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## Research Article

### MICROBIOLOGICAL QUALITY OF SOME HERBAL DRUGS IN THE SOUTHWEST REGIONAL CAPITAL OF CAMEROON

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#### ABSTRACT

With the ever-increasing use of herbal medicinal products, safety has become a major challenge as many contaminants are known as harmful for consumers. The present study aimed at identifying types of traditional herbal products sold in Buea, South-West region of Cameroon and the microbial contaminants they could contain. Fifty-three (53) samples including 24 in liquid and 29 in powder forms were collected and submitted to microbial analysis. Microbial screening and result interpretation were conducted according to the recommendations of the European pharmacopoeia. Specific microbes were further characterized with differential and selective techniques. Overall, 73.6% of the samples were not compliant. More details indicated that 81.1% of the herbal samples contained varied TAVC count (loads:  $1.28 \times 10^3$  -  $1.49 \times 10^6$  CFU/mL) and that 69.8% contained enterobacteria (loads range:  $4.16 \times 10^3$  -  $1.51 \times 10^6$  CFU/mL). Close to 66% were contaminated by *Staphylococci* ( $1.60 \times 10^3$  -  $3.31 \times 10^5$  CFU/mL) out of which 18.2% of the isolates were positive for DNase while all were negative for coagulase. Furthermore, 16.9% of the samples were contaminated by *Enterococci* with microbial loads ranging from  $1.60 \times 10^3$  to  $3.25 \times 10^5$  CFU/mL. Lastly the fungal counts ranged from  $1.60 \times 10^3$  to  $1.60 \times 10^6$  CFU/mL for 49.1% of the samples. Additional investigation on a few liquid samples indicated that the microbial loads increased with time and that the herbal products generally became unsuitable for consumption 48 hours after preparation. The present work highlighted that large numbers of herbal products sold in Buea were contaminated with potentially harmful microorganisms which made the products unsuitable for use in the management of human diseases. Application of good practices in harvesting, manufacturing, handling and storing herbal medicinal products was deemed important for quality products in order to serve the roles expected by the WHO.

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#### INTRODUCTION

Plants and plant derivatives have been used for therapeutic purposes in Africa for generations. Several investigations regard this use of plant products throughout the world as first line in the healthcare pyramids of several communities prior to conventional drugs. Herbal medicinal products can be referred

to as crude preparations of various kinds of medicinal plants; encompassing dried whole plants or any parts of it such as leaf, stem, root, flower, or seed which possess medicinal properties and used to control illnesses and disturbances (Stević *et al.*, 2012). Nowadays, herbal products are used globally in traditional medicine, especially in low-income communities

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(Adewunmi *et al.*, 2004) where according to the WHO, as many as 80 % of the population rely on traditional medicine based on medicinal plants for their primary medical care (Temu *et al.*, 2003).

Herbal medicinal products (HMPs) have taken advantage of the relatively high cost of conventional pharmaceutical forms, and the inaccessibility to formal conventional medical services to a majority of needy people and the trade of fake, substandard or counterfeit drugs used in conventional medicine. Also, adverse reactions of modern medications and development of drug resistance in infectious diseases agents have placed the HMPs as a ready alternative to conventional pharmaceutical forms in the management of diseases. With this increased usage, safety, efficacy and quality of these products which emerge as important concerns for health authorities and professionals (Gupta *et al.*, 2009; Adenike Okunlola *et al.*, 2007) deserve special regards.

Although often regarded as natural and safer, herbal remedies may actually channel sets of potential health threats like sources of adverse effects which may be related to such aspects as adulteration, replacement, contamination, misidentification, lack of standardization, incorrect preparation and/or dosage, unsuitable labelling and/or advertisement. As the plant materials used in herbal preparations are organic in their nature, they may provide nutritional resources for infectious disease aetiologies or other common microorganisms that eventually cause deterioration and variation of the product inherent properties, leading to low quality and/or little or no therapeutic efficacy. The quality of herbal products could, therefore, depend on such provisions as production environment, collection method, harvest, post-harvest handling, transport and storage methods. The commonly used herbal materials include chewing sticks, pastes, powders, mixtures and suspensions. Contaminants which could become serious threat to human are amongst others bacteria from such groups as *Salmonella*, *Escherichia coli*, *Staphylococcus aureus*, *Shigella* and other Gram-positive and Gram-negative strains of opportunistic bacteria, along-side with moulds and fungi (Nandna Khurana *et al.*, 2011; Ngari *et al.*, 2013).

