

101.547-06 – including *Taq* pol., IFU-01
101.547-06u – without *Taq* pol., IFU-02

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“Instructions for Use” (IFU)

Lot No.: **56S**

Lot-specific information

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Olerup SSP® HLA-B*49

Product number:	101.547-06 – including <i>Taq</i> polymerase 101.547-06u – without <i>Taq</i> polymerase
Lot number:	56S
Expiry date:	2016-January-01
Number of tests:	6
Number of wells per test:	16
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. 56S.

CHANGES COMPARED TO THE PREVIOUS *OLERUP SSP*® HLA-B*49 LOT (34R).

The HLA-B*49 kit is updated for new alleles to enable separation of:

- Confirmed¹ alleles as listed in the IMGT/HLA database
- Polymorphisms in exons outside of the region encoding the peptide binding domain
- Null and Alternatively expressed alleles

The Lot-specific information for HLA-B*49 including and without *Taq* polymerase is described in one common Product Insert.

¹As described in section Uniquely Identified Alleles.

The HLA-B*49 specificity and interpretation tables have been updated for the HLA-B alleles described since the previous *Olerup SSP*® HLA-B*49 lot was made (**Lot No. 34R**).

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

Well	5'-primer	3'-primer	rationale
2	Modified	Added	Modified 5'-primer for improved specificity, 3'-primer added for the B*49:26 allele, exchanged positive control primer pair.

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PRODUCT DESCRIPTION

HLA-B*49 SSP typing

CONTENT

The primer set contains 5'- and 3'-primers for identifying the B*49:01 to B*49:26 alleles.

PLATE LAYOUT

Each HLA-B*49 test consists of 16 PCR reactions in a 16 well cut PCR plate.

1	2	3	4	5	6	7	8
9	10	11	12	13	14	15	16

The 16 well PCR plate is marked with 'HLA-B*49' in silver/gray ink.

Well No. 1 is marked with the Lot No. '56S'.

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 16 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*49 SSP subtypings will be influenced by two B*07, five B*08, most B*13, several B*15, two B*18, three B*27, six B*35, two B*37, the B*38:30, ten B*39, several B*40, fourteen B*41, the B*42:04, most B*44, the B*45, three B*46, the B*47, the B*50, most B*51, the B*52, three B*53, most B*54, most B*55, most B*56, five B*57, three B*58, the B*59, the B*73 and the B*78 alleles when present on the other haplotype. In addition, primer mix 3 will amplify the C*03:87, C*05:27 and C*05:39 alleles and primer mix 16 will amplify the C*02:02:23 allele.

UNIQUELY IDENTIFIED ALLELES

All the HLA-B*49 alleles, i.e. **B*49:01 to B*49:26**, recognized by the HLA Nomenclature Committee in April 2013¹ will be amplified by the primers in the HLA-B*49 SSP kit.

The HLA-B*49 kit enables separation of the confirmed HLA-B*49 alleles as listed in the IMGT/HLA database. An HLA allele is listed as confirmed by IMGT/HLA if it has been sequenced by more than a single laboratory or from multiple sources. Current allele confirmation status for HLA-B*49 alleles is listed below.

The HLA-B*49 kit also enables identification of polymorphisms in exons outside of the region encoding the peptide binding domain and of null and alternatively expressed alleles.

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The HLA-B*49 subtyping kit cannot distinguish the silent mutations in the B*49:01:01 to B*49:01:04 alleles.

The B*49:07 and 49:21 alleles can be distinguished by the different sizes of the specific PCR products generated by primer mix 9.

The B*49:08 and 49:16 alleles can be distinguished by the different sizes of the specific PCR products generated by primer mix 10.

The B*49:13 and 49:14 alleles can be distinguished by the different sizes of the specific PCR products generated by primer mix 15.

¹HLA-B alleles listed on the IMGT/HLA web page 2013-April-17, release 3.12.0, www.ebi.ac.uk/imgt/hla.

