UDS International Journal of Development [UDSIJD] ISSN: 2026-5336
Volume 10 No. 2, 2023
https://www.udsijd.org

VARIATION IN BIOCHEMICAL AND MINERAL QUALITY OF SHEA (*VITELLARIA PARADOXA* L.) LATEX ALONG GEOGRAPHICAL GRADIENT IN GHANA

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Abstract

The research investigated the biochemical and mineral composition of Shea latex in three geographical locations in the transitional and savannah agro-ecological zones, considering a north-south gradient: Yagaba (10^{0}13'37''N, 01^0 16'11''W), Nyankpala (09^{0}25'93''N, 01^{0}00'42''W) and Kawampe (08^{0}44'70''N, 01^{0}33'58''W). The biochemical compositions entailed phytochemical and proximate contents determinations. Crude protein and crude fibre levels were significantly highest in July (1.33% and 8.86%, respectively) and in August (1.29% and 8.80%, respectively). However, the mean monthly (May-October, 2014) proximate content of the Shea latex did not differ significantly for ash, carbohydrates, crude fat, and moisture. Shea latex crude protein level was significantly highest in Yagaba (1.22%) than Nyankpala (1.11%). Nyankpala and Kawampe had significantly highest levels of 40.66 mg/L and 39.53 mg/L for potassium (K), respectively and significantly highest levels of 0.02 mg/L each for zinc (Zn) than Yagaba (0.01 mg/L). The Shea latex crude protein levels were consistently relatively low and significant variations manifested with respect to geographical location and tapping period. Shea latex sodium (Na): potassium (K) ratio of less than one across the study segments (tapping period and geographical location) was also revealed. The shea latex phytochemical quality screening of diverse polar solvents extracts revealed the presence of reducing sugars, terpenoids, alkaloids and tannins. The study suggests that Shea latex may have nutritional and medicinal benefits, and the potential for the hypo-allergenic latex products manufacturing.

Keywords: Minerals, Proximate Analysis, Phytochemical Quality, Shea Latex, *Vitellaria paradoxa*

Introduction

Latex is an emulsion with a varied composition that includes terpenoid compounds, alkaloids, and polymeric substances like gums and resins, oils, starch, and a considerable number of enzymatic activities and proteins (Han, Shin, Yang, Kim, Oh & Chow, 2000; Kekwick, 2001; Ko, Chow & Han, 2003).

Shea latex is a milky colloidal matrix that exudes upon cutting the bark of the shea tree. According to previous studies, this exudate may contain important biochemical constituents including protein, fat, carbohydrates and mineral constituents (proximate components) (Fosu & Quainoo, 2013; Quainoo, Abdul-Aziz, Abubakari & Abagale 2015). Shea is an important multipurpose plant species with differing natural abundance in the transitional and Guinea Savannah regions of Ghana (Hall, Aebischer, Tomlinson, Osei-Amaning & Hindle, 1996). Shea has immense commercial value with high demand for its butter for diverse usage such as; manufacture of body creams and lotions and as cooking oil. Activities and structure associated with biotechnological applications and latex-borne substances have been proposed for latex compound. In traditional medicines, a lot of latescent species are used to treat a wide range of illnesses. Shea butter is also widely used as an alternative to cocoa butter in the production of the highly patronized chocolate among few others. Substances that are latex-borne serve as a potential tool for crop protection in agricultural biotechnology. The use of plants that are latescent in indigenous medicinal systems for diverse purposes also considers latex fluids as a good source for pharmaceuticals.

In Ghana and elsewhere much research attention in the shea sector is given to the shea nuts, fruits and butter (Mahamadi, Jules, Tahir & Sibiri, 2009) to the neglect of the other untapped potential products of the plant such as; the shea cake, latex, roots, bark, leaves and shells. These products, apart from the indigenous knowledge generated about them, in indigenous medicine, very little are known about their commercial potential. There is the need therefore, to bridge the knowledge gap on the natural products of Shea by directing research attention towards these neglected products. This research investigated variation in proximate, mineral and phytochemical qualities of Shea latex based on tapping periods and geographical locations, from the Shea parkland areas in Ghana.

