

Olerup SSP® HLA-B*13

Product number:	101.515-12u – without <i>Taq</i> polymerase
Lot number:	25K
Expiry date:	2012-August-01
Number of tests:	12
Number of wells per test:	23
Storage - pre-aliquoted primers:	dark at -20°C
- PCR Master Mix:	-20°C
- Adhesive PCR seals	RT
- Product Insert	RT

This Product Description is only valid for Lot No. 25K.

CHANGES COMPARED TO THE PREVIOUS *OLERUP SSP*® HLA-B*13 Lot

The HLA-B*13 specificity and interpretation tables have been updated for the HLA-B alleles described since the previous *Olerup SSP*® HLA-B*13 lot was made (Lot No. 97F).

Seven wells have been added to the HLA-B*13 kit, wells **17 to 23**.
The amplification patterns for some rare HLA-B*13 alleles only differ
by the length of the specific PCR products.

The primers of the wells detailed below have been exchanged, added or modified compared to the previous lot.

Well	5'-primer	3'-primer	rationale
3	-	-	Exchanged positive control primer pair.
5	Modified	Added	Modified 5'-primer for increased yield of specific PCR product, primer added for the B*13:31 allele.
10	Modified	Modified	Modified primers for increased yield of specific PCR product.
12	Modified	-	Modified 5'-primer for increased yield of specific PCR product.
17	New	New	New primer pair for the B*13:27 allele.
18	New	New	New primer pairs for the B*13:28 and B*13:32 alleles.

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19	New	New	New primer pair for the B*13:34 allele.
20	New	New	New primer pair for the B*13:36 allele.
21	New	New	New primer pairs for the B*13:29 and B*13:30 alleles.
22	New	New	New primer pair for the B*13:25 allele.
23	New	New	New primer pair for the B*13:37 allele.

PRODUCT DESCRIPTION

HLA-B*13 SSP subtyping

CONTENT

The primer set contains 5'- and 3'-primers for identifying the B*13:01 to B*13:37 alleles.

PLATE LAYOUT

Each test consists of 23 PCR reactions in a 24 well PCR plate. Well 24 is empty.

1	2	3	4	5	6	7	8
9	10	11	12	13	14	15	16
17	18	19	20	21	22	23	empty

The 24 well cut PCR plate is marked with 'HLA-B*13' in silver/gray ink.

Well No. 1 is marked with the Lot No. '25K'.

A faint row of numbers is seen between wells 1 and 2 or wells 7 and 8 of the PCR trays. These stem from the manufacture of the trays, and should be disregarded.

The PCR plates are heat-sealed with a PCR-compatible foil.

Please note: When removing each 24 well PCR plate, make sure that the remaining plates stay sealed. Use a scalpel or a similar instrument to carefully cut the foil between the plates.

INTERPRETATION

The interpretation of HLA-B*13 subtypings will be influenced by several B*07, most B*08, the B*14:13, most B*15, most B*18, several B*27, most B*35, two B*37, the B*38, most B*39, the B*40, the B*41, the B*42, several B*44, the B*45, the B*46, two B*47, the B*48, five B*49, the B*50, several B*51, two B*52, most B*53, most B*54, most B*55, most B*56, most B*57, most B*58, the B*59, the B*67, the B*73:01, the B*78:07, the B*81 and the B*82 alleles when present on the other haplotype. In addition, the C*02:06 allele will be amplified by primer mix 12, the C*03:34 allele will be amplified by primer mix 3 and the C*15:24 allele will be amplified by primer mixes 7 and 10.

UNIQUELY IDENTIFIED ALLELES

All the HLA-B*13 alleles, i.e. **B*13:01 to B*13:37**, recognized by the HLA Nomenclature Committee in April 2010¹ will give rise to unique amplification patterns by the primers in the HLA-B*13 subtyping kit.

The HLA-B*13 kit cannot distinguish the B*13:01:01-13:01:03 alleles or the 13:02:01-13:02:07 alleles.

The B*13:15 and B*13:33 alleles can be distinguished by the different sizes of the specific PCR products generated by primer mix 16.

¹HLA-B alleles listed on the IMGT/HLA web page 2010-April-01, release 3.0.0, www.ebi.ac.uk/imgt/hla.

RESOLUTION IN HOMO- AND HETEROZYGOTES

The 35 HLA-B*13 alleles will generate 36 different amplification patterns, that can be combined in 630 homozygous and heterozygous combinations. 272 of these genotypes do not give rise to unique amplification patterns. The different lengths of the specific PCR products generated by primer mixes 5, 8, 10, 13, 15, 16, 18 and 21 were not considered in these calculations.

+++++---+ - - - - - + - - - - -	13:18,13:22:01 = 13:22:01,13:31 = 13:23,13:31
+++++---+ - - - - - - - - - -	13:01,13:31 = 13:07N,13:16 = 13:07N,13:31 =
	13:18,13:20 = 13:18,13:22:02 = 13:20,13:31 =
	13:22:02,13:31
+++++---+ + - - - - - - - - - -	13:08Q,13:20 = 13:08Q,13:22:02
+++++---+ - + - - - - - - - - - -	13:09,13:20 = 13:09,13:22:02
+++++---+ - - - - - + - - - - -	13:19,13:20 = 13:19,13:22:02
+++++---+ - - - - - + - - - - -	13:13,13:16 = 13:20,13:35 = 13:22:02,13:35
+++++---+ - - - - - + - - - - -	13:02,13:22:01 = 13:14,13:20 = 13:14,13:22:01
	= 13:14,13:22:02 = 13:16,13:22:01 =
	13:16,13:23
+++++---+ - - - - - + - - - - -	13:15,13:20 = 13:15,13:22:02 = 13:16,13:17
+++++---+ - - - - - - - - - - + - - - - -	13:20,13:27 = 13:22:02,13:27
+++++---+ - - - - - - - - - - - + - - - - -	13:16,13:28 = 13:20,13:32 = 13:22:02,13:32
+++++---+ - - - - - - - - - - - - + - - - - -	13:20,13:34 = 13:22:02,13:34
+++++---+ - - - - - - - - - - - - - + - - - - -	13:16,13:29 = 13:20,13:30 = 13:22:02,13:30
+++++---+ - - - - - - - - - - - - - - + - - - - -	13:20,13:37 = 13:22:02,13:37
+++++---+ -	13:01,13:16 = 13:02,13:20 = 13:02,13:22:02 =
	13:16,13:20 = 13:16,13:22:02
+++++---+ - - - - - + - - - - -	13:07N,13:35 = 13:13,13:18
+++++---+ - - - - - + - - - - -	13:07N,13:14 = 13:18,13:23
+++++---+ - - - - - + - - - - -	13:07N,13:15 = 13:17,13:18
+++++---+ - - - - - - - - - - - + - - - - -	13:07N,13:32 = 13:18,13:28
+++++---+ - - - - - - - - - - - - + - - - - -	13:07N,13:30 = 13:18,13:29
+++++---+ -	13:01,13:18 = 13:02,13:07N = 13:07N,13:18
+++++---+ - + - - - + - - - - -	13:02,13:21 = 13:09,13:13 = 13:09,13:21 =
	13:21,13:35
+++++---+ - - - - - + - - - - -	13:13,13:14 = 13:23,13:35
+++++---+ - - - - - + - + - - - - -	13:13,13:15 = 13:17,13:35
+++++---+ - - - - - + - - - - - - + - - - - -	13:13,13:32 = 13:28,13:35
+++++---+ - - - - - + - - - - - - - + - - - - -	13:13,13:30 = 13:29,13:35
+++++---+ - - - - - + - - - - - - - - - - - - - - - -	13:01,13:35 = 13:02,13:13 = 13:13,13:35
+++++---+ - - - - - + + - - - - - - - - - - - - - - -	13:14,13:17 = 13:15,13:23
+++++---+ - - - - - + - - + - - - - - - - - - - - -	13:14,13:28 = 13:23,13:32
+++++---+ - - - - - + - - - - - + - - - - - - - - - -	13:14,13:29 = 13:23,13:30
+++++---+ - - - - - + - - - - - - - - - - - - - - - -	13:01,13:14 = 13:02,13:23 = 13:14,13:23
+++++---+ - - - - - + - - - - - - + - - - - - - - - - -	13:15,13:28 = 13:17,13:32
+++++---+ - - - - - + - - - - - - - + - - - - - - - - - -	13:15,13:29 = 13:17,13:30
+++++---+ - - - - - + - - - - - - - - - - - - - - - -	13:01,13:15 = 13:02,13:17 = 13:15,13:17
+++++---+ - - - - - - - - - - - + - - + - - - - - - -	13:28,13:30 = 13:29,13:32
+++++---+ - - - - - - - - - - - + - - - - - - - - - -	13:01,13:32 = 13:02,13:28 = 13:28,13:32
+++++---+ - - - - - - - - - - - - + - - - - - - - - - -	13:01,13:30 = 13:02,13:29 = 13:29,13:30
+++++---+ + -	13:03,13:07N = 13:07N,13:20 = 13:07N,13:22:02
+++++---+ - + - - - + - - - - - - - - - - - - - - - -	13:03,13:21 = 13:20,13:21 = 13:21,13:22:02
+++++---+ - - + - - + - - - - - - - - - - - - - - - -	13:10,13:22:01 = 13:22:01,13:26
+++++---+ - - + - - - - - - - - - - - - - - - - - - -	13:03,13:26 = 13:10,13:20 = 13:10,13:22:02 =
	13:20,13:26 = 13:22:02,13:26
+++--+- - - - + - - - - - - - - - - - - - - - -	13:11,13:20 = 13:11,13:22:02
+++--+- - - - - + - - - - - - - - - - - - - - - -	13:03,13:13 = 13:13,13:20 = 13:13,13:22:02

