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Evaluation of Quality of Some Commonly Used Herbal Medicinal Plants in Local Markets of FCT, Abuja, Nigeria ^{1*}Florence D. Tarfa, ²Samali, A; ³Ibekwe Nneka N and ⁴Adigwe.O.P

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Abstract

The safety of herbal medicines has become a major concern to both national health authorities and the general public. Hence their production, quality, distribution and use are regulated. Some medicinal plants commonly used for malaria, diabetes and Tuberculosis and respiratory conditions were evaluated for quality. The objective of the study is to assess some quality specifications, physicochemical and heavy metals of some medicinal plant extracts. The samples were identified, purchased, processed and analyzed using appropriate standards methods specified by WHO and AOAC; while the heavy metals content was analyzed using Atomic Absorption Spectrophometer (AAS). All the analyzed samples passed WHO permissible limit established for heavy metals content, however 42.8 and 21.4% of the samples failed moisture and ash content permissible limits respectively. High level of moisture content will favor microbial and fungal growth and shortens shelf-life of the samples. The WHO maximum permissible limit in 80% of the samples, this could be associated to improper handling that has resulted to introduction of inorganic substances such as silica and other inorganic matters. The levels of heavy metals content were all below the WHO maximum permissible limits. The moisture, ash and heavy metals are among the important parameters of medicinal plants that require frequent routine monitoring in order to ensure safety.

Keywords: Medicinal plant, Physicochemical, Heavy metal, safety, Nigeria

INTRODUCTION

Herbal medicines are derived from medicinal plants that have been acclaimed for efficacy and used widely among different communities and cultures throughout the African continent and the world at large, despite insufficient scientific evidence for their quality, safety and efficacy [1].

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Scientific reports have shown that several Nigerian medicinal plants possess therapeutics and active biological substances with potency to prevent, manage and treat different disease conditions [2-4]. Herbal medicines, also known as botanical medicines refer to the medicinal products of plant roots, leaves, barks, seeds, berries or flowers that can be used to promote health and treat diseases since antiquity. The medicinal significance of the plant are due to the bioactive phytochemicals present that generate definite physiological action on humans and animals [5-6]. Phytochemicals are plant secondary metabolites that protect plants from diseases, damage and contribute to the plant's colour, aroma, flavour and protect plant cells from environmental hazards such as pollution, stress, drought, ultraviolet (UV) exposure and pathogenic attacks.[6-8].They are non-nutritional natural bioactive compounds like flavonoids, tannins, phenols, and alkaloids plays a vital role in drug development and functional foods [9]. Medicinal plants and vegetables may be contaminated with heavy metals. Heavy metals are non-biodegradable and persistent environmental contaminants which may be deposited on the surface and then adsorbed into plant tissues. The heavy metal accumulation is traceable to anthropogenic activities such as agricultural practices, high automobile activities, food, chemical and pharmaceutical industries and sewage treatment plants [10]. Heavy metals concentrations exceeding physiological requirement enters the food chain, becomes toxic and bioaccumulates [11-12]. This study was designed to assess some quality parameters of herbal medicines use for the treatment of Diabetes mellitus, malaria, Tuberculoses and respiratory conditions which are sold in Karmo Market Idu of FCT-Abuja, Nigeria.

MATERIALS AND METHODS

Collection of the Herbal Medicines

Total of ten (10) different local herbal medicines were purchased at the sales-points of some markets within FCT, Abuja-Nigeria after being identified by Mallam Muazzam and Lateef Akeem of Department of Medicinal Plant Research and Traditional Medicine, National Institute for Pharmaceutical Research and Development (NIPRD), Abuja.

Determination of Moisture Content

The determination of moisture content was based on the method of the Association of Official Analytical Chemist (AOAC) [13]. Using a pan of pre-dried Automated Moisture Meter analyzer (OHAUS, MB200) which was electrically powered, calibrated and set at the temperature of 105°C and 180 minutes. The results which automatically displayed after time elapsed were recorded for each of the samples which were analyzed in triplicate and the mean obtained.

