CHAPTER 4

CONCLUSIONS

1. DNA fragments of TTR exon 2, 3 and 4 amplified by PCR from 96 individual lymphocytes or lymphoblasts of Thai people showed single bands with the fragment size of 311 bp, 205 bp and 258 bp, respectively.
2. Nucleotide sequences of TTR exons purified from the controls and the cases were determined by dye-terminator sequencing technique. The result showed that nucleotide sequence of the TTR exon2, 3 and 4 amplified from the controls were the same as that previously deposited in GenBank.
3. By using the SSCP in screening for mutation on nucleotide sequence and the DNA sequencing, a novel single point mutation (from T to C) was found on exon 4 of the TTR gene that was purified from a case with mental retardation. This mutation led to substitution by proline of leucine at position 110 of the gene.
4. Recombinant Leu110Pro was successfully synthesized and secreted by using the expression system of *P. pastoris*. The recombinant protein can be purified from other endogenous proteins of the *Pichia* by preparative native-PAGE.
5. The recombinant Leu110Pro showed the same mobility on native-PAGE as the TTR in human plasma, and moved faster than the plasma albumin. By SDS-PAGE, subunit mass of the Leu110Pro was estimated to 22.7 kDa.