FUDMA Journal of Sciences (FJS)
ISSN online: 2616-1370
ISSN print: 2645 - 2944
Vol. 4 No. 1, March, 2020, pp 37 – 42

PHYTOCHEMICAL SCREENING AND FREE RADICAL SCAVENGING ACTIVITIES OF METHANOL LEAF AND FLOWER EXTRACT OF *Securidaca longipedunculata*

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ABSTRACT
Securidaca longipedunculata is one of the known medicinal plants in Africa belonging to the Polygalaceae family. The objective of this study was to investigate the phytochemical contents as well as the free radical scavenging properties of methanol leaf and flower extract of the plant. The plant material was extracted using methanol for 72 hours. Extraction yielded 24.432% and 19.88% (w/w) of leaf and flower respectively. The preliminary phytochemical screening for leaf extract revealed the presence of phenols, tannins, flavonoids, saponins, glycosides, steroids, terpenoids, quinones and fixed oil while alkaloids, coumarin and xanthoproteins were not detected. The flower extract showed the presence of alkaloids, tannins, flavonoids, terpenoids and phenols while steroids and saponins were absent. The leaf and flower extracts revealed significant DPPH free radical scavenging activity. The total flavonoid content of leaf extract was found to be 17.0867 ± 0.268 mg Quercetin Equivalent/g extract while total phenolics content was found to be 22.73 ± 0.04583mg Gallic Acid Equivalent/g extract. The total flavonoid content of flower extract was found to be 6.0 ± 0.273 mg Quercetin Equivalent//g extract while total phenolics content was found to be 18.17 ± 0.017mg Gallic Acid Equivalent /g extract. Our study showed that *S. longipedunculata* leaf and flower extracts have important bioactive constituents and possess potent *in vitro* antioxidant activity. These parts of the plant may therefore be used as source of important pharmacological compounds for the treatment of oxidative stress related ailments.

Keywords: Phytochemicals, Antioxidants, Free radicals, *Securidaca longipedunculata*, DPPH

INTRODUCTION

Attention have shifted to the use of medicinal plants for the management and treatment of diseases hence the need by scientists to validate their ethno-medicinal applications as remedy against diseases (Taiwo *et al.*, 2017). Also, most herbal medicines are thought to be safe, devoid of much adverse side effects especially when compared with synthetic drugs (Shemishere *et al.*, 2018). Currently, it has been recorded that 80% of the world's population depend on traditional medicine for their primary health care needs. The greater part of this traditional medicine involves the use of plant extracts or their active principles (WHO, 1993). According to World Health Organization (WHO) medicinal plants are plants that have bioactive compounds that can be used for therapeutic purposes or those that synthesizes metabolites that can be used to produce useful drugs (Paul *et al*., 2018). Plant-derived substance have recently become of great interest due to their versatile applications especially plant based constituents that can be derived from any part of a plant ranging from leaves to the roots (Benslama and Harrar, 2016; Tiwari *et al*., 2011).

The therapeutic properties of medicinal plants rely on the bioactive compounds which are also known as phytochemicals (Yadav *et al*., 2014; Padmanabhan and Jaugle, 2012; Banu and Cathrine, 2015). Phytochemicals means naturally occurring chemicals in plants that confer a protective function to plants against bacteria, viruses, fungi, damage by free radicals, insects and herbivores that feed on them and any other environmental threat (Molyneux, 2007*;* Curan, 2017). Most environmental threats and stressors potentiate their damages by the generation of free radical. Free radicals also known as reactive oxygen species (ROS) have a potential to damage cell components resulting to diseases. They are involved in initiation and progression of many diseases among humans like cancer, cardiovascular diseases etc. (Ahmed *et al*., 2015; Njoya, *et al*., 2017). Notwithstanding, plants have the ability to counteract the damaging effects of free radical production through the use of some plant chemicals known as antioxidant phytochemicals. These phytochemicals interfere with oxidative process by scavenging free radicals by acting as electron donors thereby neutralizing their oxidative activity that could lead to disease conditions (Padmanabham and Jaugle, 2012; Garcia, 2012; Shirazi, 2014; Baba, 2015). Antioxidative phytochemicals present in medicinal plants such as flavonoids and phenolics participate in the management of diseases especially those caused by free radical damage by preventing the oxidative damage by free radicals (Halliwell and Gutteridge, 1989; Baba, 2015). Due to the potency of these plant compounds in the management and treatment of diseases, they are being included in the preparation of some drugs and a good number of prescribed medicines in developing and industrialized countries are derived directly or indirectly from plants (Oladeji, 2016). Medicinal plants can be the solution to the challenges faced by pharmacological sciences these days which include toxicity of drugs and drug resistance, these challenges are wake up calls to search for natural sources (medicinal plants) in order to discover and develop new therapeutic agents with improved efficacy (Kirby, 1996).

