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Research Article

**ESTIMATION OF TOTAL PHENOLIC AND FLAVANOID
CONTENT IN ALCOHOLIC ROOT EXTRACT OF
CLERODENDRON PANICULATUM BY
SPECTROPHOTOMETRIC METHOD**Leena. P. N^{1*}, Dr. N. A. Aleykutty², Dr. K. G. Prasanth³

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Abstract:

India has a great wealth of various naturally occurring plant drugs which have great potential pharmacological activities. The root of Clerodendron paniculatum Linn is used in south India as a remedy for various diseases. The study comprising taxonomy of the species, macro and microscopical characters, phytochemical and uv analysis of the root powder. Besides chromatographic details of root extract helps in the identification of the plant constituents also contribute towards establishing pharmacopoeial standards. Phenolics are compounds among the most widely occurring secondary metabolites in the plant kingdom, acting mainly as phytoalexins, attractants for pollinators, and contributors to plant pigmentation, antioxidants. Polyphenols in plant extracts react with specific redox reagents (Folin-Ciocalteu reagent) to form a blue complex that can be quantified by visible-light spectrophotometry. Flavonoids are potent antioxidants and have aroused considerable interest recently because of their potential beneficial effects on human health in fighting diseases. The capacity of flavonoids to act as antioxidants depends upon their molecular structure. The position of hydroxyl groups and other features in the chemical structure of flavonoids are important for their antioxidant and free radical scavenging activities. Quercetin, the most abundant dietary flavonol, and is detected by Aluminium chloride method

Key Words: *Clerodendron paniculatum* root, Aluminium chloride, Folin-Ciocalteu method.

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INTRODUCTION:

Clerodendron paniculatum Linn, (Family :Verbenaceae) is a species, found in India, It is reported as folk remedy for tumours , leprosy , fever , infection , inflammation .The roots have been reported to possess laxative ,diuretic ,analgesic ,anti inflammatory, anti tumour and antibacterial activities¹. In the present study the root portions of *Clerodendron paniculatum* was extracted with ethanol and ethyl acetate as solvent by cold extraction.method. These extracts were subjected to preliminary phytochemical screening for the detection of various plant constituents,determination of total phenolic content and flavanoid content.[1-4]

MATERIALS AND METHODS:

Materials *Clerodendron paniculatum* The plant was collected from Pathanamthitta district of Kerala and identified by Thomas Mathew, HOD of Botany, Marthoma College Tiruvalla, Kerala .Voucher no. VSCI-13 was deposited in the Pharmacognosy department, Pushpagiri College of pharmacy, Tiruvalla.

Methods

The root portion of the plant was washed with running water to remove soil and other matter and dried in shade for 20 days, powdered, extracted 500gm with ethanol and ethyl acetate by cold extraction to yield the respective extracts. The extracts were reduced to molten mass by rotary vaccum evaporator and the yield was 18%, 21% respectively.

Phytochemical screening

Preliminary Phytochemical screening was performed as per standard procedure and various phytochemical constituents were identified [4-5]

Estimation of Total Phenolic content

Folin-ciocalteu method was used to determine the total phenolic content of the sample as described by singleton and Rossi (1965). The total phenolic content of the extracts was evaluated by spectrophotometric method measuring the absorbance at 765 nm. One milliliter of sample (concentration 1 mg/mL) was mixed with 1 mL of Folin and Ciocalteu's phenol reagent. After 3 min, 1 mL of saturated sodium carbonate solution was added to the mixture and made up to 10 mL with distilled water. The reaction was kept in the dark for 90 min, after which the absorbance was read at 725 nm. Gallic acid was used for constructing the standard curve (0–100 mcg/ml, $Y = 0.0162x + 0.0215$, $R^2 = 0.9985$) and the results were expressed as μg of gallic acid equivalents/ mg of extract (GAE).Total Phenolic content (% w/w) = $GAE \times V \times D \times 10^{-6} \times 100/W$ GAE – Gallic acid equivalent (mcg/ml), V– total volume of sample (ml), D – dilution factor, W sample weight(g). [6-7]

