Development of Alternative Methods for the Putapaka Swarasa of *Calotropis procera*

A DISSERTATION REPORT
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In

Rasashastra and Bhaishajya Kalpana (Ayurvedic Pharmaceutics)

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This is to submit that this written submission in my project report entitled "Development of

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ideas in my own words and where other's ideas and words have been included, I have

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List of Abbreviations

Temp.- Temperature

⁰C - Degree celcius

mg.- Miligram

kg.- Kilogram

μg.- Microgram

ml.- Miligram

Me.- Methanol

DL.- Dry latex

INTRODUCTION

Putpaka is method used to provide adequate heat for converting material in to the desired form. The word "puta" describes quantum of heat whereas paka describes the process. This method generally used to prepare bhasma of minerals and metals in Rasashastra. The amount of heat is varied according the material used to convert into bhasma. Hence, it has been classified on the basis of the source of energy i.e. Bhanu, Surya and Agniputa. Agniputa, further classified according to the quantum of heat as Mahaputa, Gajjaputa, Varahputa, Kukkuutaputa, Kapotputa, and Gorvarputa. Mahaputa has highest temperature in all these types of puta. The application of puta is based on the type of drug and material to be processed for example Gajaputa is used to prepare the bhasma of abhraka, and loha.

Putapaka is not only used for the treatment of minerals and metals. Though, herbal drugs can also be treated by using putapka in Bhaishajya Kalpana. In addition to this, Putapaka is also classified as a part of netra chikitsa. However, process described in netra chikitsa and bhaishajya kalpana is same. The purpose of putapaka is to extract the Swarasa from the drug with less water content such as Vasa, Arka, Gambhari, Vata, and Erand patra. But, this process is rarely used due to its tedious steps, less yield, short shelf life, and rapid urbanization of the society. Hence, present study has been designed to develop modern and alternate method for the putapaka of herbal drugs.

CHAPTER 2 REVIEW OF LITERATURE

2.1 Review literature of putapaka

2.1.1 Putpaka method:

Putpaka is a classical method that is mentioned in different brances of Ayurveda. In different branches it have different role. In Rasashastra it is the best method to convert minerals and metals into bhasma form. The bhasma prepared in this way are very light and assimilate in body very well. From Baishjaya kalpna view it is use to extract the juice (swarasa) of that dravya that cannot produce swarasa in wet or fresh form. The swarasa produced from this method having great therapeutic efficacy. In the same way it is the best method that is use to treat various eye disorders and it is part of ophthalmic medication. The swarasa produced from putpaka method are utilized to cure eye diseases and is a part of netra kalpa.

2.1.2 References found in various Samhita's: In various Samhita's references are available regarding putpaka.

S.No	Samhita	Description
1	Shusruta samhita ¹	Described putpaka for eye disorders. Also for the treatment of atisara and various formulations for them.
2.	Astang samgrah ²	Described putpaka for netra chikitsa, types of putpaka, material used for treatment and indications of putpaka dravya for diseases.
3.	Sharangdhara samhita ³	Described putpaka for eye disorders. Also for the treatment of atisara and various formulations for them.

Table1: Reference found in Samhita

2.1.3 Putpaka role in Ayurveda:

2.1.3.1 Putpaka in Rasashastra: In Rasashastra putpaka is a method which define the quantum of heat required by rasa dravya for their proper paka or bhasmikaran. The

medicine that is supakava or paka properly is considered best because only after that they are suitable for internal use.⁴

2.1.3.2 Classification of putpaka:

In Rasashastra putpaka is classified in to three main types: ⁵

- a) Agni puta (paka through agni)
- b) Surya puta (paka through sun rays)
- c) Chandra puta (paka through moon rays)

Agni puta is further divided into two types on the basis of providing energy that and direct and indirect. Lavak puta, kapot puta, kukutputa, varah puta gaja puta are the main examples of application of direct agni. Whereas, baluka puta, lavana puta and gorvarna puta are the examples of indirect agni. Specific puta is described for the specific material like gajaputa is used for the preparation of bhasma of loha and kukutputa for swarn bhasma etc.⁶

Application of puta for minerals and metals⁷:

S.No	Puta name	No. of Vanopalas	Temparature required ⁰ C	Uses
1	Maha puta	1500	1000	Abhraka,heerak, etc.
2	Gaja puta	700-600	1000	Abhraka,kurmapristha, manikya, etc.
3	Varah puta	150	950	Raupya, swarnamakshika, etc.
4	Kukkuta puta	100	950	Tutha, mrigshringa, etc.
5	Kapotha puta	8	600-800	Raupya, swarna, parad, etc.

