

**IN VITRO ANTIOXIDANT, CYTOTOXIC, AND ANTI-TUBERCULAR ACTIVITIES OF *CLEOME GYNANDRA* WHOLE PLANT EXTRACTS**Alluri Pavani Gayatri<sup>1</sup>, Sai Krishna Nallajerla<sup>1</sup>, Mohan Gandhi Bonthu<sup>2</sup><sup>1</sup> Department of Pharmacology, AKRG College of Pharmacy, Nallajerla, West Godavari district Andhra Pradesh 534112.<sup>2</sup> Department of Pharmaceutical Analysis, V.V. Institute of Pharmaceutical Sciences, Gudlavalleru, Krishna district, Andhra Pradesh 521356.

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DOI: <https://doi.org/10.47957/ijciar.v6i3.156>

Received: 28 July 2023 Revised: 11 Aug 2023 Accepted: 03 Sept 2023

**Abstract**

The current study investigates the antioxidant, anticancer, and anti-TB characteristics of *Cleome gynandra*, a traditional medicine herb. The research shows that *C. gynandra* has significant radical scavenging activity, suggesting it may be a natural antioxidant that can reduce oxidative stress and cancer. Several cell lines were cytotoxic by plant extracts. The ethanol extract had the maximum cytotoxicity against MCF-7 cells, while the diethyl ether extract had the highest against HCT-116 cells. The hexane extract cytotoxicity affected HepG2, HCT-116, and L6 cells. The aqueous extract of *C. gynandra* had the lowest cytotoxicity of the cell lines studied. The cytotoxic effects of *C. gynandra* extracts were highly selective, especially when targeting MCF-7 and HCT-116 cell lines. The study found that the hexane extract had the strongest anti-tuberculosis (TB) effects, with an IC<sub>50</sub> of 1.01 µg/ml. The ethanol extract had a substantially lower IC<sub>50</sub> value of 7.599 µg/ml, while the ether extract had an IC<sub>50</sub> value of 32.7 µg/ml. Finally, the aqueous extract had the highest IC<sub>50</sub> value of 793.3 µg/ml. The results suggest that *C. gynandra* could be used to generate natural anticancer drugs with fewer side effects. The plant's potential anticancer effect's mechanism needs further in vitro and in vivo research.

**Keywords:** *Cleome gynandra*, antioxidant, anticancer, selective cytotoxicity, cell lines, DPPH assay, SRB assay, natural compounds, phytochemicals.

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**Introduction**

The pandemic of cancer transcends borders. The COVID-19 pandemic has disrupted cancer detection and treatment due to hospital closures, work adjustments, and increased concerns about virus exposure. The user's text is empty. Cancer will be the top or secondary cause of death in 112 of 183 nations in 2019, according to the WHO. The International Agency for Research on Cancer (IARC) estimates the global prevalence of 36 cancers in 185 countries [2]. According to 2020 GLOBOCAN forecasts, 19.3 million new cancer cases will be diagnosed worldwide, resulting in 10.0 million deaths. The prevalence of lung, prostate, and colon cancer is higher in men, whereas breast, colon, and lung cancer are more common in women. Breast cancer has surpassed lung cancer as the most common malignant neoplasm, with 2.3 million new cases (11.7%). Lung (11.4%), colon (10.0%), prostate (7.3%), and stomach (5.6%) follow. Over 1.8 million people (18%) died from lung cancer, the leading cause of cancer deaths. Colon, liver, stomach, and breast cancers in women followed. Death rates varied by gender, with industrialized countries having two to three times higher mortality rates than transitional countries [3]. Cancer incidence is rising due to urbanization, lifestyle changes, and longer life expectancies. Asia has about 50% of newly diagnosed cancer cases and 60% of cancer-related deaths. According to the source, Europe had 22.8% more cancer cases and 19.6% more cancer deaths than the Americas [2]. In 2023, the US will see 1,958,310 new cancer diagnoses and 609,820 cancer deaths. Over the next two decades, new cancer cases are expected to rise 50% [4]. Around 28 million people will be diagnosed with cancer annually by 2040, resulting in 16 million fatalities [5, 6]. In contrast, tuberculosis, caused by the bacteria *Mycobacterium tuberculosis*, mostly affects the respiratory system and can have serious medical consequences if not treated. In 2019, 10 million cases and 1.4 million deaths worldwide indicated the issue's global health importance [4]. Current tuberculosis (TB) treatment includes isoniazid, rifampin, ethambutol, and pyrazinamide for at least 6 months [5]. However, the emergence of drug-resistant *M. tuberculosis* strains, particularly MDR and XDR, has made TB management and containment difficult [6].

