

Phytochemical and FTIR Analyses of Six Medicinal Plants from Pankshin Local Government Area of Plateau State, Nigeria

ABSTRACT

Aim: To analyze the phytochemical and FTIR of six medicinal plants frequently used by the Ngas in Pankshin District of Plateau State, Nigeria in the traditional treatment of some ailments.

Methods: The medicinal plants were extracted with water, ethanol and cyclohexane/hexane, screened for the presence and levels of phytochemicals by standard methods and the FTIR analyses were carried out on the ethanolic extracts.

Results: The study revealed that the aqueous extracts contained most of the phytochemicals analyzed. The quantitative analysis of five phytochemicals in four of the plants showed that the order of the levels of phytochemicals in plants were: alkaloids; *F.phytophylla*>*C.tinctorium*>*F.abutilifolia*>*F.phytophylla*, saponins; *A.precatorius*>*F.phytophylla*>*F.abutilifolia*>*C.tinctorium*, terpenoids; *F.phytophylla*>*F.abutilifolia*>*C.tinctorium*>*A.precatorius*, flavonoids; *F.phytophylla*>*F.abutilifolia*=*C.tinctorium*>*A.precatorius* and glycosides, *F.abutilifolia*>*F.phytophylla*>*C.tinctorium*>*A.precatorius* with saponins being the most abundant phytochemical in the study and flavonoids the least. *F.phytophylla* was richest in the phytochemicals while *A.precatorius* was the least. The presence of characteristic functional groups such as —OH, C—O, —C=C and C—H indicated the presence of phenolic compounds, carboxylic acids, alcohols, carbohydrates, and proteins in the plants that could be responsible for various medicinal properties in the plants.

Conclusion: The results justified the local use of the plants in the treatment of illnesses like malaria fever, yellow fever, cough and healing of wounds among the Ngas people in Pankshin Local Government of Plateau State. These compounds can be harnessed for industrial and pharmaceutical utilization.

Key Words: Phytochemical, *C.tinctorium*, *F.phytophylla*, *F.abutilifolia*, *M.acuata*, *A.hispidium*, *A.precatorius* and FTIR,

1. INTRODUCTION

“Natural products, especially those derived from plants, have been used to help mankind sustain his health since the dawn of medicine. Ngas are the major inhabitants of Pankshin District and had its mode of herbal treatment (which was also linked to traditional worship) been the dominant method of healing therapy until the advent of modern medicine. The Ngas traditional healer provides health care services based on culture, religious background, knowledge, attitudes, and beliefs that are prevalent in his community. Illness is regarded as having both natural and supernatural causes and thus must be treated by both physical and spiritual means, using divination, incantations, animal sacrifice, exorcism, and herbs” [1]. The Ngas knowledge of herbal medicine is confined to the older generation and transmitted from one generation to another [2]. “In a pharmaceutical landscape, plants with long history of use in ethno medicine are a rich source of active phyto constituents that provide medicinal or health benefits against various ailments and diseases” [3]. There is need to appraise the values of the phyto-medicine in order to appropriately reap its benefits [4]. “China, for example, is able to provide adequate health care for its vast urban and rural population due to the amalgamation of the traditional health care with the modern medicine” [5, 6]. “There is also need for Nigeria to officially recognize and incorporate the herbal preparations into the local healthcare. This could help develop the very essence of indigenous legacy of medicinal knowledge. Today, in order to contribute to the protection of the medicinal plants, research should be supported to search for the different chemical compounds produced by these plants in order to

isolate the active molecules that can serve as models for a synthesis of new molecules that mainly respond to the safety and efficacy of conventional drugs" [7]. "The formulation of synthetic molecules could not only contribute to the conservation of species in their biotope and thus to the protection of the environment, but also to find solutions to public health" [7].

