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Review Article

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A review on comparative studies of phytochemical evaluation and pharmacological activities of Calotropis species: Calotropis procera and Calotropis gigantea

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ABSTRACT

The genus Calotropis possesses two species Calotropis procera and Calotropis gigantea. Both species, stand out as important therapeutic plants because of the abundance of phytochemicals they contain, such as steroids and alkaloids, saponins glycosides, flavonoids, triterpenes, and anthraquinones. The various components found in different parts of both species contribute to their efficacy in treating a broad spectrum of health concerns, such as helminthic infections, diabetes, diarrhoea, malaria, inflammation, sterility, spasmodic disorders, cancer risks, and a variety of skin illnesses. This review critically examines and compares the phytochemical profiles and pharmacological activities of these two species, providing insights into their specific medicinal applications and efficacy. By consolidating current research, this review aims to guide future pharmacological studies and the development of herbal formulations based on Calotropis species.

Keywords: Phytochemicals, Flavonoids, Alkaloids, Pharmacological, Antiulcer, Antifungal.

INTRODUCTION

There are many things that plants provide to Man, including shelter, clothing, food, flavours, fragrances, and not to mention medicine. The foundation of complex traditional medical systems has been plants [1]. Around 80% of the worldwide population relies on traditional medicine as their primary healthcare system, making it one of the most important health systems. It is possible to locate and extract naturally occurring substances from many plant components, including bark, leaves, flowers, roots, fruits, and seeds [2]. Numerous studies have examined the various plant components and discovered that they include phytosterols, alkaloids, cardinolides, triterpenoid, and saponins in addition to pentacyclic triterpenes [3]. Plants naturally contain bioactive substances called phytochemicals, which serve as a defence mechanism against illness [4]. Calotropis procera, a perennial shrub of the Apocynaceae family, thrives in dry environments and is typically found in arid and semi-arid regions [5]. Calotropis is a term derived from the Greek language, which means "beautiful." This term refers to the plant's flowers. On the other hand, "procera" is a Latin word that refers to the epidermal wax present on the plant's leaves and stems [6]. Calotropis gigantea is a perennial potential herb found predominantly in barren and wastelands throughout India. Calotropis gigantea, sometimes referred to as swallowwort or milkweed, is a widespread wasteland plant found in India. Elephantiasis, indigestion, fever, rheumatism, diarrhoea, and asthma are among the frequent illnesses for which Calotropis gigantea has historically been used as a remedy. It can also be used with other plants for medical purposes [7]. In addition to treating colds, fever, diarrhea, rheumatism, indigestion, eczema, and jaundice, Calotropis species treat asthma, bronchitis, asthmatic pain, leprosy, ulcers, piles, spleens, tumors, livers, abdomens, and dyspepsia. The stem and roots of this plant were used to treat various diseases, including skin diseases, intestinal worms, leprosy, and leukoderma; the roots were used to treat elephantiasis, rheumatism, and asthma, cough, and diarrhea. Paralysed areas can be treated with oil massage; latex and leaves are used to relieve edoema and joint discomfort; and Calotropis juice can be used to purify [8]. There is still a deficiency of thorough reviews of the phytochemistry and distinct pharmacological activities of Calotropis procera and Calotropis gigantea, despite the fact that other authors have examined these aspects of the plants. Thus, an attempt was made to provide thorough and up-to-date information on its qualities in this study.

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PHYTOCHEMISTRY

Phytochemistry of Calotropis Procera

Numerous studies have reported finding different metabolites in different portions of the plant, including flavonoids, tannins, terpenoids, saponins, alkaloids, steroids, and cardiac glycosides [9].

