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Phytochemical studies and thin layer chromatography of leaves and flower extracts of *Senna siamea lam* for possible biomedical applications

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Senna siamea **is a medium-size, evergreen plant which has been utilized as a source of food, medicine and other agricultural purposes in different communities. However, there is dearth of information in regard to its possible biomedicinal uses, especially in Nigeria. Thus the preliminary phytochemical analysis and thin layer chromatography (TLC) separation was done using methanol, n-hexane and ethyl acetate (1:3:1) as solvent system while iodine vapour as spotting agent. The phytochemical screening of methanol extracts of leaves revealed the presence of cardiac glycoside, flavonoid, saponin, alkaloid and tannins while chloroform extracts of leaves revealed saponin only. Ethyl acetate and petroleum ether extracts revealed absence of all these phytochemicals. The chloroform, ethyl acetate and petroleum ether extracts of flower revealed absence of saponin, flavonoids, tannins and alkaloids but with traces of saponin and anthraquinones. TLC separation showed nine (9) spots each of chloroform and ethyl acetates, six (6) spots of methanol, three (3) spots of petroleum ether from leaves extracts. While, three (3) spots each of ethyl acetate and methanol, six (6) spots of chloroform were identified for flower extracts. No water spot separated from both leaves and flower extracts. From our findings, it can be concluded that *S. siamea lam* contains some significant phytochemicals that can exhibit desired therapeutic activities such as hypoglycemia, anti-arrthymia and antimicrobial. However, there is the need to conduct further pharmaceutical analyses on test extracts in order to establish these biomedical applications.**

Key words: *Senna siemea,* thin layer chromatography, antimicrobial, phytochemical.

INTRODUCTION

Plants have been found to be the source of energy for the animal kingdom. In addition, plant can synthesize a large variety of chemical substances that are of physiological significance (Kretovich, 2005). The active phytochemical principles produced by plants include, alkaloids, phenolic, anthraquinones, flavonoids, phenols, saponins, steroid, tannins, terpenes etc (Namukobea et al., 2011).

Medicinal plants are those that contains one or more of its phytochemicals that can be used for the synthesis of useful therapeutic agents (Sofowora, 2000). The wide

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range of medicinal plant parts like flowers, leaves, barks, stems, fruits, roots extracts are used as powerful raw drug possessing a variety of pharmacological activities (Momin et al., 2012). In the last two centuries, there have been serious investigations into the chemical and biological activities of plants and these have yielded compounds for the development of synthetic organic chemistry and the emergence of medicinal chemistry as a route for the discovery of more effective therapeutic agents (Roja and Rao, 2000).

Senna siamea is native to Southeast Asia from India, Sri Lanka, and Thailand to Indonesia, Burma, and Malaysia and forms part of the warm and wet tropical forests. The species has been introduced in Africa and America. *S. siamea* is effective in managing constipation association with a number of causes including surgery, childbirth and the use of narcotic pain relievers (Hill, 1992). It is used locally as antimalarial drugs especially when decocted (the leaves and bark) (Lose et al., 2000). In traditional medicine, the fruit is used to charm away intestinal worms and to prevent convulsion in children. The young fruits and leaves are also eaten as vegetables in Thailand. The flowers and young fruits are used as curries (Kiepe, 2001) and as an antimalarial (Otimenyin et al., 2010). The stem bark extract was reported to have analgesic and anti-inflammatory effects (Ntandu et al., 2010). Isolated compounds, emodu and lupeol from the ethyl acetate fraction of the stem bark of *S. siamea* were reported to be the active principles responsible for the antiplasmodial property with IC_{50} values of 5 µg/ml, respectively (Ajaiyeoba et al., 2008). Sub-chronic studies of the aqueous stem bark extract of the plant in rats did not show significant toxic effect after seven weeks of administration (Mohammed et al., 2012)

This study was designed to determine the phytochemical compositions as well as to perform thin layer chromatography separation of the leaves and flowers extracts of *S. siamea* in order to create awareness of its possible medicinal and nutritional values.