Cleome is a genus of the family Cleomaceae (formerly Capparaceae) is a major group of angiosperms, comprising many species found in tropical and sub-tropical areas of the globe. Cleomaceae family, encompassing flowering plants are Brassicales (or Cruciales) order, including more than 764 species belonging to 12 genera of which Cleome is the largest genus with about 601 species of ecological, ethnobotanical, and of course medicinal importance [6,8,9]. Different species of Cleome are therapeutically utilized in Island, North and Central America, Philippines, and Indo-China. In India, of 15 species 12 are reported in Maharashtra [10]. Cleome genus is under constant advancement; numerous species demonstrate a progressive movement from C3 photosynthesis to C4 photosynthesis and this developmental movement is indistinguishable to Brassicaceae individuals like *Arabidopsis thaliana*. There is extremely inadequate and scattered work in the genus, Cleome. Especially, the anatomical and physiological examinations in the species are uncommon [11]. *C. gynandra* is an opulently accessible species and matures as a weed in common sterile and in crop grounds throughout the world. As a weed, it is generally found growing on fertile soils, particularly in those previously blended with animal fertilizer, or with homestead disposed. Ideal growing conditions for *C. gynandra* require suitable soil moisture, high-intensity light and temperatures of ~25° C. In different countries, it is used to treat many diseases as traditional medicine, and it is additionally utilized as a part of different conventional culinary systems for its astounding antioxidant and nutritional activities [12-14]. A good number of phytochemicals have been isolated from different parts of white mutant *C. gynandra* which confirms its current understanding of nutritional claims and pharmacological evidence, whereas a few compounds, namely, clenbuterol, stearin compound, bicyclohexyl derivatives, and (5Z, 8Z)-3-hydroxypropyl dodeca-5,8-dienoate only been isolated from the pink mutant variety, only available in N.E states. The pharmacological importance of *C. gynandra* is referred in Ayurveda; Gulma (tumor, irregularity, or diverticulosis), Krmiroga (worm infection), Asthila (Prostate enlargement), Kandu (pruritus), and Karnaroga (ear infections). The indigenous information of numerous traditional medicines has been figured, reported, and eventually wind up noticeably with composed frameworks of the drug, for example, Ayurveda, Unani, Siddha, and other indigenous traditional system throughout the world [8]. The following are some therapeutic investigations reported by various researchers from India and from other nations as well. The main goal of this study was to test *C. gynandra*'s anticancer potential in different cell lines. The study also used the anti-microbial activity assay against *M. kansasii* to evaluate *C. gynandra*'s in vitro anti-TB activity.



**Fig. 1: Morphological View of *Cleome gynandra***

## **Experimental**

### ***Plant material and Extraction***

The entire *Cleome gynandra* plant was shade-dried and ground into a fine powder. A 5 g powdered sample was extracted with 50 ml each of ethanol, diethyl ether, hexane, and de-mineralized water. Ethanol, dimethyl ether, and hexane were incubated on a rocker shaker for 24 hours. In aqueous extraction, the sample mixture was boiled to one-fourth its original volume. After filtration using Whatman filter paper 1, the extract was dried in a 40 °C oven. The specimen was kept in a 4 °C micro centrifuge tube.

### ***DPPH radical scavenging assay***

In a 96-well plate, a 10 µl aliquot of the test chemical stock solution (0-2500 µg/mL) was mixed with 0.2 mL of 0.2 mM DPPH solution. The experiment was done in triplicate, with duplicate control solution samples of 0.2 mL DMSO/Methanol and 5µL test drug at varying dosages from 0 to 2500 µg/mL. After that, the plate was incubated in darkness for 30 minutes. The incubation period ended with a microplate reader (iMark, BioRad) measuring decolorization at 495 nm. Ascorbic acid was used as a reference. IC50 values and % inhibition relative to the control were calculated for scavenging activity.

