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Research Article

Qualitative and Quantitative Analyses of Secondary Metabolites (Phytochemicals) in Neem Stem Bark

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Abstract

The astronomical increase in the costs of chemical fertilizers, agrochemicals, animal feeds and most especially synthetic drugs, has led to an increase in debt and poverty rate coupled with the resultant environmental degradation caused by exploring the resources. However, Neem tree has proven to be a great substitute with great potential in the 21st century. In this research, the bioactive chemical constituents of Neem stem bark were extracted, separated and analyzed to determine the chemical composition of the fractions using various methods. A fresh neem sample was collected at Shalom Farm, Ojoo area, Ibadan in Oyo state, the sample was grinded into powdered form and prepared via extraction using various solvents; n-hexane, ethyl acetate and ethanol and the chemical constituents were separated using GC/MS technique and later on the neem stem sample was digested with nitric acid and hydrogen peroxide in the ratio 4:1 (acid ratio). Qualitative and quantitative phytochemical analyses was carried out on the sample extract and finely ground neem stem sample (as the case may be). The results showed that the prominent metabolites in Neem stem bark are Saponins and Alkaloids, while Flavonoids, Anthraquinones, Steroids etc. are present in trace amount and also Cardenolides and Chalcones were not detected.

It is very important that more research be carried out on Secondary metabolites, which has proven to be very helpful to the chemical industries to produce plant based chemicals to minimize environmental degradation and to promote green chemistry.

Keywords: Neem; phytochemicals; solvents; metabolites.

Introduction

Azadirachta indica, commonly known as the neem tree, is a tropical evergreen tree that is native to the Indian subcontinent (Aneesa, 2016). For thousands of years, neem has been recognized for its wide array of beneficial properties, including those in agriculture for pest control and in traditional medicine for various common human ailments. *A. indica* originally provoked world-wide interest due to its capacity as a non-toxic infection-control agent for use in farming (Govindachari, 1992).

Almost every part of *A. indica* (e.g., the stem, bark, roots, leaves, gum, seeds, fruits, flowers, etc.) have been used as house-hold remedies for human illnesses. Moreover, millions of people globally use neem twigs as a source of chewing sticks for dental hygiene (Brahmachari, 2004).

More recently, the neem tree has gained attention from modern medicine and infectious disease researchers as a potential source for new antimicrobials, in addition to the applications of *A. indica* in the fields of oncology, dentistry, dermatology, and endocrinology, among others; for reviews on some of these individual topics, see (Iman *et al*., 2021., Yarmohammadi *et al*., 2021).

Every part of the tree has been used as traditional medicine for household remedy against various diseases (Biswas *et al*., 2002). It elaborates a vast array of biologically active compounds that are chemically diverse and structurally variable with different ingredients isolate from different parts of the tree (Subapriya and Nagini, 2005). The active ingredients include alkaloids, lavonoids, phenolic compounds, carotenoids, steroid and ketones, which have antihelminthic, antimicrobial, antiulcer, antifertility, antidiabetic, anti-inflammatory and antitumor properties and the plant is used in combination with oil for more effectiveness to reduce toxicity (Udeinya *et al*., 2004).

United Nations has declared Neem tree as the "Tree of the 21st century" while The US National Academy of Science has designated this incredible tree as "Neem: A tree of solving global problems" in its report published in 1992. In Indian context it is famous as "Village dispensary", "Divine tree", "heal all" while in European literature "Materia medica" neem has been regarded as "Panacea of all Disease". Neem extracts have been shown to possess anti-bacterial, anti-fungal, potent antiviral and anticancerous properties (Udeinya *et al*., 2004). In 1942 for the first time, Nimbin, a bitter compound had been isolated from Neem (Biswas *et al*., 2002). Above 300 compounds have been derived from diverse parts of neem. There are mainly two types of metabolites viz., primary including protein, fatty substance or carbohydrate or sugar derivative and secondary metabolites containing various alkaloids, steroids, flavonoids, saponins etc. The compounds isolated have been broadly classified into two major groups-isoprenoids and non-isoprenoids. Compounds such as diterpenoids, triterpenoids and steroids including azadirone, protomeliacins, limonoids, gedunin, vilasinin type of compounds, C-secomeliacins such as azadirachtin, nimbin, salanin and its derivatives comprise isoprenoids group while non-isoprenoids contain proteins/amino acids, polysaccharides, sulphurous compounds, polyphenolics such as flavonoids, their glycosides, dihydrochalcone, coumarin, tannins, aliphatic compounds etc. (Biswas *et al*., 2002).

