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PHYSICOCHEMICAL AND PHYTOCHEMICAL EVALUATION OF *CHLOROPHYTUM BORVILIANUM* AND *SAPINDUS MUKORROSSAI*

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ABSTRACT: The study is aimed at the development of physicochemical parameters and to investigate the active principle present in *Chlorophytum borvilianum* and *Sapindus mukorrossai*. The present work embodies the investigations carried out to establish methods for quality control of drugs as per WHO guidelines: complete botanical evaluation which comprises of macroscopy, quantitative microscopy, powder microscopy, physicochemical parameters like loss on drying, extractive values, saponification value, total ash values, acid insoluble ash value, water-soluble ash value, pH value, loss on drying, bulk density and fluorescence analysis of powdered drug to investigate the phytochemicals present in the extracts in the preliminary level.

INTRODUCTION: Herbal medicines are the promising choice over modern synthetic drugs. These are believed to be safe and having a minimum or no side effects. Generally, herbal formulations involve the use of fresh or dried plant parts. It is essential to have correct knowledge of such crude drugs in preparation, safety and efficacy aspect of the herbal product. Pharmacognosy is a simple and reliable tool, by which complete information of the crude drug can be obtained [1]. Medicinal plants are of great importance to the health of individuals and communities. The medicinal value of these plants lies in some chemical substances that produce a definite physiological action on the human body. *Chlorophytum borivilianum* (Liliaceae) commonly called Safed musli is considered as one of the most important medicinal plant used in our traditional Systems of medicines to treat various diseases of human beings. The plant is used for dysuria [2]. Leaves are eaten by the tribal people of Western Ghats as an expectorant [3]. Root is used as an aphrodisiac, diuretic, and astringent useful in dysentery, as an antidiabetic and as appetizing agent [4]. Tubers are used as lactagogue. [5]

Sapindus mukorossi is a deciduous tree belongs to the family Sapindaceae grew in upper reaches of Indo-Gangetic plains, Shivaliks and sub-Himalayans tracks at altitudes from 200 m to 1500m. Also known as a soap-nut tree, it is one of the most important trees of tropical and subtropical regions of Asia.

MATERIAL AND METHOD: The roots and fruits of drugs namely *Chlorophytum borvilianum* and *Sapindus mukorrossai* were collected from the local market, Nayaganj, Kanpur, U.P. These drugs were authenticated by Dr. Prabodh Shukla, Pharmacognosy, RIMS. The collected roots and fruits were washed, shade dried and pulverized with a mechanical pulverizer for the size reduction. It was then passed with 60 mesh size sieve, and the fine powder was collected and used for the studies.

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Pharmacognostic Studies: Macroscopic studies of dried roots and fruit of both *Chlorophytum borvilianum* and *Sapindus mukorrossai* were carried out, which comprise of organoleptic parameters [6], *viz.* color, odor, taste, texture, etc **Table 1**. Powder microscopy was performed to investigate the microscopical features as per Evans [7] **Fig. 1**, **2** and **3**.

Quantitative Microscopy: In transverse sections it is not possible to study nature of epidermal cells, trichomes, and stomata; stomatal index, vein islet number and vein termination number which play an important role in identifying characteristics of crude drugs and adulterants [8, 9].

But, these can be determined by quantitative microscopy. These quantitative microscopic values are comparatively constant for a particular species and can be used to make a difference in closely related species. For the study of epidermal cells and stomatal index, leaf segments were boiled in chloral hydrate solution, and the epidermis was peeled with forceps and then mounted on a slide with 5% (v/v) glycerin in water and observed under a microscope. Stomatal index was calculated as Number of stomata/Total number of epidermal cells multiplied by 100. Microscopic descriptions of tissue are supplemented with micrographs wherever necessary.

For determination of vein islet number and vein termination number a 1 mm square leaf piece was cleared by boiling in chloral hydrate solution and then observed under a microscope for determining vein islet and terminations [10] **Table 4**.