The primary phytochemical groups present in Calotropis procera leaf extracts include 7.4% of linoleic acids, fatty acid ethyl esters (21.4%), 8.1% of amino acids and palmitic acid esters (10.2%) [10]. It has been possible to separate terpenoids (ursane, olenane type, pentacyclic triterpenes, etc.) from latex, root bark, and flowers. It has recently been found that the root bark of Calotropis procera contains oxypregnane glycosides [11,12]. HPLC was used to analyse the leaves and bark in order to determine the following total content (IC50 equivalent mg/g dry weight) of flavonoid (18.33-92.92 catechin), sinapic acid (17.3 \pm 2.11 to 9586.44 \pm 0.78 mg/kg), phenolic (20.41-100.18 gallic acids), vanillic acid (9.43 \pm 0.21 to 5051.7 \pm 18.47 mg/kg), and protocatechuic acid (2.46 \pm 0.40 to 139.05 \pm 1.37 mg/kg) [13]. Querigenin-3, sterol, calactin, calotoxin, and polysaccharides with D-arabinose, glucose, glucosamine, L-rhamnose, lupeol, syriogenin, cardenolide, gigantin, giganteol, isogiganteol, epimoretenol, alactuceryl isovalerate and acetate, are all present in the flower. The primary ingredients found in latex include calotoxin, calactin, uscharin, proceroside, trypson, calotropagenin, calotropin, syriogenin, tetraxasterol, uscharin, uzarigenin, voruscharin, β -amyrin, epimoretenol lupeol, labenzyme, and traces of ortho hydroxy phenol [14]

Phytochemistry of Calotropis gigantea

Previous studies on phytochemical profiles of various parts of *Calotropis gigantea* indicate that the plant is rich in resin, sterols, alkaloids, glycosides, carbohydrates, tannins, flavonoids, saponins, and peroxide. Other major phytoconstituents include beta-amyrin, fatty acids, acetates, and Usharin, a combination of tetracyclic triterpene compounds, giganteol, and sterols. The phytochemicals identified in *Calotropis procera* and *Calotropis gigantea* are summarized in Table 1.

Table 1. Recent reports on the Phytochemistry of Calotropis procera and Calotropis gigantea

Entry	Plant parts	Phyto Chemicals		
		Calotropis procera(a)	Calotropis gigantea(b)	
1	Flowers [15-20a,21b]	α -Amyrin ($C_{30}H_{50}O$) β -Amyrin ($C_{30}H_{50}O$) Stigmasterol ($C_{29}H_{48}O$) Cyclosadol ($C_{31}H_{52}O$) 7'-Methoxy-3' -O-demethyl-tanegool-9- O- β D-glucopyranoside Rosmarinic acid ($C_{18}H_{16}O_8$) Methyl rosmarinate ($C_{19}H_{18}O_8$) Methyl ferulate D-arabinose Glucose Glucose Glucosamine Cyclosadol ($C_{31}H_{52}O$) 3-proteinase Proceroside Proceragenin 3-Epimoretenol	α- calotropeol β-calotropeol Amyrin Glycosides Mudarine Asclepin Akundarin	
2	Leaves [21b,17a,20a,22-25a]	Lupeol-3-O-acetate $(C_{32}H_{52}O_2)$ Rutin Quercetin $(C_{15}H_{10}O_7)$ Azaleatin $(C_{16}H_{12}O_7)$ Isorhamnetin Kaempferol Tridecyl ester Methyl caffeate $(C_{10}H_{10}O_4)$ Caffeic acid $(C_9H_8O_4)$ 9-Octadecenoic acid (Z) - $(C_{18}H_{34}O)$ Tetratetracontane $(C_{44}H_{90})$ $6,10,14$ -Trimethyl-pentadecanone-2 $(C_{18}H_{36}O)$ α -rhamnose	Sapogenins, Calotropin Uscharin, Calotoxin, Alkaloids Mudarine	
3	Roots [26b,3,27,28a]	α -Amyrinacetate ($C_{32}H_{52}O_2$) Phytyl iso-octyl ether Procesterol Methyl myrisate ($C_{15}H_{30}O_2$)	Cardiac glycosides Calotropnaphthalene Calotropisesquiterpenol Calotropisesterterpenol Calotropbenzofuranone Calotroposides A-G (oxy pregnane- oligoglycosides)	
4	Latex [29-32a,33b]	Lupeol $(C_{30}H_{50}O)$ Stigmasterol $(C_{29}H_{48}O)$ Eucommin A (+)-Pinoresinol 4-O-β-D-glucopyranoside (+)-Pinoresinol 4-O-[6" -O protocatechuoyl]-β-D-glucopyranoside 12β-Hydroxycoroglaucigenin 15β-Hydroxy calactin 15β-Hydroxy uscharin Decane $(C_{10}H_{22})$	Calotoxin Calactin Calotropin Uscharin α-Calotropeol β-Calotropeol β-amyrin Calcium oxalate	

