



**American
Red Cross**

**Testing Performed by the
AABB Molecular Accredited
American Red Cross
National Molecular Laboratory**

Starting Material and Shipping Requirements for National Molecular Laboratory

All samples must be clearly labeled with the full name of the individual and a unique identification number. The information on the tube must match the information on the service request form. Requests should include date of collection.

Whole Blood should be collected in 5-10 ml EDTA (lavender top) tubes. Samples less than 10 days old are preferred. DNA yield from older specimens may be low or QNS. Whole blood specimens submitted for cDNA analysis should be collected with more than 90 days since transfusion of red cell products.

Whole blood sample can be shipped at room temperature or refrigerated using ice packs or wet ice sealed in plastic bags, according to DOT regulations for biological specimens.

Buccal swabs may be acceptable depending on the testing requested; contact the laboratory for more information. Buccal swabs should use a cotton-tipped applicator and be air dried completely before putting in a clean tube and shipping on ice packs or wet ice sealed in plastic bags.



Human Erythrocyte Antigen (HEA) Genotyping Panel

Test Code SREF-IVD

Recommended CPT Code 0001U x 1

An FDA-cleared multi-analyte test performed on DNA from whole blood provides a predicted phenotype for 36 antigens. This testing is useful when antigen status cannot be determined serologically, either because of recent transfusion, positive DAT or when reagents are not available. The HEA genotyping panel is useful in patients being treated with the anti-CD38 drug daratumumab. The extended antigen phenotype prediction is also useful in patients on chronic transfusion regimens. In alloimmunized patients, it can be used to assess the additional alloimmunization risk.

RHD Genotyping for D variants

Most Common Test Code SREF-RHD

Additional test codes: SREF200B, SREF250D, SREFSEQD, SREF500D, SREF550D, SREF750D

Recommended CPT Code 81403 x each

RHD Genotyping is used to identify *RHD* variant alleles and predict the D antigen phenotype. There are many *RHD* variant alleles and variants can have altered antigen expression and/or altered epitopes. *RHD* variants are found in all ethnic groups but are most common in individuals of African descent. Weak or altered antigen expression can lead to a serologic weak D phenotype. This is especially important in pregnant women and women of child-bearing age, as the decision regarding RhIg prophylaxis is informed by *RHD* variant type. Specifically, individuals who are weak D types 1, 2 and 3 are not at risk of D alloimmunization. Weak or altered RhD antigen expression can also result in a typing discrepancy, either between test methods, reagents, laboratories or between a current and historic type. *RHD* genotyping is performed using a commercially available “Research Use Only” genotyping panel that can detect many weak, Del and partial alleles including weak D types 1, 2 and 3 plus one or more lab-developed tests to rule out weak or partial alleles not interrogated on the commercial panel. cDNA analysis can be used to resolve equivocal allele status of compound heterozygous samples or suspected splice site variants.

***RHCE* Genotyping for C, c, E and e variants including hr^B and hr^S status**

Most Common Test Codes SREFRHD and SREF450

Additional test codes: SREF200C, SREF250H, SREFSEQC, SREF500C, SREF550C, SREF750C

Recommended CPT Code 81403 x each

RHCE Genotyping is used to identify *RHCE* variant alleles and can predict C, c, E, e, V, VS, hr^B and hr^S antigen phenotypes. There are many *RHCE* variant alleles and variants can have weakened or altered antigen expression, be associated with loss of a high prevalence antigen or gain of a low frequency antigen. *RHCE* variants are found in all ethnic groups but are most



common in individuals of African descent. Weak antigen expression can lead to typing discrepancies, for example with e and C antigens. Patients who express altered antigens can be at risk of alloimmunization (eg., e+ with anti-e or C+ with anti-C). *RHCE* genotyping is performed using a commercially available “Research Use Only” genotyping panel that can detect many common altered alleles plus one or more lab-developed tests to rule out variant alleles not interrogated on the commercial panel. cDNA analysis can be used to resolve equivocal allele status of compound heterozygous samples or suspected splice site variants.

