CHAPTER VI
CONCLUSIONS

The present work demonstrated the design and synthesis of multifunctional chitosan under the key concept of water-based reaction system and mild condition. The first work, multifunctional chitosan was successfully synthesized in a one step conjugating reaction via CS-HOBt in water soluble system. Based on the introduction of Phenylalanine (hydrophobic group) and poly(ethylene glycol)methyl ether (hydrophilic group) to chitosan, the self-assembly as chitosan nanoparticles were formed in the size range ~20-50 nm. The entrapment of allergen, *Dermatophagoides pteronyssinus*, from chitosan nanoparticles was successfully prepared via electrostatic interaction and/or like-dissolve-like hydrophobic interaction. The in vitro preclinical implementation of allergen-entrapped CS-Phe-mPEG nanoparticles on peripheral blood mononuclear cells (PBMCs) from healthy- and allergic volunteers exhibited the immunogenicity, immune modulation, and the survival of the PBMCs from allergen. To propose the potential use of these chitosan nanoparticles in allergen delivery system, *in vitro* release studies were conducted. The different pHs of buffers affect the release profiles of allergen from allergen-entrapped chitosan nanoparticles (Chapter III).

Another strategy to produce multifunctional chitosan was the hybridization between chitosan derivative and inorganic nanoparticles, gold nanoparticles, via Cu-free Click Chemistry. In Chapter IV, we studied the possibility to form cycloaddition of chitosan-oxanorbornadiene, new type of chitosan derivative, with various kinds of azido-modified substrates without metal catalysts and purification steps. In addition, this success could be a new approach to introduce various types of functional groups, carboxylic-, disulfide-, silane- groups. In the last work, chitosan-oxanorbornadiene was successfully developed to be a water-soluble chitosan derivative by additional introduction of mPEG segments. The success of hybridization between water-soluble chitosan derivative and gold nanoparticles provided gold nanoparticle aggregation via Cu-free Click cycloadditon within ~ 0-2 minutes. After antibody was immobilized to water-soluble chitosan derivative, this system could be used in a promising naked-eye detection of antigens (Chapter V).