass (g) | Percentage yield (%) |
|---------------|----------------|----------------------|
| Leaves | 7.2403 | 16.422 |
| Cracked barks | 8.0121 | 15.341 |

Table 2 Preliminary phytochemical screening of mango leaves

| A.Q | Experiment | Test | Observation | Result |
|-----|--------------------|-----------------------------|-------------------------------|--------|
| 1 | Volatile oil | Residue Test/Spot Test | No yellow colour persist | + |
| 2 | Flavonoid | Shinoda Test | Deep yellow to colourless | + |
| 3 | Glycosides | Salkowski's | Test No ring at the interface | - |
| 4 | Alkaloids | Mayers Reagent Test | Dull white ppt formed | + |
| 5 | Triterpenoids | Terpenoids Test | Grayish colour observed | - |
| 6 | Phenols | Phenolic Test | Black coloration | + |
| 7 | Steroids | Steroid Test | Red colour unchanged | - |
| 8 | Reducing compounds | Fehlings Test | Reddish – brick ppt | + |
| 9 | Saponins | Foam/Froth Test | No foam formation | - |
| 10 | Tannins | 0.1% FeCl ₃ Test | Greenish – black | + |

Table 3 Preliminary phytochemical screening of Mango cracked barks

| A.Q | Experiment | Test | Observation | Result |
|-----|--------------------|-----------------------------|------------------------------|--------|
| 1 | Volatile oil | Residue Test/Spot Test | No yellow colour persist | + |
| 2 | Flavonoid | Shinoda Test | Intense yellow to colourless | + |
| 3 | Glycosides | Salkowski's Test | ring at the interface | - |
| 4 | Alkaloids | Mayers Reagent Test | Dull white ppt formed | + |
| 5 | Triterpenoids | Terpenoids Test | Grayish colour observed | - |
| 6 | Phenols | Phenolic Test | Black coloration | + |
| 7 | Steroids | Steroid Test | Pale blue – green colour | - |
| 8 | Reducing compounds | Fehlings Test | Reddish – brick ppt | + |
| 9 | Saponins | Foam/Froth Test | No foam formation | - |
| 10 | Tannins | 0.1% FeCl ₃ Test | Bluish – black | + |

Calibration curve for total phenolic content (TPC) and total flavonoid content (TFC)

The total phenolic content in plant extract was determined by using Folin-Ciocalteu calorimetric method. The absorbance values obtained at different concentrations of gallic acid in each plant parts was used for the construction The total phenolic content in plant extract was determined by using Folin-Ciocalteu calorimetric method. The absorbance values obtained at different concentrations of gallic acid in each plant parts was used for the construction of calibration curve. Similarly, the total flavonoid contents were determined by a calorimetric assay using aluminium chloride. Absorbance values for quercetin measured at 470 nm using aluminium chloride calorimetric assay were shown in Tabular form and calibration curves are presented in the various Tables below

(Table 4 & Table 5) (Figure 1).

Table 4 Absorbance value for gallic acid measured for calibration curve

| Concentration (µg/mL) | Absorbance for gallic acid measured |
|-----------------------|-------------------------------------|
| 25 | 0.25 |
| 50 | 0.52 |
| 75 | 0.73 |
| 100 | 1.10 |
| 125 | 1.28 |
| 150 | 1.72 |
| 175 | 1.75 |
| 200 | 2.03 |

Table 5 Absorbance value for quercetin measured for calibration curve

| Concentration (µg/mL) | Absorbance for quercetin acid measured |
|-----------------------|--|
| 25 | 0.10 |
| 50 | 0.23 |
| 75 | 0.37 |
| 100 | 0.44 |
| 125 | 0.56 |
| 150 | 0.70 |
| 175 | 0.89 |
| 200 | 0.98 |

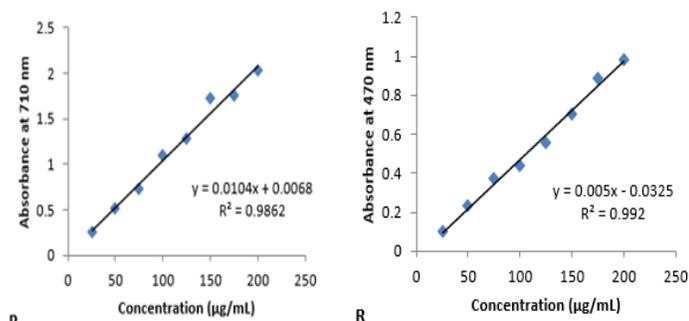


Figure 1 Total phenolic and flavonoid content for standard gallic acid and quercetin.