Securidaca longipedunculata is one of the famous medicinal plants especially in African countries. This plant belongs to

Polygalaceae family commonly known by the following names: English (Violet Tree); Hausa (Sanya popular known as 'Uwar Magunguna' meaning 'mother of all drugs); Yoruba (Ipeta) (Orwa *et al*., 2009); Igbo (Ezeogwu) (Iwu, 1986) in Nigeria. In Africa, *S. longipedunculata* is used in treatment of chest pain, headache, inflammation, infertility problems, Tuberculosis, constipation (Orwa *et al*., 2009), Malaria, stomach sickness, sleeping sickness and wound dressing (Dapar *et al*., 2007; Junaidu *et al.,* 2014). The different parts of *S. longipedunculata*; most especially the roots which seems to be the most used part of the plant, are suspected to be rich in different kinds of phytochemicals (Okoli *et al*., 2006). Despite the role of medicinal plants in treatment of diseases, there is little knowledge about them (Kirby, 1996) and this is buttressed by the findings of Oladeji, 2016 that there are about 1000 medicinal plants in Nigeria and most of their medical activities have not yet been investigated. Therefore, in continuing effort to finding suitable and non-toxic drugs for various human diseases, the acclaimed potency of this plant in the management of various diseases, stimulated our interest to screen the methanol leaf and flower extracts for their phytochemicals and Diphenyl-2-Picryl-Hydrazyl (DPPH) free radical scavenging activity.

MATERIALS AND METHOD

Sample collection and preparation

The leaf and flower samples were collected from the natural habitat of Ganwa village, Danko Wasagu local government, Kebbi State, Nigeria with the help of the village men. The plant was authenticated at the College of Agriculture Zuru, Kebbi State, Nigeria.

The leaves and flowers were stripped from the small twigs of the plant, washed with clean water and shade dried to minimize loss of volatile constituents of the leaves. After drying, the leaves were reduced to size using pestle and mortar.

The plant material was soaked in absolute methanol for 72 hours for extraction. The samples were then filtered using filter paper and the filtrate was concentrated in a rotary evaporator. The percentage yield was calculated after which the solid extracts were stored in a freezer pending when it was used.

Qualitative Phytochemical Screening

The chemical tests for the screening of bioactive chemical constituents in the plants that was studied were carried out on the extracts by using standard procedures as described by Sofowora (1993), Trease and Evans, (1989) and Harborne, (1973).

Test for Flavonoids

A measure of 2mL of extracts solution in a test tube was mixed with 5mL of dilute ammonia and 1 mL of concentrated H_2SO_4 was added to the mixture. A yellow colour was positive for flavonoids

Test for Tannins

One millilitre (1mL) of extract (filtrate) in a test tube was heated for five minutes to boil. Thereafter, drops of 15% ferric chloride was added, blue black colouration confirmed the presence of tannins.

Test for Cardiac glycosides

The test for cardiac glycoside was carried out using Killaini's test. One millilitre of extract in a test tube was mixed with 2mL of glacial acetic acid, after which 1 drop of 15% ferric chloride and 1mL of concentrated sulphuric acid was added to the mixture. Brown colouration formed at the interface indicated the presence of cardiac glycosides.

Test for Saponins

The ability of saponins to produce frothing in aqueous solution was used as a screening test for saponins. To 1mL of extract, 5mL of distilled water was added. The mixture was mixed vigorously and observed for frothing.

Test for Steroids

To 1mL of extracts in a test tube was mixed with 2mL of acetic acid and 2mL of concentrated H_2SO_4. Change of colour from violet to blue-green colouration was positive for steroids.