Allele	Status ¹	Allele	Status ¹	Allele	Status ¹
B*49:01:01	Confirmed	B*49:08	Unconfirmed	B*49:18	Confirmed
B*49:01:02	Unconfirmed	B*49:09	Confirmed	B*49:19N	Confirmed
B*49:01:03	Unconfirmed	B*49:10	Unconfirmed	B*49:20	Unconfirmed
B*49:01:04	Unconfirmed	B*49:11	Confirmed	B*49:21	Unconfirmed
B*49:02	Confirmed	B*49:12	Unconfirmed	B*49:22	Unconfirmed
B*49:03	Unconfirmed	B*49:13	Unconfirmed	B*49:23	Unconfirmed
B*49:04	Unconfirmed	B*49:14	Unconfirmed	B*49:24	Unconfirmed
B*49:05	Unconfirmed	B*49:15	Confirmed	B*49:25	Unconfirmed
B*49:06	Unconfirmed	B*49:16	Unconfirmed	B*49:26	Unconfirmed
B*49:07	Unconfirmed	B*49:17	Confirmed		

ALLELE CONFIRMATION STATUS

¹Allele status “confirmed” or “unconfirmed” as listed on the IMGT/HLA web page 2013-April-17, release 3.12.0, www.ebi.ac.uk/imgt/hla.

RESOLUTION IN HOMO- AND HETEROZYGOTES

A total of 29 alleles generate 20 amplification patterns that can be combined in 210 homozygous and heterozygous combinations. 153 of these genotypes do not give rise to unique amplification patterns. The different lengths of the specific PCR products were not considered in these calculations.

```

+-----++  -----  *49:04, *49:05 = *49:05, *49:05
-++-+-+--  -----  *49:02, *49:02 = *49:02, *49:26
+---+---+  -----+  *49:01:01, *49:15 = *49:01:01, *49:18 = *49:15, *49:15 = *49:15, *49:18
+---+---+  -----+-  *49:01:01, *49:13 = *49:13, *49:13
+---+---+  -----+-  *49:01:01, *49:12 = *49:12, *49:12
+---+---+  -----+-  *49:01:01, *49:11 = *49:11, *49:11
+---+---+  -----+-  *49:01:01, *49:10 = *49:10, *49:10
+---+---+  -++-----  *49:01:01, *49:08 = *49:08, *49:08
+---+---+  +-----  *49:01:01, *49:07 = *49:07, *49:07
+---+---+  -----  *49:01:01, *49:04 = *49:01:01, *49:20 = *49:04, *49:20 = *49:20, *49:20

```

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+---++--	*49:01:01, *49:03 = *49:01:01, *49:09 = *49:03, *49:09 = *49:09, *49:09	
+---++--	*49:01:01, *49:22 = *49:22, *49:22	
+---++--	*49:01:01, *49:06 = *49:01:01, *49:26 = *49:06, *49:06 = *49:06, *49:26	
+---++--	*49:13, *49:15 = *49:13, *49:18	
+---++--	*49:12, *49:15 = *49:12, *49:18	
+---++--	*49:01:01, *49:17 = *49:12, *49:13 = *49:12, *49:17 = *49:13, *49:17 =	
+---++--	*49:17, *49:17	
+---++--	*49:11, *49:15 = *49:11, *49:18	
+---++--	*49:10, *49:15 = *49:10, *49:18	
+---++--	*49:08, *49:15 = *49:08, *49:18	
+---++--	*49:07, *49:15 = *49:07, *49:18	
+---++--	*49:01:01, *49:19N = *49:12, *49:19N = *49:19N, *49:19N	
+---++--	*49:04, *49:15 = *49:15, *49:20 = *49:18, *49:20	
+---++--	*49:04, *49:13 = *49:13, *49:20	
+---++--	*49:04, *49:12 = *49:12, *49:20	
+---++--	*49:04, *49:11 = *49:11, *49:20	
+---++--	*49:04, *49:10 = *49:10, *49:20	
+---++--	*49:04, *49:08 = *49:08, *49:20	
+---++--	*49:04, *49:07 = *49:07, *49:20	
+---++--	*49:01:01, *49:05 = *49:05, *49:20	
+---++--	*49:03, *49:15 = *49:03, *49:18 = *49:09, *49:15 = *49:09, *49:18	
+---++--	*49:03, *49:13 = *49:09, *49:13	
+---++--	*49:03, *49:12 = *49:09, *49:12	
+---++--	*49:03, *49:11 = *49:09, *49:11	
+---++--	*49:03, *49:10 = *49:09, *49:10	
+---++--	*49:03, *49:08 = *49:08, *49:09	
+---++--	*49:03, *49:07 = *49:07, *49:09	
+---++--	*49:03, *49:04 = *49:03, *49:20 = *49:04, *49:09 = *49:09, *49:20	
+---++--	*49:15, *49:22 = *49:18, *49:22	
+---++--	*49:04, *49:22 = *49:20, *49:22	
+---++--	*49:03, *49:22 = *49:09, *49:22	
+---++--	*49:06, *49:15 = *49:06, *49:18 = *49:15, *49:26 = *49:18, *49:26	
+---++--	*49:06, *49:13 = *49:13, *49:26	
+---++--	*49:06, *49:12 = *49:12, *49:26	
+---++--	*49:06, *49:11 = *49:11, *49:26	
+---++--	*49:06, *49:10 = *49:10, *49:26	
+---++--	*49:06, *49:08 = *49:08, *49:26	
+---++--	*49:06, *49:07 = *49:07, *49:26	
+---++--	*49:04, *49:06 = *49:04, *49:26 = *49:06, *49:20 = *49:20, *49:26	
+---++--	*49:03, *49:06 = *49:06, *49:09 = *49:09, *49:26	
+---++--	*49:01:01, *49:02 = *49:02, *49:06 = *49:02, *49:22 = *49:06, *49:22 =	
+---++--	*49:22, *49:26	
+---++--	*49:15, *49:17 = *49:17, *49:18	
+---++--	*49:15, *49:19N = *49:18, *49:19N	
+---++--	*49:13, *49:19N = *49:17, *49:19N	
+---++--	*49:04, *49:17 = *49:17, *49:20	
+---++--	*49:04, *49:19N = *49:05, *49:12 = *49:05, *49:19N = *49:19N, *49:20	
+---++--	*49:03, *49:17 = *49:09, *49:17	
+---++--	*49:03, *49:19N = *49:09, *49:19N	
+---++--	*49:03, *49:05 = *49:05, *49:09	
+---++--	*49:06, *49:17 = *49:17, *49:26	
+---++--	*49:06, *49:19N = *49:19N, *49:26	