Table: 2 Classification of putpaka for rasashastriye dravya.

2.1.4 Putpaka in netra chikitsa In various Samhita's, putpaka has been mentioned for the treatment of eye disorders. Their types or methods are also described with different drug and duration of treatment.

- **2.1.4.1 Description of putpaka in Sushruta Samhita regarding Netra chikitsa:** in Sushruta samhita, putpaka is described for netra chikitsa, atisara and timir roga. The process for putpaka described in this Samhita is that firstly drug is taken and its kalka is prepared that kalka is further packed in to gambhari, kamal, kadli, eranda, patla etc. Then, kapadmitti is applied and subjected to puta and in the end swarasa is extracted⁸. The drug material is different according to diseases.
- **2.1.4.2 Description of putpaka in Astang samgraha and its classification:** ⁹ The method of putpaka is same like that in Sushruta Samhita. In this Samgraha putpaka different types like snehan, lekhan and ropan putpaka are described for different dosha of eyes. In Astanga samgraha, term prasadan is used for ropan and indicated for vataj, pittaj, raktaj and netra vrana dosha. Sources of material for particular diseases are also explained in this samgrah.

Classification of putpaka in Astang samgraha on the basis of dosha:

S.No.	Types	Matara kaal	Dosha
1	Snehan putpaka	200 matra kaal	Ati ruksha netra.
2	Lekhan putpaka	100 matra kaal	Atisnigdh netra.
3	Ropan (prasadan)	200 matra kaal	Vaataj, pittaj, rakatjanya and
	nutnolzo		natra vrana
	putpaka		netra vrana

Table3: Classification of putpaka on the basis of dosha

2.1.4.3 Classification of putpaka in Astang samgraha on the basis of drug used: 10

S	Types of	Predo	Source	of raw mat	terial	Liq.	Temp.	Mat	Ava
	putapaka	minant	Animal	Mineral	Herbal	materia		ra	d
N			Origin	and	origin	1		kaal	Hi
О				metals	origin				(Day
									s)
1	Snehana	Vaat	vasa, majja,	-	Jeevanee	Ksheer	Sukos	200	3
			masa of		ya		hna		
			Bhaumya,		gana,Ka				
			Prasaha,		kol				
			Anupa desa		yadi				
			animals		gana,				
			aiiiiiais		Madhur				
					Rasa				
					dravya				

					etc.				
2	Lekhana	Kapha	Yakrit or peshi of animals living in Jangal desha.	Lauha, shnkha, praval, moti bhasma and kasesa	Shonth, pippali maricha	Mastu	Sukos hna	100 - 200	1
3	Ropana	Pitta	Jangal desha	-	Dravyas with Tikta Ras i.e Madhu.	Breast milk and ghrita	sheeta	300	Up to 3
4	Prasadana	Tridos ha	Ykrit, asthimaja, peshi vasa, hridya of animals and birds etc.		Madhur Rasa Dravya	Mil k, ghrita	sheeta	300	2

Table 4: Classification of putpaka on the basis of drug use.

2.1.4.4 Description of putpaka in Salakya vijnana: 11

In salakya vijnana the whole process of preparing medicine from putpaka method is describe with type of drug use for snehan, lekhan and ropan putpaka. For snehan putpaka madhur aushad used whereas for lekhan putpaka kashay or for ropan putpaka tikta dravya are mentioned. After that the kalka is prepared from dravya and it is wrapped in to gambhari, kumud, erand or kadli patra after that kapadmitti is applied and it is subjected to heat. The fuel used for paka is of khadir kastha, eranda, adusa and vata and till red hot heat is provide. Then it is removed from heat and in the end the swarasa is extracted and utilised for eyes.

2.1.5 Benefits of putpaka: ¹² Putpaka have beneficial role in many diseases and method is used from old time. According to the acharyas it is the one of best method to treat eyes disorders. In other diseases like atisara, swasa, also have great role and give good results. Yet, the process is lengthy but due to its best result it is utilized. There are many aushad dravya that are utilized for eyes disorders by putpaka method and they

have good therapeutic activities. In eye treatment it give many benefits if the treatment given in perfect manner. It give benefits like

- a) If the matra of all the putpaka methods is given in right way then it is very useful for the eyes like it cures inflammation of eyes and also removes the dust.
- b) Increase the blood flow of eyes.
- c) It removes the swelling and pain of eyes.
- **2.1.6 Putpaka role in Bhaishjaya kalpna:** ¹³ Acharya Sharangdhar describe process of putpaka for extraction of swarasa of various aushadhi in Bhaishjaya kalpana.

