Evaluation of natural substances / synthetic compounds is the key step in innovative development of new drugs. Historically, evaluation of substances has been done by testing them on laboratory animals for various pharmacological actions and these are termed as *in vivo* tests. As the research towards discovery of new drugs is intensified, a large number of animals have been used in evaluation. After 1970, slowly the alternative methods of evaluation without use of whole animals, have been developed. These are termed as *in vitro* tests. *In vitro* evaluation methods also offer to the understanding of mechanism of action a cellular level. The *in vitro* evaluations involve the usage of enzymes, receptors, cell lines and protein-protein interactions and antigen-antibody interactions as the targets.

*Gymnema Sylvestre* is a known anti-diabetic plant and this activity has been studied extensively by various scientists. *Gymnema Sylvestre* is reported to contain a number of oleanane type of triterpenoids and their glycosides called Gymnemic acids. These Gymnemic acids contain a number of free hydroxyl groups. It is established fact that the polyhydroxy compounds possess the anti-oxidant activity. *Gymnema Sylvestre* has not been systematically investigated for anti-cancer activities and anti-oxidant activities. In our investigation *Gymnema Sylvestre* plant is evaluated *in vitro*
initially for anti-oxidant activity. Subsequently, on obtaining positive results the plant was evaluated for anti-cancer activity.

Alcohol, ethyl acetate and chloroform extracts of *Gymnema Sylvestre* were tested for DPPH, superoxide, nitric oxide radical scavenging activity and reducing power. In all these tests, all the three extracts have shown considerable anti-oxidant activity comparable to the standard compound BHT. Among the three extracts, the alcohol extract has shown higher activity.

Anti-cancer activity of the three extracts was evaluated in MCF7 and A549 cell lines by MTT Assay. All the three extracts have exhibited significant activity in MCF7 cells. The activity in A549 cells was not prominent as compared to MCF7 cells. Among the three extracts the activity of alcohol extract was found to be higher.

Further, investigations were in the direction of isolation of the compounds and test them for anti-cancer activity. The alcohol extract was column chromatographed and isolated two compounds which were identified as Gymnemagenin deacetylGymnemic acid (dAGA). These two compounds were evaluated for anti-cancer activity in MCF7 cell lines and the results shown that both the compounds are active. However, the activity of dAGA was better than Gymnemagenin.
The d AGA was also tested on normal cells (HEK 293- Human Embryonic Kidney Cells) and found that it does not have any significant cytotoxic activity.

To know the mechanism of anti-cancer activity of dAGA at cellular level, various flowcytometric experiments such as Annexin-V assay, mitochondrial membrane potential determination, cell cycle analysis, caspase activity and western blot technique experiments such as cytochrome C and Bax / Bcl-2 ratio estimations were conducted.

In Annexin V Assay, it was observed that there is a significant increase in the apoptotic cells as compared to the control cells. The cell cycle analysis results have shown that dAGA has caused cell cycle arrest at sub G0 /G1 phase. Mitochondrial potential determination also has shown significant decrease in the potential indicating the induction of apoptosis. Multi caspase Assay revealed that there is considerable increase in the caspase activity which consequently led to the increase in the cytochrome C levels in cytosol. In Bcl-2 / Bax ratio determination, the dAGA has caused the down regulation of Bcl-2 indicating the induction of apoptotic pathway.

Based on the findings in this study one of the major compounds of Gymnema sylvestre - deacetyl Gymnemic acid proved to be potent anticancer compound. Mechanism of anticancer activity appears to be via the activation of apoptotic pathways. This study prompts the next stage of investigations for drug development.