"Medicinal plants are considered a repository of numerous types of bioactive compounds with varied therapeutic properties. In fact, medicinal plants produce much diversified natural substances. At least 12,000 such compounds have been isolated so far; a number estimated to be less than 10% of the total" [8]. "They accumulate these secondary metabolites which represent an important source of molecules usable by man in pharmacological field. Secondary metabolism of plant is characterized by a structural diversity, which confers very important chemical and biological properties. The therapeutic potential of plants has been well explored over a very long time. The vast array of therapeutic effects associated with medicinal plants includes anti-inflammatory, antiviral, antitumor, antimalarial and analgesic" [9]. "This efficacy depends upon the current knowledge about taxonomic features of plant species, plant parts and biological property of medicinal plants, which in turn depends upon the occurrence of primary and secondary metabolites" [10]. Plants are applied in different forms such as poultices, concoctions of different plant mixtures, infusions as teas or tinctures or as component mixtures in porridges and soups administered in different ways including oral, nasal (smoking, snuffing or steaming), topical (lotions, oils or creams), bathing or rectal (enemas) [11]." These remedies are socially accepted because of their efficacy, safety, cost-effectiveness and easy availability. The risks of cancer, cardiovascular disease, diabetes and other diseases related with ageing can be reduced by using bioactive ingredients as food supplement. In modern research, there is an international trend towards the use of the natural phytochemicals present in teas, herbs, beans, fruits, berry crops and vegetables as a remedy for most of the common ailments" [4]. "A number of various environmental factors such as climate, altitude, rainfall and other conditions may affect growth of plants, which in turn affect the quality of herbal ingredients present in a particular species even when it is produced in the same country" [2]. "These conditions may produce major variations in the bioactive compounds present in plants" [12]. The current study is therefore aimed at investigating the folkloric claims of extracts from six plants in treating different ailments by Ngas natives.

2. MATERIALS AND METHODS

2.1 PLANTS COLLECTION

Six medicinal plants (*Cochlospermum tinctorium*, *Ficus phytophylla*, *Ficus abutilifolia*, *Mussaenda acuata*, *Acanthospermum hispidum*, *Abrus precatorius*) were collected from their natural habitat of Pankshin Local Government Council of Plateau State, Nigeria. The samples were identified at the herbarium section of the College of Forestry, Jos. The plant samples were washed under running tap water, air dried in a shade at room temperature, milled into a fine powder using a mixer grinder and sieved to give particle size of 50-150mm then stored in air tight containers for extraction.

2.2 CHEMICALS: The entire chemicals used in the present study were of analytical grade.

2.3 PREPARATION OF PLANT EXTRACTS

"Three solvents (water, ethanol, and cyclohexane) were used for extractions. 100g of homogenized fine powders of leaf, bark and root were separately soaked in different conical flasks containing 250ml each of distilled water, 40% ethanol and cyclohexane (v/v) and were allowed to stand for an hour on a water bath with occasional shaking. These were then kept on rotary shaker at 200rpm for 24h. Finally, each sample extract was obtained using Soxhlet apparatus" [12, 13].

2.4 QUALITATIVE AND QUANTITATIVE ANALYSES OF PHYTOCHEMICAL CONSTITUENTS

The qualitative analysis of the extracts was carried out in accordance with standard methods as described by [14, 15, and 16]. The levels of alkaloids, flavonoids, saponins, cardiac glycoside and terpenoids contents of the plant were determined by the methods described by [16, 15, 13, and 17]

2.5 FOURIER TRANSFORMS INFRARED (FT-IR) ANALYSIS

Dried powder of the plant extracts was used for FT-IR analysis. The dried extract powder was encapsulated in 100 mg of KBr pellet, in order to prepare translucent sample discs. The powdered sample of each extracts was loaded on a Universal ATR sampling accessory infrared spectrophotometer. The ATR inspecting gadget used was a DuraSample IR single-pass jewel covered inner reflection adornment (Smiths Detection, Danbury, CT,USA) and a predictable contact weight was connected by

method for a hardened steel bar and an electronic load show. The information retrieved was taken to ensure that the window (2 mm in measurement) of the ATR testing device was secured totally by fiber. FT-IR spectra of the extracts were recorded at room temperature (25 ± 2 °C), a scan range from 550 to 4000 cm^{-1} at a resolution of 4 cm^{-1} and the number of scans per sample was four [18, 19].

2.6 STATISTICAL ANALYSIS

The data obtained from this study were expressed as Mean \pm Standard error of mean using statistical package for social sciences (SPSS) version 23.0.