### **Superoxide radical scavenging assay**

In this experiment, superoxide radicals convert Nitroblue Tetrazolium (NBT) into NBT diformazan. The tetrazolium salt used by superoxide dismutase (SOD) transforms into a water-soluble formazan color when reduced by a superoxide anion. Sodium oxalate dihydrate (SOD) inhibits xanthine oxidase, which speeds oxygen-based reduction. The substrate for this experiment is nitro blue tetrazolium (NBT), which is reduced to a purple formazan molecule. The non-enzymatic interaction between phenazine methosulfate (PMS) and nicotinamide adenine dinucleotide (NADH) generates superoxide radicals that aid this reduction. The reaction mixture contained 20 mM phosphate buffer solution at pH 7.4, 73 mM NADH, 50 mM NBT, and 15 mM PMS. The reaction mixture was incubated at room temperature for 10 minutes with 1 mL of the chemical. The absorbance was measured at 562 nm using a UV-visible spectrophotometer and a blank. The experiment was repeated three times and provided as percentage inhibition.

Using a non-linear regression curve to plot scavenging activity against concentration logarithms, extracts or isolated compounds' 50% inhibitory concentration (IC<sub>50</sub>) values were found. The experiment's positive control was ascorbic acid. The mean standard deviation (SD) of triplicate experiments is shown.

### **Cell culture**

The American Type Culture Collection (Rockville, MD) provided this study's cell lines. MCF-7, HaCaT, A549, HepG2, HCT-116, and L6 are human breast cancer, lung carcinoma epithelial cells, liver cancer, and colorectal carcinoma cell lines. 8000 cells were cultured in 96-well plates for 24 hours. The culture medium was Dulbecco's Modified Eagle's Medium (DMEM, Cat No. AT149-1L) with 10% FBS and 1% antibiotic solution. The cells were kept at 37 °C with 5% CO<sub>2</sub>.

### **Sulforhodamine B (SRB) Assay**

The SRB test (Ottokemi, Cat. No. 3520-42-1) assessed extract cytotoxicity on cell lines. The cells were cultivated at 8000 per well in a 96-well plate. The culture media was DMEM with 10% FBS and 1% antibiotic solution. The cells were cultured for 24 hours at 37°C with 5% CO<sub>2</sub>. On the following day, cells were treated with various dosages from 1 to 1000 µg/mL. An incomplete medium prepared many concentrations. After a 24-hour incubation period, each well was incubated for 1 hour with 100 µL of 10% Tri Chloro Acetic Acid (TCA, Fisher Scientific 28444). Rinse the wells with deionized water (DM water) and air-dry at room temperature. Each well was incubated for 1 hour with 0.04% sulfate-reducing bacteria (SRB). After 60 minutes of incubation, the plate was washed with 1% (v/v) acetic vinegar to remove any remaining color. The plate was then air-dried at room temperature. A pH 10.5 Tris-base solution was added to the well and orbitally shaken for 10 minutes to dissolve the protein dye. The solution was measured at 510 nm using an Elisa plate reader (iMark, Biorad, USA) [19].

### **Test Organism and Media:**

The *M. kansasii* strain was tested for anti-TB activity by the extracts. The bacterial cells were grown in 7H9 broth at 37°C and 100 RPM until mid-logarithmic.

### **Minimum Inhibitory Concentration Anti-TB:**

MIC activity against *M. kansasii* was measured using the anti-microbial activity test [16]. The study used a typical dilution approach to insert 500 µl of diluted log cultures of bacteria into micro centrifuge tubes. Next, 10 µl of treatment dilutions with different concentrations were applied to the tubes. The tubes were incubated for 24 hours after making treatment dilutions of various concentrations according to the excel sheet. After incubation, the contents were transferred to a 96-well plate. After that, an Elisa Plate Reader (iMarkBiorad) assessed optical density at 490nm and 595nm. The study used Ciprofloxacin at a concentration of 100µg as the Positive Control.

