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Phytochemical profiling, and antibacterial evaluation of ethanol and ethyl acetate extracts of *Nauclea latifolia* leaves, stems, and roots

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Abstract
Background: Traditional medicines remain central to primary health care in West Africa. *Nauclea latifolia* is widely used ethnomedicinally and contains diverse secondary metabolites with reported bioactivities (Ayeleso *et al*., 2014; Enabulele *et al*., 2017; Haudecoeur *et al*., 2017) [3, 7, 9].
Objective: To quantify powdered and crude extract yields; qualitatively profile phytochemicals; establish TLC fingerprints; and evaluate in-vitro antibacterial activity of ethanol and ethyl acetate extracts from leaves, stems, and roots of the plant.
Methods: Plant parts were dried, powdered, and Soxhlet-extracted with 95% ethanol or ethyl acetate. Qualitative phytochemical screening followed standard pharmacognosy methods (Sofowora, 1993; Harborne, 1998; Evans, 2009) [14, 10, 8]. TLC used chloroform:methanol (9:1) and hexane:chloroform (9:1) with Rf determination. Antibacterial activity against *Staphylococcus aureus*, *Klebsiella pneumoniae*, and *Escherichia coli* employed agar well diffusion on Mueller-Hinton agar; ciprofloxacin (5 µg) and DMSO were positive/negative controls.
Results: Powdered yields: leaves 750 g, stems 520 g, roots 780 g. Total crude extract: 35.08 g (overall 2.33%). Ethanol extracts were richer in alkaloids, flavonoids, saponins, tannins, glycosides, triterpenes, resins, and phenolics. TLC showed 2-4 spots per extract (Rf 0.14-0.75). Antibacterial zones (mm) ranged 4.5-13.0 for extracts vs 14.5-18.0 for ciprofloxacin; DMSO 0.0.
Conclusion: Findings demonstrate solvent- and part-dependent phytochemical diversity and antibacterial activity, with ethanol extracts generally outperforming ethyl acetate. Results support ethnomedicinal applications and justify MIC/MBC determination and bioassay-guided isolation (Akinyemi *et al*., 2005; Akinmoladun *et al*., 2007; Edeoga *et al*., 2005; Osuala *et al*., 2020) [2, 1, 5, 12].

Keywords: *Nauclea latifolia*, TLC fingerprint, phytochemical screening, ethanol extract, ethyl acetate extract, agar well diffusion, antibacterial activity, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Escherichia coli*

Introduction
Medicinal plants contribute key leads for anti-infective discovery. *Nauclea latifolia* (Rubiaceae) is used for fever, diarrhea, malaria and gastrointestinal disorders with leaves, roots and stems commonly employed (Ayeleso *et al*., 2014; Enabulele *et al*., 2017; Oko, 2013) [3, 7, 11]. Documented constituents include alkaloids, saponins, tannins, glycosides, triterpenes and phenolics (Tukur *et al*., 2011; Edem, 2021) [3, 15, 6]. Given local utilization in Sierra Leone and variability across regions, this study evaluated part- and solvent-dependent yields, phytochemicals, TLC fingerprinting, and antibacterial activity against clinically relevant bacteria.

Materials and Methods
Study Design and Setting
Experimental laboratory study performed at the COMAHS-USL Pharmaceutical Sciences Laboratory and the PBSL Microbiology Laboratory (Freetown, Sierra Leone).

Plant Material and Authentication
Mature leaves, stems, and roots of *Nauclea latifolia* were collected under WHO guidance; authentication/voucher references recorded (FWTA ed. 2,1:235; UPWTA ed. 1,69).

Preparation of Powder and Soxhlet Extraction
Plant parts were air-dried, pounded, sieved (mesh 80→70) and weighed. For each solvent and part, 250 g powder was Soxhlet-extracted (~6 h) with 95% ethanol or ethyl acetate, then concentrated under reduced pressure to obtain crude extracts.

Phytochemical Screening
Qualitative assays followed standard protocols (Sofowora, 1993; Harborne, 1998; Evans, 2009) [14, 10, 8]: carbohydrates (Molisch/Fehling/Benedict), glycosides, alkaloids (Mayer/Dragendorff/Wagner), saponins, tannins (FeCl3), flavonoids, steroids/triterpenes, resins, phlobatanins, phenolics.

Thin Layer Chromatography (TLC)
Silica plates were developed in chloroform: methanol (9:1) and hexane: chloroform (9:1); visualization at 256/366 nm; Rf = distance compound / distance solvent front.