Fluorescence Analysis: Crude drugs show their characteristic fluorescence when exposed to ultraviolet radiation and are dependent on its chemical constituents. This analysis is useful to identify adulterants during crude drug evaluation. In the present study, one gram of crude drug was taken in a watch glass and subjected for fluorescent analysis as such and after treatment with different reagents [11, 12, 13] **Table 5**.

Physicochemical Studies:

Determination of Ash Values: For determining ash content of drug, about 3 g of powder was spread in a pre-ignited and weighed silica crucible. Then the crucible was incinerated gradually to make the crucible free from carbon. After cooling, the crucible was weighed to get the total ash content and then the ash was subjected for determining the acid insoluble and water soluble ash. The percentage of total ash was calculated by taking the air dried sample as standard [14].

Determination of Acid-Insoluble Ash: Twenty-five (25) ml of hydrochloric acid (~70g/l) TS was added to the crucible containing the total ash, covered with a watch-glass and boiled gently for 5 minutes. The watch-glass was rinsed with 5 ml of hot water and this liquid added to the crucible. The insoluble matter was collected on an ashless filter-paper (Whatmann 41) and washed with hot water until the filtrate was neutral.

The filter paper containing the insoluble matter was transferred to the original crucible, ignited by gradually increasing the heat to 550 °C for 3 h in a muffle furnace (Nabertherm) to constant weight. Allowed the residue to cool in a suitable desiccator for 30 minutes, and then weighed without delay. Acid-insoluble ash content was calculated as mg per g of air-dried material.

Determination of Extractive Values: Considering the diversity and chemical nature of the drug, five different solvents *viz.* water, ethanol, chloroform, ethyl acetate, diethyl ether, and petroleum ether were used for determination of extractive values. About 5 g of powdered leaf material was subjected for cold maceration extraction with 100 ml of above solvents.

Determination of extractive values of a crude drug is beneficial in its evaluation process wherever evaluation of chemical components in drugs is not possible by any other means [14]. After extraction, the extracts are concentrated in rota vaporizer and dried in the vacuum desiccator. Then the extractive values are calculated as percentage w/w of solvent soluble extractive with reference to the air dried drug.

Determination of Water-Soluble Ash: Twenty-five (25) ml of water was added to the crucible containing the total ash, covered with a watch-glass and boiled gently for 5 min. Insoluble matter was collected on an ashless filter-paper. Washed with hot water and ignited in a crucible for 15 min at a temperature not exceeding 450 °C in a muffle

furnace. Allowed the residue to cool in a suitable desiccator for 30 min, and then weighed without delay. The weight of the residue was subtracted in mg from the weight of total ash. Water - soluble ash content was calculated as mg per g of air-dried material.

Determination of Moisture Content: Moisture content was determined by loss of weight on drying (LOD) method [14]. For this 5 gm of drug (powdered leaf material) was taken and kept in an oven at 105 °C till a constant weight was obtained. Amount of moisture present in the sample was calculated as a reference to the air-dried leaf material.

Determination of pH Range: The pH of different formulations in 1% w/v (1g: 100ml) and 10% w/v (10g: 100ml) of water-soluble portions of whole powder of drugs, were determined using standard simple glass electrode pH meter [15] **Table 2**.

Preliminary Phytochemical Screening: Table 3
The preliminary phytochemical screening of the water (hot) extracts of plant powder of drugs were carried out using standard laboratory procedures, to detect the presence of different secondary metabolites (phytochemical constituents) such as alkaloids, flavonoids, saponins, tannins, steroid glycosides, phenols, coumarins, reducing sugars, protein, anthraquinones, quinines, Fixed oils and fats [16-19].

Determination of phenolic compounds two to three drops of 1% ferric chloride ($FeCl_3$) solution was added into 2 ml portions (1%) of each extract. Phenolic compounds produce a deep violet color with ferric ions.

