Screening for mutations in exons encoding the ligand-binding domain of the LDL receptor gene using PCR-CFLP and PCR-SSCP

Pongrapeeporn K.-U.\textsuperscript{a}, Leowattana W.\textsuperscript{c}, Nuchpramool W.\textsuperscript{d}, Kerdsaeng K.\textsuperscript{a}, Thepsuriyanon P.\textsuperscript{e}, Kiartivich S.\textsuperscript{c}, Yamwong P.\textsuperscript{b}, Ong-Ajyooth S.\textsuperscript{a}, Amornrattana A.\textsuperscript{b}, Kasemsuk L.\textsuperscript{a}, Laungsuwan S.\textsuperscript{c}, Sribhen K.\textsuperscript{c}

\textsuperscript{a} Department of Biochemistry, Mahidol University, Bangkok, 10700, Thailand
\textsuperscript{b} Dept. of Prev. and Social Medicine, Mahidol University, Bangkok, 10700, Thailand
\textsuperscript{c} Department of Clinical Pathology, Faculty of Medicine, Siriraj Hospital, Bangkok, 10700, Thailand
\textsuperscript{d} Department of Clinical Chemistry, Faculty of Medical Technology, Mahidol University, Bangkok, 10700, Thailand
\textsuperscript{e} Amersham Pharmacia Biotech.

Abstract
Primary hypercholesterolemia includes both monogenic disorders and polygenic conditions. Two well defined monogenic disorders are familial hypercholesterolemia (FH) and familial defective apolipoprotein (apo) B-100 (FDB). Both disorders convey high risk of premature coronary artery disease. FH and FDB are caused by mutations in LDL receptor and apo B-100 genes, respectively. In the present study, mutations in both genes in Thai subjects with primary hypercholesterolemia were screened. For apo B-100 gene, a common mutation R3500Q was screened. This mutation was not observed in the patients (n = 45). For LDL receptor gene, mutations in the exons encoding the ligand-binding domain were screened. By PCR-CFLP analysis, 18 abnormal CFLP patterns in exon 4, the hot spot for mutations, were found in patients (n=45). One of the DNA samples with abnormal CFLP patterns was previously identified and reported as a possible disease-causing mutation, namely D151Y. For the other exons, the screening technique was PCR-SSCP. Abnormal SSCP patterns in DNA samples from patients (n=20) were found as follows, two in exon 3, one in exon 5 and another one in exon 6. Further characterization by DNA sequencing and family studies for these abnormal patterns are underway.

Keywords: Hypercholesterolemia; LDL Receptor Gene; Mutation; PCR-CFLP; PCR-SSCP

Journal of the Medical Association of Thailand. 2001; 84(Suppl. 3) : S619-S627