MATERIALS AND METHODS

These include the test plant (the fresh leaf and flower of *S. siamea*), beaker, conical flask, measuring cylinder (large and small), glass funnel, glass stirrer, cotton wool, spatula, bunsen burner, top mettler weighing balance, test tubes, stainless steel tray, thermostat water bath, oven, syringe and needle, aluminum foil paper, hand gloves, mortar and pestle, analytical weighing balance, test-tube holder, refrigerator, meter rule, sieves (No. 5), bottles, UV fluorescence analysis cabinet tripod stand, wire gauze, capillary tubes, retort stand, thin layer chromatography (TLC) paper, TLC tank, test tube rack, tiles and filter paper.

Reagent used

Dragendoff's reagent, methanol, chloroform, 1% aqueous hydrochloric acid, Mayer's reagent, sodium chloride solution, glacial acetic acid, concentrated sulphuric acid, 10% Ferric chloride solution, Molisch's reagent, Fehling's solution A and B, lead sub-acetate solution, 10% sodium hydroxide, 10% ferric chloride in 95% alcohol, Barfoed's reagent, 3,5 dinitro benzoic acid I, iodine solution, dilute hydrochloric acid, Wagner's reagent, concentrated hydrochloric acid, 3.2% ferric chloride in glacial acetic acid, 10% lead acetate, 10% tannic acid, 1% w/v picric acid, 5% sodium hydroxide, bromine water, potassium iodide solution, 3% hydrogen peroxide, 1 M sodium hydroxide , acetic anhydride.

Sterilization

All work surfaces were comprehensively disinfected with cotton wool soaked in antiseptic fluid to minimize contamination during work process.

Dry heat sterilization

An hot air oven was used to sterilize the conical flasks, forceps, office punch, wire loop and filter paper discs (wrapped in foil paper) and beaker at 160°C for 45 min.

Moist heat sterilization

All materials used in the course of this research project that are not sensitive to moist heat sterilization were adequately sterilized using autoclave and detergents. Materials such as glass wares, beakers and conical flasks etc. were properly washed with detergent and water so as to remove dirt and contaminants and were allowed to dry prior to usage. These materials were then sterilized in a portable laboratory autoclave at 121°C for 15 min.

Collection, authentication and processing of plant materials

The fresh leaves and flowers of *S. siamea* were collected from the botanical garden of University of Maiduguri. Plant materials were identified and authenticated by a taxonomist, Professor S. S. Sanusi of the Department of Biological Sciences, University of Maiduguri, Nigeria in respect with the description in published literatures (Dalziel, 1958; Keay et al., 1989). The plant materials were dried under shade at our Pharmaceutical Chemistry Laboratory for about four weeks and then made into powdered form, using mortar and pestle and then sieved.

Extraction

The method of extraction in this experiment was by maceration. The general process on a small scale, consist of placing the powdered plant material (250 g) of leave was soaked in 500 ml methanol while that of flower was soaked with different solvents that is water, petroleum ether, methanol, chloroform and ethyl acetate (in order of decreasing polarity) in 1 L capacity conical flasks stopper and kept for 48 h with intermittent shaking. The cold extracts thus obtained were filtered with Whatman No. 1 filter paper into different conical flask and allowed to dry at room temperature under normal atmospheric pressure. 50 g of the powdered leaves were soaked in 100 ml distilled water and the extract was obtained using the aforementioned method.

Phytochemical analysis

Phytochemical analysis for the qualitative detection of alkaloids, anthraquinone, carbohydrates, flavonoids, tannins and saponins

Table 1. Leaves extraction results.

Parameter	Methanol	Chloroform	Ethyl acetate
Volume of solvent used (ml)	500	500	500
Weight of dried powdered (g)	200	200	200
Weight of solvent extract (g)	53.5	4.3	5
Extractive value (%)	26.8	2.15	2

Table 2. Leaves methanol extract partitioned

Parameter	Methanol/Water	Chloroform	Ethyl acetate	Petroleum ether
Volume of solvent used for partitioning (ml)	150	150	150	150
Weight of partitioned solvent extract (g)	47.6	18.8	9.9	20.8

was carried out on the extracts as described by Trease and Evans (2010), Sofowora (1993) and Harbone (1973).