Determination of Tannins:
Ferric Chloride Test: A small quantity of the extract was boiled with water and filtered. Two drops of ferric chloride were added to the filtrate, formation of a blue-black or blackish green color in the presence of ferric chloride precipitate was taken as evidence for the existence of tannins.

Determination of Flavonoids:
Shinoda Test: The extract was dissolved in methanol (50%, 1-2 ml) by heating. To an alcoholic solution of each of the extract, three pieces of magnesium chips were added followed by a few drops of concentrated hydrochloric acid. The appearance of orange, pink or red to purple color indicates the presence of flavonoids.

Determination of Steroid Glycosides:
Libermann Burchard'S Test: Extract was dissolved in equal volumes of anhydrous acetic acid and chloroform ($CHCl_3$) and cooled to 0 °C. The mixture was transferred to a dry test tube, and concentrated sulfuric acid (H_2SO_4) was introduced to the bottom of the tube. Formation of a reddish brown or violet-brown ring at the interface of the two liquids indicates the presence of steroids.

Determination of Alkaloids:
Mayer"s Test: One ml portions of each extract was acidified with 2-3 drops of 1M hydrochloric acid and treated with 4-5 drops of Mayer"s regent (Potassium mercuric iodide) formation of a yellow or white colored precipitate or turbidity indicates the presence of alkaloids.

Dragendroff"s Test: Extracts were dissolved individually in dilute hydrochloric acid and filtered. Filtrates were treated with Dragendroff"s reagent (solution of potassium bismuth iodide). Formation of red precipitate indicates the presence of alkaloids.

Detection of Proteins:
Xanthoproteic Test: The extracts were treated with a few drops of conc. nitric acid. Formation of yellow color indicates the presence of proteins.

Determination Saponins:
Foam Test: 0.5 g of extract was shaken with 2 ml of water. If foam produced persists for ten minutes, it indicates the presence of saponins.

Detection of Reducing Sugar:
Fehling"S Test: To a test tube 1 ml each a Fehling"s A, and B solutions were added and mixed. To this, ~2 ml of plant extract was added and heated on a boiling water bath for ~10 minutes. Formation of brick red or orange precipitate indicates the presence of reducing sugar/ carbohydrates.

RESULTS: The organoleptic characters of the dried roots and fruit of both *Chlorophytum borvilianum* and *Sapindus mukorrossai* powder are tabulated as **Table 1**.

TABLE 1: ORGANOLEPTIC EVALUATION PARAMETERS

S. no.	Organoleptic parameters	Inference	
		Chlorophytum borvilianum	***Sapindus mukorrosai***
1	Colour	Pale white	Brown
2	Odour	Characteristics	Characteristics
3	Taste	Mucilaginous	Bitter
4	Nature	Crystalline	Crystalline
5	Texture	Smooth	Rough
6	Solubility	Forms colloidal solution in water & insoluble in alcohol & chloroform	Soluble in water

TABLE 2: PHYSICOCHEMICAL STUDY OF POWDERED DRUGS

S. no.	Parameters	Percentage (%w/w)	
		Chlorophytum borvilianum	***Sapindus mukorrosai***
1		Ash Value	
	Total ash	7.19	3.60
	Acid-insoluble ash	3.66	0.24
	Water soluble ash	2.05	14.69
2		Successive extractive values	
	Pet. ether	1.4	1.1
	Diethyl ether	1.3	1.4
	Chloroform	1.1	1.8
	Ethyl acetate	2.1	2.2
	Ethanol	2.7	3.2
	Aqueous	5.8	4.8
3	Loss on drying	12.66	9.6
4		pH values	
	1% water solution	5.4	5.9
	10% water solution	4.5	5.3
5	Bulk density	0.8	1.2
6	Saponification values	182.3	148.66

Note: values are the average of three experiments.