Role in general health and diseases:

- AIDS: The National Institutes of Health reports neem extracts killed the AIDS virus and patents have been awarded for these extracts as an AIDS treatment (Anyaehie, 2009)
- Heart disease: Neem delays the coagulation of blood, calms erratic heart beats and helps reduce elevated heart rates and high blood pressure
- Diabetes: Neem leaf extracts taken orally reduces the insulin requirements by 30-50% in non-ketonic and insulin-sensitive diabetic patients
- Periodontal disease: German researchers have proven neem extracts prevent tooth decay and periodontal disease (Prashant *et al*., 2007; Chava *et al*., 2012) leading to good oral health (Balappanavar *et al*., 2013). Neem leaf extract has a antimicrobial effect on Enterococcus faecalis and Candida albicans. Therefore, it can be a potential endodontic irrigant (Vinothkumar *et al*., 2013)
- Skin diseases: On skin conditions that have reached chronicity neem has got a significant effect. These conditions include: Acne and psoriasis; eczema and ringworm; even stubborn warts that can be easily cleaned up with the use of organic neem oil which is of high quality. For the treatment of skin diseases in Siddha medicine both neem oil as well as leaves have been used. For clearing and beautifying as well as rejuvenating the skin as an excellent component of cosmetics neem oil can be used (Thas, 2008)
- Ulcers: Neem extracts give significant protection from discomfort and speed the healing of gastric and duodenal lesions (Maity *et al*., 2009)
- Sexually transmitted diseases: The efficacy of neem in the treatment of sexually transmitted diseases have been highlighted by few researchers and overwhelming positive responses have been reported. The efficacy of neem extract in Neisseria gonorrhea infection is also well proven (Shokeen *et al*., 2009) Anti-infection (against infective agents as bacteria, viruses, fungi, protozoa).

Materials and Methods

Sample Collection

A fresh Neem stem sample was collected at Shalom farm, Ojoo area, Ibadan, Oyo state. After collection, the outer part of the stem sample was peeled using a well-sharpened knife and then the sample was washed with water and then air-dried for five (5) weeks. After air-drying, the sample was reduced to simpler sizes by the use of a thoroughly –washed mortar and pestle and the grinded into powdered form using a grinding machine.

Sample Authentication

It was ensured that it was Neem stem sample that was collected and not any other plant sample

Sample Preparation

100g of the powdered form of the sample was extracted in a container using three (3) solvents namely; n-hexane, ethyl

acetate and ethanol. The sample was first extracted with 500ml of n-hexane solvent for seventy-two hours (3 days) shaking the container at least thrice each day for uniform and accurate extraction and then filtered after 72hours of extraction. This process was also repeated for the two remaining solvents and an extract was gotten from the sample. The extract gotten from the sample was then concentrated using a rotary evaporator and cream-like concentrate was gotten from the extract with the weight recorded. The concentrate was then used to carry out Column Chromatography in which the 3 solvents (with changing polarities) acting as the mobile phase and silica gel acting as the stationary phase yielded about 29 fractions. The weight of the concentrate is recorded below.

Weight of ethanolic concentrate of Neem stem sample: 21.1g

Apart from the extraction and column chromatography carrtoied out on the powdered form of the Neem stem sample, some other analyses were also carried out on the sample. They include digestion, heavy metal analysis, vitamins analysis and phyto-chemical analysis.

Digestion

The Neem stem sample was digested with nitric acid and hydrogen peroxide in the ratio 4:1 (acid ratio). To a well-rinsed labelled and dried digestion flask, 1.0g of Neem stem samples were added to each digestion flask as labelled. 8ml of concentrated nitric acid (HNO_3) and 2ml of hydrogen peroxide (H_2O_2) was added to each sample in the digestion flask and the solution was heated on a hot plate in a fume chamber for 2 hours. After the 2 hours of heating, the samples were allowed to cool and filtered into 100ml volumetric flask and the volume was made up to 100ml mark with deionized water and stored in a polyethylene container prior to analysis.

