Pharmacognostic profile of root of *Cryptolepis sanguinolenta* (lindl.) Schlechter

***Odoh, U. E. and Akwuaka, C. I.**

Department of Pharmacognosy and Environmental Medicine, Faculty of Pharmaceutical Sciences, University of Nigeria, Nsukka.

ABSTRACT

Medicinal plants are traditionally found to be useful for many ailments. The present study highlights the pharmacognostical as well as phytochemical studies including parameters such as macroscopic, microscopic characters, physicochemical evaluation, chemomicroscopy and preliminary phytochemical studies of the root of *Cryptolepis sanguinolenta*. The morphological studies shows a root light to medium brown in color with hard and brittle texture, prisms of calcium oxalate crystals, sclereids and parenchyma cells. The transverse section showed parenchyma cells, vascular cambium, ray lines and phelloderm. Physico-chemical standards and their percentage values w/w were found to be: total ash (14.02 ± 0.12), acid insoluble ash (5.08 ± 0.18, %), water soluble ash (4.02 ± 0.27 %), sulphated ash (4.26 ± 0.11 %), alcohol soluble extractive (6.20 ± 0.45 %), water soluble extractive (28.40 ± 0.75 %) and moisture content (6.80 ± 0.25 %). Chemomicroscopical investigation revealed presence of lignin, tannin, oils, cellulose and calcium oxalate. Phytochemical analysis of the root revealed the presence of carbohydrates, alkaloids, glycosides, saponins, resins, proteins, steroids and terpenoids. These findings will help in identification, standardization of the root of *Cryptolepis sanguinolenta* (Lindl.) Schlechter and also distinguish it from its adulterants.

Keywords: *Cryptolepis sanguinolenta*, chemomicroscopic analysis, phytochemical analysis, physiccochemical standards.

INTRODUCTION

Herbal medicine is the oldest form of healthcare known to mankind. Throughout history, herbs had been used by all cultures as sources of medicine. Primitive man observed and appreciated the great diversity of plants available to him. The plants provided food, clothing, shelter and medicine. The use of plants as medicine is older than recorded history. Traditional and folklore medicine which was bequeathed from generation to generation is rich in domestic recipes and communal practice. About 1400 herbal preparations are widely used according to a recent survey in Member States of the European Union[1].

Cryptolepis sanguinolenta (Lindl.) Schlechter (Fam: Asclepiadaceae) the plant of interest is a shrub that grows in the rainforest and distributed throughout the West Coast of Africa. It is known as yellow dye root, *Akpaoku* (Igbo), *Gangamau* (Hausa), *Delboi* (Fulani).It is a twining, scrambling shrub, with characteristic thin stems and tuberous root stock. The leaves are opposite, thinly herbaceous, elliptic-oblong to ovate or lanceolate in shape up to 7 cm long and 3 cm wide. The margin is entire, the apex is curved and acuminate and the base is symmetrical and obtuse or rounded. The midrib projects prominently on the lower side and is pinnately nerved. The dried leaves have a slight bitter taste. It is used in hypertension, microbial infections, fever and stomach ache[2]. Clinical studies have shown that extract of the plant produced cures in patients with the concomitant elimination of parasitaemia in the blood[2]. The principal constituent of *Cryptolepis sanguinolenta* (Lindl) Schlechter is the indoloquinoline alkaloid, cryptolepine which occurs at a yield of 0.52 % in the roots, 0.48 % in the stem and 1.03 % in the leaves. The compound occurs with related bases and their derivatives. Cryptolepine has been found to produce antihyperglycemic and cytotoxic effects through GC-rich DNA sequence intercalation that provides basis for the design of new anticancer drug[3]. Another indolequinoline alkaloid quindoline synthesized from cryptolepine[4]. Dwuma –Badu *et al*[5] reported the isolation of quindoline from the

***Address for correspondence:**
E-mail: estellamaris5@yahoo.com

DOI: 10.5530/pj.2012.28.8

roots of *Cryptolepis sanguinolenta*. Three new indole alkaloids hydroxycryptolepine, cryptoheptine and cryptoquindoline have been isolated and structurally elucidated as well[6]. Recently, a tetracyclic alkaloid, isocryptolepine has been isolated from the roots of *Cryptolepis sanguinolenta*[7]. Neocryptolepine and biscryptolepine (11-cryptolepin-11-y1)-cryptolepine), were isolated from the root bark of this plant and their structures elucidated[8, 9]. This study aims at establishing the macroscopic, microscopic, chemomicroscopic and physicochemical profiles of the root of *Cryptolepis sanguinolenta* that would be useful in preparing a monograph for identification of the plant.

