CHAPTER V

CONCLUSIONS

Liposomal powders can be prepared by the spray drying technique using mannitol as an additive. The spray drying process had no destructive effect on the stability of the major structural phospholipid. Addition of mannitol decreased aggregation of particles but could not preserve integrity of the liposome bilayer during the spray drying method. The HPC/mannitol weight ratio of 1:1 was an appropriate ratio to give fine liposomal powders. The reconstitution of the spray-dried HPC liposomal powders gave larger liposomes than the initial liposomes independent of HPC/mannitol ratios. Addition of glycine into the HPC liposomal formulation led to an increase in the yield of the spray-dried powders in the collector of spray dryer due to its anti-adherent property. However, the formulation with 10 %w/w glycine resulted in strong cohesion of particles which prevented formation of liposomes after reconstitution with HBS.

The lysozyme-loaded liposomal powders spontaneously formed liposomes and lysozyme was efficiently entrapped into the resultant liposomes after reconstitution with HBS. It could be speculated that after the powders reached the deep lung, mannitol would dissolve in the lung lining fluid and the liposomes would be subsequently formed, followed by entrapment of lysozyme into liposomal structure. Cholesterol incorporated into HPC lipid bilayer had an essential influence on liposome size and entrapment efficiency of lysozyme in the reconstituted liposomes. Temperature used for reconstitution of the powders also affected these properties. Loading of lysozyme and mannitol into liposomes by the DRV method before spray drying gave similar physicochemical properties of the powders and the reconstituted liposomes when compared to spray drying of preformed blank liposomes simultaneously with lysozyme solution. Thus, entrapment of the protein prior to spray drying seemed not to be necessary. The spray-dried liposomal powders with HSCP/Chol molar ratio of 8:2 and mannitol in the weight ratio of 1:1 was the most

suitable formulation for further optimization of spray drying condition factors to develop the lysozyme-loaded liposomal powders for possible pulmonary delivery. Although the spray drying process caused lysozyme aggregation, the biological activity of lysozyme remained unchanged after reconstitution of the powders. Therefore, the liposomal powder might be used as a carrier for lysozyme delivery to the lung for prevention and treatment of respiratory disorders (Cantor and Shteyngart, 2004, 2008).

Factorial design and response surface methodology could be successfully applied to study relationships among the spray drying variables and their effects on the properties of the spray-dried liposomal powders with lysozyme as a model protein. The spray drying conditions used in this study did not have a significant effect on moisture content of the spray-dried powders. The spray-drying process yield was mainly influenced by the inlet temperature followed by the pump speed and the total solid content. The particle size of the spray-dried powders was highly affected by the inlet temperature and the total solid content. The entrapment efficiency of lysozyme in the reconstituted liposomes was highly influenced by the inlet temperature and the pump speed. The optimum spray-drying condition selected for preparation of the spray-dried liposomal powders was the inlet temperature of 110 °C, the pump speed of 5 % and the total solid content of 2.975 %w/w. The powders prepared with this condition gave mean responses as follows: process yield of 60.46 %, mass median diameter of 6.00 μm and entrapment efficiency of 13.83 μg LSZ/mg lipid. In addition, the optimum formulation had appropriate theoretical aerodynamic diameter and tap density which might be considered the plausibility of using these particles for pulmonary drug delivery. However, it should be aware that the results of optimization in this present study might be applicable only to the spray drying of the liposomal powders using a lab-scale spray dryer apparatus.

Application of this Research

The scientific information obtained from this present study may be applied in the following aspects.

1. The results of formulation parameters can be used to further develop spray-dried liposomal powders suitable for pulmonary delivery of other protein/peptide drugs for local effect, for example, the delivery of subunit vaccines to induce cell-mediated responses for tuberculosis prevention.

2. The quadratic models obtained for yield, particle size and entrapment efficiency responses can be applied for predicting the influence of spray drying process conditions on these responses for development of dry liposomal powders of other protein/peptide drugs for aerosols.