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Levels of Pathogens, Heavy Metals, Essential Ions and Organochlorides in Herbal Medicines and Plant Species Used for Hepatic Diseases in Ghana

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ABSTRACT

Background and Objective: Herbal medicines are raw or processed plant parts used singly or in combination for management of diseases including hepatic or liver diseases. Herbal medicines have become the primary option for the treatment of liver illnesses because of their safety, availability and less side effects. Inappropriate sourcing of raw plant materials, non-adherence to good manufacturing practices and climate-induced endogenous toxins biosynthesis have resulted in the contamination of herbal products with pathogens and toxins. The objective of the study was to determine the levels of pathogenic bacteria, heavy metals, essential ions and organochlorides in common medicinal plants and their medicinal products. **Materials and Methods:** In all, 10 herbal drugs meant for liver diseases were purchased from herbal shops, clinics and practitioners in Tamale, Bolga, Wa, Kumasi and Ashaiman. Plant species and parts used in manufacturing the medicines were compiled. The plants were sourced from Gumbini, Forestry Commission Yard in Tamale and aqueous extraction was done. The pH, total dissolved solids (TDS), Cadmium (Cd), lead (Pb), Magnesium (Mg), Calcium (Ca), Aldrin, Dieldrin, Endrin, Endosulfan, microbial culture and sensitivity were determined. **Results:** There was no bacteria growth in the plant extracts. *Klebsiella* spp. were isolated from 4/10 (40%) of the products and were resistant to trimethoxyprinem/sulfohyoxozole. Only two product samples were above the WHO recommended level for lead and Aldrin. There was no significant difference between the plant extracts and their medicinal product levels of these markers except pH ($p = 0.032$). **Conclusion:** Raw materials should be tested and good manufacturing practices including shelving be adhered to avoid contamination.

KEYWORDS

Aldrins, endrins, endosulphans, organochlorides, hepatic diseases

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INTRODUCTION

Herbal medicinal drugs (HMDs) are plant, phytochemical, or botanical products used for alleviation of diseases[1,2]. They are also known as botanical medicine, phytochemical drugs, Ayurveda medicine and traditional or complementary medicines[3,4]. Herbal medicines include finished, raw, semi-processed parts of finished or whole plants[5].

Herbal medicines have been used for the past 2000 years to treat various human diseases including microbial infections, dyslipidaemia, hypertension, diabetes, stroke, convulsions, mental disorders and insomnia[6]. In ancient medicine, *Rauwolfia vomitoria* was used to treat hypertension, stroke and insomnia in Nigeria[6].

The World Health Organization (WHO) estimates 70-85% of Sub-Saharan Africans depend on herbal medicines for the management of primary healthcare conditions[7]. In China, 5000 medicinal herbs have been reported for the treatment of human diseases with 40% of the population depending on herbal medicines for management of primary healthcare[8]. India has 160 phytochemical products from 101 plants of which 33 are patented[9,10]. The market value of herbal medicine in 2012 was 87 billion United State dollars and is projected to reach 120 billion in 2020[11,12].

Hepatic diseases are conditions that affect the structure and functions of the human liver[13]. They include hepatitis, cirrhosis, jaundice, cholestasis, hepatobillary disease and steatosis[13]. The causes of hepatic disease include microbial pathogens and their toxins e.g, aflatoxin, viruses, substance abuse e.g. alcohol, pharmaceutical drugs, industrial chemicals, herbal drugs and ionic radiations[14].

Climate-induced toxins biosynthesis by medicinal plants for defense also results in liver diseases[15]. Adulteration of herbal medicinal drugs with pharmaceutical drugs, unrelated herbal products and industrial chemicals to enhance their taste, colour and texture also cause liver injury[16].

