



PHYTOCHEMICAL SCREENING AND ANTIMICROBIAL ACTIVITY OF *STERCULIA SETIGERA* LEAVES

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ABSTRACT

The plant *Sterculia setigera* (family: *Sterculiaceae*) is known by different indigenous cultural communities in Nigeria. It is a savannah tree, widespread in savannah areas of tropical Africa. The aim of this research is to carry out the phytochemical screening and antimicrobial activity of the leaf of *sterculia setigera*. The leaves of *sterculia setigera* was extracted with 70% methanol. The Phytochemical screening of the extract showed the presence of alkaloids, cardiac glycosides, saponins, phtobatannins, steroids, starch, resins. The Agar well diffusion method was used for the antimicrobial activity. The crude extract showed visible inhibitory effect when compared to the positive control. The crude extract of the leaves of *sterculia setigera* has zone of inhibitions of; 26 mm, 18 mm, 12 mm, 10 mm, for *Staphylococcus aureus* at 400 mg/ml, 200 mg/ml, 100 mg/ml, 50 mg/ml concentrations, 20 mm, 16 mm, 12 mm, 08mm, for *Pseudomonas aeruginosa* at 400 mg/ml, 200 mg/ml, 100 mg/ml, 50 mg/ml concentrations, 22 mm, 18.5 mm, 14.2 mm, 7.5 mm, for *Escherichia coli* at 400 mg/ml, 200 mg/ml, 100 mg/ml, 50 mg/ml concentrations and 23 mm, 20 mm, 16 mm, 10 mm, for *Candida albican* at 400 mg/ml, 200 mg/ml, 100 mg/ml, 50 mg/ml concentrations respectively. The mean zone of inhibition showed that *sterculia setigera* leaves exhibited activities against the test organisms. It was in this study that *E. coli* and *S. aureus* was more susceptible to the plant extracts compared to *P. aeruginosa*. The present study therefore offers a scientific basis for the traditional use of plant *Sterculia setigera* for the treatment of *Mycobacterium tuberculosis*, malaria, jaundice, measles, syphilis and leprosy. The evaluation of phytochemical and antibacterial screening of *Sterculia setigera* concluded that methanolic extract contain saponins, phtobatannins, resins, cardiac glycosides, alkaloids, steroids, starch. These constituents could be responsible for the inhibition of the test organism. The present study therefore offers a scientific justification for the traditional use of plant *Sterculia setigera* for the treatment of *Mycobacterium tuberculosis*, malaria, jaundice, measles, syphilis and leprosy. Further studies are necessary in order to clarify the properties of *Sterculia setigera* leaves to obtain information enough to provide validation for its medicinal use.

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INTRODUCTION

The plant, *Sterculia setigera* (family: *Sterculiaceae*) is known by different indigenous cultural communities in Nigeria: Hausa-“Kukuki”, Fulani-“bo'boli”, Yoruba-“Ose-awere”, Nupe- “Kokongiga”, Idoma- “Ompla, Tiv- “Kume-ndul” [1].

It is a savannah tree, widespread in savannah areas of tropical Africa. The seeds are with yellow aril and the tree is found in open savannah wood-lands, often characterized by stony hills [2].

This plant is used in traditional medicine by various indigenous communities. For instance, the Yourba people of Nigeria use a black soap prepared from black powder obtained from burnt mixture of the fruits and seeds in dermatosis. In Sudan, dried bark water extract is used for jaundice and dried stem bark for treating wounds. Stem bark decoction is used to treat diarrhea [2] by the Igedes, its bark as a mixture is macerated and used against dysentery by some tribes in central Nigeria [3].

The study on Anti-TB Activity of *Sterculia setigera* del., leaves (*Sterculiaceae*) Folkloric claims on antibacterial activity of *S.*

setigera were investigated *in-vitro* on a micro-scale using the Alamar Blue Assay. Three of four successive solvent extractions of the plant leaves extracts inhibited the growth of a virulent strain of *Mycobacterium tuberculosis*, H37Rv (ATCC27294) in the concentrations tested (1-128 µg/ml). The minimum inhibitory concentration (MIC) determined for the hexane, dichloromethane and ethyl acetate extracts were 84 µg/ml, 62 µg/ml and 128 µg/ml respectively [4].