Materials and Methods

Shea latex was tapped from Yagaba and Nyankpala in the Guinea-Savanah and Kawampe in the transitional agro-ecological zones of Ghana between May and October (2014). These locations were purposively selected following a north – south climatic gradient to give a representative coverage of the Shea parkland of Ghana.

Experimental Set Up

Five Shea trees were identified and tagged as experimental trees for tapping of the Shea latex in both fallowed and cultivated fields in each of the three experimental sites. The Shea latex was tapped form each tagged Shea tree into transparent vessels monthly from May to October 2014. At each tapping section the collected latex samples were air - dried under room temperature for a period of three (3) days prior to laboratory analysis. Proximate, minerals content and phytochemical quality analysis were conducted on the samples following the standard protocols briefly described below

Proximate Analysis

The moisture content of the shea latex was determined by oven drying the latex samples at 105 ^{o}C for 4 h. Crude Protein content of shea latex samples was determined using the Kjeldahl method (AOAC, 2000) to first determine total nitrogen. The total nitrogen value was then multiplied by 6.25 to obtain crude protein. Crude fat analysis was done in accordance with the AOAC (2000) standard procedure using the Soxtec Extractor. Total ash content of the Shea latex was estimated following the incineration of the latex at 550

^{o}C in a muffle furnace. Crude fibre was determined following a sequential acid and alkaline hydrolysis of defatted shea latex samples in a fibertec apparatus. The residue collected after the hydrolytic process was treated to an acetone bath and subsequently dried and ashed at 500 ^{o}C. Carbohydrates computation was by difference such that; Carbohydrates = 100 - (%dry matter+ % crude protein + % crude fat + % crude fibre + % ash).

Mineral Analysis

A 2.0 g each of the shea latex samples were weighed into cleaned porcelain crucible, moistened with concentrated HNO_3, and ashed at 500 ^{0}C in a muffle furnace for 2 h (Benton & Vernon, 1990). Aqua regia of 5.0 mL was added to each resultant ash samples and heated to obtain colourless solutions. These solutions were transferred into 100.0 mL volumetric flasks by filtration through ashless filter papers and the volumes were made up to the marks with deionized water. Aliquots of these solutions were aspirated into an atomic absorption spectrophotometer (Shimadzu AAS model AA 6300) for the mineral analysis.

Phytochemical Analysis

Ten grams of each Shea latex sample was cold extracted using solvents of different polarities thus; water, petroleum ether, methanol and acetone over a period of 72 h with occasional shaking. The extracts were filtered through Whatman's No. 1 filter paper and the filtrates were concentrated to about 1/4th total volume over water bath. The phytochemical constituents were then variously test screened from the extracts following different protocols as follows; Alkaloids were tested in accordance with the procedure described by Adebayo & Abdul-Mumeen (2012); Simple sugars, Terpenoids, Steroids and Anthraquinones test were done using the methods described by Sofowora (1993); Flavanoids and Tannins were tested according to Trease & Evans (2002) procedures; Soluble starch test was conducted following Vishnoi (1979) procedure.

Statistical Analysis

Analysis of variance was done on proximate and mineral composition to ascertain their significance based on the geographical gradient, tapering period, cultivated and fallow land fields using genstat version 18. LSD were used to separate means.

Results and Discussion

Comparison of Proximate Composition of Shea latex

The monthly mean values obtained for crude protein and fibre (Table 1) varied significantly ($P < 0.05$) from May-October. The highest value for crude protein was obtained in July which was significantly different ($P < 0.05$) from values obtained in May, June, September and October. Crude fibre content of the shea latex was highest in the month of July, was significantly different ($P < 0.05$) from values obtained in May, June, September and October. The highest and lowest values of shea latex moisture content were recorded in the months of September and May respectively (Table 1). These differences observed in the crude protein and fibre contents of the shea latex may be attributed to variable climatic factors such as rainfall volume (soil moisture levels), different sun shine periods and temperature which varied across the geographic gradient over the study period.

According to studies conducted by researchers, the crude proteins in plant latex has health purposes. Nascimento et al. (2016) indicated that, latex protein extracts from a shrub known as *C. procera*, showed a keen anti-inflammatory activity against important human conditions such as protection against the likes of yeast infection and acute inflammation (de Alencar et al., 2017) and arthritis (Chaudhary, Ramos, Vasconcelos &

Kumar, 2016). de Oliveira et al. (2019) reported on the capacity of the proteins in latex to reduce the levels of blood glucose in humans which validated the traditional use associated with this effect. According to the current studies, it is evident that latex borne substances are suitable for biotechnology.