Sourcing of raw materials from slumps, mining areas and dumpy grounds also contaminates the products with heavy metals, microbial toxins and organophosphate chloride residues[17]. Lack of standardisation and clinical trials during the production of herbal medicines can produce products with levels of bioactive compounds beyond their lethal doses[18]. Lack of adherence to good manufacturing practices during sampling, production, packaging and storage leads to microbial contamination[19].

In a study on 4 heavy metals, lead (Pb), Cadmium (Cd), Arsenic (As) and Mercury (Hg), in 10 Ghanaian herbal plants, grown in 5 different locations, the levels of the heavy metals varied[20]. The Pb was found in 44% of the samples, as was found in 2(4%) of the plants[21]. In a similar study on 14 Chinese medical herbal products including *Ginseng radix*, DDT was found in 14 of them[22]. A study in Zambia reported high levels of DDT residues in vegetables[23]. Studies in Nigeria also reported high levels of cadmium, copper, iron, nickel, selenium, zinc, lead and mercury in random samples of traditional medicine[24]. High levels of Pb, Cd and chromium were found in herbs sourced from samples grown in polluted lands than in unpolluted lands[25].

Indicators of contaminants in herbal products include pH, total dissolved solids (TDS), heavy metals, essential ions, organochlorides and microbial pathogens[26]. Organochlorides are found in insecticides, rodenticides, solvents, fumigants and pesticides and are found in DDT and heptachlor[27]. It gets into the body through inhalation, ingestion and skin contact[28]. Organochlorides are chloride antagonists thereby inhibiting Ca^{2+} and Mg^{2+} ATPases which are enzymes involved in nerve impulse transmission[29]. This results in Ca^{2+} accumulation at nerve end plates triggering sustained release of excitory neurotransmitters[30]. Organochlorides and heavy metals induce oxidative stress by covalently binding to superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and glutathione and inactivating them[31]. Organochlorides also alter the transcription of genes involved in detoxification and stress response[32].

The pH is an indication of the acidity or alkalinity of a product[33]. Acidity or alkalinity is a factor of phytochemical composition and nature, therefore a measure of deterioration[34]. Most herbal products are in the acidic range because of the flavonoids[35]. Therefore, a shift toward alkalinity is an indication of phytochemical decomposition[36]. Total dissolved solids (TDS) measure the soluble ionic composition of the products[37]. The TDS is indirect measure that indicates of the ionic quality of the water as well as deterioration in shelves[38].

The objective of the study was to determine the level of microbial pathogens, heavy metals, water quality markers, organochloride markers and essential ions in herbal medicinal products and herbal plants used as raw materials.

MATERIALS AND METHODS

Selection and confirmation of herbal medicinal products: The methods followed protocols applied by Mintah *et al*[39]. Herbal medicinal products were purchased from major herbal shops, markets and clinics in Tamale, Sawla, Kumasi and Ashiaman. The plant species used in the manufacturing were compiled from the package information. Inclusion criteria were available of Food and Drugs Registration number, company batch number, not expired and diseases targeted should include a liver disease. Excluded were medicines not meant for any liver diseases, expired and no verifiable FDA and company batch number.

Collection and identification of the herbal Plants materials: The plants sourced were obtained at Regional Forestry Commission yard, Gumani, Tamale, the Department of Parks and Gardens yard, Waterworks, Tamale and the Tamale Central Market. The plants were identified by Botanist at the Department of Botany, University for Development Studies, Nyankpala. The study was carried out at the Central Laboratory of Kwame Nkrumah University of Science and Technology from June 2021 to December, 2022.

Solvent extraction of plant materials: For herbal medicines, a 10 mL sample of each of 10 herbal products was taken into sterile containers for laboratory analysis at Tamale Teaching Hospital and Central Laboratory of Kwame Nkrumah University of Science and Technology.

For the plant materials, for the hydroethanolic and methanolic extracts, 100 g of powdered shade-dried plant was socked in 70% ethanol and methanol respectively for 72 hrs with regular vortexing. The mixture was filtered with Whatman filter paper after which the filtrate was concentrated with rotar evaporator (IKA Heindolph, Yamato Scientific Co. The LTD, Japan) at 70°C until all the solvent evaporated. The condensate was dried in an oven at 50°C till a solid extract was obtained for analysis.

