Design and Synthesis of Novel α-Galactosylceramide Analogs as Immunomodulators and Development of Synthetic Peptide Vaccine

Chapter 1: In this section a brief review over the immunology, its cellular components and their effctor functions to fight against invasive pathogens has been presented. Immunology is a vast subject which cannot be covered entirely in this small section. The more details can be obtained from the concerned references mentioned meanwhile. The basic concepts of immunology presented in this section are very much helpful understand the immune response associated with α- alactosylceramide which we are working on. The next section deals with NKT cell biology and its ligands in detail.

Chapter 2: In conclusion chapter deals with the detailed discussion about design and synthesis of a focused library of cholesterol attached α-GalCer analog. Syntheses were achieved applying several appropriate schemes mentioned earlier and purified compounds were tested in-vitro on mouse splenocytes. Estimation of cytokines IFN-Y & IL-4 revealed that compounds such as PZ1 had shown Th2 biased response, PZ2 and DO1 shown Th1 biased response while rest other compounds were moderately producing both Th1/Th2 cytokines which can be useful in many disease treatment.

Chapter 3: In Chapter 3, a detailed description about design and syntheses of focused library of ten novel α-GalCer analogs with sugar swapped to secondary hydroxyl group is presented. All the compounds were subjected to in vitro stimulation of mouse splenocytes and their immunomodulatory activities were estimated. Cytokines IL-12, IL-4 & IFN-γ after 48 h and kinetic release studies demonstrated that most of the compounds were found to exhibit a Th1 biased immune response. Th1 biased analogs can be useful against intracellular pathogens and many diseases associated with them. Further SZ subseries compounds expressed superior immunomodulatory behaviour than SE analogs.

Chapter 4: In conclusion we have presented here the synthesis and preliminary immunological studies of a focused library of novel phenylalkyl substituted triazolyl α-GalCer analogues useful as immunomodulators. The bioassays disclose that T1204B, T1206B showed significantly improved Th1 as well as Th2 cytokine expressions. Also the studies revealed that the compounds are following all the unique traits of α- galactosylceramide. Balanced Th1/Th2 activation makes these molecules promising leads for their possible application as vaccine adjuvants and ligands to subdue autoimmunity.

Chapter 5: Here we presented the design of a peptide based three component synthetic cancer vaccine bearing three arms as Th epitope, B cell epitope and and Pam2Cys adjuvant (figure 5.18). All the epitopes were basically peptide fragments in nature thus the preparation was accomplished by solid phase peptide synthesis on Wang resin. The major portion of synthesis was performed manually on a Merrifield rector and partly on automated peptide synthesiser equipped with UV-detection facility. Standard protocols for various steps including coupling, capping, Fmoc deprotection and cleavage were followed as mentioned in several literatures. The synthesis was preceded with continuous monitoring of substitution value measured from UVabsorbance at 290 nm on Fmoc deprotection. The formation of peptide was also governed by Mass analysis in between at various stages during entire peptide syntheses. Final analysis of the vaccine construct was done with MALDI which revealed that final peptide was of poor quality containing mixture of several peptides.
To sum-up, effort has been made to construct the peptide based cancer vaccine by employing a novel design, following systematic solid phase peptide synthesis protocols. Even though the quality of final peptide conjugate was not suitable for immunological evaluations, the overall study gave insights into the various possibilities and difficulties associated with the development of such complex peptide conjugate based wholly synthetic vaccines.