UNDER PEER REVIEW

3. RESULTS AND DISCUSSION

Table 1: Results of the phytochemical screening of cyclohexane, ethanolic and aqueous extracts of six plant samples

Class of Phytochemicals	Extract type/plant species																										
	Cyclohexane									Ethanolic									Aqueous								
	A	B	C	D	E	F	G	H	I	A	B	C	D	E	F	G	H	I	A	B	C	D	E	F	G	H	I
Plants																											
Saponin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Steroids	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	+	-	-	-	-	-	-	-	-	-
Terpenoid	+	+	+	+	+	+	+	NA	NA	+	+	-	+	+	+	+	NA	NA	+	+	+	+	+	+	+	NA	NA
Phenol	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	+	+	+	-	-	-	-	+
Alkaloid	+	+	+	+	-	-	-	+	+	+	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+	+
Flavonoid	-	-	-	-	+	+	+	+	+	-	-	-	-	+	+	+	+	-	+	+	+	+	+	+	+	+	+
Glycoside	+	+	+	+	+	+	+	NA	NA	+	+	+	+	+	+	+	NA	NA	+	+	+	+	+	+	+	NA	NA
Quinone	+	+	+	+	+	+	+	+	+	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Tannin	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

Key

- A= *Cochlospermum tinctorium* leaves extracts
- B = *Cochlospermum tinctorium* roots extracts
- C = *Ficus phytophylla* leaves extracts
- D = *Ficus phytophylla* bark extracts
- E= *Ficus abutilifolia* leaves extract
- F= *Ficus abutilifolia* roots extract
- G= *Abrus precatorius* leaves extract
- H = *Mussaenda acuata* leaves extracts
- I = *Acanthospermum hispidum* (aerial parts) extracts
- NA = Not Analysed
- + = Presence of phytochemicals
- = Absence of phytochemicals

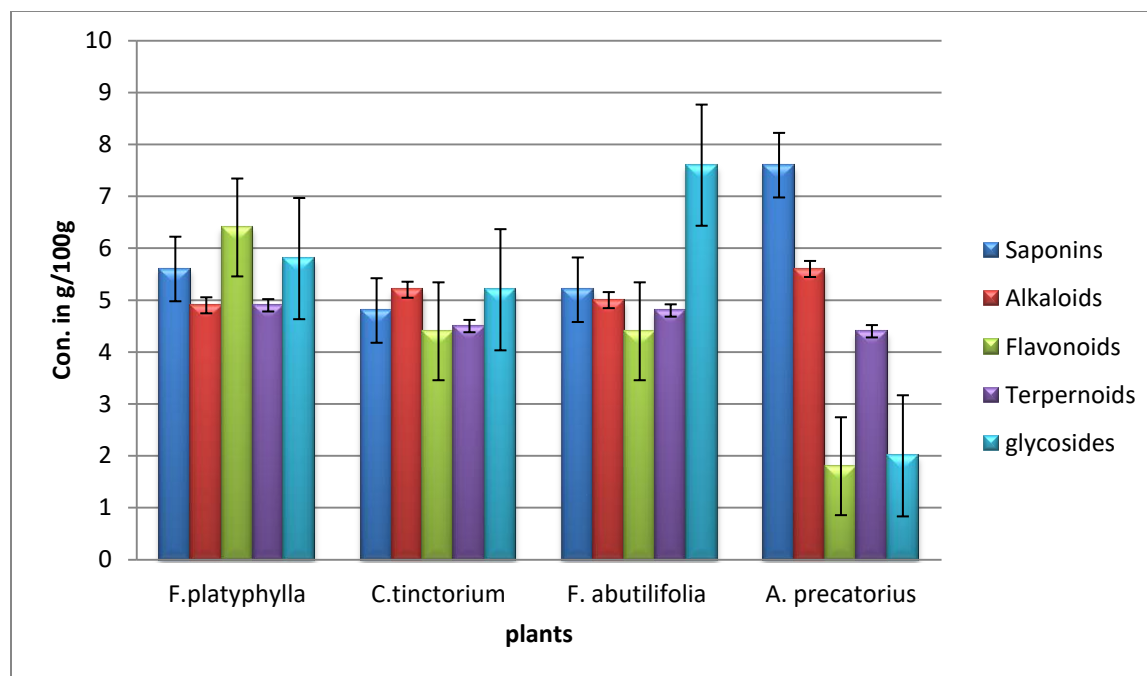


Figure 1: Results of quantitative analysis of the phytochemical constituents/100g in parts used for medication in four plants' species.

