Detection of hydrogen cyanide in selected anti-malarial plants in Southwest Nigeria

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ABSTRACT

Background: *Cyanogenic glycosides* are toxic secondary metabolites produced by plants. The safety of anti-malaria plant decoctions consumed by Nigerian indigenes is of great concern.

Objectives: This study aimed to identify and quantify cyanogenic glycosides in selected anti-malarial plants.

Methods: Phytochemical screening, qualitative and quantitative tests were carried out on each sample to detect presence of cyanide. Instrumental methods including Fourier-Transform Infrared Spectroscopy (FTIR) and Ultra-Violet Spectroscopy (UV) were employed to detect hydrogen cyanide in the plants. Data were analyzed using a standard calibration curve.

Results: The yield percentages varied, *Lawsonia inermis* leaves had the highest yield at 10.21 % w/w. phytochemical screening indicated presence of steroids, saponins, flavonoids, and cardiac glycosides but absence of cyanogenic glycosides. Both qualitative and quantitative results showed negative cyanide concentration in all samples. FTIR analysis suggested the presence of nitriles in *Lawsonia inermis,* however, UV analysis confirmed the absence of cyanide.

Conclusion: These findings indicate that the selected plants, do not contain harmful cyanide levels.

Keywords: Anti-malaria, cyanide, FTIR, medicinal plants, phytochemical

Détection de cyanure d'hydrogène dans certaines plantes antipaludiques du sud-ouest du Nigéria

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RÉSUMÉ

Contexte: Les *glycosides cyanogènes* sont des métabolites secondaires toxiques produits par les plantes. L'innocuité des décoctions de plantes antipaludiques consommées par les populations autochtones du Nigéria suscite de vives préoccupations.

Objectifs: Cette étude visait à identifier et à quantifier les glycosides cyanogènes dans certaines plantes antipaludiques.

Méthodes : Des analyses phytochimiques, qualitatives et quantitatives ont été réalisées sur chaque échantillon afin de détecter la présence de cyanure. Des méthodes instrumentales, notamment la spectroscopie infrarouge à transformée de Fourier (FTIR) et la spectroscopie ultraviolette (UV), ont été utilisées pour détecter le cyanure d'hydrogène dans les plantes. Les données ont été analysées à l'aide d'une courbe d'étalonnage standard.

Résultats: Les pourcentages de rendement ont varié, les feuilles de Lawsonia inermis présentant le rendement le plus élevé (10.21 % p/p). L'analyse phytochimique a révélé la présence de stéroïdes, de saponines, de flavonoïdes et de glycosides cardiotoniques, mais l'absence de glycosides cyanogènes Les résultats qualitatifs et quantitatifs ont tous indiqué une concentration négative en cyanure dans l'ensemble des échantillons. L'analyse FTIR a suggéré la présence de nitriles dans Lawsonia inermis ; toutefois, l'analyse UV a confirmé l'absence de cyanure.

Conclusion: Ces résultats indiquent que les plantes sélectionnées ne contiennent pas de niveaux nocifs de cyanure.

Mots-clés: Antipaludique, cyanure, FTIR, plantes médicinales, phytochimie

INTRODUCTION

Africa is a continent that comprises of many developing countries including Nigeria and Ghana. The traditional method of treating malaria is mostly utilize in these countries because of the benefits of lesser costs, fewer side effects, availability and efficacy.[1] Several medicinal plants including *Moringa oleifera, Sarcocephalus latifolius,*[2] *Vernonia amygdalina, Lepidium sativum,*[3] *Alstonia boonei Enantia chlorantha, Azadirachta indica, Morinda lucida, Cymbopogon citratus* and *Carica papaya*,[4] have been revealed for malaria management in different parts of Africa.

It has been reported that cyanogenic glycosides are nitrile containing compounds, produced by more than 3,000 plant species from about 130 diverse families, many of which are utilized by humans.[5] Møller,[6] stated that cyanogenic glycosides are bioactive plant products obtained from amino acids with cyanohydrins and oximes (alpha-hydroxynitriles) as main intermediates. When sugar molecules are attached to cyanohydrins, there is formation of cyanogenic glycosides. Cyanogenesis in plant is the release of hydrogen cyanide (HCN) because of plant tissue damage.[7,8] The released hydrogen cyanide is known to be toxic to humans. However, the human body can reportedly endure a low level of hydrogen cyanide (HCN) by conversion of the toxic compound (HCN) to thiocyanate, which is excreted in urine.[9] The toxic threshold range for blood cyanide as stated by Schrenk *et al.*,[10] is between 0.5 mgL^{-1} and 3.0 mgL^{-1}.

