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PHYSICO- CHEMICAL AND PHYTOCHEMICAL ANALYSIS OF DIFFERENT PARTS OF *CALOTROPIS PROCERA* COLLECTED FROM INDO-GANGETIC PLAIN (PATNA REGION)

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ABSTRACT

Calotropis procera Linn. (Family: Asclepiadaceae) is primarily harvested because of its distinctive medicinal properties. In the present research work, samples were collected from Indo- Gangetic plain in Patna region for analysis of Chemical constituents. Different parts (Leaves, Stems and Flowers) of the *Calotropis procera* were subjected to screen secondary metabolites such as Alkaloids, Steroid, Terpenoids, Resin, Glycosides. Physico- chemical studies thus indicate more potentiality of *Calotropis procera*. The Phytochemical are biologically active and structurally unique compounds which may be useful for generation of new medicines. The result indicated that secondary metabolites can be explored for formulation and development of newer and safer drugs.

KEYWORDS: Physico- Chemial, Phytochemicals, Traditional Medicine System.

INTRODUCTION

There are many plants which are being used as raw material for medicines. Medicinal plants are used from the ancient time as the major sources of drugs. Herbal medicines can be beneficial and no harmful side effects like conventional drugs. However, since a single plant may contain many substances, the effects of taking a plant as medicine can be complex. Various parts of the medicinal plants are reservoir of the phytochemicals which are medicinally highly potential. *Calotropis procera* Linn. is a herbal plant which is used in several traditional medicines to treat a variety of diseases. It is commonly referred to as ark, swallow-wart or milkweed having well known pharmaceutical and therapeutic applications. In ancient Ayurvedic medicine the plant *Calotropis procera* Linn. was known as "Rakta

arka". The extracts from different parts of the plant have significant therapeutic values. Chemical constituents of plants such as Alkoloids, Steroid, Terpenoids, Resin, Glycosides are reported in Ayurvedic literature. These chemicals are reported to be Analgesic, Resilient, Anti- inflammatory, Schizontocidal Activity (P. Sharma. *et.al* 1999), Emetic, Expectorant, Stomachic, Digestive, Laxative and Depurative. Further, these phytoconstituents are also reported potentially active for the treatment of several diseases such as Skin disease, Jaundice (Jan, *et.al.* 2009), Leucoderma, Eczema, Ulcer, Piles, Dysentery(Khan, *et.al* 2009), Dropsy, ring worm (Kuta, F.A.2008) and Removing Thorn from body (Rai, *et.al*. (2000). *Calotrois*root bark is very largely used as a treatment for elephantiasis and leprosy. The latex is as potent as standard anti-inflammatory drug Phenylbutazone (PBZ) in inhibiting inflammatory response induced by various inflammagens in acute and chronic models of inflammation. The Fresh leaves are used in treatment of Rheumatoid, Arthritis and Healing of wounds (Patil, *et.al*. 2009). The pungent latex extracted from the leaf and flowers of *C. procera* is processed and used in the commercial preparation of eye tonic (Vohra, 2004). (Henrich *et. al*., 2004), (Gurib-Fakim,2005), (Bruneton, 1999).

MATERIALS AND METHODS

Different parts (Leaves, Stems and Flowers) of *Calotropis procera were* collected from Indo-Gangetic Plain in Patna region. The fresh, healthy and diseases free leaves, stems and flowers of *Calotropis procera* were plugged by wearing gloves. The plant material was identified as per standard taxonomical norms at the field using standard keys and descriptions.

Physico- chemical analysis of *Calotropis procera:* Physico chemical analysis of *Calotropis procera* was carried out using standard AOAC method to analyze moisture, total ash, sulphate ash, acid insoluble ash, alcohol soluble extractive, water soluble extractive, loss on drying and sugar (Official methods of analysis, 1990). These tests were performed using weight difference method.

Preparation of extract: The samples were washed with distilled water to remove dust after that 70% Ethanol were used for surface sterilization of leaves. After that the samples were dried under shade at room temperature for 7 days. Leaves samples were ground using a grinder machine (Jaipan, Supper Deluxe, and India). Finally, the dry leaves samples were pulverized into powdered form. The fine powder is kept in air tight Amber bottle for further use.