Avoiding this microbial contamination can be achieved through implementation of basic best practice guidelines such as Good Manufacturing Practice (GMP) which makes sure that the products are continually manufactured with appropriate high-quality standards. GMP also aims at reducing the risks associated with contamination and mislabelling in order to ensure a certain standard of safety and efficacy. This requires rigorous documentation of production methods and the use of particular manufacturing and testing tools (Abbott, 2014).

In these regards, the WHO Good Manufacturing Practices are used by pharmaceutical manufacturers and regulators in over one hundred countries throughout the world, primarily in countries with overall limited resources. The principal disadvantage to demanding GMPs is that they may have prohibitive costs and may put many small to medium-sized manufacturers out of business (Abbott, 2014).

In Buea (South-West Cameroon), herbal practitioners have capitalized on the poor health conditions of the masses and the high cost of synthetic orthodox medicine by organizing herbal

trade fare indiscriminately. The probability that a patient on herbal remedies contracts more deadly diseases cannot be totally overruled, however, considering the unhygienic and crude method of production and storage. In this light, this research will focus on evaluating bacteria and fungi contaminants in some herbal medicinal products sold in Buea, the regional capital city of the South-West region of Cameroon. Findings thereof will serve as database useful in guiding policies for safer traditional drug production.

## MATERIAL AND METHODS

### *Setting and populations*

Capital of the South-West region of Cameroon, Buea is located on the eastern slopes of Mont Cameroon. The most recent census estimates a population of 90,088, primarily Bakweri people. However, due to its position as a university town and regional capital, significant numbers of other ethnic groups are also present ( UCCC. Map of Buea municipality, 2014).

### *Type of study and sample collection*

This investigation was a cross-sectional descriptive study conducted over a nine months' period, from November 2016 through July 2017. During this period of time, many samples of herbal products (liquid and powder) were purchased at random from herbal traders in some bus agencies and in market places (mainly Central and Muea markets); then kept in clean plastic bags pending analysis. Interviews were also held with these traders. When the respondents were uncomfortable with the interview questions, discussions and informal interviews techniques were used. Codes were thereafter, provided to collected specimens in addition to the following pieces of information: drug form, plant and parts used, indication and dosage. Additional details from research participants included gender, age, general education, formal training in the field of traditional medicine production, sample characteristics and aspects of quality control.

### *Isolation and enumeration of bacteria and fungi*

A pilot test was conducted for method validation prior to sample analysis. This test was performed in order to identify the concentration at which the colony counting procedure would be more efficient. In this light, several dilutions were made from the herbal raw material; ranging from the mother solution (1/2) to the fifth dilution (1/32). Each of these was plated on appropriate agar in Petri dishes and allowed to incubation at 37°C for 24, 48 and 72 hours according to the microbial types targeted. Upon completion of incubation, colony counting followed. This preliminary /pilot test revealed that the appropriate dilution for the experiment was the fifth (1/32). Accordingly, 20µl of the fifth dilution was transferred onto appropriate agar in Petri dishes (in triplicates). These preparations were incubated as indicated above in the pilot test. Agar media used for enumeration included: Plate count agar for total mesophilic aerobes, McConkey for faecal coliforms, Mannitol salt for *Staphylococcus*, Bile Esculine Azide for fecal *Enterococcus* and Sabouraud dextrose agar for fungi. For *Staphylococcus*, coagulase and DNase tests were further performed. All identifications were conducted according to standard morphological and biochemical protocols.

### Microbial loads in the course of time

To assess the microbiological loads in the course of time, three freshly prepared herbal liquid samples were selected at random, processed the same ways as described above, then incubated for one, three, five and ten days. For this experiment, each of the samples was fractioned into four parts each of which would serve one of these four days.

## RESULTS

### Traders' characteristics

The total number of traders in the town was estimated at 37 from both sexes during the present investigation. Their characteristics were summarized as displayed in figure 1.