		1,3,5-Tri-isopropylbenzene (C ₁₅ H ₂₄)	
		2,3,4-Trimethylhexane (C ₉ H ₂₀)	
5	Stem bark [34a,35b]	Luteolin ($C_{15}H_{10}O_6$)	α- calotropeol
			β- calotropeol
			β- amyrin
			Giganteol
6	Root bark [36a,37b]	Stigmasterol (C ₂₉ H ₄₈ O)	β-amyrin,
		Calotropterpenyl ester	Giganteol
			Isogiganteol
			Cardenolides
8	Stem [38a]	5-Hydroxy-3,7-dimethoxyflavone-4'-O-β-glucopyranoside	
		$(C_{23}H_{24}O_{11})$	
		Desglucouzarin (C ₂₉ H ₄₄ O ₉)	
		Uzarigenone (C ₂₃ H ₃₂ O ₄)	
		Caroglaucigenin	Palmitic acid,
		Frugoside	Oleic acid
9	Seed [39a,40b]	Carotoxigenine	Linoleic acid
		Calotropin	Linolenic acid
			Stigmasterol
			Phytosterol
			Melissyl alcohol

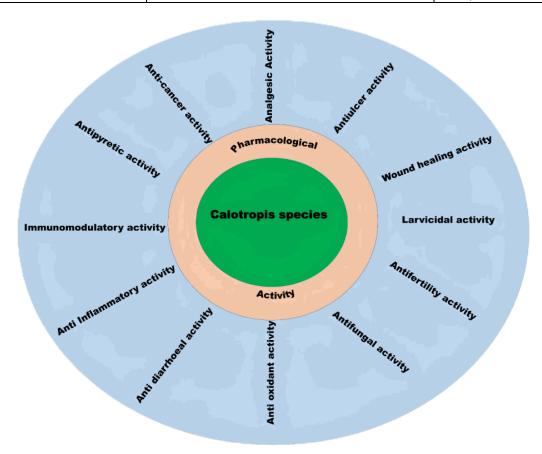


Figure 1: Pharmacological Activity of Species Based on Calotropis

PHARMACOLOGICAL ACTIVITY

Pharmacological Activity of Calotropis Procera and Calotropis gigantea

A variety of diseases have been treated with the therapeutic properties of *Calotropis procera* and gigantea since ancient times. Here some of the very vital pharmacological activities are being discussed in detail.

Analgesic Activity

It has been documented that *Calotropis procera*'s dry latex (DL) possesses analgesic properties [20]. When in contrast to an oral dosage of aspirin (100), a dose of 415mg/kg of DL was more efficient in

preventing writhing caused by acetic acid. There is analgesic action in the alcoholic extract of *Calotropis gigantea* flowers. The activity was carried out using the writhing test caused by acetic acid and the hot plate technique, and an oral dosage of this extract produced a noteworthy outcome. In albino rats, the central nervous system (CNS) action of an alcoholic peeling extract *Calotropis gigantea* roots (at oral dosages of 250 and 500 mg/kg body weight) was studied. Eddy's hot plate method and the writhings brought on by acetic acid both showed notable analgesic benefits [41].

Wound Healing Activity

Aqueous extract of latex $^{[42,43]}$, ethanolic extracts of bark $^{[44]}$, and aqueous extracts of Leaves $^{[45]}$ of *Calotropis procera* have been

reported for wound healing activity. Sterile preparation of Latex was applied twice a day for seven days on the wound of guinea pig. Increased collagen fibres, DNA, and protein were seen in the animal's treated region. The ability of Calotropis gigantea root bark extract to treat wounds in Wistar albino rats was studied. Rats were given the extract topically in order to simulate excision wound healing in the form of an ointment; Oral dosages of 100, 200, and 400 mg/kg of the extract were used for incision wound healing models. The findings suggest that rats' wound healing was sped up by extract therapy [7]. Assessed the wound-healing properties of using both excision and incision wound models, Calotropis gigantea crude latex was applied to albino rats. When compared to controls, who had a wound area of 76.22%, treated animals had an 83.42% decline in wound zone, indicating that Calotropis gigantea latex, at a dosage of 200 mg/kg/day, had considerable activity that heals wounds. In comparison to controls, it has been demonstrated that extract-treated wounds epithelize faster [46].

