

Original Article



Characterization of Ceiba petandra and Lannea kerstingii Stem Bark Extract Creams

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Olusola Isaac Aremu*, Adeola Tawakalitu Kola-Mustapha, Taiye Hameedat Ayotunde

Department of Pharmaceutics and Industrial Pharmacy, Faculty of Pharmaceutical Sciences, University of Ilorin, Ilorin, Nigeria.

Abstract

A study on physicochemical characteristics of Ceiba petandra and Lannea kerstingii stem bark extracts cream, being developed for the treatment of topical dermatophyte infections, has been carried out to assure its safety, effectiveness, and stability during storage and usage. The stem barks of Ceiba petandra and Lannea kerstingii were extracted with methanol and ethyl acetate respectively. The obtained extracts and their combinations were formulated into creams using modified Aqueous Cream BP base at concentrations ranging from 0.4 to 3.2%w/w. Physical and chemical characteristics of the creams such as stability, viscosity, pH, diffusion, irritancy and elegance were evaluated. Ketoconazole cream was used as the reference product. Creams prepared with each extract alone exhibited favorable characteristics in terms of spreadability, non-irritancy, stability, and diffusivity. Creams formulated with Ceiba petandra were most stable with pH ranges of 6.21 to 7.13. Those formulated with Lannea kerstingii were more acidic. Creams formulated with the combinations of the two extracts exhibited some levels of incompatibility, which increased with increasing extract concentrations. The rate of diffusion of the extracts from cream base increased with time and was generally more at 37 °C as compared to that at 25 °C. Cream formulations of the stem bark extracts of Ceiba petandra and Lannea kerstingii, but not their combinations using modified Aqueous Cream (BP), possess satisfactory physicochemical characteristics.

Keywords: Ceiba petandra, Lannea kerstingii, stem bark extract, Creams, Characterization.

*Corresponding author: Olusola Isaac Aremu, Department of Pharmaceutics and Industrial Pharmacy, Faculty of Pharmaceutical Sciences, University of Ilorin, Ilorin, Nigeria.
Tel: +2348033259890
Email address: solabank@yahoo.com

Introduction

Medicinal plants have been used for the treatment of illness since ancient times (Gaja, 2012). Numerous plant-derived therapeutic agents for the modern medicine have been provided by medicinal plants (Trease and Evans, 2000). There has been a growing interest worldwide in the study of medicinal plants as a source of pharmacologically active compounds. About 20% of plants across the world have been subjected to the pharmacological and biological test. A large number of antibiotics introduced into the market are from natural or semi-synthetic sources (Monthana and Lindequist, 2005). Despite tremendous progress in human medicines, infectious diseases caused by bacteria, fungi, viruses and parasites are still a major threat to public health. Their impact is particularly large in developing countries due to relative unavailability of medicines and the emergence of widespread drug resistance (Zampini et al, 2009). During the last two decades, the development of drug resistance, as well as the appearance of undesirable side effects of certain antibiotics (Zampini et al, 2009), has led to the search of new antimicrobial agents mainly among plant extracts with the goal to discover new chemical structures, which overcome the aforementioned disadvantages. Current research on natural molecule and products primarily focuses on plants since they can be sourced more easily and be selected based on their ethnomedicinal uses (Okemo et al, 2003). Available literature has shown that several plants have been used for medicinal purposes; *Ceiba petandra* and *Lannea kerstingii* belong to the group of such plants. Plants contain many compounds, and it is likely that they work together to produce the desired medicinal effect. For most plants, the specific compound that causes a therapeutic effect is unknown. Several plants are often used together to enhance effectiveness and synergistic actions and to reduce toxicity

(Hawkins and Ehrlich, 2007).

The plant *Ceiba petandra* belongs to the family called Malvaceae and subfamily of Bombacoideae. Its synonyms are *Bombax pentandrum*, *Ceiba caribaea*, *Eriodendron anfractuosum*, *Bombax ceiba*. It is commonly called Kapok tree or white silk cotton (English), Rimi, Riimaayee (Hausa, Nigeria), Vamber (Tiv, Nigeria), Kīng (Yandang, Nigeria), Ogungun, Araba (Yoruba, Nigeria), Akpu-ota (Igbo, Nigeria) (Doughari and Loryule, 2009). In Nigerian folk medicine, *Ceiba petandra* is used for the treatment of diabetes and infections. The leaves are used as a laxative, and as an infusion for colic in man and in livestock (Burkill, 2000). The leaves are used as curative dressings on sores. A decoction of boiled roots is used to treat edema. The gum is eaten to relieve stomach upset. In Ivory Coast, it is used to remove foreign matter from the eyes, or as emollient and sedative in Gabon (Burkill, 2005). In many countries in Africa, the bark and the stem are taken as remedies for diarrhea, localized oedemas, wash sores and to relieve stomach complaints, hernia, blennorrhoea, heart trouble, asthma, and gargles for gingivitis and sometimes toothache (Burkill, 2000).