Phytochemical Analyses

Alkaloids Determination

This is a distillation and titrimetric procedure. 2g of finely-ground neem stem sample was weighed into a 100ml beaker and 20ml of 80% absolute alcohol was added to give a smooth paste. The mixture was transferred to a 250ml flask and more alcohol added to make up to 100ml and 1g of magnesium oxide was added. The mixture was digested in a boiling water bath for 1.5hours under a reflux air condenser with occasional shaking. The mixture was filtered while hot through a small Buchner funnel. The residue was returned to the flask and re-digested for 30minutes with 50ml alcohol after which the alcohol will be evaporated, adding hot water to replace the alcohol lost. When all the alcohol has been removed, 3 drops of 10%HCl was added. The whole solution was later transferred into a 250ml volumetric flask, 5ml of Zinc acetate solution and 5ml of potassium ferrocyanide solution was added, thoroughly mixed to give a homogenous solution. The flask was allowed to stand for a few minutes, filtered through a fry filter paper and 10ml of the filtrate was transferred into a separating funnel and the alkaloids present were extracted vigorously by shaking with five successive portions of chloroform. The residue obtained was dissolved in 10ml hot distilled water and transferred into a Kjedahl tube with the addition of 0.20g sucrose and 10ml conc. H2SO4 and 0.02g selenium for digestion to a colourless solution to determine %N bu Kjedahl distaillation method. %Nitrogen gotten is converted to % total alkaloid by multiplying by a factor of 3.26 i.e.

%Total alkaloid = %N X 3.26

Tannins Determination

0.20g of neem stem sample was measured into a 50ml beaker, 20ml of 50% methanol was added and covered with parafilm and placed in a water bath at 77Oc – 80Oc for 1 hour. It was shaken thoroughly to ensure a uniform mixing. The extract was quantitatively filtered using a double layered Whatman No 41 filter paper into a 100ml volumetric flask, 20ml of water added, 2.5ml Folin-Denis reagent and 10ml of 17% Na_2CO_3 were added and mixed properly. The bluish-green color (will develop at the end of range 0-10ppm) was treated similarly as 1ml sample above. The absorbance of the Tannic acid standard solution as ll as samples were read after color development on a spectronic 21D Spectrophotometer at a wave length of 760nm. %Tannin was calculated using the formula given the box 1 below.

Box 1: Formula used for calculation of %Tannin

$$\%\text{Tannin} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{Wt. of sample} \times 10{,}000}$$

Phlobatannins Determination

0.50g of neem stem sample extract was weighed into 5oml beaker, 20ml of 50% Methanol was added and covered with parafilm and placed in a water bath at 77Oc – 80Oc for 1 hour. It was shaken thoroughly to ensure a uniform mixing and was later filtered through a Whatman No 1 filter paper into a 50ml volumetric flask using aqueous methanol to rinse, and make up to mark with distilled water. 1ml of the sample extract was pipetted into a 5oml volumetric flask, 20ml of water, 2.5 ml Folin-Dennis reagent and 10ml of 17% Sodium Carbonate solution were added to the solution in the 50ml flask. The mixture was homogenized thoroughly for 20minutes. 0-5mg/ml of Phlobatannin standard concentration was prepared from 100mg/ml phlobatannin stock solution and treated like sample above. The absorbances of standard solutions as well as sample were read on a Spectronic 21D spectrophotometewr at a wavelength of 55onm. % Phlobatannin was calculated using the formula given in the box 2.