Thin layer chromatography (TLC)

Commercially available standard TLC plate was used with standard particle size range to improve reproducibility. The absorbent silica gel coated on an aluminum foil of 22 cm length, 11.5 cm breadth and 0.3 cm thick plate for leaves while 22 cm length, 11.9 cm breadth and 0.3 cm thick plate for flower. Small spot of the solution containing the sample was applied on the plate 1.5 cm from the bottom marked by a line ruled using a pin. For a multiple spotted plate, the spots are applied 1 cm apart to avoid cross contamination and interference as they move up the plate.

Spotting and development

The sample spotted on the plate was allowed to dry before the plate was placed into the chromatographic tank which was covered immediately after which its atmosphere is completely saturated with solvent (mobile phase). The reaction was then monitored as the solvent moved up the plate (elutes the sample) using mobile phase solvent ratio 1:3:1 of methanol, n-hexane and ethyl acetate, respectively. When the solvent reaches the top of the plate, it is removed, marked and dried.

Visualization

Following separation of the solvent, the plate was removed and dried; the spots detected using various techniques and reagents. This includes visualization in daylight; viewing under UV at 254 and 366 nm i.e. short and long wavelengths and spraying with spotting reagent, using iodine vapor tank.

Findings

The phytochemical screening of methanol extracts of leaves revealed the presence of cardiac glycoside, flavonoid, saponin, alkaloid and tannins while chloroform extracts of leaves revealed saponin only. Ethyl acetate and petroleum ether extracts revealed absence of all these phytochemicals. The chloroform, ethyl acetate and petroleum ether extracts of flower revealed absence of saponin, flavonoids, tannins and alkaloids but with traces of saponin. Anthraquinones glycosides was absent in all the extracts.

Extraction process for leaves and flowers

This is seen in Tables 1 and 2.

Extractive value = weight of plant (part) extract/weight of dry powdered sample × 100

Volume of methanol used = 1 L.
Weight of dried powdered = 300 g.
Weight of methanol extract = 101 g.
Extractive value = 101/300 × 100 = 33.7%

Phytochemical screening results

This is seen in Tables 3 to 9.

Thin layer chromatography (TLC)

Extracts of leaves and flowers were individually applied on the origin, they dissolved and moved with the solvent, each extract separated into bio constituents and moved to different locations. After all the spots became clear. UV fluorescence lamp at 254 nm was used to visualize and identify all the various spots. However, at 366 nm and daylight, spots were not clearly visualized. On exposure to iodine vapour, spots of various extracts became darker. TLC separation showed nine (9) spots each of chloroform and ethyl acetates, six (6) spots of methanol, three (3) spots of petroleum ether from leaves extracts. While, three (3) spots each of ethyl acetate and methanol, six (6) spots of chloroform were identified for flower extracts. No water spot separated from both leaves and flower extracts.

Summary of TLC results

Leaves TLC

Length of the plate = 22 cm.
Breath of the plate =11.5 cm.
Thickness of the plate = 0.1 cm.

Table 3. Test for carbohydrate.

Tests	Extracts	Solvents used				
		Water	Methanol	Chloroform	Ethyl acetate	Petroleum ether
Molisches test (for carbohydrate)	leaves	++	++	-	++	+
	flowers	++	++	+	+	
Iodine test (for starch)	leaves	-	+	++	+	-
	flowers	-	+	++	+	
Fehling's test (for reducing sugar)	leaves	-	++	+	+	+
	flower	-	++	-	++	
Combined reducing sugar test	leaves	-	+	++	++	+
	flowers	+	++	-	+	
Barfoed test (for monosaccharides)	leaves	-	-	-	-	-
	flowers	-	-	-	-	-

++ More abundance, + Abundance, - Absence

Table 4. Tests for tannins (hydrolysable and condensed).

Tests	Extracts	Solvents used				
		Water	Methanol	Chloroform	Ethyl acetate	Petroleum ether
Lead sub- acetate test	Leaves	+	++	-	++	-
	Flowers	+	++	-	-	-
Ferric chloride test	Leaves	++	+	-	+	-
	Flowers	++	+	-	+	-
Bromine water test	Leaves	-	-	-	-	-
	Flower	-	-	-	-	-

++ More abundance, + Abundance, - Absence

Solvent front of the plate = 18.3 cm.
R_f value = distance move by the solute ÷ distance move by the solvent.

Tables 10 to 13 shows the TLC results of leaves.