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+++---+-----+-----	13:01,13:22:01 = 13:03,13:23 = 13:20,13:22:01 = 13:20,13:23 = 13:22:01,13:23 = 13:22:02,13:23
+++---+-----+-----	13:03,13:17 = 13:17,13:20 = 13:17,13:22:02
+++---+-----+-----	13:03,13:28 = 13:20,13:28 = 13:22:02,13:28
+++---+-----+-----	13:03,13:36 = 13:20,13:36 = 13:22:02,13:36
+++---+-----+-----	13:03,13:29 = 13:20,13:29 = 13:22:02,13:29
+++---+-----+-----	13:03,13:25 = 13:20,13:25 = 13:22:02,13:25
+++---+-----+-----	13:01,13:03 = 13:01,13:20 = 13:01,13:22:02 = 13:03,13:20 = 13:20,13:20 = 13:20,13:22:02
+++---+-----+-----	13:01,13:04 = 13:04,13:26
+++---+-----+-----	13:07N,13:10 = 13:07N,13:26
+++---+-----+-----	13:01,13:07N = 13:07N,13:07N
+++---+-----+-----	13:10,13:21 = 13:21,13:26
+++---+-----+-----	13:01,13:21 = 13:13,13:21 = 13:21,13:21
+++---+-----+-----	13:10,13:13 = 13:13,13:26
+++---+-----+-----	13:10,13:23 = 13:23,13:26
+++---+-----+-----	13:10,13:17 = 13:17,13:26
+++---+-----+-----	13:10,13:28 = 13:26,13:28
+++---+-----+-----	13:10,13:36 = 13:26,13:36
+++---+-----+-----	13:10,13:29 = 13:26,13:29
+++---+-----+-----	13:10,13:25 = 13:25,13:26
+++---+-----+-----	13:01,13:10 = 13:01,13:26 = 13:10,13:26 = 13:26,13:26
+++---+-----+-----	13:01,13:13 = 13:13,13:13
+++---+-----+-----	13:01,13:23 = 13:23,13:23
+++---+-----+-----	13:01,13:17 = 13:17,13:17
+++---+-----+-----	13:01,13:28 = 13:28,13:28
+++---+-----+-----	13:01,13:36 = 13:36,13:36
+++---+-----+-----	13:01,13:29 = 13:29,13:29
+++---+-----+-----	13:01,13:25 = 13:25,13:25
+++---+-----+-----	13:12,13:31 = 13:19,13:31
+++---+-----+-----	13:02,13:31 = 13:03,13:18 = 13:03,13:31 = 13:16,13:18 = 13:16,13:31 = 13:18,13:31 = 13:31,13:31
+++---+-----+-----	13:03,13:08Q = 13:08Q,13:16
+++---+-----+-----	13:03,13:09 = 13:09,13:16
+++---+-----+-----	13:03,13:19 = 13:12,13:16 = 13:16,13:19
+++---+-----+-----	13:03,13:35 = 13:16,13:35
+++---+-----+-----	13:03,13:14 = 13:14,13:16
+++---+-----+-----	13:03,13:15 = 13:15,13:16
+++---+-----+-----	13:03,13:27 = 13:16,13:27
+++---+-----+-----	13:03,13:32 = 13:16,13:32
+++---+-----+-----	13:03,13:34 = 13:16,13:34
+++---+-----+-----	13:03,13:30 = 13:16,13:30
+++---+-----+-----	13:03,13:37 = 13:16,13:37
+++---+-----+-----	13:02,13:03 = 13:02,13:16 = 13:03,13:16 = 13:16,13:16
+++---+-----+-----	13:12,13:18 = 13:18,13:19
+++---+-----+-----	13:02,13:18 = 13:18,13:18
+++---+-----+-----	13:08Q,13:12 = 13:08Q,13:19
+++---+-----+-----	13:02,13:08Q = 13:08Q,13:08Q
+++---+-----+-----	13:09,13:12 = 13:09,13:19
+++---+-----+-----	13:12,13:35 = 13:19,13:35
+++---+-----+-----	13:12,13:14 = 13:14,13:19
+++---+-----+-----	13:12,13:15 = 13:15,13:19
+++---+-----+-----	13:12,13:27 = 13:19,13:27
+++---+-----+-----	13:12,13:32 = 13:19,13:32
+++---+-----+-----	13:12,13:34 = 13:19,13:34

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++-+-----	-----+---	-----+--	13:12,13:30 = 13:19,13:30
++-+-----	-----+---	-----+--	13:12,13:37 = 13:19,13:37
++-+-----	-----+---	-----+--	13:02,13:12 = 13:02,13:19 = 13:12,13:19 =
			13:19,13:19
+++-----	-----+--	-----+--	13:02,13:35 = 13:35,13:35
+++-----	-----+--	-----+--	13:02,13:14 = 13:14,13:14
+++-----	-----+--	-----+--	13:02,13:15 = 13:15,13:15
+++-----	-----+--	-----+--	13:02,13:27 = 13:27,13:27
+++-----	-----+--	-----+--	13:02,13:32 = 13:32,13:32
+++-----	-----+--	-----+--	13:02,13:34 = 13:34,13:34
+++-----	-----+--	-----+--	13:02,13:30 = 13:30,13:30
+++-----	-----+--	-----+--	13:02,13:37 = 13:37,13:37
+++-----	-----+--	-----+--	13:03,13:22:01 = 13:22:01,13:22:01 =
			13:22:01,13:22:02
+++-----	-----+--	-----+--	13:03,13:22:02 = 13:22:02,13:22:02

13:01 = 13:01:01-13:01:03

13:02 = 13:02:01-13:02:07

13:15 = 13:15 and 13:33

SPECIFICITY TABLE

HLA-B*13 SSP subtyping

Specificities and sizes of the PCR products of the 23 primer mixes used for HLA-B*13 SSP subtyping

Primer Mix	Size of spec. PCR product ¹	Size of control band ²	Amplified HLA-B*13 alleles ³	Other amplified HLA Class I alleles ⁴
1	140 bp	800 bp	*13:01:01-13:04, 13:06-13:08Q, 13:10-13:23, 13:25-13:37	*44:60
2^{5,6}	95 bp	800 bp	*13:01:01-13:02:07, 13:07N-13:11, 13:13-13:21, 13:23, 13:25-13:37	*40:91, 51:93
3	245 bp	800 bp	*13:01:01-13:01:03, 13:07N, 13:13, 13:17, 13:20-13:23, 13:25-13:26, 13:28-13:29, 13:36	*07:12, 07:14, 07:18:01-07:18:02, 27:19, 27:30, 35:15, 35:33, 35:88, 40:04, 40:30, 40:34, 40:59, 40:64, 40:68, 40:99, 44:16, 44:21, 44:64:01-44:64:02, 48:17, 57:13, 57:25, C*03:34
4	245 bp	1070 bp	*13:02:01-13:02:07, 13:08Q-13:09, 13:14-13:16, 13:18-13:19, 13:27, 13:30-13:35, 13:37	*07:78, 07:84, 27:14, 39:50, 40:06:01:01-40:06:02, 40:44, 40:53, 40:70, 40:75, 40:83, 40:86, 40:93, 40:95-40:96, 45:10, 49:07, 51:10, 51:16, 51:31, 51:34, 51:82, 51:93, 52:08, 55:09, 55:22, 55:24, 73:01
5^{5,7,10}	125 bp, 150 bp, 180 bp	800 bp	*13:03, 13:16, 13:20, 13:22:01-13:22:02, 13:31	*07:78, 15:42, 15:73, 15:86, 35:60, 40:71, 44:10, 46:11, 46:18, 51:15, 51:62, 54:01-54:03, 54:05N, 54:07-54:08N, 54:10, 54:12-54:13, 54:17-54:22, 55:01:01-55:03, 55:05, 55:07, 55:09-55:12, 55:15-55:16, 55:18-55:19, 55:21-55:22, 55:24-55:26, 55:29-55:31, 55:33-55:40, 56:01:01-56:02, 56:04, 56:07-56:08, 56:10, 56:13-56:14, 56:16-56:17, 56:19N-56:20, 56:23-56:29, 59:01, 59:04-59:05, 82:01-82:03