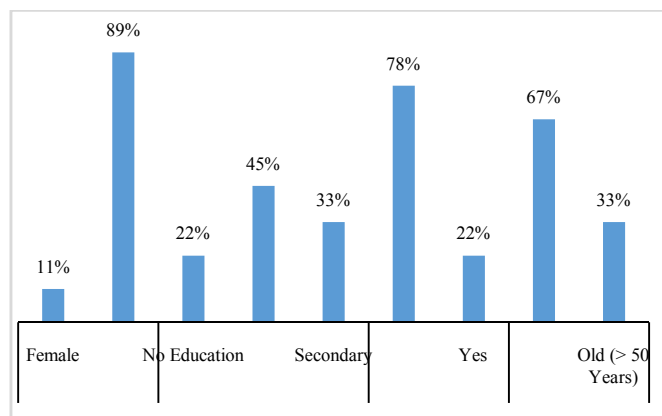


Figure 1 Traders' characteristics

Trading was overwhelmed by male, representing 90% of the total. About 66.6% (2/3) of participants belong to the 21-50 age group while 33.3% (1/3) were above 50 years. With regards to education, 78% underwent at least primary education. Further, close to 78% did not receive any formal training in traditional healing practices. For some, it was a gift from birth while others said that dealing in traditional medicine was inherited from their fore-fathers.

### Characteristics of the samples

A total of 53 samples were collected and used in this research. More detailed related pieces of information were summarized and presented in table I.