3.1 Qualitative and Quantitative analyses of plants extracts

From the result of qualitative analysis (Table 1), the three solvents showed the presence of saponins, terpenoids (except in ethanolic extracts of *F. phytophylla* leaves), alkaloids (except in cyclohexane extracts of *F. abutilifolia* leaves, *F. abutilifolia* roots, *A. precatorius* leaves extract, the ethanolic extracts of *M. acuata*. and *A. hispidum*), Flavonoids (except in the cyclohexane extracts of *C. tinctorium* leaves, *C. tinctorium* roots, *F. phytophylla* leaves, *F. phytophylla* bark, and the ethanolic extract of aerial parts of *Acanthospermum hispidum*), glycosides, quinones (except in the ethanolic extracts of *C. tinctorium* leaves, *C. tinctorium* roots, *F. phytophylla* leaves and *F. phytophylla* bark), tannins (except in the cyclohexane extracts of *C. tinctorium* leaves, *C. tinctorium* roots, *F. phytophylla* leaves, *F. phytophylla* bark), steroids (except in the ethanolic extracts of *M. acuata* and *A. hispidum*) and phenols (except in the aqueous extracts of *C. tinctorium* leaves, *C. tinctorium* roots extracts, *F. phytophylla* leaves and *F. phytophylla* bark). The order of the presence of the phytochemicals in the extracts of the three solvents was aqueous > ethanolic > cyclohexane.

"The leave samples of the selected plants (Figure 1) showed that *C. tinctorium* had the lowest average concentrations (4.80 ± 2.32 g/100g dw) of saponins while *A. precatorius* displayed highest source of saponins (7.60 ± 3.23 g/100g dw). Saponins are widely distributed in the botanical kingdom and have many pharmacological actions and biological activities. Saponins have the pharmacological effects like anti-inflammatory, molluscicidal, antimicrobial, antispasmodic, antidiabetic and anticancer, hypocholesterolemic, antioxidant, anticonvulsant and analgesic, anthelmintic, antitussive and cytotoxic activities" [20]. "Generally saponins are toxic, but consumption of saponins by human beings may be beneficial in reducing heart disease (binding of saponins with plasma membrane and cholesterol)" [21]. The presence of steroidal saponins could develop resistance to viral disease such like cancer. Saponins have expectorant action which is very useful in the management of upper respiratory tract inflammation and stop bleeding and in treatment of wounds.

"Variable concentrations of alkaloids were recorded in the leaves of studied plants. *A. precatorius* showed highest alkaloids (5.6 ± 1.99 g/100g dw) and *F. platyphylla* was lowest (5.00 ± 1.23 g/100g dw). Alkaloids are believed to function as defensive elements against predators, especially mammals because of their general toxicity and deterrence capability as well as analgesic, anti-inflammatory and adaptogenic activities which help to alleviate pains, developed resistance against diseases and endurance against stress" [21].

“Analysis of leaves showed that *F. platyphylla* was the richest source of flavonoids (4.90 ± 2.12 g/100g dw) and had more than two folds higher flavonoids than it was in *A. precatorius* (1.80 ± 0.54 g/100g dw). Flavonoids are generally nontoxic and manifest a diverse range of biologically beneficial activities. Epidemiological studies have provided data that high dietary intake of flavonoids with fruits and vegetables could be associated with low cancer prevalence in humans. These include carcinogen inactivation, anti-proliferation, cell cycle arrest, induction of apoptosis and differentiation, inhibition of angiogenesis, antioxidation and reversal of multidrug resistance mechanisms. The flavonoids are polyphenolic compounds found as integral components of the human diet. They are universally present as constituents of flowering plants, particularly of food plants. Several plants and spices containing flavonoid derivatives have found application as disease preventive and therapeutic agents in traditional medicine in Asia for thousands of years” [21].

The yellow root or rhizomes of *Cochlospermum tinctorium* are used in many local remedies. From this work, the aqueous extract of the roots contained all the phytochemicals analyzed (except steroid). The plant is usually administered for treating yellow fever by Ngas people of Pankshin District. Other literatures have it that, the aqueous extract of the roots mixed with shear butter and other oils is applied for burns. A root decoction or infusion is taken with other herbs for stomach troubles and urethral discharges [22]. “The root decoction is drunk for orchitis worms and fever in general. It is also used as liniment for epilepsy, pneumonia, intercostals pains, bronchial infections and swelling, and it is said to make a good sitz-bath cure for piles, and to be used for drops for conjunctivitis. The powdered root in water or millet beer is said to cure jaundice and local application to cure bites of poisonous snakes” [22, 23]. Haidara et al., 2016 [23] reported that, “*C. tinctorium* is one of the plants used in rural areas of Mali for the management of malaria, abdominal pains and dermatitis and to treat jaundice, gastro intestinal diseases or ailments, schistosomiasis and dysuria. According to them, this species is also used to treat infectious diseases in Guinea Conakry and in the West African sub-region for the management of various affections, including pain and inflammation”