TABLE 3: PRELIMINARY SCREENING OF MAJOR PHYTOCHEMICALS

S. no.	Chemical constitutents	Tests / reagent	*Chlorophytum borvilianum* (aqueous extract)	*Sapindus mukorrosai* (aqueous extract)
1	Alkaloids	Dragendorff's	-	-
		Wagners	-	-
		Mayer's	-	-
2	Carbohydrate	Molisch's	+	+
		Fehling's	-	+
		Benedict's	+	+
3	Flavonoids	Mg ribbon and dil. HCl	-	+
4	Glycosides	NaOH test	+	+
5	Tannins/Phenols	Ferric chloride	+	+
		Liebermann's	+	+
		Lead acetate	+	+
6	Protein's	Xanthoproteic	+	-
		Biuret	+	-
7	Starch	Iodine	+	+
8	Saponins	Frothing with $NaHCO_3$	+	+
9	Steroids / Terpenes	Salkowski	-	+
10	Amino acid	Ninhydrin solution	+	-
11	Resin	Acetic anhydride	+	+

Indication: (-) Absent & (+) Present

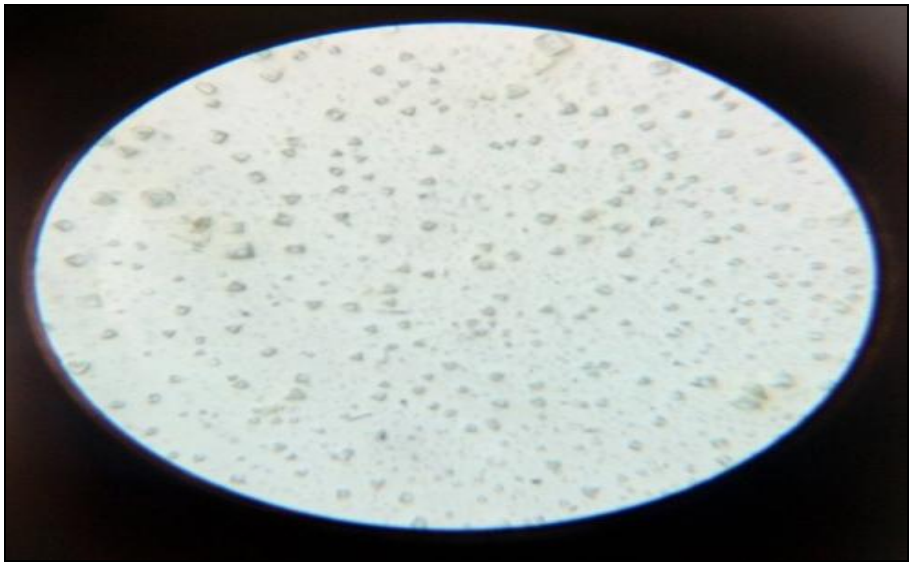

FIG. 1: CALCIUM OXALATE CRYSTALS OF *SAPINDUS MUKOROSSAI*

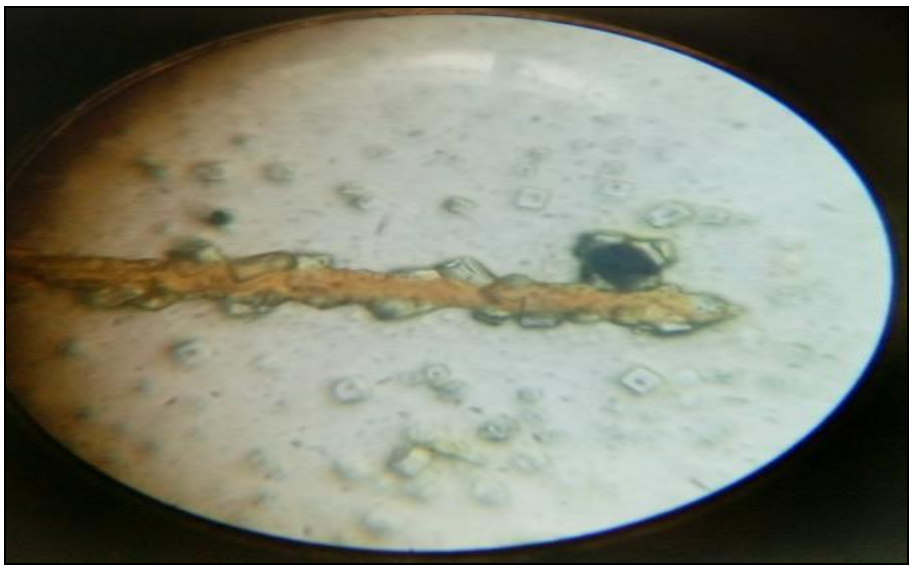

FIG 2: CALCIUM OXALATE CRYSTALS ATTACHED WITH FIBER IN *SAPINDUS MUKOROSSAI* POWDER

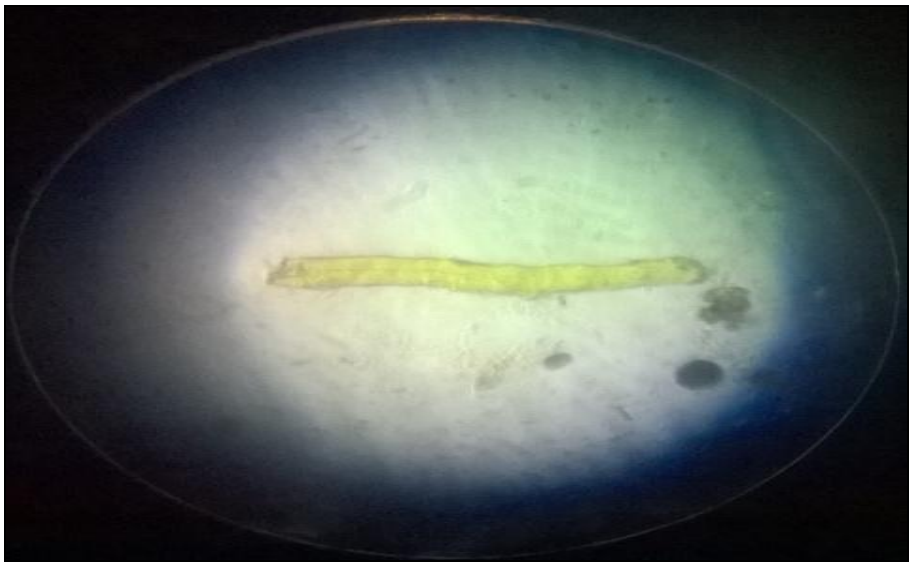

FIG. 3: FIBER IN *SAPINDUS MUKOROSSAI* POWDER

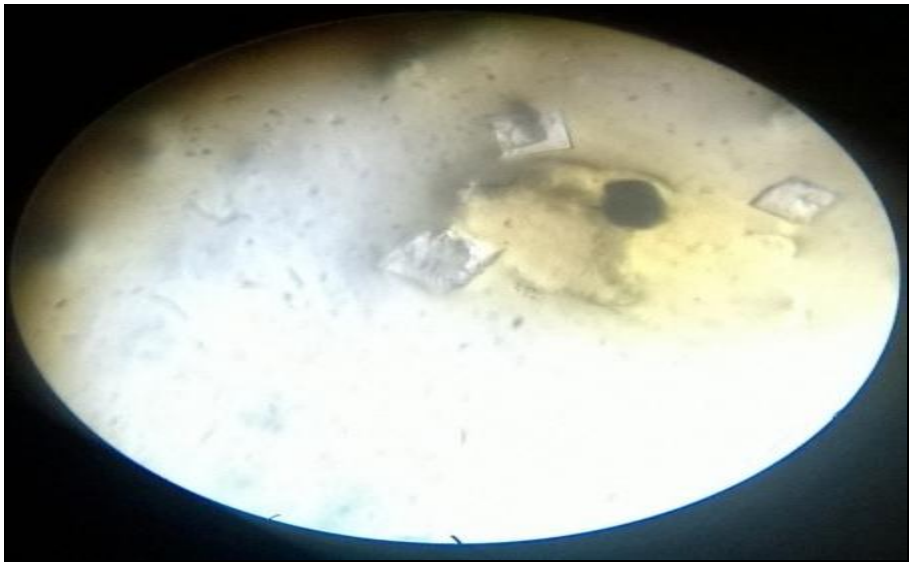

FIG. 4: STONE CELL WITH CALCIUM OXALATE CRYSTALS

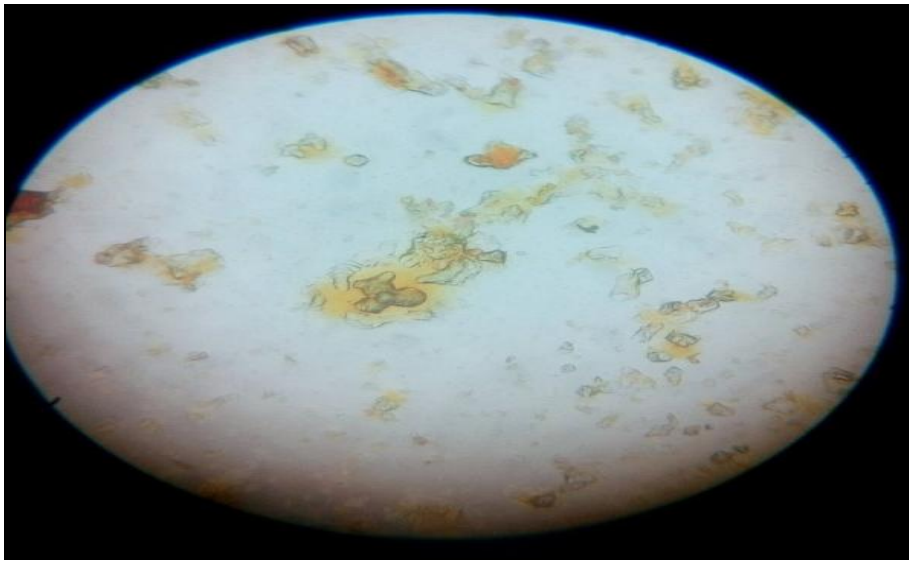

FIG. 5: POWDER MICROSCOPY OF *CHLOROPHYTUM BORVILIANUM*

TABLE 4: DETERMINATION OF PHYTOCONSTANTS

S. no.	Leaf constant	Report (/mm^2)	
		Chlorophytum borvilianum	***Sapindus mukorrosai***
1	Vein islet number	12.4	11.2
2	Vein termination number	11.6	12.9
3	Stomatal index (upper epidermis)	7	5.4
4	Stomatal index (lower epidermis)	18	15

TABLE 5: FLUORESCENCE ANALYSIS OF CRUDE DRUGS WITH DIFFERENT CHEMICAL REAGENTS

S. no.	Powdered drug + Chemical reagent	Daylight	UV Short	UV long
1	Safed musli + Conc. HCl	Brown	Green	Black
	Reetha + Conc. HCl	Dark brown	Dark brown	Black
2	Safed musli + Dil. HCl	Brown	Green	Black
	Reetha + Dil.HCl	Dark brown	Dark green	Black
3	Safed musli +Iodine solution (2%)	Dark brown	Dark green	Black
	Reetha + Iodine solution (2%)	Dark brown	Dark green	Black
4	Safed musli + Glacial acetic acid	Yellow	Green	White
	Reetha + Glacial acetic acid	Dark brown	Brown	Black