Antifertility activity

There have been reports of the antifertility action of *Calotropis procera*'s ethanolic extracts of its roots and leaves [47,48]. The antifertility and hormonal effects of *Calotropis procera* roots have been studied in albino rats using the ethanolic extract of those roots. At a dosage of 250 mg/kg, significant antiimplantation (inhibition 100%) and uterotropic action was seen. The antifertility properties of *Calotropis gigantea* have also been linked to the presence of certain phytochemical substances, including tannins, psoralen, gigantein, stigmasterol, and sapononis [49].

Anti-diarrhoeal activity

An imbalance between the digestive tract's secretory and absorptive mechanisms combined with haste causes diarrhoea, which causes an excessive amount of fluid to be lost in the stools. Several sections of Calotropis procera, including the bark [50], latex [51], leaves, stems, roots, flowers, and buds [52], have demonstrated their anti-diarrheal properties. The anti-diarrheal properties of Calotropis procera's DL have been assessed. Similar to atropine and phenyl butazone, a single 500 mg/kg dosage of DL resulted in a notable reduction in the frequency of defecation. Electrolyte content in intestinal fluid and caused intestinal fluid buildup and protection against 80 percent of rats given castor oil experienced diarrhoea. Intestinal transit decreased (by 27-37%) in DL animals in contrast to animals that were not treated with castor oil. The anti-diarrheal properties of the hydroalcoholic (50:50) extract of Calotropis gigantea's aerial component were investigated in rats using a model of castor oilinduced diarrhoea. The extract demonstrated notable decreases in both the frequency of droppings and faecal production at intraperitoneal dosages of 200 and 400 mg/kg body weight. Significant inhibitions in intestinal content volume and weight were also demonstrated by the extract [53].

Antiulcer Activity

Several Calotropis procera components exhibit antiulcer activity, including the hydroalcoholic and chloroform extract of the stem bark. the leaves' ethanolic extract, and the root chloroform extract [54,55]. Due of its ability to reduce inflammation and prevent ulcers, stem bark extract from Calotropis procera was dissolved in hydroalcoholic and chloroform. For the research of anti-ulcer activity, albino rats with ulcers were employed, and for anti-inflammatory efficacy, the paw oedema model generated by carragennan was employed. When compared to conventional medications, the extract shown notable activity. In experimental Wistar albino rats, the activity of 100% alcohol extract of Calotropis gigantea flower was examined using pylorus ligation and indomethacin-induced ulcer models. When compared to the control, the ulcer index and ulcer intensity score in both models showed a substantial (P<0.01) suppression when the alcoholic extract of Calotropis gigantea was administered at dosage rates of 500 mg/kg and 1000 mg/kg. The extract demonstrated the greatest decrease in ulcer index in pylorus ligated ulcer animals at a dosing rate of 500 mg/kg [56].

Antifungal and anti-bacterial activity

There have been reports of antifungal activity in the aqueous [57], methanol, acetone, and ethanol extracts of the leaves [58,59], root bark [59,60], and the latex of *Calotropis procra* [60]. Using agar well diffusion and paper disc methods, the antimicrobial effect of ethanol, aqueous, and chloroform extracts of Calotropis procera's leaves and latex was investigated on five bacteria: S. pyogenes, E. coli, S. aureus, S. albus, and S. pneumoniae; three fungi: Aspergillus niger and flavus, and Microsporum boulardii; and one yeast, C. albicans [61]. The findings showed that ethanol proved to be most effective extractive solvent for the antibacterial qualities of Calotropis procera's leaves and latex, with chloroform and water coming in second and third, respectively. It has been discovered that Calotropis gigantea possesses antifungal activity against harmful plants fungus such as Fusarium mangiferae, a terrible hazard to mango farming [62]. It has been observed that the aqueous extract of Calotropis gigantea leaves had effective activity against Pseudomonas aeruginosa, E. coli, Klebsella pneumonia, Bacillus cereus, and S. aureus [63]. C. krusei, S. aureus, B. cereus, E. coli has all been shown to be substantially barred by the aqueous extract of Calotropis gigantea's latex [64]. It has been discovered that Calotropis gigantea possesses antifungal combat towards plant pathogenic fungus such as Fusarium mangiferae, that puts mango cultivation in grave peril [62].