Box 2: Formula used for calculation of % Phlobatannin

$$\%\text{Phlobatannin} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{Wt. of sample} \times 10{,}000}$$

Saponins Determination

1g of finely ground neem stem sample was weighed into a 250ml beaker and 100ml of isobutyl alcohol was added. The mixture was shaken into a UDY shaker for 5hours to ensure uniform mixing. Thereafter, the mixture was filtered through a Whatman No 1 filter paper into a 100ml beaker and 20ml of 40% saturated solution of magnesium carbonate was added. The mixture obtained with saturated MgCO3 was again filtered through a Whatman No 1 filter paper to obtain a clear colorless solution. 1ml of the colorless solution was pipetted into 5oml volumetric flask and 2ml of 5% FeCl3 solution was added and made upto mark with distilled water. It was allowed to stand for 30minutes for blood red color to develop. 0-10ppm standard Saponin solutions were prepared from saponin stock solution. The standard solutions were treated similarly with 2ml of 5% FeCl3 solution as done for 1ml sample above. The absorbance of the sample as well as standard saponin solutions were read after colour development in a Jenway Spectrophotometer at a wavelength 0f 380nm. The % saponin can be calculated using the equation given in box 3 below.

Box 3: Formula used for calculation of % Saponin

$$\%\text{Saponin} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{Wt. of sample} \times 10{,}000}$$

Flavoniods Determination

0.50g of finely-ground neem stem sample was weighed into a 100ml beaker and 80ml of 95% ethanol was added and stirred with a glass rod to prevent lumping. The mixture was filtered through a Whatman No 1 filter paper into a 100ml volumetric flask and made up to mark with ethanol. 1ml of the extract was pipetted into a 50ml volumetric flask, four drops of conc. HCl was added via a dropping pipette after which 0.5g of magnesium turnings added to develop a magenta red coloration. Standard flavonoid solution of range 0-5ppm were prepared from 100ppm stock solution and treated in a similar way with HCl and magnesium turnings like sample. The absorbance of magenta red coloration of sample and standard solutions were read on a digital Jenway V6300 Spectrophotomter at a wavelength of 520nm. The % flavonoid is calculated using the formula is given in box 4.

Box 4: Formula used for calculation of % Flavonoids

$$\%\text{Flavonoids} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{Wt. of sample} \times 10{,}000}$$

Anthraquinones Determination

0.50g of finely-ground neem stem sample was weighed into a 250ml beaker and 60ml of benzene was added and stirred with a glass rod to prevent lumping. This was filtered into 100ml volumetric flask using a Whatman No 1 filter paper. 10ml of filtrate was pipetted into another 100ml volumetric flask and 0.2% zinc dust was added followed by the addition of 50ml hot 5% NaOH solution. The mixture was hated lust below boiling point for 5 minutes and then rapidly filtered and washed once in water. The filtrate was again heated with another 50ml of 5% NaOH to develop a red color. Standard anthraquinone solutions of range 0-5mg/l was prepared from 100mg/l stock anthraquinone and treated in a similar way with 0.2% zinc dust and NaOH like sample. The absorbance of sample as well as that of standard concentrations were read on a digital spectrophotometer at a wavelength of 640nm. The % Anthraquinone is calculated using equation given in the box 5.

Box 5: Formula used for calculation of % Phlobatannin

$$\%\text{Phlobatannin} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{Wt. of sample} \times 10{,}000}$$

Steroids Determination

0.50g of sample extract was weighed into a 100ml beaker. 20ml of Chloroform-Methanol (2:1) mixture was added to dissolve the extract upon shaking for 3ominutes on a shaker. The whole mixture was later filtered through a Whatman No1 filter paper into another dry clean 100ml conical flask/beaker. The resultant residue was repeatedly treated with Chloroform-Methanol mixture until free of steroids. 1ml of the filtrate was pipetted into 30ml test tube and 5ml of alcoholic KOH was added and shaken thoroughly to obtain a homogenous mixture. The mixture was later placed in a water bath set at 37oC-40Oc for 90mins. It was cooled to room temperature and 10ml of petroleum ether was added followed by the addition of 5ml of distilled water. This was evaporated to dryness on a water bath. 6ml of Liebermann Burchard reagent was added to the residue in dry bottle and absorbance taken at a wavelength of 620nm on a spectronic 21D digital Spectrometer. Standard steroids of concentration of 0-4mg/ml were prepared from 100mg/ml stock steroid solution and treated similarly like sample as above. % Steroid was calculated using the formula shown in the box 6.