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6^{5,9}	105 bp	1070 bp	*13:04	*15:04, 15:16, 15:67, 15:95, 15:137, 15:155, 35:37, 39:06:01-39:06:02, 39:33-39:34, 39:50, 39:57, 40:86, 45:02, 45:09, 49:04-49:05, 50:09, 51:13:01-51:13:02, 51:37, 51:63, 51:92, 52:14, 54:14, 55:13, 55:23, 55:32, 56:22, 59:03, 73:01
7⁸	190 bp	1070 bp	*13:06	*07:34, 08:01:01-08:06, 08:08N-08:10, 08:12:01-08:13, 08:15-08:20, 08:22-08:24, 08:26-08:27, 08:29-08:36, 08:38-08:39, 08:41-08:48, 08:50-08:61, 14:13, 15:42, 15:83, 15:86, 15:188, 18:26, 35:60, 35:87, 37:12, 38:01:01, 38:01:03-38:07, 38:09-38:21, 39:01:01:01-39:01:01:02L, 39:01:03-39:06:02, 39:08-39:14, 39:16, 39:18-39:20, 39:22-39:24, 39:26-39:28, 39:30-39:31, 39:34-39:37, 39:39, 39:41-39:42, 39:44-39:46, 39:48, 39:51-39:54, 39:56-39:57, 40:39, 40:51, 41:01-41:03:02, 41:05-41:09, 41:11-41:12, 42:01:01-42:02, 42:04-42:08, 42:10-42:13, 46:18, 51:21, 51:36, 54:01-54:02, 54:04-54:05N, 54:07-54:08N, 54:10-54:13, 54:17-54:22, 55:01:01-55:05, 55:07, 55:10-55:13, 55:15-55:21, 55:25-55:26, 55:28-55:40, 56:10, 56:12, 56:23, 58:20, 59:01-59:03, 59:05, 67:01:01-67:02, 78:07, C*15:24
8^{8,11}	185 bp, 225 bp	1070 bp	*13:07N, 13:18, 13:31	*07:78, 07:84, 15:04, 15:137, 39:33, 51:05, 51:29, 51:54, 51:61, 51:82, 54:10, 54:20, 55:09, 55:21, 55:37
9	230 bp	1070 bp	*13:08Q	
10^{5,12}	105 bp , 210 bp	800 bp	*13:09, 13:21	*07:05:01-07:06, 07:32, 07:34, 07:40, 07:53, 07:69, 07:78, 07:80, 07:90, 07:97, 08:01:02, 08:06, 08:20, 15:53, 15:150, 15:183, 15:188, 27:07-27:08, 27:11-27:12, 27:18, 27:20, 27:24, 27:26-27:27, 27:33, 27:40, 27:42-27:44, 35:09:02, 35:18, 35:31, 35:63,

				35:84, 35:88, 35:90, 40:01:01-40:16, 40:18-40:40, 40:42-40:99, 41:01-41:04, 41:06-41:11, 44:09, 44:31, 44:46, 44:75, 44:90, 45:01-45:11, 46:10, 47:02, 47:03w, 48:01:01-48:01:02, 48:03:01-48:07, 48:09-48:11, 48:13, 48:15-48:16, 48:18-48:22, 50:01:01-50:02, 50:04-50:09, 51:01:09, 51:10, 51:24:02-51:24:03, 51:81, 54:11, 55:04, 55:08, 58:27, 73:01, 81:01-81:04N, C*15:24
11	280 bp	800 bp	*13:04, 13:10, 13:26	*07:09, 07:11, 07:17, 08:28, 08:35, 08:37, 15:01:01:01-15:01:04, 15:01:06-15:08, 15:11:01-15:16, 15:18:01-15:21, 15:23-15:29, 15:31-15:36, 15:38:01-15:40, 15:43-15:44, 15:46-15:47, 15:49-15:57, 15:60-15:62, 15:64-15:72, 15:74-15:76, 15:78:01-15:82, 15:84-15:85, 15:87-15:89, 15:91-15:98, 15:101-15:129, 15:131-15:132, 15:134-15:136, 15:138-15:149N, 15:151-15:161, 15:163-15:167, 15:169-15:176, 15:178-15:187, 15:189-15:191, 18:01:01-18:15, 18:17N-18:25, 18:27-18:40, 18:42-18:49, 27:41, 35:01:01-35:01:19, 35:05:01-35:05:02, 35:07-35:08:04, 35:10-35:11, 35:14:01-35:17, 35:19-35:21, 35:23-35:30, 35:32, 35:35, 35:37, 35:40N-35:43, 35:45-35:54, 35:57-35:58, 35:61-35:64, 35:66-35:69, 35:71-35:72, 35:76-35:80, 35:82, 35:86, 35:89-35:94, 35:97, 35:99, 39:07, 39:43, 40:03, 40:20, 40:38, 40:52, 40:59-40:60, 42:09, 44:17, 44:43, 45:09, 46:01:01-46:10, 46:12-46:17, 46:19-46:22, 48:02:01-48:02:02, 48:14, 49:04-49:05, 51:37, 51:45, 51:63, 53:01:01-53:03, 53:05-53:06, 53:08:01-53:18, 53:20-53:23, 54:06, 54:09, 54:14, 55:14, 55:23, 55:32, 56:03, 56:09, 56:18, 57:01:01-57:01:07, 57:06, 57:08, 57:10-57:11, 57:13-57:16,

				57:18-57:27, 57:29-57:31, 58:01:01-58:02, 58:04-58:07, 58:09-58:19, 58:21-58:26
12^{5,8}	90 bp	1070 bp	*13:11	*15:18:03, 15:73, 39:17, 40:48, 40:71, 51:62, 56:02, 56:04, 56:10, C*02:06
13^{5,13}	90 bp, 180 bp	800 bp	*13:12, 13:19	
14⁵	130 bp	1070 bp	*13:13, 13:21, 13:35	*40:91, 51:93
15^{5,14}	105 bp, 250 bp	800 bp	*13:14, 13:22:01, 13:23	*07:78, 15:73, 18:09, 27:01, 37:10, 38:02:01-38:04, 38:08, 38:15, 38:18, 40:47-40:48, 40:96, 44:15, 44:55, 49:02, 54:22, 56:01:03
16^{5,15}	90 bp, 205 bp	1070 bp	*13:15, 13:17, 13:33	*35:46, 49:09, 51:62
17	230 bp	1070 bp	*13:27	
18¹⁶	155 bp, 205 bp	1070 bp	*13:28, 13:32	
19	195 bp	1070 bp	*13:34	
20^{5,7}	105 bp	1070 bp	*13:36	*18:48, 35:46-35:47, 38:12, 40:08, 40:25, 40:68, 44:12, 51:74
21^{5,17}	120 bp, 190 bp	1070 bp	*13:29-13:30	
22⁹	135 bp	1070 bp	*13:25	
23	200 bp	1070 bp	*13:37	

¹Alleles are assigned by the presence of specific PCR product(s). However, the sizes of the specific PCR products may be helpful in the interpretation of HLA-B*13 SSP typings.

When the primers in a primer mix can give rise to specific PCR products of more than one length this is indicated if the size difference is 20 base pairs or more. Size differences shorter than 20 base pairs are not given. For high resolution SSP kits the respective lengths of the specific PCR product(s) of the alleles amplified by these primer mixes are given.

Nonspecific amplifications, i.e. a ladder or a smear of bands, may sometimes be seen. GC-rich primers have a higher tendency of giving rise to nonspecific amplifications than other primers.

PCR fragments longer than the control bands may sometimes be observed. Such bands should be disregarded and do not influence the interpretation of the SSP typings.

PCR fragments migrating faster than the control bands, but slower than a 400 bp fragment may be seen in some gel read-outs. Such bands can be disregarded and do not influence the interpretation of the SSP typings.

Some primers may give rise to primer oligomer artifacts. Sometimes this phenomenon is an inherent feature of the primer pair(s) of a primer mix. More often it is due to other factors such as too low amount of DNA in the PCR reactions, taking too long time in setting up the PCR reactions, working at elevated room temperature or using thermal cyclers that are not pre-heated.

²The internal positive control primer pairs amplify segments of the human growth hormone gene. The two different control primer pairs give rise to either an internal positive control band of 1070 base pairs, for most wells, or a band of 800 base pairs, for some wells.

Well number 1 contains the primer pair giving rise to the shorter, 800 bp, internal positive control band in order to help in the correct orientation of the HLA-B*13 subtyping.

In addition, wells number 2, 3, 5, 10, 11, 13 and 15 contain the primer pair giving rise to the

shorter, 800 bp, internal positive control band in order to allow kit identification.

In the presence of a specific amplification the intensity of the control band often decreases.

³The B*13:15 and B*13:33 alleles can be distinguished by the different sizes of the specific PCR products generated by primer mix 16.