Determination of the presence and sensitivity of pathogenic microbes: Samples were inoculated in enrichment media which was composed of blood agar, mannitol salt agar and chocolate gar and incubated for 24 hrs before being plated on MacConkey media with a sterile lobe and incubated for 24 hrs at 37°C. Gram staining was followed using crystal violet, iodine solution and neutral red. Biochemistry test was then followed using indole, citrate, urea and triple sugar ions (TSI) where there was significant growth. Disc diffusion method was used on Mueller Hinton Agar was then used for sensitivity with McFarland standard which is a mixture of 0.1 8 M H_2SO_4 and 0.048 M $BaCl_2$. The zones of inhibition of the extracts were measured and compared to the standard.

Determination of pH and total dissolve solids: An electronic pH meter (Telatemp Corporation, New York, USA) was used for the determination of the pH. It also consists of a hydrogen ion-sensitive glass electrode, which measure the pH corresponding relative to the difference between the internal electrode and reference electrode. The probe was cleaned in deionised water before immersed into the samples after uniform mixing.

For the total dissolved solids (TDS), a cleaned and dried evaporating dish was weighted. A 50 mL sample was taken after uniform mixing and filtered with cleaned and dried filter paper. The sample was filtered three times after cleaning the filter each time. The filtrate was then transferred to the evaporating disk using a pipette and dried in an oven. The weight of the dried evaporating dish with the dried residue was taken. The dissolved solids (mg/L) were calculated from the formula: 1000(A-B)/mL[39] sample, where A is the weight of the evaporating dish with the residue and B is the weight of the disk.

Determination of essential ions and heavy metals: An inducible coupled spectrophotometry was used for the analysis of magnesium, lead, chromium and calcium. The procedure involved harmonization of the plant samples, preparation of the working solutions, sample analysis and validation. For the sample digestion and analysis, for each plant and herbal product, 1 g of the sample was placed in a 100 mL beaker and an acid mixture of 4 mL of H_2SO_4, 2 mL of $HClO_4$ and 2 mL of HNO_3 was added. The mixture was heated to 250°C and left at this temperature until a clear solution was obtained. The clear solution was then filtered into a 100 mL volumetric flask using a 0.45 pm pore size membrane filter paper (Whatman filter paper No. 41). The filtrate was then topped to the mark with distilled water. Digested samples were transferred into plastic bottles and stored at 4°C for analysis. Samples were analysed along standards at wavelengths of 228.0 and 283.0 nm, respectively.

Determination of organochlorides: The organochloride markers aldrin, eldrin, endosulphan and dieldrin were measured using a gas chromatograph Agilent 7890C GC (Agilent Technologies, California, USA) with a triple quadrupole mass spectrometer Agilent 7000C MS (Agilent Technologies, California, USA). For quality assurance and control of the results, standards were obtained at the Pesticide Residues Laboratory, Ghana Standards Authority which is an ISO/IEC 17,025: 2017 accredited laboratory for determination of toxicants in fruits, vegetables and cereals and run along the samples. The concentrations of the extracts were calculated by comparing the peak area of the analytes with an area of the standards.

Statistical analysis: Data was inputted in Excel version 16 (Microsoft INC, New York, USA). Discrete data was recorded as a percentage and continuous data as Mean±SD.

RESULTS

pH and TDS of the plants and the herbal medicines: The pH of the herbal drugs and that of the extracts of the plants were determined and indicated in Fig. 1. The pH of drug samples ranges from 2.79 to 4.41 while that of the plant extracts ranges from 3.18 to 5.25. Both pH of drugs and plant extract were in the acidic range.