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++--++++	-----	*49:05, *49:06 = *49:05, *49:26
+++--+-	-----+	*49:02, *49:15 = *49:02, *49:18
+++--+-	-----	*49:02, *49:04 = *49:02, *49:20

*49:01:01 = **49:01:01-49:01:04 and 49:23 and 49:25

*49:07 = *49:07 and 49:21

*49:08 = *49:08 and 49:16

*49:10 = *49:10 and 49:24

*49:13 = *49:13 and 49:14

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www.olerup-ssp.com**SPECIFICITY TABLE****HLA-B*49 SSP subtyping**

Specificities and sizes of the PCR products of the 16 primer mixes used for
HLA-B*49 SSP subtyping

Primer Mix	Size of spec. PCR product ¹	Size of control band ²	Amplified HLA-B*49 alleles	Other amplified HLA-B alleles ³
1	145 bp	800 bp	*49:01:01-49:01:04, 49:04-49:25	*40:13, 40:19, 40:109, 40:117, 44:18, 44:25, 44:50, 44:95, 51:112, 53:30, 57:45, 57:51
2⁷	145 bp, 190 bp, 255 bp	800 bp	*49:02, 49:06, 49:26	*13:23, 13:55, 18:09, 27:01, 37:10, 39:31, 40:47, 40:96, 40:157, 40:201, 44:15, 44:55, 44:103, 58:13
3^{4,8}	115 bp, 145 bp	800 bp	*49:02, 49:22	*15:33, 15:248, 40:47, 40:96, 40:110, 40:157, 40:201, 44:02:01:01-44:02:12, 44:02:14-44:02:18, 44:02:20-44:05:02, 44:05:04, 44:07, 44:10, 44:12-44:17, 44:19N-44:24, 44:26-44:39, 44:41:01-44:43:02, 44:45, 44:47-44:49, 44:51-44:56N, 44:58N-44:59:02, 44:61N-44:74, 44:76-44:82, 44:84-44:89, 44:91-44:94, 44:96-44:128, 44:132-44:133, 44:135-44:151, 44:153-44:155, 44:157-44:169, 44:171N-44:175, 47:04, 51:126, 57:54, C*03:87, C*05:27, C*05:39
4⁹	170 bp, 205 bp	1070 bp	*49:03, 49:09	*13:15, 18:04, 40:01:11, 40:58, 44:02:10, 44:34:01, 51:07:01, 51:62, 51:106, 52:01:02, 52:01:04, 52:01:09, 52:02-52:03, 52:09, 52:21, 52:25, 56:34
5⁴	105 bp	1070 bp	*49:01:01-49:03, 49:06-49:17, 49:19N-49:26	*07:78, 13:02:01-13:02:14, 13:03, 13:08Q-13:09, 13:14-13:16, 13:18-13:19, 13:27, 13:30-13:34, 13:37-13:38, 13:40-13:42, 13:44-13:45, 13:48-13:49N, 13:53-13:56N, 13:58, 13:65-13:67, 15:42, 35:60, 44:15, 44:18, 45:01, 45:03-45:08, 45:10-45:14, 46:11, 46:18, 50:01:01-50:02, 50:04-50:08, 50:10-50:11, 50:13, 50:15, 50:18-50:19, 50:31-50:32, 51:15, 52:25, 54:01:01-54:03, 54:05N, 54:07-54:08N, 54:10, 54:12-54:13, 54:16-54:28, 55:01:01-55:01:06, 55:01:08-55:03, 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,

