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# Comparative Evaluation of Phytochemicals, TPC and TFC in Aqueous and Ethanolic Extracts of *Calotropis gigantea* L. Leaves and Roots

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**Abstract:** ***Calotropis gigantea L. is a traditional medicinal plant that has been reported to have analgesic, antipyretic activity, antimicrobial, anti-diarrheal and cytotoxic, properties and has been considered to cure cardiovascular diseases, skin diseases, use as anticancer, antifertility and antidote for snakebites in traditional system of medicine. The present work was intended to comparatively evaluate the phytochemicals, total polyphenolic content and total flavonoidal content of leaves and root extracts of C. gigantea L.The aqueous and ethanolic extracts of this plant was reported to have alkaloids, tannins, saponins, flavonoids and glycosides. Terpenoids were present in ethanolic leaf extract only. The extraction yield were 43.72% and 19.92% for aqueous and ethanolic leaf extracts respectively, while that of roots is 16.12% and 8.1% aqueous and ethanolic extracts respectively. Leaves of rich in TPC especially the aqueous extracts with 23.1% of TPC while the roots extracts were higher in TFC values especially the aqueous root extracts with 17% which is yet higher than aqueous leaf extracts which was reported as 10.4% in present work. It is clear from present study, that the extraction procedures, and types of solvents used for extraction not only has clear impact on yield of crud extracts but also on type of phytoconstituents.***

**Keywords:** *Calotropis gigantea L.* , TPC and TFC

## 1. Introduction

*Calotropis gigantea* L. is a traditional medicinal plant it belongs to the family Apocyanaceae of *Asclepiadaceous* habitat of Asian countries that includes India, Indonesia, Malaysia, Thailand, Srilanka and china. The plants is branched, tall, erect, large, produce white or violet flower in bunches, and enduring with latex throughout. This plant has been reported to have analgesic, antipyretic activity, antimicrobial, anti-diarrheal and cytotoxic, properties (Manivannan and Shopna 2017). The plant has also been used in cough, cold, asthma, nausea vomiting, indigestion, leprosy, rheumatism, eczema etc. (Agharkar, 1991; Gaurav, *et al*., 2010; Hemalatha, *et al*., 2011). *Calotropis gigantea* L. has been considered to cure cardiovascular diseases, skin diseases, use as anticancer, antifertility and antidote for snakebites in traditional system of medicine (Suresh and Karki, 2012; Park, *et al*., 2002; Upendra, *et al*., 1992; Kitagawa, 1992).

The pharmacological properties of any plant are due to its phytochemical constituents especially the polyphenols. Structurally, polyphenols fall into different families including anthocyanins, coumarins, lignins, flavonoids, tannins, quinones, acids and phenols (Robards and Antolovich1997; Koffi *et al*., 2010). Thus the present study was intended to evaluate the phytochemicals, total polyphenolic content and total flavonoidal content of leaves and root extracts of *C. gigantea* L.on comparative basis.

## 2. Methodology

**Sampling and Extraction**

The leaves and roots of well identified *C. gigantea* L. were collected from the natural habitat around Indore City (M.P.) India. The samples were cleaned, dried and grounded into fine powders which were later subjected to defatting with petroleum ether followed by Soxhlet extraction with pure distilled water and ethanol separately. The extracts were concentrated by solvent evaporation to asses the yield of extraction and other phytochemical investigations

**Phytochemical Analysis**

A small portion of the dry extracts were subjected to the phytochemical test using Harbourne's (1983) methods to test for alkaloids, tannins, terpenoids, saponins, flavonoids and glycosides.

**Test for alkaloids:** About 0.2 g extract warmed with 2% $H_2SO_4$ for two minutes, filtered and few drops of Dragendorff's reagent added orange red precipitate indicates the presence of alkaloids.

**Test for glycosides:** Benedict's test was performed in which the filtrates were treated with Benedict's reagent and heated gently. Orange red precipitate indicates the presence of reducing sugars.

**Test for tannins:** Small quantity of extracts mixed with water, heated, filtered and ferric chloride added. A darkgreen solution indicates the presence of tannins.

**Test for saponins:** About 0.2g of the extracts shaken with 5ml of distilled water and then heated to boil frothing appearance of creamy mix of small bubbles.) shows the presence of saponins.

**Test for flavonoids:** Extract of about 0.2 g of the extracts shaken with 5ml of distilled water and then a few drops of

10% lead acetate solution is added. A yellow or dirty white precipitate shows the presence of flavonoids.

**Figure 1:** *C. gigantea L.* Plant, Leaves, Roots and their Aqueous & Ethanolic Extraction by Soxhletion

**Table 1:** Phytochemical Analysis of Leaves and Roots of *C. gigantea* Aqueous &Ethanolic Extracts

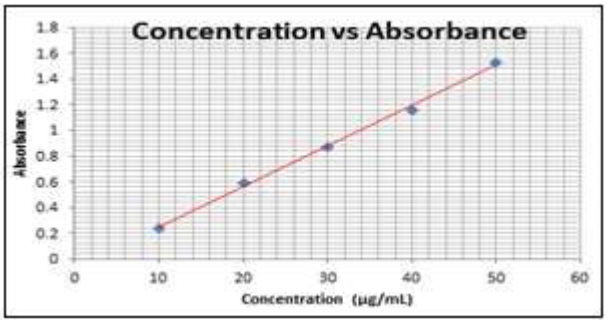
| S.N | Constituents | *C. gigantea* Leaf Extract | | *C. gigantea* Root Extract | |
|---|---|---|---|---|---|
| | | Aq Extract | Eth. Extract | Aq Extract | Eth. Extract |
| 1 | %Yield | 43.72% | 19.92% | 16.12% | 8.1% |
| 2 | Alkaloids | + | + | + | + |
| 3 | Tannins | + | + | + | + |
| 4 | Terpenoids | – | + | – | – |
| 5 | Saponins | + | + | + | + |
| 6 | Flavonoids | + | + | + | + |
| 7 | Glycosides | + | + | + | – |

**Table 2:** The percentage concentration TPC on the basis of absorbance at 765 nm in extracts at 100 µg/ml concentration.