TDS pattern between the drug samples and the plant extracts: Total dissolved solids concentration was measured in the herbal drug samples and their plant extracts and displayed in Fig. 2. The TDS of medicines ranges from 148 mg/kg to 1259.5 mg/kg. The TDS of the plant extracts ranges from 6.1 mg/kg to 1200 mg/kg. The WHO recommended limits for TDS in water and water-based products are below 300 mg/kg. All the plant extracts were above this limit whereas 6/10 (60%) of the products were below this range.

Level of essential ions and heavy metals in the drugs and the plants: The essential ions Mg^{2+} and Ca^{2+} and heavy metal Pb and Cd were determined and displayed in Table 1-4.

Distributions of magnesium (Mg): Table 1 indicates the Mg^{2+} distribution between the drug and plant extracts. From Table 1, the Mg^{2+} concentration of the drugs ranges from 6.88±0.01 to 8.42±0.02 mg/L while those of the drug samples range from 6.92±0.002 to 8.22±0.01 mg/L. The US Environmental Protection Agency limits for Ca^{2+} and Mg^{2+} is 0.3 to 135 mg/L. Both medicines and extracts are within this range for magnesium. For calcium, 1/10 (10%) of each of medicines and extracts were below this range.

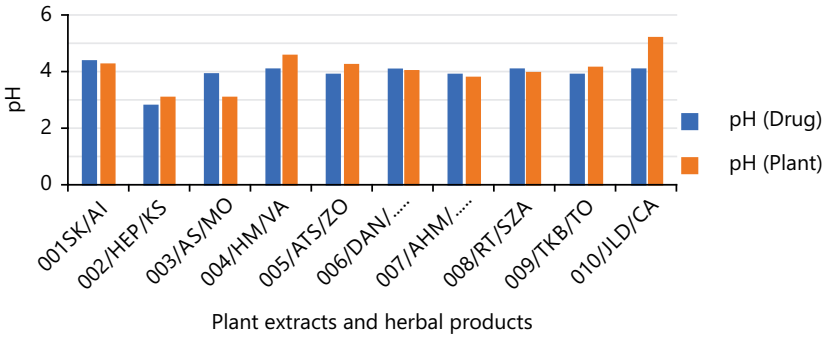

Fig. 1: pH variation between the medicinal products and the plant extracts

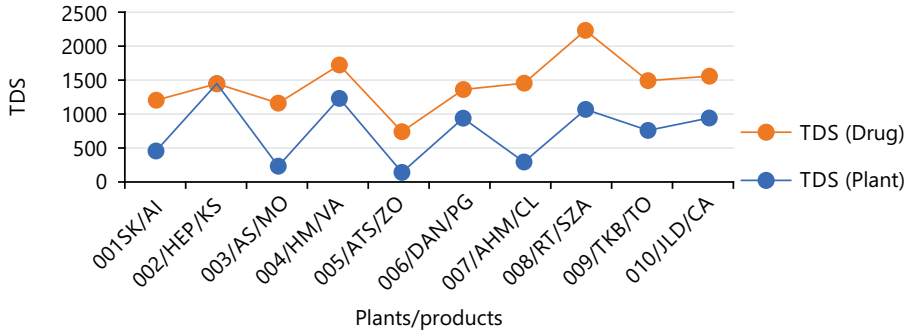

Fig. 2: Total dissolved solids concentrations in the products and extracts

Table 1: Magnesium (Mg) concentration in the products and extracts

Sample	Mg (products)/mg/L	Mg (plant) mg/L
001/SK/AI	7.71±0.02	7.67±0.12
002/HEP/KS	8.42±0.02	7.92±0.02
003/AS/MO	7.93±0.01	7.79±0.02
004/HM/VA	7.23±0.02	7.23±0.02
005/ATS/ZO	6.88±0.01	7.40±0.01
006/DAN/PG	7.69±0.01	6.92±0.02
007/AHM/CL	7.47±0.02	8.22±0.10
008/RT/SZA	8.03±0.00	7.58 ±0.01
009/TKB/TO	7.76±0.01	7.37±0.02
010/JLD/CA	7.58±0.14	7.73±0.02