The plant *Lannea kerstingii* belongs to the family Anacardiaceae. It is synonymous to *Lannea barteri*. *Lannea kerstingii* is locally called kondro (Bale, Ivory Coast), tūdi (Hausa, Nigeria), kanchimbelli (Sisaala, Ghana), nimbiligh (Tiv, Nigeria) (Burkill, 2000). It is utilized in traditional medicine by various cultures worldwide, although application varies by region (Michael, 2004). In Burkina Faso, a root decoction is taken to cure a hernia and a leaf decoction to cure hemorrhoids. The barks are used externally to treat ulcers, sores, and leprosy. Macerated roots are used in a poultice for the treatment of wounds. Leaves decoction is drunk to cure piles (Umberto, 2014).

Another major area of concern in herbal medicine is their formulation into acceptable and more usable ready dosage forms. Along

with other dosage forms, herbal medicines are formulated in the form of creams. Creams are semisolid emulsions intended for external applications. They are often composed of two phases, the aqueous and oily phase. Emulsions are of two types, oil-in-water, and water-in-oil emulsions. Oil-in-water (o/w) emulsions are most useful as water-washable bases, whereas water-in-oil (w/o) emulsions are emollient and cleansing agents. The active ingredient is often dissolved in one or both phases, thus creating a three-phase system (Barry, 2002; Betageri and Dan, 2002). Aqueous Cream BP is a light, hydrocarbon-based emulsion, which is officially registered in the British Pharmacopoeia and categorized by the British National Formulary as a non-proprietary emollient preparation. The negative effects of aqueous cream BP on the skin barrier are most likely associated with the presence of Sodium Lauryl Sulphate (SLS). Aqueous solutions of SLS have been shown to cause cutaneous irritation and elevate transepidermal water loss of healthy skin at concentrations of 1% and less (Tupker and Willis, 1997). For these reasons, the emulsifying wax in the formula for aqueous cream BP may be substituted with a safe alternative. The most important consideration with respect to pharmaceutical and cosmetic emulsions (creams) is the stability of the finished product. The stability of a pharmaceutical emulsion is characterized by the absence of coalescence of the internal phase, absence of creaming, and maintenance of elegance with respect to appearance, odor, color and other physical properties. An emulsion is a dynamic system, however, any flocculation and resultant creaming represent potential steps towards complete coalescence of the internal phase (Im-emsap et al, 2002). The present study, therefore, has sought to formulate, characterize and evaluate cream formulations containing *Ceiba petandra* and *Lannea kerstingii* extracts.

Materials and Methods

Materials

The stem barks of *Ceiba petandra* and *Lannea kerstingii* were collected from Zaria and authenticated at the herbarium section of the Department of Biological Sciences, Ahmadu Bello University Zaria, Nigeria. They were assigned voucher numbers 1832 and 7059 respectively.

Chemicals used include methanol (Guangdong Chemical Reagent, China), ethylacetate (Guangdong Chemical Reagent, China), cento stearyl alcohol (BDH Chemicals Ltd., Poole England), liquid paraffin (BDH Chemicals Ltd., Poole England), polysorbate 80 (BDH Chemicals Ltd., Poole England), white soft paraffin (BDH Chemicals Ltd., Poole England), chlorocresol (BDH Chemicals Ltd., Poole England), nutrient agar (Biomalk, India), 5% w/v ferric chloride solution (BDH chemicals Ltd., Poole England), phosphate buffer 6 (BDH Chemicals Ltd., Poole England), dimethyl sulphur oxide (DMSO) (Guangdong Chemical Reagent, China).