P represents total phenolic content for standard gallic acid, values expressed in terms of gallic acid equivalent; R represents total flavonoid content for standard quercetin, values expressed in terms of quercetin equivalent.

Estimation of total phenolic and total flavonoid contents in extracts

The phenolic concentration in extract was deduced from the calibration curve using the linear regression equation $y = 0.0104x + 0.0068$, $R^2 = 0.986$. The total phenolic content was calculated using the formula $C = cV/m$ and expressed as mg gallic acid equivalents (GAE) per g of extract in (mg/g). The concentrations of flavonoid in each test samples were calculated from the calibration curve using regression equation $y = 0.005x - 0.0325$, $R^2 = 0.992$. Total flavonoid contents of the extracts were calculated using the formula, $C = cV/m$ and expressed as mg quercetin equivalents (QE) per gram extract in (mg/g) (Table 6-8).

Table 6 Total phenolic and total flavonoid content in different extract

| Mango plant parts | 50% ethanol mg GAE/g phenolic content Mean TPC ± s.d | 50% ethanol mg QE/g phenolic content Mean TPC ± s.d |
|-------------------|---|--|
| Leaves | 364.0±1.30 | 63.41±1.26 |
| Cracked barks | 387.3±0.69 | 69.04±2.01 |

Table 7 The percentage inhibition and IC_{50} values of the ethanol extract samples

| Plant parts | % inhibition | IC_{50} (µg/ |
|---------------|--------------|----------------|
| Leaves | 8.21 | 68.47 |
| Cracked barks | 7.17 | 70.63 |

Table 8 Atomic Absorption Spectrophotometry (AAS)

| Metal | Concentration(mg/mL) | |
|-------|----------------------|---------------|
| | Leaves | Cracked barks |
| Cu | 0.74±0.20 | 0.70±0.12 |
| Pb | 0.31±0.01 | 0.37±0.01 |
| Ca | 0.82±0.10 | 0.89±0.10 |
| Zn | 0.50±0.02 | 0.40±0.02 |
| Fe | 0.37±0.01 | 0.45±0.04 |
| Mn | 0.20±0.01 | 0.21±0.01 |
| Ni | 0.05±0.01 | 0.05±0.01 |
| Mg | 0.61±0.09 | 0.71±0.09 |
| Na | 0.37±0.00 | 0.37±0.00 |
| Co | 0.09±0.00 | 0.07±0.00 |

Discussion

As described in Table 1, the percentage yield of mango leaves crude extract was slightly higher (16.422 %) as compared to mango cracked barks. According to,¹⁷ the usage of ethanol as extraction solvent will give higher percentage yield of ethanol extract of mango plant part which in ranged from 10 to 19% for dry powder compared to methanol and aqueous solution extract. The percentage yield in this study is consistent with literature values.

The table above showed the total phenolic and flavonoid content in mango plant leaves and cracked barks. There are significant amounts of phenol and flavonoids in both plant parts. The total phenol and flavonoid content in the 50% ethanol extract of the *Mangifera indica* leaves and barks reported to be 364.0 ± 1.30 mg GAE/g and 63.41 ± 1.26 mg QE/g and 387.3 ± 0.69 mg GAE/g and 69.04 ± 2.01 mg QE/g respectively.¹⁷ The nature and content of polyphenolics varies dramatically among plants, which possess beneficial properties, such as antioxidant, immune modulatory actions and anticancer and antibacterial activity.²³