2.1.6.1 Classical method of putpaka in Bhaishjaya kalpana:¹⁴

Acharya Sharangdhara describe this process in to various steps and every step have its own importance. In the whole process there are following steps:

Collection and cleaning of herb

- Preparation of kalka.
- Wrapping of kalka.
- Application of kappadmitti
- Subjected to heat
- Extraction of juice

Collection of herb and cleaning: Fresh drug is taken and properly cleaned with the help of water. Washing removes the impurities like dust.

Preparation of kalka: After the washing, kalka is prepared by crushing drug with liquid material and this lead to increase in surface area that gives more yield.

Wrapping of kalka: Kalka is then wrapped in leaves like in gambhari, jamun etc. The wrapping of kalka in leaves have own importance it protect the contact of material i.e kalka with kappadmitti that is applied after wrapping.

Application of kappadmiti: After the wrapping the layer of kappadmiti is applied. The thickness of the kapadmiti is about 2 angul. In the last the kalka is subjected to heat for paka. During the heating process kapadmitti have importantrole that it avoid the burning of material and provide close environment to the kalka due to that evaporation is prohibited and maximum extraction is done.

Extraction of juice: In the end the bolus is removed from the heat and then kappadmitti is removed carefully and bolus is squeezed through cotton cloth.

2.1.7 Dose of putpaka swarasa : 1 pala or 4 tola. ¹³

2.1.8 Anupan : Madhu. 14

2.1.9 Various traditional formulations prepared by putpaka method and their indication:¹⁵

Formulations Indications

Arlu Kutaj putpaka: Deepan and atisara roga.

Titir putpaka : Atisara.

Beejpur putpaka : Chardi

Vasa putpaka : Raktpit, kasa, jawara and kashay

Kantkari putpka : Kaphanashak and coughing

Bibhitaki putpka : Swarbhed, coughing and pratishaye

Shunthi putpaka : Deepan

Suran putpaka : Arsha

Mrigshring putpaka: Swasa, jawara, aamvata and kasa

2.2. LITERATURE REVIEW OF DRUG

2.2.1. Arka (Calotropis procera):

In Ayurveda word 'Arka'is used in many contexts like for Sun, God indra, Tamra and saphatika etc. or also as plant. Arka is known in our country from the earliest time¹⁶. This plant has number of ethnomedicinal use. In old time this herb is used for the treatment of cholera, indigestion and for worm infection. This plant is also used with combination of other herbs to treat the diseases like cold, eczema and diarrhoea¹⁷. Arka is use in Ayurvedic formulation like Arka lavana and Arka taila and both they have great therapeutic activities. Arka is one of the upvisha dravya beyond that it is one of the important drug of Ayurveda and it is used from Vedic period.

2.2.2 Reference found in Vedic Kala:

Vedic kaal: ¹⁸ The mythical origin of Arka is available in Kapishthalakatha Samhita. In the same way it is found in Taittiriya Samhita also. There are different name given to this plant in Vedic period. In Shatpatbrahman, it is explain by the name of Agni and anna. Agni, prana and aditya are also the name of this plant that are mentioned in vedic time. Shatpatbhrahman, relate arka with body parts like arkaparna with karna, arkapushpa with akshi etc.

2.2.3 References found in Various Samhita's:

S.No	Samhita's	Description
1	Charaka samhita	Charaka mention it in ksheerivriksha and added it into Shatashodhana vriksha ¹⁹ . Different uses of the plant also described like Arka is best for dantapawana ²⁰ . Arka root is included in shirovirechan dravya.
2	Sushruta samhita	Sushruta mention arka for kshara preparation method ²¹ . Plant ankur use for the raktamokshana site when the blood is not discharge properly ²² . In this samhita Arka included in Arkadi gana. Arka pushpa is described as kaphapittahara and also kustaghana ²³ . In sushruta samhita various uses of arka are also available.
3	Astang hrudya	Like charaka Samhita it is mention for kaphanasak and dantapawana ²⁴ . In this Samhita the arka lepa with other

		medicine is described for infected wounds ²⁵ .Arka
		ksheera has been indicated for virechana in Alarka.
4	Kasyap samhita 3 rd	In this samhita the combination of Arka with other
	century	drugs indicated for pariseka and abhyanga in the
		patients of kaphaja sotha ²⁶ .

