

# Neutrophil Antigen Genotyping

## Versiti offers DNA-based typing for the neutrophil alloantigen systems HNA-1, HNA-3, HNA-4 and HNA-5.

The neutrophil alloantigens HNA-1a, HNA-1b, HNA-1c, HNA-3a, and HNA-4a have all been reported to cause neonatal alloimmune neutropenia (NAN).<sup>2,3,4,5</sup> Antibodies against HNA-1a, -1b, and HNA-3a have all been implicated in cases of TRALI,<sup>7,8</sup> with HNA-3a antibodies being the most frequent cause of fatal TRALI reactions.<sup>7</sup> Population studies suggest the HNA-1c and HNA-5a antigens may be more important as a cause of alloimmunization than previously appreciated.<sup>8,9,10</sup>

Antigen	Common Name	Glycoprotein Location	Antigen Frequency (%)	
			Caucasian	African American
<b>Neutrophil-specific:</b>				
HNA-1a	NA1	CD16 (FcγRIIIb)	54	47
HNA-1b	NA2	CD16 (FcγRIIIb)	88	84
HNA-1c	SH	CD16 (FcγRIIIb)	5	22
HNA-2	NB1	CD177	97	unknown
<b>Shared antigens:</b>				
HNA-3a	5b	CTL2	95	unknown
HNA-3b	5a	CTL2	70	unknown
HNA-4a	MART	CD11b (Mac-1)	92	unknown
HNA-5a	OND	CD11a (LFA-1)	85	unknown

### Indications for testing:

- Confirmation of antibody specificity in patient samples.
- In cases of NAN, parental genotyping provides information on the probability of future infants being affected.<sup>8</sup>

- Genotyping of blood donors can be used to determine those at risk of producing HNA-3 antibodies to serve as a TRALI risk reduction practice.

### Test method:

Allele-specific PCR. Genotypes for HNA-1a/1b/1c, HNA-4a/4b, and HNA-5a/5b are assigned after PCR products have been analyzed by agarose gel electrophoresis.<sup>10</sup>

Genotypes for HNA-3a/3b are determined by TaqMan® PCR followed by genotyping with allele-specific hydrolysis probes.<sup>1</sup>

### Specimen requirements:

7-15 ml amniotic fluid, 1 ml cord blood, 5x10<sup>6</sup> cultured amniocytes, or 3-5 ml EDTA (lavender top) whole blood. For evaluation of NAN, include both maternal and paternal whole blood.



SHIP

### Shipping requirements:

Ship whole blood at room temperature. Place the specimen and a completed test requisition in plastic bags, seal, and place in a Styrofoam container. Seal the Styrofoam container, place in a sturdy cardboard box and tape securely. Ship the package in compliance with your overnight carrier guidelines. Please notify the laboratory if shipping on Friday, Saturday, or the day before a holiday.

Send to:

Versiti Client Services  
Platelet and Neutrophil Immunology Laboratory  
638 N. 18th Street  
Milwaukee, WI 53233  
800-245-3117, ext. 6250



ORDER

**Required forms:**

Please complete all pages of the [requisition form](#).

**CPT Codes/Billing/Turnaround time:**

**Test codes:**

Panel: 5201

HNA-3: 5203

HNA-4: 5204

HNA-5: 5205

HNA-1: 5250

**CPT code:** For recommended CPT codes, visit the [versiti.org/test-catalog](https://www.versiti.org/test-catalog)

**Turnaround time:** 7 days

**References:**

1. Curtis BR, Cox NJ, Sullivan MJ, et al. The neutrophil alloantigen HNA-3a (5b) is located on choline transporter-like protein 2 and appears to be encoded by an R>Q154 amino acid substitution. *Blood* 2010;115: 2073-6.
2. Bux J, Jung KD, Kauth T, Mueller-Eckhardt C. Serological and clinical aspects of granulocyte antibodies leading to alloimmune neonatal neutropenia. *Transf Med* 1992;2:143-149.
3. Curtis BR, Reno CK, Aster RH. Neonatal alloimmune neutropenia attributed to maternal immunoglobulin G antibodies against the neutrophil alloantigen HNA-1c (SH): a report of five cases. *Transfusion* 2005;45:1308-1313.
4. Fung YL, et al. Alloimmune neonatal neutropenia linked to anti-HNA-4a (Mart). *Transfus Med* 2003;13:49-52.
5. Hessner MJ, Curtis BR. Prenatal genotyping for identification of fetuses at risk for immune cytopenic disorders. In: *Molecular Diagnostics for the Clinical Laboratorian*. Coleman WB and Tsongalis GJ (editors), Humana Press, 2 edition, 2005.
6. Bux J, Kissel K, Nowak K, et. al. Autoimmune neutropenia: clinical and laboratory studies in 143 patients. *Ann Hematol* 1991;63:243-252.
7. Reil A, Keller-Stanislawski B, Gunay S, Bux J. Specificities of leucocyte alloantibodies in transfusion-related acute lung injury and results of leucocyte antibody screening of blood donors. *Vox Sang* 2008;95: 313-7.
8. Silliman CC, Ambruso DR, Boshkov LK. Transfusion related acute lung injury. *Blood* 2005;105:2266-2273.
9. Sachs UJH, Reil A, Bauer C, Bux J, Santoso S. Genotyping of human neutrophil antigen-5a (Ond). *Transfus Med* 2005;15:115-117.
10. Hessner MJ, Curtis BR, Endean DJ, and Aster RH. Determination of neutrophil antigen NA gene frequencies in five different ethnic groups by polymerase chain reaction with sequence-specific primers (PCR-SSP). *Transfusion* 1996;36:895-899.

