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ANALYSIS OF HEAVY METALS AND MICROBIAL LOAD IN DHADHU VIRTHI KULIGAI

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ABSTRACT

Standardization of herbal medicine is necessary in present days to ensure the quality of the drug. The present study aimed at analysis of heavy metals and microbial load contaminant in the siddha drug Dhadhu Virthi Kuligai. The results revealed that 0.0373ppm of Lead is present in the drug and total bacterial count was found to be 3 x 10^3 CFU/g. Other toxic metals such as Arsenic, Mercury and cadmium are absent in the drug and microbial contaminants such as *E.coli*, *Enterobacteriaceae*, *Salmonella spp.* and *Staphylococcus aureus* are also absent in DVK. Hence, the drug is free of toxic heavy metals and microbial contaminants.

KEYWORDS: Dhadhu Virthi Kuligai, Heavy metals, microbial load.

INTRODUCTION

Plant materials are used throughout developed and developing countries as home remedies, over the counter drug products and raw materials for the pharmaceutical industry, and represent a substantial proportion of the global drug market.[1] Over the recent times, the popularity of herbal medicine is increased to such an extent that around 20% of world population is now using herbal medicine in different forms for different purposes.[2]

Countries have their own set of laws and regulations for herbal as well as traditional medicines. WHO recommends that each country or area must adopt a regulatory system to manage the appropriate use of herbal medicine.[3] For the acceptance of these drugs, a minimum level of quality control is required. But when the question of bulk production, marketing and distribution to the public arises, to ensure uniformity, quality control becomes an imperative need.[4] Thus, the present study deals with the standardization of Dhadhu Virthi Kuligai (DVK), which is a Siddha classical herbal drug used to treat diseases of male reproductive system such as oligospermia, Asthenozoospermia etc, as mentioned in Noikalukku Siddha Parikaaram.[5]

MATERIALS AND METHODS

Procurement and Authentication of Raw Drugs

The drugs were purchased from authorized country raw drug store in Chennai. All the plant materials, *Mucuna pruriens, curculiogo orchiodes, Hygrohila auriculate, Asparagas racemosus* and *Acacia nilotica* were identified and authenticated by Botanist, National Institute of Siddha, Tambaram Sanatorium, Chennai.

Preparation of Dadhu virthi kuligai

All these ingredients were powdered and soaked in lime juice for 24 hours and ground well with the same juice for 3 days and allowed to dry. Again, ground with tender coconut water for 3 days, until it attains a waxy consistency, then it was made into small size pills (5 grains-325mg).

DETAILS REGARDING EXPERIMENT

Determination of heavy metal analysis

Thermo fisher M series, 650902 VI.27 model Atomic absorption spectrophotometer was used in the determination of heavy metal elements present in DVK Flame technique was used for Lead and Cadmium, various wavelengths were followed for different metals such as Lead (217nm) and Cadmium (228.8nm), for Mercury (253.7nm) and Arsenic (193.7nm) Cold vapor technique and flame vapor technique was used respectively.[6] The hallow cathode lamp for Pb, Cd, Hg and As analysis were used as light source to provide specific wavelength for the elements to be determined.

Microbial Analysis

Microbial analysis was carried for determination of microbial contamination as per procedures described in Pharmacopoeia of India. Total bacterial count, total fungal count, identification of specified organisms such as Escherichia coli, Salmonella sp., Staphylococcus aureus and Enterobacteriaceae were analyzed.[8-11]

RESULTS AND OBSERVATIONS

Table 1: Instrumental condition for analysis.

Parameter	Pb	Cd	Hg	As
Wavelength	217 nm	228 nm	253.7 nm	193.7nm
Slit width	0.5 mm	0.5 mm	0.5 mm	0.5 mm
Ligh Source	HCL	HCL	HCL	HCL
Lamp current	4.0 mA	3.0 mA	3.0 mA	6.0 mA
Carrier gas and flow rate	Air and Acetylene & 1.1 L/min	Air and Acetylene & 1.1 L/min	Argon & 1.1 L/min	Acetylene, Argon & 1.1L/min
Flow rate	2 ml/min	2 ml/min	5 ml/min	5ml/min
Technique	Flame	Flame	Cold vapour	Flame vapour

Note: HCL- Hollow Cathode Lamp

Table 4: Analysis of Heavy Metals.

S. No.	Name of the Element	Results	Limits (As per PLIM)
1	Lead	0.0373 ppm	10 ppm
2	Cadmium	Not Detected	0.3 ppm
3	Arsenic	Not Detected	03 ppm
4	Mercury	Not Detected	01 ppm

Table 4.1: Analysis of Microbial Load.

S. No.	Parameters	Results	Permissible Limit for Internal use
1	Total Bacterial Count (TBC)	3×10^3 CFU/g	10^5 CFU/g
2	Total Fungal Count (TFC)	Absent	10^3 CFU/g
3	*Enterobacteriaceae*	Absent	10^3 CFU/g
4	*Escherichia coli*	Absent	10 CFU/g
5	*Salmonella Spp.*	Absent	Absent
6	*Staphylococcus aureus*	Absent	Absent

The procedures recommended for analysis of Microbial Load as per WHO, 1998.

The daily Per Day exposure limit of Lead in raw herbal materials is 10 ppm as per PLIM guidelines[12] and WHO recommendations of limits of lead is 10 mg/kg.[13] The presence of lead is very negligible in *Dhadhu Virthi Kuligai* as compared to the recommended limits. Other toxic metals such as Arsenic, Mercury and Cadmium are in below detectable level. The

Total Bacterial Count present in the drug are $3x10^3$ CFU/g and the permissible limit in herbal medicines as per WHO is 10^5 CFU, which indicates that TBC is within the WHO permissible limits. There is no fungi were detected in the drug and the bacteriae Enterobactiaceae, *E.coli*, *Salmonella Spp* and *Staphylococcus aureus* were also not detected in DVK.

CONCLUSION

Analysis of Heavy metal showed that the only toxic metal found was Lead that too was within WHO permissible limits. Microbial load analysis showed that Total Bacterial Count was present within the WHO permissible limit and Fungi Enterobacteriaceae, *E.coli*, *Salmonella Spp* and *Staphylococcus aureus* was absent. So, it can be considered that the drug is not contaminated with any toxic or microbial contaminants.

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