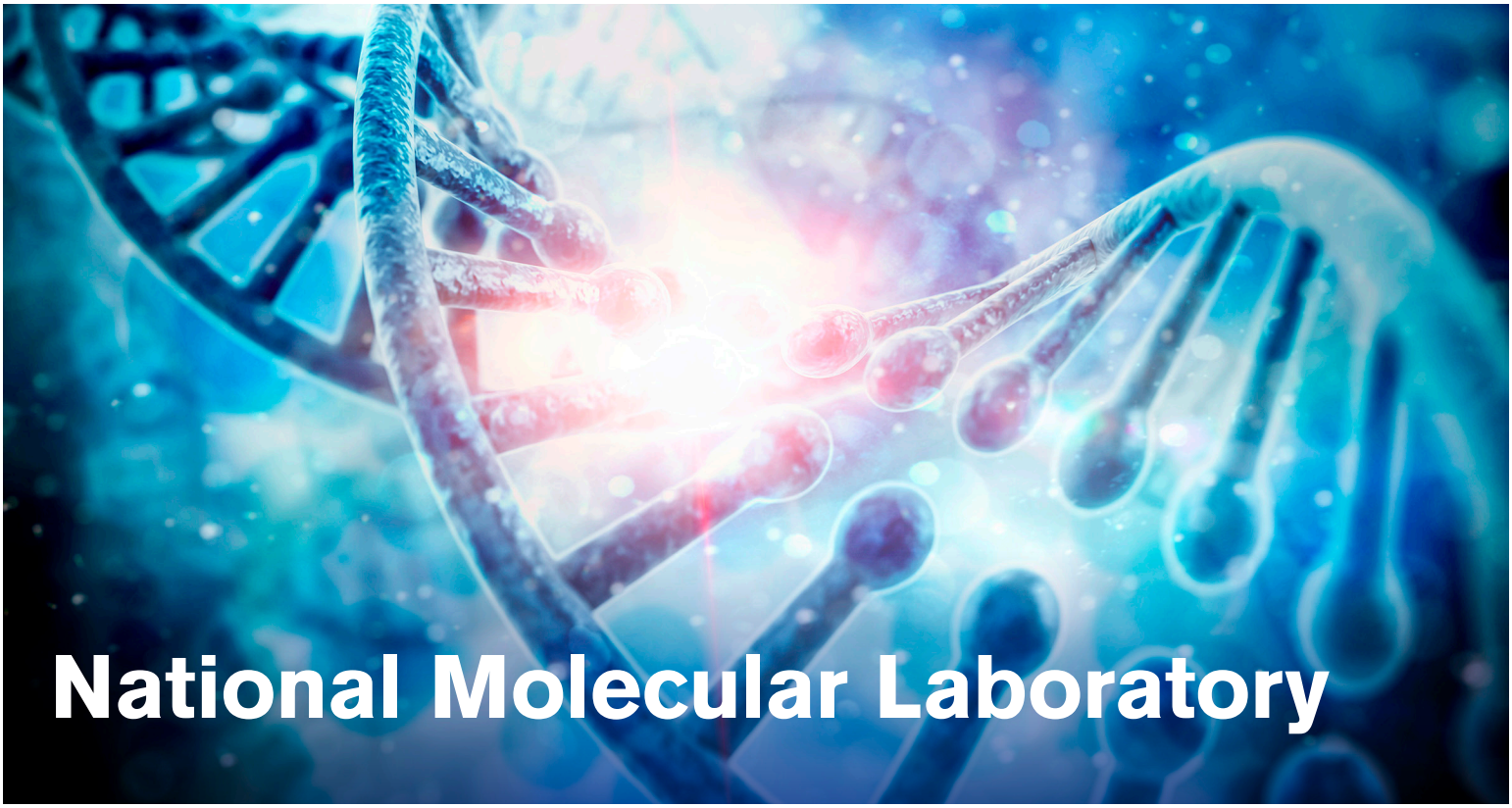

National Molecular Laboratory

The American Red Cross National Molecular Laboratory (NML) is an inclusive central testing facility at the forefront of research and testing efforts related to molecular testing. The laboratory is recognized for its ability to characterize complex blood group alleles including identification of novel and null alleles. We provide a predicted RBC phenotype based on an FDA approved test. The facility also uses advanced genomic methodologies to predict blood group and platelet antigen phenotypes. The laboratory has been performing RBC genotyping of donors since 2008 and has screened more than 300,000 blood donors.