### **Statistical analysis**

The mean value and standard error of the mean (SEM) were given from triplicate trials.

## **Results**

### **Radical scavenging activity of *C. gynandra***

The radical scavenging activity of all the extracts was assessed using the DPPH assay. The extracts that were observed demonstrated a correlation between concentration and the activity of scavenging radicals. The IC<sub>50</sub> value for the ethanol extraction of *C. gynandra* was found to be 23.63µg/ml, demonstrating comparable efficacy to ascorbic acid (IC<sub>50</sub> value of 9.138µg/ml). Subsequently, the hexane extract exhibited an IC<sub>50</sub> value of 112.8µg/ml, while the ether extract displayed an IC<sub>50</sub> value of 215.3µg/ml. The IC<sub>50</sub> value for the hexane extract was 183.21µg/ml.

### **Superoxide radical scavenging assay**

The superoxide radical-quenching assay was used to evaluate the extracts' free radical-scavenging abilities. The graph compares scavenging activity IC<sub>50</sub> values. Depending on concentration, all extracts eliminated free radicals. *C. gynandra*'s ethanol extraction yielded an IC<sub>50</sub> value of 11.32µg/mL, similar to ascorbic acid's 9.135µg/mL. The hexane extract has an IC<sub>50</sub> value of 92.30µg/mL, while the aqueous extract had 213.52µg/mL. The aqueous extract had the lowest IC<sub>50</sub> value of 328.38µg/mL.

### **Cytotoxic potential of *C. gynandra***

The sulforhodamine B (SRB) assay was used to evaluate *C. gynandra*'s cytotoxicity against MCF-7, HaCaT, A549, HepG2, HCT-116, and L6 cells. All four extracts suppressed cell viability across all cell lines with dosage. The aqueous extract was the least active, while the ethanol extract was most active. The hexane extract showed significant cytotoxic effects on HCT-116 and HepG2 cell lines (IC50 = 43.98 µg/ml and 49.45 µg/ml, respectively), but had a weaker reaction on A549 cell lines (IC50 = 81.43 µg/ml). Its efficiency against HaCaT and MCF-7 cell lines was limited. The diethyl ether extract significantly cytotoxic to HCT-116 cells, with an IC50 value of 30.38 µg/ml. It showed cytotoxic action against MCF-7 and L6 cell lines, with IC50 values of 68.51 and 76.31 µg/ml, respectively. Additionally, the A549 cell line demonstrated cytotoxicity with an IC50 of 78.83 µg/ml. Additionally, the ethanol extract of *C. gynandra* showed significant cytotoxic effects on MCF-7 cells, with an IC50 value of 30.03 µg/mL. It did not work on other cell lines. None of the extracts cytotoxic against HaCaT cells.

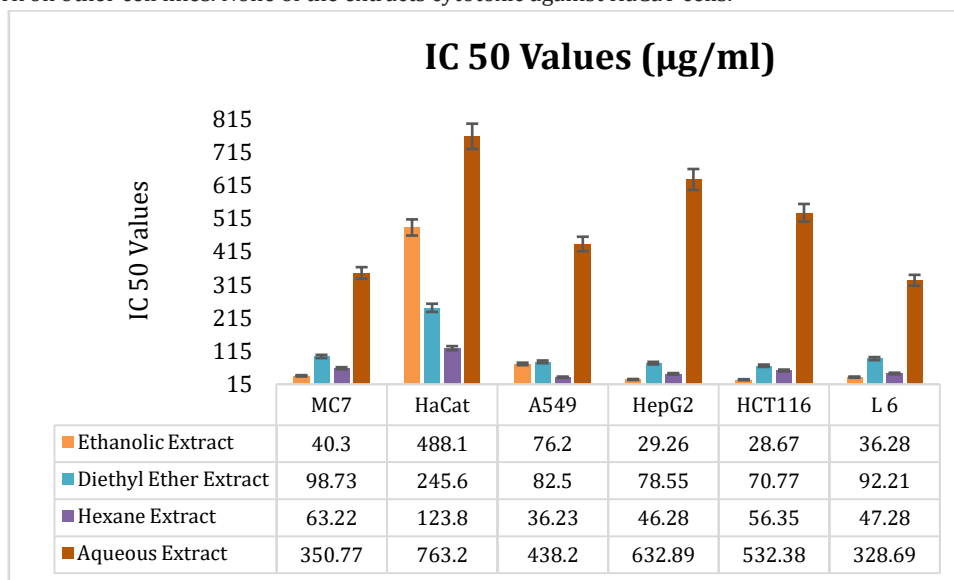


Figure 2. Radical scavenging activity of different extracts of *C. gynandra*. The data is expressed as mean ± SEM, (n=3)