Table5: Arka description in Samhita's

2.2.4 References found in Nighantu about Arka: ²⁷

S.No.	Nighantu	Time period	Varga
1.	Dhanvantari Nighantu	10-13 th century	Karaveeradi varga
2.	Shodhala Nighantu	12 th century	Karaveeradi varga
3.	Shadarasa Nighantu	20 th century A.D	Tiktadravya skanda
4.	Madanpala Nighantu	14 th century	Abhyadi varga
5.	Kaiyadev Nighantu	15 th century	Oshadhi varga
6.	Bhavaprakash Nighantu	16 th century	Guduchyadi varga
7.	Raj Nighantu	17 th century	Karaveeradi varga
8.	Priya Nighantu	21 st century	Shatapushpadi varga
9.	Adarsh Nighantu	20 th century	Arkadi varga
10.	Gunaratnamala	20 th century A.D	Guduchyadi varga

Table: 6 References for Arka in Nighantu

2.2.5 Taxonomic classification: ²⁸

Kingdom Plantae

Subkingdom Tracheobionta

Super division Spermatophyta

Division Magnoliophyta

Class Magnoliopsida

Subclass Asteridae

Order Gentianales

Family Asclepiadaceae

Subfamily Asclepiadoideae

Genus Calotropis

Species <u>calotropis procera</u>

2.2.6 Vernacular names: ²⁹

Sanskrit Ravi, Bhanu, Tapana

Hindi Aak, Madar, Akavana

English Madar Tree

Gujrati Aakado

Marathi Rui

Oriya Arakha

Punjabi Ak

Tamil Vellerukku, Erukku

Kannada Ekka, Ekkaddagida, Ekkegida

2.2.7 Ayurvedic properties of Arka:

Rasa Kattu, Tikta

Guna Laghu Veerya Usna Vipaka Katu

Doshkarma Kaphavatshamak, kaphapittashamak

2.2.8 Description of the plant:

Arka is a weed and milk producing plant so it is called as milk weed. This plant is found at the area of height 1000m. Arka grow in dry area or where the rain fall is less. Plant is also known for ornamental value in mostly dry area³⁰. This plant is related to species of flowering plant and member of Apocynaceae family.



Figure: 1 Habitat of *Calotropis procera*(Ait)

2.8.1 Macroscopy: 31

Bark and branches: The bark of plant is thick, little rough and having yellowish brown colour. The twigs of the plant are of green colour and fleshy. It also having hairs on the surface.

Leaves: The leaves of the plant is simple but opposite to each other and thick. Leaves are greyish white in colour young leaves of the plant is cottony. Leaves produce white latex when a cut is applied on the leaves.

Inflorescence: It arises from the bases of leaves in pedunculate cymes of 3-20.

Flower: Flower having five triangular white petals and five thick ovate petals. The colour of flower vary from base to tip that is white at the base and purple at the tips. The stigma of the flower is surrounded by stamens that are purple in colour from tip. **Fruit:** colour of fruit is green and ovoid in shape. Inside the fruit papery light brown colour seeds are present when it get split into two parts.

Root³²: They are rough, fissured longitudinally, corky and soft, externally yellowish grey white from the internal side it is white in colour.



Figure: 2 Leaves, flowers and fruit of calotropis procera

2.8.2 Microscopy: ³³

Leaf³³: The transverse section of leave when seen under microscope it shows the lamina of leaf having single layer of epidermis. Epidermis is covered with thick striated cuticle. Some of the epidermal cells are also present on the upper and lower surface. In the leaf the parenchymatous cells are also present that are circular in shape. Intracellular space are present in ground tissues. Below the upper epidermis three layer of closely arranged palisade parenchyma cells are present.

Stem: ³⁴ In the Stem the outermost layer is uniseriate having thick cuticle. The cells present are drum to rectangular and compactly arranged. There is presence of small amount of choloroplast in the cells. Endodermis layer of uniseriate cells form a wavy ring around the vascular bundles.

Root:³⁵ A transverse section of root shows outermost cork tissues that are consist of 6-9 rows of tangentially elongated and radially arranged cells followed about 3-6 rows of moderately thick walled parenchymatous cells, vascular cambium present within the phloem consisting of 2-5 rows of thin walled, tangentially elongated cells. Xylem form the central part of root composed of vessels.