⁴Due to the sharing of sequence motifs between HLA-B alleles some non-HLA-B*13 alleles will be amplified by primer mixes 1 to 8, 10 to 12, 14 to 16 and 20. In addition, the C*02:06 allele will be amplified by primer mix 12, the C*03:34 allele will be amplified by primer mix 3 and the C*15:24 allele will be amplified by primer mixes 7 and 10.

⁵Short specific PCR fragments are less intense and not as sharp as longer specific bands.

⁶Primer mix 2 may give rise to a weak band of about 400 bp. This band should be disregarded when interpreting HLA-B*13 SSP subtypings.

⁷Primer mixes 5 and 20 may give a lower yield of specific PCR product than the other HLA-B*13 primer mixes.

⁸Primer mixes 7, 8 and 12 have a weak tendency of nonspecific amplifications.

⁹Primer mixes 6 and 22 may have a tendency of giving rise to primer oligomer formation.

¹⁰Primer mix 5: Specific PCR fragment of 125 bp in the B*13:22:01-13:22:02 and in the B*15:86, 35:60, 46:18, 54:01, 54:02, 54:05N, 54:07-54:08N, 54:10, 54:12-54:13, 54:19, 54:21, 54:22, 55:02, 55:07, 55:10, 55:12, 55:16, 55:18-55:19, 55:22, 55:26, 55:30, 55:34, 55:35, 55:37, 55:39, 56:10, 56:23, 59:01 and 59:05 alleles. Specific PCR fragment of 150 bp in the B*13:16, 13:20 and 13:31 alleles. Specific PCR fragment of 125 and 150 bp in the B*07:78, 15:42, 54:20, 55:01:01-55:01:06, 55:03, 55:05, 55:09, 55:11, 55:15, 55:21, 55:24-55:25, 55:29, 55:31, 55:33, 55:36, 55:38 and 55:40 alleles. Specific PCR fragments of 125 and 180 bp in the B*13:03 and in the B*40:71, 44:10, 46:11, 51:15, 54:03, 56:01:01-56:02, 56:04, 56:07-56:08, 56:13-56:14, 56:16-56:17, 56:19N-56:20, 56:24, 56:26-56:29, 59:04 and 82:01-82:03 alleles. Specific PCR fragments of 125, 150 and 180 bp in the B*15:73, 51:62 and 56:25 alleles. All bands may not always be visible.

¹¹Primer mix 8: Specific PCR fragment of 185 bp in the B*13:07N allele. Specific PCR fragment of 225 bp in the B*13:18 and 13:31 and in the B*07:78, 07:84, 15:04, 15:137, 39:33, 51:05, 51:29, 51:54, 51:61, 51:82, 54:1, 54:20, 55:09, 55:21 and 55:37 alleles.

¹²Primer mix 10: Specific PCR fragment of 105 bp in the B*13:21 and the B*07:05:01-07:06, 07:32, 07:34, 07:40, 07:53, 07:69, 07:78, 07:80, 07:90, 07:97, 08:01:02, 08:06, 08:20, 15:150, 15:188, 27:07, 27:11, 27:20, 27:24, 27:27, 27:43, 35:09:02, 35:18, 35:31, 35:88, 40:10, 40:12, 40:13, 40:19, 40:21, 40:47, 40:58, 40:76, 40:96, 44:31, 48:01:01-48:01:02, 48:03:01-48:07, 48:09-48:11, 48:13, 48:15-48:16, 48:18-48:22, 51:01:09, 51:10, 51:24:02-51:24:03, 51:81, 54:11, 55:04, 55:08, 58:27, 81:01-81:04N and in the C*15:24 alleles. Specific PCR fragment of 210 bp in the B*13:09 and the B*15:53, 15:183, 27:08, 27:12, 27:18, 27:26, 27:40, 27:42, 27:44, 35:63, 35:84, 35:90, 40:03, 40:09, 40:14:01, 40:14:02, 40:18, 40:20, 40:24, 40:26, 40:28, 40:31-40:33, 40:42, 40:44, 40:59, 40:60, 40:71, 40:72:01, 40:77, 40:87, 40:95, 40:98, 41:01-41:04, 41:06, 41:07, 41:09-41:11, 44:09, 44:46, 44:75, 44:90, 45:01-45:11, 46:10, 47:02, 47:03w, 50:01:01-50:02 and 50:04-50:09 alleles. Specific PCR fragments of 105 and 210 bp in the B*27:33, 40:01:01-40:02:10, 40:04-40:08, 40:11, 40:14:03-40:16, 40:22N, 40:23, 40:27, 40:29-40:30, 40:34-40:40, 40:43, 40:45-40:46, 40:48-40:51, 40:53-40:57, 40:61-40:70, 40:72:02-40:75, 40:78-40:86, 40:88-40:94, 40:97, 40:99, 41:08 and 73:01 alleles.

¹³Primer mix 13: Specific PCR fragment of 90 bp in the B*13:19 allele. Specific PCR fragment of 180 bp in the B*13:12 allele.

¹⁴Primer mix 15: Specific PCR fragment of 105 bp in the B*13:22:01 and in the B*07:78, 15:73, 40:48, 54:22 and 56:01:03 alleles. Specific PCR fragment of 250 bp in the B*13:14 and 13:23 and in the B*18:09, 27:01, 37:10, 38:02:01-38:04, 38:08, 38:15, 38:18, 40:47, 40:96, 44:15, 44:55 and 49:02 alleles.

¹⁵Primer mix 16: Specific PCR fragment of 90 bp in the B*13:17 and 13:33 and the B*35:46 alleles. Specific PCR fragment of 205 bp in the B*13:15 and in the B*49:09 and 51:62 alleles.

¹⁶Primer mix 18: Specific PCR fragment of 155 bp in the B*13:28 allele. Specific PCR fragment of 205 bp in the B*13:32 allele.

¹⁷Primer mix 21: Specific PCR fragment of 120 bp in the B*13:29 allele. Specific PCR fragment of 190 bp in the B*13:30 allele.

‘w’, may be weakly amplified.

INTERPRETATION TABLE

HLA-B*13 SSP subtyping

Amplification patterns of the B*13:01 to *13:37 alleles

	Well ⁶											
	1	2	3	4	5	6	7	8	9	10	11	12
Length of spec.	140	95	245	245	125	105	190	185	230	105	280	90
PCR product(s)					150			225		210		
					180							
Length of int.	800	800	800	1070	800	1070	1070	1070	1070	800	800	1070
pos. control ¹												
5'-primer(s) ²	209	506	355	357	420	357	412	106	357	142	419	369
	5'-ggC 3'	5'-gCT 3'	5'-TCA 3'	5'-Tgg 3'	5'-TTA 3'	5'-Tgg 3'	5'-ATA 3'	5'-CCA 3'	5'-Tgg 3'	5'-TCA 3'	5'-gTC 3'	5'-TAC 3'
								357		412		
								5'-Tgg 3'		5'-ATA 3'		
3'-primer(s) ³	309	559	559	559	506	419	559	251	548	311	3 rd	420
	5'-gTg 3'	5'-CTC 3'	5'-CTC 3'	5'-CTC 3'	5'-TgC 3'	5'-Cgg 3'	5'-CgT 3'	5'-CCC 3'	5'-ggC 3'	5'-ggT 3'	5'-TAT 3'	5'-gCT 3'
					527	419		539		477		
					5'-CCT 3'	5'-CgA 3'		5'-TCC 3'		5'-gCg 3'		
					538							
					5'-CCA 3'							
					559							
					5'-Cag 3'							
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
HLA-B allele ⁴												
*13:01:01-13:01:03	1	2	3									
*13:02:01-13:02:07	1	2		4								
*13:03	1				5							
*13:04	1					6					11	
*13:06	1						7					
*13:07N	1	2	3					8				
*13:08Q	1	2		4					9			
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

INTERPRETATION TABLE

HLA-B*13 SSP subtyping

Amplification patterns of the B*13:01 to *13:37 alleles

Well ⁶												
13	14	15	16	17	18	19	20	21	22	23		
90	130	105	90	230	155	195	105	120	135	200		Length of spec.
180		250	205		205			190				PCR product(s)
800	1070	800	1070	1070	1070	1070	1070	1070	1070	1070		Length of int.
												pos. control ¹
268	419	97	144	368	209	406	193	160	209	357		5'-primer(s) ²
5'-Agg 3'	5'-gTA 3'	5'-TCC 3'	5'-gCA 3'	5'-gTT 3'	5'-ggC 3'	5'-gCA 3'	5'-CCA 3'	5'-ACT 3'	5'-ggC 3'	5'-Tgg 3'		
420		103	420		357			229				
5'-TTA 3'		5'-CCT 3'	5'-TTA 3'		5'-Tgg 3'			5'-TAA 3'				
		420										
		5'-TTA 3'										
317	506	309	193	559	323	559	259	309	302	515		3'-primer(s) ³
5'-ggA 3'	5'-TgA 3'	5'-gTg 3'	5'-CgT 3'	5'-CTC 3'	5'-AgC 3'	5'-CTC 3'	5'-gTT 3'	5'-gTg 3'	5'-ggC 3'	5'-CCC 3'		
559		486	583		523							
5'-CCC 3'		5'-gCC 3'	5'-gTg 3'		5'-ACC 3'							
13	14	15	16	17	18	19	20	21	22	23		Well No.
												HLA-B allele ⁴
												*13:01:01-13:01:03
												*13:02:01-13:02:07
												*13:03
												*13:04
												*13:06
												*13:07N
												*13:08Q
13	14	15	16	17	18	19	20	21	22	23		Well No.