Test for Terpenoids

The test for terpenoids was done using the Salkowski's test. To 1mL of the extract in a test tube was added 2mL chloroform and 3mL concentrated H_2SO_4. Reddish brown colouration at the interface, confirmed the presence of terpenoids.

Test for Alkaloids

Mayer's test was used for alkaloids test. To 1mL of extracts was mixed with 3drops of Mayer's reagent. Cream coloured precipitate formation confirmed the presence of alkaloid. Mayer's reagent was prepared by dissolving 1.3g of HgCl and 5g KI in water and making it up to 100mL with distilled water.

Test for Anthraquinones

Few drops of magnesium acetate were added to the test sample. Pink colour formation was a positive test for anthraquinones.

Test for Phenols

To 2mL of sample in a test tube, few drops of 10% aqueous $FeCl_3$ solution were added. Formation of bluish-green or red colour indicated the presence of phenols.

Test for Coumarins

Few drops of alcoholic NaOH were added to 2mL of the extract solution. Yellow colour confirmed the presence of coumarin.

Test for Quinones

To 2mL of test sample, concentrated NaOH was added. Colour formation indicated the presence of quinones.

Test for Xanthoproteins

To 1ml of test sample, few drops of concentrated HNO_3 was added and mixed very well. After this, 2ml of ammonia solution were added. The formation of red precipitate indicated the presence of xanthoprotein.

Test for Fixed Oil

Little amount of sample was placed between two filter papers. Grease spots indicated the presence of fixed.

Determination of Total Phenolic Content

The total phenolic content was determined using the Folin - Ciocalteau method as described by Cicco *et al.* (2009). Concentrations ranging from 0.2 - 1 mg/mL of gallic acid or extracts, were prepared in methanol. 4.5mL of distilled water was added to 0.5 mL of the extract and mixed with 0.5 mL of a tenfold diluted Folin Ciocalteau reagent. Five milliliters of 7%

sodium carbonate was then added to the tubes and another 2mL of distilled water was added. The mixture was allowed to stand for 90 min at room temperature; absorbance was then read at 760 nm. All determinations were performed in triplicates with gallic acid utilized as the positive control. The total phenolic content was expressed as Gallic Acid Equivalent (GAE).

Determination of Total Flavonoids

The total flavonoid content was determined using the method of Miliauskas *et al.* (2004). 2ml of 2% $AlCl_3$ in ethanol was mixed with 2mL of varying concentrations of the standard (0.1-1.0mg/mL), in methanol. The extract at a concentration of 2ml of 1mg/ml was also mixed with the 2% $AlCl_3$ in ethanol. The absorbance was measured at 420 nm after one hour incubation at room temperature. Similar concentrations of quercetin, the positive control were used. The total flavonoid content was calculated as mg quercetin equivalent /g (QE) of extract.

Estimation of Diphenyl-2-Picryl-Hydrazyl (DPPH) Radical Scavenging Activity

The free radical scavenging capacity of the leaf and flower extracts against 1,1–diphenyl–2–picrylhydrazyl (DPPH) radical was determined by a slightly modified method of Brand-Williams *et al*. (1995). 0.5 mL of 0.3 mM DPPH solution in methanol was added to 2mL of various concentrations (0.2 - 1.0 mg/mL) of the extracts. The reaction tubes were shaken and incubated for 15 min at room temperature in the dark; absorbance read at 517 nm. All tests were performed in triplicate.

Ascorbic acid was used as standard control, with similar concentrations as the test samples prepared. A blank containing 0.5mL of 0.3 mM DPPH and 2mL methanol was prepared and treated as the test samples. The radical scavenging activity was calculated using the following formula: DPPH radical scavenging activity (%) = $[(A_0-A_1)/(A_0)] \times 100$, where A_0 was the absorbance of DPPH radical + methanol; A_1 was the absorbance of DPPH radical + sample extract or standard. The 50% inhibitory concentration value (IC_{50}) was calculated as the effective concentration of the extract that is required to scavenge 50% of the DPPH free radicals.