RH Characterization

Test Codes SREF-RHD and SREF450

Additional Tests may include SREF750, SREF-SEQ, SREF500, SREF550

Recommended CPT Code 81403 x each

RHD variant and *RHCE* variant alleles are often co-inherited and are not uncommon in individuals of African descent. Patients who are found to have D variants may be at risk of expressing altered *RHCE* antigens and visa-versa. Patients of African descent who are likely to be multiply or chronically transfused may benefit from *RH* characterization. *RH* characterization includes use of *RHD* and *RHCE* commercially available “Research Use Only” genotyping panels and may also include performance of one or more lab-developed tests to rule out variant alleles not interrogated on these commercial panels. cDNA analysis can be used to resolve equivocal allele status of compound heterozygous samples or suspected splice site variants.

RHD Zygosity

Most Common Test Code SREF-RHZ

Additional test codes may include SREF200D, SREF250E, SREF250B

Recommended CPT code 81403 x each

It is not uncommon to be hemizygous for *RHD* alleles. The *RHD* zygosity status of the partner of an RhD negative woman with anti-D can be used to assess risk of alloimmunization. *RHD* zygosity includes one or more lab-developed tests.

ABO Common Allele Determination

Test Code SREF250A

Recommended CPT Code 81403 x 1

Determination of ABO common alleles (A, A2, B, O1, O2) can be helpful in investigating discrepancies involving ABO blood type. This testing cannot rule out the presence of rare subgroups. ABO common alleles are tested using a lab-developed test.

ABO Variants

Test Code SREF550A

Recommended CPT Code 81403 x 1

When ABO common allele determination does not resolve an ABO discrepancy or unexpected serologic result, ABO variant testing may be warranted. ABO variant alleles (eg., B(A), Ax, Aw)



are identified by Sanger sequencing. This testing is currently performed by a subcontracted laboratory using patient DNA prepared at the Red Cross.

Non Rh, Non ABO Variants

Test Codes include SREFSEQ series, SREF500 series, SREF750 series (call for specific codes)

Recommended CPT Code 81403 x each

Typing discrepancies or suspected alloantibodies to non-Rh blood group systems may benefit from molecular testing. Blood group systems include MNS, LU, KEL, FY, JK, DO, KN, YT, and XK. The *KLF1* gene can be interrogated to rule out a possible *In(Lu)* phenotype. For example, a patient who is Jk(a+) with allo-Jk^a may express a Jk^a variant that is associated with alloimmunization. In addition, variant testing can be useful in resolving discrepancies between serologic testing and the phenotype predicted by the HEA genotyping panel (described above).

This testing is performed using lab-developed tests using gel-based genotyping including sequence-specific primer PCR (SSP-PCR) and sequence-based typing (SBT). cDNA analysis can be used to resolve equivocal allele status of compound heterozygous samples or suspected splice site variants in RH, MNS, JK, KEL and LU systems.

Human Platelet Antigen (HPA) Genotyping Panel

Test Codes SR81400A, SREFHPA2, SREFHPA3, SREFHPA4, SREFHPA5, SREFHPA6, SREFHPA9, SREFHP15

Recommended CPT Codes 81105-81112, x 1 each

A single multi-analyte test performed on genomic DNA provides a predicted phenotype for antigens HPA-1a/1b, HPA-2a/2b, HPA-3a/3b, HPA-4a/4b, HPA-5a/5b, HPA-6a/6b, HPA-9a/9b and HPA-15a/15b. This testing is useful when antigen status cannot be determined serologically. The extended antigen phenotype prediction is useful in workups of suspected Fetal/Neonatal Alloimmune Thrombocytopenia (FNAIT). In platelet refractory patients in whom HLA antibodies have been ruled out, it can be used to assess alloimmunization and select platelet products.

Bombay and Para-Bombay

Test Codes SREFFUT1, SREFFUT2

Recommended CPT Codes 81403 x each

When Bombay or Para-Bombay type is suspected, FUT1 or FUT1 testing may be warranted. FUT variant alleles are identified by Sanger sequencing. This testing is currently performed by a subcontracted laboratory using patient DNA prepared at the Red Cross.