Extracts of *Trichilia heudelotii* (Meliaceae) Planch, a Nigerian medicinal plant have antibacterial and antifungal activity

Abstract

There is no overemphasizing the need for novel phytotherapeutic agents to combat the menace of drug resistance in microbial pathogens. Many plant species so far, have yielded some bioactive compounds with great promise for such drugs. *Trichilia heudelotii* (Meliaceae) is commonly used in traditional medicine in Nigeria for the treatment of many microbial infections ranging from gastrointestinal infections to gonorrhea. This study is thus designed to determine the phytochemicals present in *T. heudeloti* and evaluate the plant's extracts' antimicrobial activity on some bacterial and fungal isolates.

The phytochemical screening was done using standard procedures. Soxhlet extracts using n-hexane and methanol were drying *in vacuo*. The methanol extract was partitioned into petroleum ether, chloroform, and aqueous fractions. The antibacterial and antifungal activity of the extracts determined using the agar-well diffusion method. The MICs were determined for the extracts using the agar dilution method.

The qualitative phytochemical screening revealed the presence of tannins, saponins, alkaloids, cardenolides, and anthraquinones in the leaf, stem bark, and root bark of *T. heudelotii.* The extracts showed considerable activity against Gram-positive and Gram-negative organisms, and fungi with dermatophytes including *Klebsiella* spp, *Escherichia coli*, *Proteus* spp, *Pseudomonas aeruginosa, Staphylococcus aureus, Bacillus subtilis, Neisseria gonorrhoeae, Mycobacterium smegmatis, Candida albicans, Aspergillus niger, Rhizopus stolonifer, Penicillium* spp, *Microsporium canis, and Trichophyton mentagrophytes.* The mean diameter of zones of inhibition exhibited by the extracts ranged between 10mm$\pm$ 0.5 and 32$\pm$ 0.33mm. The methanol extracts compared favourably with the gentamycin (standard control). The minimum inhibitory concentration (MIC) ranged between 0.157mg/mL and 20mg/mL. The

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crude methanol extracts and methanol residue showed the highest activity of all the extracts while the hexane extract showed the lowest activity and the Petroleum ether fraction was inactive.

These results showed the potential of *T. heudelotii* as a possible candidate for bioactive compounds for the discovery and development of new drugs for the treatment of diseases caused by test pathogens.

Keywords*:* Antimicrobial activity; *Trichilia heudelotii*; Phytochemical Screening; Drug-discovery

Introduction

Many indigenous plants have been used by the common man since time immemorial for curing various ailments and thus lessening human suffering, without the actual knowledge of the active ingredient that causes such relief. Some plants, which are of vital importance, have been fully exploited and their uses in the pharmaceutical industry are well known. But a great majority of them remain yet untouched (Khameneh, *et al*., 2019).

In recent times, researchers have been turned towards the search for antimicrobial agents from higher plants since plant compounds such as berberine, emetine, quinine, and sanguinarine which have found specialized uses, have been identified (Bribi, 2018). Secondary metabolites from higher plants also serve as defense agents against invading micro-organisms (Faehnrich *et al*., 2021)

Herbal remedies play a fundamental role in traditional medicine in rural areas of Africa, particularly Nigeria, where they often constitute the therapeutic treatment of choice for the people. The extensive use of traditional medicine in Africa, composed mainly of medicinal plants, has been argued to be linked to several reasons, some of which are cultural and economic. As such, the WHO encourages African countries to promote and integrate traditional medical practices into their healthcare systems (Mahomoodally, 2013). Currently, it`s reported that the global market value of medicinal plant products is in the neighbourhood of 100 Billion USD per annum (Sofowora *et al* 2013).