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				55:33-55:38, 55:40-55:41, 55:43, 55:45-55:48, 55:50, 55:52, 55:54-55:57, 56:01:01-56:01:05, 56:07-56:08, 56:13-56:14, 56:16-56:17, 56:19N-56:20:02, 56:23-56:30, 56:33- 56:39, 59:01:01:01-59:01:01:02, 59:04- 59:05
6⁵	160 bp	1070 bp	*49:01:01- 49:03, 49:06- 49:17, 49:19N- 49:26	*13:01:01-13:03, 13:06-13:09, 13:11- 13:12:02, 13:14-13:17, 13:19-13:20, 13:22:01-13:23, 13:25, 13:27-13:30, 13:32-13:34, 13:36-13:40, 13:42-13:45, 13:47-13:50, 13:52-13:58, 13:60-13:61, 13:63N-13:67, 15:42, 15:86, 15:224, 35:60, 39:17, 40:48, 40:71, 44:10, 46:11, 46:18, 50:01:01-50:02, 50:04- 50:08, 50:10-50:13, 50:15-50:19, 50:31- 50:32, 51:15, 51:62, 51:106, 52:25, 54:01:01-54:03, 54:05N, 54:07-54:08N, 54:12-54:13, 54:17-54:19, 54:21-54:28, 55:01:01-55:03, 55:05, 55:07, 55:10- 55:12, 55:15-55:16, 55:18-55:19, 55:22, 55:24-55:26, 55:29-55:31, 55:33-55:36, 55:38-55:45, 55:47-55:48, 55:50, 55:53- 55:55N, 55:57, 56:01:01-56:01:04, 56:02, 56:04, 56:07-56:08, 56:10, 56:14, 56:16-56:17, 56:19N-56:20:02, 56:23- 56:30, 56:33-56:39, 59:01:01:01- 59:01:01:02, 59:04-59:05
7^{4,10}	105 bp, 255 bp	1070 bp	*49:04- 49:05, 49:20	*13:04, 13:46, 15:04, 15:16:01-15:16:03, 15:67, 15:95, 15:137, 15:155, 15:222, 15:254, 35:37, 39:06:01-39:06:02, 39:33-39:34, 39:50, 39:57, 39:62, 39:64, 40:86, 40:159, 44:15, 44:18, 44:20, 44:47, 44:100, 45:01-45:03, 45:05- 45:07, 45:09, 45:11-45:14, 46:32, 50:02, 50:09, 51:13:01-51:13:02, 51:23, 51:37, 51:63, 51:92, 51:97, 51:108, 52:14- 52:15, 54:14, 55:13, 55:23, 55:32, 55:58, 56:22, 59:03, 73:01-73:02
8^{4,6,11}	70 bp, 155 bp	1070 bp	*49:05, 49:19N	*07:78, 07:84, 08:09, 08:84, 13:02:01- 13:02:05, 13:02:07-13:04, 13:08Q- 13:09, 13:14-13:16, 13:18-13:19, 13:27, 13:30-13:35, 13:37-13:38, 13:40-13:42, 13:44-13:49N, 13:53-13:56N, 13:58- 13:59, 13:62, 13:65-13:67, 15:04, 15:16:01-15:16:02, 15:42, 15:67, 15:83, 15:95, 15:137, 15:155, 15:222, 15:254,

