African Journal of Pharmaceutical Research & Development

Vol. 4 No.2 pp.49-54 (2012)

Cryptolepis sanguinolenta Root Tablets: Effect of Binder Type and Concentration on the Tablet Properties

[1]Chime SA., [2]Brown SA., [1]Odilora OC, [3]Chimene M, [3]Anele N and [1]Godswill C Onunkwo

[1]Department of Pharmaceutical Technology and Industrial Pharmacy, University of Nigeria, Nsukka 410001, Nigeria
[2]Department of Pharmaceutics and Pharmaceutical Technology, University of Port Harcourt, Nigeria
[3]International Center for Ethnomedicine and Drug Development Nsukka, Nigeria

ABSTRACT

The objectives of the study were to formulate *Cryptolepis sanguinolenta* root powder into tablets and to evaluate the effect of different binders and binder concentrations on the properties of tablets. The tablets were formulated by the wet granulation method using gelatin and sodium carboxymethyl cellulose (SCMC) as binders at concentrations of 2%, 4%, 6% and 8%w/w. The tablets were evaluated using the relevant official and unofficial tests. Also the phytochemistry of the powdered root extract of *C. sanguinolenta* was evaluated. Phytochemical analysis showed that *C. sanguinolenta* root contains alkaloids, terpenoids, steroids, proteins, carbohydrate, resins, reducing sugars and glycosides. Tablets formulated with SCMC significantly exhibited higher disintegration times than those formulated with gelatin ($p<0.05$).Tablets hardness ranged from 3.51 ± 0.12 to 5.02 ± 0.10 kgf for A1 and A4 tablets formulated with 2 and 8% gelatin and 2.00 ± 0.11 to 5.00 ± 0.17 kgf for B1 and B4 tablets formulated with 2 and 8% SCMC. All the tablet batches exhibited friability of < 1% ($p<0.05$). Therefore the powdered root of *C. sanguinolenta* could be formulated as normal release tablets using gelatin and SCMC in order to standardize the preparation and also enhance patient's compliance.

KEYWORDS: *Cryptolepis sanguinolenta,* antimalarial,antidiabetic, tablets, alkaloids

INTRODUCTION

In recent times, focus on plant research has increased all over the world and a lot of evidence has been collected to show immense potential of medicinal plants used in various traditional systems [1]. Plants may become the bases for the development of new medicines or they may be used as phytomedicines for the treatment of diseases [2]. It is estimated that today, plant materials are present in, or have provided the models for 50% Western drugs [3]. In view of the widespread use of herbal products, important technical aspects such as standardization and quality control will be of immense benefit in order to enhance their efficacy and improve patient's compliance [4].

Cryptolepis sanguinolenta is a thin-stemmed twining and scrambling shrub up to 8 m long, containing yellow-orange juice which becomes red upon drying. It is a member of the family Apocynaceae (subfamily: Periplocoideae). The plant is native to West Africa and is found in countries like Ghana, Nigeria, Cote d'Ivoire, Guinea, Guinea-Bissau, Mali, Senegal, Sierra Leone, Angola, Congo, Uganda, and Cameroon [5]. It is a medicinal plant used by some traditional herbalist in the treatment of fever, urinary, and upper respiratory tract infections [5]. The use of this plant as a medical therapy has increased as it has been proposed that the root and leave extracts have hypotensive, antipyretic, anti-inflammatory, antidiarrhoeal, *in vitro* antibacterial and antimalarial effects [6]. It is commonly called *nibima*, *Kadze*, *gangamau*o, or yellow-die root. It is called *paran pupa* in the Yoruba- speaking areas of Nigeria [5]. Studies have documented the antidiabetic potentials of *Cryptolepis sanguinolenta* [6-8]. Crude extracts of *C. sanguinolenta* and their fractions, as well asindoquinoline alkaloids isolated from the plant, havebeen shown to have activity against *Plasmodium falciparum* both *in vitro* and *in vivo* [9-12]. In addition tostudies indicating anti-plasmodial effect, extracts of *C. sanguinolenta*

*Author for correspondence; *E-mail address:* emmymarachi@yahoo.com;salome.chime@unn.edu.ng, Tel.: + 2348061355342

roothave been shown to have anti-microbial [13-14], and anti-inflammatory activities [10]. Extracts from various morphological parts of *Cryptolepis sanguinolenta* are widely used traditionally in folklore medicine in many parts of the world for the management, control, and treatment of diabetes mellitus. The hypoglycemic activity of *Cryptolepis sanguinolenta* is associated with its influence to reduce intestinal glucose absorption and transport [10]. The biological activities of its different morphological parts have been attributed to its alkaloid constituents. Cryptolepine, an alkaloid, is the major bioactive principle of the plant [15]. In addition to cryptolepine, other minor alkaloids and their salts that have been isolated include the hydrochloride and the 11-hydroxy derivatives of cryptolepine, iso- and neo-cryptolepine, quindoline, biscryptolepine, cryptoquindoline, cryptospirolepine, cryptosanguinolentine, cryptotakienine, and cryptomisrine [16-19].

