CHAPTER VI

CONCLUSIONS AND RECOMMENDATIONS

This chapter will be focused on conclusions of all experimental details on batch fermentation, in which the effect of controlled pH and initial starch hydrolysate (equivalent to glucose) concentrations were studied together with growth curve observation and the continuous fermentation coupling with microfiltration, which dilution rates were investigated. Moreover, the prolonged cultivation time was also investigated.

6.1 Conclusions

1. The optimum pH and initial glucose concentration at 37 °C for batch fermentation with maximum aeration rate are 7.0 and 1.8 g/l, respectively.

2. Protease production is a mixed growth association, in which protease is synthesized between log phase and stationary phase.

3. Maximum specific growth rate (μ_{max})obtained from Lineweaver-Burk plot is identical to that from growth curve estimation (0.52 hr^{-1}). Furthermore, Monod constant (Ks) obtained from both methods are relatively closed to each other (0.98 g/l from Lineweaver-Burk plot and 1.0 g/l from growth curve estimation)

4. The suitable pressure applied at feed inlet and recirculation flow rate for the continuous fermentation coupling with microfiltration of Bacillus subtilis TISTR 25 are 0.0^{+} kg_f/cm^2 and 0.4 m^3/hr, respectively.

5. The dilution rate of 0.3 hr^{-1} is the highest operating value in continuous fermentation coupling with microfiltration, any further dilution rate cannot be achieved , because of less filtration rate due to viscous biomaterial.

6. The age of cell positively affects protease production – fresh culture can produce protease better than the old one. The twice cell bleeding continuous fermentation coupling with microfiltration can improve the protease productivity 1.53-fold as high as that of batch fermentation (based on cultivation time).

6.2 **Recommendations**

1. According to the results of twice cell bleeding continuous fermentation coupling with microfiltration which can improve the productivity of protease, more experiment should be focused on stability of interval cell bleeding continuous fermentation with microfiltration because the stability of protease production is an important criteria to determine the actual production time in a real industrial production.

2. With the same reason, the study of continuous cell bleeding with continuous fermentation coupling with microfiltration should be focused to clarify the optimum bleeding ratio.

3. Because the total cell recycle system causes a cell aging problem, a continuous fermentation ,chemostat, should be studied to find an optimum dilution rate to efficiently overcome the aging problem.