Phytochemical Screening, Minerals evaluation and Proximate Analysis of Neem Bark (*Azadirachta indica*) Mango bark (*Mangifera indica)* and their Blend

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Abstract
Mango bark and neem bark is an evergreen domestic plant with immense nutritional and medicinal properties. Mango bark and neem bark have been reported to have inhibitory activity against several bacteria and fungi. The objective of the study was to investigate the phytochemical constituents, mineral content and proximate composition of neem bark, mango bark and their blend. Neem bark and mango bark was used for the experiment, also both were combined together. The experiment includes: T1 (neem bark), T2(mango bark) and T3(neem bark and mango bark blend at the same ratio). The phytochemical screening, mineral evaluation, proximate analysis was conducted using laboratory method. The result showed highest crude protein T2(11.23%), moisture content T1(12.42%), ash T2(7.20%), crude fat T2(9.58%), crude fibre T2(15.65%), nitrogen free extract T3(54.27%). Highest calcium T1(0.20%), sodium T2(0.13%), magnesium T1(0.20%), potassium T1(0.38%), phosphorous T1(0.32%), Manganese T1(13.50), Pb T2(4.80%), iron T1(81.35%). The result indicated the presence of Tannin, Saponin. Glycoside, Terpene, Steroid, Flavonoids, Alkaloids, Phenol and Anthraquinones. Quantitatively, highest tannin T2, T3(0.02%), saponin T1(0.13%), glycoside T1(0.10%), terpene T2, T3(0.002%), steroid T2(0.003%), flavonoids T2(0.002%), alkaloids T2(0.16%), phenol T1(0.20%) and anthraquinones T1(0.01%). This study concluded that the neem bark had better value in minerals content while mango bark higher value in proximate composition and no much variation in definite pattern in terms phytochemical. It is therefore recommended that the two test ingredients can be combine at equal proportion to form a complementary action.

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Keywords: Mango bark, neem bark and their blend

Introduction

Neem (Azadirachta indica)is a tropical plant native to Nigeria, known as "Ogwu-iba" in Igbo and "Dogonyaro" in Hausa. Renowned for its medicinal properties, neem acts as an anti-coccidial agent in broilers and a natural pesticide as reported by (Islas *et al*., 2020) [13]. Neem (Azadirachta indica) is one of the most prominent herbal medicines with different biologically active compounds such as azadirachtin, nimbin, salanin, meliacin, and triterpenoids as stated by (Ansari *et al*., 2012) [2]. Neem (Azadirachta indica) is a versatile medicinal plant, as it shows a wide spectrum of biological activity. The chemical composition of its active principals has had considerable progress regarding biological activity and medical applications, thus being considered a valuable source of natural products for the development of medicines, industrial products, and in the integrated management of agricultural plagues by (Ansari *et al*., 2012) [2]. Therefore, Neem has been used in animal feed as a growth

promoter due to its distinguished antimicrobial capacity as well as its anticoccidian effect in birds (Tipu *et al*., 2006) [24]. Mangifera indica Linn (mango tree) belongs to the plant family known as Anacardiaceae, and originated in the region bounded by Bangladesh and northeastern India (Warschefsky *et al*., 2019) [28]. Mango trees are tropical plants used for food and medicine that grow well in arid environments (Ajayi *et al*., 2005) [3]. Numerous studies have been done on the various applications of mango fruits, peels, juice, and stem bark; however, there are few publications on the significance of M. indica stem bark and its potential for usage as a treatment for gastrointestinal protozoal infections. M. indica has been reported to have several pharmacological properties, including antimicrobial effects as reported by (Doughari *et al*., 2008) [9]. Mangifera indica stem bark (MISB) contains flavonoids, phenols, alkaloids, saponins, and tannins (Somkuwar 2013) [22].

Due to the usefulness of neem bark and mango bark as phyto-additives since they contained well documented phytochemical but no research work has been carried out to analyzed the combination of neem bark and mango bark in terms of the proximate composition, minerals present, qualitative and quantitative analysis. Therefore, the focus of this research is to evaluate the phytochemical constituents, proximate composition and minerals present of neem bark, mango bark and their blend.

