



## Anticancer properties of *Cissus quadrangularis*

Aayush Dwivedi<sup>1</sup>, I. Seethalakshmi<sup>2</sup> and D. Sharmila<sup>1\*</sup>

<sup>1</sup>Department of Industrial Biotechnology, Bharath University, Selaiyur, Chennai

<sup>2</sup>Life Tech Research Institute, Vadapalani, Chennai

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### ABSTRACT

Anticancer activity of ethanol and chloroform extract were screened by MTT assay in which IC<sub>50</sub> concentration of plant extract was found as 62.5µg/ml against HeLa cell line and 125µg/ml in Vero cell line. Cell death in HeLa cell line was studied and confirmed as apoptosis by DNA fragmentation experiments.

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### INTRODUCTION

Plant extracts are useful sources of new medicines, thereby finding applications in the pharmaceutical industry. The herbal products today symbolize safety in contrast to the synthetics that are regarded as unsafe to human and environment. Although herbs had been priced for their medicinal, flavouring and aromatic qualities for centuries, the synthetic products of the modern age surpassed their importance [1]. The *Cissus quadrangularis* stem is also reported to contain a water-soluble glycoside, which produces a fall in blood pressure in anaesthetized cats. Fresh stems of *Cissus quadrangularis* produces irritating action on the skin, which may be attributed to the presence of calcium oxalate and 31 methyl tritriacontanoic acid along with taraxerylacetate, taraxerol and iso-pentacosanoic acid [2].

*Cissus quadrangularis* found to contain vitamins and steroids, which are found to have specific effect on bone fracture healing. The anabolic steroidal principles from *Cissus quadrangularis* marked influence in the rate of fracture healing by influencing early regeneration of all connective tissues involved in the healing and quicker mineralization of callus [3].

*Cissus quadrangularis* stem is traditionally used for the treatment of gastritis, bone fractures, skin infections, constipations, eye diseases, piles, anemia, asthma, irregular menstruation, burns and wounds. The leaves and young shoots are powerful alternatives powder is administered in treatment of hemorrhoids and certain bowel infections. The juice of stem is useful in scurvy and in irregular menstruation whereas the stem paste boiled in lime water is given in asthma. It is also used as a powerful stomachic. *Cissus quadrangularis* has potent fracture healing property and antimicrobial, antiulcer, antioxidative, antiosteoporotic, gastroprotective, cholinergic activity as well as beneficial effects on cardiovascular diseases [4, 5]. *Cissus quadrangularis* plant contains a high percentage of calcium ions (4% by weight) and phosphorous. Recently a study has been undertaken which showed that the plant extract when reacted with CO<sub>2</sub>, leads to formation of calcite crystals of highly irregular morphology, indicating that bioorganic molecules present in the extract modulate the crystal morphology [6]. The present study was carried out to study the anticancer effect of *Cissus quadrangularis*.

### EXPERIMENTAL SECTION

The anticancer activity of *Cissus quadrangularis* was assayed using cell lines. HeLa cell line was obtained from National centre for cell sciences Pune (NCCS). The cells were maintained in Minimal Essential Media supplemented with 10% FBS, penicillin (100 U/ ml) and streptomycin (100 µg/ ml) in a humidified atmosphere of 50 µg/ ml CO<sub>2</sub> at 37°C. The subculturing was done using TVPG (2 % trypsin, 0.2 % EDTA and 10 % glucose). The cells were occasionally checked for cytotoxicity using an inverted microscope after the MTT assay [3-(4, 5-dimethyl thiazol-2yl)-2, 5-diphenyl tetrazolium bromide] and the cell viability in percentage was calculated using the formula (Cell viability (%) = Mean OD/Control OD x 100).

The DNA of the treated cells were isolated and separated by electrophoresis to study the anticancer effect by DNA fragmentation.

### RESULTS AND DISCUSSION

The present study was carried out to find out the anticancer properties of *Cissus quadrangularis*. Plant sample were collected and extracts of ethanol and chloroform were prepared. Solvents are used to isolate the bioactive compounds and the most commonly used solvent is ethanol. The use of non polar solvent completely indicates that active compound for solvent dissolves in polar solvent only. MTT is a yellow tetrazolium salt that is converted into a blue formazan by dehydrogenases of a living cell. The assay is based on the principle that the amount of formazan produced is directly proportional to the number of live cells. The MTT assay was performed for 48 hrs. The results obtained by MTT assay for HeLa cancer cell line (Figure-3 and Figure-4) was compared to the result of the cytotoxicity activity of the extract on Vero cell line (Figure-1 and Figure-2). The IC<sub>50</sub> value was found to be at the concentration of 62.5 µg/ml and 125µg/ml for HeLa and Vero cell line respectively.