Process of extraction: Maceration process is used for the extraction of samples. Petroleum ether, Methanol and ethyl acetate solvents were used in this technique. The sample (Leaves, stems and flowers) powder was weighed 500 gm and kept in a container in contact with 1000 ml petroleum ether for seven days, with vigorous shaking at regular interval. Material was filtered a first with muslin cloth and then with filter paper. Filtrate was collected and dried in water bath till no further reduction in mass of extract was observed. Dried extract was weighed and packed in air tight container and the marc was air dried then kept in a container in contact with 500 ml methanol for seven days, with vigorous shaking at regular interval. Material was filtered a first with muslin cloth and then with filter paper. Filtrate was collected and dried in water bath till no further reduction in mass of extract was observed. Further, the marc was air dried then kept in a container in contact with 250 ml ethyl acetate for seven days, with vigorous shaking at regular interval. Material was filtered a first with muslin cloth and then with filter paper. (Anonymous. The Indian Pharmacopoeia. Govt. of Indian publication, New Delhi, 1966, 947-950). Rotatory evaporator was used for drying plant extract.

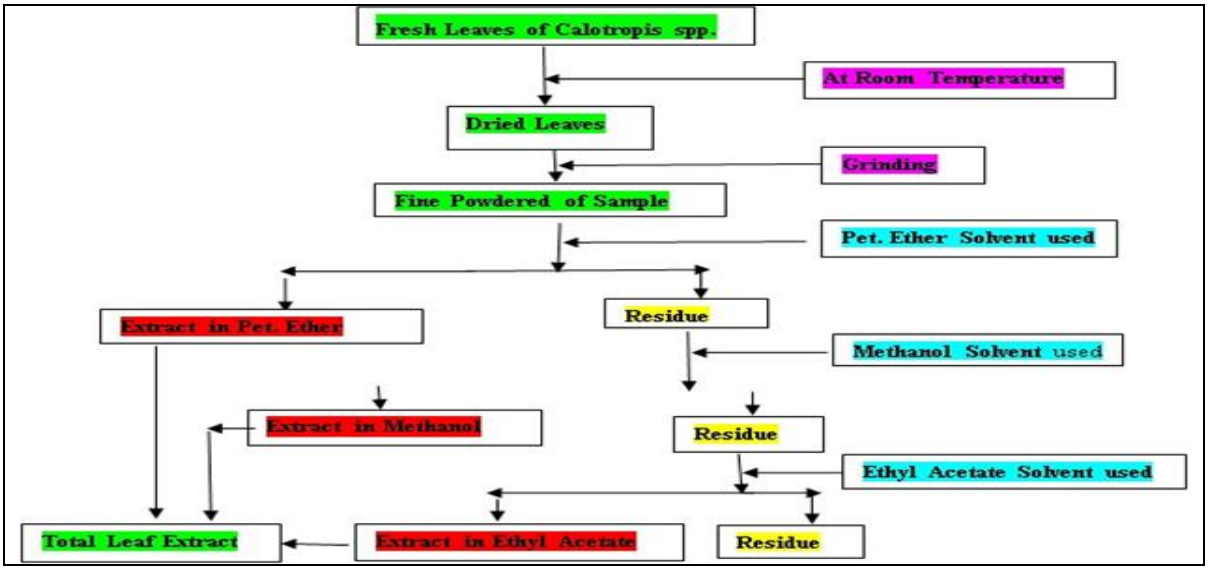

Figure no. 1: Flow Chart of Maceration Process.

Techniques Employed for Phytochemical Screening: Phytochemical Screening was carried out using standard methods to detect the bioactive compounds (Debela, 2002).

Mayer's test: About 0.5-1 ml of sample was taken in a tube. Few drops of Mayer's reagent were added. It is shaken and allowed to stand for some time. Appearance of cream colour ppt. indicates that alkaloids were present in the sample.

Hager's test: About 0.5-1 ml of sample was taken in a tube. Few drops (1-2) of Hager's reagent (saturated solution of picric acid) were added. Appearances of yellow colour ppt. after some time mark the presence of alkaloids in the sample.

Legal test: Sample was treated with small amount of pyridine in a test tube. Few drops of alkaline sodium nitroprusside solution were added. If blood red colour appears, then alkaloid was present in the sample.

Sodium nitroprusside test: About 0.5 – 1 ml of sample was taken in a test tube. A pinch of sodium nitroprusside powder and 2-3 drops of sodium hydroxide solution (10 percent) were added. Test tube is shaken and allowed to stand for 2-3 minutes. Appearance of red colour indicates presence of glycosides in the samples.

Ferric chloride test: Few drops of ferric chloride were added to 0.5 ml of test solution in a test tube. Appearance of blue- green colour confirms the presence of tannins and phenols in the samples.

Vanillin Hydrochloride Test: If test solution (0.5 – 1 ml) on treatment with few drops of vanillin hydrochloride reagent gives purplish red colour, then tannins and phenols are present in the sample.