Determination of Total Ash Content

Porcelain crucible was washed and placed in the muffle furnace for 10 minutes and dried, removed and placed in a desiccator to avoid moisture contact and was cooled. After cooling, it was

weighed (M₁). 2g of each of the sample were weighed in the crucibles (M₂) and was placed in the Muffle furnace at a 550°C and allowed to stand 3 hours for complete combustion to ash to be achieved. The ash samples were removed placed in the desiccators for cooling and the weighed (M₃) [13].

Sample Digestion for Mineral Element Determination

The procedure described by Samali *et al*, [14], quantity of 0.5 g of the finely powdered sample was weighed and digested into a digestion flask using nitric acid and perchloric acid (7:3). The digested sample was made-up to mark in 50 mL volumetric with deionized water and was analyzed using Atomic Absorption Spectrophotometer (Model:GBC Avanta) after equipment calibration with reference standard (BDH) solutions and blank sample runs.

Procedure for Elemental Analysis

For the determination of Cu, Cr, Fe, Pb, Mn, Ni and Zn in the samples. The Hitachi Model 80-80 polarize Zeeman atomic absorption spectroscopy (AAS) instrument was optimized and calibrated using standard solution of the elements of interest based on the operating conditions in Table 1 followed by sample analysis. The data obtained were processed using relation:

$Metal (\mu g/g) = \underline{C \times V \times d.f}$

W (g)

Where; C is the concentration obtained from the Atomic Emission Spectrophotometer (mg/L); V is the volume of the undiluted sample solutions in mL; W is the sample's weight in grams and d.f is the dilution factor.

Element	wavelength	Flame type	Slit width	Working concentration range (µg/mL)	R ²
Cu	324.70	Air-acetylene	0.5	0.5-8.0	0.997
Cr	357.9	Air-acetylene	0.5	0.5-8.0	0.978
Fe	248.30	Air-acetylene	0.2	1.5-24.0	0.979
Pb	217.0nm	Air-acetylene	0.5	0.625-20.00	0.998
Mn	279.5nm	Air-acetylene	0.2	0.625-10.00	0.997
Ni	232nm	Air-acetylene	0.2	0.625-20.00	0.997
Zn	213.9nm	Air-acetylene	0.5	0.5-8.0	0.987

Table 1: Instrument Operating Condition for the Analysis.

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Sample Extraction Procedures

The sample extraction process used was cold maceration of each powdered sample (100 gram) in 70% hydro ethanol 1000 mL and kept at room temperature for 72 hours and filtered through a muslin cloth, followed by Whatman's filter paper no.1 and concentrated in a Rotary evaporator (Bibby, Germany) and further concentrated over

a water bath (Karl Kolb, Germany) at 40 C. To dryness and store in a refrigerator.

Chromatographic fingerprints of the extract

The extracts were spotted on analytical TLC plates of silica gel G60 F254, 0.25 mm layer and developed in a chromo-tank containing solvent systems of different mixtures from which solvent systems that gave best resolutions were identified and utilized for the study. The developed spots of the constituents on the TLC plates were detected

 Table 2: Physicochemical and Mineral Content.

by visualization under ultra violet light (UV) at both 256 nm and 366 nm after spraying with vanillin sulfuric,,Ferric chloridde,the mixture of anisaldehyde (0.5 ml), glacial acetic (10ml), methanol (85 ml) and concentrated sulphuric acid (0.5 ml) and dried at 100-110°C for 5-10 minutes. The retention factor (Rf)-values of the identified spots were calculated and recorded Biradar *et al.*, [15 -17].

Rf =

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RESULTS

Result of moisture, ash and mineral content were reported in Table 2, while the attempt to provide TLC fingerprints for the herbs, Rf-values were reported in Table 3.

Samples	Moisture %	Ash							
code	w/w %w/w	% w/w	Cu	Cr	Mn	Ni	Pb	Fe	Zn
PN	8.26	3.79	0.56	1.15	18.20	0.44	0.05	40.17	5.12
СВ	9.32	13.93	0.18	1.18	1.45	0.15	0.15	11.71	2.57
AM	10.30	12.00	0.34	0.81	9.13	0.24	0.07	9.23	1.78
CA	20.07	10.68	0.27	0.33	36.21	0.29	0.09	3.06	6.91
DM	8.29	10.36	0.14	0.93	7.54	0.37	0.09	3.41	1.85
HR	11.38	25.84	0.37	3.70	9.78	0.69	0.15	18.00	3.81
HTF	14.25	21.21	0.44	0.51	3.24	0.75	0.08	18.80	2.70

