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Phytochemicals and Antimicrobial Activity of Crude Extracts of Fresh Leaves and Stem Bark of *Faidherbia albida.*

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ABSTRACT: The Hausas and Fulanis of Northern Nigeria have a folklore claim about the usage of *Faidherbia albida* to treat diarrhea, inflamed eyes, skin infections, bleeding, rheumatism, pneumonia, and vomiting. The purpose of this study is to assess the phytochemical and antibacterial activities of crude extracts of fresh leaves and stem bark of *Faidherbia albida* sourced from Bodeno, Guyuk Local Government Area, Adamawa State, Nigeria using appropriate standard techniques after methanol extraction. Data obtained reveals that the stem bark extract contained alkaloids, flavonoids, phenols, tannins, saponins, and quinones but no terpenoids or resins. The extracts of the leaves contained alkaloids, phenols, terpenoids, and saponins, but no tannins or resins. The zone of inhibition of microorganism growth ranged from 9 to 13 mm for the stem bark extract and 11 to 16 mm for the leaf extract. The stem bark extract inhibited *Escherichia coli, Staphylococcus aureus,* and *Klebsiella pneumoniae* more effectively than the leave extract with the exception of *Pseudomonas* while suppression of *Salmonella typhii* is same. Fourier Transform Infra-Red Spectroscopy (FT-IR) examination of the stem bark extract revealed the following distinctive peaks. The hydroxyl vibrations in alcohol were attributed to 3294.9 cm^{-1} and 2930.9 cm^{-1}, respectively, whereas the peak at 1613.4 cm^{-1} displays the distinctive N-H bending of the amino group. The band at 1521.1 cm^{-1} represents the N-O stretch of a nitro group, while the band at 1444.0 cm^{-1} represents the C=C stretch of an alkene. This study established baseline information on the efficacy of crude extracts of *Faidherbia albida.*

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According to Rani *et al.* (2020), one of the main reasons of death in developing countries is thought to be microbial infection. The worldwide issue of public health protection and a key global conflict is the emergence of antibiotic resistance. Medicinal plants are a group of compounds (organisms) that accumulate numerous active components that can be used to treat a variety of human or animal ailments (Doughari and Saa-Aondo, 2021). Herbs' lengthy history of usage in medicine is a reliable sign of their worth and potential applications. The value of therapeutic plants in contemporary medicine is growing rapidly every day. Plants have long been utilized by humans to treat common infectious diseases, and certain traditional remedies are still used today to treat a variety of illnesses (Sathiya and Muthuchelian, 2008). Ameesh

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and Murugan (2016) reported that, about 80% of the world population rely on traditional medicine, predominantly plants. These practices incorporated ancient beliefs and were passed on from one generation to another by oral tradition and guarded literature as reported by Christopher *et al.* (2021).

The unparalleled availability of chemical variety in natural goods, whether in the form of pure chemicals or standardized plant extracts, offers countless chances for novel therapeutic leads (Parekh and Chanda, 2007). The need to find new molecular structures as lead compounds from the plant kingdom has resulted in a surge in scientific interest in medicinal plants in recent times, owing to the increased efficacy of new drugs derived from plants and growing concerns about the side effects of modern medicine (Nair *et al*., 2005). Numerous investigations have been conducted to ascertain the rationale behind the application of *F. albida* in conventional medicine. According to Irvine (2003) and Maitera and Chukkol (2016), *F. albida* is used to treat a variety of conditions, including gripe, earache, deafness, wounds, ulcers, psychiatric disorders, postpartum difficulties, opthalmia, and liver issues. Moreover, diarrhea, rheumatism, skin infections, bleeding, pneumonia, and vomiting are treated with it (Kubmarawa *et al*., 2007). According to Irvine (2003), the bark of *F. albida* is used as a liniment for pneumonia and as a treatment for leprosy. An infusion of the bark is also used as a febrifuge for coughs and as support during difficult childbirths. Hence, the objective of this paper is to assess the phytochemical and antibacterial activities of crude extracts of fresh leaves and stem bark of *Faidherbia albida* sourced from Bodeno ward, in Guyuk Local Government Area, Adamawa State, Nigeria.

MATERIALS AND METHODS

*Plant collection and preparation***:** Fresh leaves and stem bark of *Faidherbia albida* were collected from Bodeno ward, in Guyuk local government area of Adamawa state, Nigeria. The leaves and stem bark of *Faidherbia albida* were air dried at room temperature and then pulverized into fine powder using mortar and pestle and weighed.

