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PHYTOCHEMICAL AND PHARMACOLOGICAL ACTIVITIES OF *POLYGALA CHAINENSIS*, *CLEOME CHELIDONII*

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Keywords:

Anthelmintic activity, Anti-oxidant activity, *Polygala chainensis*, *Cleome chelidonii*

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ABSTRACT: Natural products are an important source of bioactive compounds and have potential for the development of novel therapeutics. Natural products and their derivatives represent more than 50% of all drugs in clinical use in the world. Herbal plants contain and produce a verity of chemical substances used as a remedy for treating diseases. Anthelmintic and antioxidants have been treated with some medicinal plants or their extract based on folklore medicine. For the research *Polygala chainensis* and *Cleome chelidonii* selected based on its availability, high therapeutic value; activity has not been scientifically investigated. Very few pharmacological activities have been reported on whole plant of *Polygala chainensis* and *Cleome chelidonii*. The leaves of *Cleome chelidonii* and *Polygala chinensis* plants was observed for the phytochemical investigation and pharmacological evaluation. The percentage of yield and phytochemical was observed in methanolic extraction so this extraction was used for pharmacological activity. The extract of *Cleome chelidonii* and *Polygala chinensis* plants shown anthelmintic activity. The anthelmintic activity of *Polygala chinensis* was better than *Cleome chelidonii*. Significant DPPH free radical scavenging activity was found in methanolic extract the extract of *Cleome chelidoni* IC_{50} value is 28.06 ± 1.01 µg/ml and *Polygala chinensis* IC value is 30.1 ± 1.01 µg/ml compare with reference standard ascorbic acid IC value is 44.7 ± 2.01 µg/ml. The methanolic extract of *Polygala chinensis* was beater than *Cleome chelidonii* for antioxidant activity.

INTRODUCTION: Natural products are an important source of bioactive compounds and have potential for the development of novel therapeutics. Natural products and their derivatives represent more than 50% of all drugs in clinical use in the world. Over the decades there has been a growing interest in drugs of plant origin. During this period, utilization of medicinal plants has almost doubled in Western Europe and substances derived from higher plants constitute approximately 25% of prescribed medicines. Helminthiasis is one of the most important animal diseases worldwide, inciting heavy production losses in grazing animals.

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The disease is especially prevalent in developing countries in association with poor management practices and inadequate control measures. An integrated approach is required for the effective control of helminths which include strategic and tactical use of anthelmintics and careful management of grazing lands, including control of

stocking rates and appropriate rotation strategies. Antioxidants are radical scavengers which protect the human against free radicals that may cause pathological condition such as ischemia, asthma, arthritis, inflammation, neurodegeneration, Parkinson, mongolism, aging, process, and perhaps dementia. Flavonoids and flavones are widely distributed secondary metabolites with antioxidant and antiradical properties. Plant-based natural constituents can be derived from any part of plant bark, leaves, flowers, roots, fruits, seeds, *etc.* that is any part of the plant may contain active components.

Cleome chelidonii (Cleomaceae) (commonly known as mountain bee plant or celandine spider flower) is found throughout in India from the Santal hills, Orissa and Gujarat, Southwards. It is mostly found in Tirupathi, Ratnagiri areas. *Cleome chelidonii* mainly contains glucocapparin and glucocleomin as phytochemical constituents. *Cleome chelidonii* was also reported for its antioxidant and anti-inflammatory activity [3]. *Polygala* was traditionally used by Americans to treat snakebite and as an expectorant to treat cough and bronchitis. *Polygala* is considered as a powerful tonic herb that can help to develop the mind and aid in creative thinking. Hence, an attempt is known to evaluate the efficacy of *Polygala chinensis* as an anti-inflammatory agent in traditional healing system [4].

Anthelmintic and antioxidant have been treated with some medicinal plants or their extract based on folklore medicine. For the research work, *Polygala chainensis* and *Cleome chelidonii* selected based on its availability, high therapeutic value; activity has not been scientifically investigated. Very few pharmacological activities have been reported on whole plant of *Polygala chainensis* and *Cleome chelidonii* so i selected the plants to submit the research. Which involves phytochemical screening and pharmacological (Anthelminthic and antioxidant activity) evaluation to provide scientific validation to its folklore claims.

MATERIALS AND METHODS:

Collection, Identification, and Authentication of Plants: *Cleome chelidonii* was grown widely throughout India. The plant was identified and authenticated by Prof. P. Suresh Government Degree College Ibrahimpatnam. A voucher specimen (GP-PC- 2013/02.) was stored in Department of Pharmacognosy. *Polygala chinensis* was grown widely throughout India. The plant was identified and authenticated by Prof. P. Suresh Government Degree College Ibrahimpatnam. A voucher specimen (GP-PC- 2013/03.) was stored in the Department of Pharmacognosy.