Tablet dosage forms are the most popular and preferred drug delivery systems in terms of precision of unit dose, low cost, patient compliance, and good physical and chemical stability [20]. Therefore the objectives of the work were to formulate the dried powdered root of *Cryptolepis sanguinolenta*intotablets and to study the effect of binders and binder concentration on the properties of the tablets.

MATERIALS AND METHODS

Materials

Maize starch, sodium carboxymethyl cellulose, gelatin, ethanol (BDH, England), lactose (Merck, Germany), magnesium stearate (May and Baker, England), distilled water (Lion water, Nsukka, Nigeria). *Cryptolepis sanguinolenta* root powder was obtained from a batch processed in our laboratory. All other reagents and solvents were analytical grade and were used as supplied.

Collection and authentication of plant material

Cryptolepis sanguinolenta roots were collected from the Army Barrack's field along Edem road Nsukka, Enugu State, Nigeria in the month of June, 2011. The plant material was authenticated by Mr. A.O. Ozioko, a consultant taxonomist with the International Center for Ethnomedicine and Drug Development (InterCEDD) Nsukka. The voucher specimen of the plant was deposited in the herbarium of the Department of Pharmacognosy and Environmental Medicines, University of Nigeria, Nsukka.

Processing of Cryptolepis sanguinolenta root powder

Cryptolepis sanguinolenta root were washed thoroughly in water, cut into tiny pieces and then dried The stem bark was washed, cut into small sizes and then dried under a shed below 38 ^{o}C. The dried roots were milled severally using a hammer mill and then sieved with a 120 µm mesh sieve.

Phytochemical Screening

Phytochemical tests were carried out on the powdered extract for the presence of alkaloids, tannins, saponins, flavonoids, resins, fats and oils, steroids, glycosides, terpenoids, acidic compounds, carbohydrates, reducing sugars and proteins. The tests were carried out using standard procedure [21-22].

Table 1: Composition of *Cryptolepis sanguinolenta* tablet

Batch	*C. sanguinolenta*root powder (mg)	Gelatin (mg)	SCMC (mg)	Maize starch (mg)	Magnesium stearate (mg)	Lactose qs (mg)
A1	50.0	6.0	-	15.0	3.0	300.0
A2	50.0	12.0	-	15.0	3.0	300.0
A3	50.0	18.0	-	15.0	3.0	300.0
A4	50.0	24.0	-	15.0	3.0	300.0
B1	50.0	-	6.0	15.0	3.0	300.0
B2	50.0	-	12.0	15.0	3.0	300.0
B3	50.0	-	18.0	15.0	3.0	300.0
B4	50.0	-	24.0	15.0	3.0	300.0

Key: A1- A4 contain 2, 4, 6 and 8%w/w gelatin, B1 – B4 contain 2, 4, 6, and 8%w/w SCMC, SCMC: sodium carboxymethyl cellulose.

Preparation of granules

Granules were prepared by wet granulation method using gelatin and SCMC respectively as binders at concentrations of 2%, 4%, 6% and 8%w/w. Details of granulation are given in Table I. Lactose and maize starch BP (10% w/w) were dried and mixed for 10 min in a tumbler mixer with the powdered root of *Cryptolepis sanguinolenta*. The powder mixtures were moistened with the appropriate amount of binder solution. The homogeneous wet mass was then screened through a 1.7 mm sieve and the wet granules dried in a hot air oven at 55°C for 1 h. Thereafter, the dried granules were screened through a 1.0 mm sieve.

Preparation of tablets

Initially granules were treated with lubricant i.e. magnesium stearate. Tablets were prepared by compressing the lubricated granules at 46-48 kgf using a 9.0 mm punch and die set fitted into an automated F3 Manesty Single Punch tabletting machine.

EVALUATION OF TABLETS

Disintegration time test

Disintegration time test was conducted using an Erweka ZT 120 basket and rack assembly and 0.1 N HCl maintained at 37.0 ± 1.0 ^{o}C as the disintegration medium. Ten tablets from each batch were used for the test and the procedure being as stipulated in the BP, 2009 for normal release tablets [23].