Table I Characteristics of sample

Sample code	Drug form	Plant/product vernacular name	Part used	Therapeutic claim	Dosage	Route of administration	Package
HL1	Liquid	Simba	Bark	venereal diseases	1 cup daily	Oral	Transparent plastic bottle
HL2	Liquid	Cinchona	Bark	Malaria	1 cup × 2 daily	Oral	Transparent plastic bottle
HL3	Liquid	yellow canda stick + black stone	Bark	malaria and typhoid fever	1 cup × 2 × 3 days in adults; 1/2 cup × 2 × 3 days in children	Oral	Transparent plastic bottle
HL4	Liquid	baobab + pear leaves	Bark	gastric pain	1 cup × 3 daily	Oral	Transparent plastic bottle
HL5	Liquid	Alme	leaves	Cough	1 tablespoon × 2 daily	Oral	Transparent plastic bottle
HL6	Liquid	Simba	Bark	Toothache	apply 3 times daily on tooth	Oral	Glass bottle
HL7	Liquid	Simba	Bark	Toothache	apply 3 times daily on tooth	Oral	Glass bottle
HL8	Liquid	Simba	Bark	dental carries	apply 3 times daily on tooth	Oral	Glass bottle
HL9	Liquid	Simba	Bark	dental carries	apply 3 times daily on tooth	Oral	Glass bottle
HL10	Liquid	Simba	Bark	venereal diseases	1 cup daily	Oral	Transparent plastic bottle
HL11	Liquid	Simba	Bark	Pile	1 cup × 2 daily	Oral	Transparent plastic bottle
HL12	Liquid	Simba	Bark	menstrual pains	1 cup × 3 daily	Oral	Transparent plastic bottle
HL13	Liquid	Simba	Bark	sexual weakness	1 cup daily	Oral	Transparent plastic bottle
HL14	Liquid	Simba	Bark	waist pain	1 cup × 2 daily	Oral	Transparent plastic bottle
HL15	Liquid	N/A	leaves	conception problems	enema	Anal	Transparent plastic bottle
HL16	Liquid	N/A	Bark	Convulsion	1 cup × 2 daily	Oral	Transparent plastic bottle
HL17	Liquid	N/A	Bark	stomach ache	1 cup × 3 daily	Oral	Transparent plastic bottle
HL18	Liquid	N/A	leaves	Poison	lick 3 times daily	Oral	Transparent plastic bottle
HL19	Liquid	N/A	Bark	Worms	1 tablespoon daily × 3 days before eating	Oral	Transparent plastic bottle
HL20	Liquid	Kahi	Fruits	Hernia	1 teaspoon in pap daily	Oral	Transparent plastic bottle
HL21	Liquid	1000 diseases	Bark	Typhoid	1 glass every evening × 10days	Oral	Transparent plastic bottle
HL22	Liquid	N/A	Bark	Stomach ache	1 glass × 2 × 3 days	Oral	Transparent plastic bottle
HL23	Liquid	N/A	leaves	Venereal diseases	1 glass × 2 × 5 days	Oral	Transparent plastic bottle
HL24	Liquid	N/A	Bark	Typhoid	1 glass × 2 × 5 days	Oral	Transparent plastic bottle
HP1	Powder	Ginseng	Bark	gastric pain	half litre × 3 daily	Oral	Small transparent plastic bags
HP2	Powder	Mahogany	Bark	menstrual pains	1 cup × 2 daily × 3days	Oral	Small transparent plastic bags
HP3	Powder	Moringa	leaves	Diabetes	1 glass daily × 1 week	Oral	Small transparent plastic bags
HP4	Powder	ohsio + red limestone	Bark	venereal diseases	1 tablespoon in pap × 2 × 11days	Oral	Small transparent plastic bags
HP5	Powder	ararabi (99 powder)	Bark	Pile	1 cup × 3 daily × 1 week	Oral	Small transparent plastic bags
HP6	Powder	Kahi	Bark	ungi infection	1 teaspoon daily in children; 1 tablespoon daily in adults	Oral	Small transparent plastic bags
HP7	Powder	Simba	Bark	bloating stomach	teaspoon × 3 × 3 days	Oral	Small transparent plastic bags
HP8	Powder	Kojoli	Bark	Pile	1 teaspoon × 2 × 81 days	Oral	Small transparent plastic bags
HP9	Powder	Jimtal	Bark	stomach ache	1 teaspoon × 3 daily	Oral	Small transparent plastic bags
HP10	Powder	Dislego	Bark	frontal headache	niff once daily for 3days	Oral	Small transparent plastic bags
HP11	Powder	Kelele	leaves	Filarial	1 tablespoon in pap × 2 × 7 days	Oral	Cardboard paper
HP12	Powder	Dalehi	Bark	stomach ache	1 cup × 2 × 3 days	Oral	Cardboard paper
HP13	Powder	limee + 1000 diseases	Fruits	Worms	chew 1 each at once	Oral	Cardboard paper
HP14	Powder	Pasokori	Bark	sexual weakness	1 teaspoon × 3 daily	Oral	Cardboard paper
HP15	Powder	Flasko	Bark	stomach ache	1 tablespoon in pap 2 hours before meal daily	Oral	Small transparent plastic bags
HP16	Powder	karej + red limestone	Bark	internal pile	1 cup × 2 × 7 days	Oral	Small transparent plastic bags
HP17	Powder	nari + honey	Bark	Pile	1 tablespoon × 3 daily	Oral	Cardboard paper
HP18	Powder	Potki	Bark	snake bite	1 tablespoon daily	Oral	Cardboard paper
HP19	Powder	jintari + limestone	Bark	Toothache	gaggle once daily	Oral	Small transparent plastic bags
HP20	Powder	Neem	leaves	Jaundice	1 cup × 2 × 7 days	Oral	Cardboard paper
HP21	Powder	Nivaquine	Bark	Worms	1 cup × 2 daily × 3 days	Oral	Cardboard paper
HP22	Powder	N/A	Bark	Rheumatism	1 glass × 2 daily	Oral	Small transparent plastic bags
HP23	Powder	N/A	leaves	Dysentery	1 tablespoon × 2 daily	Oral	Small transparent plastic bags
HP24	Powder	N/A	Bark	one problems	1 teaspoon daily in children; 1 tablespoon daily in adults	Oral	Small transparent plastic bags
HP25	Powder	N/A	Bark	Filarial	1 glass × 2 daily	Oral	Small transparent plastic bags
HP26	Powder	N/A	Bark	body weakness	pump very early in the morning	Anal	Small transparent plastic bags

Sample Code	Form	Material	Part	Disease	Dose	Route	Packaging
HP27	Powder	N/A	Bark	Snoring	1 teaspoon in a glass of water	Oral	Small transparent plastic bags
HP28	Powder	N/A	Bark	Typhoid	1 glass × 2 daily	Oral	Small transparent plastic bags
HP29	Powder	N/A	Bark	venereal diseases	1 glass daily × 1 week	Oral	transparent plastic bags

N/A: Not Applicable

The 53 specimens consisted of 24 liquids and 29 powders. Mostly, they were used for management of conditions like venereal diseases, pile, gastro-intestinal disorders, malaria, and typhoid fever. Still others were used against jaundice, bone disorders, snake bite, dysentery, diabetes, filariasis, dental problems, rheumatism, sexual weakness, infertility, hernia, waist pain and worms. As raw material, tree barks were most common (81%), followed by leaves (15%) and fruits (4%). They could be prepared separately or as combinations of several items. In decreasing order, common administration ways included oral (94%), anal (4%) and nasal (2%).