Trichilia heudelotii (Meliaceae), a tree about 12-20m high which is common in most countries in West Africa, including Nigeria, is a plant that has shown great potential as a possible source of bioactive agents for phytotherapy (da Silva *et al.*, 2021). The bark and leaves of *Trichilia heudelotii* have several

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medicinal applications, in decoction form, pulp, or as dry powder. Ethno-pharmacologically, they are used in the treatment of wounds, cuts and sores, gastro-intestinal pains and disorders, gonorrhea, and epidermal infections (Irvine 1961; Benjamin *et al.,* 2018; Okpalanwaka *et al*., 2020).

Fig 1: The leaf of *Trichilia heudelotii* (Meliaceae) Planch (Source: Picture by Adeniyi Bolanle A. UIH -23365).

Previous studies reported the wound-healing ability, antioxidant, and some other pharmacological potential of *T. Heudelotii* (Bankole *et al*, 2016 and Benjamin *et al*, 2018). However, there is a pulsity of information on the antibacterial (particularly antimycobacterial) and antifungal properties of *T. Heudelotii* available. This study therefore reports the antimicrobial activities of *T. heudelotii* against clinically important bacterial (including Mycobacterial) and fungal pathogens.

Materials and methods

Plant source and authentication

Trichilia heudelotii Planch, ex Oliver (Meliaceae) leaves stem bark and root bark were collected from Ijeoma road, Ebrohime, University of Ibadan, Ibadan, Nigeria. The authentication was done at University of Ibadan Herbarium, Ibadan as UI23365.

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Phytochemical evaluation of plant samples

The powdered plant parts were screened for the presence of secondary metabolites such as alkaloids, tannins, anthraquinones and so on using standard chemical tests and procedures (Adeniyi *et al* 1996, Adeniyi, B. A. and Anyiam, F. M. (2004), and Shrestha *et al*, 2015).

Preparation of plant extracts

About 500 g of powdered plant parts were extracted successively, by Soxhlet method using the two solvents *n*-Hexane and methanol for about 12 hrs in each cycle. The extracts were then concentrated to dryness *in vacuo*, using Rotary evaporator, then weighed and stored in clean dry containers till needed.

Strains of organisms used in the study

The following strains were used in this study: *Bacillus subtilis* PHM 1502, *Bacillus subtilis* NPD 3042, *Bacillus subtilis* UCH 2032, *Staphylococcus aureus* PHM 1501, *Staphylococcus aureus* ATCC 13709, Methicillin Resistant *Staphylococcus aureus* (MRSA) UCH 2031, *Pseudomonas aeruginosa* UCH 2033, *Pseudomonas aeruginosa* ATCC 27853, *Pseudomonas aeruginosa* PHM 1503, *Escherichia coli* ATCC 25922, *Escherichia coli* NPD 3041, *Escherichia coli* UCH 2041, *Escherichia coli* UCH 2042, *Klebsiella* spp UCH 2046, *Klebsiella* spp UCH 2049, *Klebsiella* spp UCH 2040, *Klebsiella* spp UCH 2047, *Neisseria gonorrhoeae* UCHSTC 2021, *Neisseria gonorrhoeae* UCHSTC 2024, *Proteus* spp UCH 2034, *Proteus* spp UCH 2035, *Mycobacterium smegmatis* ATCC 3043, *Candida albicans* UCHSTC 2036, *Candida albicans* UCH STC 2037, *Aspergillus niger* UCH 2038, *Penicillium* spp PHM 5101, *Rhizopus stolomtes* PHM 5102, *Trichophyton mentagrophytes* ATCC 4808 and *Microsporum canis.* ATCC 11622.

Fresh overnight culture of each of the organisms was used in this study.