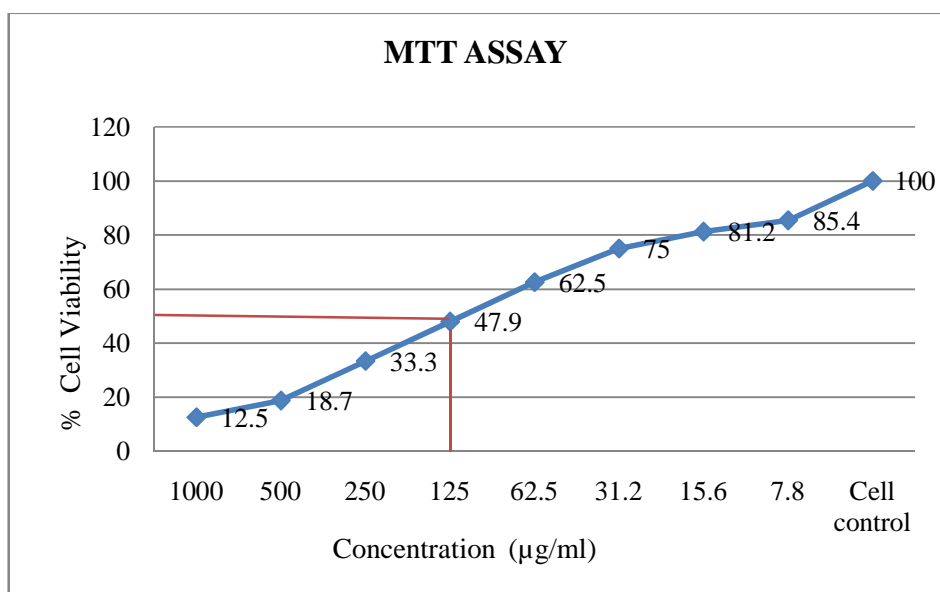


Figure-1 Cytotoxicity effect of ethanol extract of *Cissus quadrangularis* on Vero cell line

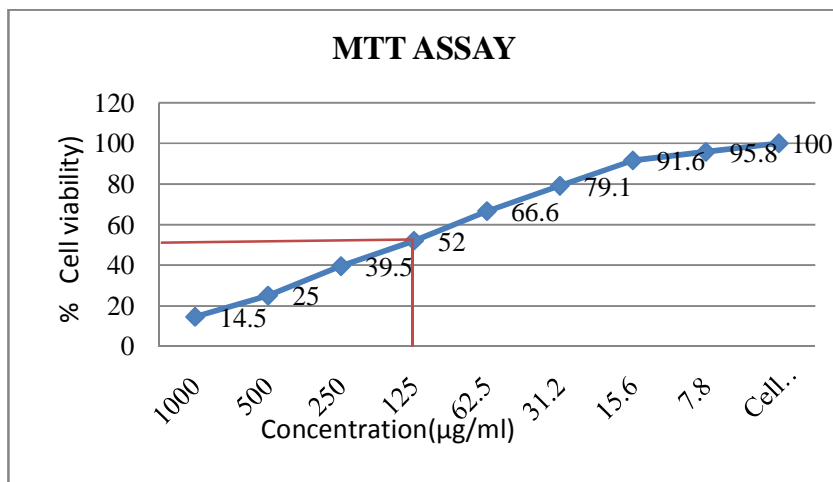


Figure -2 Cytotoxicity effect of chloroform extract of *Cissus quadrangularis* on VERO cell line

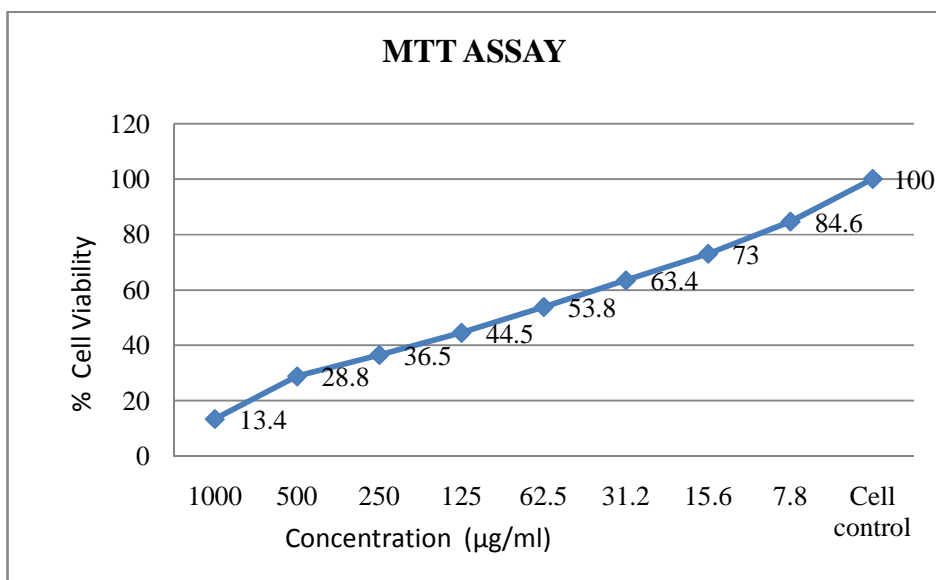


Figure-3 Anticancer effect of ethanol extract of *Cissus quadrangularis* on HeLa cell line