### Trading site and microbial load assessment

Overall, 71% of traders harvested raw materials in the wild while the remaining (29%) used products that were grown in formal farms. Most of the businesses were located in crowded areas or in slums (42.8% each) and nearby roadside disposal pits or in bus stations (14.2%). To package their products, traders generally used plastic bottles for liquid form while the powders, most often in large buckets, were retailed and sealed in small transparent plastic bags or tied in cardboard papers.

Specimen analysis yielded ranges of data most of which did not meet the standards as recommended by the European Pharmacopia. More detailed pieces of information were reorganized and summarized as shown in table II.

**Table II** Microbial load of herbal medicinal products

Sample code	TAVC (CFU/ml)	Enterobacteria (CFU/ml)	Staphylococci (CFU/ml)	Enterococci (CFU/ml)	Fungi (CFU/ml)	Compliance
HL1	1.2×10 <sup>5</sup>	7.87×10 <sup>5</sup>	3.68×10 <sup>3</sup>	0	7.57×10 <sup>4</sup>	Not compliant
HL2	6.4×10 <sup>3</sup>	1.06×10 <sup>5</sup>	3.68×10 <sup>3</sup>	0	3.2×10 <sup>3</sup>	Not compliant
HL3	4.80×10 <sup>4</sup>	3.23×10 <sup>5</sup>	0	0	0	Not compliant
HL4	4.75×10 <sup>5</sup>	0	1.01×10 <sup>4</sup>	0	4.42×10 <sup>5</sup>	Not compliant
HL5	1.04×10 <sup>5</sup>	0	3.20×10 <sup>3</sup>	0	7.52×10 <sup>4</sup>	Not compliant
HL6	0	0	0	0	0	Compliant
HL7	0	0	0	0	0	Compliant
HL8	0	0	0	0	0	Compliant
HL9	0	0	0	0	0	Compliant
HL10	8.15×10 <sup>5</sup>	7.55×10 <sup>5</sup>	1.28×10 <sup>4</sup>	2.24×10 <sup>4</sup>	3.41×10 <sup>4</sup>	Not compliant
HL11	5.32×10 <sup>5</sup>	4.85×10 <sup>5</sup>	3.20×10 <sup>3</sup>	0	2.29×10 <sup>4</sup>	Not compliant
HL12	8.06×10 <sup>5</sup>	4.86×10 <sup>5</sup>	9.60×10 <sup>3</sup>	3.25×10 <sup>5</sup>	6.56×10 <sup>4</sup>	Not compliant
HL13	1.27×10 <sup>5</sup>	3.20×10 <sup>4</sup>	5.28×10 <sup>3</sup>	0	0	Not compliant
HL14	4.92×10 <sup>5</sup>	4.03×10 <sup>5</sup>	3.2×10 <sup>3</sup>	0	1.01×10 <sup>5</sup>	Not compliant
HL15	6.08×10 <sup>5</sup>	4.78×10 <sup>5</sup>	7.36×10 <sup>3</sup>	5.28×10 <sup>3</sup>	3.41×10 <sup>4</sup>	Not compliant
HL16	7.02×10 <sup>5</sup>	7.21×10 <sup>5</sup>	0	0	3.25×10 <sup>4</sup>	Not compliant
HL17	2.49×10 <sup>4</sup>	2.24×10 <sup>4</sup>	3.2×10 <sup>3</sup>	0	0	Not compliant
HL18	1.02×10 <sup>6</sup>	1.51×10 <sup>6</sup>	0	2.34×10 <sup>4</sup>	1.69×10 <sup>4</sup>	Not compliant
HL19	1.62×10 <sup>5</sup>	1.97×10 <sup>5</sup>	0	0	0	Not compliant
HL20	0	0	1.52×10 <sup>5</sup>	0	0	Compliant
HL21	0	0	1.55×10 <sup>5</sup>	0	0	Compliant
HL22	1.28×10 <sup>3</sup>	0	0	0	0	Compliant
HL23	1.28×10 <sup>3</sup>	0	2.56×10 <sup>3</sup>	0	0	Compliant
HL24	0	8.60×10 <sup>5</sup>	0	0	0	Not compliant
HP1	2.46×10 <sup>5</sup>	TNTC	0	0	0	Not compliant
HP2	0	0	0	0	0	Compliant
HP3	4.21×10 <sup>4</sup>	TNTC	0	0	0	Not compliant
HP4	2.29×10 <sup>4</sup>	TNTC	0	0	0	Not compliant
HP5	2.13×10 <sup>4</sup>	TNTC	3.68×10 <sup>4</sup>	1.6×10 <sup>3</sup>	4.16×10 <sup>4</sup>	Not compliant
HP6	TNTC	TNTC	1.33×10 <sup>4</sup>	0	2.77×10 <sup>4</sup>	Not compliant
HP7	TNTC	TNTC	4.8×10 <sup>4</sup>	0	9.6×10 <sup>3</sup>	Not compliant
HP8	4.58×10 <sup>4</sup>	TNTC	3.12×10 <sup>4</sup>	0	1.44×10 <sup>4</sup>	Not compliant
HP9	3.84×10 <sup>4</sup>	TNTC	1.49×10 <sup>4</sup>	0	0	Not compliant
HP10	0	0	3.68×10 <sup>3</sup>	0	0	Compliant
HP11	2.88×10 <sup>4</sup>	1.3×10 <sup>6</sup>	1.60×10 <sup>3</sup>	0	1.33×10 <sup>4</sup>	Not compliant