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ML	8.70	17.12	0.17	1.04	15.60	0.49	0.07	18.86	3.28
IS	10.07	21.16	0.20	1.14	7.26	0.72	0.13	12.97	6.06
AL	7.30	5.66	0.13	0.25	5.31	0.79	0.18	0.39	1.05

Table 2: TLC Fingerprint Profile of the Medicinal Plants

Sample Code	Solvents	Sports	Retention RF	Colour	Det	Inference		
Code			Kſ	Visible	Detection (UV265n m)	Vanillin sulfuric	Feric chloride	Inference
AM	Hex/Ethyl acetate (60:40)	5	0.94 0.82 0.48 0.34 0.18	Purple blue brown brown brown	- - fluores- cence brown brown	Violet violet blue/grren blue/grren brown	yellow blue/gree n brown brown	Terpenes flavonoids Polyphenols
CA	Hex/Ethyl acetate (80:20)	6	0.96 0.90 0.80 0.76 0.30 0.14	purple purple Pink blue brown brown	Fluores- cence - - grey brown brown	violet/pink grey black brown brown	yellow blue/gree nbrown brown brown	terpenoids flavonoids Polyphenols Tannins
AL	Ethyl ace- tate/ MeOH/ H ₂ O (7:2:1	5	0.90 0.80 0.60 0.46 0.21	yellow yellow blue blue brown	- - Fluores- cence brown	violet/pink grey black brown	yellow blue/gree n brown brown	terpenoids flavonoids tannins

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СВ	Ethyl ace-	4	0.86	pink	-	violet pink	yellow	Terpenoids
	tate/		0.70	pink	-	grey	blue	Flavonoids
	MeOH/		0.63	blue	Fluores-		green	
	H ₂ O		0.50	blue	cence			
	(7:2:1)							
DM	Ethyl ace-	5	0.93	yellow	-	violet/pink	yellow	terpenoids
	tate/		0.89	yellow	-	blue/green	blue/gree	flavonoids
	MeOH/		0.73	pink	-		n	Polyphe-
	H ₂ O		0.63	blue	blue		brown	nolstannins
	(7:2:1)		0.50	blue	fluores-			
					cence			
	Ethyl ace-	4	0.69	pink	-	grey	yellow	Flavonoids
HR	tate/		0.60	pink	-	blue/green	blue/gree	Polyphenols
	MeOH/		0.45	blue	-blue		n	Brown
	H ₂ O		0.27	brown	Fluores-		brown	
	(7:2:1)				cence			
HTF	Ethyl ace-	7	0.84	yellow	-	violet/pink	yellow	Flavonoids
	tate/		0.77	pink	-	grey	blue/gree	polyphenols
	MeOH/		0.73	pink	-	black	n	tannins
	H ₂ O		0.49	blue	blue	brown	brown	
	(7:2:1)		0.40	blue	fluores-	brown		
			0.25	brown	cence			
			0.13	brown	brown			
ML	Ethyl ace-	4	0.69	pink	blue/fluo-	pink	blue/gree	Terpenes
	tate/		0.60	pink	rescence	blue/green	n	flavonoids
	MeOH/		0.45	blue		blue		polyphenols
	H ₂ O		0.27	brown		brown		tannins
	(7:2:1)							
L	1				1	1	1	

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ISI	Ethyl ace-	5	0.88	-	Fluores-	violet	yellow	Terpenes
	tate/ CCl4/		0.77	pink	cence	blue/green	blue/gree	flavonoids
	MeOH/H ₂		0.55	blue	-	brown	n	polyphenols
	0		0.32	brown	-		brown	
	(8:4:2:0.5)		0.26	brown	brown			
					brown			
PN	Ethyl ace-	4	0.75	Yellow	-	pink	yellow	Terpenoids
	tate/ CCl4/		0.65	Pink	-	blue/green	blue/gree	flavonoids
	MeOH/H ₂		0.53	blue	Fluores-	brown	n	polyphenols
	0		0.38	brown	cence		brown	tannins
	(8:4:2:0.5)				brown			

Discussion

The moisture content obtained from the samples (Table 1) ranges from 7.30 to 20.07% with only 10 % of the total sample CA failed the WHO maximum standard permissible limit (\leq 15). The total ash content of the samples ranges from 3.79 to 25.84 % w/w which indicated 80% of the samples; CB, AM, CA, DM HR, HTF ML, IS, AL failed the requirement of WHO maximum permissible limits (\leq 8.0) [18], the gross failure of the samples for total ash, this could be due to contamination of the samples by dust, sand and other inorganic materials due to crowded nature of the Karmo market and other local markets where the samples were purchased.