#### Minimum Inhibitory Concentration Anti-TB:

The investigation found that the ethanolic extract had the lowest IC50 value (1.61 µg/ml), followed by ether (5.321 µg/ml), hexane (13.8 µg/ml), and aqueous (634.20 µg/ml). The IC50 values are the extract concentration needed to inhibit microorganism growth by 50%. The ethanol extract had the highest antibacterial activity, whereas the hexane and ether extracts had lesser amounts.

#### Discussion

*Cleome* species are found worldwide in tropical and subtropical regions [20]. Many species in this genus have been used in traditional medicine to cure various diseases. These include antibacterial, antidiabetic, abortifacient, wound healing, hepatoprotective, and boil, rheumatic pain, and skin infection treatment. Research on *Cleome* species shows that this genus produces most C-glycosyl flavonoids. Several *Cleome* genus and plant extracts contain antioxidant, anti-inflammatory, antimycobacterial, and antiproliferative properties. *C. gynandra* is a notable *Cleome* species used in traditional medicine. Additionally, there is a large amount of data suggesting natural chemicals are being studied as anti-proliferative and chemo preventive medicines. These natural alternatives are being studied as chemically produced pharmaceutical equivalents to reduce toxicity and side effects. This study investigated *C. gynandra*'s antioxidant and anticancer effects on various cell lines.

Antioxidants protect the body against chronic diseases by lowering free radical-induced cellular oxidative damage. Free radical overexpression and oxidative stress influence various pathways in cancer etiology. Thus, the DPPH and Superoxide radical scavenging assays were used to investigate *C. gynandra*'s antioxidant capacity. The concentration-dependent radical scavenging activities in all extracts of *C. gynandra* showed significant antioxidant activity. The IC50 values of the extracts compared to ascorbic acid were hexane, diethyl ether, and aqueous ethanol. However, the ethanolic *C. gynandra* extract showed the lowest radical scavenging activity, with an IC50 value that surpassed the assay concentration range. Numerous investigations have found phenols, flavonoids, and proteins in *C. gynandra*, which may explain its anticancer capabilities. Researchers are also studying bioactive compounds and their derivatives as natural antioxidants. This is to defend against oxidative stress, which reactive oxygen species cause and is linked to many diseases, including cancer. Concerns about synthetic antioxidants' side effects drive interest in natural antioxidants. In this case, *C. gynandra* may be effective.

The SRB assay was used to assess *C. gynandra*'s anti-cancer efficacy against MCF-7 (human breast cancer), Hacat (immortalized human keratinocytes), A549 (lung carcinoma epithelial cells), HepG2 (liver cancer), HCT-116 (human colorectal carcinoma), and L6 (skeletal muscle cell lines). The concentration-dependent cell viability suppression of all five cell lines was reported in all *C.*

*C. gynandra* extracts. However, the extracts targeted a specific cell line for cytotoxicity. The ethanol extract of *C. gynandra* showed the maximum cytotoxicity against HCT116, with an IC<sub>50</sub> value of 28.67 µg/ml, followed by HepG2, L6 & A549. The diethyl ether extract from *C. gynandra* was cytotoxic to HCT-116 cells at 70.77 µg/ml. It moderately cytotoxic against HepG2 and A549 cells. The hexane extract from *C. gynandra* showed substantial cytotoxic effects on A549, followed by HepG2 & L6 cell lines, with IC<sub>50</sub> values of 36.23, 46.28 and 47.28 µg/ml. However, the hexane extract showed moderate cytotoxic activity in HCT116, and MCF-7 cell lines. Of the four extracts tested, the aqueous *C. gynandra* extract had the lowest cytotoxicity to all cell lines. The HaCaT cell line was not inhibited by any extract. The conclusions of this investigation match previous research. *C. gynandra* cytotoxicity was mostly selective for Hep G2, L6 and HCT-116 cell lines.