Materials and Methods

Experimental site

The experiment was carried out at the Poultry Unit of Teaching and Research Farm, Ladoke Akintola University of Technology Iseyin campus, Oyo State Nigeria. The area is in derived savannah zone of Nigeria. It lies on longitude 3.60 east of greenish meridian and latitude 7.970 North. The mean annual rainfall is 1141mm while relative humidity is between 75% and 95%. It is situated at about 313 meters above the sea level with a mean annual temperature of 270C (Google earth, 2026) [11].

Preparation of test ingredients

The test ingredient (Mango bark and Neem bark) were sourced within teaching and research farm Ladoke Akintola University of Technology Iseyin, Oyo State. Neem bark and mango bark was sliced into flakes in order to increase the surface area to aid drying. Thereafter, it was air dried until each weight remained constant. Neem bark and mango were reduced using a mortar and pestle to a lentil-size portion, the sample were grounded into a powdery consistency using the burr mill. After that, it was sieved and kept until use in an airtight container as describe by (Oyetoro, *et al*., 2020 and Okanlawon *et al*., 2024) [19, 18].

Methods of Proximate and Mineral Analysis

Samples were analyzed chemically according to the official methods of analysis described by the Association of Official Analytical Chemist (A.O.A.C., 18TH EDITION, 2005) [4]. All analysis was carried out in duplicate.

Crude Protein Determination (A.O.A.C. Official Method 988.05)

The crude protein in the sample was determined by the routine semi-micro Kjeldahl, procedure/technique. This consists of three techniques of analysis namely Digestion, Distillation and Titration.

Apparatus: Analytical Balance, Digestion tubes, Digestion Block Heaters, 50ml Burette, 5ml Pipette, 10ml Pipette, 10ml Measuring Cylinder, 100ml Beakers, Fume Cupboard.

Reagents: ConC.H_2SO_4, 0.01NHCL, 40% (W/V) NaOH, 2% Boric Acid Solution, Methyl Red – Bromocresol green mixed indicator, Kjeldahl Catalyst tablet.

Digestion

0.5g of each finely ground dried sample was weighed carefully into the Kjeldahl digestion tubes to ensure that all sample materials got to the bottom of the tubes. To this were added 1 Kjeldahl catalyst tablet and 10ml of ConC. H_2SO_4. These were set in the appropriate hole of the Digestion Block Heaters in a fume cupboard. The digestion was left on for 4 hours, after which a clear colourless solution was left in the tube. The digest was cooled and carefully transferred into 100ml volumetric flask, thoroughly rinsing the digestion tube with distilled water and the flask was made up to mark with distilled water.

Distillation

The distillation was done with Markham Distillation Apparatus which allows volatile substances such as ammonia to be steam distilled with complete collection of the distillate. The apparatus was steamed out for about ten minutes. The steam generator is then removed from the heat source to all the developing vacuum to remove condensed water. The steam generator is then placed on the heat source (i.e. heating mantle) and each component of the apparatus was fixed up appropriately.

Determination: 5ml portion of the digest above was pipetted into the body of the apparatus via the small funnel aperture. To this was added 5ml of 40% (W/V) NaOH through the same opening with the 5ml pipette. The mixture was steam-distilled for 2 minutes into a 50ml conical flask containing 10ml of 2% Boric Acid plus mixed indicator solution placed at the receiving tip of the condenser. The Boric Acid plus indicator solution changes colour from red to green showing that all the ammonia liberated have been trapped.

Titration

The green colour solution obtained was then titrated against 0.01N HCL contained in a 50ml Burette. At the end point or equivalent point, the green colour turns to wine colour which indicates that all the Nitrogen trapped as Ammonium Borate $[(NH_4)_2BO^{3]}$ have been removed as Ammonium chloride (NH_4CL).

The percentage nitrogen in this analysis was calculated using the formula:

% N = Titre value x Atomic mass of Nitrogen x Normality of HCL used x 4

or % N = Titre value x Normality/Molarity of HCL used x Atomic mass of N x Volume of flask containing the digest x $\frac{100}{1}$

Weight of sample digested in milligram x Vol. of digest for steam distillation. The crude protein content is determined by

multiplying percentage Nitrogen by a constant factor of 6.25 i.e. % CP = % N x 6.25.