5	Safed musli +Conc. HNO_3	Dark brown	Dark green	Black
	Reetha + Conc. HNO_3	Dark brown	Black	Black
6	Safed musli + Dil. HNO_3	Dark brown	Dark green	Black
	Reetha + Dil. HNO_3	Light brown	Light green	Grey
7	Safed musli + Glacial acetic acid +HNO_3	Light brown	Light green	Green
	Reetha + Glacial acetic acid +HNO_3	Dark brown	Dark green	Black
8	Safed musli + Dil. H_2SO_4	Light brown	Light green	Grey
	Reetha + Dil. H_2SO_4	Dark brown	Dark green	Black
9	Safed musli + Conc. H_2SO_4	Dark brown	Black	Black
	Reetha + Conc. H_2SO_4	Dark brown	Dark green	Black
10	Safed musli + Fehling's reagent	Light brown	Light green	Grey
	Reetha + Fehling's reagent	Dark brown	Dark brown	Black
11	Safed musli + Picric acid solution (Hager's reagent)	Yellow	Light green	Black
	Reetha + Picric acid solution (Hager's reagent)	Brown	Light brown	Black
12	Safed musli +NaOH (10%)	Light brown	Green	Black
	Reetha + NaOH (10%)	Brown	Dark green	Black
13	Safed musli + KOH (10%)	Light brown	Light green	Dark brown
	Reetha + KOH (10%)	Dark brown	Light brown	Black
14	Safed musli +Wagner's reagent	Black	Dark brown	Black
	Reetha + Wagner's reagent	Dark brown	Brown	Black
15	Safed musli + $CuSO_4$ (5%)	Bluish	Light green	Black
	Reetha + $CuSO_4$ (5%)	Bluish	Light green	Black
16	Safed musli + Lead acetate	Brown	Light green	Black
	Reetha + Lead acetate	Light brown	Light green	Black

Powder microscopy of *Sapindus mukorrossai* reveals the presence of calcium oxalate crystals, fibers, stone cells **Fig. 1**, **2**, **3**, **4** whereas powder microscopy of *Chlorophytum borvilianum* stone cells, starch granules and raphides **Fig. 4**. The Average physicochemical parameters of the *Chlorophytum borvilianum* and *Sapindus mukorrossai* powder are tabulated as **Table 2**. The Preliminary phytochemical screening for various functional groups is tabulated as **Table 3**. The list of phytoconstituents parameters is summarized in **Table 4**. Fluorescence analysis of powdered drugs is presented in **Table 5**.

DISCUSSION: Physicochemical parameters like ash values, moisture content and fluorescence analysis help determine the physiological and non-physiological ash, the possibility of microbial growth or contamination and presence of impurities respectively. The relative low acid insoluble ash value 3.66% of *Chlorophytum borvilianum* and 0.24% of *Sapindus mukorrossai* and a high ratio of water-soluble ash content 2.05% of *Chlorophytum borvilianum* and 14.69% of *Sapindus mukorrossai* indicates that the crude drug contains more physiological ash or contents (*i.e.* related to plant tissue) than the non- physiological content. The relative low moisture content indicates that the drugs give an equal possibility for microbial growth and contamination. The preliminary phytochemical investigations in *Chlorophytum borvilianum* and *Sapindus mukorrossai* aqueous extract indicated the presence of glycosides, tannins, flavonoids, saponins and triterpenoids in more quantity whereas the sterols and proteins in less quantity, but there are no fats and oils. Presence of such phytochemical constituents may be responsible for various pharmacological activities of this elite medicinal plant. Medicinal plants are considered as living factories as they produce various phytochemicals *viz.*, alkaloids, flavonoids, glycosides, phenols, saponins, sterols, etc in the form of secondary metabolites.

These serve as life-saving drugs. Due to this, day by day demand for crude drugs is increasing and also adulteration of crude drugs. Hence, standardization of a crude drug is essential to avoid and identify adulteration. In standardization of a crude drug, macroscopic and microscopic evaluation is the primary step. According to WHO, standard botanical investigations like epidermal cells, stomatal index, vein islet and termination

values, etc are mandatory for the diagnosis of the crude herbal drug.

