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Investigation of Antimicrobial and Anti Proliferative Properties of Prostate Epithelial Cancer Cells Resistance to Centipeda Minima Parts

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Abstract:- Natural remedies are continually being developed and improved. The species Centipeda minima is responsible for the production of a variety of chemical substances (C.minima). The phytochemical profile of Centida minima as well as its potential for yielding bioactive chemicals will be investigated in this study. According to the findings of the research, plenolin, a bioactive component found in plants, possesses both antibacterial and anticancer properties.

Keywords:- *Antimicrobial action, Prostate Cancer Cells (PC3), Centipeda minimum.*

I. INTRODUCTION

A Multi - protocol annual that thrives in damp conditions. Antibiotic overuse by humans and animals in agriculture leads to bacterial resistance. In recent decades, there has been a rise in interest in antimicrobials generated from plants that have minimal toxicity to mammals. Investigating substances for their antibacterial and antiallergenic effects. The aerial portions of C. minima are used to cure a variety of conditions, including malaria, piles, headaches, and colds. Triterpenes and sesquiterpene lactones are found. Anti-allergic and antibacterial properties came in first class. In traditional Chinese medicine, it is employed as a means of combating nasopharyngeal cancer. It is uncertain the components of C. minima protect against nasopharyngeal cancer. Anticancer sesquiterpene lactones occur in composites. In HL-60 cells, 6-O-angeloylenolin, which is a C. minimum sesquiterpene lactone, increased the rate of apoptosis and decreased the growth of tumours.

An investigation was conducted to investigate the antibacterial and antiproliferative properties of Centipeda minima on prostate cancer cells (PC3).

II. MATERIALS AND METHODS

The identification of Centipeda minima was confirmed by taxonomists at the Dehradun botanical garden.

A. Extraction and Analysis

Flowers were freshly plucked, then washed, and then weighed (10 g each). In a total volume of 10 millilitres containing water, methanol, acetone, and benzene, the components were macerated for a period of six hours. Using sterile Whatmann filter paper No. 1, the mixture was filtered. centrifuged at 5,000 revolutions per minute for five minutes. The supernatants were collected in a beaker, and the solvents were allowed to evaporate. Following that step, the dried extracts were frozen. In order to dissolve the extracts, dimethyl sulfoxide (also known as DMSO) was utilised. The samples were processed by moving them through a column that included cotton, activated charcoal, and silica gel in the following proportions: 1:2:1. Therefore, the extracted chemical went through the column a number of times during the process.

B. Phytochemical Screening

A qualitative analysis was conducted on the metabolites found in flower and leaf extracts. The amount of alkaloids was determined by utilising several techniques. It was determined that sterols are present in the dried ethyl acetate extract by adding acetic anhydride to the mixture and then adding sulphuric acid. In order to determine the presence of phenolics, ferric chloride neutral was added to the extract. Tin and thionyl chloride were utilised in the process of terpene identification. Add 10 percent sodium hydroxide to detect flavones. To extract the tannins, 0.5 grammes of dried leaves and blossoms were simmered in 5 millilitres of water and then filtered. After adding ferric chloride to the filtrate, the mixture was analysed. Calculations of phospholipids and glycolipids were carried out in accordance with established methods. In order to get the fixed oils, petroleum ether and benzene extract were compressed in between two filter sheets.

C. HPLC Analysis

The conformity and purity of extracts of Centipeda minima (Root, Stem, Leaf, and Flower) that had been cleaned in a column were evaluated using HPLC mobile and stationary phases. After installing the gradient programme, peak analysis was determined by keeping an eye on the graph and analysing the chromatogram in light of previously collected information.