Crude Fat or Ether Extract Determination (AOAC official method 2003.06)

Apparatus: Soxhlet apparatus and accessories, oven, desicator and analytical balance.

Reagents: Petroleum spirit or Ether (40° – 60°C b.pt).

Determination: 1gm of each dried sample was weighed into fat free extraction thimble and pug lightly with cotton wool. The thimble was placed in the extractor and fitted up with reflux condenser and a 250ml soxhlet flask which has been previously dried in the oven, cooled in the desicator and weighed. The soxhlet flask is then filled to $^3/_4$ of its volume with petroleum ether (b.pt. 40º – 60ºC), and the soxhlet flask. Extractor plus condenser set was placed on the heater. The heater was put on for six hours with constant running water from the tap for condensation of ether vapour. The set is constantly watched for ether leaks and the heat source is adjusted appropriately for the ether to boil gently. The Ether is left to siphon over several times say over at least 10 – 12 times until it is short of siphoning. It is after this is noticed that any ether content of the extractor is carefully drained into the ether stock bottle. The thimble containing sample is then removed and dried on a clock glass on the bench top. The extractor, flask and condenser is replaced and the distillation continues until the flask is practically dry. The flask which now contains the fat or oil is detached, its exterior cleaned and dried to a constant weight in the oven. If the initial weight of dry soxhlet flask is Wo and the final weight of oven dried flask + oil/fat is W_1, percentage fat/oil is obtained by the formula:

$$\frac{W_1 - Wo}{\text{Wt. of Sample}} \times \frac{100}{1 \text{ taken}}$$

Dry Matter and Moisture Determination (AOAC official method 967.08)

Apparatus: Oven, crucibles, dessicator and balance.

Reagents: Silical gel, grease.

Determination: 2g of the sample was weighed into a previously weighed crucible. The crucible plus sample taken was then transferred into the oven set at 100C to dry to a constant weight for 24 hours overnight. At the end of the 24 hours, the crucible plus sample was removed from the oven and transferred to dessicator, cooled for ten minutes and weighed.

If the weight of empty crucible is Wo

weight of crucible plus sample is W_1

weight of crucible plus oven-dried sample W_3

$$\text{(\% DM) \% Dry Matter} = \frac{W_3 - Wo}{W_1 - Wo} \times \frac{100}{1}$$

$$\text{\% Moisture} = \frac{W_1 - W_3}{W_1 - Wo} \times \frac{100}{1} \text{ or}$$

$$\text{\% Moisture} = 100 - \text{\% DM.}$$

Determination of Ash (AOAC OFFICIAL METHOD 942.05)

Apparatus: Porcelain Crucibles, a Dessicator, Analytical Balances and a Furnace.

Determination: 2.0gm of the sample were weighed into a porcelain crucible. This was transferred into the muffle furnace set at 550ºC and left for about 4 hours. About this time, it had turned to white ash. The crucible and its content were cooled to about 100ºC in air, then room temperature in a dessicator and weighed. This was done in duplicate. The percentage ash was calculated from the formula below:

$$\text{Ash content} = \frac{\text{wt. of ash}}{\text{original wt. of sample}} \times \frac{100}{1}$$

Fibre Determination (AOAC 958.06)

Apparatus: Heating mantle, crucibles, furnace, sieve cloth, fibre flask, funnel, analytical weighing balance, a dessicator.

Reagents: 0.255N H_2SO_4, 0.313N NaOH and Acetone.

Determination: 2.0gm of the sample was accurately into the fibre flask and 100ml of 0.255N H_2SO_4 added. The mixture was heated under reflux for 1 hour with the heating mantle. The hot mixture was filtered through a fibre sieve cloth. The filterate obtained was thrown off and the residue was returned to the fibre flask to which 100ml of (0.313N NaOH) was added and heated under reflux for another 1 hour. The mixture was filtered through a fibre sieve cloth and 10ml of acetone added to dissolve any organic constituent. The residue was washed with about 50ml hot water on the sieve cloth before it was finally transferred into the crucible. The crucible and the residue were oven-dried at 105ºC overnight to drive off moisture. The oven-dried crucible containing the residue was cooled in a dessicator and later weighed to obtain the weight W_1. The crucible with weight W_1 was transferred to the muffle furnace for Ashing at 550°C for 4 hours.