Statistical Analysis

All analysis were carried out in triplicates and reported as Mean ± Standard deviation. Where necessary, Student t-test was used to test level of significance at $p < 0.05$ between samples.

RESULTS

Phytochemical Screening

Table 1: Phytochemical constituent of methanol leaf and flower extract of *S. longipedunculata*

Constituents	leaf	flower
Alkaloids	-	+
Anthraquinones	-	-
Flavonoids	+	+
Phenols	+	+
Saponins	+	-
Steroids	+	-
Tannins	+	+
Terpenoids	+	+

Key: + = presence, - = not detected

Table 2: Total Flavonoid Content and Total Phenolic Content

Plant part	**Total flavonoids** (mg Quercetin Equivalent/g of extracts)	**Total phenolics** (mg Gallic Acid Equivalent/g of extracts)
Leaf	17.0867 ± 0.268^a	22.73 ± 0.04583^a
Flower	6.0 ± 0.0273^b	18.17 ± 0.017^a

Values are mean ± standard deviation (n = 3). Same superscript indicate no significant difference in each column indicates no significant difference between the plant parts ($p > 0.05$) while different superscript indicates significant difference ($p < 0.05$) between the plant parts.

Table 3: Percentage Inhibition of DPPH radical by ascorbic acid (standard) and methanol leaf and flower extracts of *S. longipedunculata*

Conc (µg/ml)	Ascorbic Acid	Leaf extract	Flower Extracts
5	97.28 ± 0.170	17.19 ± 0.061	21.39 ± 0.058
10	98.90 ± 0.098	19.40 ± 5.684	23.43 ± 0.258
15	99.27 ± 0.088	25.00 ± 1.270	23.94 ± 0.094
20	99.32 ± 0.196	25.40 ± 1.900	24.28 ± 0.221
30	99.49 ± 0.026	28.40 ± 0.170	25.47 ± 0.087
40	99.66 ± 0.098	31.51 ± 0.900	26.15 ± 0.112
50	99.66 ± 0.098	33.65 ± 1.392	31.24 ± 0.091

Values are mean ± standard deviation (n = 3)

DISCUSSION

The percentage yield of the leaf and flower extract obtained from the extraction process carried out at room temperature for 72 hours was 24.43% and 19.88% (w/w) respectively. This is closely similar to the report of Sanusi *et al.,* (2017) who using the same plant leaves sample, reported to have obtained 20.20% (w/w) ethanol extract after soaking for 24 hours. The difference may be due to the different time used for soaking and the different solvents used.

The results for phytochemical screening of the methanol leaf extract of *Securidaca longipedunculata* presented in table 1 above, revealed the presence of some of the phytochemicals screened for. The leaf extract was found to have phenols, tannins, flavonoids, saponins, steroids, and terpenoids while the flower extract had phenols, alkaloids, flavonoids, terpenoids and tannins. These phytochemicals are known to exhibit medicinal as well as physiological activities (Shreshtha *et al*., 2015).

The presence of these phytochemicals like phenolics (known with anticancer activity (Han *et al*., 2007)) in *S. longipedunculata* leaf extracts suggests the reason why the leaves are used locally for the treatment of skin cancer as reported by (Mustapha, 2013). The presence of terpenoids, saponins, tannins and phenolics suggests the reason why the leaves have anti-inflammatory activity as reported by Okoli *et al*., (2006). The antifungal activity reported by Karou *et al*., (2012) could be due to the presence of tannins (which are reported with such activity (Chung *et al*., 1998)), while antibacterial activity (Ndamitso *et al.,* 2013) due to the presence of steroids which are reported with that activity (Epand *et al*., 2007). It is important to note that the presence or absence of a particular phytochemical in a plant part is attributed to the various physiological and biosynthetic reactions taking place inside the plant, the effect of the environment should not be neglected, as the environment always modify them (Shreshtha *et al*., 2015).