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27:14, 27:81, 35:60, 39:06:01-39:06:02, 39:33-39:34, 39:50, 39:57, 39:62, 39:64, 40:06:01:01-40:06:07, 40:44, 40:53, 40:70, 40:75, 40:83, 40:86, 40:93, 40:95-40:96, 40:103, 40:109-40:110, 40:127, 40:131, 40:148, 40:159, 40:161-40:162, 40:165, 40:177, 40:190, 40:230, 41:01, 41:05-41:07, 41:09, 41:12, 41:14, 41:16-41:17, 41:20-41:22, 41:25-41:26, 42:04, 44:20, 44:47, 44:100, 45:14, 46:11, 46:18, 46:32, 50:20, 51:01:01-51:01:18, 51:01:20-51:03, 51:05, 51:07:01-51:16, 51:18-51:24:04, 51:26-51:41N, 51:43-51:44N, 51:48-51:55, 51:57-51:58, 51:60-51:61:02, 51:63, 51:65-51:80, 51:82-51:93, 51:95-51:134, 51:136-51:138, 51:140-51:147, 51:149N-51:152, 52:01:01:01-52:31, 54:01:01-54:05N, 54:07-54:08N, 54:10-54:14, 54:16-54:28, 55:01:01-55:01:06, 55:01:08-55:03, 55:05, 55:07, 55:09-55:13, 55:15-55:26, 55:28-55:48, 55:50, 55:52-55:58, 56:01:01-56:01:05, 56:05:01-56:08, 56:13-56:17, 56:19N-56:30, 56:33-56:39, 58:08:01-58:08:02, 59:01:01:01-59:05, 73:01-73:02, 78:01:01-78:07				
9¹²	155 bp, 240 bp	800 bp	*49:07, 49:21	*45:10, 50:31
10^{4,13}	80 bp, 250 bp	800 bp	*49:08, 49:16	
11	505 bp	1070 bp		*08:03, 08:52, 08:78, 37:34, 38:30, 44:06, 44:18, 44:25, 44:95, 51:08, 51:20, 51:36, 51:44N, 51:97, 51:141, 52:19, 53:22, 57:09, 57:24
12	135 bp	1070 bp	*49:10, 49:24	*15:46, 15:53, 15:106, 35:47, 35:154, 40:10:01-40:10:02, 40:40, 40:49, 40:58, 40:110, 40:129, 40:164, 40:210, 40:219, 40:229, 44:02:01:01-44:03:15, 44:03:17-44:05:04, 44:09-44:14, 44:16-44:17, 44:19N-44:39, 44:41:01-44:43:02, 44:45-44:48, 44:50-44:54, 44:56N, 44:58N-44:59:02, 44:61N-44:80, 44:82, 44:84-44:102, 44:104-44:110, 44:112-44:129, 44:132-44:133, 44:135-44:144, 44:146-44:155, 44:157-44:175,

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13⁵	150 bp	1070 bp	*49:11	47:01:01:01-47:08, 50:13, 53:30, 57:45 *13:01:01-13:03, 13:06-13:09, 13:11-13:12:02, 13:14-13:15, 13:17, 13:18 ^w , 13:19, 13:22:01-13:23, 13:25, 13:27-13:30, 13:32-13:34, 13:36-13:40, 13:42-13:45, 13:47, 13:49N-13:50, 13:52-13:58, 13:60-13:61, 13:63N-13:67, 15:86, 15:224, 35:60, 40:48, 40:71, 44:10, 46:11, 46:18, 51:15, 54:01:01-54:03, 54:05N, 54:07-54:08N, 54:10 ^w , 54:12-54:13, 54:17-54:19, 54:21-54:28, 55:02:01-55:02:06, 55:07, 55:10, 55:12, 55:16, 55:18-55:19, 55:22, 55:26, 55:30, 55:34-55:35, 55:37 ^w , 55:39, 55:41-55:43, 55:47-55:48, 55:50, 56:01:01-56:01:04, 56:02, 56:04, 56:07-56:08, 56:10, 56:14, 56:16-56:17, 56:19N-56:20:02, 56:23-56:24, 56:26-56:29, 56:33-56:36, 56:38N-56:39, 59:01:01:01-59:01:01:02, 59:04-59:05
14¹⁴	155 bp, 190 bp	1070 bp	*49:12, 49:17, 49:19N	*51:130
15^{5,15}	160 bp, 190 bp, 290 bp	1070 bp	*49:13- 49:14, 49:17	
16^{4,16}	95 bp, 135 bp	1070 bp	*49:15, 49:18	*35:01:10, 35:04:02, 40:28, 50:14, 51:56:01-51:56:02, 53:28, 55:01:04, C*02:02:23

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*49 SSP typings.