The crucible containing white or grey ash (free of carbonaceous material) was cooled in the dessicator and weight to obtain W_2. The difference $W_1 - W_2$ gives the weight of fibre. The percentage fibre was obtained by the formula:

$$\text{\% Fibre} = \frac{W_1 - W_2}{\text{wt. of sample}} \times \frac{100}{1}$$

Nitrogen-Free Extract (NFE) Determination

The NFE determined by difference. This was done by subtracting SUM of (Moisture % + % Crude Protein + % Ether Extract + % Crude Fibre + % Ash) from 100 i.e. (100 – (% M + % CP + % EE + % CF + % Ash).

Determination of Mineral Element (Aoac, 975.11)

Calcium potassium and Sodium Determination

Apparatus: Heating mantle, Crucible, Glass rod, Flame photometer, 100ml Volumetric flask, Whatman No. 1 Filter paper, Wash bottle, 10ml pipette, funnel.

Reagents: 2 MHCL.

Determination: The ash of each sample obtained was digested by adding 5ml of 2 MHCL to the ash in the crucible and heat to dryness on a heating mantle. 5ml of 2 MHCL was added again, heat to boil, and filtered through a Whatman No. 1 filter paper into a 100ml volumetric flask. The filterate was made up to mark with distilled water stoppered and made ready for reading of concentration of Calcium, Potassium and Sodium on the Jenway Digital Flame Photometer (PFP7 Model) using the filter corresponding to each mineral element.
The concentration of each of the element was calculated using the formula: %Ca or %K or %Na =

$$\frac{\text{Meter Reading (MR) x Slope x Dilution factor}}{1000}$$

NB: MR x slope x dilution factor will give you the concentration in part per million (ppm or mg/kg). You get concentration in % when you divide by 10000.

Phosphorus Determination (Spectrophotometric method) (AOAC, 975.16)
Phosphorus was determined routinely by the vanado-molybdate colorimetric or spectrophotometric method.

Apparatus: Spectrophotometer or colorimeter, 50ml volumetric flask, 10ml pipette, filter paper, funnel, wash bottle, glass rod, heating mantle, crucibles.

Reagents: Vanadate – Molybdate yellow solution, 2 MHCL.

Determination: The ash of each sample obtained was treated 2 MHCL solution as described for calcium determination above. 10ml of the filterate solution was pipetted into 50ml standard flask and 10ml of vanadate yellow solution was added and the flask was made up to mark with distilled water, stoppered and left for 10 minutes for full yellow development. The concentration of phosphorus was obtained by taking the optical density (OD) or absorbance of the solution on a Spectronic 20 spectrophotometer or colorimeter at a wavelength of 470nm.
The percentage phosphorus was calculated from using the formula:

$$\%\text{Phosphorus} = \frac{\text{Absorbance x Slope x Dilution factor}}{10000}$$

Determination of Mg, Pb, Mn, Fe, USING BUCK 200 AAS (AOAC, 975.23)
The digest of the ash of each sample above as obtained in calcium and potassium determination was washed into 100ml volumetric flask with deionised or distilled water and made up to mark. This diluent was aspirated into the Buck 200 Atomic Absorption Spectrophotometer (AAS) through the suction tube.

Quantitative determinations
Tannin
0.20g of sample was measured into a 50ml beaker 20ml of 50% methanol was added and covered with parafilm and placed in a water bath at 77-80^{o}C for 1 hour. It was shaking thoroughly to ensure a uniform mixing. The extract was quantitatively filtered using a double layered Whatman No 41-filter paper into a 100ml volumetric flask, 20ml water added, 2.5ml folin-Denis reagent and 10ml of 17% na2Co3 were added and mixed properly. The mixture was made up to mark with water mixed well and allow to stand for 20min. the bluish –green color will develop at the end of range 0-10ppm were treated similarly as 1ml sample above.
The absorbance of the Tannic acid standard solutions as well as samples were read after color development on a spectronic 21D spectrophotometer at a wavelength of 760nm. % Tannin was calculated using the formula.