Flowers TLC

Length of the plate = 22 cm.
Breath of the plate = 11.9 cm.
Thickness of the plate = 0.1 cm.
Solvent front of the plate = 17.9 cm.
R_f value = distance move by the solute ÷ distance move by the solvent.

Tables 14 to 16 shows TLC results of flowers.

DISCUSSION

S. siamea plants grow virtually everywhere in Nigeria and Maiduguri in particular. The plant has been used in this region for the treatment of typhoid fever and fever related conditions. Traditionally, it has also been used for treatment of jaundice, abdominal pain, menstrual pain, and hypoglycemic agent among diabetics. Ethno medicinally, *S. siamea* is used as laxative, blood cleaning agent, cure for digestive system and genitourinary

Table 5. Tests for anthraquinones glycosides.

Tests	Extracts	Solvents used				
		Water	Methanol	Chloroform	Ethyl acetate	Petroleum ether
Borntrager's Test	Leaves	-	-	-	-	-
	Flowers	-	-	-	-	-

++ More abundance, + Abundance, - Absence

Table 6. Tests for cardiac glycosides.

Tests	Extracts	Solvents used				
		Water	Methanol	Chloroform	Ethyl acetate	Petroleum ether
Burchard test (for glycosides steroids)	Leaves	-	-	-	+	++
	Flowers	-	-	-	+	-
Salkwoskii test (for steroidal nucleus)	Leaves	-	++	-	+	+
	Flowers	-	+	-	+	-
Keller killiani's test	leaves	++	+	-	-	++
	flower	++	-	+	+	-
Kedde test	leaves	-	-	-	-	-
	flowers	-	-	-	-	-

++ More abundance, + Abundance, - Absence

Table 7. Tests for Saponin.

Tests	Extracts	Solvents used				
		Water	Methanol	Chloroform	Ethyl acetate	Petroleum ether
Frothing test	Leaves	++	+	++	-	-
	Flowers	+	-	-	-	-
Haemolysis test	Leaves	+	+	+	-	-
	Flowers	+	+	-	-	-

++ More abundance, + Abundance, - Absence

disorders, herpes and rhinitis (Aliyu, 2006). When decocted, *S. siamea* leaves are locally used as anti-malaria drug (Lose et al., 2000). Previous studies on *S. siamea* extracts have confirmed some of the traditional uses: antiplasmodial activity (Gbeassor et al., 1990; Nsonde-Ntandou et al., 2005; Mbatchi et al., 2006). Antibacterial activities of the extract were tested against thirteen pathogenic bacteria and were compared with the standard antibiotic, kanamycin by measuring the zone of inhibition diameter and expressed in millimeter (mm) (Hailu et al., 2005; Dahiru et al., 2013).

Phytochemical screening reveals that methanolic extract contains carbohydrate, cardiac glycosides, saponins, flavonoids, tannins and alkaloids. The extracting solvent used are decreasing order of polarity in which each of them extract a number of solvent to their own polarity depending on the active metabolites the plant contained. Based on this experiment the alkaloid, tannins and saponins content of this can be responsible for its antibacterial activity (Dahiru et al., 2013)

Preliminary phytochemical analysis showed that leaf extracts of *S. siamae* possesses alkaloids, saponins, tannins and glycosides which is in support with studies done by Momin et al. (2012), Edeoga et al. (2005) and Bukar et al. (2009). Phytoconstituents such as saponins, phenolic compounds and glycosides when present in *S.*

Table 8. Tests for Flavonoids.

Tests	Extracts	Solvents used				
		Water	Methanol	Chloroform	Ethyl acetate	Petroleum ether
Shinoda's test	Leaves	-	+	-	-	-
	Flowers	-	+	-	-	
Ferric chloride test	Leaves	++	+	+	+	-
	Flowers	+	-	-	-	-
Lead acetate test	Leaves	+	++	-	-	-
	Flower	+	++	-	-	-
Sodium hydroxide test	Leaves	+	-	-	-	-
	Flowers	+	-	-	-	-

++ More abundance, + Abundance, - Absence

Table 9. Tests for alkaloids.

