GENOTYPING OF BETA THALASSEMIA TRAIT BY HIGH-RESOLUTION DNA MELTING ANALYSIS

Rattika Saetung¹, Siriwan Ongchai², Pimlak Charoenkwan¹ and Torpong Sanguansermsri¹

¹Department of Pediatrics, ²Department of Biochemistry, Faculty of Medicine, Chiang Mai University, Chiang Mai, Thailand

Abstract. Beta thalassemia is a common hereditary hematological disease in Thailand, with a prevalence of 5-8%. In this study, we evaluated the high resolution DNA melting (HRM) assay to identify beta thalassemia mutation in samples from 143 carriers of the beta thalassemia traits in at risk couples. The DNA was isolated from venous blood samples and tested for mutation under a series of 5 PCR-HRM (A, B, C, D and E primers) protocols. The A primers were for detection of beta thalassemia mutations in the HBB promoter region, the B primers for mutations in exon I, the C primers for exon II, the D primers for exon III and the E primers for the 3.4 kb deletion mutation. The mutations were diagnosed by comparing the complete melting curve profiles of a wild type control with those for each mutant sample. With the PCR-HRM technique, fourteen types of beta thalassemia mutations were detected. Each mutation had a unique and specific melting profile. The mutations included 36.4% (52 cases) codon 41/42-CTTT, 26.6% (38 cases) codon 17 A-T, 11.2% (16 cases) IVS1-1 G-T, 8.4% (12 cases) codon 71/72 +A, 8.4% (12 cases) of the 3.4 kb deletion and 3.5% (5 cases) -28 A-G. The remainder included one instance each of -87 C-A, -31 A-C, codon 27/28 +C, codon 30 G-A, IVS1-5 G-C, codon 35 C-A, codon 41-C and IVSII -654 C-T. Of the total cases, 85.8% of the mutations could be detected by primers B and C. The PCR-HRM method provides a rapid, simple and highly feasible strategy for mutation screening of beta thalassemia traits.

Keywords: beta thalassemia trait, mutation, PCR-HRM, risk couples