Extraction Procedure: Freshly collected plant materials were dried under shade, and the dried material was milled to obtain a coarse powder. The coarse powder was packed in a Soxhlet apparatus and subjected to sequential extraction with pet ether, methanol, and water. The liquid extracts were collected and evaporated under reduced pressure until a soft mass obtained. The mass obtained was weighed in each case. The extracts were thoroughly air-dried to remove all traces of the solvent.

Preliminary Phytochemical Screening: The condensed extracts were used for preliminary screening of phytochemicals such as cholesterol, alkaloid, flavonoids, saponin, cardiac glycosides and terpenoids [5].

Screening Procedure:

Test for Flavonoids: Add a few drops of concentrated HCl and Mg, turning to 1 ml of ethanol extract. The appearance of pink or magenta-red color indicates the presence of flavonoids.

Test for Cholesterol: To 2 ml of the extract 2 ml of the chloroform was added in a dry test tube. Then 10 drops of acetic anhydride and 2 to 3 drops of con. H_2SO_4 was added. A red color changed to blue-green color.

Test for Alkaloids: To the extract added 1% HCl and 6 drops of Mayer's reagent and Dragendorff reagent. Any organic precipitate indicated the presence of alkaloids in the sample.

Test for Terpenoids: 5 ml of each extract was added to 2 ml of chloroform and 3 ml of conc. H_2SO_4 to form a monolayer of reddish-brown coloration of the interface was showed to form positive results for the terpenoids.

Test for Cardiac Glycoside: 5 ml of each extract was treated with 2 ml of glacial acetic acid containing one drop of ferric chloride solution. This was underplayed with 1 ml of con. H_2SO_4. A brown ring of the interface indicated a deoxysugar characteristic of cardenolides. A violet ring might appear below the brown ring whereas the acid layer, a greenish ring might form just gradually throughout thin layer.

Test for Steroids: 2 ml of acetic anhydride was added to 0.5 gm of ethanolic extract of each sample with 2 ml of H_2SO_4. The color change from violet to blue or green indicated the presence of steroids.

Test for Saponins: The extract with 20 ml of distilled water was agitated in a graduated cylinder for 15 min. The formation of 1 cm layer of foam indicated the presence of saponins.

Pharmacological Studies *in-vitro* Antioxidant Activity:

Chemicals Required: 1, 2-diphenyl-2-picryl-hydrazyl (DPPH). Ascorbic acid.

Procedure: Antioxidant activity has been performed by the DPPH method. Scavenging activity of antioxidants was studied by using DPPH (1.1-diphenyl-2- picrylhyldrazyl free radical). Various of concentration of as 2.5, 5, 10, 25, 50, and 100 µg/ml in 0.1 ml was added to 0.9 ml of solution of DPPH in methanol. Methanol only (0.1 ml) was used as experimental control. After 30 min of incubation at room temperature, the reduction in the number of free radicals was measured by reading the absorbance at 517 nm. Reference standard (Ascorbic acid). The scavenging activity of the sample corresponded to the intensity of quenching DPPH. The percent inhibition was calculated from the following equation:

% inhibition = (Absorbance of control – absorbance of the test sample) / absorbance of control × 100 [6]

Anthelmintic Activity: The methanolic extract of *Polygala chinensis*, and *Cleome chelidonii* was tested in various doses in each group. Normal saline water was used as control. Piperazine citrate and albendazole were used as standard drugs for comparative study with methanolic extracts.

Procedure: The method of nargoud was followed for screening of anthelmintic activity. Anthelmintic activity was evaluated on adult *Pheretima posthuma*. Earthworms were divided into eight groups (5 each). The first group (I) served the standard drugs piperazine citrate and albendazole at a dose level of 10 mg/ml groups (VI) to (IX) received doses of methanolic extract of 10 mg/ml, 15 mg/ml, 20 mg/ml, 25 mg/ml, 30 mg/ml, and 35 mg/ml respectively. Observations were made for the time taken to cause paralysis and death of individual worm for two hours. Paralysis was confirmed when the worms did not revive even in normal saline water. Death was concluded when the warms lost their lost motility followed by fading away from their body colour [7].