Table 1: Percentage (%) proximate content of Shea latex based on tapping periods (monthly) and geographical location

Month	Crude fat	Crude protein	Moisture	Ash	Crude fibre	Carbohydrates
May	53.54^{a}	1.02^{c}	1.84^{a}	4.99^{a}	8.29^{b}	30.32a
June	52.99^{a}	1.05^{c}	2.03^{a}	4.97^{a}	8.35^{b}	30.61^{a}
July	55.22^{a}	1.33^{c}	1.94^{a}	5.08^{a}	8.86^{a}	30.71^{a}
August	56.16^{a}	1.29^{c}	1.97^{a}	5.11^{a}	8.80^{a}	29.81^{a}
September	54.73^{a}	1.14^{bc}	2.07^{a}	5.11^{a}	8.42^{b}	28.53^{a}
October	53.47^{a}	1.13^{c}	2.03^{a}	5.17^{a}	8.39^{b}	29.81^{a}
P values	0.2	0	0.308	0.273	0	0.683
S.E.M	1.003	0.038	0.076	0.069	0.077	1.011
Location						
Kawampe	53.39^{a}	1.14^{ab}	2.00^{a}	5.13^{a}	8.53^{a}	30.87^{a}
Nyankpala	54.68^{a}	1.11^{b}	1.99a	5.01^{a}	8.48^{a}	29.74^{a}
Yagaba	54.98^{a}	1.22^{a}	1.96a	5.07^{a}	8.55^{a}	29.28^{a}
P values	0.247	0.012	0.813	0.248	0.701	0.276
S.E.M	0.709	0.027	0.054	0.049	0.054	0.715

* Mean values in a column sharing different alphabets are significantly different ($P < 0.05$)

Effect of Geographical location on Proximate Composition of Shea Latex

Crude protein content of the shea latex was statistically variable ($P < 0.05$) with respect to location and does agree with the findings of Quainoo et al. (2015) who reported similar variation regarding crude protein content of shea latex along agro-ecological zones. This finding suggests that crude protein synthesis may be dependent on slight variable climatic indices and the soil nutrient levels that may uniquely characterize these different locations. Latex has a plastic nature, depicting differences in production depending on the surrounding environment which can aid the deposition of trace elements in shea latex. The abiotic and biotic variations, such as light, drought, pathogens, inter- and intra-specific competition, and herbivory attack, accounts for the final concentration of compounds in latex (Dicke & van Loon, 2000). Abubakari et al. (2012) reported variations in the soil nutrients potential of the two agro – ecological zones with shea growth in Northern Ghana (i.e., Guinea-Savannah and Transitional belt), from which shea latex samples for this study were collected for analysis. Minimum and maximum crude protein values were recorded at Nyankpala and Yagaba locations, respectively, with Kawampe recording the median value (Table 1).

Variation of Mineral Content in Shea Latex

There were no significant differences ($P > 0.05$) in the results obtained for all the elements analyzed in the different periods. However, potassium and sodium recorded maximum results, in September – October (Table 2). This period recorded relatively higher mean volumes of rains and lower mean temperatures in the sampled locations over the period of the study. Incidentally, minimum values of potassium and sodium were recorded in July-August, unlike the case of most of the proximate parameters discussed earlier, which also recorded relatively lower mean rainfall volumes and higher temperatures over the study period and locations. The trend suggests probable direct and inverse relationships for potassium and sodium concentrations in the shea latex with rain fall volumes and temperature, respectively.

The results of shea latex trace elements content were generally low, with copper (Cu) recording values less than 0.001 mg/L for all the samples. Manganese (Mn) and zinc recorded minimum and maximum values in different periods unlike potassium and sodium (Table 2). Manganese recorded maximum and minimum values in July-August and May-June respectively. The reverse was the case for zinc concentrations in shea latex. This trend may be attributed to lack of influence of micro-climatic factors on the accumulation of trace elements in shea latex.