Table 2: Calcium (Ca) distribution within the drug samples and the plant extracts

Sample	Ca (products)/mg/L	Ca (plant) mg/L
001/SK/AI	1.27±0.17	3.51±0.17
002/HEP/KS	1.50±0.02	6.07±0.25
003/AS/MO	0.98±0.03	3.26±0.12
004/HM/VA	0.32±0.00	0.35±0.01
005/ATS/ZO	0.03±0.01	0.03±0.00
006/DAN/PG	1.10±0.01	0.31±0.01
007/AHM/CL	0.97±0.06	0.57±0.01
008/RT/SZA	1.04±0.02	0.04±0.00
009/TKB/TO	2.59±0.12	2.23±0.05
010/JLD/CA	0.66±0.02	1.03±0.05

Table 3: Lead (Pb) concentration among the plant extracts and the drug samples

Sample	Pb (products)/mg/L	Pb (plant) mg/L
001/SK/AI	1.39±0.02	0.92±0.03
002/HEP/KS	1.41±0.02	1.01±0.02
003/AS/MO	1.41±0.02	1.05±0.01
004/HM/VA	1.44±0.02	1.06±0.04
005/ATS/ZO	1.47±0.01	1.10±0.03
006/DAN/PG	1.45±0.03	1.16±0.03
007/AHM/CL	1.50±0.09	1.21±0.02
008/RT/SZA	1.48±0.03	1.25±0.02
009/TKB/TO	1.50±0.01	1.31±0.01
010/JLD/CA	1.39±0.02	1.35±0.04

Table 4: Level of cadmium in the drugs and plant extracts

Sample	Cd (product)/mg/L	Cd (plant) mg/L
001/SK/AI	BDL	BDL
002/HEP/KS	0.003	BDL
003/AS/MO	BDL	BDL
004/HM/VA	BDL	BDL
005/ATS/ZO	BDL	0.004
006/DAN/PG	BDL	BDL
007/AHM/CL	BDL	BDL
008/RT/SZA	BDL	0.004
009/TKB/TO	BDL	0.005
010/JLD/CA	0.004	0.003

BDL: Below detectable level implies when level is below 0.001mg/L

Variation of calcium among the plants and the drugs: The variations in the Ca^{2+} concentration between the drug and plant extracts are indicated in Table 2. The Ca^{2+} concentration of the drugs ranges from 0.03±0.01 to 2.59±0.02 mg/L while those of the drug samples range from 0.04±0.00 to 6.07±0.25 mg/L.

Level of lead (Pb) among the plants and the medicinal drugs: Table 3, indicates the Pb levels among the drug samples and the plant extracts. The Pb concentration of the drugs ranges from 1.39±0.02 mg/L to 1.50±0.09 mg/L while those of the drug samples range from 0.92±0.03 to 1.35±0.04 mg/L. The US EPA recommends zero lead levels in medicines and water while the EU/WHO limit is below 1.00 mg/L. Whereas all the medicinal products were above this range, 90% of extracts were within this limit.

Levels of cadmium among the plants and the products: Detectable cadmium levels were found in 2/10 (20%) of the herbal drugs and 4/10 (40%) in plant extracts. All the herbal products as well as the plant extracts were within the WHO limits of 0.003 mg/Kg for cadmium. However, 1/10 (10%) of herbal products and 3/10 (30%) of plant extracts had cadmium levels higher than the US/EU acceptable limits of 0.001 mg/Kg.

Variation of organochlorides among the herbal products and plants: The concentrations of the soluble organochloride phosphates (OCPs) markers, dieldrin, endrin and endosulfan were measured and indicated in Table 5-8. The EU/US EPA recommended limit for aldrin, endrin, dieldrin and endosulfan (alpha, beta and sulphate) is below 2.0 µg/L. All the samples from the plant extracts and medicines were within this range.