Due to the constant reoccurrence of malaria parasites in the populace and challenges associated with the orthodox method of malaria treatment, which include increased cost, inaccessibility to health facilities, adverse effects of the medications and increased resistance of the *Plasmodium* parasites to the available drugs, there is an urgent need for the evaluation of the safety of herbal preparations consumed by humans, for malaria treatment in developing countries. Therefore, this study was carried out to provide useful information about the safety of the locally prepared anti-malarial remedies, for the consumption of humans by assessing the presence and amount of cyanide in the leaves of *Citrus aurantiifolia* (Rutaceae), *Cymbopogon citratus* (Poaceae), *Lawsonia inermis* (Lythraceae), *Mangifera indica* (Anacardiaceae) and the stembark of *Alstonia boonei* (Apocynaceae).

MATERIALS AND METHODS

Sample collection

The leaves of *Citrus aurantiifolia* (CA), *Cymbopogon citratus* (CC), *Lawsonia inermis* (LI), *Mangifera indica* (MI) and the stembark of *Alstonia boonei* (AB) were collected in April, 2024 at Sagamu Local Government Area, Ogun State, Nigeria. Identification and authentication of each plant sample was done at the Herbarium unit of the Forest Herbarium Ibadan, where voucher samples were submitted and numbers obtained. The obtained numbers were FHI 1142140, FHI 1142139, FHI 1142136, FHI 1142137, and FHI 1142170 respectively, for the leaves of *Citrus aurantiifolia, Cymbopogon citratus, Lawsonia inermis, Mangifera indica* and the stembark of *Alstonia boonei.*

Sample preparation and extraction procedures

The collected parts of the plant (stembark and leaves) were air-dried separately at room temperature for 3 weeks and then pulverized. The pulverized leaves of *Citrus aurantiifolia* (300 g), *Cymbopogon citratus* (400 g), *Lawsonia inermis* (120 g), *Mangifera indica* (500 g), and stembark of *Alstonia boonei* (500 g) were separately extracted with absolute methanol in different glass bottles, by cold maceration for three consecutive days (72 hours) with occasional agitation. The methanol extract of each plant sample was filtered and concentrated in vacuo. The concentrated crude extract of each sample was then transferred into clean, dry and properly labeled evaporating dishes of known weights, where the remaining solvent was allowed to evaporate till a constant weight was obtained.

Determination of the percentage yield of each plant sample

The percentage yield of each plant extract was calculated using the formula below and recorded;

Percentage yield (%) = WE (g) /WPS (g) × 100%
Where;
WE = Weight of obtained methanol extract in gram
WPS = Weight of powdered plant sample in gram

Qualitative analysis

Preliminary phytochemical screening

The phytochemical screening of the pulverized leaves of *Citrus aurantiifolia, Cymbopogon citratus, Lawsonia inermis, Mangifera indica* and *Alstonia boonei* stembark, were carried out using standard procedures as described by Evans.[11]

Picrate paper test

The method described by Nkafamiya and Manji,[12] was adopted to evaluate the presence of cyanide in the leaves of *Citrus aurantiifolia, Cymbopogon citratus, Lawsonia inermis* leaves*, Mangifera indica* and the stembark of *Alstonia boonei.* About 10 grams of each pulverized plant was humidified and correctly placed in labeled test tubes. Moistened sodium picrate paper was suspended in each test tube and chloroform (few drops) was added into each tube. The tubes were then stoppered tightly. A colour change on the suspended strip from yellow to reddish brown indicates the liberation of hydrogen cyanide (HCN).

Quantitative analysis

The method described and modified by Nwokoro *et al.*,[13] was implemented to determine the amount of cyanide present in the leaves of *Citrus aurantiifolia, Cymbopogon citratus, Lawsonia inermis, Mangifera indica* and the stembark of *Alstonia boonei*. About 2 mL of 2 % KOH (Potassium hydroxide) and picric acid (1 mL) were measured into 200 mL of $Na_2CO_3.H_2O$ (Sodium carbonate monohydrate) for the preparation of the alkaline picrate reagent. Ten Whatman No. 1 filter papers, with a dimension of 8 cm × 1 cm each, were immersed into the prepared alkaline picrate solution for 15 minutes. The picrate saturated papers were then separated from the solution and used instantly for cyanide determination. Various cyanide concentrations 50, 100, 150 and 200 µg/mL, were each prepared in well labeled glass vessels. About 10 grams of each powdered sample was weighed into properly labeled glass bottles. The cyanide solutions and the samples were acidified with 15 mL of 20 % HCl and heated to 80 ^oC then each bottle was sealed with a picrate saturated strip suspended over the acidified samples. The system was incubated at 28 ^oC ± 2 ^oC temperature for twenty-four hours. After 24 hours, the picrate paper strips were removed from the bottles, and each strip was eluted for thirty minutes with 5 mL of 50 % ethanol solution. The absorbance of each eluate was determined at 510 nm using a UV spectrophotometer. The absorbance of the standard cyanide solutions was used to plot the Calibration line. The concentration of cyanide in each of the samples was deduced from the Standard graph.