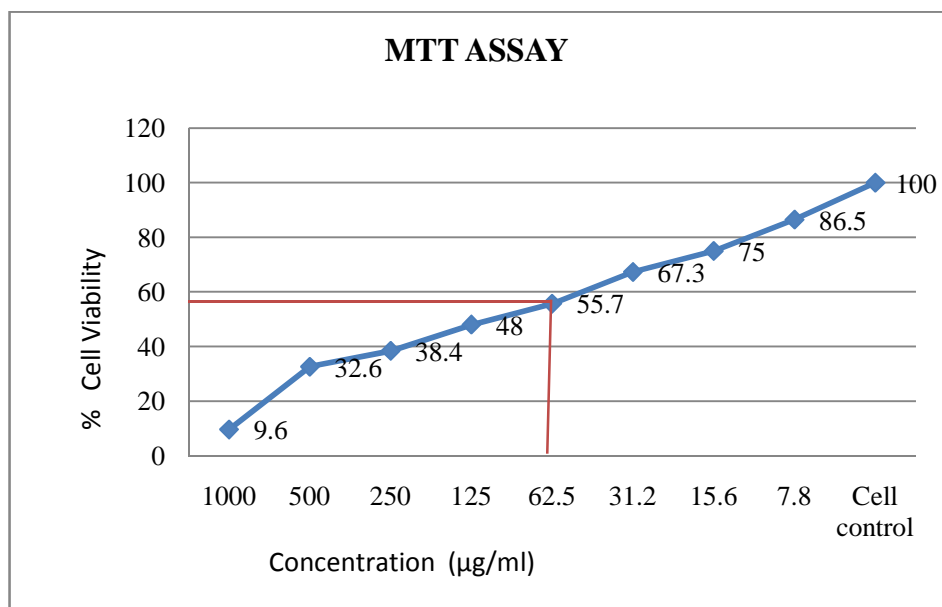


Figure-4 Anticancer effect of chloroform extract of *Cissus quadrangularis* on HeLa cell line

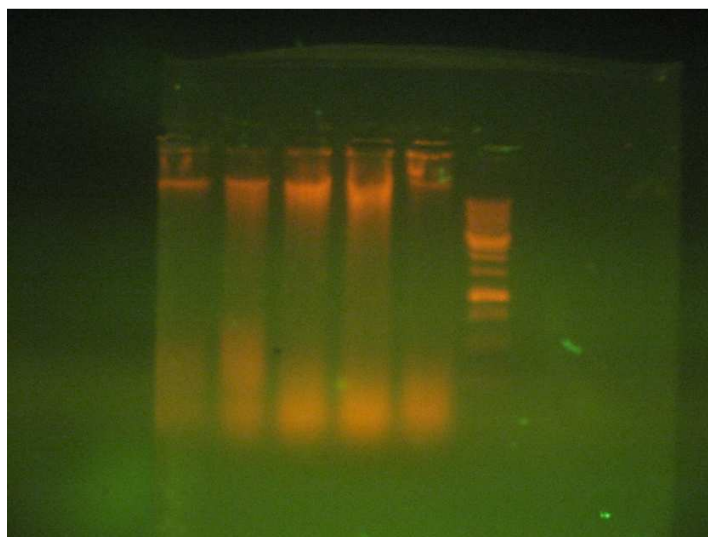


Figure-5 DNA fragmentation

The biochemical mark of apoptosis is the cleavage of chromatin into nucleosomal fragmentation. The cell suspension was centrifuged and the pellet was resuspended and used for the DNA fragmentation study. The DNA was isolated from the treated cell lines, concentrated and 31.2 µg/ml, 15.6 µg/ml and 7.8 µg/ml were loaded to the wells. The electrophoresis was carried out on agarose gel. Figure-5 shows the bands of DNA fragmentation due to the process of apoptosis.

### CONCLUSION

The present study has shown that the ethanol and methanol extracts of *Cissus quadrangularis* had anticancer activity. The IC50 value was found to be at the concentration of 62.5 µg/ml and 125 µg/ml for HeLa and Vero cell line respectively.

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