

Guidelines for the Blood Transfusion Services

18.1: Reagent manufacture/reference preparations

<http://www.transfusionguidelines.org/red-book/chapter-18-platelet-immunology/18-1-reagent-manufacture-reference-preparations>

18.1: Reagent manufacture/reference preparations

18.1.1: HPA typing reagents

- There are several human platelet antigen (HPA) genotyping and phenotyping techniques. The latter are generally based on the use of polyclonal HPA alloantibodies obtained from immunised donors or patients, or monoclonal antibodies. HPA typing techniques that do not require polyclonal antibodies derived from donors or patients are the techniques of choice.
- HPA typing reagents prepared from human source material should comply with the guidelines in section 11.1.4.10. An 'Instructions for use' sheet (package insert) should be prepared and supplied with antibody typing reagents. Information in the 'Instructions for use' should further indicate the immunoglobulin class of the antibodies and the presence of any other contaminating antibodies reactive by the recommended methods.
- HPA typing reagents used in genomic DNA and polymerase chain reaction (PCR)-based techniques should comply with the guidelines in Chapter 14.

18.1.2: Composition of platelet cell panel for HPA antibody detection

- It is recommended that laboratories make all reasonable efforts to include cells in their panel that will aid the detection and identification of clinically significant HPA antibodies. The panel should consist of platelets typed at a minimum for HPA-1, -2, -3, -5 and -15 by validated HPA typing techniques. Ideally, the panel should contain platelets that are homozygous for HPA-1a, -1b, -2a, -2b, -3a, -3b, -5a, -5b, -15a and -15b and be from Group O donors.
- HPA typing of a platelet panel donor should be based on two concordant typing results using samples obtained on different occasions.

18.1.3: Selection of normal control sera

Normal control sera should be taken from non-transfused group AB male or ABO compatible blood donors. The sera should be screened and found negative for platelet-reactive-antibodies (e.g. clinically non-significant autoantibodies or EDTA-dependent antibodies are occasionally detected in apheresis donors). An appropriate number of normal sera should be used so that a statistically relevant normal range in a given assay can be determined.

18.1.4: Selection of positive control sera

At least one positive control should be included in each assay. The selection and number of positive control sera will depend on the technique and the HPA type of the platelets being used.

In glycoprotein-specific assays a positive control for each glycoprotein used should be included as a minimum.

18.1.5: Reference preparations

- Sensitivity of techniques should be monitored on the basis of the inclusion of a 'weak positive' control. For anti-HPA-1a, -3a and -5b, the internal sensitivity control should be calibrated against the WHO International Reference Reagents for anti-HPA-1a (NIBSC code 05/106), anti-HPA-3a (NIBSC code 03/190), anti-HPA-5b (NIBSC code 99/666) and anti-HPA-15b (NIBSC code 18/220) when diluted as instructed by the manufacturer.
- In-house sensitivity standards, with similar reaction strengths to the above reagents, should be prepared for anti-HPA-1, -3 and -5, and, if possible, for anti-HPA-2 and -15 antibodies.

Table 18.1 Current HPA nomenclature

System	Antigen	Original names	Glycoprotein	CD
HPA-1	HPA-1a	Zw ^a , PIA1	GP1IIa	CD61
	HPA-1b	Zw ^b , PIA2		
HPA-2	HPA-2a	Ko ^b	GP1balpha	CD42b
	HPA-2b	Ko ^a , Sib ^a		
HPA-3	HPA-3a	Bak ^a , Lek ^a	GP1Ib	CD41
	HPA-3b	Bak ^b		
HPA-4	HPA-4a	Yuk ^b , Pen ^a	GP1IIa	CD61
	HPA-4b	Yuk ^a , Pen ^b		
HPA-5	HPA-5a	Br ^b , Zav ^b	GP1a	CD49b
	HPA-5b	Br ^a , Zav ^a , Hc ^a		
	HPA-6bw	Ca ^a , Tu ^a	GP1IIa	CD61
	HPA-7bw	Mo ^a	GP1IIa	CD61

	HPA-8bw	Sr ^a	GPIIIa	CD61
	HPA-9bw	Max ^a	GPIIb	CD41
	HPA10bw	La ^a	GPIIIa	CD61
	HPA11bw	Gro ^a	GPIIIa	CD61
	HPA12bw	Iy ^a	GPIIb β	CD42c
	HPA13bw	Sit ^a	GP1a	CD49b
	HPA14bw	Oe ^a	GPIIIa	CD61
HPA-15	HPA-15a	Gov ^b	CD109	CD109
	HPA-15b	Gov ^a		
	HPA-16b	Duv ^a	GPIIIa	CD61
	HPA-17b	Va ^a	GPIIIa	CD61
	HPA-18b	Cab ^a	GP1a	CD49b
	HPA-19b	Sta	GPIIIa	CD61
	HPA-20b	Kno	GPIIb	CD41
	HPA-21b	Nos	GPIIIa	CD61
	HPA-22b	Sey	GPIIb	CD41
	HPA-23b	Hug	GPIIIa	CD61
	HPA-24b	Cab2 ^{a+}	GPIIb	CD41

	HPA-25b	Swi ^a	GPIa	CD49b
	HPA-26b	Sec ^a	GPIIIa	CD61
	HPA-27b	Cab3 ^{a+}	GPIIb	CD41
	HPA-28b	War	GPIIb	CD41
	HPA-29b	Kha ^b	GPIIIa	CD61
	HPA-30b	Lab ^a	GPIIb	CD41
	HPA-31b	Cab4 ^b	GPIX	CD42a
	HPA-32b	Dom ^b	GPIIIa	CD61
	HPA-33b	Bl ^a	GPIIIa	CD61
	HPA-34b	Bzh ^a	GPIIIa	CD61
	HPA-35b	Efs ^a	GPIIIa	CD61

18.1.6: Quality control schemes

Laboratories should take part in regular external quality control exercises such as the UK NEQAS for Histocompatibility and Immunogenetics schemes for HPA genotyping and HPA antibody detection /specification. Effective mechanisms should be in place to correct poor performance in the quality scheme.

18.1.7: Nomenclature

The current HPA nomenclature must be used for recording platelet-specific alloantigen and alloantibody specificities¹ (see Table 18.1). Any subsequent additions can be found in the Human Platelet Antigen Database (www.versiti.org/products-services/human-platelet-antigen-hpa-database).