MATERIALS AND METHODS

Collection of plant materials

The roots of *Cryptolepis sanguinolenta* were collected in February 2010 from Nsukka, Enugu State, Nigeria. It was identified and authenticated by Mr. A. Ozioko, a taxonomist with International Centre for Ethnomedicine and Drug Development (INTERCEDD) Nsukka, Enugu State and a Voucher Specimen (UN/PCOG/010/401) deposited in the Herbarium of Department of Pharmacognosy and Environmental Medicine, University of Nigeria, Nsukka.

Plant sample preparation

Fresh roots were collected, washed and excess water allowed to drain off. Representative samples were kept for examination while the rest were dried completely. They were pulverized and the powdered sample stored in airtight container for use in the microscopic, chemomicroscopic and phytochemical studies. Transverse sections were cut from the representative sample using sledge microtone. The sections were preserved in 70 % ethanol until needed for studies.

Macroscopical Examination

The macroscopical features of the fresh root were examined using the methods described by Evans[10].

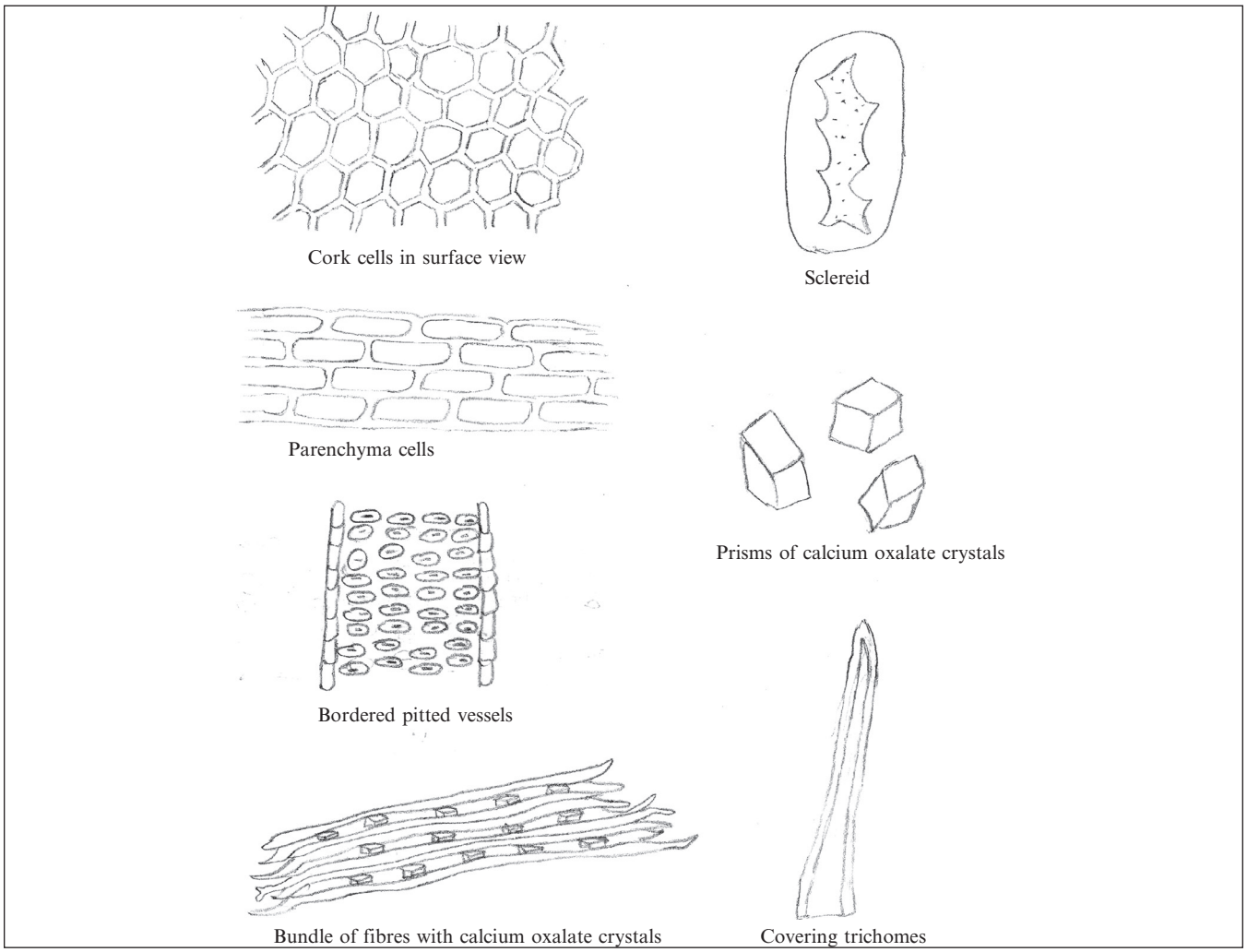
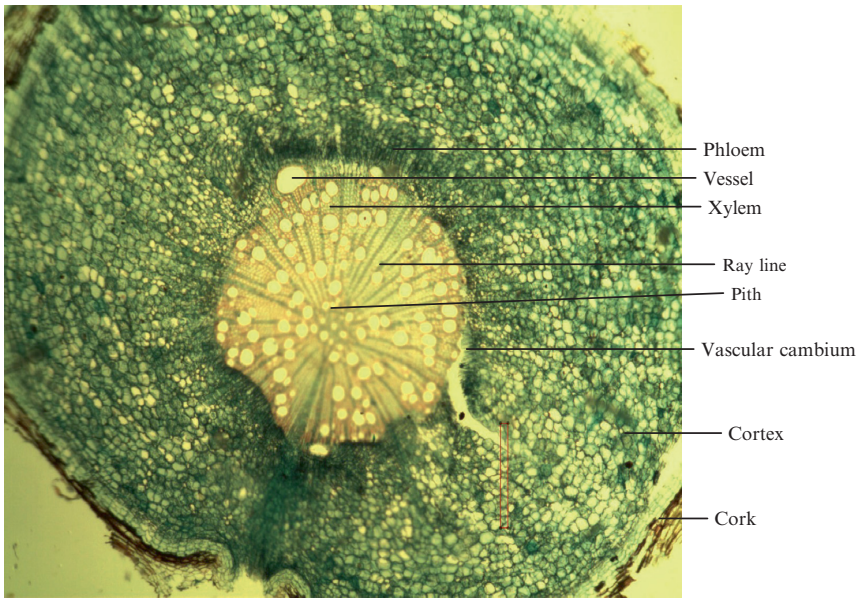


Figure 1: Microscopical features of root of *Cryptolepis sanquinolenta*



Figrue 2: Transverse section of the root of *Cryptolepis sanguinolenta*

Microscopical examination

Anatomical sections of the fresh root and powdered samples were prepared for the microscopic studies. The staining was done using standard laboratory methods[10 - 11].

Phytochemical studies

The preliminary phytochemical screening of the root powder was performed following standard qualitative chemical tests[10, 12] in other to detect the presence or absence of major secondary plant metabolites of pharmacognostic importance. The classes of phytoconstituents tested include; alkaloids, tannins, flavonoids, saponins, glycosides, proteins, fats and oils, steroids and carbohydrates.

Determination of some physicochemical standards

The water extractive and alcohol extractive, total ash, acid insoluble ash, water soluble ash and sulphated ash were determined as described by Evans[10]. The moisture content of the root powder was determined by loss on drying method[13].