The present study revealed that the plant leaf extract has total flavonoid content of 17.0867 mg QE/g of extract while the flower has 6.0 ± 0.0273 mg QE/g of extract. This result revealed that the leaf extract has more flavonoids content than the flower. Compared to *Polygala balansae* which was reported to have 29.20mgQE/g extract (Guiche *et al*., 2015), a plant of the same family with *S. longipedunculata*, the leaf extract can be said to have appreciable flavonoid content. Other plants belonging to the same family; *Polygala sabulosa* showed higher flavonoid content 24.68mgQE/g extract of the aerial part of the plant as compared to the studied plant. *Polygala cyparissia* revealed low flavonoid content 12.08mgQE/g (Brihente *et al*., 2007) extract of the aerial part of the plant compared to *S. longipedunculata.*

Phenols are also important class of phytochemicals. This study revealed the total phenolics content of the methanol leaf and flower extracts of *S. longipedunculata* to be 22.73 ± 0.04583mg GAE/g and 18.17±0.017 mg GAE/g respectively. This is below that of hydro alcoholic extract of *Polygala sabulosa* (64.92mg GAE/g) and *Polygala cyparissia* (33.97mg GAE/g) reported by Brighente *et al*., (2007)

The percentage free radical inhibition presented in table 3 revealed that the methanol leaf and flower extracts of *S. longipedunculata* has antioxidant activity which increases with increase in concentration. The results are closely in line to that presented by Abonyi *et al*., (2015) who though used modified Gyamfi *et al*., (2002) method to determine DPPH radical scavenging activity of methanol leaf extract of *S. longipedunculata* as opposed to the method used in the current study, reported moderate inhibitory activity.

CONCLUSION

Our study have shown that *S. longipedunculata* leaf and flower extracts possess phytochemicals of pharmacological importance. Moreso, both the leaf and flower extracts have been shown to possess potent free radical scavenging and antioxidant activity. We hope that these properties can be further explored in the treatment of oxidative stress related diseases. However, the toxicity status of both extracts still need to be investigated in other to ascertain their safety in consumption.

REFERENCES

Abonyi, O., Uzoegwu, P.N., Ezugwu, A. L., Uroko, R. I., Ani, C.C., Onyemuche, T. N. and Anigbogu J.U (2014). In vitro antioxidant profile of methanol leaf extract of securidaca longipedunculata. *Journal of Dental and Medical Sciences13,(11) 75-81*. doi: 10.9790/0853-131157581

Ahmed D., Khan M.M and Saeed R. (2015). Comparative analysis of phenolics, flavonoids and antioxidant and antibacterial potential of methanolic, hexanic and aqueous extracts from *Adiantum caudatum* leaves: *Antioxidants,4,* 394-409. doi:10.3390/antiox4020394

Baba A. and Malik A.,(2015)**.** Determination of total phenolic and flavonoid content, antimicrobial and antioxidant activity of a root extract of *Arisaema jacquemontii* blume**.** *Journal of Taibah University for Science***,** *9* (4) 449-454. doi.org/10.1016/jtusci.2014.11.001

Banu S. and Cathrine L., (2015). General techniques involved in phytochemical analysis *International Journal of Advanced Research in Chemical Science*, *2* (4) 25-32.

Beslama, A. and Harrar, A. (2016). Free radical scavenging activities and reducing power of two Algerian sahara medicinal plant extracts. *International Journal of Herbal Medicine, 4,* (6) 158-161.

Brand-Williams, W, Cuvelier ME, and Berset C. (1995). Use of free radical method to evaluate antioxidant activity. *Lebensm Wiss Technology*, *28*, 25-30.

Brighente, I.M.C., Dias,M., VerdiL.G. and Pizzolatti M.G (2007) Antioxidant Activity and Total Phenolic Content of Some Brazilian Species. *Pharmaceutical Biology*, *45*, (2), 156–161.doi: 10.1080/13880200601113131

Cicco, N., Lanorte M.T., Paraggio M., Viggiano M. and Lattanzio V. (2009). A reproducible, rapid and inexpensive Folin–Ciocalteu micro-method in determining phenolics of plant methanol extracts, *Microchemical Journa, 91,* 107-110

Chung K., Wong, T., Wei, C., Huang, Y. and Lin Y. (1998). Tannins and human health: A review. Critical reviews in food science and nutrition, 38 (6), 421-464.