Well No.	1	2	3	4	5	6	7	8	9	10	11	12
Length of spec.	140	95	245	245	125	105	190	185	230	105	280	90
PCR product(s)					150			225		210		
					180							
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
*13:09		2		4						10		
*13:10	1	2									11	
*13:11	1	2										12
*13:12	1											
*13:13	1	2	3									
*13:14	1	2		4								
*13:15, 13:33 ⁵	1	2		4								
*13:16	1	2		4	5							
*13:17	1	2	3									
*13:18	1	2		4				8				
*13:19	1	2		4								
*13:20	1	2	3		5							
*13:21	1	2	3							10		
*13:22:01	1		3		5							
*13:22:02	1		3		5							
*13:23	1	2	3									
*13:25	1	2	3									
*13:26	1	2	3								11	
*13:27	1	2		4								
*13:28	1	2	3									
*13:29	1	2	3									
*13:30	1	2		4								
*13:31	1	2		4	5			8				
*13:32	1	2		4								
*13:34	1	2		4								
*13:35	1	2		4								
*13:36	1	2	3									
*13:37	1	2		4								
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

Lot No.: **25K**

Lot-specific information

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13	14	15	16	17	18	19	20	21	22	23	Well No.
90	130	105	90	230	155	195	105	120	135	200	Length of spec.
180		250	205		205			190			PCR product(s)
13	14	15	16	17	18	19	20	21	22	23	Well No.
											*13:09
											*13:10
											*13:11
13											*13:12
	14										*13:13
		15									*13:14
			16								*13:15, 13:33 ⁵
											*13:16
			16								*13:17
											*13:18
13											*13:19
											*13:20
	14										*13:21
		15									*13:22:01
											*13:22:02
		15									*13:23
								22			*13:25
											*13:26
				17							*13:27
					18						*13:28
								21			*13:29
								21			*13:30
											*13:31
					18						*13:32
						19					*13:34
	14										*13:35
							20				*13:36
										23	*13:37
13	14	15	16	17	18	19	20	21	22	23	Well No.

Length of spec.	140	95	245	245	125	105	190	185	230	105	280	90
PCR product(s)					150			225		210		
					180							
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
*07:05:01-07:06, 07:32, 07:40, 07:53, 07:69, 07:80, 07:90, 07:97, 15:150, 27:07-27:08, 27:11-27:12, 27:18, 27:20, 27:24, 27:26-27:27, 27:33, 27:40, 27:42-27:44, 35:09:02, 35:18, 35:31, 35:84, 40:01:01-40:02:10, 40:05, 40:07, 40:09-40:16, 40:18-40:19, 40:21-40:24, 40:26-40:29, 40:31-40:33, 40:35-40:37, 40:40, 40:42-40:43, 40:45-40:46, 40:49-40:50, 40:54-40:58, 40:61-40:63, 40:65-40:67, 40:69, 40:72:01-40:74, 40:76-40:82, 40:84-40:85, 40:87-40:90, 40:92, 40:94, 40:97-40:98, 41:04, 41:10, 44:09, 44:31, 44:46, 44:75, 44:90, 45:01, 45:03-45:08, 45:11, 47:02, 48:01:01-48:01:02, 48:03:01-48:07, 48:09-48:11, 48:13, 48:15-48:16, 48:18-48:22, 50:01:01-50:02, 50:04-50:08, 51:01:09, 51:24:02-51:24:03, 51:81, 55:08, 58:27, 81:01-81:04N										10		
*07:09, 07:11, 07:17, 08:28, 08:37, 15:01:01-01-15:01:04, 15:01:06-15:03:03, 15:05:01-15:08, 15:11:01-15:15, 15:18:01-15:18:02, 15:18:04-15:21, 15:23-15:29, 15:31-15:36, 15:38:01-15:40, 15:43-15:44, 15:46-15:47, 15:49-15:52, 15:54-15:57, 15:60-15:62, 15:64-15:66, 15:68-15:72, 15:74-15:76, 15:78:01-15:82, 15:84-15:85, 15:87-15:89, 15:91-15:94N, 15:96-15:98, 15:101-15:129, 15:131-15:132, 15:134-15:136, 15:138-15:149N, 15:151-15:154, 15:156-15:161, 15:163-15:167, 15:169-15:176, 15:178-15:182N, 15:184-15:187, 15:189-15:191, 18:01:01-18:08, 18:10-18:15, 18:17N-18:25, 18:27-18:40, 18:42-18:47, 18:49, 27:41, 35:01:01-35:01:19, 35:05:01-35:05:02, 35:07-35:08:04, 35:10-35:11, 35:14:01-35:14:02, 35:16-35:17, 35:19-35:21, 35:23-35:30, 35:32, 35:35, 35:40N-35:43, 35:45, 35:48-35:54, 35:57-35:58, 35:61-35:62, 35:64, 35:66-35:69, 35:71-35:72, 35:76-35:80, 35:82, 35:86, 35:89, 35:91-35:94, 35:97, 35:99, 39:07, 39:43, 42:09, 44:17, 44:43, 46:01:01-46:09, 46:12-46:17, 46:19-46:22, 48:02:01-48:02:02, 48:14, 51:45, 53:01:01-53:03, 53:05-53:06, 53:08:01-53:18, 53:20-53:23, 54:06, 54:09, 55:14, 56:03, 56:09, 56:18, 57:01:01-57:01:07, 57:06, 57:08, 57:10-57:11, 57:14-57:16, 57:18-57:24, 57:26-57:27, 57:29-57:31, 58:01:01-58:02, 58:04-58:07, 58:09-58:19, 58:21-58:26											11	
*07:12, 07:14, 07:18:01-07:18:02, 27:19, 27:30, 35:33, 44:16, 44:21, 44:64:01-44:64:02, 48:17, C*03:34			3									
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

Lot No.: **25K**

Lot-specific information

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90	130	105	90	230	155	195	105	120	135	200	Length of spec.
180		250	205		205			190			PCR product(s)
13	14	15	16	17	18	19	20	21	22	23	Well No.
											*07:05:01-07:06, 07:32, 07:40, 07:53, 07:69, 07:80, 07:90, 07:97, 15:150, 27:07-27:08, 27:11-27:12, 27:18, 27:20, 27:24, 27:26-27:27, 27:33, 27:40, 27:42-27:44, 35:09:02, 35:18, 35:31, 35:84, 40:01:01-40:02:10, 40:05, 40:07, 40:09-40:16, 40:18-40:19, 40:21-40:24, 40:26- 40:29, 40:31-40:33, 40:35-40:37, 40:40, 40:42-40:43, 40:45-40:46, 40:49-40:50, 40:54-40:58, 40:61-40:63, 40:65-40:67, 40:69, 40:72:01-40:74, 40:76-40:82, 40:84- 40:85, 40:87-40:90, 40:92, 40:94, 40:97-40:98, 41:04, 41:10, 44:09, 44:31, 44:46, 44:75, 44:90, 45:01, 45:03- 45:08, 45:11, 47:02, 48:01:01-48:01:02, 48:03:01-48:07, 48:09-48:11, 48:13, 48:15-48:16, 48:18-48:22, 50:01:01- 50:02, 50:04-50:08, 51:01:09, 51:24:02-51:24:03, 51:81, 55:08, 58:27, 81:01-81:04N
											*07:09, 07:11, 07:17, 08:28, 08:37, 15:01:01:01- 15:01:04, 15:01:06-15:03:03, 15:05:01-15:08, 15:11:01- 15:15, 15:18:01-15:18:02, 15:18:04-15:21, 15:23-15:29, 15:31-15:36, 15:38:01-15:40, 15:43-15:44, 15:46-15:47, 15:49-15:52, 15:54-15:57, 15:60-15:62, 15:64-15:66, 15:68-15:72, 15:74-15:76, 15:78:01-15:82, 15:84-15:85, 15:87-15:89, 15:91-15:94N, 15:96-15:98, 15:101-15:129, 15:131-15:132, 15:134-15:136, 15:138-15:149N, 15:151- 15:154, 15:156-15:161, 15:163-15:167, 15:169-15:176, 15:178-15:182N, 15:184-15:187, 15:189-15:191, 18:01:01-18:08, 18:10-18:15, 18:17N-18:25, 18:27- 18:40, 18:42-18:47, 18:49, 27:41, 35:01:01-35:01:19, 35:05:01-35:05:02, 35:07-35:08:04, 35:10-35:11, 35:14:01-35:14:02, 35:16-35:17, 35:19-35:21, 35:23- 35:30, 35:32, 35:35, 35:40N-35:43, 35:45, 35:48-35:54, 35:57-35:58, 35:61-35:62, 35:64, 35:66-35:69, 35:71- 35:72, 35:76-35:80, 35:82, 35:86, 35:89, 35:91-35:94, 35:97, 35:99, 39:07, 39:43, 42:09, 44:17, 44:43, 46:01:01-46:09, 46:12-46:17, 46:19-46:22, 48:02:01- 48:02:02, 48:14, 51:45, 53:01:01-53:03, 53:05-53:06, 53:08:01-53:18, 53:20-53:23, 54:06, 54:09, 55:14, 56:03, 56:09, 56:18, 57:01:01-57:01:07, 57:06, 57:08, 57:10- 57:11, 57:14-57:16, 57:18-57:24, 57:26-57:27, 57:29- 57:31, 58:01:01-58:02, 58:04-58:07, 58:09-58:19, 58:21- 58:26
											*07:12, 07:14, 07:18:01-07:18:02, 27:19, 27:30, 35:33, 44:16, 44:21, 44:64:01-44:64:02, 48:17, C*03:34
13	14	15	16	17	18	19	20	21	22	23	Well No.