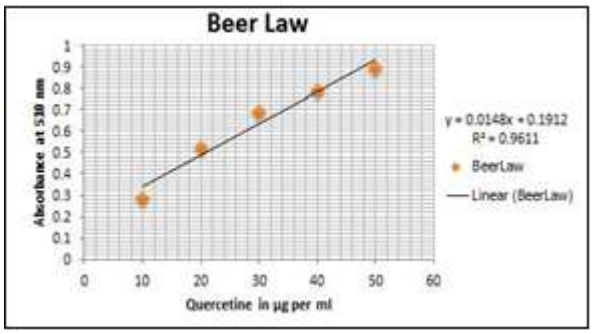
| S.N. | *C. gigantea* Extracts | Concentration (µg/ml) | Absorbance (λ) | Percentage TPC in w/W |
|---|---|---|---|---|
| 1 | Aq. Leaf extract | 100 µg/ml | 0.661 | 23.1% |
| 2 | Eth. Leaf extract | 100 µg /ml | 0.612 | 21.6% |
| 3 | Aq. Root extract | 100 µg /ml | 0.534 | 19.1% |
| 4 | Eth. Root extract | 100 µg /ml | 0.474 | 17.2% |

**Table 3:** percentage concentration of TFC in ***C. gigantea*** leaves and roots extracts at 510 nm compared to Quercetine standard plot

| S.N. | *C. gigantea* Extracts | Concentration (µg/ml) | Absorbance (λ) | Percentage TFC in w/W |
|---|---|---|---|---|
| 1 | Aq. Leaf extract | 100 µg/ml | 0.334 | 10.4% |
| 2 | Eth. Leaf extract | 100 µg /ml | 0.308 | 8.7% |
| 3 | Aq. Root extract | 100 µg /ml | 0.436 | 17% |
| 4 | Eth. Root extract | 100 µg /ml | 0.298 | 8.1% |




**Graph 1:** Gallic acid standard curve at 765 nm. The Graph is obtained from Excel 2010 linear regression function




**Graph 2:** Standard Plot of Quercetine as standard concentration vs absorbance at 510 nm. The Graph is obtained from Excel 2010 linear regression function

**Estimation of TPC**

The total phenolic content of the extracts was determined using to the Folin-Ciocalteu method with suitable modification. The extracts were suitably diluted with their respective solvents and oxidized with Folin-Ciocalteu reagent, and the reaction was neutralized with sodium carbonate. The absorbance of the resulting blue colour was measured at 765 nm after 60 min. The total phenolic content was calculated by comparing absorbance with gallic acid as standard curve (Graph 1)

**Estimation of TFC**

The total flavonoid content of crude extracts was determined spectrophotometrically by aluminum chloride method. For this, small amount of crude extracts were suitably diluted with distilled water up to 4 ml to which 0.3 mL of 5% $NaNO_2$ solution; 0.3 mL of 10% $AlCl_3$ solution was added after 5 min of incubation, and the mixture was allowed to stand for 5 min. Then, 2 ml of 1M NaOH solution were added and final volume of 10 ml with double-distilled water was made. The absorbance of the mixture was measure at 510 nm after 15 minutes of incubation. The total flavonoidal content was calculated by comparing the absorbance of the samples with standard curve of quercetine. (Graph 2)

## 3. Results & Discussion

The results of phytochemical analysis as depicted in table 1, the aqueous and ethanolic extracts of *C. gigantea L.* leaves and roots are quite close regarding the presence of the phytoconstituents. The difference among *C. gigantea L.* extracts is that terpenoids were not detected in aqueous leaf and root extracts and also absent in ethanolic root extracts. In addition, glycosides were also not detected in ethanolic root extracts. More or less similar constituents were reported in petroleum ether and methanolic extracts of leaves and ethanolic and aqueous extracts of latex (Sarkar*et al.*, 2013; Singh, *et al.*, 2014). From results a huge difference in percentage yield of extraction among aqueous and ethanolic leaf extract and also aqueous and ethanolic root extracts of *C. gigantea* were observed. The yield of extraction for aqueous extracts in both leaf and roots is higher compared to ethanolic extracts.

Otherwise leave gives higher yield of extraction compared to the root extracts. The Total phenolic content was estimated to be higher in both aqueous leaf and root extract when compared to their ethanolic extracts (Table 2). Inter

organally, leaves were found richer in TPC than roots Overall the TPC is present in enough amounts in all the extracts.

The extracts evaluated in present work are found less contented with total flavonoids compared to the TPC. Though the yield of extraction of root extracts were low but are found to be rich TFC (Table 3). Out of both plant parts both aqueous extracts are rich in TFC compared to their ethanolic extracts.

## 4. Conclusion

Both leaves and roots of *C. gigantea L.* are rich in various types of phytochemical constituents which are generally responsible for the therapeutic and pharmacological properties of any medicinal plant. It is clear from present study, that the extraction procedures, and types of solvents used for extraction not only has clear impact on yield of crud extracts but also on type of phytoconstituents. Thus the extraction and requirement of desired phytoconstituent could be managed by understanding the type solvent system needed for extraction as per the requirements.

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