RESULTS:

Percentage Yield of Extraction:

TABLE 1: PERCENTAGE YIELD OF *CLEOME CHELIDONII*

S. no.	Type of extraction	Practical yield (in gm)	Percentage yield (w/w)
1	Petroleum ether extract	3.6	7.2
2	Methanol extract	4.8	9.6
3	Aqueous extract	3.2	6.4

TABLE 2: PERCENTAGE YIELD OF *POLYGALA CHINENSIS*

S. no.	Type of extraction	Practical yield (in gm)	Percentage yield (w/w)
1	Petroleum ether extract	3.5	**7**
2	Ethanol extract	5.1	10
3	Aqueous extract	3.1	6.2

Quantitative Phytochemical Analysis of Extracts:

TABLE 3: QUANTITATIVE PHYTOCHEMICAL ANALYSIS OF *CLEOME CHELIDONII*

S. no.	Test	Pet. ether extraction	Methanol extract	Aqueous; Methanol
1	Carbohydrates	+	+	+
2	Glycosides	-	-	-
3	Proteins & Amino acids	-	-	-
4	Fixed oils & Fats	-	-	-
5	Alkaloids	+	-	-
6	Phytosterols	-	-	-

7	Flavonoids	+	+	-
8	Phenolic compounds	+	+	+
9	Saponins	-	-	-
10	Tannins	-	-	-

TABLE 4: QUANTITATIVE PHYTOCHEMICAL ANALYSIS OF *POLYGALA CHINENSIS*

S. no.	Test	Pet. ether extraction	Methanol extract	Aqueous; Methanol
1	Carbohydrates	-	-	-
2	Glycosides	-	-	-
3	Proteins & Amino acids	-	-	-
4	Fixed oils & Fats	-	-	-
5	Alkaloids	-	+	+
6	Phytosterols	-	-	-
7	Flavonoids	+	+	-
8	Phenolic compounds	+	+	+
9	Saponins	-	-	-
10	Tannins	-	-	-

Anthelmintic Activity:

TABLE 5: ANTHELMINTIC ACTIVITY OF *CLEOME CHELIDONII*

Groups	Treatment	Concentration used (mg/ml)	Time taken for paralysis (Mean & SEM)	Time taken for death (Mean & SEM)
1	Vehicle normal saline	-	-	-
2	Standard (Albendazole)	10	2.5 ± 0.25	50.6 ± .3
3	MOLC 1	10	92.6 ± 0.5	125.21 ± 0.5
4	MOLC 2	15	76.5 ± 0.5	108.3 ± .4
5	MOLC 3	20	63.2 ± 0.2	96 ± 0.1
6	MOLC 4	25	51.6 ± 0.3	72.2 ± 0.4
7	MOLC 5	30	35.6 ± 0.3	45 ± 0.3

Significant difference from control by one way ANOVA, followed by Dunnett's test (n=5), *p<0.00

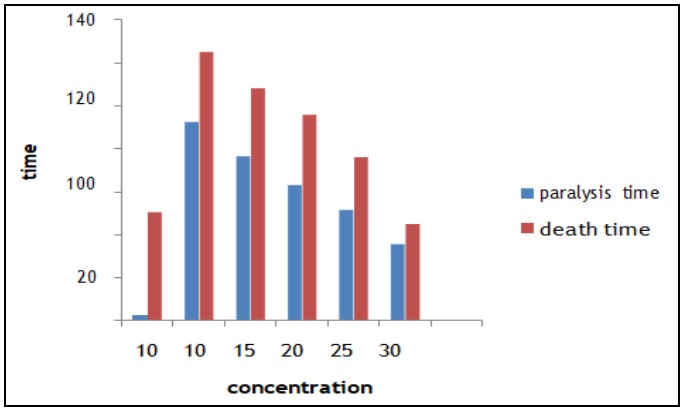

FIG. 1: ANTHELMINTIC ACTIVITY OF *CLEOME CHELIDONII*

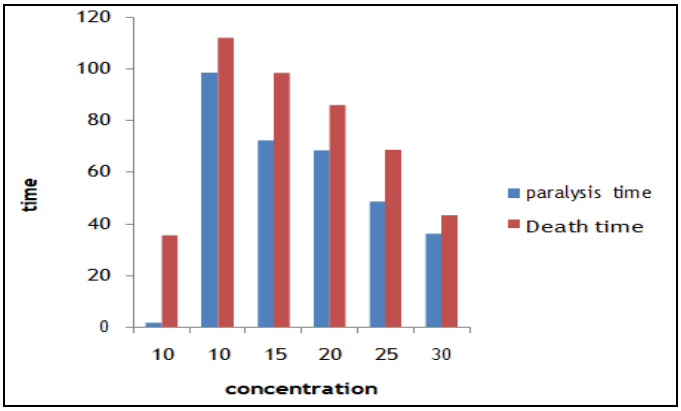

FIG. 2: ANTHELMINTIC ACTIVITY OF *POLYGALA CHINENSIS*

TABLE 6: ANTHELMINTIC ACTIVITY OF *POLYGALA CHINENSIS*

Groups	Treatment	Concentration used (mg/ml)	Time taken for paralysis (min)	Time taken for death (min)
1	Vehicle normal saline	-	-	-
2	Standard (Albendazole)	10	1.8 ± 0.25	35.6 ± 0.2
3	MOLC 1	10	98.6 ± 0.2	112 ± 0.5
4	MOLC 2	15	72.3 ± 0.2	98.5 ± 0.2
5	MOLC 3	20	68.5 ± 0.1	86 ± 0.1
6	MOLC 4	25	48.6 ± 0.3	68.6 ± 0.4
7	MOLC 5	30	33.2 ± 0.3	43.3 ± 0.3