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

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.

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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*49 SSP subtyping. In addition, wells number 2, 3, 9 and 10 contains 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.

³Due to the sharing of sequence motifs between HLA-B alleles some non-HLA-B*49 alleles will be amplified by primer mixes 1 to 9, 11 to 14 and 16. In addition, primer mix 3 will amplify the C*03:87, C*05:27 and C*05:39 alleles and primer mix 16 will amplify the C*02:02:23 allele.

⁴HLA-specific PCR products shorter than 125 base pairs have a lower intensity and are less sharp than longer PCR products.

⁵Primer mixes 6, 13 and 15 may have tendencies of unspecific amplifications.

⁶Primer mix 8 may give rise to a lower yield of HLA-specific PCR product than the other B*49 primer mixes.

⁷Primer mix 2: Specific PCR fragment of 145 bp in the B*49:06 and the B*58:13 alleles. Specific PCR fragment of 190 bp in the B*49:26 and the B*39:31 alleles. Specific PCR fragment of 255 bp in the B*49:02 and the B*13:23, 13:55, 18:09, 27:01, 37:10, 40:47, 40:96, 40:157, 40:201, 44:15, 44:55 and 44:103 alleles.

⁸Primer mix 3: Specific PCR fragment of 115 bp in the B*49:22 and the B*15:33, 15:248, 51:126 and 57:54 and in the C*03:87, C*05:27 and C*05:39 alleles. Specific PCR fragment of 145 bp in the B*49:02 and the B*40:47, 40:96, 40:110, 40:157, 40:201, 44:02:01:01-44:02:12, 44:02:14-44:02:18, 44:02:20-44:05:02, 44:05:04, 44:07, 44:10, 44:12-44:17, 44:19N-44:24, 44:26-44:39, 44:41:01-44:43:02, 44:45, 44:47-44:49, 44:51-44:56N, 44:58N-44:59:02, 44:61N-44:74, 44:76-44:82, 44:84-44:89, 44:91-44:94, 44:96-44:128, 44:132-44:133, 44:135-44:151, 44:153-44:155, 44:157-44:169, 44:171N-44:175 and 47:04 alleles.

⁹Primer mix 4: Specific PCR fragment of 170 bp in the B*49:03 and the B*18:04, 40:01:11, 40:58, 44:02:10, 44:34:01, 51:07:01, 52:01:02, 52:01:04, 52:01:09, 52:02-52:03, 52:09, 52:21 alleles. Specific PCR fragment of 205 bp in the B*49:09 and the B*13:15, 51:62, 51:106, 52:25 and 56:34 alleles.

¹⁰Primer mix 7: Specific PCR fragment of 105 bp in the B*49:04 and 49:05 and the B*13:04, 13:46, 15:04, 15:16:01-15:16:03, 15:67, 15:95, 15:137, 15:155, 15:222, 35:37, 39:06:01-39:06:02, 39:33-39:34, 39:50, 39:57, 39:62, 39:64, 40:86, 40:159, 45:02, 45:09, 50:09, 51:13:01-51:13:02, 51:37, 51:63, 51:92, 51:97, 52:14, 54:14, 55:13, 55:23, 55:32, 55:58, 56:22, 59:03 and 73:01-73:02 alleles. Specific PCR fragment of 255 bp in the B*49:20 and the B*44:15, 44:18, 44:20, 44:47, 44:100, 45:01-45:03, 45:05-45:07, 45:09, 45:11-45:14, 50:02, 51:23, 51:108 and 52:15 alleles.