Box 6: Formula used for calculation of % Steroids

$$\%\text{Steroids} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{Wt. of sample} \times 10{,}000}$$

Terpenes Determination

0.50g of neem stem sample was weighed into a 50ml conical flask, 20ml of 2:1 Chloroform-Methanol mixture was added, shaken thoroughly and allowed to stand for 15minutes. The mixture was later centrifuged for another

15minutes, supernatant obtained was discarded and the precipitate was rewashed with another 20ml chloroform-methanol mixture for re-centrifugation. The resultant precipitate was dissolved in 40ml of 10% Sodium Dodecyl Sulphate solution. 1ml of 0.01M Ferric Chloride solution was added to the above at 30seconds interval shaken well and allowed to stand for 30minutes. Standard Terpenes of concentration range 0-=5mg/ml were prepared from 100mg/ml stock terpenes solution from Sigma-Aldrich chemicals, U.S.A. the absorbance of the sample as well as that of standard concentrations of terpenes were read on a Digital Spectrophotometer at a wavelength of 510nm. The % terpenes was calculated using the formula given in the box 7.

Box 7: Formula used for calculation of % Terpenes

$$\%\text{Terpenes} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{Wt. of sample} \times 10{,}000}$$

Cardenolides Determination

0.50g of sample extract was accurately weighed into 100ml beaker followed by the addition of 50ml of chloroform to dissolve the extract. 0.20g of Sodium Bicarbonate powder (NaHCO3) was d after complete dissolution of extract in chloroform to remove any free acid. The mixture was later transferred into a 250ml separating funnel and thoroughly shaken to allow the two layers to separate. 5 drops of acetic anhydride were added to allow the mixture to be clear and free of cloudy suspensions. This was filtered through a Whatman No 1 filter paper into 100ml volumetric flask and made up to mark with chloroform. Standard cardenolide solutions of concentration 0-10mg/ml was prepared from 100mg/ml stock cardenolide solution and treated similarly as sample above to obtain the gradient factor. % Cardenolide was calculated using the formula (see box 8)

Box 8: Formula used for calculation of % Cardenolides

$$\%\text{Cardenolides} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{Wt. of sample} \times 10{,}000}$$

Phenols Determination

0.20g of neem stem sample was weighed into a 50ml beaker, 20ml of acetone was added and homogenized properly for 1 hour to prevent lumping. The mixture was filtered through a Whatman No 1 filter paper into a 100ml volumetric flask using acetone to rinse and made up to mark with distilled water with thorough mixing. 1ml of sample extract was pipetted into 50ml volumetric flask, 20ml of water was added, then 3ml of phosphomolybdic acid was added followed by the addition of 23% Na2CO3 and mixed thoroughly then made up to mark with distilled water and allowed to stand for 10minutes to develop bluish-green color. Standard phenol solutions of concentration range 0-10mg/ml was prepared from 100mg/ml stock phenol solution from Sigma-Aldrich chemicals, U.S.A. The absorbance of the sample as well as that of the standard concentrations of phenol were read on a digital spectrophotometer at a wavelength of 510nm. The % phenol is calculated using the formula given in the box 9.

Box 9: Formula used for calculation of % Phenol

$$\%\text{Phenol} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{Wt. of sample} \times 10{,}000}$$

Chalcones Determination

0.50g of sample extract was weighed into a 100ml beaker. 20ml of chloroform-methanol (2:1) mixture was added to dissolve the extract upon shaking for 3ominutes on a shaker. The whole mixture was later filtered through a Whatman No 1 filter paper into another dry clean 100ml conical flask/beaker. The resultant residue was pipetted into a 30ml test tube and 5mlof alcoholic KOH was added and shaken thoroughly to obtain a homogenous mixture. The mixture water placed in a water bath set at 37Oc-400c dor 90minutes. It was cooled at room temperature and 10ml of petroleum ether added followed by the addition of 5ml distilled water. This was evaporated to dryness on the water bath, then 6ml of Liebermann Burchard reagent was added to the residue in dry bottle and absorbance taken at a wavelength of 620nm on a Spectronic 21D Digital Spectrophotometer. Standard Chalcone standards of concentration of0-4mg/ml were prepared from 100mg/ml stock steroid solution and treated similarly like sample above. % Chalcones was calculated using the formula shown in box 10.