HP12	TNTC	1.8×10 <sup>5</sup>	8.32×10 <sup>4</sup>	0	1.07×10 <sup>5</sup>	Not compliant
HP13	0	0	0	0	0	Compliant
HP14	8.21×10 <sup>4</sup>	7.52×10 <sup>4</sup>	0	0	0	Not compliant
HP15	7.52×10 <sup>5</sup>	9.31×10 <sup>5</sup>	8.48×10 <sup>4</sup>	1.74×10 <sup>5</sup>	1.6×10 <sup>4</sup>	Not compliant
HP16	1.49×10 <sup>6</sup>	1.53×10 <sup>6</sup>	0	0	5.28×10 <sup>3</sup>	Not compliant
HP17	1.65×10 <sup>4</sup>	4.16×10 <sup>3</sup>	2.08×10 <sup>3</sup>	0	1.6×10 <sup>6</sup>	Not compliant
HP18	1.09×10 <sup>6</sup>	5.01×10 <sup>4</sup>	2.0×10 <sup>5</sup>	4.32×10 <sup>4</sup>	3.2×10 <sup>3</sup>	Not compliant
HP19	4.14×10 <sup>5</sup>	2.15×10 <sup>5</sup>	3.25×10 <sup>4</sup>	0	0	Not compliant
HP20	1.83×10 <sup>5</sup>	1.15×10 <sup>5</sup>	0	0	8.80×10 <sup>3</sup>	Not compliant
HP21	2.38×10 <sup>5</sup>	1.10×10 <sup>5</sup>	3.31×10 <sup>5</sup>	0	6.88×10 <sup>3</sup>	Not compliant
HP22	9.69×10 <sup>4</sup>	9.60×10 <sup>3</sup>	2.40×10 <sup>5</sup>	0	0	Not compliant
HP23	1.60×10 <sup>3</sup>	0	2.21×10 <sup>5</sup>	4.0×10 <sup>3</sup>	0	Compliant
HP24	3.94×10 <sup>5</sup>	1.34×10 <sup>5</sup>	2.85×10 <sup>5</sup>	0	5.28×10 <sup>3</sup>	Not compliant
HP25	6.24×10 <sup>4</sup>	1.49×10 <sup>4</sup>	2.58×10 <sup>4</sup>	0	1.6×10 <sup>3</sup>	Not compliant
HP26	6.08×10 <sup>4</sup>	4.78×10 <sup>5</sup>	2.42×10 <sup>5</sup>	8.96×10 <sup>3</sup>	0	Not compliant
HP27	1.28×10 <sup>4</sup>	0	1.52×10 <sup>5</sup>	0	0	Compliant
HP28	1.09×10 <sup>5</sup>	7.62×10 <sup>4</sup>	1.65×10 <sup>4</sup>	0	1.6×10 <sup>3</sup>	Not compliant
HP29	1.6×10 <sup>3</sup>	0	2.42×10 <sup>5</sup>	0	0	Compliant

The overall picture revealed that most specimens (73.6%) did not meet the expected specifications, then qualified as unacceptable for use. Further, they were contaminated at varying amplitude with bacteria and fungi. Amongst these, six were totally free from contaminants. Further, eight (33.3%) of the liquid forms and six (20.7%) the powders were compliant to the stated limits.