Estimation of Total Flavanoid content

Ten milligram of quercetin was dissolved in 100ml of methanol (80%) (100mcg/ml) and standard solutions of concentrations 0-50 mcg/ml were prepared. The diluted standard solutions (0.5 ml) were separately mixed with 1.5 ml of methanol (95%), 0.1 ml of aluminium chloride (10%), 0.1 ml of 1 M potassium acetate and 2.8 ml of distilled water. After incubation at room temperature for 30 min, the absorbance of the reaction mixture was measured at 415 nm with UV/VIS spectrophotometer. The amount of aluminium chloride (10%) was substituted by the same amount of distilled water in blank. About 1gm of ethanol extract, ethyl acetate extract root were dissolved separately in 25 ml of methanol (80%). Similarly, 0.5 ml of these extracts was reacted with aluminium chloride for determination of flavonoids content as described above. These parallel determinations were recorded. The amount of aluminium chloride (10%) was substituted by the same amount of distilled water in blank. Quantification was done on the basis of a standard curve of quercetin.($Y = 0.007x + 0.017$, $R^2 = 0.996$). A standard curve of extinction against quercetin concentration was prepared. Results were expressed as percentage % w/w. Flavonoids content (% w/w) = $QE \times V \times D \times 10^{-6} \times 100/WQE$ – quercetin equivalent (mcg/ml), V – total volume of sample (ml), D – dilution factor, W – sample weight (g). [6-8]

RESULT AND DISCUSSION:

Preliminary phytochemical screening was performed as per standard procedure and various phytochemical constituents were identified such as carbohydrates ,starch, mucilage ,saponins ,flavanoids ,tannins ,phenolic compounds in the different extract .The total phenolic content of the ethanolic root extract *Clerodendron paniculatum* .Linn was found to be 102.2 ± 0.65 mg/g gallic acid equivalent of crude extract. The flavanoid content of the ethanolic root extract of *Clerodendron paniculatum* Linn was found to be 56.35 ± 0.26 gm of rutin extract.

CONCLUSION:

From the present work helps to find out phytochemical investigations of the root , the total phenolic and flavanoid content of the ethanolic root extract which help to identify the active constituents which are not available in the literature and also helps to identify the marker compound.

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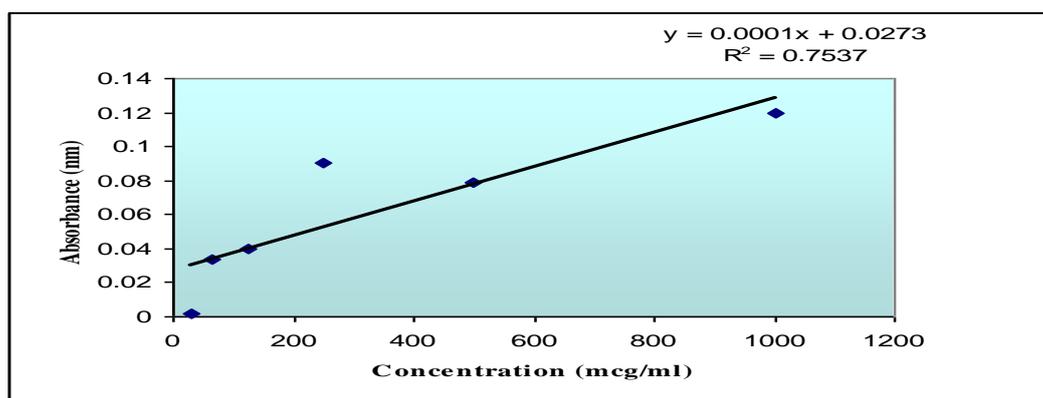


Fig 1: Calibration curve of Gallic acid

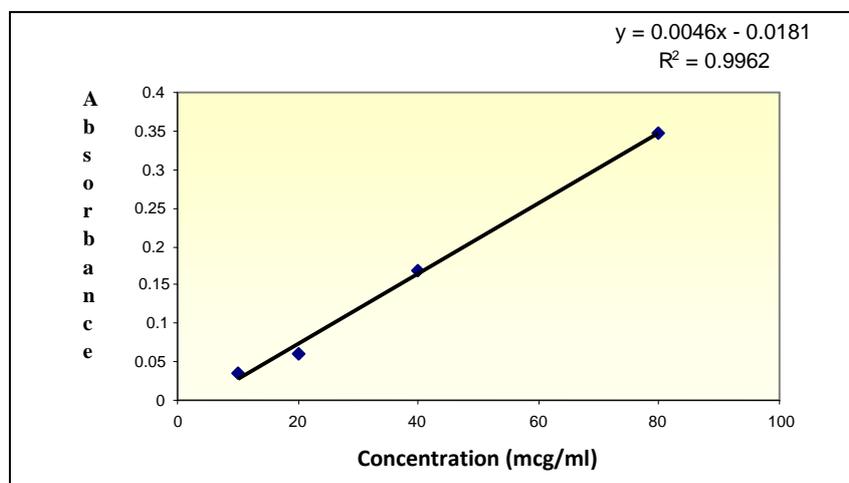


Fig 2: Flavonoid content of Ethanolic root extract of Clerodendron paniculatum Linn