Box 10: Formula used for calculation of % Chalcones

$$\%\text{Chalcones} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{Wt. of sample} \times 10{,}000}$$

Cardiac Glycosides Determination

10ml of sample extract was into a 250ml conical flask. 50ml chloroform was added and shaken on a Vortex Mixer for 1hour. The mixture was filtered into 100ml conical flask and 10ml pyridine, 2ml of 2% sodium nitroprusside were added and shaken thoroughly for 10minutes. 3ml of 20%NaOH was later added to develop a brownish-yellow color. Glycoside standards of concentrations which range from 0-5mg/ml were prepared from 100mg/ml stock glycoside solution standard. The series of standards 0-5mg/ml was treated similarly like sample above. The absorbance of sample as well as standards were read on a Spectronic 21D Digital Spectrophotometer at a wavelength of 510nm. % Glycoside was calculated using the formula given in box 11.

Box 11: Formula used for calculation of % Glycoside

$$\%\text{Glycoside} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{Wt. of sample} \times 10{,}000}$$

Results and Discussions

According to the result of the phytochemical analysis, the prominent metabolites in Neem stem bark are Saponins and Alkaloids, while Flavonoids, Anthraquinones, Steroids etc. are present in trace amount and also Cardenolides and Chalcones were not detected (Table 1& 2). Saponins aids the reduction of blood cholesterol level, reduce the risk of cancer and stimulate our immune system. It also aids the treatment of certain diseases such as malaria, eczema etc. and can also be used as a detergent, thus, it is economical when compared to other chemical detergents. They can also be used as adjuvants in vaccines but toxicity associated with sterol complexation needs to be taken note of. Saponins are widely used also for their effects on ammonia emissions in animals (Hostetman and Marston, 1995).

Table 1: Qualitative estimate of the secondary metabolites present in neem stem bark

Phytochemical	Observation
ALKALOIDS	+++
TANNINS	+++
PHLOBATANNIN	+
SAPONINS	+++
FLAVONOIDS	+
ANTRAQUINONES	+
STEROIDS	+
TERPENES	+
CARDENOLIDES	N.D
PHENOLS	+++
CHALCONES	N.D
CARDIAC GLYCOSIDES	+++

KEY: +++ Appreciable amount; ++ Moderate amount; + Trace amount; N.D Not Detected

Table 2: Quantitative estimate of the secondary metabolites present in the neem stem bark

Phytochemical	Mean (%)
ALKALOIDS	0.5915
TANNINS	0.0800
PHLOBATANNINS	0.0220
SAPONINS	0.7295
FLAVONOIDS	0.0055
ANTHRAQUINONES	0.0019
STEROIDS	0.0017
TERPENES	0.0012
CARDENOLIDES	0.0000
PHENOLS	0.1860
CHALCONES	0.0000
CARDIAC GLYCOSIDES	0.3455

According to some researches, it was also discovered that saponins can be used as dietary supplements and nutraceuticals. Alkaloids e.g. thebaine are important in production of synthetic and semi-synthetic drugs. They are also used in the production of pesticides, insecticides etc. Alkaloids e.g. Ergonovine reduces uterine haemorrhages after childbirth, also, Tubocurarine can also be used as muscle relaxant in surgery. Nearly all alkaloids are poisonous in large amounts, hence, they are required to be used minimally.

Conclusion

The qualitative and quantitative screening of the investigated phytochemicals in this work, using *Azadirachta indica* stem-bark revealed that these secondary metabolites in plants occur in different qualities and quantities. Their usefulness to pharmaceutical and other chemical industries for the production of drugs for malaria, hypertension, cancer, antidotes for many poisons, birds and insects' repellant and treatment of skin infection, have been more in the Traditional or herbal form. More technologies should be adopted to better utilize this part of neem. Goverments across the world should set policies so as to preserve this specie of plant due to the pharmacological importance of the stem bark. Lastly, government should encourage researches on medicinal plants so as to encourage local sound health care services, consequently improving the economy.

Conflicts of Interest

The authors declare that there is no conflict of interest with present publication

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