

ONLINE SUPPLEMENTARY CONTENT**Primers**

Genotyping for HPA-1 to -6, -9 and -15 systems was performed by polymerase chain reaction (PCR) amplifications using sequence-specific primers (SSP). Primer sequences and amplicon sizes for each of the HPA-systems and the internal control are detailed in Supplementary **Table SI**.

Master Mixes

The assays for HPA-1,-2 and -3 systems were modified from those described by Klüter *et al.* and performed as multiplex PCR SSP. The assays for HPA-4, -5, -6, -9, and -15 systems were performed in uniplex PCR SSP with some modifications to protocols described by Skogen *et al.*, Cavanagh *et al.* and Schuh *et al.*

Two different amplification reaction mixtures were prepared for each HPA system, mix a for amplification of allele a (containing primer forward a and the common reverse primer) and mix b for amplification of allele b (containing primer forward b and a common reverse primer). All PCR were carried out using PCR Reagent Buffer 10x, 25 mM MgCl₂ stock solution, 10 mM stock PCR nucleotide mix (dNTP solution), and Taq Polymerase 5 U/μL from the FastStart Taq DNA Polymerase dNTPack kit (Roche Diagnostics, Mannheim, Germany). The mixtures were prepared and added in 0.2 mL PCR tubes in cold blocks. DNA was also added to the mix in cold blocks. The concentration of DNA was between 10-50 ng/μL. The temperature condition was maintained until the tubes were placed in the thermocycler.

To genotype HPA-1, -2 and -3 systems, 2 μL of DNA were added to each 0.2 mL PCR tube containing 23 μL of amplification reaction mixture a or b. The following reagents were added to each mixture: 10X PCR reaction buffer, 2 mM MgCl₂, 0.4 mM dNTP, 0.148 μM of each internal control primer (HGH-1/-2), 0.375 μM of each allele-specific primer HPA 1a or HPA 1b and HPA 1II, 0.25 μM of HPA 2a or HPA 2b and HPA 2 II, 0.2 μM of HPA3a or HPA3b and HPA3II and 1.5 U of Taq DNA Polymerase 5 U/μL.

For HPA 4 system genotyping, 1 μL of DNA was added to 9 μL of the amplification mixtures containing: 10X PCR reaction buffer, 3 mM MgCl₂, 0.075 mM dNTP, 0.16 μM of each internal control primer, 1.05 μM of each allele-specific primer, HPA 4a or HPA 4b and 1.01 μM of HPA 4c and 0.5 U of Taq DNA Polymerase.

For HPA 5 system genotyping, 0.5 μL of DNA were added to 12.5 μL of each reaction mixture containing: 10X PCR

reaction buffer, 3.5 mM MgCl₂, 0.2 mM dNTP, 0.48 μM of forward control primer and 0.55 μM of reverse control primer, 0.94 μM of HPA 5a and 0.8 μM of HPA 5c to mix "a"

