AMPATHCHAT no. 82

HLA TISSUE TYPING: INTRODUCING HIGH-RESOLUTION TYPING WITH NEXT-GENERATION SEQUENCING

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KEY MESSAGES

- This method can be used to match a donor and a recipient for a solid organ or stem cell transplant.
- Sample type: One EDTA tube
- Mnemonic: **HLATYPE**
- Turnaround time: ~6 weeks from the time that the sample is received at the laboratory
- More information can be found at https://hla.alleles.org/

INTRODUCTION TO THE HUMAN LEUKOCYTE ANTIGEN (HLA) MOLECULE

The major histocompatibility complex (MHC) genes are located on the short arm of chromosome 6 and code for cell surface markers that play a vital role in the immune system. The human MHC is synonymous with the human leukocyte antigen (HLA) complex. The two major classes are MHC Class I and Class II. MHC Class I is found on the surface of all nucleated cells and platelets, whereas MHC Class II is constitutively expressed on antigen-presenting cells of the immune system (Table 1). The function of these molecules is to present proteins, whether from "self" or "non-self", to T-lymphocytes. HLA genes are inherited (one allele from each parent) and are highly polymorphic, creating diversity within a population's gene pool.[1]

TABLE 1: THE TWO MAJOR CLASSES OF MHC

Class I – Nucleated cells and platelets	Class II – Antigen-presenting cells
HLA-A	HLA-DQ
HLA-B	HLA-DR
HLA-C	HLA-DP

INDICATIONS

HLA typing is mainly used in transplant medicine to determine whether a donor and a recipient match at tissue level, thus decreasing solid organ rejection or graft versus host disease in stem cell transplants. HLA typing can also be used in other clinical scenarios, for example, certain disease associations, pharmacogenomics, bone marrow and tissue registries, and immunotherapy.[2]

METHODS

HLA typing is performed by sequencing the DNA. This method is robust, specific and reproducible, which, in turn, allows for accurate and higher-resolution typing between the donor and the recipient. HLA typing by next-generation sequencing (NGS) is considered to be the gold standard of tissue typing methods internationally.

The test is capable of reporting 11 HLA loci in a single assay: HLA-A, HLA-B, HLA-C, HLA-DRB1, HLA-DRB3, HLA-DRB4, HLA-DRB5, HLA-DQA1, HLA-DQB1, HLA-DPA1 and HLA-DPB1 (Figure 1).

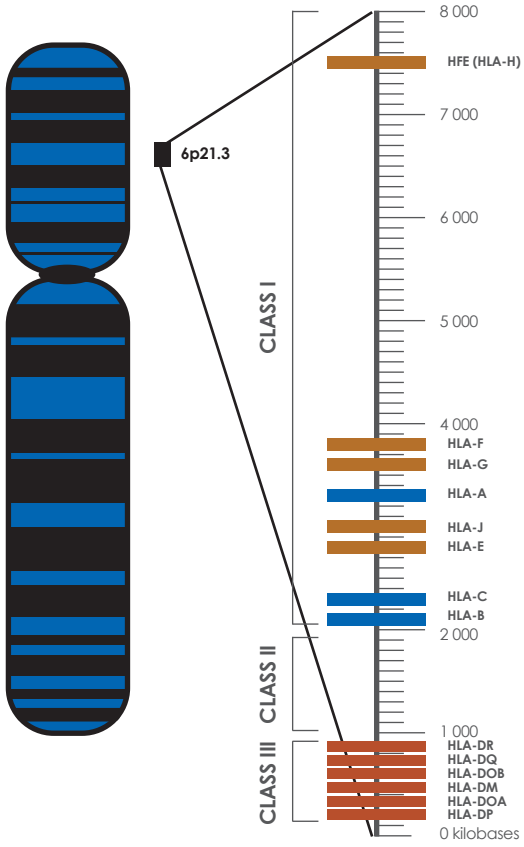

FIGURE 1: CHROMOSOME 6 ILLUSTRATING HLA ALLELES[3]

HLA NOMENCLATURE

As at the end of December 2022, 35 821 HLA alleles have been identified, but the number continues to grow daily. The naming of new HLA genes and their allele sequences is the responsibility of the World Health Organization's Nomenclature Committee for Factors of the HLA System.

Each HLA allele starts with the prefix HLA, followed by a hyphen. The name of the gene follows (e.g. A/B/C or DR/DQ/DP) with an asterisk and up to four sets of digits, separated by a colon (Figure 2).

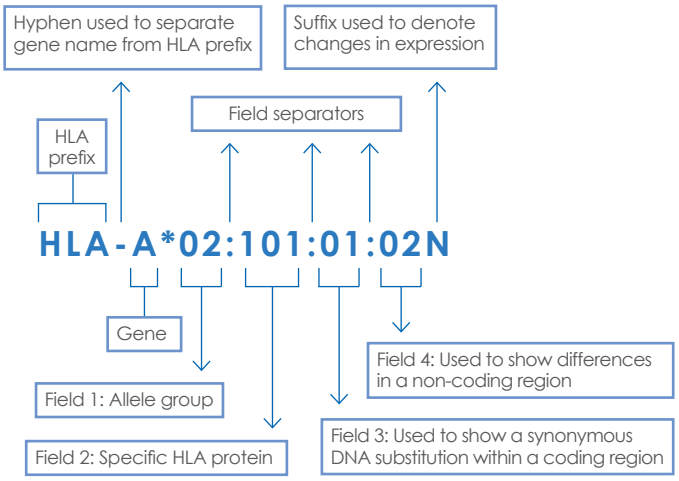

FIGURE 2: HLA NOMENCLATURE[4]

ADVANTAGES

PCR and the historic serological methods were restricted to single or double digits (only typing up to the first field of the HLA gene). Due to the polymorphism of the HLA molecule, rare and new alleles could be missed, particularly in our culturally diverse population.

DNA-based typing is able to sequence the entire HLA gene (including introns and untranslated regions). This has been shown to be beneficial for disease outcome and morbidity in the transplant setting.[5]

Other advantages include:

- Increased throughput
- High resolution (up to eight digits/four fields)
- Increased accuracy
- Decreased genotyping ambiguity
- Improved price

HLA DISEASE ASSOCIATIONS

Some diseases are more common in patients with a specific HLA allele or haplotype. The most common of these are included in Table 2.[6]

One of the strongest HLA disease associations is HLA B27, which is present in >90% of people with ankylosing spondylitis. Other diseases can be associated with more than one allele, e.g. coeliac disease and Type I diabetes mellitus.

It is important to note that these HLA alleles are only associated with the pathology, but are not regarded as a diagnostic criterion for the specific disease. Disease can still be present in the absence of the specific HLA allele.[6]

TABLE 2: HLA DISEASE ASSOCIATIONS OFFERED AT AMPATH

	Coeliac disease	Ankylosing spondylitis	Abacavir hypersensitivity	Behçet syndrome	Other
HLA allele/s	HLA DQ2 and DQ8	HLA B27	HLA B57:01	HLA B51	Specify (any loci)
Method	Array	PCR	PCR	PCR	NGS
Mnemonic	HLADQ28	HLAP	HLAB5701	HLA51	HLADIS

For queries related to HLA typing, contact the Next-generation Sequencing Laboratory at 012 678 0670 or ngs@ampath.co.za.

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