



## CHAPTER V

# CONCLUSION

Amino acid sequence comparison between the *Bacillus circulans* A11 CGTase and the thermostable CGTases revealed four major different regions I, II, III and IV at position 89-94, 265-271, 333-339, and 538-540 (*B. circulans* A11 CGTase numbering), respectively. The amino acid sequences in the relevant regions I-III in catalytic domains A/B of  $\beta$ -CGTase from *B. circulans* A11 were mutated towards those of thermostable CGTases using the unique site elimination (USE) mutagenesis method. The CGTase mutants with various combinations of the three mutation regions were constructed and assayed for their activities and thermostability. It was found that mutations in the three mutation regions, I, II and III, resulted in an increase in dextrinizing activity, a decrease in optimum temperature and no increase in thermostability. All CGTase mutants were active in CD-forming activity; all but one with altered product specificity.