CHAPTER 5

CONCLUSIONS

Chai-nat1 rice is a suitable substrate for monacolin K production because of the amylose level which is as high as 26-27%. The optimal conditions for monacolin K and citrinin analysis were successfully developed. The column of Waters Symmetry C_{18} (250mm × 4.6mm i.d., 5 µm) was chosen as the stationary phase. The isocratic elution was applied using acetonotrile/ phosphoric buffer pH 2.5 (70:30) as the mobile phase, giving monacolin K at the retention time of 8.173 min. The same stationary phase was chosen for citrinin analysis with an isocratic elution at 0.75 ml/min of acetonotrile/ formic acid buffer acid pH 2.5 (70:30), giving citrinin with retention time of 6.020 min. The procedures showed the accuracy as % recovery of mevinolin and citrinin at 98.81 and 101.73 respectively. The precision of the analysis method is also demonstrated by Relative Standard Deviation (%RSD) of 4.52 and 1.25 %R.S.D for mevinolin and citrinin respectively.

Since both monacolin K and citrinin are derived from polyketide pathway, the production of both monacolin K and citrinin presumably possess similar trends under the same cultivation conditions. The conditions that increased monacolin K could also lead to the increased production of citrinin. Thus, production of monacolin K and citrinin is closely related. Solid state cultivation was chosen as a method of choice to produce red fermented rice in this study. According to Su *et al*., 2003, solid state cultivation always produced more monacolin K than submerged cultivation, possibly because it is more stable and was easily released from rice grains under conditions of

solid state cultivation. Citrinin, another secondary metabolite-a nephrotoxic agent, is produced by *M. purpureus* or *M. ruber* in both submerged and solid state cultures according to Blanc et al.,1995.

In this study, we found out that *Monascus purpureus* BCC 6131 significantly produced monacolin K more than *Monascus ruber* TISTR 3006 and the optimum temperature for *Monascus purpureus* BCC 6131 to produce monacolin K was 30^{o}C as had been reported by Su et al., 2003. At 30^{o}C, *Monascus purpureus* BCC 6131 produced 109.86 ±5.49 ppm of monacolin K and was chosen to conduct the further study. With 30 g of polished rice, when reduce amount of water added from 30 ml to 20 ml, the production of monacolin K by *Monascus purpureus* BCC 6131 at 30^{o}C was increased from 109.86 ±5.49 ppm to 252.07±12.40 ppm. Additionally, the ratio of monacolin K to citrinin was increased from 308.20±4.19 to 785.82±7.70. The results also further revealed that neither broken rice nor unpolished rice was a better substrate than polished rice itself. Moreover, the data obtained also showed that inoculum size had significant effect on monacolin K production of *M. purpureus* BCC 6131 which supported the study of *M. purpureus* CCRC 31615 (Su *et al*., 2003). The amount of monacolin K production increased gradually when the inoculum size was increased from 1cm^2 to 4 cm^2 (352.09±20.11 ppm). It can be concluded that the optimal condition for monacolin K production from *M. purpureus* BCC 6131 at 30^{o}C is 30 g polished rice with 20 ml water added and the inoculum size of 4cm^2.

To conclude, in this study, a maximum amount of 352.09±20.11 ppm of monacolin K was produced. Even though the mount of monacolin K was still lower than it had been reported from some groups (Lee et al., 2007, sayyad et. al., 2007, Chen and Hu 2005). These data of cultivation conditions could be efficiently used to

conduct further experiment in order to improve the production of red fermented rice in future.