$$\%\text{TANNIN} = \frac{\text{absorbance of sample X average gradient factor X Dilution factor}}{\text{Wt. of Sample X 10,000}}$$

Saponin Procedure
The Spectrophotometric method of Brunner (1984) was used for Saponin Analysis.
1g of finely ground sample was weighed into a 250ml beaker and 100ml of isobutyl alcohol was added. The mixture was shaken on a UDY shaker for 5 hours to ensure uniform mixing. Thereafter the mixture was filtered through a what man No1 filter paper into a 100ml beaker and 20ml of 40% saturated solution of magnesium carbonate was added. The mixture obtained with saturated MgCO3 was again filtered through a Whatman No1 filter paper to obtain a clear colorless solution. 1ml of the colorless solution, was pipetted into 50ml volumetric flask and 2ml of 5% FeCL3 solution was added and made up to mark with distilled water. It was allowed to stand for 30min for blood red color to develop. 0-10ppm standard Saponin solutions were prepared from saponin stock solution. The standard solutions were treated similarly with 2ml of 5% FeCL3 solution as done for 1ml sample above. The absorbance of the sample as well as standard saponin solutions were read after color development in a Jenway V6300 Spectrophotometer at a wavelength of 380min

$$\%\text{Saponin} = \frac{\text{Absorbance of sample X gradient factor X dilution factor}}{\text{Wt. of sample X10000}}$$

Determination of Glycoside
10ml of extract was pipette into a 250ml Conical Flask.50ml Chloroform was added and shaken on a Vortex Mixer for 1hr. The mixture was filtered into 100ml Conical flask and 10ml pyridine, 2ml of 2% sodium nitroprusside were added, shaken thoroughly for 10 minutes. 3ml of 20% NaOH was later added to develop a brownish yellow colour.
Glycoside standard of concentrations which range from 0-5mg/ml was prepared from 100mg/ml stock Glycoside standard. The series of standards 0-5mg/ml was treated similarly like sample above.
The absorbances of sample as well as standards were read on a Spectronic 21D Digital Spectrophotometer at a wavelength of 510nm. % Glucoside was calculated using the formula:

$$\frac{\text{Absorbance of sample X gradient factor X dilution factor}}{\text{Wt. of sample X10000}}$$

Determination of Terpene

0.50g of sample was weighed into a 50ml Conical Flask,20ml of 2:1 Chloroform-Methanol mixture was added, shaked thoroughly and allowed to stand for 15minutes.The mixture was later centrifuged for another 15minutes.Supernatant obtained was discarded, and the precipitate was re-washed with another 20ml chloroform-methanol mixture for re-centrifugation.

The resultant precipitate was dissolved in 40ml of 10% Sodium Deodocyl Sulphate solution.1ml of 0.01M Ferric Chloride solution was added to the above at 30s interval Shaked well, and allowed to stand 30minutes.Standard Terpenes of concentration range 0-5mg/ml were prepared from 100mg/l stock Terpenes solution from Sigma-Aldrich chemicals, U.S.A. The absorbances of sample as well as that of standard concentrations of Terpenes were read on a Digital Spectrophotometer at a wavelength of 510nm.The percentage Terpene is calculated using the formula:

$$\frac{\text{Absorbance of sample X gradient factor X dilution factor}}{\text{Wt. of sample X 10,000}}$$

Determination of Steroids

0.50g of sample extract was weighed into a 100ml beaker.20ml of Chloroform-Methanol (2:1) mixture was added to dissolve the extract upon shaking for 30minutes on a shaker. The whole mixture was later filtered through a Whatman No.1filter paper into another dry clean 100ml Conical Flask/Beaker.

The resultant residue was repeatedly treated with Chloroform-Methanol mixture until free of Steroids.1ml of the filterate was pipetted into a 30ml test tube and 5ml of alcoholic KOH was added and shaken thoroughly to obtain a homogenous mixture.The mixture was later placed in a water bath set at 37^0C-40^0C for 90minutes.It was cooled to room temperature and 10 ml of petroleum ether added followed by the addition of 5ml distilled water.This was evaporated to dryness on the water bath. 6ml of Liebermann Burchard reagent was added to the residue in dry bottle and absorbance taken at a wavelength of 620nm on a Spectronic 21D digital Spectrophotometer.