¹¹Primer mix 8: Specific PCR fragment of 70 bp in the B*49:05 and the B*07:78, 07:84, 08:09, 08:84, 13:02:01-13:02:05, 13:02:07-13:04, 13:08Q-13:09, 13:14-13:16, 13:18-13:19, 13:27, 13:30-13:35, 13:37-13:38, 13:40-13:42, 13:44-13:49N, 13:53-13:56N, 13:58-13:59, 13:62, 13:65-13:67, 15:04, 15:16:01-15:16:02, 15:42, 15:67, 15:83, 15:95, 15:137, 15:155, 15:222, 15:254, 27:14, 27:81, 35:60, 39:06:01-39:06:02, 39:33-39:34, 39:50, 39:57, 39:62, 39:64, 40:06:01:01-40:06:07, 40:44, 40:53, 40:70, 40:75, 40:83, 40:86, 40:93, 40:95-40:96, 40:103, 40:109-40:110, 40:127, 40:131, 40:148, 40:159, 40:161-40:162, 40:165, 40:177, 40:190, 40:230, 41:01, 41:05-41:07, 41:09, 41:12, 41:14, 41:16-41:17, 41:20-41:22, 41:25-41:26, 42:04, 44:20, 44:47, 44:100, 45:14, 46:11, 46:18, 46:32, 50:20, 51:01:01-51:01:18, 51:01:20-51:03, 51:05, 51:07:01-51:16, 51:18-51:24:04, 51:26-51:41N, 51:43-51:44N, 51:48-51:55, 51:57-51:58, 51:60-51:61:02, 51:63, 51:65-51:80, 51:82-51:93, 51:95-51:134, 51:136-51:138, 51:140-51:147, 51:149N-51:152, 52:01:01:01-52:31, 54:01:01-54:05N, 54:07-54:08N, 54:10-54:14, 54:16-54:28, 55:01:01-55:01:06, 55:01:08-55:03, 55:05, 55:07, 55:09-55:13, 55:15-55:26, 55:28-55:48, 55:50, 55:52-55:58, 56:01:01-56:01:05, 56:05:01-56:08, 56:13-56:17, 56:19N-56:30, 56:33-56:39, 58:08:01-58:08:02, 59:01:01:01-59:05, 73:01-73:02 and 78:01:01-78:07 alleles. Specific PCR fragment of 155 bp in the B*49:19N allele.

¹²Primer mix 9: Specific PCR fragment of 155 bp in the B*49:21 allele. Specific PCR fragment of 240 bp in the B*49:07 and the B*45:10 and 50:31 alleles.

¹³Primer mix 10: Specific PCR fragment of 80 bp in the B*49:16 allele. Specific PCR fragment of 250 bp in the B*49:08 allele.

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¹⁴Primer mix 14: Specific PCR fragment of 155 bp in the B*49:19N allele. Specific PCR fragment of 190 bp in the B*49:12 and 49:17 and the B*51:130 alleles.

¹⁵Primer mix 15: Specific PCR fragment of 160 bp in the B*49:13 allele. Specific PCR fragment of 190 bp in the B*49:17 allele. Specific PCR fragment of 290 bp in the B*49:14 allele.

¹⁶Primer mix 16: Specific PCR fragment of 95 bp in the B*49:18 and the B*35:01:10, 35:04:02, 40:28, 50:14, 51:56:01-51:56:02, 53:28 and 55:01:04 and in the C*02:02:23 alleles. Specific PCR fragment of 135 bp in the B*49:15 allele.

‘w’, may be weakly amplified.

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INTERPRETATION TABLE								
HLA-B*49 SSP subtyping								
Amplification patterns of the B*49:01 to B*49:26 alleles								
	Well ⁸							
	1	2	3	4	5	6	7	8
Length of spec. PCR product(s)	145	145	115	170	105	160	105	70
		190	145	205			255	155
		255						
Length of int. pos. control ¹	800	800	800	1070	1070	1070	1070	1070
5'-primer ²	206	97	206	141	357	420	357	357
	5' -gAA 3'	5' -TCC 3'	5' -gAA 3'	5' -ATT 3'	5' -Tgg 3'	5' -TTA 3'	5' -Tgg 3'	5' -Tgg 3'
		454	485	420				
		5' -ACA 3'	5' -CAA 3'	5' -TTA 3'				
3'-primer(s) ³	309	246	309	272	420	538	419	387
	5' -ATC 3'	5' -TAT 3'	5' -gTg 3'	5' -Tgg 3'	5' -gCT 3'	5' -CAG 3'	5' -Cgg 3'	5' -TCC 3'
		309	559	583			419	470
		5' -gTg 3'	5' -CAG 3'	5' -gTg 3'			5' -CgA 3'	5' -TCT 3'
		559					572	
		5' -CAG 3'					5' -gCg 3'	
Well No.	1	2	3	4	5	6	7	8
HLA-B allele ⁴								
*49:01:01-49:01:04, 49:23, 49:25	1				5	6		
*49:02		2	3		5	6		
*49:03				4	5	6		
*49:04	1						7	
*49:05	1						7	8
*49:06	1	2			5	6		
*49:07, 49:21 ⁵	1				5	6		
*49:08, 49:16 ⁶	1				5	6		
*49:09	1			4	5	6		
*49:10, 49:24	1				5	6		
*49:11	1				5	6		
*49:12	1				5	6		
*49:13-49:14 ⁷	1				5	6		
*49:15	1				5	6		
*49:17	1				5	6		
*49:18	1							
*49:19N	1				5	6		8
Well No.	1	2	3	4	5	6	7	8