Aqueous extracts of *F. phytophylla* leaves and bark are used by the natives of Ngas to treat catarrh and common colds. *F. phytophylla* presented the highest flavonoid content in this work. Adeshina et al., (2010) [25] also found that, “the leaf extract of *F. platyphylla* had higher flavonoid contents in the leaves than the *F. sycomorus* hence the better antibacterial activity of its leaf extracts than for *F. sycomorus* leaf extract”.

The bark of *Ficus abutilifolia* is used by the Ngas people for the treatment of tumors, wounds and diseases associated or characterized by inflammation.

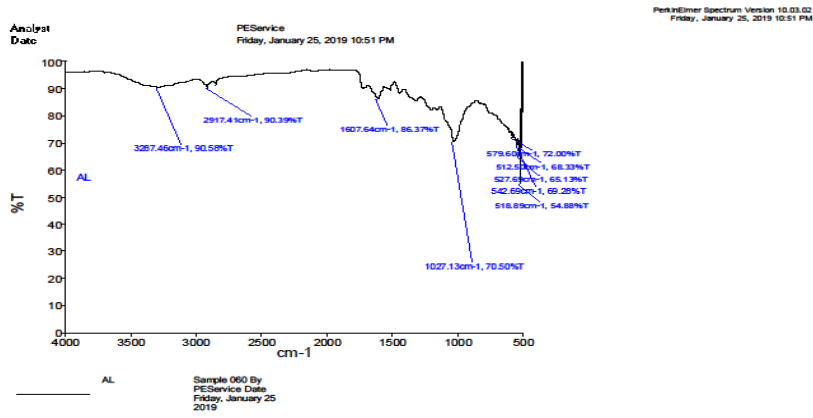
“*Acanthospermum hispidum* is used among Ngas people for treating diarrhea. Alban et al., (2014) [26] noted that only the hydroethanolic extract of *Acanthospermum hispidum* (HE7) showed an antibiotic power ($ap \leq 4$) on *E. coli*. They added that, *Acanthospermum hispidum* dominated by the presence of gallo tannins and accessorily triterpenes, steroids and mucilage, showed bactericidal activity against *E. coli*. According to them, this activity seemed to be due to its composition rich in gallo tannins and a synergistic activity with flavonoids, triterpenes, steroids and mucilage. According to Imosemi et al, (2014)” [27] “*Acanthospermum hispidum* LD50 above 5000 mg/kg is traditionally used in treating diabetes mellitus in North Western Nigeria. *Acanthospermum hispidum* DC. is found in the tribal area of Koraput and Malkangiri districts and traditionally used as anthelmintic, antidiabetic, antihypertensive, carminative and antifungal” [28].

The aqueous extract of leaves of *Mussaenda tomentosa* is used by the Ngas people for the treatment of common fever. Muruganandam et al., (2016) [29] found that, the methanol extracts of leaves of *Mussaenda tomentosa* showed the presence of phenols, flavonoids, glycosides, saponins, tannins, terpenoids and reducing sugars that have shown significant antioxidant and antidiabetic properties.

The results of Fourier Transform Infrared (FTIR) analysis

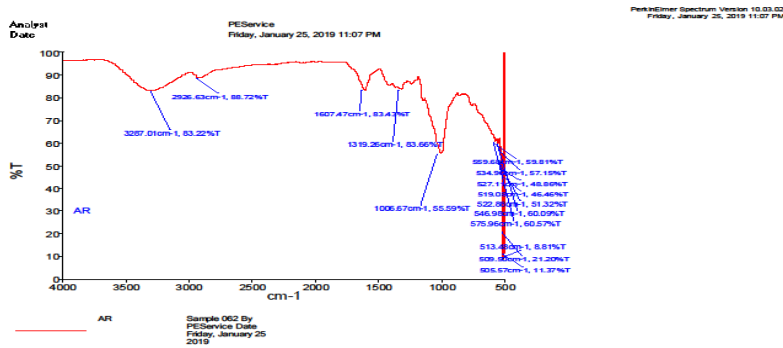
The FTIR spectrum was used to identify the functional groups of the active components based on the peak values in the region of infrared radiation. The FTIR spectra profiles of the ethanolic extracts of *C. tinctorium* (leaves and roots), *A. precatorius* (leaves), *M. acuata* (aerial parts), and *A. hispidum* (aerial parts) are presented in Figures 2 – 6.

