


WORKING INSTRUCTION

HLA-A*/-B*/-Cw*
CTS-PCR-SSP TRAY KITS

LOCUS- AND LOT-SPECIFIC MANUAL

To be applied to the following product:

Product No.	Description
120	HLA-A*/-B*/-Cw* CTS-PCR-SSP TRAY KIT 

HLA-A TYPING

Main differences between lot A12-4 (the current lot) and lot A12-3

- The primer composition of mix 1, 3 and 9 has been changed. However, there are no differences in mix specificities or PCR product sizes.

HLA-B TYPING

Main differences between lot B11-3 and lot B11-2

- Mix 20 has been modified. The allele specificity has changed. As a consequence of that, the amplification patterns of several HLA-B alleles (e.g. B*1506/1511/1515/ 1525/1536/1556/1576/1577) have been altered. Please specifically note the new amplification pattern of HLA-B*1511 (serologically B75). Mix 20 now generates only one (relatively small) fragment which is 140 bp and which should be carefully distinguished from the amplification control band (89 bp).

Main differences between Lot B12-0 (the current lot) and Lot B11-3:

- Almost every mix covers new alleles.
- Mix 10 has been altered to distinguish the B*37 alleles (including B*3706) from the common B*5108 allele. Mix 10 no longer detects B*0832, B*3538/87, B*4406, B*5108/20/36/44N.
- Mix 25 now also detects B*4406.

HLA-C TYPING

Main differences between lot C09-2 (the current lot) and lot C09-1

- The mixes 3, 4 and 5 have been modified to cover new HLA-C alleles published by the IMGT/HLA Database Release 2.21.0.
- The total number of mixes remains unchanged.

Introduction

- Intended use: This kit reveals a low/intermediate resolution typing of HLA-A, HLA-B and HLA-C by the PCR-SSP method.
- Allele coverage: IMGT/HLA Sequence Database Release 2.16.0, January 2007 for HLA-A (except for HLA-A*0318/0323 and A*110202/24); IMGT/HLA Sequence Database Release 2.24.0, January 2009 for HLA-B (except for HLA-B*0826, B*1322, B*1408, B*3817, B*3930/34/36 and B*5145); IMGT/HLA Sequence Database Release 2.21.0, April 2008 for HLA-C (except for HLA-Cw*0114 and Cw*0816). The alleles which are not covered are considered to be rare and can be detected by e. g. sequencing (you may contact us for further information on this topic).
- This manual is only valid for Lot No. **A12-4 B12-0 C09-2**.
- This manual should be used together with the Main Manual (General Information) 'Working Instruction for the CTS-PCR-SSP **TRAY and MINITRAY KITS**' (Manual No. 100A).

Table of contents

Section	Description	Page
Cover	Introduction	1
1.	Kit Composition	4
2.	Materials, Reagents and Equipment not supplied	4
3.	Sample Requirements, PCR and Gel Electrophoresis	4
4.	Result Evaluation	4
5.	Interpretation Hints	4
6.	Troubleshooting	7
7.	Precaution	7
8.	Contact	7
Appendix	Table 1 (HLA-A)	8
	Table 2 (HLA-B)	10
	Table 3 (HLA-C)	15
	Table 4 (HLA-A)	17
	Table 5 (HLA-B)	19
	Table 6 (HLA-C)	25

1. Kit Composition

- Number of PCR primer mixes per test: 96, of which:
 - 24 allele-specific mixes for HLA-A typing
 - 48 allele-specific mixes for HLA-B typing
 - 23 allele-specific mixes for HLA-C typing
 - 1 negative control mix
- Number of tests per tray: 1
- Number of trays per kit: 10
- The primer mixes are aliquoted and lyophilized in thin-walled, green PCR-trays.
- PCR buffer: 3 ml of 7.5% Mastermix (without Taq polymerase).

For storage condition, please refer to Section 1 of the ‘Working instruction for the CTS-PCR-SSP TRAY and MINITRAY KITS’ (Manual No. 100A) supplied along with this product.

2. Materials, Reagents and Equipment not supplied

Please refer to Section 2 of the ‘Working instruction for the CTS-PCR-SSP TRAY and MINITRAY KITS’ (Manual No. 100A) supplied along with this product.

3. Sample Requirements, PCR and Gel Electrophoresis

Please refer to Section 3 to 6 of the ‘Working instruction for the CTS-PCR-SSP TRAY and MINITRAY KITS’ (Manual No. 100A) supplied along with this product.

4. Result Evaluation

- 4.1 Check the approximate base pair size of the PCR product against the Primer Mix Specificity Table(s) (Appendix / Table 1, 2 and 3) to confirm the correct product size.
- 4.2 Use the Typing Table(s) (Appendix / Table 4, 5 and 6) to make the allele assignments or use the SCORE Software (www.IHWG.org) for detailed result interpretation.

5. Interpretation Hints

The quality and quantity of DNA as well as of the Taq polymerase are extremely crucial factors. If your bands are too weak, you might try to adjust these two factors until you obtain optimal results.

Please also refer to Section 7 of the ‘Working instruction for the CTS-PCR-SSP TRAY and MINITRAY KITS’ (Manual No. 100A) supplied along with this product

SPECIAL NOTES

HLA-A Locus:

- The following mixes will give **short** PCR products (< 200 bp): **mixes 17, 20, 23 and 24**. These PCR products might be difficult to be distinguished from the 89 bp amplification control (internal positive control) PCR product. In general, the allele-specific product will give a much stronger signal than that of the amplification control and will not migrate as far into the gel as the amplification control band. If you are not sure whether the (strong) signal represents an allele-specific product or just the amplification control, you might let the gel run for an additional 15 minutes at a lower voltage. By this way, the allele-specific band can be separated from the amplification control band and you will be able to clearly see a double signal at this position: a strong allele-specific band and - compared to that - a weaker shorter amplification control band. Since mix 5 corresponds to the frequent allele A*23, mix 17 to A*31 and mix 23 to A*02, it is recommended to always start the interpretation by checking these reactions very carefully first!
- Some mixes contain specific primers which may give rise to **PCR fragments of two or more different sizes** simultaneously (see Table 1). Please pay special attention to the following critical situations:
 - Mix 8** will amplify A*2501/02-06 at 400 bp. In addition, mix 8 will amplify A*3001-04/06-19 and A*0252 at 550 bp.
 - Mix 11** will amplify A*021701/02, A*2304, A*2403/10/18/22/33, A*2907 and A*9208 at 245 bp. A*2403 (serologically A2403) will be amplified only in mix 11 with a strong specific band. In addition, mix 11 will amplify A*3401/05 at 580 bp.
 - Mix 16** will amplify A*3001-04/06-11/13-17/19 at 570 bp and A*0112/19, A*0310, A*3202 at 720 bp.
 - Mix 18** will amplify A*3201-03/05-14, A*2303, A*2913, A*3107/08/10 at 410 bp. In addition, mix 18 will amplify A*2418 and A*3204 at 515 bp.
 - Mix 24** will amplify A*8001 at 170 bp and A*4301 at 445 bp.

HLA-B Locus:

- The following mixes will give **short** allele-specific PCR products (<200 bp): **mixes 5, 9, 20, 43, and 47**. These PCR products might be difficult to be distinguished from the 89 bp amplification control (internal positive control) PCR product. In general, the allele-specific product will give a much stronger signal than that of the amplification control and will not migrate as far into the gel as the amplification control band. If you are not sure whether the (strong) signal represents an allele-specific product or just the amplification control, you might let the gel run for an additional 15 minutes at a lower voltage. By this way, the allele-specific band can be separated from the amplification control band and you will be able to clearly see a double signal at this position: a strong allele-specific band and - compared to that - a weaker shorter amplification control band. Since three of these mixes correspond to quite frequent alleles (B*18 and B*35 = mix 5; B*27 = mix 9; B*15(62) = mix 43), it is recommended to start your evaluation by checking these four reactions very carefully first!
- Some HLA-B* mixes will amplify **two or even three fragments of different sizes** (e. g. mixes 9, 10, 15, 19, 25 and 35). Mixes with 2 fragments of almost the same size are not mentioned here.
- **Mix 7**
Some HLA-A specificities will cause a positive reaction with this mix
- **Mix 9**
All HLA-B*27 specificities except B*2712, 2716, 2718, 2723, 2726, 2729, 2731, 2739 and B* 2744

will amplify the **short** fragment at 155 bp. All B*27 specificities except B* 2706, 2707, 2711, 2720, 2721, 2724, 2727, 2733, 2734, 2735, 2743 as well as B*47010101-05 will give an **additional** positive signal at 525 bp.