Sample collection and preparation

Fresh samples of the plant material (leaves of *Sterculia setigera*) were collected at Sabon kaura Road behind Abubakar Tafawa Balewa University Bauchi, Nigeria. The plant was identified at the botany laboratory in the Department of Biological Sciences, ATBU Bauchi. The freshly collected leaves of *Sterculia setigera* were spread to air dry at room temperature for two weeks. After drying, the leaves were pounded with the laboratory mortar and pestle and were stored in a clean container for further analysis.

Extraction of the leaf extracts

The sample (30g) was weighed and was subjected to maceration extraction method with 30% water and 70% methanol for 48 hours. The extracts were filtered and the filtrates were evaporated for the qualitative analysis.

Antibacterial Analysis

The test organisms used in this study include *Escherichia coli*, *Staphylococcus aureus*, *Candida albicans* and *Pseudomonas aeruginosa*. The plates were incubated at 37 °C for 24 hours. After incubation the plates were checked for pure confluent growth and the diameter of the zones of inhibition of growth were measured to the nearest millimeter ruler [5].

The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) was done according to the method described by [6].

Phytochemical Screening of the Extracts:

Test for Saponins

Frothing Foam Test: The method described by [7] 1 ml solution extract was diluted with distilled water to 20 ml and shaken. If foam produced persists for ten to fifteen minutes it indicates the presence of saponins.

Test for starch (Fehling's test)

Iodine crystals (0.1 g) and 0.1 g of potassium iodide will be weighed using analytical weighing balance and 5 ml of distilled water will be measured with the aid of 10 ml measuring cylinder, the iodine crystals and potassium iodide will be dissolved into distilled water and 2 ml of the crude extract will be added. The formation of blue-black color, indicates the presence of starch [8].

Test for steroids

The extract was dissolved in 2ml of chloroform and 1 ml of acetic anhydride solution, followed by the two drops of concentrated sulfuric acid was added along the side of the test tube and left standing. The formation of pink color, indicates the presence of steroid [7].

Test for alkaloids

The presence of alkaloids in the crude extract was detected using Mayer's and Wagner's test as described by [9].

Hydrochloric acid 5 ml of 2 M was added to 0.5 g of the crude extract and warmed in boiling water bath, the solution will be filtered and will be distinctly separated into two (2) test tubes, and used for the below test:

Mayer's Reagent:

The test solution was mixed with little amount of Mayer's reagent and 1 ml of the filtrate. Formation of yellow precipitate indicates the presence of alkaloids.

Wagner's Reagent:

The test solution was mixed with little amount of Wagner's reagent and 1 ml of the filtrate. Formation of reddish-brown precipitate indicates the presence of alkaloids.

Test for cardiac glycosides

Keller-Killian test [10] was employed for the detection of Cardiac Glycoside which to the solution of the extract in 2 ml glacial acetic acid, few drops of ferric chloride and 1 ml concentrated sulfuric acid was gently pour through the wall of the test tube and observed for a brown ring colouration at the junction of two layers and a bluish green color at the upper layer. This is an indication of positive result.

Test for phtobatannins

Crude extract 0.5 ml was transferred into test tube, the boiled in 1% Hydrochloric acid, using water bath. The formation of red precipitate indicates the presence of phtobatannins [11].

Test for resin

To the solution of the extract, shake thoroughly. The formation of turbidity indicates the presence of resins [8].

RESULTS AND DISCUSSION

The leaves of *Sterculia setigera* was extracted with 70% methanol using the maceration extraction method for 48 hours. The crude extract was obtained by evaporating the solvent to dryness. Table 1 below shows the physical properties and % yield of the crude extract.