Uniformity of weight

Twenty tablets were randomly selected from each batch. The tablets were weighed individually using an electronic balance (Ohaus Adventurer, China) and the individual weights recorded. The mean weight, standard deviation and percentage deviation were calculated [23].

Tablet friability test

Twenty tablets were randomly selected from each batch of the tablet. The tablets were dedusted and weighed. The tablets were placed into the drum of the friabilator (Erweka GmbH, Germany) and rotated at 25 rpm for 4 min. The tablets were removed from the friabilator, dedusted and reweighed. The friability result was expressed as loss of mass expressed as a percentage of the initial mass [23]. The abrasion resistance B was calculated from the equation below:

$$B = 100\left[1-\frac{W}{W_o}\right] \quad \text{----------(1)}$$

where W_o and W are the initial weight and final weight of the tablets respectively.

Hardness/Crushing Strength Test

This test was carried out using a Monsanto-stokes hardness tester. Ten tablets from each batch were randomly selected. Each tablet was placed between the jaws of the hardness tester and force was applied by adjusting the knob of tester until the tablet integrity failed. The results were recorded in kgf.

Statistical analysis

Statistical analysis was carried out using SPSS version 14.0 (SPSS Inc. Chicago, IL.USA). All values are expressed as mean ± SD. Differences between means were assessed by a two-tailed student's t-test. $P<0.05$ was considered statistically significant.

RESULTS

Phytochemical constituents

Results of phytochemical analysis of *Cryptolepis sanguinolenta* root extract presented in Table 2 revealed the presence of alkaloids, terpenoids, steroids, proteins, carbohydrate, resins, reducing sugars and glycosides in substantial quantities. However, tannins, saponins, flavonoids and acidic compounds were not found in the plant root.

Table 2: Phytochemical constituents of *C. sanguinolenta* roots extract

Phytochemical constituents	Remarks
Alkaloids	+++
Carbohydrates	++
Saponins	-
Reducing sugars	+++
Steroids	+++
Tannins	-
Glycosides	+++
Proteins	+++
Flavonoids	-
Resins	+++
Fats and oils	-
Acid compounds	-
Terpenoids	+++

Key: +++ High concentration, ++ moderate concentration, - absent

TABLET PROPERTIES

Weight uniformity

The results of tablets weight uniformity test are presented in Table 3 and show that*C. sanguinolenta* tablets exhibited weights that ranged from 303.05 ± 4.43 to 308.70 ± 2.49 mg for batches A4 and A3 formulated with 8 and 6% gelatin and 266.25 ± 11.32 to 302.70 ± 2.66 mg for tablets B4 and B3 formulated with 4 and 3% SCMC. Batches A1-A4 formulated with 2, 4, 6, and 8% gelatin exhibited lower percent variations than those formulated SCMC (B1-B4) as shown in Table 3.

Disintegration time

The results of the disintegration time of *C. sanguinolenta* tablets are shown in Table 3. The tablets exhibited disintegration time range of 9.00 ± 0.05 to 20.50 ± 0.03 min for A1 and A4 tablets formulated with 2 and 8% gelatin and 10.00 ± 0.10 to 25.00 ± 0.11 min for tablets formulated with 2 and 8% SCMC (B1 and B4).

Tablets hardness

The results of tablets hardness are presented in Table 3 and show that tablets formulated with gelatin exhibited higher hardness values than those formulated with SCMC. Tablets hardness ranged from 3.51 ± 0.12 to 5.02 ± 0.10 kgf for A1 and A4 tablets formulated with 2 and 8% gelatin and 2.00 ± 0.11 to 5.00 ± 0.17 kgf for B1 and B4 tablets formulated with 2 and 8% SCMC.

Table 3: Properties of *C. sanguinolenta* root extract tablets

Batch	Weight (mg ± CV)*	Hardness (kgf ± SD)[a]	Disintegration time (min ± SD)[a]	Friability (%)*
A1(2% gelatin)	305.30 ± 3.07	3.51 ± 0.12	9.00 ± 0.05	1.04
A2 (4% gelatin)	302.55 ± 3.48	4.16 ± 0.15	15.00 ± 0.07	0.89
A3 (6% gelatin)	308.70 ± 2.49	4.65 ± 0.07	19.00 ± 0.05	1.09
A4 (8% gelatin)	303.05 ± 4.43	5.02 ± 0.10	20.50 ± 0.03	0.78
B1 (2% SCMC)	290.85 ± 2.85	2.00 ± 0.11	10.00 ± 0.10	0.74
B2(4% SCMC)	283.45 ± 7.40	2.42 ± 0.12	19.00 ± 0.05	1.27
B3 (6% SCMC)	302.70 ± 2.66	3.93 ± 0.10	20.80 ± 0.07	1.21
B4 (8% SCMC)	266.25 ± 11.32	5.00 ± 0.17	25.00 ± 0.11	0.99