- **Special notes to HLA-B*07**

The common HLA-B*07 alleles react positively with mix 1 and mix 28. Some HLA-B*07 alleles may cause additional weakly positive reactions with mix 24, especially if high amounts of Taq polymerase or high DNA-concentrations are used.

- **Special notes to HLA-B* 13**

B*1301 reacts positively with **mix 3**, whereas **B*1302** is amplified by **mix 3 and mix 34**.

The rare alleles **B*1303 and B*1304** can only be detected by **mix 34**.

Mix 34 may show **different PCR products for B*13**:

- **B*1304** will amplify **a smaller and a larger fragment** (425bp and 485bp).

- **B*130201/03/08Q/09/14-16/18/19** amplify **a smaller fragment** (425bp).

- **B*1310** amplifies **a larger fragment** (485bp).

HLA-C Locus:

- HLA-Cw*08 positive individuals give an allele-specific amplification band which is 165 bp long (mix 13). This band may be difficult to distinguish from the amplification control (internal positive control) band (89 bp). In general, the HLA-Cw*08-specific product will give a much stronger signal than that of the control band and it will not have migrated as far into the gel as the control band. By letting the gel run for additional 15 minutes at a lower voltage, the specific band will be separated from the amplification control band, and you will be able to clearly see a double signal at this position: a strong HLA-Cw*08-specific band and a weaker shorter amplification control band.
- The most common HLA-Cw*07 alleles will be detected by **two** PCR-SSP mixes, mix 11 and mix 12. HLA-Cw*0704/0711/0712/0745 are amplified by mix 11 with usual intensity and weakly by mix 12.
- PCR-SSP mix no. 5, 6, 12, 16 and 20 will also detect some HLA-B specificities (most of them are considered to be rare). Please take this into consideration when interpreting your results.

6. Troubleshooting

Please refer to Section 8 of the 'Working instruction for the CTS-PCR-SSP TRAY and MINITRAY KITS' (Manual No. 100A) supplied along with this product.

7. Precaution

Please refer to Material Safety Data Sheet for the CTS-PCR-SSP TRAY and MINITRAY KITS (Manual No. 100B) supplied along with this product.

8. Contact

If you have any particular questions concerning this kit, which are not answered in this or the Main Manual, please do not hesitate to contact me or my coworkers at:

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Table 1: Sizes of the PCR products and allele specificities for each HLA-A* CTS-PCR-SSP primer mix (**Lot-No A12-4**) based on IMGT/HLA Sequence Database Release 2.16.0, January 2007

Amplification Control (Internal Positive Control): 89 base pairs (bp)

Position on Tray	Primer mix	Allele specificity	Serology	Product size (bp)
H1	MIX 1 A12-4	A*01010101-04N/06-20	A1, Null, -	650
G1	MIX 2 A12-0	A*1117, A*3601-04	-, A36	580
F1	MIX 3 A12-4	A*02010101-22/24-3501/36-47/49/51/53N/54/56-59/61-63/66-79/81-87/89-97/99, A*9201/02/04/06-09	A2, A203, -, A210, Null	455
E1	MIX 4 A12-2	A*03010101-09/11N-17/19-22/25/26, A*1125, A*3204, A*3602:	A3, Null, -	see below
		A*0305/06, A*1125	-	555
		A*03010101-04/07-09/11N-17/19-22/25/26, A*3204, A*3602	A3, Null, -	580
D1	MIX 5 A12-0	A*2301-14, A*2424, A*2907	A23(9), -, Null	195
C1	MIX 6 A12-0	A*0246/48/70, A*2301/02/04-08N/10-14, A*24020101-07/09N-11N/13-15/17-20/22/23/25-28/30/32-41/43-66/68, A*3108:	-, A23(9), Null, A24(9), Low A24, A9	see below
		A*2417/41/62, A*3108	-	435
		A*0246/48/70, A*2301/02/04-08N/10-14, A*24020101-07/09N-11N/13-15/17-20/22/23/25-28/30/32-41/43-66/68	-, A23(9), Null, A24(9), Low A24, A9	470
B1	MIX 7 A12-0	A*24020101-0206/0208-0211/0213-0302/05/07-11N/14/15/17/20/21/23/25-27/29-31/33-43/45N-47/48N?/49-59/62-64/66-68	A24(9), Low A24, Null, A9, -	510
A1	MIX 8 A12-0	A*0252, A*250101/0102w/02-06, A*300101-04/06-19:	-, A25(10), A10, A30(19)	see below
		A*0252, A*300101-04/06-19	-, A30(19)	555
		A*250101/0102w/02-06	A25(10), A10, -	405
H2	MIX 9 A12-4	A*250101-06, A*260101-18/20-30/32-34, A*3401-08, A*6601-06:	A25(10), A10, -, A26(10), Null, A34(10), A66(10)	see below
		A*260701/0702	A26(10)	450
		A*250101-06, A*260101-06/08-18/20-30/32-34, A*3401-08, A*6601-06	A25(10), A10, -, A26(10), Null, A34(10), A66(10)	445
G2	MIX10 A12-0	A*260101/0103-32/34, A*4301:	A26(10), -, A10, Null, A43	see below
		A*2603/06/21/30	A26(10), -	410
		A*260101/0103-02/04/05/0701-20/22-29/31/32/34, A*4301	A26(10), -, A10, Null, A43	410
F2	MIX11 A12-0	A*021701/1702, A*2304, A*240301/0302/10/18/22/33, A*2907, A*3401/05, A*9208:	A2, -, Low A24, A9, A34(10)	see below
		A*3401/05	A34(10), -	585
		A*021701/1702, A*2304, A*240301/0302/10/18/22/33, A*2907, A*9208	A2, -, Low A24, A9	245