From Table 7, The DPPH assay was based on the capability of an antioxidant to donate hydrogen radical which is stable free radical with deep violet colour. When an odd electron become paired in the presence of free radical scavenger of antioxidant agent, DPPH radicals get reduced to corresponding hydrazine, DPPH-H form and the solution gets decolorized from its initial deep violet to light yellow colour.¹⁷ The higher the IC_{50} value, the lower the antioxidant activity of the sample and vice versa.²⁴ reported that a sample that has IC_{50} less than $50 \mu\text{g/mL}$ is a very strong antioxidant, a sample that is in the range of 50 to $100 \mu\text{g/mL}$ is a strong antioxidant, whereas a sample that is in the range of 101 to $150 \mu\text{g/mL}$ is a medium antioxidant and lastly sample that had more than $150 \mu\text{g/mL}$ is a weak antioxidant. The value obtained for Mango plant leaves and cracked barks in Table 7 were $68.47 \mu\text{g/mL}$ and $70.63 \mu\text{g/mL}$ respectively indicates strong antioxidant activity.

All the elements are found to be in the permissible limit defined by WHO.²⁵ Various kinds of elements are required for different physiological, biochemical and molecular functions in human body. These essential elements are for normal functioning of body but can be harmful and toxic at high concentrations.²⁶ Hence, elemental analysis in plants is necessary for safety of herbal drugs to treat various diseases.²⁷ suggested that trace elements have potential factors for improving metabolic disorders including diabetes mellitus and also trace elements, including as Cr, Zn, Se, V, Mg and Mn act as cofactors of antioxidative enzymes and play an important role in protecting the pancreatic β -cells of islets of Langerhans, which are sensitive to free radical damage according to.²⁸ Copper plays a major role in the formation of the heme.²⁹ It is also necessary for myelin sheath formation in the nervous system. Calcium is needed in the body for transmission of nerve impulses, membrane permeability, excitability, muscle contraction, blood clotting and hardness to bones and teeth. Iron is an essential component of

cytochromes, myoglobin, peroxidase, haemoglobin and electron transport. Manganese is necessary for normal metabolism of lipid, protein and carbohydrate and also aids in synthesis and secretion of insulin.³⁰ It also plays a role in micro component of metallo-enzymes such as arginase, glutamine synthetase, phosphoenolpyruvate decarboxylase, manganese superoxide dismutase, galactotransferase of connective tissue biosynthesis, urea formation and it aids in defence mechanisms against free radicals.²⁹ Magnesium serves as co-factor of various enzymes in carbohydrate oxidation and also assists in insulin secretion, transport of glucose that aids in regulation of blood glucose.³¹ Zinc acts as a catalyst for variety of enzymes, for example the alcohol dehydrogenase, ribonucleic polymerases, alkaline phosphate and carbonic anhydrase. It is necessary for skin integrity, bone metabolism and growth. Sodium helps in maintenance of osmolality with plasma and maintains the pH of blood within normal limit. In addition, it is also useful in conduction of nervous impulses, muscle contraction and conduction of impulse in the heart.³² Cobalt is used in the body to help absorb and process vitamin B₁₂. It also helps treat illnesses such as anaemia, and certain infectious diseases.

Conclusion

The result of the phytochemical screening showed the presence of bioactive compounds in the ethanol extract of both leaves and cracked barks of mango plants. The presence of phenols, flavonoids and antioxidant constituent of the leaf and cracked bark extract suggests that mango plants have some significant inhibitory action against disease causing organisms and therefore might be a potential source of natural antioxidants to cure the diseases in which there is an increased free radical production. Also, *Mangifera indica* is a good source of trace elements that are necessary for the normal physiological and biochemical functions of living organisms to maintain a healthy life.

Limitation

Due to financial constraints, the root and the unripe peel of the fruit could have also been analysed to determine the different concentrations available at each part of the mango plant. Future studies could consider this aspect.

Acknowledgments

The authors acknowledge the contribution of colleagues from Analytical Laboratory, Natural Product Laboratory FRST/FSTS UNIMAS and Nuclear Power Research Institute, Ghana Atomic Energy Commission (GAEC).

Conflicts of interest

The authors declare no conflict of interest in this study.

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