Lot No.: **25K**

Lot-specific information

www.olerup-ssp.com

Length of spec. PCR product(s)	140	95	245	245	125	105	190	185	230	105	280	90
					150			225		210		
					180							
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
*07:34, 08:01:02, 08:06, 08:20, 15:188, 40:39, 40:51, 41:01-41:03:02, 41:06-41:09, 41:11, 54:11, 55:04, C*15:24							7			10		
*07:78				4	5			8		10		
*07:84, 51:82				4				8				
*08:01:01, 08:01:03-08:05, 08:08N-08:10, 08:12:01-08:13, 08:15-08:19N, 08:22-08:24, 08:26-08:27, 08:29-08:34, 08:36, 08:38-08:39, 08:41-08:48, 08:50-08:61, 14:13, 15:83, 18:26, 35:87, 37:12, 38:01:01, 38:01:03-38:01:04, 38:05- 38:07, 38:09-38:11, 38:13-38:14, 38:16-38:17, 38:19-38:21, 39:01:01:01-39:01:01:02L, 39:01:03-39:05:02, 39:08-39:14, 39:16, 39:18- 39:20, 39:22-39:24, 39:26-39:28, 39:30-39:31, 39:35-39:37, 39:39, 39:41-39:42, 39:44-39:46, 39:48, 39:51-39:54, 39:56, 41:05, 41:12, 42:01:01-42:02, 42:04-42:08, 42:10-42:13, 51:21, 51:36, 54:04, 55:17, 55:20, 55:28, 56:12, 58:20, 59:02, 67:01:01-67:02, 78:07							7					
*08:35							7				11	
*15:04						6		8			11	
*15:16, 15:67, 15:95, 15:155, 35:37, 49:04-49:05, 51:37, 51:63, 54:14, 55:23						6					11	
*15:18:03											11	12
*15:42, 15:86, 35:60, 46:18, 54:01-54:02, 54:05N, 54:07-54:08N, 54:12-54:13, 54:17-54:19, 54:21, 55:01:01-55:03, 55:05, 55:07, 55:10-55:12, 55:15- 55:16, 55:18-55:19, 55:25-55:26, 55:29-55:31, 55:33-55:36, 55:38-55:40, 56:23, 59:01, 59:05					5		7					
*15:53, 15:183, 35:63, 35:90, 40:03, 40:20, 40:38, 40:52, 40:60, 46:10										10	11	
*15:73					5							12
*15:137, 39:33						6		8				
*18:09											11	
*18:48, 35:47											11	
*27:01, 37:10, 38:08, 44:15, 44:55, 49:02												
*27:14, 49:07, 51:16, 51:31, 51:34, 52:08				4								
*35:15, 57:13, 57:25			3								11	
*35:46											11	
*35:88, 40:04, 40:30, 40:34, 40:64, 40:99			3							10		
*38:02:01-38:04, 38:15, 38:18							7					
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

Lot No.: **25K**

Lot-specific information

www.olerup-ssp.com

90	130	105	90	230	155	195	105	120	135	200	Length of spec. PCR product(s)
180		250	205		205			190			
13	14	15	16	17	18	19	20	21	22	23	Well No.
											*07:34, 08:01:02, 08:06, 08:20, 15:188, 40:39, 40:51, 41:01-41:03:02, 41:06-41:09, 41:11, 54:11, 55:04, C*15:24
		15									*07:78
											*07:84, 51:82
											*08:01:01, 08:01:03-08:05, 08:08N-08:10, 08:12:01-08:13, 08:15-08:19N, 08:22-08:24, 08:26-08:27, 08:29-08:34, 08:36, 08:38-08:39, 08:41-08:48, 08:50-08:61, 14:13, 15:83, 18:26, 35:87, 37:12, 38:01:01, 38:01:03-38:01:04, 38:05- 38:07, 38:09-38:11, 38:13-38:14, 38:16-38:17, 38:19-38:21, 39:01:01:01-39:01:01:02L, 39:01:03-39:05:02, 39:08-39:14, 39:16, 39:18- 39:20, 39:22-39:24, 39:26-39:28, 39:30-39:31, 39:35-39:37, 39:39, 39:41-39:42, 39:44-39:46, 39:48, 39:51-39:54, 39:56, 41:05, 41:12, 42:01:01-42:02, 42:04-42:08, 42:10-42:13, 51:21, 51:36, 54:04, 55:17, 55:20, 55:28, 56:12, 58:20, 59:02, 67:01:01-67:02, 78:07
											*08:35
											*15:04
											*15:16, 15:67, 15:95, 15:155, 35:37, 49:04-49:05, 51:37, 51:63, 54:14, 55:23
											*15:18:03
											*15:42, 15:86, 35:60, 46:18, 54:01-54:02, 54:05N, 54:07-54:08N, 54:12-54:13, 54:17-54:19, 54:21, 55:01:01-55:03, 55:05, 55:07, 55:10-55:12, 55:15- 55:16, 55:18-55:19, 55:25-55:26, 55:29-55:31, 55:33-55:36, 55:38-55:40, 56:23, 59:01, 59:05
											*15:53, 15:183, 35:63, 35:90, 40:03, 40:20, 40:38, 40:52, 40:60, 46:10
		15									*15:73
											*15:137, 39:33
		15									*18:09
							20				*18:48, 35:47
		15									*27:01, 37:10, 38:08, 44:15, 44:55, 49:02
											*27:14, 49:07, 51:16, 51:31, 51:34, 52:08
											*35:15, 57:13, 57:25
			16				20				*35:46
											*35:88, 40:04, 40:30, 40:34, 40:64, 40:99
		15									*38:02:01-38:04, 38:15, 38:18
13	14	15	16	17	18	19	20	21	22	23	Well No.

Length of spec.	140	95	245	245	125	105	190	185	230	105	280	90
PCR product(s)					150			225		210		
					180							
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
*38:12							7					
*39:06:01-39:06:02, 39:34, 39:57, 55:13, 59:03						6	7					
*39:17, C*02:06												12
*39:50				4		6						
*40:06:01:01-40:06:02, 40:44, 40:53, 40:70, 40:75, 40:83, 40:93, 40:95, 45:10, 51:10				4						10		
*40:08, 40:25										10		
*40:47										10		
*40:48										10		12
*40:59			3							10	11	
*40:68			3							10		
*40:71					5					10		12
*40:86, 73:01				4		6				10		
*40:91		2								10		
*40:96				4						10		
*44:10, 46:11, 51:15, 54:03, 56:01:01-56:01:02, 56:01:04, 56:07-56:08, 56:13-56:14, 56:16-56:17, 56:19N-56:20, 56:24-56:29, 59:04, 82:01-82:03					5							
*44:12, 51:74												
*44:60	1											
*45:02, 50:09						6				10		
*45:09						6				10	11	
*47:03										w		
*49:09												
*51:05, 51:29, 51:54, 51:61								8				
*51:13:01-51:13:02, 51:92, 52:14, 56:22						6						
*51:62					5							12
*51:93		2		4								
*54:10, 54:20, 55:21, 55:37					5		7	8				
*54:22					5		7					
*55:09				4	5			8				
*55:22, 55:24				4	5							
*55:32						6	7				11	
*56:01:03					5							
*56:02, 56:04					5							12
*56:10					5		7					12
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

Lot No.: **25K**

Lot-specific information

www.olerup-ssp.com

90	130	105	90	230	155	195	105	120	135	200	Length of spec. PCR product(s)
180		250	205		205			190			
13	14	15	16	17	18	19	20	21	22	23	Well No.
							20				*38:12
											*39:06:01-39:06:02, 39:34, 39:57, 55:13, 59:03
											*39:17, C*02:06
											*39:50
											*40:06:01:01-40:06:02, 40:44, 40:53, 40:70, 40:75, 40:83, 40:93, 40:95, 45:10, 51:10
							20				*40:08, 40:25
		15									*40:47
		15									*40:48
											*40:59
							20				*40:68
											*40:71
											*40:86, 73:01
14											*40:91
		15									*40:96
											*44:10, 46:11, 51:15, 54:03, 56:01:01-56:01:02, 56:01:04, 56:07-56:08, 56:13-56:14, 56:16-56:17, 56:19N-56:20, 56:24-56:29, 59:04, 82:01-82:03
							20				*44:12, 51:74
											*44:60
											*45:02, 50:09
											*45:09
											*47:03
			16								*49:09
											*51:05, 51:29, 51:54, 51:61
											*51:13:01-51:13:02, 51:92, 52:14, 56:22
			16								*51:62
14											*51:93
											*54:10, 54:20, 55:21, 55:37
		15									*54:22
											*55:09
											*55:22, 55:24
											*55:32
		15									*56:01:03
											*56:02, 56:04
											*56:10
13	14	15	16	17	18	19	20	21	22	23	Well No.