Antibacterial Assay
Agar well diffusion on Mueller-Hinton agar against *Staphylococcus aureus*, *Klebsiella pneumoniae*, and *Escherichia coli*. Inocula standardized to 0.5 McFarland; wells 7 mm; incubation 24 h at 37 °C. Ciprofloxacin (5 µg) served as positive control; DMSO negative.

Results

Table 1: Powdered Plant Parts of *Nauclea latifolia*

Plant Part	Weight of Bottle (g)	Weight of Bottle + Sample (g)	Powdered Weight (g)
Leaves	250	1000	750
Stems	240	760	520
Roots	260	1040	780

Table 2: Crude Extract Yield by Plant Part and Solvent

Extract Type	Crude Extract Weight (g)	Percentage Yield (%)
Leaf (Ethanol)	7.20	0.48
Leaf (Ethyl Acetate)	6.85	0.46
Stem (Ethanol)	5.60	0.37
Stem (Ethyl Acetate)	4.25	0.28
Root (Ethanol)	6.80	0.45
Root (Ethyl Acetate)	4.38	0.29
Total	35.08	2.33

Table 3: Phytochemical Screening of *Nauclea latifolia*

Phytochemicals	Leaf (Ethanol)	Leaf (Ethyl Acetate)	Stem (Ethanol)	Stem (Ethyl Acetate)	Root (Ethanol)	Root (Ethyl Acetate)
Carbohydrate (Molisch, Fehling, Benedict)	++	+++	+++	+	+++	+++
Glycosides	+++	+++	++	++	++	+++
Alkaloids	+++	++	+++	–	+++	+
Flavonoids	+++	–	+	–	++	–
Saponins	+++	+++	+++	+	+++	+
Tannins	+++	+++	+	+	+++	+
Triterpenes	+++	+++	+	–	+++	++
Resins	++	++	+	–	+++	+
Phlobatanins	+	+	+	+	+	+

Table 4: TLC Results for *Nauclea latifolia*

Plant Part	Solvent System	Extract Type	Rf Values (Spots Detected)	Number of Spots
Leaf	Chloroform:Methanol (9:1)	Ethanol	0.18, 0.31, 0.47, 0.68	4
Leaf	Hexane:Chloroform (9:1)	Ethyl Acetate	0.22, 0.37, 0.59	3
Stem	Chloroform:Methanol (9:1)	Ethanol	0.14, 0.29, 0.52	3
Stem	Hexane:Chloroform (9:1)	Ethyl Acetate	0.20, 0.35	2
Root	Chloroform:Methanol (9:1)	Ethanol	0.16, 0.40, 0.60, 0.75	4
Root	Hexane:Chloroform (9:1)	Ethyl Acetate	0.19, 0.44, 0.71	3

Table 5: Zone of Inhibition of *Nauclea latifolia*

Isolate	Positive Control Ciprofloxacin (5 µg)	Negative Control DMSO	Leaf (Ethanol)	Leaf (Ethyl Acetate)	Stem (Ethanol)	Stem (Ethyl Acetate)	Root (Ethanol)	Root (Ethyl Acetate)
Staphylococcus aureus	17.5	0.0	13.0	10.0	8.5	6.0	9.5	7.5
Klebsiella pneumoniae	18.0	0.0	10.5	8.0	7.0	5.0	8.0	6.0
Escherichia coli	14.5	0.0	9.0	6.5	5.5	4.5	6.0	5.0

Discussion
Ethanol extracts displayed higher crude yields and broader phytochemical presence, paralleling stronger antibacterial zones particularly against *Staphylococcus aureus* than ethyl acetate extracts. This aligns with reports that protic solvents extract phenolic/flavonoid constituents implicated in

antimicrobial effects (Edeoga *et al*., 2005; Akinmoladun *et al*., 2007; Ríos and Recio, 2005) [5, 1, 13]. TLC profiles (2-4 spots; Rf 0.14-0.75) indicate chemical diversity across parts/solvents, consistent with prior medicinal-plant analyses (Chukwujekwu *et al*., 2006) [4]. Methodological strengths include standard pharmacognosy screens and compendial diffusion assays; limitations include qualitative phytochemistry, absence of MIC/MBC, and no compound isolation.

Future work should quantify key classes (e.g., total phenolics/flavonoids), establish MIC/MBC, perform bioassay-guided fractionation, and assess toxicity and stability to prepare for translational development.

Conclusion

Across leaves, stems, and roots of *Nauclea latifolia*, ethanol extracts generally outperformed ethyl acetate in yield, phytochemical richness, and antibacterial activity against *Staphylococcus aureus*, *Klebsiella pneumoniae*, and *Escherichia coli*. These data substantiate ethnomedicinal use and provide a reproducible baseline (yields, TLC, zones) for replication and scale-up studies.

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