Table 1: Results for the Crude Extraction of Sterculia setigera leaves

Extract weight	% yield	solvent used	plant part	color
Crude extract 1. 1 g	3.66 %	70% methanol	leaves	green

Weight of sample: 30 g

Residue: 23.0 g

Key: (g) = gram

% = percentage

Table 2: Phytochemical Constituents of Sterculia setigera Leaves

S /N	Secondary Metabolites	Test/Reagent	Result
1.	Alkaloid	Mayer's Wagner's	+ +
2.	Saponins	Distilled water	+
3.	Steroid	Chloroform	+
4.	Cardiac Glycoside	Glacial acetic acid	+
5.	Resin	Distilled water	+
6.	Starch	Felhing's solution	+
7.	phtobatannins	Hydrochloric acid	+

Key: Present = (+), Precipitate = ppt

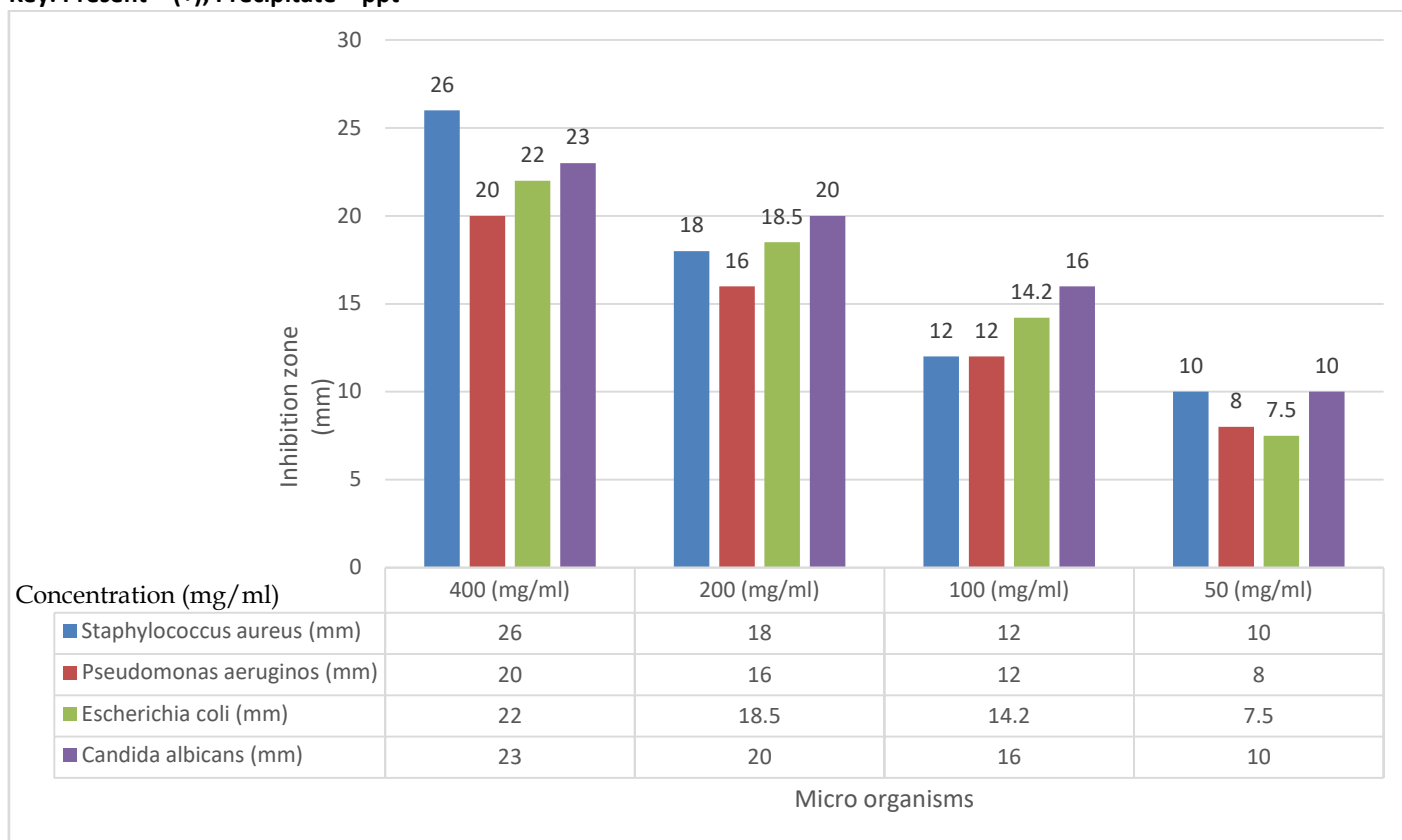


Figure 1: Chart Representing Mean Zone of Inhibitions of Crude Extract Sterculia setigera Leaves Against Different Organisms.

