

*BloodCenter of Wisconsin offers high resolution typing for
Class I HLA-A, -B, and -C loci and Class II HLA-DRB1, -DQB1, and -DPB1 loci
by DNA sequencing.*

BACKGROUND:

DNA sequencing is universally regarded to be the gold standard for identification of HLA alleles. By sequencing portions of HLA genes that govern compatibility, all common allele combinations can be resolved and new alleles are more easily identified.

REASONS FOR REFERRAL:

HLA matching is important for successful allogeneic hematopoietic stem cell transplantation.

METHOD:

PCR and Direct DNA Sequencing

The informative class I and II HLA exons of an individual sample are amplified using the polymerase chain reaction (PCR) with locus-specific primers. The nucleotide sequences for each haplotype are determined in both directions using one of two methods. One method generates dye-terminator sequencing fragments from the PCR products which are analyzed on an automated, high throughput capillary sequencer with allele-assignment software. A second method uses a Next Generation Sequencing platform where the PCR products for each sample are fragmented and barcoded prior to sequencing on the instrument using reversible dye-labeled terminators. Output sequences are assembled for each sample and HLA types are determined for each locus by allele-assignment software. HLA types are reported for each locus by trained and certified histocompatibility experts.

LIMITATIONS:

- Some rare allele combinations and alleles whose sequences differ outside the informative exons may not be completely resolved by this technique.

SPECIMEN REQUIREMENTS:

4 buccal swabs or 14 ml EDTA (lavender top) whole blood or 2-5 ml EDTA bone marrow or high quality DNA ($\geq 500\text{ng}$ at $25\text{ng}/\mu\text{l}$)

SPECIMEN REQUIREMENTS (PEDIATRIC):

3-4 buccal swabs or 5 ml EDTA (lavender top) whole blood
Contact the laboratory for instructions if submitting cord blood

SHIPPING REQUIREMENTS:

Place the room temperature specimen and requisition into plastic bags, seal and place in an insulated container. Seal the container and place in a sturdy cardboard container and tape securely. Ship the package in compliance with your overnight carrier guidelines. Address the package to:

Client Services/Histocompatibility Laboratory
BloodCenter of Wisconsin
638 N. 18th St.
Milwaukee, WI 53233-2121
800-245-3117, ext. 6250

TURNAROUND TIME: 5-7 days

CPT CODES:

HLA-A High Resolution: 81380
HLA-B High Resolution: 81380
HLA-C High Resolution: 81380
HLA-DRB1 High Resolution: 81382
HLA-DQB1 High Resolution: 81382
HLA-DPB1 High Resolution: 81382