Spectroscopic analysis

Fourier Transform Infrared spectroscopy (FTIR) test

The FTIR analysis was carried out to determine the absorption spectrum of cyanide (C≡N) as a functional group in the leaves of *Citrus aurantiifolia, Cymbopogon citratus, Lawsonia inermis, Mangifera indica* and the stembark of *Alstonia boonei* (Enaam *et al*).[14] The Infrared spectrum of each extract was obtained using a Nicolet Avatar FTIR 330 spectrophotometer.

Ultra-Violet spectroscopy (UV) test

The UV spectrophotometer was warmed and set-up, by inputting the necessary values and details of each plant sample. The equipment was calibrated, before the absorbance of each sample was read. Once the equipment set-up was completed, methanol was carefully transferred into the cuvette and placed in the inner sample holder to serve as a blank. Each dilute extract was dispensed into the cuvette and placed in the outer sample chamber. The instrument was allowed to run and after some seconds, the peaks at specific wavelengths of absorption were displayed as result and the UV spectrum of each plant sample was printed out afterwards.

RESULTS

Percentage yield of each plant sample

The yield percentages of the methanol extracts varied, with *Lawsonia inermis* leaves (120 g) yielding the highest at 10.21 % w/w (Table 1).

Table 1: Percentage yield of the dried methanol extract of researched plants.

S/N	Name of plant	Plant part	Weight of sample (g)	Weight of extract (g)	Percentage yield ($\%\ ^w/_w$)
1	*Citrus aurantiifolia*	Leaves	300	15.97	5.32
2	*Cymbopogon citratus*	Leaves	400	6.01	1.50
3	*Lawsonia inermis*	Leaves	120	12.25	10.21
4	*Mangifera indica*	Leaves	500	28.66	5.73
5	*Alstonia boonei*	Stembark	500	8.51	1.70

Preliminary phytochemical screening

The phytochemical screening indicated the presence of saponins, steroids, flavonoids and cardiac glycosides in all samples (Table 2).

Table 2: Results of the phytochemical screening of the researched plants

S/N	Metabolites	*Citrus aurantiifolia*	*Cymbopogon citratus*	*Lawsonia inermis*	*Mangifera indica*	*Alstonia boonei*
1	Saponins	+	+	+	+	+
2	Anthraquinones	-	-	+	-	-
3	Flavonoids	+	+	+	+	+
4	Cardiac glycosides	+	+	+	+	+
5	Steroids	+	+	+	+	+
6	Tannins	+	+	+	+	+
7	Alkaloids	+	+	-	+	+

Keys: + = present, **-** = not detectable

Sodium picrate paper test

All the plant samples tested negative to sodium picrate paper test indicating absence of cyanogenic glycosides (Table 3).

Table 3: Results of the sodium picrate paper test carried out on researched plants

S/N	Plant sample	Plant part	Observation	Inference
1	*Citrus aurantiifolia*	Leaves	No colour change was observed on the suspended picrate paper strip	Cyanogenic glycosides are absent
2	*Cymbopogan citratus*	Leaves	No colour change was observed on the suspended picrate paper strip	Cyanogenic glycosides are absent
3	*Lawsonia inermis*	Leaves	No colour change was observed on the suspended picrate paper strip	Cyanogenic glycosides are absent
4	*Mangifera indica*	Leaves	No colour change was observed on the suspended picrate paper strip	Cyanogenic glycosides are absent
5	*Alstonia boonei*	Stembark	No colour change was observed on the suspended picrate paper strip	Cyanogenic glycosides are absent

Quantitative analysis

All samples showed negative values in their cyanide composition, indicating a zero level of cyanogenic glycosides (Table 4).

Table 4: Results of the concentration of cyanide present in the researched plants

S/N	Sample	Concentration (µg/mL)	Average Absorbance (510 nm)
1	Control 1 (KCN)	50	0.125
2	Control 2 (KCN)	100	0.160
3	Control 3 (KCN)	150	0.077
4	Control 4 (KCN)	200	0.147
5	Leaves of *Citrus aurantiifolia*	-53	0.033
6	Leaves of *Cymbopogon citratus*	-103	0.008
7	Leaves of *Lawsonia inermis*	-71	0.024
8	Leaves of *Mangifera indica*	-129	-0.005
9	Stem-bark of *Alstonia boonei*	-101	0.009

Fourier-Transform Infrared (FTIR) spectroscopy

The FTIR identified presence of different functional groups in the plant samples as seen in the Figures 1a - 1e below:

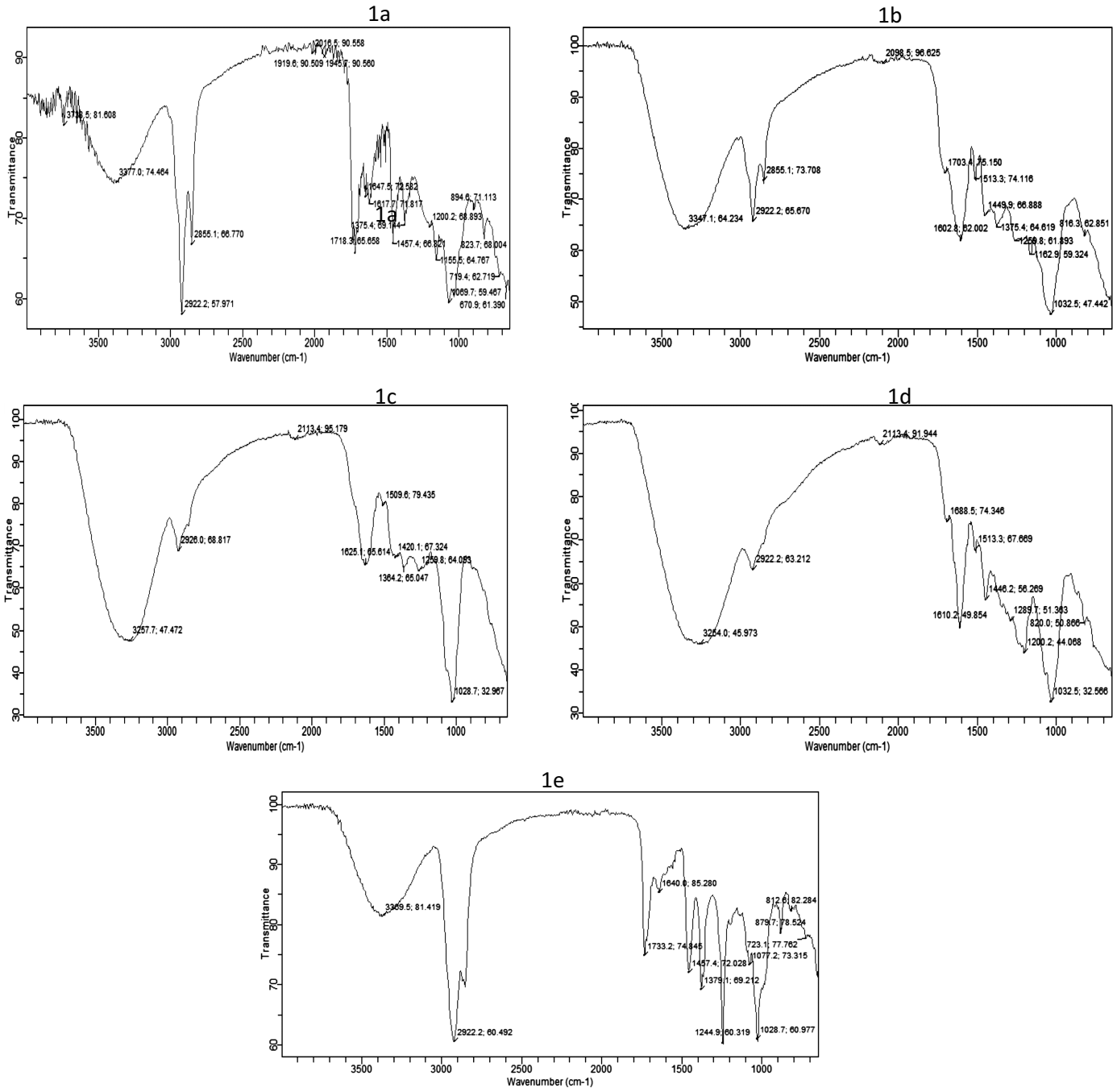

Figure 1: FTIR spectra of *Citrus aurantiifolia* (1a), *Cymbopogon citratus* (1b), *Lawsonia inermis* (1c), *Mangifera indica* (1d) leaves and *Alstonia boonei* (1e) stembark extracts

Ultra-Violet (UV) spectroscopy

The UV identified absorbance of electronic transitions at different wavelengths in each plant sample indicating presence of varied functional groups such as aromatic or conjugated compounds (Figure 2a - 2e).