Position on Tray	Primer mix	Allele specificity	Serology	Product size (bp)
E2	MIX12 A12-0	A*2502, A*2613/19/33, A*3401/05/06, A*6601/02/04/06	A10, -, A34(10), A66(10)	415
D2	MIX13 A12-2	A*0324, A*1110, A*3402-04/07/08	-, A34(10)	385
C2	MIX14 A12-0	A*0212/13/19/27/37/38/44/49/54, A*0302w/10, A*110101-0201/03-16/18-22/23w/26-29, A*2419/28/44, A*6809/26/28	A2, -, A3, A11, Null, A69(28)	520
B2	MIX15 A12-0	A*2303, A*29010101-12/14-16, A*3107/08/10, A*3203, A*7410	-, A29(19), Null	430
A2	Mix16 A12-0	A*0112/19, A*0310, A*300101-04/06-11/13-17/19, A*3202:	-, A30(19), A32(19)	see below
		A*0112/19, A*0310, A*3202	-, A32(19)	725
		A*300101-04/06-11/13-17/19	A30(19), -	570
H3	MIX17 A12-0	A*2914, A*310102-07/09-15	-, A31(19), Null	185
G3	MIX18 A12-0	A*2303, A*2418/48N?, A*2913, A*3107/08/10, A*3201-14:	-, Null, A32(19)	see below
		A*2303, A*2913, A*3107/08/10, A*3201-03/05-14	-, A32(19)	410
		A*2418/48N?, A*3204	-, Null	520
F3	MIX19 A12-0	A*0265, A*3301/0301-09, A*7404	-, A33(19)	470
E3	MIX20 A12-0	A*7401-09/11/12N	A74(19), A19, -, Null	160
D3	MIX21 A12-3	A*020109/50, A*2505, A*680101-36, A*6901	A2, -, A68(28), A28, A69(28), Null	645
C3	MIX22 A12-0	A*0234-3502/56/62/78, A*6901, A*9203	-, A69(28)	390
B3	MIX23 A12-0	A*02010101-0108/0110/0111/0112w/02-22/24-3501/3502w/36-77/79-97/99, A*240206, A*3007w/13w/16w, A*9201-04/06-09:	A2, A203, -, A210, Null, A24(9)	see below
		A*0250	-	205/225
		A*02010101-0108/0110/0111/0112w/02-22/24-3501/36-44/47/49/51-54/57-60/62-77/79-91/93-97/99, A*9201/02/04/06-09	A2, A203, -, A210, Null	135
		A*02010101-0108/0110/0111/0112w/02-07/09-19/21/22/24-3501/3502w/36-49/51-54/57-61/63-77/79-97/99, A*240206, A*3007w, A*9201/02/04/06-09	A2, A203, -, A210, Null, A24(9)	165
		A*02010101-0102/0104-0108/0110/0111/0112w/02-10/12-22/24-34/36-47/49/51-68/70-77/79/80/82N-86/88N/89/91-97/99, A*3013w/16w, A*9201-04/06-09	A2, A203, -, A210, Null	190
A3	MIX24 A12-0	A*4301, A*8001:	A43, A80	see below
		A*8001	A80	170
		A*4301	A43	445

w = weak

? = nucleotide sequence information not available for the primer matching sequence

bold: mixes which result in PCR fragments of different sizes (the specificities are first indicated all in one row, then split into groups in the following rows depending on the fragment size)

Table 2: Sizes of the PCR products and allele specificities of each HLA-B* CTS- PCR-SSP primer mix (**Lot-No B12-0**)

based on IMGT/HLA Sequence Database Release 2.24.0, January 2009

Amplification control (internal positive control): 89 base pairs (bp)

Position on tray	Primer Mix	Allele specificity	Serology	Product size (bp)
H4	Mix 1	B*070201-07/08/09-1802/20-32/34-39/41-52/55-59/61-68, B*3566, B*4025, B*420501/0502w, B*4606, B*4806, B*8101-04N, Cw*0223, Cw*0738	B7, B703, -, Null, B81	see below
		B*070201-07/08/09-12/14-1802/20-32/34-39/41-52/55-59/61-68, B*3566, B*4025, B*420501/0502w, B*4806, B*8101-04N	B7, B703, -, Null, B81	625
		B*0713, B*4606, Cw*0223, Cw*0738	-	610
G4	Mix 2	B*0729, B*080101-02/03/04-25/27-38, B*3912, B*4449, B*9542	-, B8, Null, B39(16)	see below
		B*0809	-	540
		B*0729, B*080101-02/03/04-08N/10-25/27-38, B*3912, B*4449, B*9542	-, B8, Null, B39(16)	555
F4	Mix 3	B*1301-0204/06-21/23	B13, -, Null	580
E4	Mix 4	B*180101-15/17N-25/27/28/30-32, B*352002	B18, -, Null, B35	460
D4	Mix 5	B*0765, B*0832, B*180101-08/10-15/17N-32, B*350101-0801/0803-18/2001-24/28-45/48/50-55/57-62/64-72/74-94, B*3711/14, B*391901/1902, B*5606, B*7801-05	-, B18, Null, B35, B78	135
C4	Mix 6	B*350101-0402/05-0902/11/1401-15/17-19/21/23-25/27/29/30/32-38/40N-42/45-50/52-58/62/64-66/6801/6802/70/71/74/75/77/78/81/84/89-94, B*530101-06/08/10-16	-, B35, Null, B53	400
B4	Mix 7	A*250101-08, A*260101-06/08-18/20/21/23-30/33-37, A*340101/0102/05/06, A*6601-08, A*68020101-0202/15/18N/23/27/28/31/34/40/44, B*0712/14w/1801/1802w, B*1502/13/21/44/80w/88/89, B*350101-0103/0104w/0105-0403/06-0802/0803w/0901/0902/11/12w/1401-15/18-21/23-27/29/33-36/38/39w/40N-42/45-50/52-57/59w/61w/62/64/65Q/70/71/74/76-78/81-85/88/90-94, B*3708, B*3942w, B*4068, B*4406w/12w, B*5104/42w/46w/56w, B*530101-13/15/16, B*5609/11/12w, B*8301w, B*9512w/21/39/44	A25(10), A10, -, A26(10), Null, A34(10), A66(10), A68(28), B75(15), B77(15), B35, B44(12), B51(5), B53	375
A4	Mix 8	B*1513, B*2730w, B*4406w/25w/50w, B*5104/42w/46w/56w, B*530101-02/04-06/08/10/15/16, B*5705, B*580101/0102/04/05/09-13/17N/19	B77(15), -, B44(12), B51(5), B53, B58(17), Null	325
H5	Mix 9	B*2701-21/23-45, B*47010101-05	B27, B2708, -, B47	see below
		B*2702-11/13-15/17/19-21/24/25/27/28/30/32-38/40-43/45	B27, B2708, -	155
		B*2701-0510/08-10/12-19/23/25/26/28-32/36-40/42/44/45, B*47010101-05	B27, B2708, -, B47	525
G5	Mix 10	B*370101-06/08-14	B37, -, Null	see below
		B*370101-0104/3703N-06/08-14	B37, -, Null	380
		B*3702	-	500

Position on tray	Primer Mix	Allele specificity	Serology	Product size (bp)
F5	Mix 11	A*2496, B*3710, B*380101-16, B*511301/1302, B*5214, B*5304/07, B*5820, B*5903	-, B38(16), B16	390
E5	Mix 12	B*0745w, B*1548w/69, B*3535, B*39010101-20/22-29/31/32w/35/37-42/44-46, B*670101-02, B*9508w/36w	-, B62(15), B3901, B3902, B39(16), B16, Null, B67	515
D5	Mix 13	B*670101/0102	B67	545
C5	Mix 14	B*400101-04/060101-11/13-16/18-20/22N-25/27/29-38/40/42-45/47-50/52-62/64-70/7201-91/94, B*4416/21/31/6401/6402, B*47010101-05	B60(40), B61(40), B40, -, Null, B47	635
B5	Mix 15	B*0727w/50w, B*0804w/17, B*1321, B*1507/33/45/55w/68/78, B*1814w/21w/22w, B*3572w, B*3707/09/12, B*400101-0602/09-12/1401-16/18-24/25w/27/29-40/42-60/62-67/69-74/76/78-86/88-94, B*4101-08, B*4431/53/54, B*480101/0102/0301-05/07-19, B*9507/16/26/41/50	-, B62(15), B15, B60(40), B61(40), B40, B4005, Null, B41, B48	see below
		B*0727w/50, B*0804/17, B*1507/45/55w/68, B*1814w/22w, B*3572w, B*3707/09, B*400201-03/05/09/15/16/18/19/27/29/31/32/35/37/39/40/45/50/56-58/71/78/80/82/85/89-91/94, B*4102/04, B*4454, B*480101/0102/04/05/07-16/18/19, B*9526	-, B62(15), B61(40), B40, B4005, B41, B48	375
		B*0727w/50, B*0804/17, B*1321, B*1533/78, B*1821, B*3707/09/12, B*400101-0602/09-12/1401-16/18-24/25/27/29-34/36-40/42-60/62-67/69/70/7201-74/76/78-81/83-86/88-94, B*4101-08, B*4431/53, B*480101/0102/0301-05/07-11/13/15-19, B*9507/16/41/50	-, B15, B60(40), B61(40), B40, B4005, Null, B41, B48	475
A5	Mix 16	B*2724/36, B*400101-0106/07/22N/23/25/30/31/33/34/36/38/42/43/45-48/51/52/54/55/59-63/65-67/69/73/74/76/79-81/84/87/88/92	-, B60(40), Null	685
H6	Mix 17	B*1546/53, B*3519/47/63, B*3706, B*400201-0602/08/09/11/13-16/18-20/24/26-29/32/35/37/39/40/44/50/53/56-58/64/68/70-7202/75/77/78/82/83/86/89-91/94, B*4101-08, B*44020101-05/07/09-30/32-39/41-43/45-51/53-56N/58N/59/61N-65, B*4501-09, B*47010101-05, B*490101-02/04/05, B*5001/02/04, B*9506/43	B72(70), -, B35, B61(40), B40, B4005, B41, B44(12), B12, Null, B45(12), B47, B49(21), B50(21)	575
G6	Mix 18	B*2718/29, B*3702, B*47010101-05	-, B47	420
F6	Mix 19	B*0712/14w/1801/1802w, B*1562/80w, B*2719w/24/30w/36, B*3525/26/49, B*3942w, B*4012/46, B*4440w/44w, B*480101-04/06/07/09/11/15-19, B*5609/11/12w, B*8101-04N, B*8301w, B*9524	-, B48, B81, Null	
		B*0712/14w/1801/1802w, B*1562/80w, B*2719w/30w, B*3525/26/49, B*3942w, B*4440w/44w, B*4802/17, B*5609/11/12w, B*8301w	-	430
		B*2724/36, B*4012/46, B*480101/0102/0301-04/06/07/09/11/15-19, B*8101-04N, B*9524	-, B48, B81, Null	575
E6	Mix 20	B*1502/08/1101-1103/13/15/21/31/44/55/76/88/89, B*3546, B*9512/21/39/44/48	B75(15), B77(15), B62(15), -	140