Our NML is AABB-accredited, CLIA-certified, and licensed by the Department of Health of multiple states. NML laboratory leadership contributes to several industry programs including the AABB Molecular Testing Program Unit and the International Society of Blood Transfusion (ISBT) Working Party for Red Cell Immunogenetics and Blood Group Terminology.

Blood Group and Platelet Antigen Testing Indications

Indications

- Multi-transfused patients, including those with sickle cell disease and thalassemia
- Patients with warm autoantibodies, and those receiving immunotherapies including anti-CD38
- Extended genotype matching of patients requiring long-term transfusion support
- Molecular characterization of the major antigens in the RH system
- Identification of RH variants, including those encoding weak and partial D phenotypes
- Determination of paternal RHD gene zygosity
- Evaluation of risk of hemolytic disease of the fetus and newborn (HDFN)
- Evaluation of fetal and neonatal alloimmune thrombocytopenia (FNAIT)
- Support of serology-related discrepancy resolutions and complex serologic evaluations
- Cost-effective and efficient screening of blood donors for prediction of antigens
- Accurate typing of panel and rare donor cells for reference laboratories

Description

Peripheral blood or buccal swab specimens are used to obtain genomic DNA for genotyping. Testing can predict one specific blood group antigen or can involve use of a genotyping panel that can predict multiple human erythrocyte antigen (HEA) or human platelet antigen (HPA) phenotypes simultaneously.

Sanger sequence analysis is used to characterize new alleles in many blood group systems, resolve complex Rh blood group alleles, and identify weak ABO subgroups. Plasmid cloning is used to resolve complex allele assignments. Testing is available for the following systems:

Blood Group Antigens, including

- HEA panel (including antigens in RH, KEL, FY, JK, MNS, LU, DO, LW, DI, CO, SC systems)
- ABO, including subgroups
- *RHD* gene zygosity
- *RHD* gene variants including weak D types 1, 2 and 3 and partial D types
- C, c, E, e, VS, V, hr^B, hr^S, Hr^B
- FY – Fy^a, Fy^b, Fy^x, GATA, Fy null
- KEL – K, k, Js^a, Js^b, Kp^a, Kp^b, K_{mod}, K_0
- MNS – M, N, S, s, U^{+var}, U-
- JK – Jk^a, Jk^b including weak and null phenotypes
- DO – Do^a, Do^b, Hy, Jo^a, Gy^a
- LU – Lu^a, Lu^b and In(Lu)
- CO – Co^a, Co^b
- KN – Kn^a, Kn^b, McC^a, McC^b, Sl^a, Sl3, Vil, KCAM
- YT – Yt^a, Yt^b
- CR – Cr^a, Cr^b

Platelet Antigens, including

- HPA-1a/1b
- HPA-2a/2b
- HPA-3a/3b
- HPA-4a/4b
- HPA-5a/5b
- HPA-6a/6b
- HPA-7a/7b
- HPA-8a/8b
- HPA-9a/9b
- HPA-11a/11b
- HPA-15a/15b

Test Methods

- Sequence-specific primer (SSP)-Polymerase chain reaction (PCR) PCR-restriction fragment length polymorphism (RFLP)
- Multiplex PCR-based hybridization and elongation, including use of PreciseType™ | HEA
- Multiplex PCR and single base primer extension by MALDI-TOF mass spectrometry
- Sequence-based typing (SBT) using Sanger Sequencing
- Transcript analysis by Sanger Sequencing of cDNA

The Red Cross can help resolve complex antibody identification problems and aid in the selection of compatible donors with state-of-the-art molecular testing. Here are two cases to illustrate the utility of molecular testing:

Sample Case 1:

A sample from a pregnant woman is found to be RhD negative when tested as part of a preoperative work-up by the hospital. Previously the patient's red blood cells typed RhD positive. The sample is submitted for D variant testing as part of the discrepancy resolution. The sample is found to be weak D+, carrying the *RHD*weak D type 2* allele. Based on the *RHD* genotype carried by the patient, they are not predicted to be at risk of RhD alloimmunization and do not need Rh immunoprophylaxis.

Sample Case 2:

A patient with sickle cell disease presents with anti-C; their RBCs type D+C+E+c+e+. The physician requests molecular genotyping for red cell antigens. The HEA genotyping panel indicated that the patient might carry an r's allele. RH genotyping predicts the patient to express an altered C antigen, supporting that the anti-C detected is an alloantibody. In addition, the patient is predicted to express a partial e antigen and lack the high prevalence antigen hr^B. This information can be used in selection of blood products to minimize risk of further Rh alloimmunization.