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Antimicrobial susceptibility assay

The agar cup diffusion bioassay method (Adeniyi *et al*, 1995; Saleem *et al*., 2015) was used in this study. A volume of 20 ml of molten nutrient agar was cooled to 45-50 ^{o}C and poured into sterile Petri dishes previously inoculated with 0.2 ml of 1×10^{-2} dilution of overnight culture of test organisms. However tryptic soy agar and Sabourand dextrose agar was used for *Mycobacterium smegmatis* and the fungal isolates respectively. The content of the Petri dish was gently mixed and allowed to set before being placed in a drying oven to remove moisture from the agar surface. Equidistant wells of 8 mm were bored into the agar using a sterile cork-borer. The wells were filled with 60 µl of the reconstituted dried hexane and methanol extracts in the required concentrations (12 and 6 mg/mL). All the test samples were reconstituted in 50% v/v aqueous methanol which was used as a negative control while gentamycin and griseofulvin were used as positive controls. The plates were left at room temperature for 45 min and then incubated for 24 hrs at 37 ^{o}C for bacteria strains or 48 hrs at 25 ^{o}C (fungal strains). *Mycobacterium smegmatis* was incubated for 48 hrs at 37 ^{o}C. The zones of inhibition were measured after the incubation periods.

Determination of Minimum inhibitory concentrations (MIC): The MIC were determined for the extracts using agar dilution method utilizing several concentration of plant extracts.

Partitioning of the Methanol Extracts

The methanol extracts of leaves, root-bark, and stem-bark of *Trichilia heudelotii* were partitioned using petroleum ether, chloroform and distilled water**.** About 50 g of each extract was dissolved in 200 ml of distilled water, (1:4) and was transferred into a separating funnel. Aliquots of petroleum ether were poured into the funnel, shaken vigorously and then allowed to stand and partition. The aqueous portion was tapped off into a clean dry container leaving the petroleum ether extract and this was also dispensed into another container. The process was repeated for chloroform, and the aqueous portion left was the methanol residue. All the extracts were allowed to evaporate to dryness and weight yield calculated.

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Microbiological Screening of the Fractions

The various fractions obtained from the methanol extracts of *Trichilia heudelotii* were screened against the bacterial and fungal isolates at a concentration of 6 mg/mL. Microbiological screening was carried out as described earlier.

Thin Layer Chromatography (TLC)

Thin layer chromatography of the crude methanol extracts of leaf, stem-bark, root-bark *Trichilia heudelotii* was carried out using a chromo plate of plastic coated with a thin uniform layer of silica gel which is the stationary phase. The plate was then spotted with a small amount of the solution of the different extracts and then placed in a solvent system of hexane and ethyl acetate (ratio 1:4 as the mobile phase) in a closed eluting tank. The extracts moved by capillarity across the plates at different rates depending on their solubility and so became separated (Klimek-Turek *et al*., 2016). The Retention factor (Rf) values were calculated for each component. The same experiment was repeated using methanol, ethyl acetate and hexane in ratio 1:2:4. as the mobile phase

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Results

The phytochemical screening revealed the absence of anthraquinones in the leaf and root bark but was present in the stem bark of *Trichilia heudelotii* (Table 1). The leaves, stem bark, root bark were also found to contain alkaloids, cardenolides, saponins and tannins. Table 2 shows the results of the antimicrobial screening of the crude methanol and hexane extracts of leaf, stem bark and root bark of *Trichilia heudelotii* at concentrations of 12 and 6 mg/mL.

The results of the antimicrobial screening of the partitioned methanol extracts of *Trichilia heudelotii* showed that the methanol residue (highest zone of inhibition = 25 mm) of all the extract partitions was generally more active than the other fractions (Table 3). The pet ether fractions were all inactive while the chloroform fraction (highest zone of inhibition = 19 mm) showed less activity.

Bacillus subtilis (*Bs*) was susceptible to all extracts of *Trichilia heudelotii.* The root bark extract of *Trichilia heudelotii* had an MIC of 0.157 mg/mL against *Bs* PHM 1502 with the methanol residue showing an MIC of 0.625 mg/mL (Table. 4).