Methods

Preparation of *Ceiba petandra* and *Lannea kerstingii* extracts

After identification, the stem barks of the two plants were removed and air-dried for three weeks and thereafter pulverized. The obtained materials were sieved with 60-mesh size to obtain finely powdered mass, which was then stored in polyethene bag for further use.

Extraction of *Lannea kerstingii* powdered bark was carried out as described by Njinga et al, 2013. A hundred (100) g of the stem bark of *Lannea kerstingii* was extracted exhaustively using sequential solvent extraction. Thereafter 140 g of the powdered stem bark of *Ceiba petandra* was extracted with methanol (Guangdong Chemical Reagent, China) after

defatting with petroleum ether for 24 hours. The methanolic extract was concentrated using rotary evaporator at 40 °C and 85 rpm (Buchi Rotary Evaporator, Germany). The concentrate was stored in the refrigerator prior to evaluation. Test fractions of the plant extracts were prepared in 30% DMSO to obtain concentrations of 100, 200, 400, 600 and 800 mg/mL for the individual plants and a combination of both at 1:3 ratio of *Ceiba petandra* to *Lannea kerstingii*.

Formulation of creams:

Aqueous cream base (BP) in which the sodium lauryl sulfate (SLS) was substituted with polysorbate 80, was used for the formulation of the herbal cream product. Replacement of SLS was due to its reported allergic, hypersensitivity, and keratolytic effects. The modified aqueous cream was used to prepare 25g of oil in water (o/w) emulsion based cream of concentrations 0.4, 0.8, 1.6, 2.4, and 3.2% w/w of each plant extract and a combination of the two extracts at 1:3. This was presented in Table 1.

Table 1: Composition of the 25 g modified aqueous cream incorporated with the stem bark extract of *Ceiba petandra*, *Lannea kerstingii* and a combination of the two extracts.

Ingredients	Quantity of ingredient in each formulation			
Formulations	(LKEC)	(CPEC)	(CLEC)	(CFWE)
Liquid paraffin (g)	1.50	1.50	1.50	1.50
White soft paraffin (g)	3.75	3.75	3.75	3.75
Cetostearyl alcohol (g)	2.00	2.00	2.00	2.00
Polysorbate 80 (g)	0.25	0.25	0.25	0.25
Chlorocresol (g)	0.10	0.10	0.10	0.10
<i>Lannea kerstingii</i> stem bark extract (mL)	1.00	0.00	0.75	0.00
<i>Ceiba petandra</i> stem bark extract (mL)	0.00	1.00	0.25	0.00
Purified Water (mL)	16.40	16.40	15.40	17.40
Total weight (g)	25.00	25.00	25.00	25.00

KEYS:

LKEC - *Lannea kerstingii* extract cream.

CPEC - *Ceiba petandra* extract cream.

CLEC - *Ceiba petandra* and *Lannea kerstingii* extract cream.

CFWE - Cream formulation without extracts (negative control).

Physical Evaluation.

The appearance, color, odor, texture, spreadability, ease of removal, ease of application, oily/tacky feel, homogeneity and phase separation of the different concentration of creams prepared were observed and recorded. Rheological evaluation of the creams was carried out, using a viscometer to measure the viscosity at a higher temperature.

Chemical Evaluation.

Determination of pH.

The pH of each cream formulation was determined by a pH meter (Hanna, England). All measurements were an average of three determinations and expressed as mean \pm S.D.

Evaluation of Drug-Vehicle Compatibility.

The cream samples were observed for any

instability between the drug and the type and amount of base used. Observation for each sample was recorded.

Evaluation of Cream Stability

The accelerated stability test was conducted to determine the color and pH change of the different concentrations of cream at 40, 25 and 4 °C.

Evaluation of Drug Release

0.25 mL was measured and poured into a 25 mL volumetric flask and the cream was buffered with phosphate buffer 6 to fill up to 25 mL mark. The volumetric flask was shaken thoroughly to obtain a homogeneous mixture.

This procedure was repeated for the different creams formulated. Sterilized nutrient agar was poured into sterile Petri dishes and allowed to solidify. The surface of each plate was flooded with 5%w/v ferric chloride solution, and excess solution was drained and allowed to dry. Cylindrical holes were made on each plate with the aid of number 6 cork borer. 0.5 mL of the different concentrations of buffer and cream sample prepared was poured into the holes. This was done in triplicate and the plates were placed on the bench for 1 hour to allow proper diffusion. The plates were then incubated at 27 and 37 °C. The zones of color changes were measured with a vernier caliper for each sample after 1, 2, 3, 6, 12, and 24 hours.