3.2 FTIR Analysis



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Figure 2: (FTIR) Spectrum of *C. tinctorium* (leaves)



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Figure 3: Fourier Transform Infrared (FTIR) Spectrum of *C. tinctorium* (Roots)

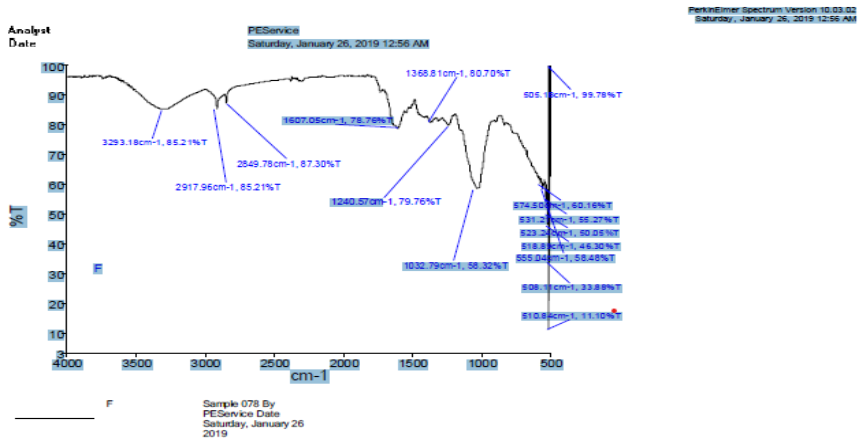


Figure 4: Fourier Transform Infrared (FTIR) Spectrum of *A. precatorius* (leaves)

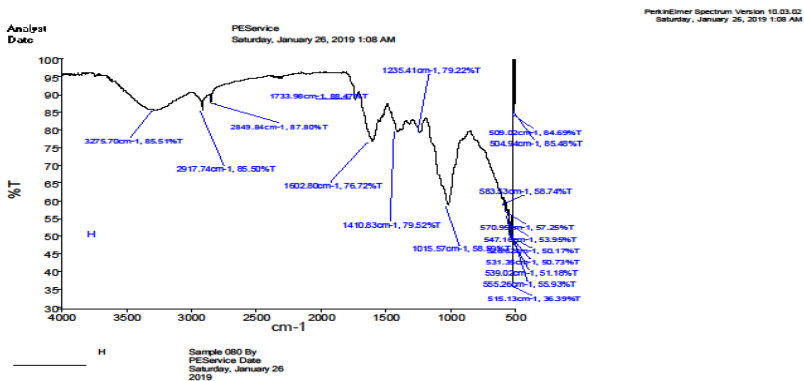


Figure 5: Fourier Transform Infrared (FTIR) Spectrum of *M. acuata* (aerial parts)

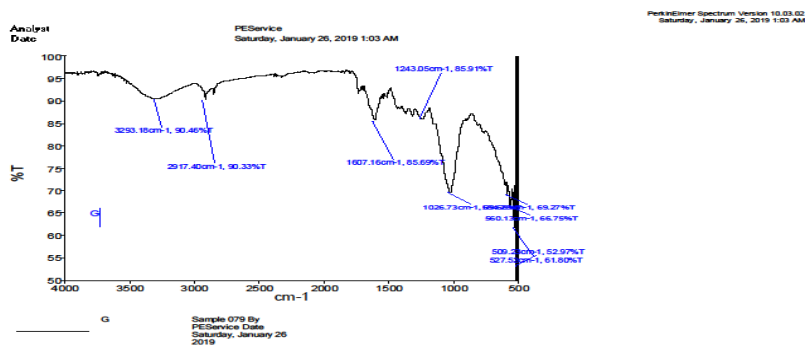


Figure 6: Fourier Transform Infrared (FTIR) Spectrum of *A. hispidum* (aerial parts)

