

CHAPTER V

CONCLUSIONS AND RECOMMENDATION

In this work, polycaprolactone (PCL) was used as a scaffold; it was prepared by solvent casting technique. Chloroform and 40:60 (v/v) EtOH:THF were used as solvents to dissolve polycaprolactone and obtain different surface topology of films because of different solubility parameter. Polycaprolactone is a high hydrophobicity polymer, it is not suitable for cell growth; however, polycaprolactone can be improved to enhance hydrophilic surface via aminolysis by 1,6-hexamethylenediamine (HMD) to the ester group on PCL surface and activation of amino groups was reacted by *N,N'*-disuccinimidyl carbonate (DSC) as a coupling agent. After that proteins (BSA or collagen) were immobilized on the PCL surface to improve hydrophilicity of surface.

Surface topography of films was characterized by SEM and AFM. The results indicated that the film casted from chloroform had smooth surface and lower average roughness than film casted 40:60 (v/v) EtOH/THF. The protein adsorption test concluded that the film casted from 40:60 EtOH:THF could higher amount of protein adsorbed than the film casted from chloroform. Furthermore, at high protein concentration (3000 $\mu\text{g/mL}$), modified film casted from 40:60 EtOH:THF showed the highest protein adsorbed on surface. Because porous surface could more adsorb the amount of protein than smooth surface, modified surface can enhance the amount of protein adsorbed compared with the neat film. Surface wettability of 40:60 (v/v) EtOH:THF was the highest hydrophobic, but after surface was immobilized with protein, the film became more hydrophilic and showed lower water contact angle.

Biological evaluation of neat and modified surface, MC3T3-E1 cells were cultured on the PCL films. Indirect cytotoxicity test showed all types of materials did not release toxic substance to harmful the cells. For cell attachment and proliferation, the adsorbed BSA (3000 $\mu\text{g/mL}$) surface casted from 40:60 (v/v) EtOH:THF had the highest the percent viability of cells because of roughness surface. Moreover, the pre-adsorbed BSA (200, 1500 and 3000 $\mu\text{g/mL}$) and adsorbed collagen (100 $\mu\text{g/mL}$) systems did not promote cells growth as well as the adsorbed BSA system (3000 $\mu\text{g/mL}$) did. Images of SEM were used to observe cell morphology on the materials.

It can be seen that cells were still round after 4 hrs of cell seeding and began to extend their cytoplasm after 24 hrs. Although a cell attachment and proliferation provided a high value, it showed a low value of Alkaline phosphatase activity. The adsorbed BSA film casted from 40:60 (v/v) EtOH:THF provided slightly higher value than film casted from chloroform. However the values of both PCL films had a lower the value than TCPS.

Recommendations for the future work would be to study the effect of modified rough surface at high protein concentration (more than 3000 $\mu\text{g}/\text{mL}$) in other proteins adsorption and various types of cell.