The total aerobic viable count (TAVC) ranged from 1.28×10<sup>3</sup> through 1.49×10<sup>6</sup> CFU/mL; with 81.1% contaminations involving bacteria. More specifically, bacteria from the *Enterobacteriaceae* family were involved in 69.8% with loads found between 4.16×10<sup>3</sup> and 1.51×10<sup>6</sup>CFU/mL for faecal coliforms. The *Staphylococcus* counts were recorded between 1.60×10<sup>3</sup> and 3.31×10<sup>5</sup> CFU/mL, done from 66% of the specimens. DNase and coagulase test results further revealed *Staphylococcus aureus* in 18.2% of the studied products. For more details, *Enterococcus* spp. was isolated from 17% of the samples with loads recorded between 1.60 × 10<sup>3</sup> and 3.25 × 10<sup>5</sup> CFU/mL, and fungi from 49.1% with lowest and highest count values found between 1.60×10<sup>3</sup> and 1.60 × 10<sup>6</sup> CFU/mL.

### Microbial loads of herbal liquids in the course of time

Of the sealed samples studied, all were found to be contaminated with bacteria other than those from genus *Enterococcus*. In the course of time for ten days, the microbial bio-burden was monitored as summarized in Table III.

**Table III** Bacterial loads in the course of time

TAVC	Period of time			
	Day 1	Day 3	Day 5	Day 10
HL22	1.28×10 <sup>3</sup>	4.48×10 <sup>3</sup>	1.45×10 <sup>3</sup>	1.49×10 <sup>4</sup>
HL23	1.28×10 <sup>3</sup>	1.71×10 <sup>3</sup>	1.01×10 <sup>4</sup>	2.56×10 <sup>4</sup>
HL24	0	2.20×10 <sup>6</sup>	2.23×10 <sup>6</sup>	3.26×10 <sup>6</sup>
<i>Enterobacteriaceae</i> count	Day 1	Day 3	Day 5	Day 10
HL22	0	5.04×10 <sup>3</sup>	2.89×10 <sup>3</sup>	9.60×10 <sup>3</sup>
HL23	0	0	0	0
HL24	0	9.52×10 <sup>5</sup>	9.22×10 <sup>5</sup>	1.13×10 <sup>6</sup>
<i>Staphylococci</i> count	Day 1	Day 3	Day 5	Day 10
HL22	1.3×10 <sup>3</sup>	0	0	0
HL23	2.56×10 <sup>3</sup>	2.35×10 <sup>3</sup>	1.01×10 <sup>4</sup>	2.2×10 <sup>3</sup>
HL24	0	7.17×10 <sup>4</sup>	6.14×10 <sup>4</sup>	1.02×10 <sup>5</sup>

The general picture highlighted that one out of the three (HL23) would be suitable for consumption if coliforms were the only bacteria indicator targeted upon completion of incubation. But, the TAVC developed above the acceptable limits, making it unsuitable like the others. Otherwise, the bacterial loads increased with incubation time.

## DISCUSSION

Gender division of labour is usually common amongst human communities. The findings from the current survey indicated that the majority of participants were male (89%). This contradicted the finding reported in South Africa where 74% of plant harvesters, street vendors and traditional healers were female (Dold *et al.*, 2002). The predominance of male in Buea could be due to the difficulties associated with the sourcing and transportation of herbal materials. Moreover, ethnobotanical researchers have lamented the little or lack of interest amongst young members of communities when it came to assimilate and pass on the medicinal plant legacy (Begossi *et al.*, 2002). This lack of interest seemed to deviate according to the findings in Buea. In fact, it was shown that most of the respondents belonged to the 21-50 years age-group. This tendency might be explained differently rather than ruling out the need for them to learn and pass the legacy. In fact, unemployment rate that is high in the locality could guide several people in traditional medicine, simply as means for making a living. Admitting that involving the youth in the herbal trade could be a sure way of preserving the traditional knowledge and cultural set up of the society, this would necessarily not insure efficacy of traditional medicine practices because they are not trained for the business. In addition, although most participants have undergone at least primary education, traditional knowledge is passed orally, then theoretically diluted from generation to generation.