Table 1: Results of the phytochemical screening of *Trichilia heudelotii*

Phytochemicals	**Plant parts**		
	Leaves	**Stem-bark**	**Root-bark**
Alkaloids	++	++	++
Cardenolides	++	++	++
Anthraquinones	--	++	--
Saponins	++	++	++
Tannins	++	++	++

Key

++ Present -- Absent

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Table 2: Antimicrobial activity of methanol and hexane extracts of *Trichilia heudelotii*

Organism	Methanol extracts						Hexane extracts						Controls		
	Leaf (mg/mL)		Stem bark (mg/mL)		Root bark (mg/mL)		Leaf (mg/mL)		Stem bark (mg/mL)		Root bark (mg/mL)		Gent.	Gris.	MeOH
	12	6	12	6	12	6	12	6	12	6	12	6	5µg/mL	50 µg/mL	50%v/v
	Diameter of zone of inhibition in mm*														
Bs PHM 1502	15 ± 0.1	13 ± 0.1	19 ± 0.2	17 ± 0.3	25 ± 0.1	23 ± 0.3	15 ± 0.1	12 ± 0.2	13 ± 0.2	10 ± 0.2	10 ± 0.2	R	20 ± 0.3	NT	R
Bs NPD 3042	12 ± 0.3	11± 0.1	18 ± 0.1	15 ± 0.2	18 ± 0.2	15 ± 0.2	10 ± 0.1	R	12 ± 0.1	R	11 ± 0.1	R	17 ± 0.2	NT	R
Bs UCH 2032	13 ± 0.2	10 ± 0.3	22 ± 0.3	20 ± 0.3	21 ± 0.3	15 ± 0.1	13 ± 0.3	10 ± 0.2	R	R	R	R	20 ± 0.1	NT	R
Sa PHM 1501	12 ± 0.2	10 ± 0.2	15 ± 0.2	14 ± 0.3	15 ± 0.2	13 ± 0.3	15 ± 0.2	13 ± 0.2	13 ± 0.3	R	R	R	20 ± 0.3	NT	R
Sa ATCC 13709	15 ± 0.3	13 ± 0.3	15 ± 0.1	13 ± 0.1	15 ± 0.1	14 ± 0.2	13 ± 0.3	11 ± 0.3	14 ± 0.2	R	R	R	24 ± 0.2	NT	R
Sa UCH 2031	12 ± 0.2	11 ± 0.1	13 ± 0.2	11 ± 0.2	17 ± 0.3	15 ± 0.3	12 ± 0.1	10 ± 0.1	R	R	R	R	15 ± 0.1	NT	R
Ps UCH 2033	11 ± 0.1	10 ± 0.1	14 ± 0.3	13 ± 0.3	14 ± 0.2	13 ± 0.2	15 ± 0.3	13 ± 0.2	R	R	R	R	R	NT	R
Ps ATCC 27853	14 ± 0.3	13 ± 0.3	13 ± 0.1	11 ± 0.1	15 ± 0.2	13 ± 0.3	13 ± 0.1	R	R	R	R	R	R	NT	R
Ps PHM 1503	13 ± 0.2	10 ± 0.1	11 ± 0.1	10 ± 0.1	13 ± 0.1	10 ± 0.1	R	R	R	R	R	R	R	NT	R