III. ANTI BACTERIAL ACTIVITY

A. Pour Plate Technique

A conical flask measuring 250 millilitres was loaded with sterile water and nutrients for the growth of microorganisms. A solution with a pH of 7.2 received 2 grammes of agar. The nutritional agar medium was autoclaved at 121 degrees Celsius for fifteen to twenty minutes at a pressure of fifteen pounds. The bacterial strains, including E. coli, P. aeruginosa, S. subtilis, and S. aureus, were obtained from in Dehradun. The loopful of culture obtained from the stock cultures was streaked on Petri plates using the streak plate method so that fine isolated colonies could be obtained. At a temperature of 37 degrees Celsius, petri dishes were kept warm for a full day. Pour plates allow for the growth of isolated colonies on the surface of the agar medium. The agar well diffusion method was utilised in order to examine the antibacterial activity of the pour plate. Inoculums obtained from the broth were placed on a plate made of sterile agar medium. Once more, the loop was subjected to flame sterilisation, and bacteria was poured into it. The bacteria were spread out across the plate by rotating it at a 90°C angle. As many times as required. The next step was incubation. The disc method was utilised in order to investigate whether or not the HPLC compound possessed any antibacterial properties. The culture development on the plates was hindered by the tested substances.

IV. ANTI CANCER ACTIVITY

A. Cell Lines and culture conditions:

The isolated compound's anticancer efficacy was assessed by cell viability and morphology. Human Prostate Aden carcinoma epithelial Cancer cell line (PC3) was grown in Dulbecco modified eagles medium. Bacterial and fungal pollutants were avoided when preparing culture medium. By adjusting the Trypsin/EDTA volume, we may make adherent and semi-adherent cell line suspensions. 100-200l of cells are extracted, counted, and resuspended in freezing medium to achieve excellent recovery after freezing.

B. Cryo Presevation of Cell line

Evaluation of cell density as well as bacterial and fungal contamination was carried out with the use of an inverted microscope. Trypsin/EDTA was used to suspend adherent and semi-adherent cells, and then those cells were resuspended in trypsin. Utilizing suspension cell lines on a direct level.

C. Cell Treatment

Effects against cancer of components isolated from the Centipeda minima column. These compounds were manufactured as 10mM DMSO stock solutions and stored at 40C throughout the production process. Each cell is exposed to the medication for a period of forty-eight hours. The medium used to culture the control cells do not contain DMSO.

V. RESULT AND DISCUSSION

Qualitative Test	*Centipeda minima*
Terpenes	+ ++
Fixed Oils	+
Flavones	+++
Alkaloids	+ +

Table 1: Examination of the phytochemical profile of Centipeda minima

+: Low, ++: Medium & +++: High concentration

Terpenes can be identified by the pink colour of the crude extract that remains following treatment with tin and thionyl chloride. When the crude extract becomes orange after being treated with 10 percent NaoH, this indicates the presence of flavones. The presence of fixed oil can be determined using benzene extract that has been dyed with petroleum ether. When treated with Mayer's reagent, a colour that resembles cream suggests the presence of an alkaloid (Table 1).

A. HPLC Analysis

Pleneolin was the chemical that was cleaned by the column (Figure 1), and its max height was 6.5871. Evaluation of Plenolin's antibacterial properties

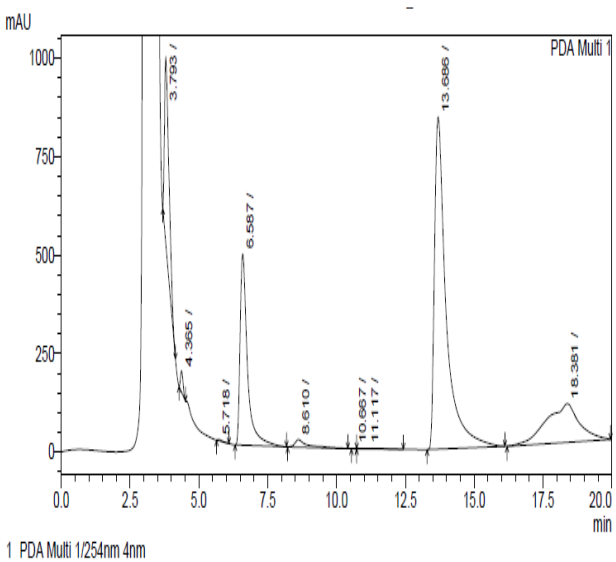


Fig. 1: HPLC analysis of Centipeda minima extracts reveals the active component as peaks in the spectrum

B. Anti-Bacterial Activity

Flower extracts of C. minima had a greater antibacterial effect than leaf extracts. We extracted the substance using acetone, methanol, and water. The flower and leaf extracts of C. minima displayed the greatest inhibition against B. subtillis, with an inhibition zone of between 6.9 and 9.2 millimetres. Column extracts demonstrated a zone of inhibition that was between 3.3 and 6.6 millimetres smaller than crude extracts against all of the bacteria that were tested. The C. minima column and crude extracts have an

inhibitory effect on E. coli. Additionally, both P. aeruginosa and E. fecalis showed sensitivity to crude and column extracts of C. minima.