Position on tray	Primer Mix	Allele specificity	Serology	Product size (bp)
D6	Mix 21	B*0709/11/17, B*0828/35/37, B*1503/18/23/29/47/49/51/52/54/61/62/64/68/69/72/74/80/91/93/98, B*2741, B*3525/26/49, B*3907/43, B*4209, B*4802/14, B*5514/23/32, B*5603/09/18, B*9503/08/14/15/19/23/24/27/31/32/34/51, Cw*0746	B7, -, B72(70), B71(70), B15, B22	495
C6	Mix 22	B*4101-07, B*4202/09	B41, -, B42	790
B6	Mix 23	B*0704/19/25, B*4201/02/04-09, B*4506, B*5520, B*5613, B*8201/02, B*8301	B7, -, B42	see below
		B*0704/19/25, B*4201/02/04-09, B*4506, B*5520, B*5613, B*8201/02, B*8301	B7, -, B42	550
		B*0704/19/25, B*4201/02/04-06/08/09, B*4506, B*5520, B*5613, B*8201/02, B*8301	B7, -, B42	540
A6	Mix 24	B*070201w-04w/07w-10w/12w/14w-16w/1801w-31w/33w/35w-38w/41w-52w/54w- 64w/66w-68w, B*0807w, B*1510w/37w/99w, B*2706w/21w/34w, B*3929w, B*4009w/18w/24w/31w-33w/42w/44w, B*4104w, B*44020101-05/07/09/11-13/16/19N- 30/32-42/44-56N/58N/59/61N/63-65, B*4808w, B*5527w, B*8301w	B7, B703, -, Null, B71(70), B27, B61(40), B44(12), B12	490
H7	Mix 25	B*0720, B*1514/91, B*3545/71, B*4406/09/34/46, B*4501-03/05-07/09, B*4617, B*5002, B*5123/42, B*8201/02, B*8301, B*9531	-, B76(15), B44(12), B12, B45(12)	see below
		B*4406/34, B*5123/42	B44(12), -	715
		B*0720, B*1514/91, B*3545/71, B*4409/46, B*4501-03/05-07/09, B*4617, B*5002, B*8201/02, B*8301, B*9531	-, B76(15), B12, B45(12)	545
G7	Mix 26	B*1546/53, B*4005/15/16/23/26/28/32/51, B*490101/02/04/05, B*5001/02/04, B*9506/43	B72(70), -, B4005, B61(40), B49(21), B50(21), B45(12)	605
F7	Mix 27	B*1309, B*1542/73/86, B*3560, B*3917, B*4048/71, B*4501/03-08, B*4611/18, B*5001/02/04, B*5401-03/05N/07/08N/10/13/16/17, B*550101-03/05/07/09- 12/15/16/18/19/21/22/24-26/29-31/33/34, B*5601/02/04/08/10/13/14/16/17/19N/20/23/24, B*8201/02, Cw*0312/19, Cw*1215, Cw*1507	-, B45(12), B50(21), B54(22), Null, B55(22), B22, B56(22)	390

Position on tray	Primer Mix	Allele specificity	Serology	Product size (bp)
E7	Mix 28	B*070201-0207/04-07/09w/10/11w/12/15/1801-26/29-31/33-36/39-49N/51-68, B*1576w, B*3576w, B*4201/02/04-06/08/09, B*4506, B*5401-05N/06w/07w/08N/09w/10-13/14w/16/17, B*550101-05/07-17/19-22/23w/24-26/27w/28-31/32w/33/34, B*5601/02/03w/04-07/09w/10/11w/12/13/15/16/18w/19N-21/22w/24, B*670101/0102, B*8101-04N, B*8201/02, B*9501w	B7, -, Null, B42, B54(22), B55(22), B22, B56(22), B67, B81	see below
		B*070201-0207/04-07/09w/10/11w/12/15/1801-26/29-31/33-36/39-49N/51-68, B*1576w, B*3576w, B*4201/02/04-06/08/09w, B*5404/06w/07w/09w/11/14w, B*550104/04/08/13/14/17/20/23w/27w/28/32w/34, B*5603w/0501-06/09w/11w/12/15/18w/21/22w, B*670101/0102, B*8101-04N, B*9501w	B7, -, Null, B42, B55(22), B22, B67, B81	410
		B*4506, B*5401-03/05N/07?/08N/10/12/13/16/17, B*550101-03/05/07/09-12/15/16/19/21/22/24-26/29-31/33/34, B*5601/02/04/07/10/13/16/19N/20/24, B*8201/02	-, B54(22), Null, B55(22), B22, B56(22)	420
D7	Mix 29	B*5401-05N/07/08N/10-17, B*5507	B54(22), -, Null	430
C7	Mix 30	B*0720w/24/60, B*1576, B*3576, B*4506w, B*5403/06, B*5508, B*5601-07/09/11/13/15/16/20-22/24, B*8201w/02w, B*8301w, B*9501	-, B56(22), B22	560
B7	Mix 31	B*4418, B*490101/0102/03, B*5115/62, B*5412, B*5901/04	-, B49(21), B59	390
A7	Mix 32	B*1571, B*1815/19/21/30, B*270401/0402/06/10/15/18/20/21/24/25/40, B*3707, B*4005/15/16/23/26/28/32/51, B*4614, B*490101/02-05, B*5001/02/04	-, B27, B4005, B61(40), B49(21), B50(21), B45(12)	715
H8	Mix 33	B*460101-05/07N-18, B*5614, B*6702	B46, -, Null	470
G8	Mix 34	B*130201-04/08Q-10/14-16/18/19, B*15010101-02/04-08/1101-16/19-21/24-28/31-36/38-40/42-45/50/55-57/60/63/65-67/70/71/75-79N/81-85/87-89/92/94N-97, B*3546, B*4408/57/60, B*460101-03/05-18, B*5202, B*570101/0103/04/06/08/10/11/13-15/18/19, B*9501/02/04/05/07/09-13/16-18/20-22/25/26/28/29/35-42/44-49N	B13, -, B62(15), Null, B75(15), B76(15), B77(15), B63(15), B15, B44(12), B46, B57(17)	see below
		B*130201-04/08Q/09/14-16/18/19, B*1504/42/83, B*4611/18, B*5202, B*9537	B13, -, B62(15)	430
		B*1304/10, B*15010101-02/04-08/1101-16/19-21/24-28/31-36/38-40/43-45/50/55-57/60/63/65-67/70/71/75-79N/81/82/84/85/87-89/92/94N-97, B*3546, B*4408/57/60, B*460101-03/05-10/12-17, B*570101/0103/04/06/08/10/11/13-15/18/19, B*9501/02/04/05/07/09-13/16-18/20-22/25/26/28/29/35/36/38-42/44-49N	-, B62(15), Null, B75(15), B76(15), B77(15), B63(15), B15, B44(12), B46, B57(17)	485
F8	Mix 35	B*570101-10/12-19, B*5814	B57(17), -	see below
		B*570101-04/06-10/12-19, B*5814	B57(17), -	360
		B*5704/05	B57(17), -	405
Position	Primer	Allele specificity	Serology	Product