Chemomicroscopic examination

Chemomicroscopical examination was carried out to detect the presence or absence of various chemical compounds such as starch, cellulose, tannins, and lignin, fat and oil, mucilage and calcium oxalate crystals[10].

RESULT

Macroscopical examination

The root surface of *Cryptolepis sanguinolenta* is light to medium brown in colour. It is hard and brittle, with longitudinal ridges apparent in the dried samples. The roots are branched with little or no rootlets. The root breaks with a short fracture exposing a smooth surface which is yellow in colour and has a bitter taste.

Microscopic examination

The microscopical analysis of the root revealed the following features: cork with thin walled cells, polygonal and elongated in surface view, sclereids thickened with branched pits showing distinct strations. A unicellular covering trichomes, bundle of fibres with calcium oxalate crystals and fibres with bordered pitted vessels. There were also parenchyma cells.

Phytochemical studies

Phytochemical screening of the root powder revealed the presence of alkaloids, reducing sugars, flavonoids, saponins, glycosides, proteins, steroids, terpenoids and carbohydrates (Table 1).

Physicochemical standards

The water extractive and alcohol extractive, total ash, acid insoluble ash, water soluble ash and sulphated ash were shown in Table 2.

Chemo-microscopical Examination

The chemomicroscopy of *Cryptolepis sanguinolenta* revealed the presence of cellulose, starch, suberized wall, fibres, secretory cells and ducts and calcium oxalate crystals (Table 3).

DISCUSSION

The identification and evaluation of the root of *Cryptolepis sanguinolenta* have been carried out and the various

characteristics and features associated with it duely determined by the various analysis and tests performed. The macroscopic examination reveals the physical appearance of the root, which can be seen with the naked eyes. This however gives an idea of the part and cannot be relied solely for the identification of the root of the plant. The microscopic examination gives hints about the characteristic features that could be found in different morphological parts of plants. These features and their arrangements are not always the same in all morphological parts. The presence of abundant prisms of calcium oxalate which indicates the presence of the calcium salt of oxalic acid that is present usually at about 1.0 % in plants[10], the covering trichomes can be used as identifying characters of the plant. The results obtained for ash values, which are of tremendous importance in quality control is used to detect foreign organic matter and detection of adulteration of sand or earth[14] were within the British Pharmacopoeia (BP) specification. The ash values obtained were adequate within the limits of experimental error since the total ash, acid-insoluble, water soluble ash and sulphated ash were determined were within the BP specification. The extractive values are moderate. The moisture content of the crude drug is not high (fall within the limit of the general requirement 8 – 14 %), indicating less probability of microbial degradation. Excess moisture in crude drug may lead to the breakdown of important constituent and the growth of microorganisms especially during storage of drug[15].

Table 1: Phytochemical analysis of the root of *Cryptolepis sanguinolenta*

Constituent	Inference
Carbohydrates	++
Reducing sugars	+
Alkaloids	+++
Glycosides	+
Saponins	+
Tannins	+
Flavonoids	+
Resins	+
Proteins	+
Fats and Oils	–
Steroids	+
Terpenoids	++
Acidic compounds	–

Key: + = slightly present; ++ = moderately present; +++ = highly present; – = Absent

Table 2: Qualitative evaluation of the root of *Cryptolepis sanguinolenta*

Parameter	Value % (w/w)
Total ash	14.02 ± 0.12
Acid insoluble ash	5.08 ± 0.18
Water soluble ash	4.02 ± 0.27
Sulphated ash	4.26 ± 0.11
Alcohol soluble extractive	6.20 ± 0.45
Water soluble extractive	28.40 ± 0.75
Moisture content	6.80 ± 0.25

Values are Mean ± SEM, n = 3.

CONCLUSION

The results obtained from this study can serve as diagnostic parameters for proper identification as well as preparation of a monograph on *Cryptolepis sanguinolenta*

ACKNOWLEDGEMENT

The authors thank Department of Pharmacognosy and Environmental Medicine and Department of Botany, both in the University of Nigeria, Nsukka for providing the facilities for the research.