Table 2: Means of mineral elements of Shea latex based on tapping periods (bi-monthly)

Bi-Monthly	K (mg/L)	Na(mg/L)	Mn (mg/L)	Zn (mg/L)	Cu (mg/L)	Na/K ratios
May-June	35.872[a]	10.306[a]	0.114[a]	0.015[a]	Absent	0.287
July-August	35.583[a]	10.061[a]	0.117[a]	0.013[a]	Absent	0.282
Sep-Oct	35.956[a]	10.317[a]	0.115[a]	0.014[a]	Absent	0.287
P.values	0.732	0.399	0.831	0.182		
S.E.M	0.348	0.149	0.004	0.001		

* Mean values in a column sharing different alphabets are significantly different ($P < 0.05$)

** **Each value is a mean of 18 replicated determinations**

Effect of Geographical Location on Mineral Content in Shea Latex

The essential elements (potassium and sodium) contents of the shea latex recorded relatively higher values than the trace elements (manganese, zinc and copper) in all instances (Table 3). Yagaba and Nyankpala recorded significantly different ($P < 0.05$) values for potassium. This may be attributed to prevalently different levels of potassium in soils from these locations and climatic variations. The essential elements (potassium and sodium) which work together in muscle contraction and nerve transmission both showed promise in shea latex as their levels were relatively higher than the trace elements (manganese, zinc and copper) in all instances. However, levels of sodium (Na) in shea latex from across the locations were not significantly different (Table 3). Nyankpala recorded the least value followed by Yagaba and Kawampe. The Na/K ratios were in all instances less than 1 which according to NRC (1989) is good for regulating high blood pressure. This study ranged the Na/K ratio for shea latex between 0.25 - 0.37 as against 0.17 - 0.35 range reported for shea fruit pulp by Okulo et al. (2010). These Na/K ratios of shea latex of less than 1 showed that, chewing the shea latex as a gum as is the case among indigenes of shea parkland communities in northern Ghana may have health benefits, particularly, in the management of cardio-vascular conditions, just like eating the shea fruits would do (Okulo et al., 2010). The levels of the trace elements (copper, zinc, and manganese) were generally low with zinc levels in the shea latex from Yagaba varying significantly from the other locations (Table 3). The low level of these minerals in the shea latex may be due to absence or presence of miniscule quantities of these elements in soils of the locations. It could also be that, latex in general do not absorb and accumulate these trace elements very efficiently. This observation is buttressed in the *Hevea brasilienesis* latex study by George, Andy & Joseph (2014) who reported similarly low levels of these trace elements such as; copper ranging between 0.015 - 0.017 mg/L with a mean of 0.016 mg/L and zinc had a value of 0.082 mg/L. Copper (Cu) recorded less than 0.001 mg/L (absent) for all the shea latex samples from the different locations in this current study. Manganese as well did not only record low values as; 0.11 mg/L (Nyankpala) < 0.12 mg/L (Kawampe) <0.12 (Yagaba) but also did not vary significantly across the locations (Table 3).

Table 3: Mean mineral elements contents of Shea latex based on geographical location

Location	K (mg/L)	Na (mg/L)	Mn (mg/L)	Zn (mg/L)	Cu (mg/L)	**Na/K ratios**
Kawampe	39.528a	10.361a	0.117a	0.015a	Absent	**0.262**
Nyankpala	40.656a	10.156a	0.111a	0.017a	Absent	**0.250**
Yagaba	27.228b	10.167a	0.118a	0.009b	absent	**0.373**
P. values	0.000	0.553	0.376	0.000		
S.E.M	0.348	0.149	0.004	0.001		

* Mean values in a column sharing different alphabets are significantly different (P < 0.05

**** Each value is mean of 36 replicated determinations**

Interactive Effect of Geographical Location and Period of Tapping on the Phytochemical Quality of Shea Latex