Results:

Table 2: Organoleptic properties of extracts

Extract	Color	Odor	Texture	pH
Methanol extract of <i>Ceiba petandra</i>	Light brown	Faint	Fine	5.60
Ethyl acetate extract of <i>Lannea kerstingii</i>	Dark brown	Burnt wood	Gritty	4.12
Combination of extracts	Brown	Burnt wood	Gritty	6.25

Table 3: Physical characterization of cream formulations with the extracts in (%w/w).

Formulations		Color and odor	Texture and skin feel	Ease of application and removal
(LKEC)	0.4	Light pink and odorless	Hard and smooth, feels smooth and non-irritant	Easy to apply and easily removed with water
	0.8	Light pink and chloro-cresol smell	Hard and smooth, feels smooth and non-irritant	Easy to apply and easily removed with soap water
	1.6	Light pink and burnt wood smell	soft and smooth feels smooth and non-irritant	Easy to apply and easily removed with water
	2.4	Light pink and odorless	soft and smooth feels smooth and non-irritant	Easy to apply and easily removed with water
	3.2	Peach and odorless	Hard and smooth, feels smooth and non-irritant	Not easy to apply and easily removed with water
(CPEC)	0.4	Light pink and odorless	soft and smooth feels smooth and non-irritant	Easy to apply and easily removed with water
	0.8	Light pink and odorless	soft and smooth feels smooth and non-irritant	Easy to apply and easily removed with water
	1.6	Peach and extract smell	soft and smooth feels smooth and non-irritant	Easy to apply and easily removed with water
	2.4	Pink and odorless	Hard and smooth, feels smooth and non-irritant	Easy to apply and easily removed with water
	3.2	Light brown and odorless	Hard and smooth, feels smooth and non-irritant	Easy to apply and easily removed with water

(CLEC)	0.4	Pink and chlorocresol smell	Hard and smooth, feels smooth and non-irritant	Easy to apply and easily removed with water
	0.8	Peach and extract smell	Hard and smooth, feels smooth and non-irritant	Easy to apply and easily removed with water
	1.6	Light brown and burnt wood smell	Hard and smooth, feels smooth and non-irritant	Not easy to apply and easily removed with water
	2.4	Brown and extract smell	Hard and smooth, feels smooth and non-irritant	Not easy to apply and easily removed with water
	3.2	Dark brown and burnt wood smell	Hard and smooth, feels smooth and non-irritant	Not easy to apply and easily removed with water
(CFWE)		White and odorless	Soft and smooth feels smooth and non-irritant	Easy to apply and easily removed with water
KC		White and odorless	Soft and smooth feels smooth and non-irritant	Easy to apply and easily removed with water
KEY: LKEC - <i>Lansea kerstingii</i> extract cream; CPEC - <i>Ceiba petandra</i> extract cream; CLEC - <i>Ceiba petandra</i> and <i>Lansea kerstingii</i> extract cream; CFWE - cream formulation without extracts (negative control); KC - Ketoconazole cream (positive control)				

Chemical Evaluation of the Cream Formulations.

Table 4: pH Determination and Viscosity Values of the Cream Formulations

Formulations (% w/w)	Viscosity evaluation at 50°C (cm ² /sec)	pH	Drug-vehicle compatibility
(LKEC) 0.4	61.79 ± 0.65	6.25 ± 0.75	Stable
0.8	99.20 ± 0.34	6.21 ± 0.12	Stable
1.6	71.48 ± 0.81	7.09 ± 0.78	Stable
2.4	101.87 ± 0.57	6.49 ± 0.16	Stable
3.2	59.79 ± 1.22	7.13 ± 1.52	Stable
(CPEC) 0.4	53.21 ± 0.94	5.16 ± 0.21	Stable
0.8	87.84 ± 1.38	5.63 ± 0.77	Stable
1.6	114.56 ± 0.65	5.31 ± 1.17	Stable
2.4	80.16 ± 1.70	5.80 ± 1.54	Stable
3.2	111.89 ± 1.39	6.27 ± 0.64	
(CLEC) 0.4	43.42 ± 1.92	6.83 ± 0.29	Stable
0.8	62.79 ± 0.56	5.87 ± 0.54	Stable
1.6	53.44 ± 0.11	5.13 ± 0.65	Unstable
2.4	54.776 ± 1.96	5.32 ± 0.92	Unstable
3.2	129.26 ± 1.52	5.16 ± 0.91	Unstable
(CFWE)	46.09 ± 0.17	6.22 ± 0.84	Stable
Ketoconazole cream(KC)	77.49 ± 0.11	5.98 ± 0.15	Stable
KEY: LKEC - <i>Lansea kerstingii</i> extract cream; CPEC - <i>Ceiba petandra</i> extract cream; CLEC - <i>Ceiba petandra</i> and <i>Lansea kerstingii</i> extract cream ; CFWE - Cream formulation without extracts.			