Discussion

Extraction and phytochemical screening of bioactive agents from medicinal plant permits demonstration of their physiological ac-

tivities. Table 1 shows that the plant extract to be green in color and 1.1 g crude extract was recovered from the 30 g of the sample used for the extraction.

The result of the phytochemical analysis of *sterculia setigera* leaf showed that, saponins, phtobatanins, resin, starch, steroids, cardiac glycoside, alkaloids are all present in the leaf of the plants (Table 2). The observed phytochemicals in the leaves extracts corresponds to the report of [12]. [5] reported the presents of phtobatanins and alkaloids in the methanol extracts of the stem bark of *sterculia setigera*. The medicinal activity of the plant maybe related to its chemical constituents for example phytobatanins are plant secondary metabolites well known for their antimicrobial properties and can aid in wound healing and burns [13]. Similarly, [14] revealed the presence of some secondary metabolites but alkaloids and flavonoids were not detected in the report. Also, about 80% of the western pharmaceuticals have their origin in plants [15]. Flavonoids have important dietary significant because being phenolic compound they are strongly antioxidant and this probably explains why Africans eat the young leaf of *Sterculia setigera* as vegetable. Since, they are known to be synthesized by plant and responsible for microbial inhibition in curing ailment and also therapeutic agent for relieving pain.

Figure 2, shows the antibacterial activity of crude extract of leaves of *Sterculia setigera* with zone of inhibitions of 26 mm, 18 mm, 12 mm, 10 mm on *Staphylococcus aureus* at 400 mg/ml, 200 mg/ml, 100 mg/ml, 50 mg/ml concentration and 20 mm, 16 mm, 12 mm, 08 mm on *Pseudomonas aeruginosa* at 400 mg/ml, 200 mg/ml, 100 mg/ml, 50 mg/ml concentration while 22 mm, 18.5 mm, 14.2 mm, 7.5 mm on *Escherichia coli* at 400 mg/ml, 200 mg/ml, 100 mg/ml, 50 mg/ml concentration and 23 mm, 20 mm, 16 mm, 10 mm on *Candida albicans* at 400 mg/ml, 200 mg/ml, 100 mg/ml, 50 mg/ml concentration. The extract of the plant has shown wide range of activity against various microorganisms. The standard antibiotic Chloramphenicol (250 mg/ml) was used as positive control for the bacteria and Ketoconazole (200 mg/ml) was used for the fungus, while Dimethyl sulfoxide (DMSO) was used as a negative control of both bacteria and fungus. Based on the results obtained, *Sterculia setigera* leaves have demonstrated varying degree of antibacterial activity against *Escherichia coli*, *Pseudomonas aeruginosa*, *Saphylococcus aerues* and *Candida albican* (fungi). Therefore, this signifies that some bacteria that have not been tested with *sterculia setigera* leaves extract in this research may also be susceptible to the antibacterial effect of *sterculia setigera* leaves.

The minimum inhibitory concentration (MIC) to Minimum bactericidal and fungicidal concentration (MBC and MFC) results of *sterculia setigera* leaf extract is 100 mg/ml to 400 mg/ml (for *Escherichia coli*), 200 mg/ml to 400 mg/ml (for *Staphylococcus aureus* and *Pseudomonas aeruginosa*) and 400 mg/ml to above (for *Candida albican*) respectively.

These were higher than that reported by [16] about the ethanolic extract of *Sterculia setigera* leaf. But was Similar to [17] report on the antifungal effect of the bioactive fractions of the leaf of *S. setigera*.