*C. gynandra* preparations were tested for tuberculosis resistance using a minimum inhibitory concentration (MIC) assay against *Mycobacterium kansasii*. The study found that the ethanol extract had the strongest anti-tuberculosis (TB) effects, with an IC<sub>50</sub> of 1.61 µg/ml. The ether extract had a substantially lower IC<sub>50</sub> value of 5.321 µg/ml, while the hexane extract had an IC<sub>50</sub> value of 13.8 µg/ml. Finally, the aqueous extract had the highest IC<sub>50</sub> value of 634.23 µg/ml. The Hexane and Ethanol extracts may include substances that suppress *M. kansasii* growth better than the other extracts. The solubility and polarity of the compounds in the extracts may affect their ability to interact with the bacterial cell membrane and inhibit growth. The plant extracts showed significant anti-TB and anti-diabetic effect against *Mycobacterium kansasii*, as measured by glucose absorption and alpha-amylase inhibition.

The study has potential, but more research is needed to determine the safety and efficacy of *C. gynandra* extracts as diabetes and tuberculosis treatments in animal models. To better understand the plant extracts' anti-diabetic and anti-TB activities, we need more data on *C. gynandra*'s bioactive compounds and their pharmacological effects. The study generally supports *C. gynandra*'s pharmacological qualities.

## Conclusion

The current study unveiled that the botanical species *C. gynandra* had significant antioxidant, anticancer, and anti-tuberculosis characteristics. The botanical specimen shown significant effectiveness in the scavenging of radicals, a critical attribute of a naturally occurring molecule that has substantial importance in relation to diverse diseases, such as cancer. In addition, the plant demonstrated selective cytotoxicity against several cell lines employed in the study, with the degree of effectiveness dependent on the particular extract fraction. Additional research is required to better understand the specific mechanism by which the plant may demonstrate potential anticancer effects. This necessitates conducting in vitro and in vivo trials.

## Funding

No Funding

## Acknowledgement

The authors are thankful to the management of AKRG College of Pharmacy, Nallajerla, for providing necessary research facilities.

## Informed Consent

Not Applicable

## Ethical Statement

Not Applicable

## Author Contribution

Study conception and design: Alluri Pavani Gayatri, Mohan Gandhi Bonthu; data collection: Alluri Pavani Gayatri; analysis and interpretation of results: Alluri Pavani Gayatri, Mohan Gandhi Bonthu, N. Saikrishna; draft manuscript preparation: Alluri Pavani Gayatri, Mohan Gandhi Bonthu. All authors reviewed the results and approved the final version of the manuscript.

## Conflicts of Interest

The authors declare no conflicts of interest.

## References

1. Greenwell M., Rahman P.K. Medicinal Plants: Their Use in Anticancer Treatment. *Int. J. Pharm. Sci. Res.* 2015;6:4103–4112. doi: 10.13040/IJPSR.0975-8232.6(10).4103-12.
2. Siegel R.L., Miller K.D., Jemal A. Cancer Statistics, 2017. *CA Cancer J. Clin.* 2017;67:7–30. doi: 10.3322/caac.21387.
3. Banerjee P., Erehman J., Gohlke B.O., Wilhelm T., Preissner R., Dunkel M. Super Natural II—A database of natural products. *Nucleic Acids Res.* 2015;43:D935–D939. doi: 10.1093/nar/gku886. C. Melissa, R. Siegel. *Ame Cancer Soc*: Atlanta, GA, USA;

(2018);<https://www.cancer.org/content/dam/cancer-org/research/cancer-facts-and-statistics/annual-cancer-facts-and-figures/2018/cancer-facts-and-figures-2018.pdf>