Ec ATCC 25922	15 ± 0.3	13 ± 0.1	14 ± 0.3	12 ± 0.2	15 ± 0.3	13 ± 0.2	13 ± 0.2	11 ± 0.1	12 ± 0.1	11 ± 0.1	10 ± 0.2	R	R	NT	R
Ec NPD 3041	R	R	19 ± 0.2	17 ± 0.3	17 ± 0.3	15 ± 0.1	12 ± 0.3	10 ± 0.2	R	R	R	R	16 ± 0.3	NT	R
Ec UCH 2041	R	R	R	R	R	R	15 ± 0.3	11 ± 0.1	13 ± 0.1	R	15 ± 0.1	12 ± 0.2	R	NT	R
Ec UCH 2042	R	R	R	R	R	R	13 ± 0.1	12 ± 0.1	12 ± 0.3	11 ± 0.2	11 ± 0.3	10 ± 0.1	17 ± 0.1	NT	R
Kb UCH 2046	12 ± 0.1	10 ± 0.1	R	R	R	R	R	R	R	R	R	R	R	NT	R
Kb UCH 2049	12 ± 0.2	11 ± 0.1	R	R	14 ± 0.3	12 ± 0.2	R	R	R	R	R	R	R	NT	R
Kb UCH 2040	R	R	R	R	R	R	R	R	R	R	R	R	R	NT	R
Kb UCH 2047	R	R	R	R	R	R	R	R	R	R	R	R	R	NT	R
Ng UCH STC 2021	14 ± 0.2	12 ± 0.2	20 ± 0.2	18 ± 0.3	22 ± 0.2	20 ± 0.2	17 ± 0.2	15 ± 0.3	R	R	10 ± 0.3	R	20 ± 0.3	NT	R
Ng UCH STC 2024	R	R	16 ± 0.3	14 ± 0.3	R	R	R	R	R	R	12 ± 0.2	R	18 ± 0.4	NT	R
Pr UCH 2034	13 ± 0.1	12 ± 0.2	15 ± 0.2	13 ± 0.3	18 ± 0.2	16 ± 0.3	15 ± 0.1	13 ± 0.3	12 ± 0.1	R	11 ± 0.1	R	10 ± 0.2	NT	R
Pr UCH 2035	14 ± 0.3	12 ± 0.2	15 ± 0.3	14 ± 0.3	16 ± 0.1	14 ± 0.1	12 ± 0.1	10 ± 0.2	11 ± 0.2	10 ± 0.2	R	R	17 ± 0.1	NT	R
Ms ATCC 3043	17 ± 0.1	16 ± 0.3	17 ± 0.2	17 ± 0.1	32 ± 0.3	29 ± 0.3	R	R	R	R	R	R	R	NT	R
Ca UCH STC 2036	15 ± 0.2	13 ± 0.2	13 ± 0.3	11 ± 0.3	12 ± 0.1	10 ± 0.1	R	R	R	R	11 ± 0.1	R	NT	R	R
Ca UCH STC 2037	R	R	12 ± 0.1	10 ± 0.1	12 ± 0.3	10 ± 0.1	R	R	R	R	13 ± 0.2	R	NT	R	R
An UCH 2038	20 ± 0.2	14 ± 0.2	15 ± 0.2	12 ± 0.3	17 ± 0.3	11 ± 0.1	13 ± 0.2	11 ± 0.1	R	R	14 ± 0.1	R	NT	R	R
Pn BMC 5101	R	R	R	R	12 ± 0.2	10 ± 0.3	10 ± 0.1	R	R	R	R	R	NT	R	R
Rz BMC 5102	R	R	R	R	R	R	R	R	R	R	R	R	NT	R	R
Tm ATCC 4808	R	R	16 ± 0.3	14 ± 0.1	R	R	R	R	R	R	R	R	NT	R	R
Mc ATCC 11622	20 ± 0.3	15 ± 0.3	15 ± 0.3	12 ± 0.1	15 ± 0.3	13 ± 0.1	R	R	R	R	R	R	NT	R	R