¹The internal positive control primer pairs amplify segments of the human growth hormone gene. The two different control primer pairs give rise to either an internal positive control band of 1070 base pairs, for most wells, or a band of 800 base pairs, for some wells.

Well number 1 contains the primer pair giving rise to the shorter, 800 bp, internal positive control band in order to help in the correct orientation of the HLA-B*13 subtyping.

In addition, wells number 2, 3, 5, 10, 11, 13 and 15 contain the primer pair giving rise to the shorter, 800 bp, internal positive control band in order to allow kit identification.

²The nucleotide position, in the 2nd or 3rd exon, matching the specificity-determining 3'-end of the primer is given. Nucleotide numbering as on the www.ebi.ac.uk/imgt/hla web site. The sequence of the 3 terminal nucleotides of the primer is given.

³The nucleotide position, in 2nd or 3rd exon or the 3rd intron, matching the specificity-determining 3'-end of the primer is given in the anti-sense direction. Nucleotide numbering as on the www.ebi.ac.uk/imgt/hla web site. The sequence of the 3 terminal nucleotides of the primer is given.

⁴The sequence of the B*13:05 allele has been shown to be identical to B*13:04.

⁵The B*13:15 and B*13:33 alleles can be distinguished by the different sizes of the specific PCR products generated by primer mix 16.

⁶Primer mix 5: Specific PCR fragment of 125 bp in the B*13:22:01-13:22:02 and in the B*15:86, 35:60, 46:18, 54:01, 54:02, 54:05N, 54:07-54:08N, 54:10, 54:12-54:13, 54:19, 54:21, 54:22, 55:02, 55:07, 55:10, 55:12, 55:16, 55:18-55:19, 55:22, 55:26, 55:30, 55:34, 55:35, 55:37, 55:39, 56:10, 56:23, 59:01 and 59:05 alleles. Specific PCR fragment of 150 bp in the B*13:16, 13:20 and 13:31 alleles. Specific PCR fragment of 125 and 150 bp in the B*07:78, 15:42, 54:20, 55:01:01-55:01:06, 55:03, 55:05, 55:09, 55:11, 55:15, 55:21, 55:24-55:25, 55:29, 55:31, 55:33, 55:36, 55:38 and 55:40 alleles. Specific PCR fragments of 125 and 180 bp in the B*13:03 and in the B*40:71, 44:10, 46:11, 51:15, 54:03, 56:01:01-56:02, 56:04, 56:07-56:08, 56:13-56:14, 56:16-56:17, 56:19N-56:20, 56:24, 56:26-56:29, 59:04 and 82:01-82:03 alleles. Specific PCR fragments of 125, 150 and 180 bp in the B*15:73, 51:62 and 56:25 alleles. All bands may not always be visible.

Primer mix 8: Specific PCR fragment of 185 bp in the B*13:07N allele. Specific PCR fragment of 225 bp in the B*13:18 and 13:31 and in the B*07:78, 07:84, 15:04, 15:137, 39:33, 51:05, 51:29, 51:54, 51:61, 51:82, 54:1, 54:20, 55:09, 55:21 and 55:37 alleles.

Primer mix 10: Specific PCR fragment of 105 bp in the B*13:21 and the B*07:05:01-07:06, 07:32, 07:34, 07:40, 07:53, 07:69, 07:78, 07:80, 07:90, 07:97, 08:01:02, 08:06, 08:20, 15:150, 15:188, 27:07, 27:11, 27:20, 27:24, 27:27, 27:43, 35:09:02, 35:18, 35:31, 35:88, 40:10, 40:12, 40:13, 40:19, 40:21, 40:47, 40:58, 40:76-40:96, 44:31, 48:01:01-48:01:02, 48:03:01-48:07, 48:09-48:11, 48:13, 48:15-48:16, 48:18-48:22, 51:01:09, 51:10, 51:24:02-51:24:03, 51:81, 54:11, 55:04, 55:08, 58:27, 81:01-81:04N and in the C*15:24 alleles. Specific PCR fragment of 210 bp in the B*13:09 and the B*15:53, 15:183, 27:08, 27:12, 27:18, 27:26, 27:40, 27:42, 27:44, 35:63, 35:84, 35:90, 40:03, 40:09, 40:14:01, 40:14:02, 40:18, 40:20, 40:24, 40:26, 40:28, 40:31-40:33, 40:42, 40:44, 40:59, 40:60, 40:71, 40:72:01, 40:77, 40:87, 40:95, 40:98, 41:01-41:04, 41:06, 41:07, 41:09-41:11, 44:09, 44:46, 44:75, 44:90, 45:01-45:11, 46:10, 47:02, 47:03w, 50:01:01-50:02 and 50:04-50:09 alleles. Specific PCR fragments of 105 and 210 bp in the B*27:33, 40:01:01-40:02:10, 40:04-40:08, 40:11, 40:14:03-40:16, 40:22N, 40:23, 40:27, 40:29-40:30, 40:34-40:40, 40:43, 40:45-40:46, 40:48-40:51, 40:53-40:57, 40:61-40:70, 40:72:02-40:75, 40:78-40:86, 40:88-40:94, 40:97, 40:99, 41:08 and 73:01 alleles.

Primer mix 13: Specific PCR fragment of 90 bp in the B*13:19 allele. Specific PCR fragment of 180 bp in the B*13:12 allele.

Primer mix 15: Specific PCR fragment of 105 bp in the B*13:22:01 and in the B*07:78, 15:73, 40:48, 54:22 and 56:01:03 alleles. Specific PCR fragment of 250 bp in the B*13:14 and 13:23 and in the B*18:09, 27:01, 37:10, 38:02:01-38:04, 38:08, 38:15, 38:18, 40:47, 40:96, 44:15, 44:55 and 49:02 alleles.

Primer mix 16: Specific PCR fragment of 90 bp in the B*13:17 and 13:33 and the B*35:46 alleles. Specific PCR fragment of 205 bp in the B*13:15 and in the B*49:09 and 51:62 alleles.

Primer mix 18: Specific PCR fragment of 155 bp in the B*13:28 allele. Specific PCR fragment of 205 bp in the B*13:32 allele.

Primer mix 21: Specific PCR fragment of 120 bp in the B*13:29 allele. Specific PCR fragment of 190 bp in the B*13:30 allele.

'w', may be weakly amplified.