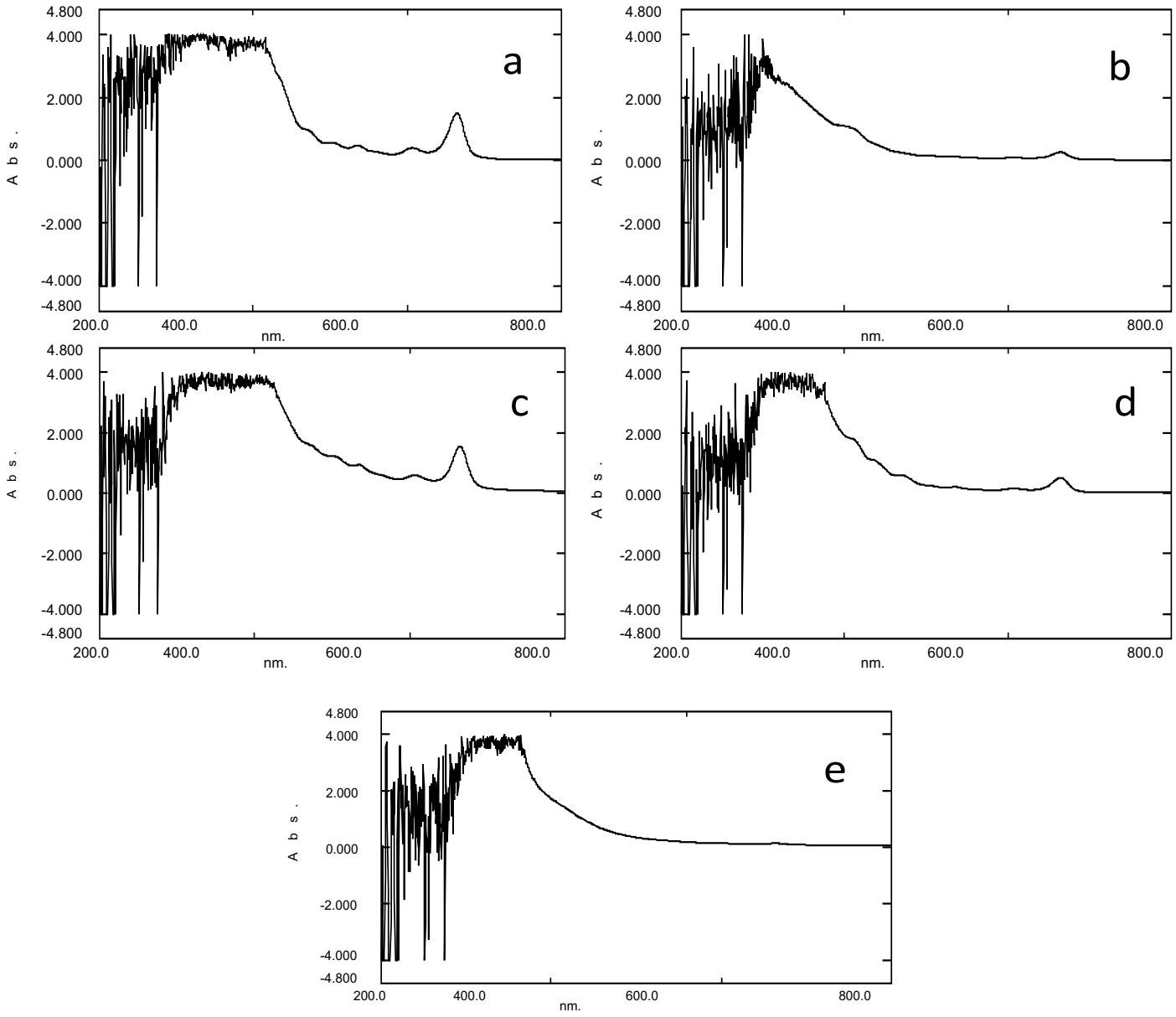

Figure 2: UV spectra of *Citrus aurantiifolia* (a), *Cymbopogon citratus* (b), *Lawsonia inermis* (c), *Mangifera indica* (d) leaves and *Alstonia boonei* (e) stembark extracts

DISCUSSION

Cyanogenic glycosides, also known as alpha-hydroxynitrile glycosides, are a distinctive class of secondary metabolites, contained in various plant species, having the nitrile moiety which liberates hydrogen cyanide after enzymatic degradation.[15]

The percentage yield of each plant sample was calculated to reflect the extractive value of the plants using methanol. Table 1 presented the percentage yield of the crude extract of *Citrus aurantiifolia, Cymbopogon citratus, Lawsonia inermis, Mangifera indica,* leaves as well as *Alstonia boonei* stembark, as 5.32 % w/w, 1.50 %w/w, 10.21 % w/w, 5.73 % w/w, and 1.70 % w/w, correspondingly based on their dry weights. Though, *Lawsonia inermis* has the lowest pulverized weight, it had much yield compared to others.