Shinoda test or Magnesium hydrochloride reduction test

Test solution (0.5 0- 1 ml), few reagent of magnesium ribbon were added and concentration hydrochloric acid was added drop-wise. Pink scarlet, crimson and red of occasionally green to blue colour appears after few minutes, if flavonoid is present in the sample.

Alkaline reagent test: To the test solution (0.5 – 1 ml), few drops of sodium hydroxide solution (10 percent) were added. Formation of an intense yellow colour, which turns colourless on addition of few drops of dilute acid, indicates presence of flavonoids.

Salkowski test

About 0.5 – 1 ml of test solution was treated with chloroform in a test tube. Few drops of concentration sulfuric acid were added, shaken well and then wait for some time. Appearance of red colour at the lower layer indicates the presence of steroids and formation of yellow lower layer indicates the presence of the triterpenoids.

Libermann – Buchard test: Sample (0.5 – 1 ml) was treated with few drops of acetic anhydride in a test tube. Boil and cool, concentration sulphuric acid was added from the sides of the test tube, shows a brows ring at the junction of two layers and the upper layer turns green who shows the presence of steroids and formation of deep red colour indicates the presence of triterpenoids.

RESULT AND DISCUSSION

Herbal medicines are very popular in traditional medicine system. The Phytochemicals may be related with its ethno-medicinal use in the treatment of various diseases. Phytochemicals, also known as Secondary metabolites, natural products or plant constituents are responsible for medicinal properties of plants to which they belong. This present study tends to investigate the Phytochemical contents of the different solvent extract (Petroleum ether, Methanol and Ethyl acetate) of different parts of (Leaves, Stems and Flowers) *Calotropis procera*. The result of screening indicates that all 6 secondary metabolites are present in ethyl acetate extract of leaves, stems and flowers.

Table. 1: Phytochemical Screening of *Calotropis procera.*

Sl. No.	Secondary Metabolites					
	Alkaloids	**Flavanoids**	**Glycoside**	**Tannin and Phenolic compound**	**Triterpenoids and Steroids**	**Saponin**
Pet ether extract of Leaves	-	+	+	+	+	-
Pet ether extract of Stems	-	+	+	+	+	-
Pet ether extract of Flowers	-	+	+	+	_	-
Methanolic extract of Leaves	-	+	-	+	+	+
Methanolic extract of Stems	-	+	+	+	+	+
Methanolic extract of Flowers	+	+	+	-	+	+
Ethyl Acetate extract of Leaves	+	+	+	+	+	+
Ethyl Acetate extract of Stems	+	+	+	_	+	+
Ethyl Acetate extract of Flowers	+	_	+	+	+	+

(+) indicates presence **(-)** indicate absence.

Whole plant of *Calotropis procera* is analysed for Physico Chemical test. The result of Physico chemical analysis was revealed that the moisture contents in *Calotropis procera* was recorded was 88.5. Similarly, total ash was 9.1, water soluble ash is 1.8, sulphate ash 2.5, acid soluble as 1.6, water soluble extractive is 6.4, alcohol soluble extractive is 8.5, sugar is

2.66 and loss on drying is 9. This indicates that the Physico- chemical studies thus indicate more potentiality of *Calotropis procera* containing higher amount of extractive values.

Table no. 2: Physico Chemical Analysis Result of *Calotropis procera.*

Parameters (*Calotropis procera*)	Value (%W/w)
Moisture	88.5
Total ash	9.1
Water soluble ash	1.8
Sulphate ash	2.5
Acid insoluble ash	1.6
Water soluble extractive	6.4
Alcohol soluble extractive	8.5
Loss on dryings	9
Sugar	2.66

Secondary metabolites are responsible for medicinal activity of plants. *Calotropis procera* is high potential medicinal plant which can be explored further for its various uses in pharmaceuticals, nutraceuticals, fibre, health, environment and many other areas. The results suggest that the Phytochemical properties for curing various ailments and leads to the isolation of new and novel compounds. This type of study provides the health application at affordable cost.

CONCLUSION

Calotropis procera is one such plant which is believed to be loaded with highly useful secondary metabolites which can be isolated, identified, purified and characterised and used in formulation and development of newer herbal drugs which are cheaper, without side effect, assessable and effective for all people especially the poor one living in remote rural and tribal area. This research work is a sincere attempt to explore the potentiality of secondary metabolites present in the *Calotropis procera* leaf for their use in formulation of new, safe and herbal drug.

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