CELL LINE VALIDATION SHEET																					
HLA-B*13 SSP subtyping kit																					
					Well																
						1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
				Prod. No.:		201072201	201072202	201072203	201072204	201072205	201072206	201072207	201072208	201072209	201072210	201072211	201072212	201072213	201072214	201072215	201072216
	IHWC cell line		B*																		
1	9001	SA	*07:02		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
2	9280	LK707	*52:01	*73:01	-	-	-	+	-	+	-	-	-	-	+	-	-	-	-	-	-
3	9011	E4181324	*52:01		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
4	9275	GU373	*15:10	*53:01	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
5	9009	KAS011	*37:01		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
6	9353	SM	*39:01	*51:01	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-
7	9020	QBL	*18:01		-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
8	9025	DEU	*35:01		-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
9	9026	YAR	*38:01		-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-
10	9107	LKT3	*54:01		-	-	-	-	+	-	+	-	-	-	-	-	-	-	-	-	-
11	9051	PITOUT	*44:03		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
12	9052	DBB	*57:01		-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
13	9004	JESTHOM	*27:05		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
14	9071	OLGA	*15:01	*15:20	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
15	9075	DKB	*40:01		-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-
16	9037	SWEIG007	*40:02		-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-
17	9282	CTM3953540	*08:01	*55:01	-	-	-	-	+	-	+	-	-	-	-	-	-	-	-	-	-
18	9257	32367	*14:01	*56:01	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
19	9038	BM16	*18:01		-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
20	9059	SLE005	*40:01		-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-
21	9064	AMALA	*15:01		-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
22	9056	KOSE	*35:03		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
23	9124	IHL	*40:02	*56:02	-	-	-	-	+	-	-	-	-	-	+	-	+	-	-	-	-
24	9035	JBUSH	*38:01		-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-
25	9049	IBW9	*14:02		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
26	9285	WT49	*58:01		-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
27	9191	CH1007	*07:05	*51:01	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-
28	9320	BEL5GB	*44:02	*44:03	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
29	9050	MOU	*44:03		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
30	9021	RSH	*42:01		-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
31	9019	DUCAF	*18:01		-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
32	9297	HAG	*41:02		-	-	-	-	-	-	+	-	-	-	+	-	-	-	-	-	-
33	9098	MT14B	*40:01		-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-
34	9104	DHIF	*38:01		-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-
35	9302	SSTO	*44:02		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
36	9024	KT17	*15:01	*35:01	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
37	9065	HHKB	*07:02		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
38	9099	LZL	*15:01		-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
39	9315	CML	*08:01	*27:05	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-
40	9134	WHONP199	*13:02	*46:01	+	+	-	+	-	-	-	-	-	-	-	+	-	-	-	-	-
41	9055	H0301	*14:02		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
42	9066	TAB089	*46:01		-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
43	9076	T7526	*46:01		-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
44	9057	TEM	*38:01		-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-
45	9239	SHJO	*42:01	*50:01	-	-	-	-	-	-	-	+	-	-	+	-	-	-	-	-	-
46	9013	SCHU	*07:02		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
47	9045	TUBO	*51:01		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
48	9303	TER-ND	*35:01	*44:03	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-

CELL LINE VALIDATION SHEET												
HLA-B*13 SSP subtyping kit												
					Well							
						17	18	19	20	21	22	23
					Prod. No.:	201072217	201072218	201072219	201072220	201072221	201072222	201072223
	IHWC cell line		B*									
1	9001	SA	*07:02			-	-	-	-	-	-	-
2	9280	LK707	*52:01	*73:01		-	-	-	-	-	-	-
3	9011	E4181324	*52:01			-	-	-	-	-	-	-
4	9275	GU373	*15:10	*53:01		-	-	-	-	-	-	-
5	9009	KAS011	*37:01			-	-	-	-	-	-	-
6	9353	SM	*39:01	*51:01		-	-	-	-	-	-	-
7	9020	QBL	*18:01			-	-	-	-	-	-	-
8	9025	DEU	*35:01			-	-	-	-	-	-	-
9	9026	YAR	*38:01			-	-	-	-	-	-	-
10	9107	LKT3	*54:01			-	-	-	-	-	-	-
11	9051	PITOUT	*44:03			-	-	-	-	-	-	-
12	9052	DBB	*57:01			-	-	-	-	-	-	-
13	9004	JESTHOM	*27:05			-	-	-	-	-	-	-
14	9071	OLGA	*15:01	*15:20		-	-	-	-	-	-	-
15	9075	DKB	*40:01			-	-	-	-	-	-	-
16	9037	SWEIG007	*40:02			-	-	-	-	-	-	-
17	9282	CTM3953540	*08:01	*55:01		-	-	-	-	-	-	-
18	9257	32367	*14:01	*56:01		-	-	-	-	-	-	-
19	9038	BM16	*18:01			-	-	-	-	-	-	-
20	9059	SLE005	*40:01			-	-	-	-	-	-	-
21	9064	AMALA	*15:01			-	-	-	-	-	-	-
22	9056	KOSE	*35:03			-	-	-	-	-	-	-
23	9124	IHL	*40:02	*56:02		-	-	-	-	-	-	-
24	9035	JBUSH	*38:01			-	-	-	-	-	-	-
25	9049	IBW9	*14:02			-	-	-	-	-	-	-
26	9285	WT49	*58:01			-	-	-	-	-	-	-
27	9191	CH1007	*07:05	*51:01		-	-	-	-	-	-	-
28	9320	BEL5GB	*44:02	*44:03		-	-	-	-	-	-	-
29	9050	MOU	*44:03			-	-	-	-	-	-	-
30	9021	RSH	*42:01			-	-	-	-	-	-	-
31	9019	DUCAF	*18:01			-	-	-	-	-	-	-
32	9297	HAG	*41:02			-	-	-	-	-	-	-
33	9098	MT14B	*40:01			-	-	-	-	-	-	-
34	9104	DHIF	*38:01			-	-	-	-	-	-	-
35	9302	SSTO	*44:02			-	-	-	-	-	-	-
36	9024	KT17	*15:01	*35:01		-	-	-	-	-	-	-
37	9065	HHKB	*07:02			-	-	-	-	-	-	-
38	9099	LZL	*15:01			-	-	-	-	-	-	-
39	9315	CML	*08:01	*27:05		-	-	-	-	-	-	-
40	9134	WHONP199	*13:02	*46:01		-	-	-	-	-	-	-
41	9055	H0301	*14:02			-	-	-	-	-	-	-
42	9066	TAB089	*46:01			-	-	-	-	-	-	-
43	9076	T7526	*46:01			-	-	-	-	-	-	-
44	9057	TEM	*38:01			-	-	-	-	-	-	-
45	9239	SHJO	*42:01	*50:01		-	-	-	-	-	-	-
46	9013	SCHU	*07:02			-	-	-	-	-	-	-
47	9045	TUBO	*51:01			-	-	-	-	-	-	-
48	9303	TER-ND	*35:01	*44:03		-	-	-	-	-	-	-

CERTIFICATE OF ANALYSIS

Olerup SSP® HLA-B*13 SSP

Product number: 101.515-12u – without *Taq* polymerase
Lot number: 25K
Expiry date: 2012-August-01
Number of tests: 12
Number of wells per test: 23

Well specifications:

Well No.	Production No.	Well No.	Production No.	Well No.	Production No.
1	2010-722-01	9	2010-722-09	17	2010-722-17
2	2010-722-02	10	2010-722-10	18	2010-722-18
3	2010-722-03	11	2010-722-11	19	2010-722-19
4	2010-722-04	12	2010-722-12	20	2010-722-20
5	2010-722-05	13	2010-722-13	21	2010-722-21
6	2010-722-06	14	2010-722-14	22	2010-722-22
7	2010-722-07	15	2010-722-15	23	2010-722-23
8	2010-722-08	16	2010-722-16		

The specificity of each primer solution of the kit has been tested against 48 well characterized IHWC cell line DNAs.

No DNAs carrying the alleles to be amplified by primer solutions 9, 13, 14, 16 to 19 and 21 to 23 were available. The specificities of the primers in primer mixes 13, 14 and 16 were tested by separately adding one additional 5'-primer, respectively one additional 3'-primer. In primer solutions 9, 18, 22 and 23 it was only possible to test the 5'-primers, the 3'-primers were not possible to test. In primer solutions 17, 19 and 21 it was only possible to test the 3'-primers, the 5'-primers were not possible to test. In primer solution 13, one 5'-primer and one 3'-primer was not possible to test. Additional primers in primer solutions 5, 8 and 15 were tested by separately adding one 5'-primer and/or one 3'-primer. In primer solution 8, one 3'-primer was not possible to test.

Results: No false positive or false negative amplifications were obtained.

Date of approval: 2010-August-23

Approved by:

Quality Control, Supervisor

Lot No.: **25K**

Lot-specific information

www.olerup-ssp.com

Declaration of Conformity

Product name: *Olerup* SSP® HLA-B*13
Product number: 101.515-12u
Lot number: 25K

Intended use: HLA-B*13 high resolution histocompatibility testing

Manufacturer: *Olerup* SSP AB
Hasselstigen 1
SE-133 33 Saltsjöbaden, Sweden
Phone: +46-8-717 88 27
Fax: +46-8-717 88 18

We, *Olerup* SSP AB, hereby declare that this product, to which this Declaration of Conformity relates is in conformity with the following Standard(s) and other normative document(s) ISO 9001:2008 and ISO 13485:2003, following the provisions of the 98/79/EC Directive on *in vitro* diagnostic medical devices, Annex II List B, conformity assessed using Annex IV, as transposed into the national laws of the Member States of the European Union.

The Technical Documentation File is maintained at *Olerup* SSP AB, Hasselstigen 1, SE-133 33 Saltsjöbaden, Sweden.

The Authorized Representative located within the Community is: *Olerup* SSP AB.

Notified Body: Lloyd's Register Quality Assurance Limited, Hiramford, Middlemarch Office Village, Siskin Drive, Coventry CV3 4FJ, United Kingdom. (Notified Body number: 0088.)

Saltsjöbaden, Sweden
2010-August-23

Olle Olerup
Managing Director

Lot No.: **25K**

Lot-specific information

www.olerup-ssp.com

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