Standard Steroids of concentration of 0-4mg/ml was prepared from 100mg/ml stock steroid solution ande treated similarly like sample as above. % Steroid was calculated using the formula:

$$\frac{\text{Absorbance of Sample X Gradient X Dilution Factor}}{\text{Wt of sample X 10000}}$$

Flavonoids Determination

0.50g of finely ground sample was weighed into a 100ml beaker and 80ml of 95% Ethanol added and stirred with a glass rod to prevent lumping. The mixture was filtered through a Whatman No.1. filter into a 100ml volumetric flask and made up to mark with Ethanol. 1ml of the extract was pipetted into 50ml volumetric flask, four drops of Conc.HCL added via a dropping pipette after which 0.5g of magnesium turnings added to develop a magenta red coloration. Standard flavonoid solution of range 0-5ppm were prepared from 100ppm stock solution and treated in a similar way with HCL and magnesium turnings like sample. The absorbance of magenta red coloration of sample and standard solutions were read on a digital Jenway V6300 Spectrophotometer at a wavelength of 520nm. The percentage flavonoid is calculated using the formula.

$$\frac{\text{Absorbance of sample X average gradient factor X dilution factor}}{\text{Wt. sample X 10,000}}$$

Alkaloids Procedure

This is a distillation and titrimetric procedure 2g of finely ground sample was weighed into a 100ml beaker and 20mls of 80% absolute alcohol added to give a smooth paste. The mixture was transferred to a 250ml flask and more alcohol added to make up to 100ml and 1g magnesium oxide added. The mixture was digested in a boiling water bath for 1.5hrs under a reflux air condenser with occasional shaking. The mixture was filtered while hot through a small buchner funnel. The residue was returned to the flask and redigested for 30min with 50ml alcohol after which the alcohol will be evaporated, adding hot water to replace the alcohol lost. When all the alcohol has been removed, 3 drops of 10% HCL was added. The whole solution was later transferred into a 250ml volumetric flask 5ml of zinc acetate solution and 5ml of potassium ferrocyanide solution was added, thoroughly mixed to give a homogenous solution.

The flask was allowed to stand for a few minutes, filtered through a dry filter paper and 10ml of the filterate was transferred into a separatory funnel and the alkaloids present were extracted vigorously by shaking with five successive portions of chloroform. The residue obtained was dissolved in 10ml hot distilled water and transferred into a kjeldahl tube with the addition of 0.20g sucrose and 10ml Conc.H2SO4 and 0.02g selenium for digestion to a colorless solution to determine %N by Kjeldahl distillation method. %Nitrogen got is converted to % total alkaloid by multiplying by a factor of 3.26 i. e % Total alkaloid = %N X 3.26

% Alkaloids = %N X 3.26

Determination of Phenol

0.20g of sample was weighed into a 50ml beaker,20ml of acetone was added and homogenize properly for 1hr to prevent lumping. The mixture was filtered through a Whatman No.1 filter paper into a 100ml Volumetric Flask using acetone to rinse and made up to mark with distilled water with thorough mixing.

1ml of sample extract was pipetted into 50ml Volumetric flask,20ml water added,3ml of phosphomolybdic acid added followed by the addition of 5ml of 23% NaCO3 and mixed thoroughly, made up to mark with distilled water and allowed to stand for 10min to develop bluish-green colour.

Standard Phenol of concentration range 0-10mg/ml was prepared from 100mg/l stock Phenol solution from Sigma-Aldrich chemicals, U.S.A. The absorbances of sample as well as that of standard concentrations of Phenol were read on a Digital Spectrophotometer at a wavelength of 510nm.The percentage Phenol is calculated using the formula:

$$\frac{\text{Absorbance of sample X gradient factor X dilution factor}}{\text{Wt. of sample X 10,000}}$$

Results and Discussion

Table 1 shows the proximate composition of neem bark, mango bark and their blend. Highest crude protein T2(11.23%), moisture content T1(12.42%), ash T2(7.20%), crude fat T2(9.58%), crude fibre T2(15.65%), nitrogen free extract T3(54.27%).