***Mean for 20 tablets, [a]Mean for 10 tablets ± SD, CV: coefficient of variation, SD: standard deviation, A1-A4 contain 2, 4, 6 and 8%w/w gelatin, B1 – B4 contain 2, 4, 6, and 8%w/w SCMC; SCMC: sodium carboxymethyl cellulose.**

CONCLUSION

The extracts have different classes of compounds which are known to complex or trap metal ion, thereby inhibiting chain of reactions capable of being induced by metal ions.

The methanol extract (F2) and the seed powder are rich in antioxidant vitamins, though the concentration in 100 g of the samples was more in the seed powder. In fact, many medicinal plants contain large amounts of antioxidants such as polyphenols. Many of these phytochemicals possess significant antioxidant capacities that are associated with lower occurrence and lower mortality rates of several human diseases (29, 30). The results strongly suggest the presents of polyphenolic compounds like flavonoids, glycosides and vitamins C, E and β-carotene as the important components of this plant, and some of its pharmacological effects could be attributed to the presence of these valuable constituents. The seed and F2 were moderately rich in mineral element composition, like magnesium and zinc which are useful in endogenous antioxidant enzyme production. The seed is rich in potassium which indicates diuretic property and also has a high concentration of iron suggesting that it may be helpful in management of anaemia.Zinc and some of the element affect both non-specific and specific immune function as they participate in major biochemical pathways involved in the perpetuation of genetic materials.

From the result, the extracts F1 and F2 protected the liver from the effect of carbon tetrachloride in a dose dependent manner. F1 in doses of 200 and 500 mg/kg had little protection. The hepatoprotective effect of the extract was more pronounced in the methanol extract (F2). The reason may be that the total lipid extraction procedure could not extract the antioxidant phenolic compounds, mostly hydrophobic in nature and are known free radical scavenge. The results of F2 were comparable to that of silymarin and much better than that of vitamin E. The methanol extract had significant antioxidant and hepatoprotective

activities, which may be useful therapeutic agents for treating radical-related pathological damage. The result suggests that methanol extract of *Diospyros preussii* seed has antioxidative activity table 4 and can protect the organs from peroxidation damage. This could be due to the presence of antioxidant related vitamins, mineral and phytochemical constituents present in the seed. Further studies are ongoing to narrow down these activities to their particular components.

Conflict of interest declaration

The authors declare no conflict of interest.

ACKNOWLEDGMENTS

The authors are grateful to Mr. A. O. Ozioko of the Centre for Ethnomedicine and Drugs Development, a subsidiary of Bioresources Development and Conservation Program (BCDP), Nsukka, Enugu State for his assistance in sourcing the plant materials and authentication.