Tests	Extracts	Solvents used				
		Water	Methanol	Chloroform	Ethyl acetate	Petroleum ether
Mayers reagent	Leaves	-	-	-	-	-
	Flowers	-	-	-	-	-
Dragendorff A × B	Leaves	-	-	-	-	-
	Flowers	-	-	-	-	
Wagners reagent	Leaves	+	+	-	-	-
	Flower	+	+	-	-	-
10% w/v tannic acid	Leaves	-	+	-	-	-
	Flowers	-	-	-	-	-
1% w/v picric acid	Leaves	-	+	+	-	+
	Flowers	-	+	-	-	-

++ More abundance, + Abundance, - Absence

Table 10. Leaves methanol extract TLC results.

Spots positions (cm)	R_f values (cm)	Day light	UV-254 nm	UV-366 nm	Iodine vapour
16.3	0.89	Green	Green	Blue black	Light Green
15.2	0.83	Green	Green	Blue black	Light Green
14.1	0.77	Light green	Light green	Blue black	Yellow
11.5	0.62	Light yellow	Light yellow	-	Yellow
10.8	0.59	Yellow	Light brown	-	White
9.3	0.50	White	White	-	Brown

- No colour

siamea have been reported to inhibit bacterial growth and to be protective to plants against bacterial and fungal infections (Gonzalel and Mather, 1982; Okwute, 1992). Cardiac glycosides have also been found useful in

Table 11. Leaves Chloroform extract TLC results.

Spots positions (cm)	R_f values (cm)	Day light	UV-254 nm	UV-366 nm	Iodine vapour
16.8	0.91	White	White	White	Yellow
16.1	0.87	Green	Violet	Blur black	Green
14.6	0.79	Green	Green	Blue black	Green
14.4	0.78	Light green	Green	-	Yellow
13.7	0.74	Light green	Light green	-	Yellow
13.5	0.73	Light brown	White	-	Yellow
11.9	0.65	White	Violet	-	Brown
10.8	0.59	Light brown	Light brown	-	Brown
9.8	0.53	Light yellow	Violet	-	Brown

- No colour

Table 12. Leaves ethyl acetate extract TLC results.

Spots positions (cm)	R_f values (cm)	Day light	UV-254 nm	UV-366 nm	Iodine vapour
15.9	0.86	Green	Light green	Blue black	Yellow
14.8	0.80	Green	Green	Blur black	Green
13.9	0.75	Light green	Green	Blue black	Green
13.1	0.71	Light green	Light brown	-	Yellow
11.3	0.61	Light green	Light brown	-	Yellow
10.5	0.57	Yellow	Light brown	-	Yellow
9.4	0.51	Brown	Brown	-	Yellow
8.7	0.47	Green	Brown	-	Brown
7.6	0.41	Light green	Violet	-	Brown

- No colour

Table 13. Leaves petroleum ether extract TLC results.

Spots positions (cm)	R_f values (cm)	Day light	UV-254 nm	UV-366 nm	Iodine vapour
16.3	0.89	Green	Green	Blue black	Green
15.0	0.81	Green	Green	-	Light green
14.6	0.79	Yellow	Violet	-	Yellow

- No colour

Table 14. Flowers methanol extract TLC results.

Spots positions (cm)	R_f values (cm)	Day light	UV-254 nm	UV-366 nm	Iodine vapour
11.9	0.66	Yellow	Violet	Blue black	Brown
10.8	0.60	Light brown	Brown	-	Brown
9.7	0.54	Yellow	Violet	-	brown

- No colour

treatment of heart failure and supraventricular arrhythmias (Zamotaev et al., 2005). The traditional uses indicate that both the leaves and flowers have been used together for therapeutic purposes. From this study, findings showed that the leaves have more phyto-chemicals than the flowers and since all active metabolites in the flowers are also present in the leaves, using leaves alone might suffice for treatments.

Presence of alkaloids, tannins, saponins, glycosides, steroids, phenolic compounds and flavonoids in all the

Table 15. Flowers chloroform extract TLC results.

Spots positions (cm)	R_f values (cm)	Day light	UV-254 nm	UV-366 nm	Iodine vapour
15.9	0.88	Green	Light green	White	Brown
15.5	0.86	Light green	Yellow	White	Brown
14.5	0.81	White	White	White	Brown
12.9	0.72	Yellow	Violet	White	Brown
11.9	0.66	Yellow	Violet	White	Brown
8.9	0.49	Light brown	Brown	White	brown

- No colour

Table 16. Flowers ethyl acetate extract TLC results.

