

CHAPTER VI

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

In this study, a decellularized protocol of porcine dermis was developed. A periodic pressurized technique has been effectively utilized in the decellularized process during an enzymatic treatment, providing advantages over typical enzymatic methods. When periodic pressurization was applied, enzymatic treatment time could be shortened since enzyme solution could penetrate into porcine skin more thoroughly under pressurized condition. The percentage of cell removal from porcine skin was then greatly increased as observed from both quantitatively determined by DNA assay and qualitatively determined by H&F staining. The enzyme refreshment introduced in decellularized process could further improve the percentage of cell removal since the inhibition of enzyme-substrate interaction was prevented. We also proved that the number of pressurized periods could affect the percentage of cell removal. More pressurized periods could increase the percentage of cell removal and shorten enzymatic treatment time, which could prevent the disruption of dermal structure as observed by SEM. Dispase II can be used to remove cells from porcine skin better than trypsin. However, *in vivo* study showed that numerous fibroblasts from host tissue could migrate into the ADM prepared by both enzymes, and neo-collagen fibrils and neo-capillaries were produced. ADM from porcine skin produced by the periodic pressurized technique was proofed to be effectively used in skin tissue engineering.

6.2 Recommendations

1. Since porcine skin has a thick layer of subcutaneous fat, it was difficult to harvest 1 mm-thick layer of porcine dermis. Electrical microtome should be used in skin preparing process.
2. In order to improve the productivity of this process, the periodic pressurized technique should be automatically performed.
3. The optimal of porcine skin weight and enzyme solution volume used in decellularized process should be further investigated.