Antipyretic Activity

Aqueous extract of dry latex [65], and ethanolic extract of flowers [66], of *Calotropis procera* have been reported as antipyretic activity. An extract of Gigantea roots in water and ethanol (50:50) has been reported to have antipyretic properties. An Albino Swiss rat and a rabbit, induced with yeast or TAB (Typhoid) vaccine, were used to determine anti-pyretic activity. It was found that extract was significant in reducing fever and normalizing body temperature at 200 and 400 mg/kg body weight dosages [67].

Anti-Inflammatory Activity

There have been reports of anti-inflammatory properties for dry latex [68], hydro-alcoholic and chloroform extracts of stem bark [55], ethyl acetate, C₆H₁₄, CH₂Cl₂, n-butanol, and water-based extracts of latex [69], leaves [70], flowers [66], and the ethanolic and ethanolic extracts of Calotropis procera's root bark [71]. When it came to carrageenan, the anti-inflammatory properties of DL's aqueous and methanolic extracts outperformed those of phenylbutazone (PBZ), whereas they were similar to those of chlorpheniramine and PBZ when it came to histamine and prostaglandin E2, respectively. Compared to other solvent extracts, the ethanolic extract of *Calotropis gigantea* leaves had a stronger anti-inflammatory effect. The anti-inflammatory properties of Calotropis gigantea ethanol extract were found to be efficient toward carrageenan-induced paw edoema in Wistar albino rats. Significant reduction in inflammation was seen with oral dosing of 400 mg/kg of Calotropis gigantea; this potency was observed to be exceeded that of 100 mg/kg of Ibuprofen [72].

Anti-cancer activity

Aqueous extracts of dry latex [73], Hexane, dichloromethane, ethyl acetate, acetone, and methanol extract of Stem [74,75], of *Calotropis procera*. have been reported as Anticancer activity. When the DL of *Calotropis procera* was examined in a transgenic hepatocellular cancer model in mice, it was discovered to provide full protection against the development of hepatocarcinogenesis. AML12 cells were determined to be alive, whereas Huh-7 and COS-1 cells showed severe cell death and a significant decrease in serum vascular endothelial growth factor levels. Cardiac glycosides isolated from *Calotropis gigantea* can inhibit tumor/cancer cell growth. Cardiac glycosides such as calactin, calotropin, asceplin, and cymarin can

affect the expression of p53 and Bcl-2 genes in breast cancer CF-7 in vitro. Tumor suppression activities on the mediated test sample more likely occurred through Bcl-2 regulation (disrupting antiapoptotic activity) than through p53 (facilitating apoptosis) [76].

Immunomodulatory activity

The ethanolic extract of *Calotropis procera* root bark can be used in complementary medicine to treat immunodeficiency disorders by modulating several immunological parameters [77]. Using immunological assays such as peritoneal macrophage count, humoral-mediated antibody titer, delayed-type hypersensitivity, and vascular permeability, and haematological profile, immunomodulatory activity was assessed in mice at three dosage levels. There is no doubt that *Calotropis gigantea*'s water-soluble latex and whole aqueous extract have immunomodulatory qualities [78].

