SUMMARY

*Brassica oleraceae* was extracted using methylene chloride by maceration. The crude extract is assayed for its anticancer activity by MTT assay. Crude extract was found to possess marked growth inhibitory activity against HEp-2 cells in comparison to normal vero cells. The crude extract of *Brassica oleraceae* was purified using solid phase C₁₈ silica cartridge. Three different fractions were collected and further purified by enrichment. Fraction 3 was found to contain sulforaphane and the amount was quantified using external standard. Purified extract found to contain 75µg/ml of sulforaphane. The different fractions (fraction 1, fraction 2 and fraction 3) were assayed for its anticancer activity using HEp-2 and Vero cells. Fraction 3 was found to possess significant anticancer activity in HEp-2 cells when compared to other fractions. Results revealed that apart from its cytotoxic effect the purified extract of *Brassica oleraceae* could induce apoptosis in HEp-2 cells. Cell plots revealed that purified extract treated cells have 65.34 apoptotic cells lower than that of 15.1 in untreated cells. The increase in percentage of apoptotic cells proves the induction of apoptosis. The block in the transition of treated cells from S phase to G₂-M phase further prevents the active proliferation of malignant cells.

The up regulation of p53, bax and casp-3 and down regulation of bcl-2 indicates that the induction of apoptosis is mediated through p53 dependant signalling apoptotic pathway in HEp-2 cells. The increased expression of p53 induces a series of signalling events thus promote the conversion of caspases to its active form. Caspases thus brings about apoptosis by complete destruction of cancer cells. Our results revealed that the crude extract fraction was found to contain higher radical scavenging activity than compared to purified extract. The results also suggest that percentage scavenging effect found in HEp-2 cells were higher than that found in Vero cells. The results showed that crude extract exert a marked reduction in GST activity in HEp-2 cells whereas there is no marked reduction in the enzyme activity in purified extract treated cells. The elevated GSH as seen in untreated HEp-2 cells were decreased significantly after treatment with the crude extract fraction. There is no marked decrease in the GSH content in purified extract treated cells. Crude extract and Purified extract of *Brassica oleraceae* were found to possess potent antibacterial activity against (gram positive—*Bacillus cereus*, *Staphylococcus aureus*; gram negative—*Escherichia coli*, *Vibrio cholerae*, *Pseudomonas*
*aeruginosa* and antifungal activity against (*Candida albicans* and *Aspergillus flavus*) species by diffusion method. Crude extract (500 µg) was found to possess good equivalent antibacterial and antifungal activity against all the tested species in comparison with positive control (Streptomycin 10µg and Nystatin 10µg).

Structure activity relationship of sulforaphane with various apoptosis proteins such as p53, bax, bcl-2 and caspases were studied by docking studies using molecular docking server. The structure activity relationship of various apoptosis proteins such as p53, bax, bcl-2 and casp-3 with SFN proved their stability by studying various attractive forces involved in the binding. Molecular models were used to study the interactions of SFN with various apoptotic proteins. SFN was successfully docked to all the proteins and their binding energies were comparable with 5-fluorouracil (positive inducer of apoptosis). The results were appreciable and in agreement with our earlier research. The purified extract of *Brassica oleraceae* was found to have significant anticancer activity in HEp-2 cells and apoptosis is mediated through p53 signalling pathway. The extract was found to possess significant antioxidant and antimicrobial activities.