Summary

The present study has explored the effect of magainin II in comparison with 5-FU on colon cancer cell lines.

- The cell viability and IC\textsubscript{50} concentration determined by MTT assay for 5-FU were found to be 32.8\,\mu\text{M}, 26.52 \, \mu\text{M}, 24.6\mu\text{M}, 75.21 \, \mu\text{M}, 18.94 \, \mu\text{M} for HT-29, COLO 205, COLO 320 DM, HCT-15 and HCT 116 respectively. The IC\textsubscript{50} concentration of magainin II for HT-29, COLO 205, COLO 320 DM, HCT-15 and HCT 116 were found to be 293.9 nM, 147.9 nM, 89.34 nM, 386.2 nM and 76.72 nM respectively.

- An increased release of LDH and reduction in cell viability was observed in IC\textsubscript{50} magainin II treated colon cancer cell lines.

- Less toxicity towards normal cells (Vero cell line and MDCK cell line) were observed on treatment with magainin II.

- The qualitative measurement (Hoechst 33342 and DAPI staining) by inverted phase contrast fluorescent microscope showed apoptotic changes in cell and nuclear morphology on treatment with IC\textsubscript{50} magainin II on colon cancer cells.

- DNA laddering was observed in IC\textsubscript{50} magainin II treated colon cancer cell lines.

- The qualitative measurement (Acridine orange/ethidium bromide staining) distinguished early apoptosis from late apoptosis on magainin II treated colon cancer cell lines.

- Similar results were observed quantitatively using Annexin V/PI by FACS.

- The IC\textsubscript{50} magainin II induces cell cycle arrest at G1 phase in colon cancer cell lines by FACS.

- The IC\textsubscript{50} magainin II treated HCT116 cell line showed increased ROS release when compared with other colon cancer cell lines.
The increased caspase-3 activity was observed in IC$_{50}$ of magainin II treated colon cancer cell lines.

The colon cancer cell lines showed the alteration in mitochondrial membrane potential on treatment with IC$_{50}$ magainin II.

Semi quantitative RT-PCR showed increased mRNA expression of FasL, p53, Bax and decreased Bcl-2 mRNA expression in IC$_{50}$ magainin II treated wild p53 colon cancer cell line. Mutant p53 colon cancer cell lines showed increased mRNA expression of p53, Bax, decreased Bcl-2 mRNA expression and no changes in FasL mRNA expression on treatment with IC$_{50}$ magainin II.

The up-regulation of Bax, p53, down-regulation of Bcl-2 and increased release of cytochrome-C proteins were observed in IC$_{50}$ magainin II treated colon cancer cell lines.

Magainin II reduced telomerase activity in mutant and wild p53 colon cancer cell line.

The results of the present study, has shown that magainin II is efficacious than 5-FU, by exhibiting significant cytotoxicity in wild p53 colon cancer cell lines than mutant p53 colon cancer cell lines. This study also revealed that the mechanism of action of magainin II is different between wild and mutant p53 colon cancer cell lines. The molecular mechanism is regulated initially via extrinsic pathway by stimulation of Fas through p53 mediated caspase activation, followed by secondary activation of intrinsic pathway, as evident by suppression and activation of Bcl-2 and Bax expression respectively. It also increases the ROS generation, decreases telomerase activity, causes alteration of the mitochondrial membrane potential, increases the release of cytochrome-C into cytosol with subsequent activation of caspase-3 and leading to apoptosis. The knowledge of the pathway of magainin II induced apoptosis
as anti cancer agents, could be used to tailor better therapies for colon cancer treatment. The *in vitro* antitumor activity of magainin II in colon cancer is promising but *in vivo* studies are required to investigate magainin II as an efficient agent against colon cancer.