Levels of soluble aldrin and dieldrin among the herbal drugs and the plant extracts: The concentration of aldrin in the herbal drug samples ranges from non-detectable limits up to 1.38±0.02 µg/dL and up to 1.48±0.04 µg/gL in the plant extracts.

Table 5: Soluble aldrin concentrations among the herbal products and the plant extracts

	Organochlorides	
Sample	Aldrin (drug) µg/dL	Aldrin (plant) µg/dL
001/SK/AI	BDL	1.03±0.01
002/HEP/KS	1.03±0.02	1.05±0.01
003/AS/MO	1.04±0.02	1.05±0.02
004/HM/VA	1.04±0.01	1.04±0.02
005/ATS/ZO	1.04±0.02	BDL
006/DAN/PG	1.03±0.02	BDL
007/AHM/CL	BDL	1.04±0.01
008/RT/SZA	BDL	BDL
009/TKB/TO	BDL	BDL
010/JLD/CA	BDL	1.10±0.01

BDL: Below detectable limited, that of aldrin below 0.1µg/dL

Table 6: Dieldrin concentrations among the herbal products and plant extracts

	Organochlorides	
Sample	Dieldrins (products) µg/dL	Dieldrins(plant) µg/Dl
001/SK/AI	BDL	0.92±0.03
002/HEP/KS	1.79 ± 0.01	1.01±0.02
003/AS/MO	BDL	1.05±0.01
004/HM/VA	BDL	1.06±0.04
005/ATS/ZO	BDL	1.10±0.03
006/DAN/PG	BDL	1.16±0.03
007/AHM/CL	BDL	1.21±0.02
008/RT/SZA	BDL	1.25±0.02
009/TKB/TO	BDL	1.31±0.01
010/JLD/CA	BDL	1.35±0.04

BDL: Below detectable limited, that of aldrin below 0.1µg/dL

Table 7: Endrin concentration in the herbal products and plant extracts

Plant material/product	Endrin (drug) mg/L	Endrin (plants) mg/L
001/SK/AI	1.83±0.01	1.72±0.04
002/HEP/KS	1.72±0.03	1.69±0.07
003/AS/MO	1.72±0.03	1.79±0.03
004/HM/VA	1.70±0.04	1.72±0.02
005/ATS/ZO	1.72±0.03	1.72±0.01
006/DAN/PG	1.70±0.05	1.72±0.03
007/AHM/CL	1.72±0.03	1.74±0.03
008/RT/SZA	1.72±0.01	1.72±0.06
009/TKB/TO	1.73±0.02	1.70±0.03
010/JLD/CA	1.72±0.06	1.70±0.06

Table 8: Endosulphan concentration in the herbal drug and plant extracts

Sample number	Endosulphans (drug) mg/L	Endosulphans (plants) mg/L
001/SK/AI	1.87±0.03	1.86±0.01
002/HEP/KS	1.84±0.06	1.80±0.06
003/AS/MO	1.86±0.03	1.76±0.01
004/HM/VA	1.66±0.01	1.81±0.03
005/ATS/ZO	1.86±0.02	1.84±0.03
006/DAN/PG	1.87±0.01	1.86±0.02
007/AHM/CL	1.86±0.03	1.76±0.01
008/RT/SZA	1.56±0.03	1.90±0.03
009/TKB/TO	1.56±0.01	1.86±0.02
010/JLD/CA	1.86±0.06	1.76±0.03

Table 9: Presence and sensitivity of pathogenic bacteria in the herbal products

Product	Bacteria isolated	Drug sensitivity	Drug resistance
001SK	*Klebsiella* spp	Azm	Mem, tri/sxt, cip,azm
002/HEP	NBG	NIL	NIL
003/AS	*Klebsiella* spp	Sxt	mem, gen,Azm, cip,amc
004/HM	NBG	NIL	NIL
005/ATS	*Klebsiella* spp	Sxt	Gen, mem, cip,azm, amc
006/DAN	NBG	NIL	NIL
007/AHM	NBG	NIL	NIL
008/RT	NBG		NIL
009/TKB	NBG	NIL	Gen, mem, cip,azm, amc
010/JLD	*Klebsiella* spp	Sxt	NIL

