DISCOVERY OF THREE NOVEL HLA ALLELES BY NEXT GENERATION SEQUENCING IN GREEK POPULATION

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Objectives: HLA mismatch is a critical negative factor in Hematopoietic Stem Cell (HSC) transplantation. The aim of this study was the presentation of three novel HLA alleles identified using Next Generation Sequencing (NGS) technology.

Methods: Since July 2021 up to date, 1920 samples were typed at up to 11 HLA genetic loci using NGS technology. HLA genotyping using commercial NGSgo®-MX6-1 kit (GenDx) was performed on Illumina-MiSeq platform. The Fastq-files were analyzed by NGSengine software v.2.21.0 using the IPD-IMGT/HLA Database v3.47.1.2. In order to confirm the presence of the novel HLA alleles, HLA genotyping supplied by CareDX (AlloSeq Tx17 kit) was also conducted. The sequencing data were analyzed using AlloSeq Assign Tx17.1 v1.0.3 software.

Results: a) HLA-DPB1*02:01:68 (GenBank AN:OP019279) differs from the closely related HLA-DPB1*02:01:02:01 by one single nucleotide substitution (SNS) (T>C), exon 3, gDNA 10216, codon 179c GAT>GAC, resulting in a synonymous mutation coding for Aspartic-acid. b) HLA-A*01:426 (GenBank AN:OP712622) differs from the closely related HLA-A*01:01:01:01 by one SNS (G>A), exon 4, gDNA 1617, codon 199a GCC>ACC, resulting in the amino acid change of Alanine to Threonine. c) HLA-A*02:09:01:04 (GenBank AN:OP795782) differs from the closely related HLA-A*02:09:01:01 by three SNS, intron 5; T>G, gDNA 2263; A>T, gDNA 2266; C>T, gDNA 2268. Official names have been assigned by WHO Nomenclature Committee in September and November 2022.

Conclusion: Our study highlights how NGS technology enhances HLA typing in Clinical Histocompatibility Labs by revealing extensive genetic diversity, enabling detailed analysis of HLA sequence variations, and aiding in selecting optimal donor-recipient pairs for successful transplants.