Diameter of cork borer: 8mm; * Result is average of triplicate experiment

KEY

Ba - Bacillus subtilis, Ps - Pseudomonas aeruginosa, Ec - Escherichia coli, Kb - Klebsiella spp, Pr - Proteus spp, Ng - Neisseria gonorrhoeae, .
Sa - Staphylococcus aureus, Ms - Mycobacterium smegmatis, Ca - Candida albicans, An - Aspergillus niger, Pn – Penicillium spp,
Rz - Rhizopus stolomtes, Tm - Trychophyton mentagrophytes, Mc - Microsporum canis,
R- Resistant, MeOH – Methanol, Gent. - Gentamycin, Gris – Griseofulvin. NT- Not Tested; PHM: Pharmaceutical Microbiology; NPD: Nigeria Pharmaceutical Research Institute Development; UCHSTC: University College Hospital Sexually Transmitted Disease Clinic. ATCC: American Type Culture Collection.

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Table 3: Antimicrobial activity of partitioned methanol extracts of *Trichilia heudelotii*

Organism	Leaf (6 mg/mL)			Stem bark (6 mg/mL)			Root bark (6 mg/mL)		
	Pet ether	Chloroform	Methanol residue	Pet ether	Chloroform	Methanol residue	Pet ether	Chloroform	Methanol residue
Diameter of zone of inhibition in mm*									
Bs PHM 1502	R	10 ± 0.1	12 ± 0.2	R	13 ± 0.3	16 ± 0.2	R	13 ± 0.1	19 ± 0.2
Bs NPD 3042	R	13 ± 0.2	14 ± 0.3	R	12 ± 0.1	15 ± 0.1	R	15 ± 0.3	20 ± 0.1
Bs UCH 2032	R	R	10 ± 0.1	R	12 ± 0.2	17 ± 0.3	R	16 ± 0.1	19 ± 0.3
Sa PHM 1501	R	12 ± 0.1	14 ± 0.1	R	R	12 ± 0.1	R	R	12 ± 0.1
Sa ATCC 13709	R	14 ± 0.3	16 ± 0.3	R	R	13 ± 0.2	R	12 ± 0.1	12 ± 0.2
Sa UCH 2031	R	12 ± 0.1	12 ± 0.2	R	12 ± 0.3	14 ± 0.3	R	R	13 ± 0.3
Ps UCH 2033	R	12 ± 0.2	12 ± 0.1	R	R	12 ± 0.1	R	R	11 ± 0.1
Ps ATCC 27833	R	13 ± 0.3	14 ± 0.3	NT	NT	NT	NT	NT	NT
Ps PHM 1503	R	R	12 ± 0.1	R	R	10 ± 0.1	NT	NT	NT
Ec ATCC 25922	R	R	12 ± 0.2	R	10 ± 0.1	13 ± 0.3	R	13 ± 0.1	16 ± 0.2
Ec NPD 3041	NT	NT	NT	R	R	12 ± 0.1	R	10 ± 0.2	12 ± 0.1
Kb UCH 2046	R	13 ± 0.3	15 ± 0.3	NT	NT	NT	NT	NT	NT
Kb UCH 2049	R	12 ± 0.2	14 ± 0.1	NT	NT	NT	R	R	12 ± 0.1
Ng UCHSTC 2021	R	10 ± 0.1	13 ± 0.2	R	13 ± 0.3	20 ± 0.3	R	13 ± 0.3	25 ± 0.3
Ng UCHSTC 2024	NT	NT	NT	R	12 ± 0.2	16 ± 0.2	NT	NT	NT
Pr UCH 2034	R	R	15 ± 0.3	R	10 ± 0.1	14 ± 0.1	R	12 ± 0.1	14 ± 0.2
Pr UCH 2035	R	13 ± 0.1	17 ± 0.2	R	13 ± 0.3	15 ± 0.2	R	10 ± 0.1	12 ± 0.1
Ms ATCC 3043	R	14 ± 0.3	19 ± 0.3	R	12 ± 0.1	19 ± 0.3	R	19 ± 0.2	23 ± 0.3
Ca UCH STC 2036	R	12 ± 0.1	15 ± 0.1	R	10 ± 0.1	13 ± 0.2	R	10 ± 0.1	12 ± 0.1
Ca UCH STC 2037	R	R	R	R	12 ± 0.2	13 ± 0.3	R	14 ± 0.3	15 ± 0.3
An UCH 2038	R	13 ± 0.1	15 ± 0.1	R	R	12 ± 0.1	R	R	12 ± 0.2
Pn BMC 5101	R	R	R	R	R	R	R	10 ± 0.1	13 ± 0.3
Rz BMC 5102	R	R	R	R	R	R	R	R	R
Tm ATCC 4808	R	R	R	R	12 ± 0.3	16 ± 0.2	R	R	R
Mc ATCC 11622	R	14 ± 0.2	16 ± 0.1	R	R	12 ± 0.1	R	R	14 ± 0.2