Spots positions (cm)	R_f values (cm)	Day light	UV-254 nm	UV-366 nm	Iodine vapour
15.9	0.88	Light green	Green	-	Brown
15.5	0.86	Yellow	Yellow	-	Brown
14.6	0.81	Light yellow	Violet	-	brown

- No colour, Solvent system- methanol: n-hexane: ethyl acetate (20: 60: 20), Running time- 53 min, Adsorbent used- silica aluminum sheet, Locating reagent- Day light, UV 254 nm, UV 366 nm and iodine vapour

extracts confirmed the presence of rich bioactive principles in the leaf. Tannins, steroids and glycosides had been reported in ethanol extract of the leaf of *S. siamea* (Bukar et al., 2009; Muhammad et al., 2012) while alkaloids, saponins, phenolics and flavonoids by Momin et al. (2012). Secondary metabolites are mostly produced by plant during adverse condition for protection against herbivores (Chitra et al., 1999). Alkaloids, flavonoids, tannins and saponins were known to show medicinal activity as well as exhibiting physiological activity (Edeoga et al., 2005). The presence of phenolic group in plants is to protect them from microbial, insect and herbivores damage (Conco, 2000). Many of these active compounds also possess other functional attributes like anti-inflammatory, antimutagenic, hypocholestemic and antiplatelet aggregation properties (Praveena et al., 2012). These phytochemical compounds carry out their activity by combining with protein, lipids or other components of the bacterial cell membrane that are relevant to one or more vital physiological roles thereby disrupting the integrity and functional behaviour of the membrane (de Kruijff et al., 2000).

Conclusion

From our findings, it can be concluded that *S. siamea lam* contains some significant phytochemicals that can exhibit desired therapeutic activities such as hypoglycemia, anti-arrthymia and antimicrobial. However there is the need to conduct further pharmaceutical analyses on test extracts in order to establish these biomedical applications.

Conflict of interest

There are none to declare.

REFERENCES

Ajaiyeoba EO, JS Ashidi, Okpako LC, PJ Houghton, CW Wright (2008). Antiplasmodial compounds from Cassia siamea stem bark extract. Cassia siamea L. (Fabaceae). Phytother. Res. 22(2):254-255.

Aliyu BS (2006). West African Ethnomedicinal Plants. Triumph Publishing Company, Kano, Nigeria.

Bukar A, Mukhtarand MD, Hassan AS (2009). Phytochemical screening and antibacterial activity of leaf extracts of *Senna siamea* (lam) on *Pseudomonas aeruginos*". Bayero J. Pure Appl. Sci. 2(1):139-142.

Chitra S, Patil MB, Rav K (2009). Wound healing activity of Hyptissuaveolens (L) Poit (Laminiaceae). Int. J. Pharm. Tech. Res. 1: 737-744.

Conco WZ (1999). Zulu Traditional Medicine, its role in modern society. Comm. Health 5: 8-13.

Dahiru D, Malgwi AR, Sambo HS (2013). Growth Inhibitory Effect of Senna siamea Leaf Extracts on Selected Microorganisms. Am. J. Med. Med. Sci. 3(5):103-107.

Dalziel JM (1958). Flora of West Tropical Africa. Vol. I Part II, R. W. J. Keay (Ed.). Crown Agents, London pp. 485-503.

de Kruijff B, Cullis RP,Verkelij JA, Hope JM,Van Echteld AJC, Taraschi FT (2000). Lipid polymorphism and membrane function. Enzym of Biol Membranes. (Martonosi, A. N.,Ed.), 2nd ed. Plenum Press, New York pp. 131-204.

Edeoga HO, Okwu DE, Mbaebie BO (2005). Phytochemical consttituent of some Niger ian Medicinal plants. Afr. J. Biotechnol. 4(7):685-688.

Gbeassor M, Kedjagni AY, Koumaglo K, de Souza C, Agbo K, Aklikokou K, Amegbo KA (1990). *In vitro* Antimalarial Activity of Six Medicinal Plants. Phytother. Res. 4(3):115-117.