REFERENCES

1. Dahanuka SA, Kulkarni RA, Rege NN. Pharmacology of medicinal plants and natural products. Indian J Pharmacol 2002; 32: 508-512.
2. Iwu MW, Duncan AR, Okunji CO. New antimalarials of plant origin. In: Janick J, editor. Perspective on new crops and new uses. Alexandria: VA ASHS Press.1999; 457 – 462.
3. Rodders J, Speedie M, Tyler V. Pharmacognosy and harmacobiotecknology. Baltimore: Williams and Wilkins.1996; 1-4.
4. Patwardhan B. Ethnopharmacology and drug development. J Ethnopharmacol 2005; 100:50-52.
5. AjayiAF, AkhigbeRE, AdewumiOM, OkelejiLO, MujaiduKB, Olaleye SB.Effect of ethanolic extract of *Cryptolepis sanguinolenta* stem on *in vivo* and *in vitro* glucose absorption and transport: Mechanism of its antidiabetic activity. Ind J Endocrinology 2012; 16(7): 91-96.
6. Iwu M. Handbook of African medicinal plants. Boca Raton, FL: CRC Press; 1993.
7. Bierer DE, Dubenko LG, Zhang P, Lu Q, Imbach PA, Garofalo AW et al. Antihyperglycemic activities of cryptolepine analogues: An ethnobotanical lead structure isolated from *Cryptolepis sanguinolenta*. J Med Chem 1998; 41:2754-64.
8. Bierer DE, Fort DM, Mendez CD, Luo J, Imbach PA, Dubenko LG et al. Ethnobotanical-directed discovery of the antihyperglycemic properties of cryptolepine: Its isolation from *Cryptolepis sanguinolenta*, synthesis, and *in vitro* and *in vivo* activities. J Med Chem 1998; 41:894-901.
9. Luo J, Fort DM, Carlson TJ, Noamesi BK, nii-Amon-Kotei D, King SR et al. *Cryptolepis sanguinolenta*: An ethnobotanical approach to drug discovery and the isolation of a potentially useful new antihyperglycaemic agent. Diabet Med 1998; 15:367-74.
10. Bugyei KA, Boye GL and Addy ME. Clinical efficacy of a tea-bag formulation of *Cryptolepis sanguinolenta*root in the treatment of acute uncomplicated *falciparum*malaria. Ghana Medical Journal 2010, 44 (1):3-10.
11. Cimanga K, De Bruyne T, Pieters L, Vlietinck AJ, Turger C.A. *In vitro* and *in vivo* antiplasmodial activity of cryptolepine and related alkaloids from *Cryptolepis sanguinolenta*. J Nat Prod 1997; 60:688-691.
12. Grellier P, Ramiaramanana L, Milleriox V, Deharo E, Shrevel J, Frappier F. Antimalarial activity of cryptolepine and isocryptolepine, alkaloids isolated from *Cryptolepis sanguinolenta*. Phytother Res 1996;10:317-321.
13. Boakye-Yiadom K. Antimicrobial properties of some West African medicinal plants II. Antimicrobial activity of aqueous extracts of *Cryptolepis sanguinolenta*(Lindl.) Schlechter. Quart J Crude Drug Res 1979; 17:78-80.
14. Paulo A, Duarte A, Gomes ET. *In vitro* antibacterial screening of *Cryptolepissanguinolenta*alkaloids. J Ethnopharmacol 1994; 44:127-130.
15. Ajayi A. F., Akhigbe R.E. Antifertility activity of *Cryptolepis sanguinolenta* leaf ethanolic extract in male rats. J Human Rep Sci 2012; 5(1): 43-47.
16. Tackie AN, Boye GL, Sharaf MH, Cryptospirolepine, an unique spiro-nonacyclic alkaloid isolate from *Cryptolepis sanguinolenta*. J Nat Prod 1993; 56:653-70.
17. Pousset JL, Martin MT, Jossang A, Bodo B. Isocryptolepine from *Cryptolepis sanguinolenta*. Phytochem 1995; 39:735-6.
18. Sharaf MH, Schiff PL Jr, Tackie AN, Phoebe CH Jr, Martin GE. Two new

indoloquinoline alkaloids from *Cryptolepis sanguinolenta*: Cryptosanguinolentine and cryptotackieine. J Heterocyclic Chem 1996; 33:239-43.

19. Sharaf MH, Schiff PL Jr, Tackie AN, Phoebe CH Jr, Johnson RL, Minick D. The isolation and structure determination of cryptomisrine, a novel indolo [3,2-b] dimeric alkaloid from *Cryptolepis sanguinolenta*. J Heterocyclic Chem 1996; 33:789-97.
20. Yüksel N, Türkmen B, Kurdoğlu AH, Başaran B, Erkin J, Baykara T. Lubricant efficiency of magnesium stearate in direct compressible powder mixtures comprising cellactose® 80 and pyridoxine hydrochloride. FABAD J Pharm Sci 2007; 32: 173-183.
21. Harborne JB. Phytochemistry. Academic Press, London; p. 89-131.
22. Trease GE and Evans WC. Pharmacology. 15th Edn Saunders Publishers, London; 2002, p. 42-44, 221-306, 331-393.
23. British Pharmacopoaeia. The Commision Office London; 2009; Vol. 111: 6578- 6585.
24. Ikewuchi CC and Ikewuchi JC. Chemical profile of *Pleurotus tuberregium* (Fr) Sing's Sclerotia. The Pacific J Sci Tech 2008; 10(1):28-30.
25. Edeoga HO, Okwu DE, Mbaebie BO. Phytochemical Constiuents of some Nigerian medicinal plants. Afri J Biotechnol 2005; 4 (7):685-688.
26. Ofoefule SI. A text book of pharmaceutical technology and industrial pharmacy. Samakin (Nig.) Enterprises; 2002: 26 – 65.