REFERENCES

1. Herbal Gram. Cryptolepis: An African Traditional Medicine that provides Hope for Malaria Victims. The Journal of the American Botanical Council. 2003, 60: 54 – 59, 67.
2. Iwu MM. Handbook of African Medicinal Plants, CRC Press Inc., London, 1993.
3. Bierer DE, Fort MDM, Mendez CD, Luo J, Imbacha PA, Dubenko LO, et al. Ethnobotanical –directed discovery of the antihyperglycaemic properties of cryptolepine : its isolation from *Cryptolepis sanquilolenta*, synthesis and in vitro and invivo activities. J. Med. Chem.1998; 41 (6): 894 – 901.

Table 3: Chemomicroscopy of the root of Cryptolepis sanguinolenta

Test reagent	Observation	Inference
Iodinated zinc chloride solution	Blue colour observe d on epidermal cells	Cellulose present
Iodinated zinc chloride solution	Blue-black coloration observed in the xylem vessels	Starch present
Iodinated zinc chloride solution	Brown coloration observed	Suberized wall present
Phloroglucinol + Conc. HCl	No red colour observed in xylem vessels and phloem fibres	Lignin absent
80% H_2SO4	Bright crystals of calcium oxalate dissolved	Calcium oxalate crystals present
Sudan III	Pink-red coloration observed	]Fibres present
Ferric chloride solution	No greenish colour in parenchyma cells	Tannins absent
Sudan IV	No pink colour observed	Lipid absent
Picric acid solution	Yellow coloration observed	Secretory cells and ducts present

4. Gellert E, Raymond-Hamet R, Sehlitter E. Die Konstitution des alkaloids Cryptolepine. Helvetica Chimica Acta. 195; 34: 642 – 651.
5. Alexandra P, Elsa TG, Peter JH. An indolequinoline alkaloid quindoline synthesized from cryptolepine. Journal of Natural Products.1995; 58 (10): 1485 –1491.
6. Dwuma-Badu D, Ayim JS, Fiagbe NI, Knapp JE, Schiff PL, Slatkin T. Constiteunts of West African Medicinal Plants XX:Quindoline from *Cryptolepis sanquinolenta*. J. Pharm.Sci.1978; 67 (3): 433 – 444.
7. Dubovitskii SV, Radchenko OS, Novikov VL. Synthesis of isocryptolepine, an alkaloid from *Cryptolepis sanguinolenta*. Russian Chemical Bullentino, 1996, Vol. 45.
8. Cimanga K, Pieters L, Claeys M,Vanden Berghe D, Vlietinck AJ. Biological activities of cryptolepine, an alkaloid from *Cryptolepis sanquinolenta*. Planta Medica. 1996; 57 (2): A98 A99.
9. Cimanga K, De Bruyne, Pieters L, Claeys M, Vlietinck AJ. Anew alkaloid from *Cryptolepis sanquinolenta*. Tetrahedron.Letters. 1996;Volume 37, Issue 10, 1703 – 1706.
10. Evans WC. Trease and Evans Pharmacognosy, 14th ed.W. B. Saunders Company Ltd: London; 1996.
11. Brain KR, Turner TD. The Practical Evaluation of Phytopharmaceutical. Wright Scienctechnica: Bristol; 1975.
12. Harbourne JB. Textbook of Phytochemical Methods and Guide to Mordern Technique of Plant Analysis. 2nd ed. Chapman and Hall Ltd: London;1973.
13. African Pharmacopoeia. Vol.2, General Methods of Analysis. (OAU/ STRC) Lagos; 1986.
14. Kunle OF, Jegede IA, Ibrahim H, Okogun JI. Pharmacognostic studies on the leaf of *Lippia multiflora* Moidenke. Journal of Phytomedicine and Therapeutics. 2002; 7, (1 & 2): 40 – 45.
15. Adesina GO, Onaolapo JA, Ehinmidu JO, Odama LE. Anti-microbial stability of *Alchornea cordifolia* leaf extract. International Conference of Research and Development, Ghana. 2008.