Antioxidant Activity

There have been reports of antioxidant activity in methanolic and aqueous extracts of leaves [79], roots extracted both methanolically and aqueously [80], methanolic extracts of flowers and fruits [79-81], and bark extracts from *Calotropis procera* in ethanol [44]. The reducing power, nitric oxide scavenging, and DPPH radical scavenging processes of the hydroalcoholic leaf extract of *Calotropis gigantea* have demonstrated the antioxidant activity of the leaf extracts. The extract exhibited a maximal scavenging activity of 85% for DPPH radicals at 400 µg/mL concentration, 54% for nitric oxides at 100 µg/mL concentration, and an increase in reducing power as the extract concentration rose. Furthermore, when the antioxidant potency of *Calotropis gigantea*'s leaf and flower extracts was examined, it was found that the methanolic extract had a notable 64% free radical scavenging activity, compared to only 30% and 37% for the acetone and chloroform extracts, respectively [82].

Larvicidal Activity

When Testing of *Calotropis procera* against mosquito larvae was done of the Anopheles labranchiae species, it demonstrated strong larvicidal action, with an LC₅₀ (24 hours) ranging from 28 to 325 ppm. Studies were conducted on the harmful effects of crude extracts of *Calotropis procera* (from both leaves and flowers) toward Heterotermes indicola and Coptoter mesheimi, two species of termites ^[83]. Previous research on *Calotropis gigantea*'s ethanolic extract has demonstrated larvicidal effects on Ae. aegypti larvae, with an LD₅₀ value of 351.43 ppm. For the larvicidal action, the LC₅₀ value of all sections showed no discernible difference between *Calotropis gigantea* and *Calotropis procera*, indicating that both species had the same impact on Ae. aegypti larvae ^[84]. The primary substances that may be in charge of the larvicidal properties displayed by the Calotropis species are calcotropin and calotoxin.

COMPARATIVE STUDY

Many plant components, including the leaves, stem, flowers, and root bark of Calotropis gigantea and Calotropis procera, are used as medicines to cure common illnesses. However, their comparative studies have been less explored. Comparative phytochemistry and pharmacological research of Calotropis gigantea and Calotropis procera were presented by R. A. Sharma et al [85]. Numerous active chemicals were present in the phytochemical makeup of these plants. All components of Calotropis gigantea and Calotropis procera, including the root, stem, leaf, and flower, are recommended for use in the treatment of numerous ailments by the Ayurvedic medical system. Some comparative (qualitative or quantitative) studies of phytochemical analysis of plants parts have been investigated [86-88]. Four different solvents were used in the phytochemical examination of root extracts by Pratibha and colleagues: ethanol, water, chloroform, and ethyl acetate [86]. Their research showed that the root extracts of both species included phenolic chemicals, alkaloids, and flavonoids. However, the amounts of phenolic and flavonoid

components in the Calotropis gigantea root extract in ethanol were noticeably greater.

An examination of the physicochemical and phytochemical characteristics of both Calotropis species was carried out by Srivastava and associates [87]. Calotropis procera had more extractive qualities than Calotropis gigantea, according to the findings of the physicochemical investigations. HPTLC was used to examine the phytochemical composition and, in particular, to determine the amount of β-sitosterol present. It was discovered that both species possessed β-sitosterol; however, there was a little difference between the roots of Calotropis gigantea (2.79%) and Calotropis procera (1.07%). The H. V. Patel group conducted a relative study of the antioxidant interest of extracts from the leaves, roots, and latex of Calotropis gigantea and Calotropis procera [19]. After evaluating the larvicidal and antioxidant properties using radical (DPPH) scavenging against Ae. Aegypti larvae, it was determined that the methanolic leaf extract of Calotropis procera exhibited high larvicidal and antioxidant properties in comparison to Calotropis gigantea.

CONCLUSION

Overall, Calotropis procera and Calotropis gigantea provide many research opportunities thanks to their rich chemistry, allowing their isolated compounds to be developed for a wide range of pharmacological applications. A variety of methods are needed to explore the medicinal properties of this plant's phytochemicals. To fully understand this plant's pharmacological and phytochemical properties, more investigation is required. Pharmacological and phytochemical constituents found in Calotropis procerea and Calotropis gigantea make them potential sources for new drugs in pharmaceuticals. It also identifies critical gaps in knowledge and explores future research opportunities for both Calotropis procera and Calotropis gigantea in light of the information presented in this review.

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Conflict of interest

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