Significant difference from control by one way ANOVA, followed by Dunnett's test (n=5), *p<0.001

Antioxidant Activity:

Determination of Total Phenolic Content:

TABLE 7: STANDARD GRAPH OF ASCORBIC ACID

Concentration (µg/ml)	Absorbance
0	0
20	0.015
40	0.029
60	0.049
80	0.057
100	0.071
1 mg ethanol extract	0.027

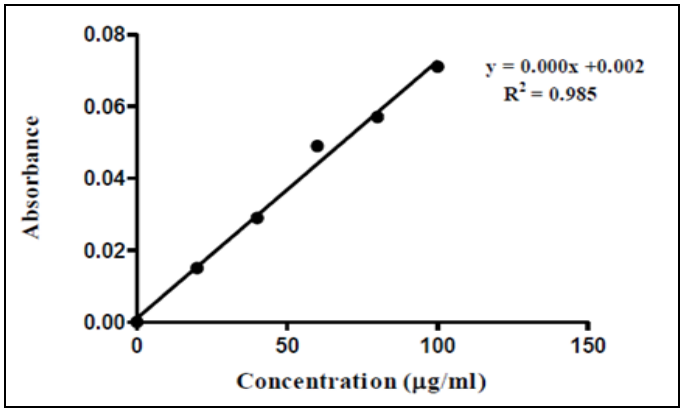

FIG. 3: STANDARD GRAPH OF ASCORBIC ACID

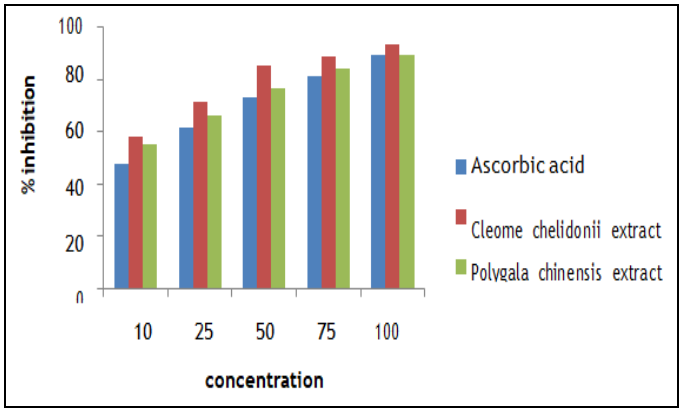

FIG. 4: DPPH RADICAL SCAVENGING ACTIVITY

1, 1-Diphenyl-2-Picrylhydrazyl Assay:

TABLE 8: EFFECT OF ETHYL ACETATE & ETHANOL EXTRACTS ON DPPH RADICALS

Concentration (µg/ml)	Percentage inhibition		
	Ascorbic acid	*Cleome chelidonii* extract	*Polygala chinensis* extract
0	0	0	0
10	47.90 ± 1.445	58.29 ± 1.456	55.42 ± 2.011
25	61.66 ± 1.132	71.35 ± 3.325	66.37 ± 1.425
50	73.32 ± 1.496	85.42 ± 1.256	76.66 ± 1.526
75	81.22 ± 2.485	88.52 ± 1.523	84.21 ± 2.065
100	89.52 ± 44.7	93.23 ± 0.125	89.31 ± 0.235
IC_{50} (µg/ml)	2.425 ± 2.01	28.06 ± 1.01	30.1 ± 1.01

CONCLUSION: The leaves of *Cleome chelidonii* and *Polygala chinensis* plants were observed for the phytochemical investigation and pharmacological evaluation. The percentage of yield and phytochemical was observed in methanolic extraction so this extraction was used for pharmacological activity. The extract of *Cleome chelidonii* and *Polygala chinensis* plants shown anthelmintic activity. The anthelmintic activity of *Polygala chinensis* was better than *Cleome chelidonii*. Significant DPPH free radical scavenging activity was found in methanolic extract the extract of *Cleome chelidonii* of IC_{50} value is 28.06 ± 1.01 µg/ml and *Polygala chinensis* IC value is 30.1 ± 1.01 µg/ml compare with reference standard ascorbic acid IC value is 44.7 ± 2.01 µg/ml. The methanolic extract of *Polygala chinensis* was beater than *Cleome chelidonii* for antioxidant activity.

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CONFLICT OF INTEREST: Nil

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