Curran K. (2017). The ethno-herbalist review of phytochemicals. Retrieved on August 30, 2018 from www.ethnoherbalist.com/phytochemicals-food-benefits/

Dapar, L.P., Maxwell, C., John, A.C., Noel, W.N., Steven, G.S. and Naanlep, T.M. (2007). The histopathologic effects of *Securidaca longepedunculata* on heart, liver, kidney and lungs of rats. *African Journal of Biotechnology, 6*(5): 591-595.

Epand, R., Savage P. and Epand R. (2007). Bacterial lipid composition and the antimicrobial efficacy of cationic steroid compounds (Ceragenins), *Biochimica Et Biophysica Acta (BBA) - Biomembranes*, *1768(10),* 2500-2509. doi:10.1016/j.bbamem.2007.05.023

Garcia E., Oldoni T., Alencar S., Reis A., Loguecio D. and Grand R. (2012). Antioxidant activity by DPPH assay of potential solutions to be applied on bleached teeth. *Brazil Dental Journal*, *23* (1): 22-27

Guiche E.R., Tahrouch, S., Amri, O., Mehrach, K.E. and Hatimie, A (2015). Antioxidant activity and total phenolic and flavonoid contents of 30 medicinal and aromatic plants located in the south of morocco. *International Journal of New Technology and Research*, 1 (3) 7-11

Gyamfi, M.A and Aniya, Y.(2002) Antioxidant properties of Thonningianin A, isolated from the African medicinal herb, *Thonningia sanguine*. Biochemical Pharmacology, 63: 1725–37.

Halliwell, B. and Guteridge, J.M.C. (1989). *Free Radicals in Biology and Medicine*, (2nd ed.) Clarendon Press: Oxford, UK.

Han, X., Shen, T., and Lou, H. (2007). Dietary polyphenols and their biological significance, *Snternational Journal of Medical Sciences*, *8*(9), 950-988. doi:10.3390/i8090950

Harborne, J., (1973). *Phytochemical methods* (pp. 49-88), London: Chapman and Hall,.

Iwu, M. M. (1986). African ethnomedicine. CECTA Nigeria Limited, Enugu, pp 34-37.

Junaidu, S., Shehu, K., Aliero, A.A., Bawa, J.A. and Suleiman, I. (2014). Evaluation of antifungal and phytochemical properties of Violet tree (*Securidaca longepedunculata* Fres). *Global Journal of Science Frontier Research. 14*(5).

Karou, S.D., Tchacondo, T., Tchibozo, M.A.D., Anani, K., Ouattara, L., Simpore, J. and de Sousa, C., (2012). Screening of Togolese medicinal plants for few pharmacological properties. *Journal ofPharmacognosy Research, 4*, 116–122.

Kirby, G.C. (1996). Medicinal plants and the control of parasites. *Transactions of the Royal Society of Tropical Medicine and Hygiene journal*, *90*, 605-609

Miliauskas, G, Venskutonis, P.R. and Van Beek, T. A (2004). Screening of radical scavenging activity of some medicinal and aromatic plant extracts. *Food Chemistry*, *85*, 231-237.

Molyneux, R.J., Lee, S.T., Gardner, D. R., Panter, K. E and James, L. F (2007). "Phytochemicals: The good, the bad and the ugly?". Phytochemistry. 68 (22–24), 2973–85. *doi*:*10.1016/j.phytochem.2007.09.00*

Molyneux, A.A., (2013). Ethno-medicinal field study of anti-fertility medicinal plants used by the local people in Keffi local government, Nasarawa State, Nigeria. *International Journal of Medicinal Plants Research* 2, 215–218.

Mustapha, A.A., (2013). Ethno-medico-botanical uses of *Securidaca longipedunculata* (Fresen) (Family Polygalaceae) from Keffi local government, Nasarawa State, Nigeria. *Journal of Natural Remedies,13*, 133–137.

Ndamitso, M. M., Mohammed, A., Jimoh, T.O., Idris, S., Oyeleke, S.B. and Etsuyangkpa, M.B., (2013). Phytochemical and antibacterial activity of *Securidaca longepedunculata* on selected pathogens. *Africa Journal of Microbiology Research*, 7:5 652–5656.