Microbes	Zone of Inhibition in mm Mean ± SD	
	Crude	Column
E.coli	7.6±0.6	4.4±0.6
S.aureus	7.7±0.8	4.5±0.2
B.subtilis	7.2±1.2	5.8±0.8
P.aerugens	8.4±0.4	5.6±0.8

Table 2: Evaluation of the antibacterial activity of crude and column extracts of Centipeda minima against various pathogenic microorganisms

These bacteria are able to digest plenolin, which is produced by Centipeda minima.

P.aeruginosa>B.subtilis>S.aureus>E.coli (Table 2). According to phytochemical research, C. minima contains more than ten different forms of the sesquiterpene lactones known as pseudo-guanianolide or guaianolide. Sesquiterpenoids are anti-PAF and antibacterial kill Bacillus and Sreptococcus. Flavonoids have the potential to have a bioactive effect. Active components of this plant include something called sesquiterpenes. This research is essential to the development of plant-based antimicrobials since bacteria are getting resistant to the antibiotics that are now in use.

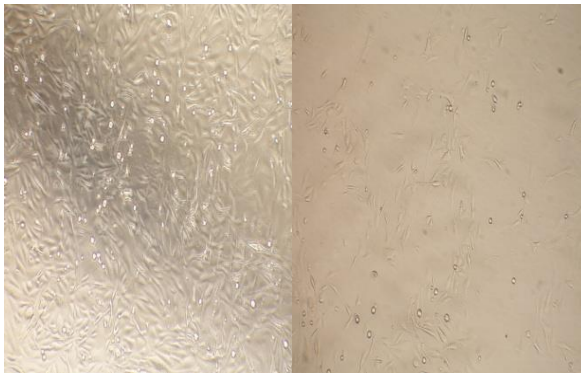
C. Anti-Cancer Properties:

S.No	Sample	Solvents	Cell Death
1	Control		100%
2	Root	Water	8.49%
3	Stem	Water	7.86%
4	Leaf	Acetone	91.18%
5	Flower	Water	7.76%

Table 3: Prostate cancer cell (PC3) death percentage of Centipeda minimum

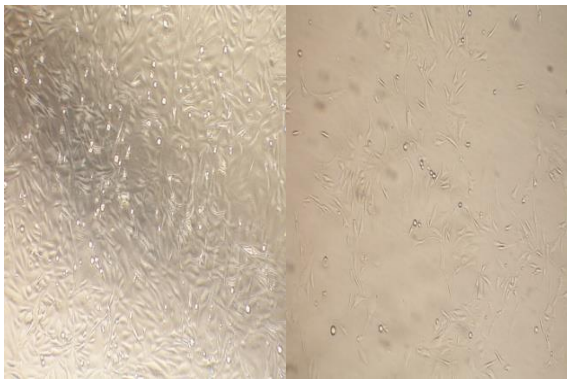
2. (a) 2.(b)

Fig: 2: In PC3 cells, the control was compared with a leaf extract that included acetone



3. (a) 3. (b)

Fig. 3: In PC3 cells, the control group was compared to the stem in water



4. (a) 4.(b)

Fig. 4: In PC3 cells, the control group was compared to the root in water

VI. CONCLUSION

The anticancer effects of leaf extract in acetone are more potent than those of stem, flower, and root extract in water. One might attribute the reduction in the total tumour incidence to the ability of these medications to suppress carcinogenesis and, as a consequence, the formation of subsequent tumours. I. viscosa leaf extract causes similar cytogenetic abnormalities to develop in A. cepa root tips. These abnormalities include cytoplasmic shrinkage, nuclear condensation, DNA breaks, membrane blebbing, cytoskeleton alterations, and the production of apoptotic bodies. These findings have the potential to contribute to the development of new therapeutic treatments.

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