Table SI - Primers

HPA System	Primer	Amplicon size
HPA 1a	5'-ACT TAC AGG CCC TGC CTC T-3'	189 bp
HPA 1b	5'-ACT TAC AGG CCC TGC CTC C-3'	
HPA 1II	5'-GTG CAA TCC TCT GGG GAC T-3'	
HPA 2a	5'-CCC CCA GGG CTC CTG AC-3'	241 bp
HPA 2b	5'-CCC CCA GGG CTC CTG AT-3'	
HPA 2II	5'-GCC AGC GAC GAA AAT AGA GG-3'	
HPA 3a	5'-GGG GGA GGG GCT GGG GA-3'	293 bp
HPA 3b	5'-GGG GGA GGG GCT GGG GC-3'	
HPA 3II	5'-GGC CCT GGG ACT GTG AAT G-3'	
HPA 4a	5'-GCT GGC CAC CCA GAT GCG-3'	120 bp
HPA 4b	5'-GCT GGC CAC CCA GAT GCA-3'	
HPA 4c	5'-CAG GGG TTT TCG AGG GCC T-3'	
HPA 5a	5'-AGG AAG AGT CTA CCT GTT TAC TAT CAA AG-3'	252 bp
HPA 5b	5'-AGG AAG AGT CTA CCT GTT TAC TAT CAA AA-3'	
HPA 5c	5'-CTC TCA TGG AAA ATG GCA GTA CAC T-3'	
HPA 6a	5'-GAC GAG TGC AGC CCC CG-3'	238/239 bp
HPA 6b	5'-GGA CGA GTG CAG CCC CCA-3'	
HPA 6c	5'-CCT ATG TTT CCC AGT GGT TGC A-3'	
HPA 9a	5'-CTC CTT TGC CCC CCC AGG-3'	185 bp
HPA 9bw	5'-CTC CTT TGC CCC CCC AGA-3'	
HPA 9c	5'-GAG AGC CTG CTC ACT ACG AG-3'	
HPA 15a	5'-TTC AAA TTC TTG GTA AAT CCT GG-3'	225 bp
HPA 15b	5'-TTC AAA TTC TTG GTA AAT CCT GT-3'	
HPA 15c	5'-ATG ACC TTA TGA TGA CCT ATT-3'	
HGH-1	5'-CAG TGC CTT CCC AAC CAT TCC CTT A-3'	429 bp
HGH-2	5'-ATC CAC TCA CGG ATT TCT GTT GTG TTT C-3'	

and 1.88 μM of HPA 5b and 1.62 μM of HPA 5c allele specific primers to mix “b”, and 0.5 U of Taq DNA Polymerase.

For HPA 6 genotyping, 1 μL of DNA was added to 24 μL of the amplification mixtures containing: 10X PCR reaction buffer, 2 mM MgCl_2 , 0.2 mM dNTP, 0.1 μM of each internal control primer, 0.2 μM of each allele-specific primer: HPA 6a or HPA 6b and HPA 6c and 0.5 U of Taq DNA Polymerase.

For HPA 9 genotyping, 0.5 μL of DNA were added to 9.5 μL of the amplification reaction mixture containing: 10X PCR reaction buffer, 4.5 mM MgCl_2 , 0.075 mM dNTP, 0.15 μM of each internal control primer, 1 μM of each allele-specific

primer HPA 9a or HPA 9bw and HPA 9c and 0.5 U of Taq DNA Polymerase.

For genotyping HPA 15, 2 μL of DNA were added to 8 μL of each reaction mixture containing: 10X PCR reaction buffer, 2 mM MgCl_2 , 0.2 mM dNTP, 0.072 μM of each internal control primer, 0.6 μM of each allele-specific primer, HPA 15a or HPA 15b primer and HPA 15c and 0.5 U of Taq DNA Polymerase.

Amplification conditions

Amplification of the HPA systems was carried out in a Biometra UNO II thermocycler. The conditions are detailed in Supplementary **Table SII**

	HPA-1-2-3			HPA-4 and HPA-5			HPA-6	
	1 Cycle	10 Cycles	22 Cycles	1 Cycle	10 Cycles	22 Cycles	1 Cycle	30 Cycles
Denaturalisation	95° 10´	95° 30´´	95° 30´´	95° 5´	95° 25´´	95° 25´´	95° 10´	95° 30´´
Annealing		65° 60´´	58° 60´´		68° 45´´	61° 45´´		58° 60´´
Extension		72° 30´´	72° 30´´		72° 30´´	72° 30´´		72° 30´´
Final extension	72° 4´			72° 10´			72° 10´	
	HPA-9			HPA-15				
	1 Cycle	10 Cycles	22 Cycles	1 Cycle	5 Cycles	20 Cycles	8 Cycles	
Denaturalisation	95° 5´	95° 25´´	95° 25´´	95° 5´	95° 25´´	95° 25´´	95° 25´´	
Annealing		63° 45´´	58° 45´´		70° 45´´	58° 45´´	51° 45´´	
Extension		72° 30´´	72° 30´´		72° 30´´	72° 30´´	72° 30´´	
Final extension	72° 7´			72° 7´				

°: degrees centigrade; ´ minutes; ´´ seconds