Phytochemical screening refers to the identification of the different classes of chemical constituents present in various plants. It helps to reveal several bioactive compounds contained in each plant qualitatively. Results presented in Table 2 showed the preliminary phytochemical examination of each of the plant samples. The leaves of *Citrus aurantiifolia* were found to contain mainly saponins, flavonoids, cardiac glycosides, tannins and alkaloids. Steroids were detected in moderate quantities while anthraquinones were not detected. This

result was compared to previous reports, which revealed that the bioactive components present in *Citrus aurantiifolia* include essential oils that consist mainly of citral (7.49 %), β-pinene (9.02 %), d-limonene (35.98 %) and α-terpineol (8.12 %).[16,17] It also comprises flavonoids - apigenin, rutin, quercetin and kaempferol,[18] phenolic acids, which consists largely of tannic acid and minorly of ferrulic and coumaric acids[19] as well as alkaloids, mainly synephrine, tyramine and octopamine.[20] The leaves of *Cymbopogon citratus* was observed to contain flavonoids, tannins, cardiac glycosides, steroids, alkaloids and saponins, but anthraquinones were not detected in the plant sample. Several studies reported that *Cymbopogon citratus* contained phenolics (caffeic, elemicin, p-coumaric acids, hydroquinone and catechol), steroids and fatty alcohols (beta-sitosterol, triacontanol and hexaconsanol), tannins (hydrolysable tannins), flavonoids (luteolin, leuteolin, quercetin, kaempferol, apigenin) and terpenoids (myrcene, citral, geranial- responsible for the lemony smell that characterizes the species).[21,22] The leaves of *Lawsonia inermis* were observed to be rich in saponins, flavonoids, steroids, tannins, and cardiac glycosides. Alkaloids were not detected in the plant sample. The leaves of *Lawsonia inermis* were reported to contain flavonoids (luteolins, apigenin and their glycosides), Steroids (β-sitosterol), tannins, gallic acid, fat, resin and mucilage.[23,24] Kirkland and Marzin,[25] also revealed the presence of 2- Hydroxy-1,4-naphthoquinone compound, responsible for the natural dye of the plant. The leaves of *Mangifera indica* were shown to be rich in saponins, cardiac glycosides, steroids, tannins flavonoids, and alkaloids however, anthraquinones were not detected. Various studies revealed that the critical constituents present in *Mangifera indica* Linn., are polyphenolics, flavonoids and triterpenoids and the plant is further reported to contain mangiferin (xanthone glycoside), isomangiferin, tannins and gallic acid.[26] The stem-bark of *Alstonia boonei* was observed to be rich in saponins, cardiac glycosides, steroids, alkaloids, tannins and flavonoids. Anthraquinones were not detected in the leaves. Reportedly, the leaves, stem bark and root bark of *Alstonia boonei* are rich in tannins, alkaloids, saponins, steroids, triterpenes, cardiac glycosides, cyanogenic glycosides, carbohydrates, and reducing sugars in varying amounts.[27 - 32]

The sodium picrate paper test is the standard qualitative test for the detection of cyanogenic glycosides. It is based on the reaction between the liberated hydrogen cyanide (HCN) gas and the sodium picrate paper to produce a reddish-brown complex (Sodium isopurpurate). Data in Table 3 represented the results for the sodium picrate paper test carried out on the leaves of *Citrus* aurantiifolia, C*ymbopogon citratus, Lawsonia inermis, Mangifera indica* and the stembark of *Alstonia boonei,* to detect the presence of cyanogenic glycosides. Cyanogenic glycosides were, however, not detected in any of the plant samples. The results were compared to previous research works that suggested the presence of cyanogenic glycosides in some of the plant samples such as in the stembark of *Alstonia boonei*[33] and in the leaves of *Citrus aurantiifolia*.[34] The disparity in results may be attributed to several factors including environmental factors and geographical location of the plants. Also, reports have shown that cyanogenic glycosides are present in the seed kernels of *Mangifera indica*.[35] Medicinal properties vary by plant part, one part of a plant could be harmless while another part of the same plant is toxic due to different phytoconstituents[36].

A linear connection was observed between cyanide concentrations of 0 - 200 (µg/mL). The determination of cyanide content was carried out to obtain the actual concentrations of hydrogen cyanide present in the plant samples. For the evaluation of cyanide content using the suspended strip method, a significant factor is the quantity of picrate impregnated in the strip. It was therefore ensured that the ten strips, used for analysis were prepared under equal environments so that the strips used for the analysis were as constant as possible. The cyanide present in the test samples vaporized as hydrogen cyanide (HCN) when 20 % HCl solution was added, followed by heating at eighty degrees Celsius. The volatility of HCN from biological substances containing cyanogenic glycosides on addition of hot mineral acid has been reported.[14,37] The alkaline picrate solution served as a trapping agent of the liberated HCN.

From the quantitative analysis results of the leaves of *Citrus aurantiifolia, Cymbopogon citratus, Lawsonia inermis, Mangifera indica* and the stembark of *Alstonia boonei* presented in Table 4, the cyanide concentration obtained for each plant sample was negative. This could imply that cyanogenic glycosides were not detected in significant amounts validating the results of the qualitative tests carried out on the plant samples.