on tray	Mix			size (bp)
E8	Mix 36	B*5714, B*580101-02/04-20	-, B58(17), Null	485
D8	Mix 37	B*1513/16w/67w/95w, B*2730w, B*4406w/18w/25w/50w, B*490101w/0102w/03w-05w, B*510101-05/07-2402/26-40/42w/43/44N/46w/48-53/55/56w/57/58/60/61/63, B*520101-14, B*530101-02/04-06/08/10/15/16, B*5412, B*5621, B*5705, B*580101/0102/04/05/08-13/17N/19, B*5901-04	B77(15), B63(15), -, B44(12), B49(21), B51(5), B5102, B5103, Null, B52(5), B53, B58(17), B59	325
C8	Mix 38	B*350402, B*510101-0103/0105-0203/03-06/08-0902/11N/12/1302/14/16-21/23/26-41N/43/46/48-61/63, B*7801-0202/04	B35, B51(5), B5102, B5103, Null, -, B52(5), B78	445
B8	Mix 39	B*070503, B*1509, B*350402, B*510101-0103/0105-0203/03-0902/11N/12/1302/14/16-23/26-41N/43/44N/46/48-61/63, B*550104, B*560501-06/21, B*7801-04	B7, B70, B35, B51(5), B5102, B5103, Null, -, B52(5), B55(22), B78	455
A8	Mix 40	B*150102, B*400106/26/28, B*4462, B*5107w, B*520101-05/07-13, B*7805/06	B62(15), B60(40), -, B51(5), B52(5)	445
H9	Mix 41	B*070503, B*150102/09, B*350402, B*400106/26/28, B*550104, B*560501-06, B*7801-06	B7, B62(15), B70, B35, B60(40), -, B55(22), B78	405
G9	Mix 42	B*1509/10/18/21/23/37/51/72/80/90/99, B*3526/82/85, B*3932/33, B*5122, B*7803, B*9508/15/19/33/34	B70, B71(70), B75(15), -	570
F9	Mix 43	B*1309, B*15010101-02/04-08/1101-12/14/15/19-21/25-28/30-35/38-40/42/44/45/48/50/55/56/58/60/63/65/66/70/71/73/75-79N/81-86/88/92/94N/96/97, B*3546, B*4021, B*5712, B*9501/02/04/05/07/10-13/16-18/20-22/25/26/28/29/35-42/44-50	-, B62(15), Null, B75(15), B76(15), B15	130
E9	Mix 44	B*1512/14/19, B*4408/57/60, B*4617, B*5707	B76(15), B44(12), -	645
D9	Mix 45	B*1516-1702/67/95, B*5713, B*5806/08/19	B63(15), -	525
C9	Mix 46	B*1401-04/07N/09	B64(14), B65(14), -, Null	400
B9	Mix 47	B*140201-03/05-0602/09, B*3526, B*3805, B*3904	B65(14), -, B39(16)	185
A9	Mix 48	B*7301	B73	295

w = weak

bold: mixes which result in PCR fragments of different sizes (the specificities are first indicated all in one row, then split into different groups in the subsequent rows depending on their fragment size)

Table 3: Sizes of the PCR products and allele specificities of each **HLA-Cw*** CTS-PCR-SSP Primer Mix (Lot-No C09-2) based on IMGT/HLA Sequence Database Release 2.21.0, April 2008

Amplification control (internal positive control): 89 base pairs (bp)

Position on tray	Primer mix	Allele specificity	Serology	Product size (bp)
H10	Mix 1	Cw*010201-13/15-20, 'B*3907?	-, Cw1	245
G10	Mix 2	Cw*020201-11/12w/13-21, Cw*0432, Cw*0608, Cw*1701-04, Cw*1803	Cw2, -	530
F10	Mix 3	Cw*030201-15/17-40/42-48	Cw10(w3), Cw9(w3), -, Null	570
E10	Mix 4	Cw*030201-06/08-13/15-24/26-44/46-48, Cw*0411/29, Cw*140201/0203-03/05-09	Cw10(w3), Cw9(w3), -, Null	440
D10	Mix 5	B*1596, Cw*030201-0203/0401-10/14/17/19/23-29/33-3802/40/42/44/46-48	-, Cw10(w3)	530
C10	Mix 6	B*6702, Cw*04010101-0105/03-14/1502-20/23-32, Cw*1210	-, Cw4, Null	470
B10	Mix 7	Cw*041501	-	540
A10	Mix 8	Cw*050101/0103/0104/03-19, Cw*0810	Cw5, -, Null	425
H11	Mix 9	Cw*0113, Cw*050101-0104/03-11/13-17/19, Cw*0741w, Cw*0802/04/05/07/12/13/15:	-, Cw5, Null, Cw8	see below
		Cw*050101-0104/03-11/13-17/19, Cw*0802/04/05/07/12/15	-, Cw5, Null, Cw8	590
		Cw*0113, Cw*0741w, Cw*0802/04/05/07/12/13/15	-, Cw8	465
G11	Mix 10	Cw*06020101-14/15?/16N/17, Cw*0731	Cw6, -, Null	305
F11	Mix 11	Cw*070101-21/22w/23-25/2701-32N/35-40/42-54	Cw7, -, Null	715
E11	Mix 12	B*0751, Cw*070101-03/0401w/0402w/05-10/11w/12w/13-19/21/22w/23-29/31-33N/35-40/42-44/45w/46-54, Cw*0814	-, Cw7, Null	670
D11	Mix 13	Cw*080101-09/11/12/14/15	Cw8, -	165
C11	Mix 14	Cw*0104w, Cw*120201-0203/08/10/16/18/19:	-	see below
		Cw*1219	-	370
		Cw*0104w, Cw*120201-0203/08/10/16/18	-	460
B11	Mix 15	Cw*0104, Cw*120201-030102/0303-0306/0402-08/10-13/15-18/20/21	-	420
A11	Mix 16	B*0713/15, B*6702, Cw*0212, Cw*0315/27/3801/3802, Cw*0403w/06w/16w, Cw*0603w, Cw*0726, Cw*0805, Cw*120201-0306/0401w/0402w/06-08/10-15/18-20, Cw*1503w/16w, Cw*1701w-04w	-	230
H12	Mix 17	Cw*0205, Cw*0516, Cw*06020101-10/12-14/15?/16N/17, Cw*120401-05/09, Cw*1602/09	-, Cw6, Null	330

Position on tray	Primer mix	Allele specificity	Serology	Product size (bp)
G12	Mix 18	Cw*020201-0206/04-11/12w/13-15/17/19-21, Cw*0508, Cw*06020101-03/07-10/12-17, Cw*120401-05/21, Cw*1803	Cw2, -, Cw6, Null	510
F12	Mix 19	Cw*041501, Cw*140201-0203/0204w/03-09	-, Null	565
E12	Mix 20	B*350802, Cw*0510, Cw*150201-13/15-21	B35, -	435
D12	Mix 21	Cw*160101-02/0401/06-08/09/10/11	-	470
C12	Mix 22	Cw*1701-04:	-	see below
		Cw*1701-04	-	915
		Cw*1704	-	605
B12	Mix 23	Cw*1801-03, Cw*0708	-	555
A12	Mix 24	Negative Control	-	-

w = weak

? = nucleotide sequence information not available for the primer matching sequence

bold: mixes which result in PCR fragments of different sizes (the specificities are first indicated all in one row, then split into different groups in the subsequent rows depending on their fragment size)