Table 5: Drug Release of Cream Formulations.

Cream formulations	Concentrations	Time (hours) at 25°C						Time (hours) at 37°C					
		1	2	3	6	12	24	1	2	3	6	12	24
LKEC	0.4	0	0	0	0	0	0	0	0	0	0	0	0
	0.8	0	0	0	0	0	0	0	0	0	0	0	0
	1.6	0	0	0	0	0	0	0	0	0	0	0	0
	2.4	6	11	12	15	18	18	1	6	8	14	16	16
	3.2	2	7	11	14	14	14	5	9	11	14	18	18
CPEC	0.4	0	0	0	0	0	0	0	0	0	0	0	0
	0.8	0	0	0	0	0	0	0	0	0	0	0	0
	1.6	0	0	0	0	0	0	0	0	0	0	0	0
	2.4	2	7	8	10	10	10	1	7	11	14	15	15
	3.2	1	5	6	9	10	10	6	9	10	11	11	11
CLEC	0.4	0	0	0	0	0	0	0	0	0	0	0	0
	0.8	0	0	0	0	0	0	0	0	0	0	0	0
	1.6	0	0	0	0	0	0	0	0	0	0	0	0
	2.4	5	9	12	13	13	13	0	5	6	7	10	10
	3.2	5	10	11	15	16	16	0	6	6	5	8	8
CFWE		0	0	0	0	0	0	0	0	0	0	0	0
KC		2	6	9	12	13	13	6	10	11	13	15	15
KEY: LKEC - <i>Lannea kerstingii</i> extract cream; CPEC - <i>Ceiba petandra</i> extract cream CLEC - <i>Ceiba petandra</i> and <i>Lannea kerstingii</i> extract cream ; CFWE - Cream formulation without extracts (negative control); KC - Ketoconazole cream (positive control).													

Discussion

Many herbs and medicinal plants have been found useful for the management of various diseases and conditions. Their application and acceptability have been hampered largely by their presentation forms. *Ceiba petandra* and *Lannea kerstingii* are known for their medicinal properties. In the present work, extracts were obtained from these plants and used in the formulation of a dosage form that can be used in the treatment of skin disorders. The stem bark of the plants was selected on the basis of data obtained from the literature for the treatment of skin disorders. Aqueous cream BP with slight modification was chosen as the base for formulation because of ease of penetration to the skin (Muller et al, 2003).

The extracts from the stem bark of both plants had a brown color with an odor like a burnt wood. Creams formulated with *Lannea kerstingii* stem bark extract appeared pink; those from *Ceiba petandra* appeared peach while those from the combination of extracts appeared brown. The color of the various cream formulations is a reflection of the color of corresponding extracts. This may, however, be improved upon by introducing suitable colorant in the formulation in order to enhance elegance and acceptability. Creams formulations with the extract from *Lannea kerstingii* stem bark were easy to apply and easily removed with soap and water. This is an encouraging outcome because patients can

easily comply with the usage of non-greasy, non-occlusive, easily washable dermal formulations. A similar feature of ease of application and washability was observed with cream formulations from *Ceiba petandra* stem bark extract. The cream formulations from the combination of stem bark extracts of the two plants gave a different picture from the aforementioned. The explanation to this observation was possible because of the increase in the density of the particles present in the formulation, as the concentration of the extracts had increased. At higher concentrations of particles, the stiffness problem could easily set in which would make dermal application difficult. To overcome this, particle size of ingredients in a formulation would have needed to be carefully controlled. The ease of application decreased with the increase in the concentration of the extracts. All creams formulated were non-irritating when applied to the skin. Some of the formulated creams appeared hard but had a smooth feel.