Diameter of cork borer: 8mm

* Result is average of triplicate experiment

KEY

Ba - Bacillus subtilis, Ps - Pseudomonas aeruginosa, Ec - Escherichia coli, Kb - Klebsiella spp, Pr - Proteus spp, Ng - Neisseria gonorrhoeae . Sa -Staphylococcus aureus, Ms - Mycobacterium smegmatis. Ca - Candida albicans, An - Aspergillus niger, Pn – Penicillium spp, *Rz - Rhizopus stolomtes, , Tm - Trychophyton mentagrophytes, Mc - Microsporum canis* R- Resistant, NT- Not Tested

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Table .4: Minimum Inhibitory Concentration (MIC) of the extracts of *Trichilia heudelotii (*mg/mL)

	Leaf (mg/mL)				Stem Bark (mg/mL)				Root Bark (mg/mL)			
Organism	**Methanol**	**Chloroform**	**Methanol Residue**	**Hexane**	**Methanol**	**Chloroform**	**Methanol Residue**	**Hexane**	**Methanol**	**Chloroform**	**Methanol Residue**	**Hexane**
Bs PHM 1502	2.5	5	2.5	10	1.25	2.5	0.625	100	0.157	0.625	0.625	20
Bs NPD 3042	5	10	5	20	0.625	5	1.25	200	0.313	0.625	0.157	20
Bs UCH 2032	10	NT	10	10	0.313	5	0.625	NT	0.625	0.625	0.313	NT
Sa PHM 1501	10	5	5	2.5	2.5	NT	5	NT	5	NT	5	NT
Sa ATCC 13709	1.25	1.25	0.625	10	1.25	NT	2.5	NT	5	1.25	1.25	NT
Sa UCH 2031	10	5	2.5	10	0.625	2.5	0.313	NT	0.625	NT	1.25	NT
Ps UCH 2033	10	10	5	10	10	NT	5	NT	2.5	NT	10	NT
Ps PHM 1503	5	NT	5	20	10	NT	10	NT	5	NT	2.5	NT
Ps ATCC 27833	2.5	2.5	1.25	NT	5	R	5	NT	2.5	NT	1.25	NT
Ec ATCC 25922	5	NT	5	5	5	10	5	100	5	2.5	1.25	20
Ec NPD 3041	NT	NT	NT	10	10	NT	5	NT	1.25	10	1.25	NT
Ec UCH 2041	NT	NT	NT	5	NT	NT	NT	200	NT	NT	NT	2.5
Ec UCH 2042	NT	NT	NT	2.5	NT	NT	NT	50	NT	NT	NT	10
Kb UCH 2046	5	5	0.625	NT	NT	NT	NT	NT	NT	NT	NT	NT
Kb UCH 2049	5	10	0.125	NT	NT	NT	NT	NT	5	NT	5	NT
Ng UCHSTC 2021	5	10	2.5	1.25	0.625	1.25	0.313	NT	0.313	2.5	0.157	20
Ng UCHSTC 2024	NT	NT	NT	NT	0.625	1.25	0.313	NT	NT	NT	NT	20
Pr UCH 2034	2.5	NT	0.625	2.5	0.313	2.5	0.313	20	0.625	2.5	0.625	20
Pr UCH 2035	2.5	5	0.625	10	0.625	5	0.625	10	0.313	10	2.5	NT
Ms ATCC 3043	0.625	2.5	0.313	NT	1.25	5	0.313	NT	0.157	1.25	0.313	NT
Ca UCHSTC 2036	5	10	50	NT	10	10	10	NT	10	10	10	20
Ca UCHSTC 2037	NT	NT	NT	20	10	10	5	NT	10	5	2.5	NT
An UCH 2038	2.5	10	10	5	5	NT	10	NT	10	NT	10	20
Pn BMC 5101	NT	NT	NT	20	NT	NT	NT	NT	10	10	10	NT
Tm ATCC 4808	NT	NT	NT	NT	5	10	5	NT	NT	NT	NT	NT
Mc ATCC 11622	1.25	5	2.5	NT	10	NT	10	NT	5	NT	2.5	NT