The peaks observed in the FTIR spectra of the leaves of *Citrus aurantiifolia* were 3377.0 cm^{-1}, which indicates O-H stretching (alcohols or carboxylic acids) or N-H stretching (amines or amides), 2922.2 cm^{-1} and 2855.1 cm^{-1} representing C-H stretching vibrations. Peak at

1718.3 cm^{-1} suggests the presence of a C=O (carbonyl) group, from an aldehyde, ketone, ester, or carboxylic acid. 1647.5 cm^{-1} and 1617.7 cm^{-1} indicates C=C stretching (alkenes) or N-H bending in amides. The presence of C-O stretching alcohols, ethers or esters is shown by 1069.7 cm^{-1} (Figure 1a). The UV spectroscopy showed absorbance values across wavelengths from 200 nm to 800 nm. This range covers the UV region and extends into the visible spectrum. There are several peaks and troughs throughout the spectrum. The highest absorbance values are generally in the 200 - 300 nm range. The absorbance gradually decreases as the wavelength increases beyond 300 nm then a peak in the 600 - 650 nm region. Around 215 nm, there is high absorbance (> 3.5) while around 240 - 250 nm there are multiple peaks with absorbance > 3 and at 280 - 300 nm region, the peaks had absorbance > 3.5. These peaks correspond to electronic transitions in the molecule, such as $\pi \rightarrow \pi^*$ or $\pi \rightarrow \pi^*$ transitions, which are common in organic compounds with conjugated systems or heteroatoms (Figure 2a).

The FTIR spectrum of the leaves of *Cymbopogon citratus* (Figure 1b) revealed that the peak at 3347.1 cm^{-1} suggests the presence of O-H stretching, likely from alcohols or carboxylic acids. It could also indicate N-H stretching from amines or amides. The presence of C-H stretching vibrations, which is typical of alkyl groups, is seen from the presence of 2922.2 cm^{-1} and 2855.1 cm^{-1}. The peak at 1703.4 cm^{-1} indicates the presence of a C=O (carbonyl) group, from aldehydes, ketones, esters, or carboxylic acids. C=C stretching in aromatic rings or C=N stretching is shown at 1602.8 cm^{-1}. The presence of aromatic ring stretching is seen at 1513.3 cm^{-1}. Other notable peaks are 1449.9 cm^{-1} and 1375.4 cm^{-1} for C-H bending vibrations, 1250.8 cm^{-1} due to C-O stretching in esters or C-N stretching in amines and 1032.5 cm^{-1} for C-O stretching in alcohols, ethers, or esters. The UV spectrum showed several absorption peaks in the UV region, particularly between 200 - 300 nm. There's a strong absorption peak around 205 - 215 nm, with absorbance values exceeding 3.0. Another significant absorption region was observed around 275 - 285 nm, with absorbance values reaching up to 4.0. The absorption gradually decreases as the wavelength increases, with relatively low absorption in the visible region (above 400 nm). The compound likely contains aromatic rings, as suggested by the UV absorption in the 250 - 280 nm range and the FTIR peaks at 1602.8 cm^{-1} and 816.3 cm^{-1}. The strong UV absorption below 220 nm could be due to $\sigma \rightarrow \sigma^*$ or $\pi \rightarrow \sigma^*$ transitions, while the absorption around 275 - 285 nm might be attributed to $\pi \rightarrow \pi^*$ transitions in aromatic or conjugated systems (Figure 2b).

The FTIR spectrum of the leaves of *Lawsonia inermis* showed several peaks at different wavenumbers (Figure 1c). The O-H or N-H stretching, typical of alcohols, phenols, or amines is seen at 3257.7 cm^{-1}, and 2926.0 cm^{-1} for C-H stretching, common in organic compounds. Other notable peaks include 2113.4 cm^{-1} (C=C stretching in alkynes or C=N stretching in nitriles), 1625.1 cm^{-1} (C=C stretching in alkenes or C=O stretching in amides), 1509.6 cm^{-1} (aromatic C=C bending), 1420.1 cm^{-1} and 1364.2 cm^{-1} (C-H bending in alkanes), 1259.8 cm^{-1} (C-N stretching in amines or C-O stretching in esters) and 1028.7 cm^{-1} (C-O stretching in alcohols or ethers). In the UV spectrum (Figure 2c), there are several peaks and troughs in the 200 - 250 nm range, indicating complex absorption patterns in the far UV region. A broad, strong absorption is seen from about 280 nm to 400 nm, with multiple peaks. The absorption gradually decreases from 400 nm to 530 nm. The UV spectrum indicates the presence of conjugated systems or aromatic structures, which typically absorb in the 200 - 400 nm range. The complex absorption pattern suggests multiple chromophores or a mixture of compounds. The broad absorption in the UV spectrum could indicate the presence of extended conjugated systems.