Drug-vehicle incompatibility was observed in the creams made with a combination of extracts as the drug was seen to separate from the cream after a week of the formulation. Phase separation was not seen in the creams formulated with *Lannea kerstingii* and *Ceiba petandra* extracts alone. The phase separation, observed in the cream formulations with a combination of the extracts, may be due to the difference in the density of the combined extract and that of the base. This separation is referred to as creaming. According to Stoke's equation, the velocity of the creaming rate is dependent on the size of the particles, density and the viscosity of the systems. The creaming rate will be reduced when the particles are homogeneous, small, the density difference between the particles is small, and the system is viscous (Anita, 2005).

Time variable rheological behavior of semisolid may signal physical or chemical changes. The viscosities of all formulations studied and that of reference compound (ketoconazole) is presented in Table 4. It was

observed that as the concentration of the extract increased, the viscosities also increased ($p < 0.05$) except for few concentrations. Viscosity changes may signal sometimes phase changes whereby there are slow rearrangement and contraction of internal structure. Eventually, here and there, globules of what is often clear liquid internal phase are squeezed out of the matrix. The main concern with a system that has undergone such separation is that a patient will not be applying a medium of uniform composition because of the unequal concentration of the drug relative to the other (Anita, 2005). The increase in concentrations observed for some concentrations of LKEC and CPEC was as a result of an increase in extract concentrations and not any compromise with the structure except for the formulation of combinations of extracts, where there was a complete breakdown in structural arrangement probably due to incompatibility. However, the viscosities at lower concentrations compare favorably with that of reference compound (ketoconazole). Measurements are important as they can predict the performance and behavior of a product.

The pH of a medium is the measure of acidity or alkalinity of phase. "Acid" and "basic" are two extreme that describes the chemical properties. It is important to obtain the pH of formulated products as the slightest difference in pH of a formulation can make a major change in how a cream interacts with the skin (Para and Peyre, 2003). Report from the literature posited that pH of the skin is often between 5.4 - 5.9 and very ideal is 4.0 - 4.9 (Schmid and Korting, 2006.) It is important that the base chosen must possess desirable characteristics including acceptable pH. The pH of the two extracts and combination were between the ranges of 4.12 - 6.25 as can be seen in Table 4, while the pH of the formulated creams was between the ranges of 5.13 - 7.13 and that of the negative and positive controls were 6.22 and 5.98 respectively. Most of the pH values obtained fell within the acidic range except for 1.6 and 3.2% w/w LKEC formulations. Generally, the pH values obtained were the limits which may not have altered skin penetration, efficacy, and

stability of the formulations.

The results of accelerated stability evaluation after 3 weeks revealed that all formulations were unstable at a lower temperature of 4°C. All formulations of *Lannea kerstingii* extract and 0.4%, 0.8%w/w *Ceiba petandra* extract were stable at 25 and 40 °C respectively. This suggests that the formulations will be able to withstand storage conditions without them losing their structural integrities. The formulation of a combination of extracts at all concentrations was unstable at both 25 and 40 °C respectively.

The result of drug release from the different concentrations of creams prepared was shown in Table 5. The zones of color change increased in diameter with an increase in concentration and time for all concentrations studied. The amount released and the rate of release of a drug suspended in a vehicle such as cream may be related to time and to variables of the system (Aulton,1996).The rate of release of the extract is greatly influenced by diffusion coefficient, concentration, and solubility of the extract in the cream base. Employing the following, equation,

$$\frac{dm}{dt} \cong \left(\frac{AD_v C_s}{r_t} \right)^{\frac{1}{2}}$$

Equation (1)

Where dm/dt is the rate of release, A is the total amount of cream base, D_v is the diffusion coefficient in the base, C_s is the solubility of the drug in the vehicle and t is time. It may be possible to predict the in-vitro availability of medicament from vehicle base. Temperature also had an effect on the rate of release of extract at various concentrations, higher at 37 °C than 25 °C as can be seen in Table 5. This is possibly so because temperature is able to alter diffusive tendency of drugs in cream base at elevated temperature due to increase in fluidity of the base

Conclusion

From the foregoing cream formulations of the stem bark extracts of *Ceiba petandra* and *Lannea kerstingii* but not their combinations using modified Aqueous Cream (BP) possess satisfactory physicochemical characteristics to support further development.

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