KEY

Bs Bacillus subtilis, Ps - Pseudomonas aeruginosa, Ec - Escherichia coli, Kb - Klebsiella spp, Pr - Proteus spp, Ng - Neisseria gonorrhoeae, Sa - Staphylococcus aureus, Ms - Mycobacterium smegmatis. Ca - Candida albicans, An - Aspergillus niger, Pn – Penicillium spp, *Rz - Rhizopus stolomtes, Tm - Trychophyton mentagrophytes, Mc – Microsporum canis.* NT – Not Tested

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Discussion

The phytochemical analysis and antimicrobial screening produced results which are consistent with traditional uses of the plants. It revealed secondary metabolites with various biological activities. Alkaloids were significantly present in *T. heudelotii.* They have been found to be active against *Giardia* and *Entamoeba* the common causes of diarrhea, Ghoshal *et al.*, (1996); and against *Plasmodium berghei* Bankole *et al*., (2016); *Trypanosama brucei*, Okpalanwaka *et al*., (2020). This probably justifies the claim that the bark decoction of *T. heudelotii* is used to cure dysentery and diarrhoea (Benjamin *et al.,* 2018).

While other phytochemicals tested for were present in abundance, anthraquinones was found to be absent only in the stem-bark of *Trichilia heudelotii.* This tends to deviate from the findings of Benjamin *et al.,* (2018), who reported the presence of little to moderate amount of anthraquinones in the leaves and root-bark of *Trichilia heudelotii.* The point of deviation could be as a result of the difference in the solvent used in the extraction or partitioning of the extracts and the seasonal variation. The leaves, stem bark, root bark were also found to contain alkaloids, cardenolides, saponins and tannins. Bankole *et al.*, (2016), Benjamin *et al.,* 2018, reported similar findings in their studies.

With the minimum inhibitory concentration (MIC) ranging between 0.157 mg/mL (*Mycobacterium smegmatis* ATCC 3043) and 20 mg/mL (*Bacillus subtilis* NPD 3042), the extracts of the various parts of *Trichilia heudelotii*, displayed a broad spectrum of activity. *Mycobacterium smegmatis* was not considered a human pathogen until Vonmoos *et al* described a pleuropulmonary infection in 1986. Since then, infections due to *M. smegmatis* have been reported, which include pneumonia, bacteremia, and arthroplasty infections (Adeniyi, 2013). The methanol and methanol residue fractions of the various parts of the plant proved the most active, while the hexane fractions the least. While this study reports MIC range of 0.625 - 10 mg/mL for *S. aureus* ATCC 13709, Benjamin *et al.,* (2018) reported 5 to 10 mg/mL for *S. aureus* ATCC25923. While these results appeared close, strain variation amongst other factors could account for the little differences. This trend was observed in other common pathogens investigated in our study.

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Conclusion

The investigation has justified the folkloric claims of *Trichilia heudelotii* in treating infections such as diarrhea, urinary tract infections, sexually transmitted infections, skin diseases, and other microbial infections, as this study has been able to show that the plant has both antibacterial and antifungal activities. The activity may be due to the presence of secondary metabolites such as triterpenoids, tannins, saponins, anthraquinones, cardenolides, and alkaloids. Further studies are required to isolate identify and characterize these compounds, which could serve as a template for the formulation of new antimicrobial drugs.

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