The peaks observed in the FTIR spectrum of the leaves of *Mangifera indica* are 3254.0 cm^{-1} (O-H or N-H stretching for alcohols, phenols, or amines), 2922.2 cm cm^{-1} (C-H stretching for alkanes), 3113.4 cm^{-1} (aromatic C-H stretching), 1688.5 cm^{-1} (C=O stretching (carbonyl group, ketones or aldehydes), 1610.2 cm^{-1} (C=C stretching (alkenes or aromatics), 1513.3 cm^{-1} (Aromatic C=C bending), 1446.2 cm^{-1} (C-H bending for alkanes), 1289.7 cm^{-1} (C-O stretching for alcohols, ethers, or esters), 1200.2 cm^{-1} (C-N stretching) and 1032.5 cm^{-1} (C-O stretching for alcohols or ethers) (Figure 1d). The UV spectrum showed complex absorption patterns in the 200 - 300 nm range, indicating multiple chromophores or a mixture of compounds. Strong absorption is observed between 280 - 400 nm, suggesting conjugated systems or aromatic structures. The absorption gradually decreases from 400 nm to 490 nm. The UV-VIS spectrum supports the presence of conjugated systems or aromatic structures, as evidenced by the strong absorption in the 280 - 400 nm range. The complex absorption pattern in the 200 - 300 nm range suggests that the compound

might be a mixture or have multiple chromophores (Figure 2d).

In the FTIR spectrum of the stembark of *Alstonia boonei*, the peaks observed are 3369.5 cm^{-1} (O-H stretching; alcohols or carboxylic acids) or N-H stretching (amines or amides), 2922.2 cm^{-1} (C-H stretching), 1733.2 cm^{-1} (C=O stretching; carbonyl group; esters or ketones), 1640.0 cm^{-1} (C=C stretching; alkenes or C=O stretching for amides), 1457.4 cm^{-1} and 1379.1 cm^{-1} (C-H bending; alkanes), 1244.9 cm^{-1} (C-O stretching; alcohols, ethers, or esters), 1077.2 cm^{-1} and 1028.7 cm^{-1} (C-O stretching; alcohols, ethers, or esters) (Figure 1e). The UV spectrum showed absorption in the UV region (200 - 800 nm). There is a strong absorption peak around 205 - 206 nm, which could indicate $\pi \rightarrow \pi^*$ transitions in conjugated systems or $\pi \rightarrow \pi^*$ transitions in carbonyl compounds. Peaks around 295 - 300 nm, suggest extended conjugation or aromatic systems. The absorbance gradually decreases from 300 nm to 544 nm, indicating less absorption in the visible region (Figure 2e).

Table 5: Some functional groups, their corresponding electron transitions and wavelengths

Chromophore	Compound	Transition	λmax (nm)	ε
C–H	CH_4	$\sigma \rightarrow \sigma^*$	122	
C–C	C_2H_6	$\sigma \rightarrow \sigma^*$	135	
C=C	C_2H_4	$\pi \rightarrow \pi^*$	103	15000
			174	5500
C=C	R–C=C–R	$\pi \rightarrow \pi^*$	178	10000
C–N	Amino	$\pi \rightarrow \sigma^*$	190 - 200	2500 - 4000
C–S	R–S–R	$\pi \rightarrow \sigma^*$	235	180
C=O	Carboxylic acid	$\pi \rightarrow \pi^*$	200	50
C=O	Ester	$\pi \rightarrow \pi^*$	210	50
C=O	Amide	$\pi \rightarrow \pi^*$	205	200
C=N	$(NH_2)_2C=NH$	$\pi \rightarrow \pi^*$	265	15
C=N	$CH_3C=N$	$\pi \rightarrow \pi^*$	<170	
N=N	Me–N=N–Me	$\pi \rightarrow \pi^*$	350 - 370	15
N=O	Me_3NO	$\pi \rightarrow \pi^*$	300	100
			665	120
N=O	Me_3NO_2	$\pi \rightarrow \pi^*$	276	27
C–Br		$\pi \rightarrow \sigma^*$	208	300
C–I		$\pi \rightarrow \sigma^*$	259	400

Table 5 suggests that the cyanide (C≡N) group was not detected in any of the UV spectra of the leaves of *Citrus aurantiifolia, Cymbopogon citratus, Lawsonia inermis, Mangifera indica* and the stembark of *Alstonia boonei,* as the $\pi \rightarrow \pi^*$ transition was not observed at a wave length less than 170 nm in the UV spectra of any of the plant samples.

CONCLUSION

This study analyzed cyanogenic glycosides in the leaves of *Citrus aurantiifolia, Cymbopogon citratus, Lawsonia inermis, Mangifera indica* and *Alstonia boonei* stembark, using qualitative and quantitative procedures. It can be inferred from the results of the experiment that the plant samples, locally used in the preparation of anti-malaria herbal decoction in Nigeria, do not contain harmful levels of cyanide and may be considered safe for consumption by humans.

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