Table 4: Amplification patterns for all detectable HLA-A specificities (Lot-No A12-4) based on IMGT/HLA Sequence Database Release 2.16.0, January 2007

Allele	Serology	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
A*01010101-04N/06-11N/13-18N/20	A1, Null, -	+																							
A*0112/19	-	+															+								
A*02010101-0108/0110/0111/02-11/14-16/18/2001-22/24-26/28-33/36/39-43N/45/47/51/53N/57-59/61/63/66-69/71-77/79/81-87/89-97/99, A*9201/02/04/06/07/09	A2, A203, -, A210			+																				+	
A*020109	A2			+																		+			
A*020112	A2			+																					W
A*0212/13/19/27/37/38/44/49/54	A2, -			+											+										+
A*021701/1702, A*9208	A2, -			+								+													+
A*0234/3501/56/62	-			+																				+	+
A*023502	-																							+	W
A*9203	-																							+	+
A*0246/70	-			+				+																	+
A*0248	-							+																	+
A*0250	-																						+		+
A*0252	-																								+
A*0255/60/64/80/88N	-																								+
A*0265	-																								+
A*0278	-			+																				+	
A*03010101-0105/03N-09/11N-17/19-22/25/26, A*1125	A3, Null, -				+																				
A*0302	A3				+																				
A*0310	-																								
A*0324	-																								
A*110101-0201/03-09/11-16/18-22/26-29	A11, -																								
A*1110	-																								
A*1117, A*3601/03/04	-, A36			+																					
A*1123	-																								
A*2301/02/05-08N/10-14	A23(9), -					+	+																		
A*2303	-					+											+								
A*2304	-					+	+																		
A*2309, A*2424	-					+							+												
A*24020101-0205/0208-0211/0213/05/07/09N/11N/14/15/17/20/23/25-27/30/34-41/43/45N-47/49-59/62-64/66/68	A24(9), -						+	+																	
A*240206	A24(9)						+	+																	+
A*240207/0212/04/06/13/32/60N/61/65	A24(9), -						+																		
A*240301/0302/10/33	Low A24, A9, -						+	+					+												
A*2408/21/29/31/42/67	A24(9), -							+																	
A*2418	-						+						+												+
A*2419/28/44	-						+								+										
A*2422	-						+						+												
A*2448N	Null						+	?																	?
A*250101/03/04/06	A25(10), -									+	+														
A*250102	A25(10)									W	+														

Allele	Serology	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
A*2502	A10								+	+			+												
A*2505	-								+	+												+			
A*260101/0103-12/14-18/20-30/32/34	A26(10), -, A10									+	+														
A*260102, A*6603/05	A26(10), A10, -									+															
A*2613	-									+	+		+												
A*2619	-											+	+												
A*2631	-										+														
A*2633, A*3406, A*6601/02/04/06	-, A66(10)									+			+												
A*29010101-06/08N-12/15/16, A*7410	A29(19), Null, -															+									
A*2907	-					+						+				+									
A*2913, A*3201/05-14	-, A32(19)																			+					
A*2914	-															+		+							
A*300101-04/06/08-11/14L/15/17/19	A30(19), -									+							+								
A*3007/13/16	-									+							+							w	
A*3012/18	-									+															
A*310102-06/09/11-15	A31(19), -																		+						
A*3107/10	-															+		+	+						
A*3108	-					+										+			+						
A*3202	A32(19)																+		+						
A*3203	-															+			+						
A*3204	-				+														+						
A*3301/0301-09	A33(19), -																				+				
A*3401/05	A34(10), -									+		+	+												
A*3402-04/07/08	A34(10), -									+				+											
A*3602	-		+		+																				
A*4301	A43										+														+
A*680101-08/10-25/27/29-36	A68(28), A28, -																					+			
A*6809/26/28	A69(28), -														+							+			
A*6901	A69(28)																					+	+		
A*7401-03/05-09/11/12N	A74(19), A19, -																					+			
A*7404	-																				+	+			
A*8001	A80																								+

w = weak

? = nucleotide sequence information not available for the primer matching sequence

Table 5: Amplification patterns for all **HLA-B** alleles detected by the **HLA-B*** CTS-PCR-SSP reagents (**Lot-No B12-0**) based on IMGT/HLA Sequence Database Release 2.24.0, January 2009

Allele	Serology	Mix
A*250101-08, A*260101-06/08-18/20/21/23-30/33-37, A*340101/0102/05/06, A*6601-08, A*68020101-0202/15/18N/23/27/28/31/34/40/44, B*5309	A25(10), A10, -, A26(10), A34(10), A66(10), A68(28)	7
B*070201-0207/07/10/15/21-23/26/30/31/35/36/41-44/46-49N/51/52/55-59/61-64/66-68	B7, -	1+24w+28
B*0703/08/16/28/37/38	B703, -	1+24w
B*0704/25	B7, -	1+23+24w+28
B*070501/0502/06/34/39	B7, -	1+28
B*070503	B7	1+28+39+41
B*0709	B7	1+21+24w+28w
B*0711	B7	1+21+28w
B*0712/1801	-	1+7+19+24w+28
B*0713/32, Cw*0223, Cw*0738	-	1
B*0714	-	1+7w+19w+24w
B*0717	-	1+21
B*071802	-	1+7w+19w+24w+28
B*0719	-	23+24w+28
B*0720	-	1+24w+25+28+30w
B*0724	-	1+24w+28+30
B*0727/50	-	1+15w+24w
B*0729	-	1+2+24w+28
B*0733/54	-	24w+28
B*0740/53, B*5504/13/17/28	-, B55(22)	28
B*0745	-	1+12w+24w+28
B*0760	-	24w+28+30
B*0765	-	1+5+28
B*080101-03/05/06/08N-16/18-25/27/29-31/33/34/36/38	B8, -	2
B*0804	-	2+15w
B*0807	-	2+24w
B*0817	-	2+15
B*0828/35/37	-	2+21
B*0832	-	2+5
B*1301/06/07N/11-13/17/20/23	B13, -	3
B*130201-0204/08Q/10/14-16/18/19	B13, -	3+34
B*1303/04, B*1524/36/43/57/87, B*5711, B*9509	-, B62(15)	34
B*1309	-	3+27+34+43
B*1321	-	3+15
B*1401/04/07N	B64(14), -	46
B*140201-03/09	B65(14), -	46+47
B*1405-0602	-	47
B*15010101/010102N/0103-0108/04-06/20/25-28/32/34/35/38-40/50/56/60/63/65/66/70/75/77/79N/81-85/92/94N/96/97, B*9502/04/05/10/11N/13/17/18/20/22/25/28/29/35/37/38/40/45-47/49N	B62(15), Null, B15, -	34+43
B*150102	B62(15)	34+40+41+43
B*1502/44/88, B*9512/21/39/44	B75(15), -	7+20+34+43