*Sample extraction***:** Powdered *Faidherbia albida* leaves and stem bark were weighed 100 g each and placed into the thimble of a soxhlet extraction apparatus. About 400 ml of methanol was placed into the soxhlet and allowed to extract at 60 ^{0}C for one hour. The extract was then concentrated using the water bath set at 60 ^{0}C to one-tenth its original volume and then it was dried at room temperature**.**

*Phytochemical Screening***:** Phytochemical screening for the secondary metabolites of the pulverized plant sample was carried out using standard qualitative techniques (Brain and Turner, 1975) for the presence of saponins, tannins, alkaloids, flavonoids, glycosides, phenols and resins.

*Test organisms***:** The microorganisms that were used as test organism for this study are *Escherichia coli, Staphylococcus aureus Klebsiella pneumoniae, Salmonella typhi, Pseudomonas*.

*Antimicrobial activity screening***:** The antimicrobial activity tests of the extracts were carried out using some pathogens. The pure clinical bacteria isolates were obtained from the department of microbiology Federal Medical Centre Gombe, Nigeria. Agar well diffusion method was used to determine the antimicrobial activities of the plant extracts.

The stock solutions of the plant extract were made by weighing 0.2 g of each extract and dissolving in 10 ml of dimethylsuphoxide (DMSO) to obtain a concentration of 20mg/mL, this was the initial concentration. A sterile pasteur pipette was used to transfer 0.1 ml of inoculums into Muellar Hinton agar plate and a sterilized bent glass rod was used to spread the inoculums evenly over the entire plate, and allowed to set for 15 minutes. A sterilized cork borer of 6 mm in diameter was used to bore five wells (holes) at equidistant and 2-inch to the edge of the plate. Different concentrations of plant extracts (6.25 ug/ml, 125 ug/ml, 250 ug/ml and 500 ug/ml) were prepared and 0.5 ml from each concentration was transferred into each well. The plate was allowed to stand for 1 hr for pre-diffusion of the extracts to occur and then incubated at 37 ^{0}C for 24 hrs for bacteria. They were observed after the periods of incubation as to note the zone of inhibition of growth. The zones were recorded in millimeters of their diametrical section (Emaikwu *et al*., 2019).

FT-IR Analysis: Dried powder of the stem bark extract of *Faidherbia albida* was used for FT-IR analysis. 10mg of the dried extract powder was encapsulated in 100 mg of KBr pellet, in order to prepare translucent sample discs. The powdered sample of the stem bark extract was loaded in FTIR spectroscope (Shimadzu, IR Affinity 1, Japan), with a Scan range from 400 to 4000 cm^{-1} with a resolution of 4 cm^{-1}.

RESULTS AND DISCUSSION

Phytochemicals are biologically active, naturally occurring chemical compounds found in plants that protect plant cells from environmental threats such as pollution, stress, dehydration, UV exposure, and pathogenic attack (Nyamai *et al.*, 2016). According to

Christopher *et al.* (2019), these chemicals are classified as secondary plant metabolites and provide health benefits for humans. They are supposed to operate synergistically, helping the body to use nutrients more efficiently. Phytochemicals have several advantages, including low toxicity, low cost, and easy availability, as well as biological properties such as antioxidant activities, antimicrobial effects, modulation of detoxification enzymes, immune system stimulation, platelet aggregation reduction, hormone metabolism modulation, and antineoplastic properties (Andre *et al.*, 2010).

The preliminary phytochemical screening of both the stem bark and leaf extracts of *Faidherbia albida* revealed the presence of alkaloids, phenols, saponins, and quinones, while flavonoids and tannins were only found in the stem bark extracts and terpenoid was only found in the leaf extract, as shown in table 1. This finding is consistent with previous studies on the phytochemical composition of *Faidherbia albida*, which show the presence of similar secondary metabolites in various parts of the plant (Mahmoud *et al.,* 2013; Maitera and Chukkol, 2016), and it is also in contrast to Kubmarawa *et al.,* (2007), who stated that only volatile oil was present in the stem bark. These discrepancies could be ascribed to environmental alterations where the plants were harvested or seasonal changes that affected the plant components (Mahmoud *et al.,* 2013). It could possibly have been due to modifications during extraction and or storage (Maitera and Chukkol, 2016). It also depends on the drying process used.

