

*BloodCenter of Wisconsin offers a single test for expanded high resolution typing of Class I HLA-A, -B, and -C loci and Class II HLA-DRB1/3/4/5, -DQA1, -DQB1, -DPA1, and -DPB1 loci by next generation DNA sequencing (NGS).*

## BACKGROUND:

DNA sequencing is universally regarded to be the gold standard for identification of HLA alleles. Historically, sequence based typing of HLA genes has focused on the exons that encode the Antigen Recognition Site (ARS) of the HLA molecule. By sequencing regions beyond the ARS of HLA genes that govern compatibility, all common allele combinations can be resolved and new alleles are more easily identified.

## REASONS FOR REFERRAL:

- The HLA High Resolution Panel by NGS provides typing for all transplant relevant loci of the HLA region in a single orderable with fewer ambiguities versus Sanger based methods.
- 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 and introns of an individual sample are amplified using the polymerase chain reaction (PCR) with locus-specific primers. The nucleotide sequences for each HLA locus are determined using 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 and HLA types are assigned based on known reference alleles using validated 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 included HLA regions 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 (3-4ug at 25ng/ul)

**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, B, C, & DRB1 High Resolution: 81378  
HLA-DRB3/4/5 High Resolution: 81382  
HLA-DQA1 High Resolution: 81382  
HLA-DQB1 High Resolution: 81382  
HLA-DPA1 High Resolution: 81382  
HLA-DPB1 High Resolution: 81382