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MICROBIAL BURDEN OF SOME HERBAL ANTIMALARIALS MARKETED AT ELELE, RIVERS STATE

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Abstract

Herbal antimalarials still remain an alternative to our traditional communities who can not afford orthodox antimalarials. This study was aimed at investigating the microbial quality of six herbal antimalarials using standard microbiological methods. Of the six preparations analyzed, "schnapps", palm wine and water were the media of preparation; the water base preparations recorded higher microbial load. The mean microbial load was $159.5x10^5$ cfu/ml and $217.4x10^2$cfu/ml in water and alcohol base preparations respectively. The microbial profile of the preparations showed that the schnapps base preparations were predominantly contaminated with *Bacillus* sp (Aerobic spore bearers) and *Mucor* spp. The palm wine preparation harboured *Bacillus* sp, yeasts and *Mucor* spp while the water base preparations had several isolates such as *Staphylococcus epidermidis, Pseudomonas aeruginosa, Escherichia coli* 0157H7, *Proteus mirabilis, Enterococcus feacalis, Serratia marcensces, Staph. aureus, Bacillus* spp and *Mucor* spp. Conclusively, this study underlines the public health importance of these preparations given the high burden of such human pathogen as *Ecoli* O157H7, *Ps aeruginosa, Stahp aureus, etc.* in the preparations.

Key words: antimalarials, herbal, microbial, Elele

Introduction

Malaria is a major dreaded disease of humans causing hundreds of millions of morbidity with increasing rate of mortality. The number of people dying from malaria is now higher than it was 30 years ago (Joy et al., 2003). Traditional medicines have been used to treat malaria for thousands of years and are the source of the two main groups (artemisinin and quinine derivatives) of modern antimalarial drugs. With the problems of increasing levels of drug resistance and the inability of a large number of patients being able to afford and access effective antimalarial drugs, traditional medicines could be an important and sustainable source of treatment (Wilcox and Bodeker, 2004). Resistance has spread so fast that it now represents a serious threat to global public health (World Health Organization, 2003). Scientists now agree that the most effective treatment against malaria is a combination of drugs using artemisinin derivatives which are highly potent extracts of the Chinese plant *Artemisia annua*. Artemisinin based combination therapy (ACT) is the quickest and most reliable way of clearing malaria infection and has been shown to protect each individual drug from resistance (World Health Organization, 2002). Herbal therapy has been used in the treatments of many ailments, including malaria. Various plant parts singly or a combination of

a number of plants have been employed in the treatment of malaria; some plants which have been reported include *Azadirachta indica, Morinda lucida, Alstonia boonei, Enantia chlorantha, Crossopteryx febrifuga, Tithonia diversifolia* etc. (Tella, 1977; Ade 1983; Obih and Makinde, 1985; Makinde et al., 1987; Elufioye and Agbedahunsi, 2004). In this work, we investigated the microbial quality of some antimalarials marketed at Elele, Rivers State.

Materials and Methods

Study design

Six preparations of antimalarials herbal preparations were obtained from different marketers at Elele, Rivers state, Nigeria. The products were collected in sterile containers to avoid contamination from external source for microbial analysis. The composition of the preparations was as given below;

Preparation 1: chopped Roots of *Azadirachta indica* A. Juss family Meliaceae soaked in Schnapps.

Preparation 2: Sliced leaves *Cymbopogon citratus* (DC.) Stapf., family Graminae soaked in alcohol Schnapps.

Preparation 3: Sliced stem barks of *Azadirachta indica* A. Juss family Meliaceae soaked in palm wine

Preparation 4: Sliced stem barks of *Azadirachta indica* A. Juss family Meliaceae soaked in water

Preparation 5: Leaves of *Ocimum gratissimum* Linn. (Labiatae), *Psidium guajava* Linn. (Myrtaceae), *Cymbopogon citratus* (DC.) Stapf., Graminae), *Azadirachta indica* A. Juss (Meliaceae), *Mangifera indica* Linn. (Anarcadiaceae) and unripe leaf of *Carica papaya* Linn. (Caricaceae) all cut into pieces, boiled in water, allowed to cool and sieved into a clean container.

Preparation 6: Leaves of *Cymbopogon citratus* (DC.) Stapf., Graminae), leaves of *Azadirachta indica* A. Juss (Meliaceae), leaves of *Mangifera indica* Linn. (Anarcadiaceae) and unripe bark of the fruit of *Carica papaya* Linn. (Caricaceae) and leaves of *Vernonia amygdalina* Del. (Compositae) boiled in a pot till it changed colour, allowed to cool and then sieved into a clean container.

Sample Processing

Microbial Isolation and Identification

The isolation of microbes was done culturally using Eosin Methylene Blue agar, Potato Dextrose agar, Blood agar and Salmonella-Shigella agar plates and microbial identification was carried out using standard microbiological techniques, this includes Gram staining of the isolates, biochemical testing i.e. coagulase, catalase, citrate utilization, indole production, oxidase test, sugar fermentation tests, serology for *E. coli* O157H7 using and lacto phenol cotton blue preparations.