Table 1: Result of phytochemical screening

S/N	Plant extracts	Stem bark	Leaves
1	Alkaloids	+	+
2	Flavonoids	+	_
3	Phenols	+	+
4	Terpenoid	_	+
5	Tannins	+	_
6	Saponins	+	+
7	Quinones	+	+
8	Resins	_	_

Key: *+ = present, - = absent*

Tables 2 and 3 show the results of the minimal inhibitory concentrations of methanol leaf and stem bark extracts of *Faidherbia albida*. The results showed that the plant's stem bark and leaf extract could suppress the growth of practically all test bacteria. The stem bark extract inhibited *Escherichia coli, Staphylococcus aureus,* and *Klebsiella pneumoniae* more effectively than the leave extract, whereas the leave extracts inhibited *Pseudomonas* more than the stem bark extract. The suppression of *Salmonella typhii* by both extracts is same. The results revealed the presence of antibacterial components in both plant extracts (leaf and stem bark), indicating a strong correlation with the documented usage of these plants in traditional medicine against infectious disorders, as described by Kubmarawa *et al.* (2007). The reason behind this strong inhibition as observed in this study may not be far from the secondary metabolites present in this plant.

Table 2: Zone of inhibition of the test organisms by the leaf extract of in (mm)

Organisms	Concentration				Positive Control AUG/30ug
	500 ug/ml	250 ug/ml	125 ug/ml	6.25 ug/ml	
Escherichia coli	11	9	8	7	30
Staphylococcus aureus	8	8	7	6	23
Salmonella typhii	6	6	6	6	16
Klebsiella pneumoniae	9	6	6	6	13
Pseudomonas	16	13	9	8	6

Table 3: Zone of inhibition of the test organisms by the stem bark extract in (mm)

Organisms	Concentration				Positive Control AUG/30ug
	500 ug/ml	250 ug/ml	125 ug/ml	6.25 ug/ml	
Escherichia coli	13	8	8	7	16
Staphylococcus aureus	10	8	7	7	27
Salmonella typhii	6	6	6	6	30
Klebsiella pneumoniae	13	11	10	9	22
Pseudomonas	9	7	6	6	19

The presence of secondary metabolites in these extracts results in effective suppression of the targeted microorganisms as shown in tables 2 and 3. Also, Doughari and Saa-Aondo (2021) revealed that the presence of these phytochemicals in the extracts may have been responsible for the inhibitory effect. According to Christopher *et al.* (2021), due to the antioxidant effects of phytochemicals (saponins, tannins, alkaloids, phenols, flavonoids) and free radical scavenging properties that is present in many medicinal plants made them to exert their inhibition effect (medicinal effects) as seeing in this studies. This plant has immense potential and have broad spectrum of activity on ailments as earlier indicated.

Table 4: FT-IR spectral peak values and functional groups obtained for the stem bark extract

Peak value (cm^{-1})	Functional group	Type of vibration	Characteristic absorptions (cm^{-1})	Intensity
3294.9	O-H	Stretch, (H-bonded)	3200-3600	Strong, broad
2930.9	O-H	Stretch	2500-3300	strong, very broad
1613.4	N-H	Bending	1550-1640	
1521.1	N-O	Stretch	1515-1560 1345-1385	strong, two bands
1444.0	C=C	Stretch	1400-1600	medium-weak, multiple bands
1369.5	-C-H	Bending	1350-1480	Variable
1274.4	C-O	Stretch	1210-1320	Strong
1104.7	C-O	Stretch	1000-1300	two bands or more
807.7	=C-H	Bending	675-1000	Strong
627.2	C-Cl	Stretch	600-800	Strong

