CHAPTER VI: SUMMARY AND CONCLUSION

6.1. Summary

Since ancient times plants are being used as a source of medicine. These medicinal properties are generally ascribed to the presence of few bioactive molecules and not the whole chemical component present in the plant. Bioactivity guided fractionation is one technique which is used in the separation of the bioactive principles. In this research, two very important medicinal plants i.e. *Cephalotaxus griffithii* and *Oroxylum indicum* have been taken up for a systematic study to identify the active therapeutic molecule with special reference to cancer. However, we also evaluated two other therapeutic properties of these plants viz. antioxidant and antimicrobial property.

*Cephalotaxus griffithii* needle extract inhibited the proliferation of HeLa, HepG2 and ZR751 cells with maximum growth inhibition shown by *Cephalotaxus griffithii* needle petroleum ether extract. Mechanism study of ZR751 cell death suggests that the extract induced cell death was associated with apoptosis (mitochondria and death receptor mediated pathway) induction, cell cycle arrest and suppression of telomerase expression. Functional analysis study of p53 using siRNA suggests that p53 is an essential target for CGNP extract. Further, fractionation of CGNP produced six fractions of which fraction 6 inhibited the maximum antiproliferation on cancer cells. Again, fractionation of fraction 6 lead to the isolation of three compounds (impure), two of them showed very high antiproliferative effect on cancer cells.

*Cephalotaxus griffithii* needle essential oil exhibited the antiproliferative effect on human cervical cancer (HeLa, ME-180 and SiHa) cells. CGNO also slowed down or
inhibited the motility of HeLa cells to close a scratched wound and also reduced the HeLa cell migration towards a chemotactic stimulus. It was revealed that the HCC cell death was associated with apoptosis (mitochondria and death receptor mediated pathway). These multiple chemotherapeutic potential may be attributed to the presence of great number of chemical constituents in CGNO.

*Ce*phalotaxus* g*riffithii* bark extract possessed free radical scavenging and reducing power ability. The highest antioxidant activity was shown by *Ce*phalotaxus* g*riffithii* bark acetone extract. CGB extracts inhibited the growth of three bacterial pathogens, namely *Klebsiella pneumoniae*, *Escherichia coli*, and *Staphylococcus aureus*, out of the six organisms tested. The maximum antibacterial activity was shown by CGBA extract. Again, CGBA extract induced the maximum cytotoxicity and apoptosis of cervical cancer cells. Correlation analysis suggested that phenolic and flavonoid content present in stem bark of *C. griffithii* extracts was responsible for the high antioxidant, cytotoxic, and apoptotic activity.

*Oroxylum indicum* bark (OIB) extract exhibited free radical scavenging and reducing power ability. The highest antioxidant activity was found in OIB methanol (OIBM) extract. Further, OIBM extract displayed a broad antibacterial spectrum as well as antifungal properties. In contrast, OIB petroleum ether (OIBP) extract inhibited maximum proliferation of HeLa cells. The cell death was associated with apoptosis. Further, fractionation of OIBP extract produced five fractions of which fraction 5 exhibited the highest antiproliferative activity on cancer cells (HeLa, HepG2 and ZR751) cells. From fraction 5, a compound named oroxylin A was isolated. However, oroxylin A did not show better antiproliferative activity than fraction 5 or OIBP. Overall, this suggests that components other than oroxylin A were mainly responsible
for the anti-neoplastic effect induced by fraction 5. But, compared to cancer cells, the normal human fibroblast was less sensitive to the cytotoxic effect of oroxylin A. Oroxylin A was 2.3 times more active in killing ZR751 cells, and 1.7 times more active in killing HeLa cells than normal human fibroblast indicating the ability of oroxylin A to induce selective cytotoxicity towards cancer cells.

6.2. Conclusion

Both *Cephalotaxus griffithii* and *Oroxylum indicum* possessed antioxidant, antimicrobial and anticancer property. For CGN, the compounds (CGNC2 and CGNC3), however impure, isolated from fraction 6 of CGNP extract (cytotoxicity guided isolation) was mainly responsible for the anti-neoplastic property. Moreover, the essential oil isolated from CGN did possess anticancer property. Further, for CGB, the active anti-neoplastic principle lies in the CGBA extract and assumed to be phenolic components. For OIB, oroxylin A was isolated from fraction 5 of OIBP extract, but it did not turn out to be a main active anti-neoplastic molecule suggesting components other than oroxylin A was mainly responsible for the anti-neoplastic effect induced by fraction 5. However, oroxylin A was 2.3 times more active in killing ZR751 cells, and 1.7 times more active in killing HeLa cells than normal human fibroblast indicating the ability of oroxylin A to induce selective cytotoxicity towards cancer cells. This research now initiated and paved the way for a systematic approach using cytotoxicity guided isolation of anti-neoplastic molecule from *Cephalotaxus griffithii* and *Oroxylum indicum*. Further works are encouraged.