Conclusion

The crude extract of the leaves of *sterculia setigera* were screened for phytochemical and anti-microbial activity. The result of phytochemical revealed that various secondary metabolites present in the leaves of *Sterculia setigera* were saponins, phtobatanins, resins, cardiac glycosides, alkaloids, steroids and starch. *Sterculia setigera* leaves has demonstrated antibacterial properties which could be used for the development of alternative means of therapeutic control of clinical pathogens.

Therefore, there is need for further research to be carried out to determine the toxicity/safety level of the plant extracts, administration as well as to isolate and identify the active compound responsible for the activity.

References

- [1] Igoli JO, Ogaji OG, Tor-Anyiin TA, Igoli NP (2005). Traditional medicine practice amongst the Igede people of Nigeria. Part II. *Afr J. Traditional Complement Altern Med.*, **2**(2):134–152.
- [2] Agishi EC (2004). Etulo, Idoma, Igede, Tiv and Hausa names of plants. AGITAB Pub. Ltd., Makurdi, Nigeria. p. 188.
- [3] Igoli JO, Tor-Anyiin TA, Usman SS, Oluma HOA, Igoli NP (2002). Folk Medicine of the lower Benue Valley of Nigeria. Recent progress in medicinal plants **7**, 327-338.
- [4] Ibrahim TB, Adelakun EA, Wang Y, Shode OF (2012). Anti-TB Activity of *Sterculia setigera* Del., Leaves (Sterculiaceae). *J Pharmacog Phyto.*, **1**(3):17-2.
- [5] Tor-Anyiin TA, Akpuaka MU, Oluma HOA (2011). Phytochemical and antimicrobial studies on stem bark extract of *Sterculia setigera*, Del. *African Journal of Biotechnology*, **10**(53):11011-11015.
- [6] Akinyemi KO, Oladapo O, Okwara CE, Ibe CC, Fasure KA (2005). Screening of Crude Extracts of Six Medicinal Plants Used in South–West Nigeria unorthodox medicine anti-methicillin resistant *Staphylococcus aureus* activity. BMC Complement. *Alternative Medicine*, **5**(6): 1-7.
- [7] Devmurari VP (2010). Phytochemical screening study and antibacterial evaluation of *Symplocos racemose* Roxb. *Arch Appl Sci Res.* **2**: 354-9.

- [8] Bigyan Sharma, Sapana Thapa, Gan B Bajracharya (2017). Promising anti-oxidative potentiality and antibacterial activity of *Mallo-tus philippensis* grown in Nepal. *Journal of Pharmacognosy and Phytochemistry*. **6**(3), 629-632.
- [9] Goyal Sh (2013). Ecological role of alkaloids. *Natural products: phytochemistry, botany and metabolism of alkaloids, phenolics and terpenes*. 149-171.
- [10] Sofowora, A. (2008). *Medicinal plants and traditional medicine in Africa*. 3rd edition. Sprectrum Book Limited, Ibadan, pp 201.
- [11] Edeoga HO, Okwu DE, Mbaebie BO (2005). Phytochemical constituents of some Nigeria Medicinal plants. *Afri. J. Biotechnol.* **4**:658-688.
- [12] Hamidu AA (2012). Phytochemical constituents of the leaves of *Sterculia setigera*. *J. Pharm.*, **2**(1):062-064.
- [13] Mohan SC, Sasikala K, Anad T (2014). Antimicrobial and Wound Healing Potential of *Canthium coromandelicum* Leaf Extract – A Preliminary Study. *Research Journal of Phytochemistry*, **8**(2):35-41.
- [14] Babalola I.T. and Adelakun E.A (2013) Isolation of stigmast-5-en-3 β -ol (β -sitosterol) from dichloromethane extract of *Sterculia setigera* Leaves (Sterculiaceae). *Archives of Applied Science Research*, **5**(5):16-19
- [15] Cuellar CA, Okori O, Dennis B (2010). Preliminary phytochemical and antimicrobial evaluation of fresh and dried whole plant extract from *Commelina benghalensis*. *Revision Colmbia Ciencena Animation*, **2**(1): 104 116: 1-7.
- [16] Luois, H., Linus, M. N., Israt, A., Innocent, J. Amos, P. I. and Magu, T.O. (2018). Antimicrobial Activity of Stem, Leave and Root Plant Extract of *Sclerocarya birrea* and *Streculia setigera* against some selected Micro Organism. *An International Scientific Journal of World Scientific News*. WSN **92**(2): 309-326.
- [17] Ouedraogo KK, Zebro P, Barro N, Sawadogo LL (2013). Phytochemical Analysis and *in vitro* Antifungal Profile of Bioactive Fractions from *Sterculia setigera* (Sterculiaceae). *Current Research Journal of Biological Sciences*, **5**(2):75-80.