Azm: Azithromycin, Mem: Meropenem, tri/sxt: Trimethoprim/sulfamethoxazole, cip: Ciptoflaxcin, amc: Amycacin, gen: Gentamycin and NBG: No bacterial growth

From Table 5 and 6, more drug and plant extracts (11/20, 55.0%) contain detectable soluble aldrin than dieldrin (2/20, 10%). Only 1 of the medicine products had dieldrin above detectable limits (1.79±0.01 µg/dL) whereas the plant extracts had dieldrin levels between 0.92±0.03-1.35±0.04 µg/dL.

The concentration of endrin in the herbal drug samples ranged from 1.70 to 1.83 mg/L while endrin concentration in the plant extracts ranged from 1.69 to 1.79 mg/L. There was no significant difference between the endrin concentration of the herbal drug and plant extracts.

Presence, sensitivity and resistance pattern of clinically significant pathogens in the samples: Culture and sensitivity were performed to identify waterborne pathogenic bacteria in the samples. Resistance and sensitivity were determined by the diameter of the zone of inhibition compared to the MacFarland standard. There was no growth of pathogenic bacteria of clinical significance among the 10 plant extracts. *Klebsiella* species were isolated in 4/10 (40%) of the herbal drug samples while *Klebsiella oxytoca* was isolated in 1/10 (10%) of herbal drug samples. All the isolates in the 5 products were sensitive to meropenem (mem), ciprofloxacin (cip) and amoxicillin (amc) while 4 were sensitive to both gentamycin (Gen) and azithromycin (Azm). Two isolates were sensitive to trimethoprim/sulfamethoxozole. For resistance, the combined Three of the five isolates were resistant to trimethoprim/sulfamethoxozole while 1 isolate was resistant to azithromycin.

DISCUSSION

The objective of the study was to determine whether the level of pathogenic bacteria, toxic heavy metals, organochlorides residues (OCPs) and essential ions in herbal products and the unprocessed plants extracts are within acceptable limits. An ideal hepatoregenerative agent should contain physiological tolerant limits of heavy metals, organochloride and pathogens in order to avoid herbal-induced liver injury.

No bacteria of clinical significance were isolated in the plant extracts whereas *Klebsiella* species and *Klebsiella oxytoca* were isolated in 5 of the 10 herbal drug samples. The isolates were sensitive to 5 out 6 antibiotics tested and were resistant to combined trimethoxyprinem/sulfmethoxazole by *Klebsiella* spp. and azithromycin by *oxytoca*. *Klebsiella* species are water and food-borne bacteria that cause pneumonia, skin rashes, conjunctivitis and gastroenteritis[40]. In the absence of exogenous contamination, crude plant extracts exhibit antimicrobial activities[41]. Plant phytochemicals such as flavonoids inhibit beta-lactamases which are responsible for the synthesis of bacteria cell walls thereby inhibiting bacterial growth and action[42]. Tannins have also been reported to inhibits bacterial ribosome subunit through covalent binding which inhibit protein synthesis in bacteria[42]. *Klebsiella Pneumoniae*, *Staphylococcus aureus* and *Pseudomonas aeruginosa* were isolated from in herbal samples which were sensitive to 10 µg/dL of

ampicillin, penicillin and tetracyclin[43]. The products were purchased from various retail outlets, therefore, transportation, packaging and shelving could have contaminated these bacteria. Even though crude extracts exhibit antimicrobial activity, long shelve duration and exposure to harsh weather conditions result in decomposition of phytochemicals leading to loss of antimicrobial potential.