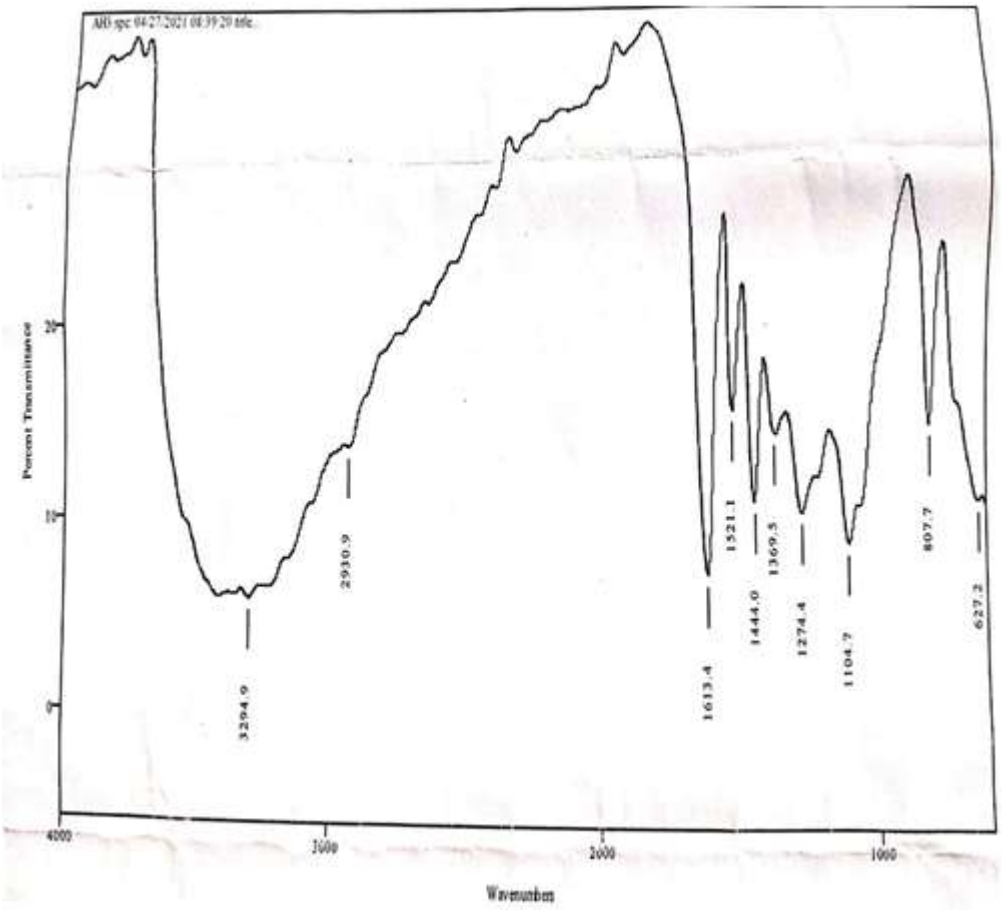

Figure 1: FT-IR Spectrum of methanol stem bark extracts of *Faidherbia albida*

The FT-IR analysis of methanol stem bark extracts, presented in table 4, demonstrated the presence of hydroxyl, amino, and nitro groups as the main functional groups in the extract. The findings of this study suggest that the stem bark of Faidherbia albida, along with its phytoconstituents, could serve as an antibiotic source. Carbohydrates, carotene, amino acids, amides, starch, and cellulose are all likely present in the stem bark extracts. The OH group has the ability to create hydrogen bonds; the presence of an OH group, particularly in a methanol extract of stem bark, indicates that the extract has a higher potential for inhibitory effect against microorganisms.

Figure 1 depicts the FT-IR spectrum of *Faidherbia Albida's* methanol stem bark extract, which shows significant peaks at 3325.84 cm^{-1}, 2926.98 cm^{-1}, 1638.67 cm^{-1}, and 1051.60 cm^{-1}. These peaks represented the stretching vibrations of -OH, -CH, C=O, and C-O groups, respectively. These functional groups are often found in a variety of secondary metabolite classes, including phenolics, flavonoids, and tannins, all of which have bioactive features such as antioxidant, anti-inflammatory, and antibacterial activity (Maitera and Chukkol, 2016). The presence of these functional groups in *Faidherbia Albida's* methanol stem bark extract indicates that it includes a

varied range of secondary metabolites with potential bioactive characteristics. This is consistent with prior research that found numerous bioactive components in *Faidherbia Albida* extracts, including as tannins, flavonoids, and alkaloids (Maitera and Chukkol, 2016).

Broad absorption bands in the range of 3325 cm^{-1} indicate strong O-H stretching vibrations, suggesting the presence of hydroxyl groups essential for hydrogen bonding. This observation is consistent with the findings of Rajathy *et al*. (2021) in *Telescopium telescopium*. The presence of numerous hydroxyl groups contributes to its reactivity and solubility (Wang *et al.*, 2020). N-O and N-H stretching vibrations in the extract of *Faidherbia Albida* have great medicinal effect. According to Gündüz *et al.* (2020), nitro compounds are notable for their wide spectrum of activities which include antineoplastic, antibiotic, anti-parasitic, antihypertensive and tranquilizer. Also, these amine groups are important for the bioactivity of the polymer, including antimicrobial and biocompatible characteristics (Pham *et al*., 2021).

*Conclusion***:** These findings support the traditional use of *Faidherbia Albida* in the treatment of various infectious diseases and suggest that the plant could be a potential source of natural antimicrobial agents. These therefore establish the ethno-medicinal claims on the plant.

Declaration of Conflict of Interest: The authors declare no conflict of interest.

Data Availability Statement: Data are available upon request from the first author.

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