Shea latex did not manifest flavonoids, steroids, anthraquinones and starch presence for the entire study period (Table 3) across the locations. However, terpenoids, tannins, alkaloids and simple sugars profoundly showed presence as more than one solvent extracted portion tested positive to each of the phytochemicals. Neither location nor period of study had impact on shea latex phytochemical quality as positive (+) and negative (-) test outcomes were consistently repetitive and follows similar patterns. Phytochemicals according to Decker, Wanner, Zenk & Lottspeich (2000) are stress and defense related proteins and the presence of a host of them in herbal medicinal substances determine the potency and relevance of such substances and also feed pharmaceutical industries the raw material they need (Savithramma, Linga & Suhrulatha, 2011). Stress, according to Decker et al. (2000), which may be caused by impact of rainstorms, abrasions from grazing activities, bushfires and drought, in this instance did not significantly induce synthesis of these phytochemicals in shea latex differently across the locations and periods. Nonetheless, the phytochemical presence demonstrated in this current study supported findings in *Hevea brasiliensis* latex phytochemical content (George et al., 2014). The presence of alkaloids in Shea latex adds up to a list of plant latex known to contain alkaloids.

Table 4: Phytochemical quality of Shea latex (May- October 2014) based on geographical location and bi-monthly tapping periods

Location	Solvent	Tannins	Sugars	Starch	Alkaloids	Terpenoids	Flavonoids	Steroids	Anthraquinones
May-June									
Yagaba	H_2O	+	+	-	-	-	-	-	-
	Pet	+	-	-	+	+	-	-	-
	Eth	+	+	-	+	+	-	-	-
	Ac	+	-	-	+	-	-	-	-
	Meth	+	-	-	+	+	-	-	-
Nyankpala	H_2O	+	+	-	-	-	-	-	-
	Pet	+	-	-	+	+	-	-	-
	Eth	+	+	-	+	+	-	-	-
	Ac	+	-	-	+	-	-	-	-
	Meth	+	-	-	-	+	-	-	-
Kawampe	H_2O	+	+	-	-	-	-	-	-
	Pet	+	-	-	+	+	-	-	-
	Eth	+	+	-	+	+	-	-	-
	Ac	+	-	-	+	-	-	-	-
	Meth	+	-	-	+	+	-	-	-
July-August									
Yagaba	H_2O	+	+	-	-	-	-	-	-
	Pet	+	-	-	+	+	-	-	-
	Eth	+	+	-	+	+	-	-	-
	Ac	+	-	-	+	-	-	-	-
	Meth	+	-	-	+	+	-	-	-
Nyankpala	H_2O	+	+	-	-	-	-	-	-
	Pet	+	-	-	+	+	-	-	-
	Eth	+	+	-	+	+	-	-	-
	Ac	+	-	-	+	-	-	-	-
	Meth	+	-	-	+	+	-	-	-
Kawampe	H_2O	+	+	-	-	-	-	-	-
	Pet	+	-	-	+	+	-	-	-
	Eth	+	+	-	+	+	-	-	-
	Ac	+	-	-	+	-	-	-	-

	Meth	+	-	-	+	+	-	-	-
September-October									
Yagaba	H_2O	+	+	-	-	-	-	-	-
	Pet	+	-	-	+	+	-	-	-
	Eth	+	+	-	+	+	-	-	-
	Ac	+	-	-	-	-	-	-	-
	Meth	+	-	-	+	+	-	-	-
Nyankpala	H_2O	+	+	-	-	-	-	-	-
	Pet	+	-	-	-	+	-	-	-
	Eth	+	+	-	+	+	-	-	-
	Ac	+	-	-	-	-	-	-	-
	Meth	+	-	-	+	+	-	-	-
Kawampe	H_2O	+	+	-	-	-	-	-	-
	Pet	+	-	-	+	+	-	-	-
	Eth	+	+	-	+	+	-	-	-
	Ac	+	-	-	-	-	-	-	-
	Meth	+	-	-	+	+	-	-	-

Meth. = methanol; Pet. = petroleum ether; Eth. = ethanol; Ac. = acetone; H_2O = water; [+] = positive; [-] = negative

Conclusions

Crude protein, crude fibre and potassium (K) levels in Shea latex may be impacted by the variable climatic indices and other environmental factors that characterized different locations and periods. The Na/K ratio of less than 1 and appreciable levels of sodium and potassium suggest shea latex may have a nutritive potential if chewed as gum. The manifested phytochemical presence in shea latex, namely; alkaloids, terpenoids, reducing sugars and tannins, which are bioactive in nature suggest some therapeutic or medicinal significance for Shea latex.

Acknowledgement

Special appreciation and thanks go to Wienco Ghana for providing the funding for the research.

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