Table 1: Proximate Composition of Neem Bark, Mango Bark and their Blend

Parameters %	T1 (Neem bark)	T2 (Mango bark)	T3 (Neem bark and Mango bark) blend
Crude protein	8.84	11.23	9.13
Moisture content	12.42	11.86	12.35
Ash	4.76	7.20	4.60
Crude fat	6.40	9.58	6.45
Crude Fibre	13.62	15.65	13.21
Nitrogen Free Extract	53.97	44.50	54.27

Table 2 shows the minerals present in neem bark, mango bark and their blend. Highest calcium T1(0.20%), sodium T2(0.13%), magnesium T1(0.20%), potassium T1(0.38%), phosphorous T1(0.32%), Manganese T1(13.50), Lead T2(4.80%), iron T1(81.35%).

Table 2: Minerals present in Neem Bark, Mango Bark and their Blend

Parameters %	T1 (Neem bark)	T2 (Mango bark)	T3 (Neem bark and Mango bark) blend
Calcium	0.20	0.17	0.18
Sodium	0.10	0.13	0.12
Magnesium	0.20	0.18	0.19
Potassium	0.38	0.37	0.35
Phosphorous	0.32	0.29	0.31
Manganese	13.50	11.35	12.21
Lead	4.80	3.25	4.00
Iron	81.35	69.55	73.70

The qualitative and quantitative phytochemical screening of neem bark, mango bark and their blend is presented in Table 3. The result indicated the presence of Tannin, Saponin. Glycoside, Terpene, Steroid, Flavonoids, Alkaloids, Phenol and Anthraquinones. Quantitatively, highest tannin T2, T3(0.02%), saponin T1(0.13%), glycoside T1(0.10%), terpene T2, T3(0.002%), steroid T2(0.003%), flavonoids T2(0.002%), alkaloids T2(0.16%), phenol T1(0.20%) and anthraquinones T1(0.01%).

Table 3: Qualitative and Quantitative of Neem Bark, Mango Bark and their Blend

	Quantitative %		
Qualitative	T1 (Neem bark)	T2 (Mango bark)	T3 (Neem bark and Mango bark) blend
Tannin	0.01	0.02	0.02
Saponin	0.13	0.12	0.12
Glycoside	0.10	0.09	0.09
Terpene	0.001	0.002	0.002
Steroid	0.002	0.003	0.002
Flavonoids	0.001	0.002	0.001
Alkaloids	0.14	0.16	0.15
Phenol	0.20	0.19	0.18
Anthraquinones	0.01	0.003	0.004

Discusion

The plants sampled in this study indicates that Mangifera indica is a good source of and protein which seem to be significantly higher than that obtained from neem bark and combination of both the neem bark and mango bark. It is evidently clear that mango bark (Mangifera indica) contained a significant amount of crude protein, crude fat, ash and crude fibre as seen in Table 1. This result as shown that its can be included in animal feed when compare to neem bark and the combination of the two-test ingredient. The proximate composition acts as an enzyme catalyst, mediates cell responses, and regulates cell development when added to animal diets. In addition, the presence of protein suggests a modest nutritional contribution to tissue repair and regeneration Springer, 2024. The phytochemical profile of mango bark and neem bark demonstrates significant potential for antioxidant, antimicrobial, and therapeutic applications, supporting its use in functional food development and postoperative dietary management. This result of this study is slightly different from the report of (Bennett *et al*., 2022; Oyetoro, *et al*., 2024) [5, 19] which may be as results of the climatic conditions, soil type, processing and storage of the test ingredient.

According to mineral analysis, the most abundant minerals in Neem bark and mango bark are Calcium, Sodium, Magnesium, Potassium, Phosphorous, Manganese, Lead and Iron. Calcium aids in the formation of strong bones and teeth, the coagulation of blood, the release of hormones and other substances, the squeezing and relaxing of muscles, and so also the minerals are essential for heart function as well as skeletal and smooth muscle contraction, making it essential for regular digestive and muscular function. It also helps the human body's cells, tissues, and organs work properly. Its importance in the treatment of hypertension, stroke, and