Mineral content of the samples indicated the presence of both the essential (Cu, Cr, Fe, Ni, Mn, Zn and the non-essential minerals (Pb) at variable concentrations. The essential minerals are usually required by the body at variable concentrations by different age groups and sexes [20] for several metabolic processes, while the non-essential ones are toxic to the body system even at trace level [18]. Result of mineral concentrations of the toxic element (Pb) obtained in all the samples (Table 1) were below the WHO Maximum permissible limit ($10\mu g/g$) [19]while the concentration of the essential minerals which ranges as Cu (0.14-0.56 $\mu g/g$), Cr (0.25-3.70 $\mu g/g$), Fe (0.39 – 40.17 $\mu g/g$), Mn (1.45-36.21 $\mu g/g$), Ni (0.15-0.79 $\mu g/g$) and Zn (1.05-6.91 $\mu g/g$) were determined by the required daily allowance [19] of individual age group and sex [18], and unregulated consumption could pose health hazard over time due to bioaccumulation.[12,25]

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Fingerprinting is now globally accepted by WHO as a quality evaluation parameter of herbal medicine. Fingerprint construction has become an important quality control tool of herbal samples in the light of constantly growing interest in natural medicines [20]. It is applied to identify

closely related plant species, to detect adulterations, to control the extraction process or to study the quality of a finished product. Chromatographic fingerprint of phytomedicine can be referred to as a set of characteristic chromatographic or spectroscopic signals, whose comparison leads to sample recognition [21]. The summary of the TLC profiling of the medicinal plants extracts indicated Rf-values of different phytocomponents from solvent systems that gave better separations and more components are reported in Table 2. The Ethyl acetate/ MeOH/ H_2O (7:2:1) solvent system is the best solvent system that favors better separation for sample AL, CB, DM, HR, HTF and ML and multi-component of the herbal medicine evaluated indicated seven (7), five (5) and four (4) spots in 60% of the samples which the Rf-values ranges from 0.13-0.84 (HTF), 0.50-0.93 (DM), 0.18-0.94 (AL), 0.50-0.86 (CB), 0.18-0.85 (HR) and 0.27-0.71 (ML); the second best solvent system was Ethyl acetate/ CC14/ MeOH/H2O (8:4:2:0.5) for IS and PN samples indicated five (5) and four (4) spots which Rf-values ranges from 0.38 to 0.75 and 0.26 to 0.88, while Hex/Ethyl acetate (80:20) solvent system favors CA sample which indicated six (6) spots with Rf-values of 0.14 to 0.96 and Hex/Ethyl acetate (60:40) solvent system favors AM which shows five (5) spots with Rf-values of 0.18 to 0.94 respectively. Previous studies of different solvent (chloroform, methanol and water) extracts of Nigerian medicinal plants have reported Rf-values for flavonoids (0.74, 0.86, 0.8, and 0.92), alkaloids (0.25, 0.56, and 0.92), tannins (0.85, 0.92) and phenol (0.8) [22]. This confirmed that there are presences of these phytochemicals in the herbal medicine evaluated in this study. According to American Herbal Pharmacopoeia (AHP), the use of single or multiple chemical markers was important to quality control [23-24].Our attempt for this samples gave us insight to group physical maker as ash, moisture ,the TLC finger print can also serve as a quality maker for qualitative control. Polarity of compounds and solvents influence Rf-values of compounds [24].This consist of all the strategies of quality of herbal products[25-26] to give standardized quality marker and products.

Conclusion

The quality of herbal medicine requires routine monitoring in order to ensure consistency, safety and efficacy are sustained to avert sample degradation and contamination which could pose health hazard to the consumers.

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