Allele	Serology	Mix
B*1503/29/47/49/52/54/61/64/74/93/98, B*3943, B*9503/14/23/27/32/51, Cw*0746	B72(70), B15, -	21
B*1507/33/45/78, B*9507/16/26/41	B62(15), B15, -	15+34+43
B*1508/1101-1103/15/31, B*9548	B75(15), B62(15), -	20+34+43
B*1509	B70	39+41+42
B*1510/37/99	B71(70), -	24w+42
B*1512/19	B76(15)	34+43+44
B*1513	B77(15)	7+8+20+34+37
B*1514	B76(15)	25+34+43+44
B*1516/67/95	B63(15)	34+37w+45
B*15170101-1702	B63(15)	45
B*1518/23/51/72, B*9515/19/34	B71(70), -	21+42
B*1521	B75(15)	7+20+34+42+43
B*1524/36/43/57/87, B*1303/04, B*5711, B*9509	B62(15), -	34
B*1530/58	B62(15), -	43
B*1542	-	27+34+43
B*1546/53, B*9506/43	B72(70), -	17+26
B*1548	B62(15)	12w+43
B*1555	-	15w+20+34+43
B*1562, B*4802, B*9524	-	19+21
B*1568, B*4814	-	15+21
B*1569, B*3907	-	12+21
B*1571	-	32+34+43
B*1573/86	-	27+43
B*1576	-	20+28w+30+34+43
B*1580	-	7w+19w+21+42
B*1589	-	7+20+34
B*1590, B*3933, B*9533	-	42
B*1591, B*9531	-	21+25
B*180101-08/10-13/17N/18/20/23N-25/27/28/31/32	B18, -	4+5
B*1809	-	4
B*1814/22	-	4+5+15w
B*1815/19/30	-	4+5+32
B*1821	-	4+5+15w+32
B*1826/29, B*3510/13/16/22/28/31/43/44/51/67/69/79/80/86/87	-, B35	5
B*2701-03/0502-0510/07-09/11-14/16/17/23/26-28/31-33/35/37-39/42-45	B27, B2708, -	9
B*270401/0402/10/15/20/25/40	B27, -	9+32
B*2706/21	B27, -	9+24w+32
B*2718	-	9+18+32
B*2719	-	9+19w
B*2724	-	9+16+19+32
B*2729	-	9+18
B*2730	-	8w+9+19w+37w
B*2734	-	9+24w
B*2736	-	9+16+19
B*2741	-	9+21
B*350101-0103/0105-0401/06-0801/0901/0902/11/1401-15/18/21/23/24/29/33/34/36/38/40N-42/48/50/52-55/57/62/64/65Q/70/74/77/78/81/84/90-94	-, B35	5+6+7

Allele	Serology	Mix
B*350104/0803	-	5+6+7w
B*350402	B35	5+6+7+38+39+41
B*350403/2001/83/88	B35, -	5+7
B*3505/17/30/32/37/58/6801/6802/75/89	B35, -	5+6
B*350802/27/56, B*5303/11-13	B35, -	6+7
B*3512/39/59/61	B35	5+7w
B*3519/47	B35, -	6+7+17
B*352002	B35	4+5+7
B*3525/49	-	6+7+19+21
B*3526	-	7+19+21+42+47
B*3535	-	5+6+7+12
B*3545/71	-	5+6+7+25
B*3546	-	6+7+20+34+43
B*3560	-	5+27
B*3563, B*4410/14/15/17/43	-	17
B*3566	-	1+5+6
B*3572	-	5+15w
B*3576	-	5+7+28w+30
B*3582/85	-	5+7+42
B*370101-0104/03N-05/13	B37, -	10
B*3702	-	10+18
B*3706	-	10+17
B*3707	-	15+32
B*3708	-	7+10
B*3709/12	-	10+15
B*3710	-	10+11
B*3711/14	-	5+10
B*380101-04/06-16, A*2496	-, B38(16), B16	11
B*3805	-	11+47
B*39010101-03/05-0602/08-11/1301-16/18/20/22-28/31/35/37-41/44-46	B3901, B3902, B39(16), B16, -	12
B*3904	B39(16)	12+47
B*3907, B*1569	-	12+21
B*3912	B39(16)	2+12
B*3917	-	12+27
B*391901/1902	-	5+12
B*3929	-	12+24w
B*3932	-	12w+42
B*3942	-	7w+12+19w
B*400101-0105/22N/30/34/36/38/43/45/47/52/54/55/59/60/62/65-67/69/73/74/76/79-81/84/88	B60(40), -	14+15+16
B*400106	B60(40)	14+15+16+40+41
B*400201-04/060101-0602/11/1401-1403/19/20/27/29/35/37/40/50/53/56-58/64/70/7201/7202/78/82/83/86/89-91/94	B61(40), B40, -	14+15+17
B*4005	B4005	15+17+26+32
B*4007/61/87	-	14+16
B*4008/13/75/77	-	14+17
B*4009/18/24/44	B61(40), -	14+15+17+24w
B*4010/49/85, B*4431	B60(40), -	14+15
B*4012, B*480101/0102/0301-04/07/09/11/15-19	-, B48	15+19
B*4015/16	-, B61(40)	14+15+17+26+32
B*4021, B*9550	-	15+43

Allele	Serology	Mix
B*4023	-	14+15+16+26+32
B*4025	-	1+14+15w+16
B*4026/28	-	17+26+32+40+41
B*4031/33/42	-	14+15+16+24w
B*4032	-	14+15+17+24w+26+32
B*4039, B*4108	-	15+17
B*4046	-	15+16+19
B*4048	-	14+15+16+27
B*4051	-	15+16+26+32
B*4063/92	-	15+16
B*4068	-	7+14+17
B*4071	-	15+17+27
B*4093, B*4805/10/12/13	-, B48	15
B*4101-0302/05-07	B41, -	15+17+22
B*4104	-	15+17+22+24w
B*4201/04/06/08, B*5520	B42, -	23+28
B*4202	B42	22+23+28
B*420501	-	1+23+28
B*420502	-	1w+23+28
B*4207	-	23
B*4209	-	21+22+23+28w
B*44020101-05/07/11/13/19N/20/22-24/26-30/32/33/35-39/41/42/45/47/48/51/55/56N/58N/59/61N/63/65	B44(12), -	17+24
B*4406	B44(12)	7w+8w+25+37w
B*4408/57/60	B44(12), -	34+44
B*4409/34/46	B12, -	17+24+25
B*4410/14/15/17/43, B*3563	-	17
B*4412	-	7w+17+24
B*4416/21/6401/6402	-	14+17+24
B*4418, B*490102	-	17+31+37w
B*4425/50	-	8w+17+24+37w
B*4440/44	-	19w+24
B*4449	-	2+17+24
B*4452N	-	24
B*4453/54	-	15+17+24
B*4462	-	17+40
B*4501/03/05/07	B45(12), -	17+25+27
B*4502/09	-	17+25
B*4504/08	-	17+27
B*4506	-	17+23+25+27+28+30w
B*460101-03/05/07N-10/12/13/15N/16	B46, -	33+34
B*4604	-	33
B*4606	-	1+34
B*4611/18	-	27+33+34
B*4614	-	32+33+34
B*4617	-	25+33+34+44
B*47010101-05	B47, -	9+14+17+18
B*480101/0102/0301-04/07/09/11/15-19, B*4012	B48, '-	15+19
B*4802, B*1562, B*9524	-	19+21
B*4805/10/12/13, B*4093	B48, '-	15
B*4806	-	1+19