arrhythmias cannot be overstated. The minerals aids in fluid balance and contributes to normal muscular contraction and nerve impulse condition. The body also uses it to regulate blood pressure and volume. It is also a key component of both human and animal diets, as well as the transfer of other minerals into and out of cells. Finally, they help create bones, aids in the proper working of the muscles, hearts, nerves, and iron use, aids blood coagulation, controls body temperature, supports nerve function, and bone growth. It aids in the utilization of vitamins B, C, and E, increases mineral absorption and metabolism, activates enzymes for glucose and amino acid metabolism, and inhibits calcium deposits in the ureters. Iron is a component of hemoglobin, which transports oxygen to tissues via blood circulation. Zinc reduces cholesterol deposits, assists in B-vitamin absorption, enzyme and insulin production, and glucose metabolism. It is also necessary for growth, healing, preventing sterility, and keeping feathers glossy and smooth (Garba *et al*., 2019) [10]. Sodium's relevance in cellular homeostasis and physiological function cannot be overstated. Calcium aids in the formation and maintenance of bone mass and strength, making it a vital dietary component (Chavan *et al*., 2014) [7].

Phytochemical study revealed that neem bark, mango bark and their blend contained alkaloids, flavonoids, saponins, terpenoids, polyphenols and tannins which corresponded with the result of (Uwague 2019) [26]. Neem bark has shown the presence of alkaloids, saponins, flavonoids, terpenoids, polyphenols, tannins and steroids in extract where the result was in contrast with (Sharma, *et al*., 2014) [20]. The presence of alkaloids, saponins, flavonoids and terpenoids, only, but as comparative to earlier studies there was only presence of alkaloids as reported by (Bigoniya, *et al*., 2012). The presence of phytochemicals such as alkaloids, flavonoids, saponins, glycosides, steroids, terpenoids, polyphenols and tannins reveal that plant parts exhibit medicinal as well as pharmacological activities. The phytochemical analysis of mango bark powder also revealed the presence of several important bioactive compounds including alkaloids, flavonoids, tannins, saponins, glycosides, phenols, reducing sugars, and proteins. The presence of alkaloids suggests potential antimicrobial and pharmacological properties, as alkaloids are known to exhibit bioactivity against a wide range of pathogens (Diso *et al*., 2023; Mehmood *et al*., 2024) [8, 15]. The detection of flavonoids and phenols indicates strong antioxidant potential, as these compounds are capable of scavenging free radicals and reducing oxidative stress, which is important in wound healing and disease prevention (Usman *et al*., 2025; Mehmood *et al*., 2024) [25, 15]. This supports the potential use of mango bark in functional food formulations and therapeutic applications. The presence of tannins in the extract suggests significant astringent and antimicrobial properties. Tannins have been reported to promote wound healing by forming protective layers over tissues and preventing microbial invasion (Usman *et al*., 2025) [25]. Similarly, the strong presence of saponins indicates potential anti-inflammatory and immune-boosting effects. Saponins are known to enhance immune responses and contribute to various therapeutic benefits including antimicrobial activity (Ogbonna *et al*., 2025) [17]. The presence of glycosides further

supports the therapeutic potential of the extract, as glycosides are known for their role in metabolic and physiological regulation (Diso *et al*., 2023) [8].

Conclusion

This study demonstrated that mango bark and neem bark powder possess significant nutritional and phytochemical potential, thereby validating its relevance as a functional and therapeutic plant material. The proximate composition revealed appreciable levels of protein and fibre, indicating its role in energy provision, metabolic regulation, and digestive health, while the moderate moisture content confirms good storage stability and the ash content reflects substantial mineral presence essential for physiological functions. Furthermore, the phytochemical screening showed the presence of important bioactive compounds such as flavonoids, tannins, saponins, alkaloids, and phenols, which are associated with antioxidant, antimicrobial, anti-inflammatory, and antidiabetic properties. These findings scientifically support the traditional use of mango bark and neem bark blend in disease management and highlight their potential application in functional foods, nutraceuticals, and phytotherapeutic formulations, thereby reinforcing the importance of natural plant-based products in improving human health and addressing chronic diseases. Lastly the study has also shown that the neem back and mango back can be combined together in other to form synergy since mango bark shows higher value in proximate composition while neem bark shows higher value in term of mineral content.

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