Njoya, M., Muvera, M., Mkounga, P., Nkengfac, E. and McGaw, J. (2017). Phytochemical analysis with free radical scavenging, nitric oxide inhibition and anti-proliferative activity of *Sarcocephalus pobeguinii* extract; *Complementary and Alternative Medicine, 7,* 119. doi:10.1186/s12906-017-1712-5

Okoli, C. O., Akah P. A and Ezugworie, U (2006). Anti-Inflammatory activity of extracts of root bark of *Securidaca longipedunculata* Fres (Polygalaceae) *African. Journal of Traditional, Complementary and Alternative Medicines, 3* (1), 54 – 63

Oladeji, O. (2016). The characteristics and roles of medicinal plants: Some important medicinal plants in Nigeria. *Natural products Indian Journal12* (3).

Orwa, C., Mutua, A., Kindt, R., Jamnadas, R. and Simon, A. (2009). Agroforestry Database: A tree reference selection guide version 4.0. Retrieved on August 21, 2018 from www.worldagroforestry.org/af/treedb/

Padmanabhan P. and Jougle N. (2012). Evaluation of DPPH radical scavenging activity and reduce,ng power of four selected medicinal plants and combinations. *International Journal of Pharmaceutical Sciences and Drug Research.4*, (2) 143-146.

Paul, A., Barma K., Roy, K. and Ray, S. (2018). A study of antimicrobial activity and medicinal value of *Adhotoda vasica, Centella asiatica, Paederia foetida, Nytanthes abhor-tri Ocimum temiflorum. International Journal of Current Microbiology and Applied Sciences, 7* (5), 1406-1413. doi.org/10..20546/ijcmas.2018.705.167

Sanusi, J., Bawa, J.A., Aghemwenhio, I. S., Rabi'u, Z. S., Sani, M. G and Liadi, S. (2017). Efficacy of S*ecuridaca Longepedunculata* on the Parameters of Blood Glucose Level and Pulse Rate of Envenomed Albino Rats. *International Journal of Pharmacological Science Research*, 7(12): 4805-11. doi: 10.13040/IJPSR.0975-8232.7 (12).4805-11.

Shirazi, U., Khattak, K., Shukri M., and Nasiriq A. (2014). Determination of total phenolic, flavonoid content and free radical scavenging activity of common herbs and spices. *Journal of Pharmacognosy and Phytochemistry*, 3 (3), 104-108.

Shemishere U.B., Taiwo, J.E., Erhunse, N. and Omoregie, E. S. (2018). Comparative study on the Proximate and Nutritional Composition of *Musanga cercropioides and Maesobotrya barteri* leaves. *J.Appl. Sci. Environ. Manage.* 22(2) 287- 291. DOI: https://dx.doi.org/10.4314/jasem.v22i2.22

Shrestha P., Adhikari Lamichhane S. and Govinda S. B., (2015). Phytochemical screening of the medicinal plants of Nepal. *Journal of Environmental Science, Toxicology and Food Technology, 1,* (6), 11-17

Sofowora, A. (1993).*Medicinal Plants And traditional Medicine In Africa.* Ibadan Nigeria: Spectrum Books, pp. 191-289

Taiwo, J.E., Shemishere U.B. and Omoregie, E. S. (2017). Phytochemical constituents and *In Vitro* Antioxidant Activities of *Melastomastrum capitatum* and *Solanum macrocarpon* leaf extracts. *Nig. J. Life Sci.* 7(2): 128-149.

Tiwari P., Kumar B., Kaur M., Kaur G. and Kaur H. (2011). Phytochemical screening and extraction. *Internationale pharmaceutica sciencia*. *1* (1). Retrieved on September 20, 2018 from http://www.ipharmsciencia.com

Trease, G. and Evans, W. (1989).*Pharmacognosy* (11th ed.,) pp. 45-50, London: Bailliere.

World Health Organization (1993). Guidelines on conservation of medicinal plants. *International Union for Conservation of Nature and Natural Resources4* retrieved on August 30, 2018 from www.apps.who.int/medicinedocs/documents/s7150e.pdf

Yadav M., Chattergi S., Gupta K. and Watal G. (2014). Preliminary phytochemical screening of six medicinal plants used in traditional medicine. *International Journal of Pharmacy and Pharmaceutical Sciences, 6* (5).