The plant extracts and medicinal products had pH in the acidic range. The major contributor to acidity in phytochemicals is flavonoids due to their abundant polyhydroxy groups. Flavonoids also have high antioxidant activity which attenuates oxidative stress. Oxidative stress is the fulcrum on which pathophysiology of liver diseases revolts. Therefore, suggests the presence of high flavonoids in the plant extracts and medicinal products. In phytochemical biotransformation, the pH moves towards the neutral to alkaline region (even though this depends on the composition). The inclusion criteria for the products included non-expiration. This also suggests possible minimal biotransformation or degradation of the medicinal products.

All the plant extracts had TDS above the recommended levels whereas the majority (60%) of the herbal medicines had TDS within the recommended levels. The TDS measures the chemical composition of constituting water and is principally determined by magnesium, calcium, arsenic, lead and nitrates. Absorption from soil nutrients is the predominant source of dissolved ions for plants. The variation in the source of raw materials for the herbal products and the fresh plant materials could account for the differences in the TDS level. Processing of herbal medicines also involves ion exchange and chelating which reduce the ionic composition of the finished products. This could also account for the comparatively lower levels of TDS level in medicines than extracts.

All the medicines and 80% of the plant extracts had high lead levels above the recommended levels. Lead is a ubiquitous metal that exist in various ionic states and obtained from various sources. Is it abundant in soils through burning of tyres, dumping of electronic waste, batteries and industrial waste. It is also a component of the some of bottles used for storing medicines. Further, plant raw materials are often not tested for heavy metal or remediation done before formulation into medicines. These could be the sources of the high lead content in both the plant extracts and the medicines.

All the detectable organochlorides were within acceptable limits. The major sources of organochlorophosphates are industrial chemicals and agricultural inputs[44]. Commercial production of medicinal plants where application of agrochemicals is required is still not being practised in Ghana. Further, the majority of the plants were not food items and therefore could not have been produced using agricultural inputs. Naturally grown herbs still constitute major a source of raw materials for herbal medicines in Ghana[45] and could be the reason for the low level of OCPs in medicines and plant extracts.

Finally, even though heavy metals and ions were found in some plant extracts and herbal medicines, their ionic states were not determined and therefore their toxicological effect can be evaluated. The WHO recommends that toxicological evaluation of heavy metals, ions and OCPs should not be determined by their concentration alone but by their ionic state[45]. The sulphate residues in OCPs exist as SO_3^{2-} and HSO_4^{2-} while the phosphate ions exist as HPO_4^{-}, PO_4^{2-} and $PO6^{2-}$. The oxidative state of calcium and magnesium varies between -1 and -2. Lead and cadmium exist in a transitional state in which where oxidative state varies between -2 to -5. The charged, ionic or oxidative state of these elements determines the number of lone pairs of electrons available and the type of chemical interaction that can be formed with phytochemicals, enzymes, co-factors and other biological molecules. The nature and extent of this chemical interaction further determine the physiological or pathological state of the affected system or organ.

CONCLUSION AND RECOMMENDATION

Crude plant extracts exhibit antimicrobial activity however their finished products can be contaminated through improper storage, transportation and shelving. Heavy metals, essential ions, chemical parametersand organochloride levels in most of the raw plants and herbal products are within the WHO recommended limits. Raw materials should be tested for heavy metals and OCPs and remediation systems should be part of good manufacturing practices (GMPs) of herbal medicines.

SIGNIFICANCE OF STATEMENT

Safe and efficacious herbal medicinal products are used to alleviate many human illnesses including that of the liver. The presence of chemical and microbial contaminants levels beyond accepted levels set by Standard Organization results in initiation of untargeted pathways which lead to illnesses. The study produced data on the level of several chemical and microbial markers which can be used to determine the safety of these herbal products for usage and their raw plant materials for formulation. The data is also saved as a base study for further research into tracer studies, microbial genomics and drug resistance as well as mechanisms of the contaminant's action.

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