4. Adhikari PP, Paul SB, Choudhury MD, Choudhury S. GC-MS studies on the steam-distillate of the medicinally important plant *Cleome gynandra* L. *Int J Appl Res Stud* 2014;3:1-4.
5. Aparadh VT, Mahamuni RJ, Karadge BA. Taxonomy and physiological studies in spider flower (*Cleome* species): A critical review. *Plant Sci Feed* 2012;2:25-46.
6. The Plant List. The Plant List, Version 1; 2010. Available from: <http://www.theplantlist.org>. [Last cited on Jan 01] [Last accessed on Aug 15].
7. Ravichandra B, Ram PS, Saritha C, Shankaraiah P. Anti-diabetic and anti-dyslipidemia activities of *Cleome gynandra* in alloxan induced diabetic rats. *J Pharm Toxicol* 2014;9:55-61.
8. Marshall DM, Muhaidat R, Brown NJ, Liu Z, Stanley S, Griffiths H, et al. *Cleome*, a genus closely related to *Arabidopsis*, contains species spanning a developmental progression from C(3) to C(4) photosynthesis. *Plant J* 2007;51:886-96.
9. Kori ML, Gaur K, Dixit VK. Investigation of immunomodulatory potential of *Cleome gynandra* Linn. *Asian J Pharm Clin Res* 2009;2:35-9.
10. Albarello N, Simões C, Rosas PF, Castro TC, Gianfaldoni MG, Callado CH, et al. In vitro propagation of *Cleome spinosa* (Capparaceae) using explants from nursery-grown seedlings and axenic plants. *Vitro Cell Dev Biol Plant* 2006;42:601-6.
11. Mishra SS, Moharana SK, Dash MR. Review on *Cleome gynandra*. *Int J Res Pharm Chem* 2011;1:681-9.
12. Chweya JA, Mnzava NA. *Cat's Whiskers, Cleome gynandra* L. Vol. 11. Rome, Italy: Bioversity International; 1997.
13. Anbazhagi T, Kadavul K, Suguna G, Petrus AJ. Studies on pharmacognostical and in vitro antioxidant potential of *Cleome gynandra* Linn. leaves. *Nat Prod Radian* 2009;8:151-7.
14. Paul SB, Choudhury S, Adhikari PP. Isolated chemical components from spider weed, *Cleome gynandra* Linn: An overview. *Assam Univ J Sci Technol* 2012;10:166-73.
15. Misra RN, Dutt S. Chemical examination of *Cleome pentaphylla* Linn. Part II. Constituents of the oil from the seeds. *Proc Natl Inst Sci India* 1937;3:45.
16. Gupta RK, Chandra S, Mahadevan B. Chemical examination of seeds of *Gynandropsis pentaphylla*. *Indian J Pharm* 1968;30:127.
17. Saleh M. Phytochemical and botanical study of *Gynandropsis* sp. Part-I. *Gynandropsis* glucosinolates. *Pharmazie* 1976;31:818.
18. Hasapis X, MacLeod AJ, Moreau M. Glucosinolates of nine cruciferae and two Capparaceae species. *Phytochemistry* 1981;20:2355-8.
19. Ali AA, Sayed HM, Mesbah MK. Pharmacognosical study of *Gynandropsis pentaphylla* growing in Egypt, Pt. III. *Bull Pharm Sci Assiut Univ* 1987;10:74.
20. Jain AC, Gupta SM. Minor phenolic components of the seeds of *Gynandropsis gynandra*. *J Nat Prod* 1985;48:332-3.
21. Rastogi RP, Mehrotra BN. *Cleome gynandra*, *Compendium of Indian Medicinal Plants*. Vol. II, III, IV. New Delhi: CSIR; 1993. p. 112, 182, 193, 198.
22. Ragunathan V, Jaswant V, Sulochana N. Rutin from flowers of *Cleome gynandra* Linn. *J Ind Chem Soc* 1997;74:821.
23. Das PC, Patra A, Mandal S, Mallick B, Das A, Chatterjee A. Cleogynol, a novel dammarane triterpenoid from *Cleome gynandra*. *J Nat Prod* 1999;62:616-8.
24. Lwande W, Ndakala A, Hassanali A, Moreka L, Nyandat E, Ndungu M, et al. *Gynandropsis gynandra* essential oil and its constituents as a stick (*Rhipicephalus appendiculatus*) repellent. *Phytochemistry* 1999;50:401-5.