Total Viable Count

A ten fold dilution was carried out on the six samples. The dilution was made as follows, 9 ml of sterile water was transferred in each of 6 rows of 10 sterile test tubes, 1 ml of each of the samples was transferred in the first test tubes on each row containing sterile distilled water and was mixed properly. One (1) ml from the first tube on each row was transferred to the 2^{nd} test tube and was mixed by shaking to obtain a 1/10 dilution. This exercise was carried though the tenth tube where 1 ml of the mixture was eventually discarded away to obtain dilutions of $1/10^2$, $1/10^3$, $1/10^4$…$1/10^{10}$. One (1) ml from each test tube was transferred into a sterile petri dish and molten nutrient agar was added, mixed well and incubated aerobically at 37 ^oC. Count was made on the plate showing evenly distributed and discrete colonies.

Results

Of the six preparations analyzed, schnapps, palm wine and water were the media of preparation, the 2 alcohol base preparations had count of $0.2x10^2$ cfu/ml and $2.0x10^2$ cfu/ml, the only palm wine preparation had a count of 6.5x104 cfu/ml while the 3 water based preparations had count of $5.6x10^5$ cfu/ml, $3.1x10^5$ cfu/ml and $4.7x10^7$ cfu/ml each. The microbial profile showed that the schnapps base preparations were mostly contaminated with *Bacillus* spp (Aerobic spore bearers) and *Mucor* spp, the palm wine preparation harboured *Baciillus* spp, yeasts and *Mucor* spp while the water based preparations had several microbial isolates such as *Staph. epidermidis, Pseudomonas aeruginosa, E. coli 0157, Proteus mirabilis, Enterococcus feacalis, Serratia marcensces, Staph. aureus, Bacillus* spp and *Mucor* spp (Table 1).

Table 1: Total viable count and distribution of microbial isolates by sample.

Samples	1	2	3	4	5	6
Microbial Load(cfu/ml)	$0.2x10^2$	$2.0x10^2$	$6.5x10^4$	$5.6x10^5$	$3.1x10^5$	$4.7x10^7$
Isolates	*Bacillus* spp (ASB)	*Bacillus* spp (ASB)	*Bacillus* spp (ASB)	*Staph. epidermidis*	*Pseudomonas aeruginosa*	*Pseudomonas aeruginosa*
	Mucor spp	*Mucor* spp	*Mucor* spp	*Bacillus* spp	*Proteus mirabilis*	*Staph. Aureus*
			Yeast cells	*Pseudomonas aeruginosa*	*Bacillus* spp (ASB)	*E. coli*
				E. coli 0157	*Serratia marcensces*	*Staph epedermidis*
				Proteus mirabilis	*Mucor* spp	*Bacillus* spp
				Mucor spp		*Mucor* spp
				Aspergillus niger		*Enterococcus feacalis*

Discussion

Herbal antimalarials are herbal drugs used in the form of concoction and decoction from roots, stems and flowers from various plants which is believed to cure malaria. They have some risks associated with their use. Findings from this study showed that the antimalarials studied were contaminated with large number of pathogenic organisms of public health importance. *Bacillus* spp and *S. aureus* had the highest frequency of occurrence. This was similar to previous findings reported by World Health Organization (WHO, 1998). The occurrence of isolates such as *E. coli* O157H7 and *Entercoccus feacalis* is of serious medical importance as its presence could indicate a potential feacal contamination. Both water and alcohol base preparations harboured organisms; however, "Schnapps" base preparations recorded lower microbial load than the water base preparations which recorded load above acceptable levels. Obviously, Schnapps contains some level of alcohol at concentration which could have some inhibitory activities on some microorganisms. Water base preparations contamination could result from the water used for the preparation, the traditional healers could have been the source of contamination as the hygienic requirement may not have been met. Soil could also be a source of contamination as medicinal plant materials normally carry a large number of microbes originating from the soil. The usage of alcohol containing medium in the preparations of these products remains worrisome in our communities where such are used also as prophylaxis by both children and adults bearing in mind the effect of alcohol on liver functions.

Clinical observations on traditional remedies are feasible and useful. Some herbal remedies may be safe and effective for the treatment of malaria. Nevertheless, better evidence from randomised clinical

trials is needed before herbal remedies can be recommended on a large scale. Preventing children's deaths is the key objective of any malaria control programme. Once a remedy has been shown to be safe and effective for uncomplicated malaria in adults, studies on mortality in children would be the necessary next step. It has already been shown that mortality can be reduced in the under 5s by training mothers to recognise malaria and to give early treatment (Kidane and Morrow, 2000).

In conclusion, some herbal antimalarials may be helpful in the management of malaria fever; serious attention should be given to this alternative branch in medicine so that its limitations do not overshadow its benefits.

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