2. ABSTRACT OF THE THESIS

Breast cancer is one of the most common cancer throughout the world with average death of 0.45 million each year. The rate of breast cancer is increasing rapidly in both the developed and developing countries. In spite of advancement in the treatment of breast cancer, the results to date remain unsatisfactory, prompting a need for better understanding the mechanism of tumor growth as well as to identify natural agents that could target cancer efficiently with least side effects. Osteopontin (OPN) is a secreted, non collagenous, sialic acid rich, chemokine like matrix-associated phosphoglycoprotein. OPN is a member of SIBLING (Small Integrin Binding LIgand N-linked Glycoprotein) family which is associated with the metastatic and oncogenic potential of various cancers. Andrographolide (Andro), a natural diterpenoid lactone isolated from *Andrographis paniculata* has been shown to possess inhibitory effect on cancer cell growth.

Here, for the first time we report that Andro inhibits breast cancer cell migration, arrests cell cycle at G2/M phase and induces apoptosis through caspase independent pathway. Our experimental evidences suggest that Andro attenuates endothelial cell motility and breast tumor-endothelial cell interactions. Furthermore, we have demonstrated that Andro inhibits OPN expression both at RNA and protein level; and suppresses NF-κB and AP1 activation in breast cancer cells. We further report for the first time that Andro curbs breast tumor growth in orthotopic NOD/SCID mice model, and this anti-tumor activity was correlated with down regulation of PI3-Kinase/Akt activation and inhibition of c-Jun, OPN and VEGF expressions. Collectively, these results demonstrate Andro as an effective anti-tumor agent in orthotopic mice model.

Current development in cancer research is focused on understanding the complex crosstalk between tumor and stromal microenvironment. The role of host derived factors in cancer progression has been considered as one of the least studied area in cancer biology. Researchers from various schools of thoughts indicated that stroma-derived OPN plays crucial role in fostering preneoplastic cell growth, as well as regulating stem cell growth. OPN a metastasis associated protein plays important role in regulation of various physiological and pathophysiological processes, but the influence of host (stromal)-derived OPN in cancer progression remain unclear. In this part of study, using
an isograft melanoma model, in wild type (OPN+/+) and OPN knockout (OPN−/−) mice, we have demonstrated that deficiency of host OPN effectively curbs melanoma growth, angiogenesis and metastasis. Melanoma cells isolated from OPN+/+ mice exhibit elevated tumorigenic feature as compared to the parental cell line or cells isolated from the tumors of OPN−/− mice. This study highlighted the crucial role of host OPN in melanoma progression. Furthermore, we have demonstrated that host OPN induced profound changes in murine melanoma cells resulting in enhanced melanoma growth, metastases and angiogenesis. This profound change is associated with the upregulation of ABCG2 expression and enrichment of SP phenotype. We report for the first time that stroma-derived OPN regulates SP phenotype in murine melanoma cells. Moreover, loss and gain of function studies demonstrated that stroma-derived OPN regulates SP phenotype specifically through ERK2 in murine melanoma cells. Our experimental findings demonstrated, at least in part, that the molecular mechanism underlying host OPN-regulated tumor growth and this observation may depict the prognostic significance of host OPN in tumor progression, and such knowledge might be helpful for the development of novel OPN-targeted therapy for the management of cancer.