2. 2.9 Distribution³⁶:

This herb is present more or less in India mostly in dry places Afganistan, Arabia, Egypt, Iran, and throughout the African continent.

2. 2.10 Chemical constituents of Calotropis procera:

The leaves and stalk of plant contain calotropin and calotropagenin whereas, flower contain calotropenyl acetate and multiflavanol compounds. *Calotropis procera* is a milk weed and its latex contains cardenolide, proceraenin. Its root bark has benzoylinesolone. In the chemical identification this plant also shows occurance of triterpenoids, calotropursenyl acetate and calofriedelenyl, a norditerpenyl esters, calotropternyl and ester oleanene triterpenes³⁷. Plant also have tannins, saponins, cardiac glycosides and bioflavonoids compounds³⁸.

2.2.11 Varieties of Arka in Nighantu:

Different varieties are mentioned in Nighantu:

S.No.	Nighantu	Varities
1	Dhanvantri Nigahntu ³⁹	Arka or Rajarka
2	Bhavprakash Nighantu ⁴⁰	sweta and Rakta
3	Rajnighantu ⁴¹	Arka, rajarka, suklarka and sweta
		mandarka

Table: 7 Varieties mentioned in Nighantu.

2.2.12 Varieties according to modern Botanist:⁴²

- a) <u>Calotropis procera</u> (Rakta)
- b) Calotropis gigantean (Sweta)

2.2.13 Shodhana of Arka:

As the latex of Arka is naturally purified, therefore no need of purification separately.

2.2.14 Therapeutic use:

Pama, vicharchika, swasa, pliha vrddhi, krmidanta, mukhaksata, arsa, sarpa visa, mukha krsnatwa, kasa.

2.2.15 Formulations⁴³:

There are many formulations prepared from arka like arkaksiradi bindu ghrta, arkaditaila, arkadi gana kwath, Arkadi vati and arka lavana.

2.2.16 Treatment of Arka toxicity:

The latex of plant is toxic and the excessive intake of arka ksira causes vomiting and diarrhoea, polyurea and burning sensation in the tongue so for the management of this sugar is mixed in the cold water is given.

2.2.17 Dose⁴⁴: Twak churna 1.5-1g and flower 1-3g

2. 2.18 Ayurvedic pharmacology: 44

Action on digestive system Deepan, pachan,krimighan and rechan.

Action on circulatory system Raktshodhak, shothhar or hridyautejak.

Action on respiratory system Kaphanisark or swashar.

Action on skin: Swedjanan or kusthaghan.

2. 2.19 Reported pharmacological actions:

S.No	Pharmacological activity	Part used	Extract	Model	Dose
1.	Anti inflamatory activity ⁴⁵	Latex from aerial part	Aq.DL,Me.	Winstar rats	100- 250mg/kg
2.	Antifertility activity ⁴⁶	Root	Ethanolic extract	Albino rats	250 mg/kg
3.	Hepatoprotective activity ⁴⁶	Flower	Ethanolic extract	Rats	200 mg/kg
4.	Antidiarrhoeal activity ⁴⁷	Latex	Dry latex	Rats	500 mg/k
5.	Antidiabetic activity ⁴⁷	Latex	Dry latex	Rats	400 mg/kg
6.	Anti hyperglycemic activity ⁴⁷	Dry latex	-	Rats	100-400 mg/kg

7.	Anti tumour	Root	Ethyl acetate	-	10μg/ml
	activity ⁴⁸				
8.	Antinociceptive	Latex		Mice	12.5, 25
	activity ⁴⁹				and
					50mg/kg
9.	Antipyretic	Aerial part	Etanolic	Mice	500mg/kg
	activity ⁵⁰		extract		
10.	Gastroprotective	Stem bark	-	Albino rats	100,200
	activity ⁵¹				and
					400mg/kg

Table: 8 Pharmacological actions of Arka

2.2.20 Toxic profile of Arka:

The latex produce by arka contain many compounds like alkaloids they are calotropin, catotoxin, calcilin and gigantean they are very harmful and having poisonous properties. The latex of arka can cause permanently eye damage. It causes the diminish eye vision and reduces the endothelial cells in the eye with less pain⁵². According to the Ayurveda the eye damage is due to its kshara effect. Externally it also effect the part of the body like it causes the breaking of skin⁵³. Internally if used in large amount it causes vomiting and diarrhoea. Hence, proper care should be taken when in contact with arka like proper washing of hands when working with plant and avoid it in contact of eyes because it can cause severe effect in eyes.