The FT-IR spectrum has proven to be a valuable tool to identify the functional group of the active components. The FT-IR spectrum profile of various plants' tissue extracts was illustrated in Figures 2 – 6 above. The characteristic band occurring at 2921.0/2857.0 cm⁻¹ in the ethanolic extract of *C. tinctorium* (leaves), at 1368.81cm⁻¹ and 2917.96cm⁻¹ in *A. precatorius* (leaves), at 3275.70cm⁻¹ and 2849.84cm⁻¹ in *A. hispidum* (leaves), at 2917.40cm⁻¹ *M. acuata* (leaves) corresponding to the C–H symmetric stretching of methylene groups in aliphatic compounds. There was absorption in the region of 1607.64cm⁻¹ and 1319.26cm⁻¹ in *C. tinctorium* leaves and root respectively indicating that cyanide groups in leaf and root extracts which means that it contains toxic substances. The very strong absorptions observed at 3287.46cm⁻¹ ethanolic *C. tinctorium* (leaves) extract, at 2926.63cm⁻¹ in *C. tinctorium* (root) extract, at 3293.18cm⁻¹ in *A. precatorius* (leaves) extracts, at 3293.18cm⁻¹ *Acanthospermum hispidum* (leaves), and at 3293.18cm⁻¹ *M. acuata* (leaves) extracts indicating the presence of phytochemical carrying hydroxyl group (–OH) of polyphenolic such as, flavonoids and tannins provide a relative ranking of extracts in term of antioxidant activity. The absorption of the leaf's extracts of *C. tinctorium* at 1027.13cm⁻¹, at 1006.67cm⁻¹ in *C. tinctorium* root, at 1240.57cm⁻¹ *Abrus precatorius* (leaf) corresponding to the C–O symmetric stretching of acidic groups in the compounds. The bands observed at 1733.98cm⁻¹ in ethanolic *A. hispidum* (aerial part) is responsible for the stretching vibration of C=O group. The absorption bands at 2849.78cm⁻¹ and at 2849.84cm⁻¹ in *A. precatorius* (leaves) and *A. hispidum* (aerial part) extracts respectively corresponds to the stretching of double bonded compounds (=C-H) and at 1410.83cm⁻¹ to the stretching of double bond (C=C) in *A. hispidum* (aerial part) extracts. The root's extracts of *C. tinctorium*, *A. precatorius* and *M. acuata* (leaves) also indicated the presence of bands at 1607.64cm⁻¹, 1607.05cm⁻¹ and 1607.16cm⁻¹ (N - H) that provides the richness of the extract of protein. Felhi et al. 2017 and Fernando et al., 2019 in their works reported “the presence of characteristic functional groups such as –OH, C–O, C≡C and C–H of phenolic compounds, carboxylic acids, alcohols, carbohydrates, and proteins in the plant, that are responsible for various medicinal properties” [18, 19].

4. Conclusion

From this work, the aqueous extracts of the leaves, stem bark and root of *Cochlospermum tinctorium*, *Ficus phytophylla*, *Mussaenda acuata*, *Acanthospermum hispidum*, *Ficus abutilifolia* and *Abrus precatorius* contain most of the phytochemicals analyzed. The quantitative analysis of phytochemicals (alkaloids, saponins, terpenoids, flavonoids, and glycoside) in *Abrus precatorius* was found to contain 5.60mg, 7.60mg, 4.40mg, 1.80mg, and 2.00mg respectively; *Ficus abutilifolia* (leaf) was found to contain 5.00mg, 5.20mg, 4.80mg, 4.40mg, and 7.60mg respectively; *Ficus phytophylla*, 4.90 mg, 5.60 mg, 6.40 mg, 4.90 mg, and 5.80 mg respectively; *Cochlospermum tinctorium*, 5.20mg, 4.80mg, 4.50mg, 4.40mg and 5.2mg respectively. The presence of these phytochemicals accounts for the use of these plants for the medicinal purposes. If traditional mode of medication is incorporated with the modern medication, it

will develop the very essence of indigenous legacy of medicinal knowledge. Awareness should be created for the use of the herbs

COMPETING INTERESTS DISCLAIMER:

Authors have declared that they have no known competing financial interests OR non-financial interests OR personal relationships that could have appeared to influence the work reported in this paper.

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