CONCLUSION: In the present investigation, a set of pharmacognostical standardization parameter studies were conducted on *Chlorophytum borvilianum* and *Sapindus mukorrossai* as per pharmacopeia and WHO guidelines. These studies revealed the presence of various important bioactive compounds and proved that these plant drugs are also medicinally important. These results may help in standardization, identification and in carrying out further research in *Chlorophytum borvilianum* and *Sapindus mukorrossai* based drugs which are used in Ayurveda and modern pharmacopeia.

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REFERENCES:

1. Modi DC, Patel JK and Shah BN: Pharmacognostic Studies of the seed of *Syzygium cumini* Linn. 2010; 2: 20-26.
2. Negi KS, Tiwari JK, Gaur RD and Pant KC: Notes on the ethnobotany of five districts of Garhwal Himalayas. Ethnobotany 1993; 5: 73-77.
3. Sebastian MK and Bhandari MM: Medicinal Plant flora of Udaipur district, Rajasthan. Bulletin of Medicinal Ethnobotany Research 1984; 5: 122-134.
4. Chetty KM and Rao KN: Ethnobotany of Sarakallu and adjacent areas of Chittoor district. Vegetos 1989; 2: 51-58
5. Bhandary MJ, Chandrashekhar KR and Kaveriappa KM: Ethnobotany of Gowlis of Uttara district, Karnataka, India. Journal of Ethnopharmacology 1995; 47: 149-158
6. Chopra R and Ghosh S: Poisonous plants of India. The Manager of Publishers, Delhi, 1946.
7. Trease GE and Evans WC: Pharmacognosy. Saunders Company Limited, New Delhi, Edition 15th, 1996: 516-547.
8. Siddiqui and Hakim MA: Format for the pharmacopoeial analytical standards of compound formulation, a workshop on standardization of Unani drugs, anuary, Central Council for Research in Unani Medicine (CCRUM), New Delhi, (appendix), 1995: 24-25.
9. Agarwal SS: Herbal Drug Technology. Hyderabad: University Press (India) Pvt. Ltd., 2007; 625–679.
10. Nazish I, Kaskoos RA, Mir SR, Amin S and Ali M: Preliminary pharmacognostical standardization of *Ruta graveolens* L. Aerial Parts. Res J Medicin Plant 2009; 3(2): 41-44.
11. Kumar D, Kumar K, Kumar S, Kumar T, Kumar A and Prakash O: Pharmacognostic evaluation of leaf and root bark of *Holoptelea integrifolia* Roxb. Asian Pac J Trop Biomed 2012; 169-175.
12. Anonymous: State pharmacopeia committee of china, Pharmacopoeia of People's Republic of China 2010; 10th, appendix IX K: 53.
13. Mukherjee PK: Quality Control of Herbal Drugs, Business horizon, New Delhi, Edition 1st, 2002: 356:187–195.
14. Choudhary N and Sekhon BS: An overview of advances in the standardization of herbal drugs. J Pharm Educ Res Vol. 2011; 2(2): 55-70.
15. Saxena PNN, Shrivastava and Saxena RC: Preliminary Physico-phytochemical study of stem bark of *Alstonia scholaris* (L) R. BR. - A Medicinal Plant. IJPSR 2012; 3(4): 1071-1075.
16. Kokate CK, Khandelwal KR, Pawer AP and Gokhale SB: Practical Pharmacognosy, Nirali Prakashan, Pune, 3rd Edition 3rd, 1995: 137.
17. Farnsworth NR: Biological and Phytochemical screening of Plants. J of Pharmaceutical Science 1996; 55: 225-276.
18. Tiwari P, Kumar B, Kaur M, Kaur G and Kaur H: Phytochemical screening and extraction: A review, Internationale Pharmaceutica Sciencia 2011; 1(1): 98-106.
19. Gupta AK, Tandon N and Sharma M: Quality Standards of Indian Medicinal Plants, Indian Council of Medical Research, New Delhi, Vol. 7, 2008: 331-339.

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