SCOPE OF STUDY

Putpaka is a classical method that is used to extract the swarasa of herbs which have less water content. Extracted swarsa is used to prepare other formulations and netra chikitsa. Size reduction, hydrodiffusion and extraction are the key operations of the putapaka. These operations improve the quality and efficacy of the end product. However, preparation of kalka, wrapping in the leaf of vata, applying thick layer of wheat flour and kapadmitti, and subjecting to heat making process of putapaka tedious and long. So, application of the putapaka became rare which may affect the quality and efficacy of the finished products. Hence, present study has been designed to establish alternate method of putapaka by comparing them with classical method on the basis of analytical study of finished products.

AIMS AND OBJECTIVES

4.1 Aims: Development of alternative method for the Putpaka swarasa of Arka.

4.2 Objectives:

- To authenticate the drug.
- To develop alternative method for putpaka.
- To evaluate and compare the putapaka and developed method on the basis of analytical parameters of arkapatra swarsa.

MATERIAL AND RESEARCH METHDOLOGY

5.1 List of equipment used:

S. no.	List of equipment
1	Beakers
2	Spatula
3	Gas stove
4	Sieves
5	Stainless steel plate
6	Ladle
7	Stainless steel containers
8	Friability apparatus
9	Disintegration apparatus
10	Crucible
11	Furnance
12	UV spectrophotometer
13	Water bath
14	Electric balance
15	Digital pH meter
16	Microscope
17	Vernier caliper
18	China dish
19	Zeta sizer
20	Transmission electronic microscope
21	Pycnometer

Table 9: List of equipment

5.2 Chemical used:

S. No	Material list
1	Wagnor reagent
2	Hager reagent
3.	Glycrine
4	Dragondroff's reagent
5	Lead acetate
6	Ethanol
7	Methanol
8	Chloroform
9	Sodium hydroide
10	Acetic acid
11	Benzene
12	Hydrochloric acid
13	Sulphuric acid
14	Nacl
15	Petroleum ether
16	Ethylacetate
17	Sodium nitropruside
18	Pyridin

Table 10: List of chemical

5.3 Research methodology:

- Identification and Collection of raw drug.
- Physicochemical analysis of raw material.
- Preparation of putpaka from classical method.
- Development of alternative method for putpaka and comparison of classical method
 of putpaka and new method develop on the basis of analytical parameters of
 arkapatra swarasa.

EXPERIMENTAL WORK

- **6.1 Collection of drug: The** leaves of plant Arka collected in August 2017 from Jalandhar India.
- **6.2 Organoleptic study:** Arka observed for colour, odour, texture etc.
- 6.3 Physicochemical Analysis of Arka (Calotropis procera).

6.3.1 Foreign matter:

100 g of sample was taken and spreaded in a stainless-steel tray. The foreign matter was detected with the unaided eye. Remaining quantity of sample was weighed and percentage of foreign matter calculated⁵⁴.

Foreign matter = (Weight of foreign matter / Weight of drug) $\times 100$

6.3.2 Total ash⁵⁴:

Incinerated of the 2.5 gm of the sample into the crucible, at temperature of 450°C for 5 hours. After shelf cooling, kept in the desiccator under vacuum. The weight of obtained ash was measured and percentage of obtained ash was calculated.

Total Ash = (Weight of ash / Weight of sample) $\times 100$

6.3.3 Acid insoluble ash⁵⁵:

Ash obtained from the above method was mixed with 25 ml dilute hydrochloric acid and boiled for 5 minutes. Then, mixture was filtered through ash less filter paper. The filtrate was subjected for the washing with hot water to make it chloride free and again ignited to constant weight. Percentage of acid insoluble ash was calculated after weighing obtained ash.

Acid insoluble ash = (Weight of residue \times Volume made) / (Weight of sample \times Volume taken) \times 100

6.3.4 Alcohol soluble extractive⁵⁵:

5gm of the sample (coarse powder) was taken in a closed conical flask with 100ml of alcohol. Conical flask was shaken frequently for 6 hours and kept undisturbed for 18 hours. Then, it was filtered by using filter paper. 25 ml of filtrate was taken in the china dish and allowed the content to evaporate. Percentage was calculated after weighing the residue.