Allele	Serology	Mix
B*4808	-	15+24w
B*490101	B49(21)	17+26+31+32+37w
B*490102, B*4418	B49(21), '-	17+31+37w
B*4902	-	17+26+32
B*4903	-	31+32+37w
B*4904/05	-	17+26+32+37w
B*5001/04	B50(21), -	17+26+27+32
B*5002	B45(12)	17+25+26+27+32
B*510101-0103/0105-0203/03/05/08-0902/11N/12/14/16-21/26-40/43/48-53/55/57/58/60/61/63	B51(5), B5102, B5103, -, B52(5)	37+38+39
B*510104/0204/10/2401/2402, B*520601/0602, B*5902	B51(5), B5102, -	37
B*5104	B51(5), -	7+8+37+38+39
B*5106/41N/54/59	B5, -	38+39
B*5107	B51(5)	37+39+40w
B*511301, B*5214, B*5903	-	11+37
B*511302	-	11+37+38+39
B*5115, B*5901/04	-, B59	31+37
B*5122	-	37+39+42
B*5123	-	25+37+38+39
B*5142	-	7w+8w+25+37w
B*5144N		37+39
B*5146/56	-	7w+8w+37w+38+39
B*5162	-	31
B*520101-0104/03-05/07-13	B52(5), -	37+40
B*5202	-	34+37+40
B*520601/0602, B*510104/0204/10/2401/2402, B*5902	B51(5), B5102, -	37
B*530101-02/05/06/08/10/15/16	B53, -	6+7+8+37
B*5303/11-13,'B*350802/27/56	-, B35	6+7
B*5304	-	6+7+8+11+37
B*5307	-	7+11
B*5314	-	6
B*5401/02/05N/08N/10/13/16/17, B*5507	B54(22), -	27+28+29
B*5403	-	27+28+29+30
B*5404/11	-	28+29
B*5406	-	28w+30
B*5407	-	27+28w+29
B*5409	-	28w
B*5412	-	28+29+31+37
B*5414	-	28w+29
B*5415	-	29
B*550101-0103/0201-03/05/09-12/15/16/19/21/22/24-26/29-31/33/34, B*5610/19N	B55(22), -, B22	27+28
B*550104	B55(22)	27+28+39+41
B*5504/13/17/28, B*0740/53	B55(22), -	28
B*5507,'B*5401/02/05N/08N/10/13/16/17	, -, B54(22)	27+28+29
B*5508, B*5607/15/22	-	28+30
B*5514	-	21+28
B*5518, B*5608/17/23, Cw*0312/19, Cw*1215, Cw*1507	-	27
B*5523	-	21+28w
B*5527	-	24w+28w
B*5532	-	21+28w

Allele	Serology	Mix
B*5601/02/04/16/20/24	B56(22), -	27+28+30
B*5603	B22	21+28w+30
B*560501/0502	-	28+30+39+41
B*5606	-	5+28+30+39+41
B*5607/15,B*5508	-	28+30
B*5608/17/23, B*5518Cw*0312/19, Cw*1215, Cw*1507	-	27
B*5609	-	7+19+21+28w+30
B*5610/19N,'B*550101-0103/0201-03/05/09-12/15/16/19/21/22/24-26/29-31/33/34	-, B22, B55(22)	27+28
B*5611	-	7+19+28w+30
B*5612	-	7w+19w+28
B*5613	-	23+27+28+30
B*5614	-	27+33
B*5618	-	21+28w
B*5622	-	28w+30
B*570101/0103/04/06/08/10/15/18/19	B57(17), -	34+35
B*570102/02-0302/09/16/17	B57(17), -	35
B*5705	-	8+35+37
B*5707	-	35+44
B*5712	-	35+43
B*5713	-	34+35+45
B*5714	-	34+35+36
B*580101/0102/04/05/09-13/17N	B58(17), -	8+36+37
B*5802/07/15/16/18	B58(17), -	36
B*5806	-	36+45
B*5808	-	36+37+45
B*5814	-	35+36
B*5818	-	21+28w
B*5819	-	8+36+37+45
B*5820	-	11+36
B*5901/04, 'B*5115	B59, '-	31+37
B*5902,'B*510104/0204/10/2401/2402, B*520601/0602	-, B51(5), B5102, -	37
B*5903,'B*511301, B*5214	-	11+37
B*670101/0102	B67	12+13+28
B*6702	-	12+33
B*7301	B73	48
B*7801-0202/04	B78, -	5+38+39+41
B*7803	-	5+39+41+42
B*7805	-	5+40+41
B*7806	-	40+41
B*8101-04N	B81, -	1+19+28
B*8201/02	-	23+25+27+28+30w
B*8301	-	7w+19w+23+24w+25+30w
B*9501	-	28w+30+34+43
B*9508	-	12w+21+42
B*9536	-	12w+34+43
B*9542	-	2+34+43

w = weak

Table 6: Amplification patterns of HLA-Cw alleles detected by the HLA-Cw* CTS-PCR-SSP primer mixes (Lot-No C09-2) based on IMGT/HLA Sequence Database Release 2.21.0, April 2008.

Allele	Serology	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23
B*0713/15	-																+							
B*0751	-												+											
B*1596	-					+																		
B*350802	B35, -																					+		
B*3907	-, Cw1	?																						
B*6702	-						+											+						
Cw*010201-03/05-12/15-20	-, Cw1	+																						
Cw*0104	-	+														w	+							
Cw*0113	-	+									+													
Cw*020201-0206/04/06-11/13-15/17/19-21	Cw2, -		+																	+				
Cw*0203/16/18	-		+																					
Cw*0205	-		+																+	+				
Cw*0212	-		w															+		w				
Cw*030201-0203/0401-06/08-10/17/19/23/24/26/28/29/33-37/40/42/44/46-48	Cw10(w3), -			+	+	+																		
Cw*030301-0305/1101-13/18/20N-22Q/30-32/39/43	Cw9(3), -			+	+																			
Cw*0307/14/25	-			+		+																		
Cw*0315	-			+	+													+						
Cw*0316/41	-			+	+																			
Cw*0327/3801/3802	-			+	+	+												+						
Cw*0345	-			+																				
Cw*04010101-0105/0401-05/07-10/12-14/1502/17-20/23-28/30/31	Cw4, -						+																	
Cw*0403/06/16	-						+											w						
Cw*0411/29	-				+		+																	
Cw*041501	-								+													+		
Cw*0432	-			+			+																	
Cw*050101/0103/0104/03-07N/09/11/13-15/17/19	Cw5, -									+	+													
Cw*050102, Cw*0741, Cw*0813	Cw5, -										+													
Cw*0508	-									+	+									+				
Cw*0510	-									+	+												+	
Cw*0512/18, Cw*0810	-									+														
Cw*0516	-									+	+								+					
Cw*06020101-0203/07/09/10/12-14/16-17	Cw6, -										+								+	+				
Cw*0603	-										+							w	+	+				
Cw*0604-06	-										+								+	+				
Cw*0608	-			+							+								+	+				
Cw*0611	-										+								+	+				
Cw*0615	Cw6										?								?	+				
Cw*070101-03/05-07/09/10/13-19/21/23-25/2701-29/32N/35-40/42-54	Cw7, -											+	+											
Cw*070401/02/11/12/45	-											+	w											
Cw*0708	-											+	+											+
Cw*0720/30	-											+												
Cw*0722	-											w	w											
Cw*0726	-											+							+					
Cw*0731	-											+	+	+										
Cw*0733N	-											+												
Cw*0741	-										w													
Cw*080101/0102/03/06/08/09/11	Cw8, -																			+				
Cw*0802/04/07/12/15	Cw8, -										+									+				
Cw*0805	-										+								+					
Cw*0814	-													+	+									
Cw*120201-0203/08/18	-																+	+	+					
Cw*12030101/030102/0303-0306/06/07/11-13/15/20	-																+	+						
Cw*120302/14	-																	+						
Cw*120401	-																	w	+	+				
Cw*120402	-																+	w	+	+				
Cw*1205	-																+		+	+				
Cw*1209	-																		+					
Cw*1210	-						+									+	+	+						
Cw*1216	-															+	+							
Cw*1217	-																+							
Cw*1219	-															+	+							
Cw*1221	-																+							
Cw*140201/0203-03/05-09	-				+																+			
Cw*140202	-																				+			
Cw*140204	-																				w			
Cw*150201-0204/04-13/15/17-21	B35, -																						+	
Cw*1503/16	-																	w				+		
Cw*160101-0103/0401/06-08/10/11	-																						+	
Cw*1602/09	-																			+				+
Cw*1701-04	-			+														w						+
Cw*1801/02	-																							+
Cw*1803	-			+																	+			+

w = weak
 ? = nucleotide sequence information not available for the primer matching sequence
bold: mixes which result in PCR fragments of different sizes (the specificities are first indicated all in one row, then split into different groups in the subsequent rows depending on their fragment size)