Abstract

Charles University in Prague
Faculty of Pharmacy in Hradec Králové
Department of Biochemical Sciences

Candidate: Bc. Daniela Putzová
Supervisor: Doc. PharmDr. Martin Beránek, Ph.D.
Title of diploma thesis: Hereditary hemochromatosis – Importance of molecular biology analysis of S65C mutation in HFE gene

Background: The aim of this work was to establish an investigative method PCR-RFLP for the detection of the mutation S65C in HFE gene which can be used for an analysis of real clinical samples and for determination of their S65C mutation occurrence.

Methods: In this study, 140 samples from patients were analyzed (68 % (n = 95) of samples from men, 32 % (n = 45) from women, in the age from 20 to 83 years) for the presence of the mutation S65C in HFE gene using PCR-RFLP method. For amplification of required segments of HFE gene, primers (5´– ACA TGG TTA AGG CCT GTT GC – 3´, 5´– GCC ACA TCT GGC TTG AAA TT – 3´) were used. The enzyme HInfI was used for the restriction analysis.

Results: S65C mutation allelic frequency was determined as 1.8 % in this cohort of patients. Five patients (3.6 %) were heterozygous for the mutation S65C; two of them (1.4 %) were identified as C282Y/S65C compound heterozygotes.

Conclusions: PCR – RFLP procedure was successfully established for the determination of S65C mutation as an alternative to the more commonly analyzed mutations C282Y and H63D. The obtained results of genotypic and allelic frequencies were comparable with those reported in earlier studies concerning Central European populations.