Different plant parts were harvested and used for the preparation of herbal drugs. Recording that about 82% of times tree barks are used as raw material represents a real threat for the activity sustainability because it threatens in turn the plant survival (Zschocke *et al.*, 2000). This highlighted the need for sensitization of users on the parts to be used and the ones to be protected.

The ethno medicinal uses of several plants recorded in this survey agreed with their pharmacological and traditional uses elsewhere. Polyherb therapy is common in herbal practices but very few studies have so far reported the use of different plant combinations in drug preparation. The use of polyherbal concoctions needs investigation with respect to bioassay and phytochemistry. Synergetic effect of these combinations should also be appropriately addressed for efficient caretaking with traditional drugs.

Several studies also raised the issue of the poor hygienic conditions under which traditional drugs were prepared and dispensed (Oyetayo *et al.*, 2008; Stević *et al.*, 2012), consistent with the finding from the present investigation; since the herbal products were typically sold either in crowded areas, slums or near disposal pits. The high unacceptability rates recorded is a reliable clue that could be explained by these working conditions and could predict higher potential of contamination for consumers, then a real source of infectious diseases as anticipated by former authors (Adounkpe *et al.*, 2017). In fact, the mode of transport and storage of herbal material was likely to compromise the quality. Most of these products were stored in recycled containers, heat labile polythene bags, then likely to alter the active chemical components of the herbal products. In line with above discussion and in connection with the route of administration,

gastro-intestinal disorders are most likely, especially amongst those with reduced immunity like children and elderly. These conclusions are supported by the rates of detection of indicators of faecal contaminations like *Enterococcus* and coliforms, and the overall unacceptability rate (74%); consistent with those reported in South-Western Nigeria (Oluwatoyin *et al.*, 2016)

The results from the present study reveal inadequate labelling amongst other procedure's weaknesses, implying the low standards of products in the market and failure of the product to meet the WHO standards. This agreed with earlier studies carried out on documentation of herbal products in other parts of the world where labelling was not appropriately conducted (Bandaranayake *et al.*, 2006) and further highlighted the need for the training of people who work in the whole network of traditional medicine; in agreement with Walther *et al.* (Walther *et al.*, 2016). Otherwise, the presence of microbial contaminants in herbal medicine can be associated with hosts of factors that can act with specific amplitude at different levels along the chain.

Together with the group of coliforms and enterococci, detection of bacteria belonging to the *Enterobacteriaceae* family can be assumed as indicator of poor hygiene and sanitation that either prevail in the working environment, as post-preparation pollution or both (Araújo *et al.*, 2012). In all cases, thinking about product sterilisation and post-production quality with limited resources, but on regular basis would emerge as a suitable alternative for their safer utilization. This conclusion further stresses the necessity for training as mentioned above.

The presence of the fungi contaminants was another key element to target in these drugs. Beyond the above development on bacteria in fact, some fungi species can synthesize harmful products like aflatoxins. According to the WHO (World Health Organization, 2000), these toxins which frequently develop during storage could have adverse effects on consumers even at minute concentration.

When the study was continued in order to determine the microbial load of some samples in the course of time, an increase in the TAVC, staphylococcal and enterobacteria counts was recorded. Similar reasons as above could have explained their presence in the preparation. The liquids are sealed, then virtually free from new contaminations, unlike powders that are frequently exposed to contaminants during retail. Otherwise, product in powder form would more frequently receive new contaminants during the retail procedures; theoretically further empowering their harmful potential higher than that of those in liquid. The population increase that occurred with time virtually varied in their types with respect to the chemical composition of the broth which in turn depends on microbial metabolism. This variation might favour positive selection of specific strains and/or species with harmful characteristics including bacteria with low and intermediate virulence likely to worsen the health status of drug users, especially amongst communities with reduced immunity. Definitely, proper means of sterilization should be developed to prevent infections related to drug's use and to make of these herbal derivatives proper alternatives to conventional drugs in resource-limited populations as expected and encouraged by the WHO.

## CONCLUSION

Findings from this investigation on the quality of traditional drugs sold in Buea Cameroon revealed several variants of products, largely provided by untrained male that otherwise, for most, have the lowest education background that could help acceptable services. The rates of compliance were very low and post-production contamination very likely. Microbial density in ready-to-use products also increased with time. All these findings highlighted the absolute necessity for the training of all practitioners involved in the whole network of drug production and their distribution for safer administration; pre-requisite that would improve their quality in order to meet the goals expected by the World Health Organization.

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