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References

- [1] J.S. Bridle, "Probabilistic Interpretation of Feedforward Classification Network Outputs, with Relationships to Statistical Pattern Recognition," *Neurocomputing – Algorithms, Architectures and Applications*, F. Fogelman-Soulie and J. Hérault, eds., NATO ASI Series F68, Berlin: Springer-Verlag, pp. 227-236, 1989. (Book style with paper title and editor)
- [2] W.-K. Chen, *Linear Networks and Systems*. Belmont, Calif.: Wadsworth, pp. 123-135, 1993. (Book style)
- [3] H. Poor, "A Hypertext History of Multiuser Dimensions," *MUD History*, <http://www.ccs.neu.edu/home/pb/mud-history.html>. 1986. (URL link *include year)
- [4] K. Elissa, "An Overview of Decision Theory," unpublished. (Unpublished manuscript)
- [5] R. Nicole, "The Last Word on Decision Theory," *J. Computer Vision*, submitted for publication. (Pending publication)
- [6] C. J. Kaufman, Rocky Mountain Research Laboratories, Boulder, Colo., personal communication, 1992. (Personal communication)
- [7] D.S. Coming and O.G. Staadt, "Velocity-Aligned Discrete Oriented Polytopes for Dynamic Collision Detection," *IEEE Trans. Visualization and Computer Graphics*, vol. 14, no. 1, pp. 1-12, Jan/Feb 2008, doi:10.1109/TVCG.2007.70405. (IEEE Transactions)
- [8] S.P. Bingulac, "On the Compatibility of Adaptive Controllers," *Proc. Fourth Ann. Allerton Conf. Circuits and Systems Theory*, pp. 8-16, 1994. (Conference proceedings)
- [9] H. Goto, Y. Hasegawa, and M. Tanaka, "Efficient Scheduling Focusing on the Duality of MPL Representation," *Proc. IEEE Symp. Computational Intelligence in Scheduling (SCIS '07)*, pp. 57-64, Apr. 2007, doi:10.1109/SCIS.2007.367670. (Conference proceedings)
- [10] J. Williams, "Narrow-Band Analyzer," PhD dissertation, Dept. of Electrical Eng., Harvard Univ., Cambridge, Mass., 1993. (Thesis or dissertation)
- [11] E.E. Reber, R.L. Michell, and C.J. Carter, "Oxygen Absorption in the Earth's Atmosphere," Technical Report TR-0200 (420-46)-3, Aerospace Corp., Los Angeles, Calif., Nov. 1988. (Technical report with report number)
- [12] L. Hubert and P. Arabie, "Comparing Partitions," *J. Classification*, vol. 2, no. 4, pp. 193-218, Apr. 1985. (Journal or magazine citation)
- [13] R.J. Vidmar, "On the Use of Atmospheric Plasmas as Electromagnetic Reflectors," *IEEE Trans. Plasma Science*, vol. 21, no. 3, pp. 876-880, available at <http://www.halcyon.com/pub/journals/21ps03-vidmar>, Aug. 1992. (URL for Transaction, journal, or magazine)
- [14] J.M.P. Martinez, R.B. Llavori, M.J.A. Cabo, and T.B. Pedersen, "Integrating Data Warehouses with Web Data: A Survey," *IEEE Trans. Knowledge and Data Eng.*, preprint, 21 Dec. 2007, doi:10.1109/TKDE.2007.190746. (PrePrint)