Gonzalel L, Mather S (1982). Identification of terpenoids from leaves of

Piptocarphaperitora and their biological activities. J. Nat. Prod. 45:495-496
Hailu T, Endris M, Kaleab A, Tsige G (2005). Antimicrobial activities of some selected traditional Ethiopian medicinal plants used in the treatment of skin disorder. J. Ethnopharmacol. 100:168-175.
Harbone JB (1973). Phytochemical methods. London, Chapman and Hall Ltd. pp 49-188
Hill AR (1992). Medicinal plants and tradition medicine in Africa. J. Int. Med. 39:42-45.
Keay RWJ (1989). Trees of Nigeria. Clarendon Press, Oxford. Medicine in Africa 2nd edition Spectrum Books Ltd, Sunshine House, Ibadan Nigeria pp. 81-93, 134-156.
Kiepe P (1995). Effects of Cassia siamea hedgerow barriers on soil physical properties. Geoderma 66:113-120.
Kretovich UL (2005). Principles of plant biochemistry permagon. Oxford Press. J. Food Sci. 54:254-260.
Lose GA, Bernard SJ, Leihner DE. (2000). Studies on agro forestry hedgerow system with Senna siamea rooting patterns and competition effects. J. Ethnopharmacol. 38, 57-60.
Mbatchi SF, Mbatchi B, Banzouzi JT, Bansimba T, Nsonde-Ntandou GF, Ouamba JM, Berry A, Benoit-Vica LF (2006). *In vitro* antiplasmodial activity of 18 plants used in Congo Br-azzaville traditional medicine. J. Ethnopharmacol. 104:168-174.
Mohammed A, Mada SB, Yakasai HM. (2012). Subchronic study of aqueous stem bark extract of Senna siamea in rats". Asian J. Biol. Sci. 5(6):314-321
Momin MAM, Bellah SF, Afrose A, Urmi KF, Rana MS (2012). Phytochemical Screening and Cytotoxicity Potential of Ethanolic Extracts of Senna siamea Leaves". J. Pharm. Sci. Res. 4(8):1817-1879.
Namukobea J, Kaseneneb JM, Kiremere BT In. Amukama M, Kamatenesi-Mugisha S, Krief V, Dumontet DJ, Kabasa JD (2011). Traditional plants used for medicinal purpose by local communities around northern sector of KiboleNational park, Uganda. J. Ethnopharmacol. 136:236-255.
Nsonde-Ntandou GF, Ndounga M, Ouamba JM, Gbeassor M, Etou-Ossibi A, Ntoumi F, Abena AA (2005). Ethnobotanical survey, chemical screening and effective treatment of certain plants used in traditional medicine to treat malaria in Brazzaville. Phytotherapia 1: 13-18.
Nsonde Ntandou GF, Banzouzi JT, Mbatchi B, Elion-Itou RD, Etou-Ossibi AW, Ramos S, Benoit-Vical F, Abena AA, Ouamba JM (2010). Analgesic andanti-inflammatory effects of Cassia siamea Lam. stem bark extract. J. Ethnopharmacol. 127:108-111.
Okwute SK (1992). Plant derived pesticidal and antimicrobial agents for use in Agriculture. A review of phytochemical and biological studies on some Nigerian plants. J. Agric. Sci. Technol. 2(1):62-70.
Otimenyin SO, Kolawole JA, Nwosu M (2010). Pharmacological basis for the use of the root of Senna siamea in traditional medicine. Int. J. Pharm. Biosci. 1:1-9.
Praveena B, Pradeep SN (2012). Antioxidant and Antibacterial Activities in the Leaf Extracts of Indian Borage (Plectranthusamboinicus). Food Nutr. Sci. 3:146-152.
Roja G, Rao PS (2000). Anticancer compound from tissue cultures of medicinal plant. J. Herbs Spices Med. Plants 7:71-102.
Sofowora A (1993). Medicinal plants and Traditional Medicines in Africa. Spectrum Books Ltd, Ibadan, Nigeria p 289
Sofowora A (2000). Phytochemical screening of Nigerian medicinal plants. J. Integr. Med. 41:234-246.
Trease GE, Evans MC (2008). A textbook of Pharmacognosy. BuillerTindall and Causel London 13th Edition 1978, pp 176-180.
Zamotaev IUN, Kremnev IUA, Podshibiakin SE (2005). Cardiac glycosides in complex treatment of patients with heart failure and supraventricular arrhythmias. Klin. Med. (83)7:59-63.