Alcohol soluble extractive value = (Weight of residue \times Volume made) / (Weight of sample \times Volume taken) \times 100

6.3.5 Water soluble extractive⁵⁵:

5gms of the sample (coarse powder) was taken in a closed conical flask with 100ml of water. Conical flask was shaken frequently for 6 hours and kept undisturbed 18 hours. Then, it was filtered by using filter paper. 25 ml of filtrate was taken in the china dish and allowed the contents to evaporate. Percentage was calculated after weighing the residue.

Water soluble extractive value = (Weight of residue \times Volume made) / (Weight of sample \times Volume taken) \times 100

6.4 Qualitative analysis of Calotropis procera⁵⁶:

6.4.1 Test for 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. Appearance of an orange, pink or red to purple colour indicates the presence of flavonoids.

6.4.2 Test for alkaloids:

6.4.2.1 Mayer's test:

One ml of aqueous 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 coloured precipitates or turbidity indicate the presence of alkaloids.

6.4.2.2 Dragendroff's test: Extract was 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.

6.4.3 Test for Tannin:

A small quantity of the extract was boiled with water and filtered. Two drops of ferric chloride were added to the filtrate. Formation of blackish green precipitates confirmed presence of tannins.

6.4.4 Test for phenolic compound:Two to three drops of 1% ferric chloride (FeCl₃) solution were added in to 2 ml of 1 % extract. Deep violet colour was not produced which should be produced in the presence of phenolic compounds.

6.4.5 Test for coumarin compounds:

Coumarins form a yellow colour with 1% KOH in absolute ethanol. 1 ml of extract in test tubes was treated with 3-4 drops of 1% KOH in absolute ethanol.

6.4.6 Test for Reducing Sugar (Fehling's test):

To a test tube 1 ml each 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.

6.4.7 Test for Quinones:

To the test sample, sodium hydroxide is added. Formation of blue, green, or red colour indicates the presence of quinones.

6.4.8 Test for Proteins (Xanthoproteic Test):

The extracts were treated with few drops of conc. nitric acid. Formation of yellow colour indicates the presence of proteins.

6.4. 9 Test for fixed oil and fats:

A drop of concentrated extract was pressed in between two filter papers and kept undisturbed. Oil stain on the paper indicated the presence of oils and fats.

RESULT AND DISCUSSION

7.1 Analysis of raw drug:

7.1.1 Organoleptic study:

S. no.	Parameters	Observation		
1	Colour	Green		
2	Odour	Characteristic		
3	Taste	Characterstic		
4	Texture	Soft and cotny		

Table: 11 Organoleptic study of Arka

7.1.2 Physicochemical studies of Arka:

S.	Parameters	Batch1	Batch2	Batch3	Batch4	Batch5	Batch6	Average	Limits
no									in
									API
1	Foreign	0.1	0.3	0.1	0.2	0.1	0.1	0.15	NMT
	matter %								2%
2	Total ash	12.7	12.5	12.7	12.8	12.7	12.5	12.65	NMT
	%								21%
3	Acid in	3.0	3.0	2.9	2.9	3.0	3.0	2.9	NMT
	soluble								NMT
	ash%								5%
4	Water	27	27	26	28	27	26	26.8	NLT
	soluble								24%
	extractive								
	value%								
5	Alcohol	7	7	6	6	8	7	6.8	NLT
	soluble								5%
	extractive								
	value%								

Table: 12 Result of physicochemical study

7.1.3 Phytochemical screening of Arka:

S. No.	Components	Chemical tests	Observation	Results
1	Phenolic compound	_,	Brown	_
2	Tannins	Ferric chloride test	Greenish black colour	
				+
3	Flavonoids	Shinoda test	Yellow colour	+
4	Coumarins	КОН	Yellow colour	+
5	Steroidal glycosides	L. Burchard's test	Reddish brown ring	+
6		Mayer's test	Red Precipitates	
	Alkaloids			+
		Dragendroff's test	Red precipitates	
7	Protein	Xanthoprotein test	Yellow colour	+
8	Quinones	Sodium hydroxide test	Green colour	
				+
9	Anthraquinone glycoside	Borntrager test	Greenish brown	_
10	Saponins	Foam test	Formation of foam	_
11	Reducing sugars	Fehling's test	Brick red colour	_
12	Fixed oil and Fats	Spot test	Oil stain on paper	_

Table: 13 Screening of phytochemicals

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