CHAPTER V

CONCLUSION AND SUGGESTION

Immobilization of bimolecules on the surface of polycaprolactone (PCL) containing amino and carboxyl group was successfully accomplished through a three-step procedure. The first step involved the surface modification of PCL film *via* aminolysis of 1,6-hexamethylenediamine or graft copolymerization of acrylic acid to covalently introduce amino or carboxyl groups on PCL surface, respectively. Using ninhydrin and toluidine blue o assays together with UV-Vis spectroscopy, the density of amino and carboxyl groups on the modified film surface obtained under the optimized condition was estimated to be 6.30×10^{-8} mol/cm^2 and 9.10×10^{-8}, mol/cm^2 respectively. The second step was the activation of amino and carboxyl groups into active succinimidyl groups which were quite stable, yet reactive enough to interact with amino groups of biomolecules by using DSC and EDCI/NHS as a coupling agent, respectively. The last step was the coupling of selected biomolecules, collagen and chitosan. Data from ATR-FTIR, XPS and water contact angle analyses indicated that the PCL film became more hydrophilic after surface modification and the immobilization of biomolecules on the surface-modified PCL film was successful.

Results from *in vitro* cell studies suggested that the alteration of surface hydrophilicity and functionality has a significant impact on adhesion and proliferation of skin cells, keratinocyte (HEK001) and fibroblast (L929). PCL was considered as a fair substrate for cell adhesion and proliferation as compared to the positive control (100% of TCPS). Introducing hydrophilic amino or carboxyl group on PCL surface increased the cell adhesion and proliferation ratio of the virgin polymer film. The cytocompatability, in some cases, were further improved after the immobilization of biomolecules, especially collagen type I, a major component found in skin. The surface modification and biomolecule immobilization seem to have a more pronounced effect on the cytocompatibility of the modified PCL films towards L929 fibroblasts than HEK001 keratinocytes. From this investigation, the graft copolymerization seems to be a better route for surface modification of PCL film than

aminolysis, mainly because it can introduce a greater quantity of active species for subsequent immobilization of biomolecules. All above results have suggested that the chemical modification and biomolecule immobilization render PCL a potential candidate for artificial skin application.

Determination of cytocompatibility of materials based on the data from cell adhesion and proliferation are preliminary and somewhat ambiguous from biomaterial scientist's perspective. It can not be guaranteed that the adhered and proliferated cells will function regularly. More advanced studies which can provide information related to how cell functions are necessary. Thus, the future plan will cover an investigation of cell morphology as well as a determination of substance secreted from the cell that can verify its function (*i.e.* von Willebrand factor (vWF)). Immobilization of other biomolecules which is believed to enhance cytocompatibility more effectively such as RGD peptide, laminin is also a subject of future investigation.