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Length of spec.	145	145	115	170	105	160	105	70
PCR product(s)		190	145	205			255	155
		255						
Well No.	1	2	3	4	5	6	7	8
*49:20	1				5	6	7	
*49:22	1		3		5	6		
*49:26		2			5	6		
*07:78, 13:31, 13:41, 54:16, 54:20, 55:09, 55:21, 55:46, 55:52, 55:56, 56:01:05, 56:13					5			8
*07:84, 08:09, 08:84, 13:35, 13:59, 13:62, 15:83, 27:14, 27:81, 40:06:01:01-40:06:07, 40:44, 40:53, 40:70, 40:75, 40:83, 40:93, 40:95, 40:103, 40:127, 40:131, 40:148, 40:161-40:162, 40:165, 40:177, 40:190, 40:230, 41:01, 41:05-41:07, 41:09, 41:12, 41:14, 41:16-41:17, 41:20-41:22, 41:25-41:26, 42:04, 50:20, 51:01:01-51:01:18, 51:01:20- 51:03, 51:05, 51:07:02, 51:09:01- 51:12, 51:14, 51:16, 51:18-51:19, 51:21-51:22, 51:24:01-51:24:04, 51:26-51:35, 51:38-51:41N, 51:43, 51:48-51:55, 51:57-51:58, 51:60- 51:61:02, 51:65-51:80, 51:82-51:91, 51:93, 51:95-51:96, 51:98N-51:105, 51:107, 51:109-51:111, 51:113- 51:125, 51:127-51:129, 51:131- 51:134, 51:136-51:138, 51:140, 51:142-51:147, 51:149N-51:152, 52:01:01:01-52:01:01:02, 52:01:03, 52:01:05-52:01:08, 52:01:10- 52:01:17, 52:04-52:08, 52:10:01- 52:13, 52:16-52:18, 52:20, 52:22- 52:24, 52:26-52:31, 54:04, 54:11, 55:17, 55:20, 55:28, 56:05:01-56:06, 56:15, 56:21, 58:08:01-58:08:02, 59:02, 78:01:01-78:07								8
*08:03, 08:52, 08:78, 37:34, 38:30, 44:06, 53:22, 57:09, 57:24								
Well No.	1	2	3	4	5	6	7	8

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155	80	505	135	150	155	160	95	Length of spec.
240	250				190	190	135	PCR product(s)
						290		
9	10	11	12	13	14	15	16	Well No.
								*49:20
								*49:22
								*49:26
								*07:78, 13:31, 13:41, 54:16, 54:20, 55:09, 55:21, 55:46, 55:52, 55:56, 56:01:05, 56:13
								*07:84, 08:09, 08:84, 13:35, 13:59, 13:62, 15:83, 27:14, 27:81, 40:06:01:01-40:06:07, 40:44, 40:53, 40:70, 40:75, 40:83, 40:93, 40:95, 40:103, 40:127, 40:131, 40:148, 40:161-40:162, 40:165, 40:177, 40:190, 40:230, 41:01, 41:05-41:07, 41:09, 41:12, 41:14, 41:16-41:17, 41:20-41:22, 41:25-41:26, 42:04, 50:20, 51:01:01-51:01:18, 51:01:20- 51:03, 51:05, 51:07:02, 51:09:01- 51:12, 51:14, 51:16, 51:18-51:19, 51:21-51:22, 51:24:01-51:24:04, 51:26-51:35, 51:38-51:41N, 51:43, 51:48-51:55, 51:57-51:58, 51:60- 51:61:02, 51:65-51:80, 51:82-51:91, 51:93, 51:95-51:96, 51:98N-51:105, 51:107, 51:109-51:111, 51:113- 51:125, 51:127-51:129, 51:131- 51:134, 51:136-51:138, 51:140, 51:142-51:147, 51:149N-51:152, 52:01:01:01-52:01:01:02, 52:01:03, 52:01:05-52:01:08, 52:01:10- 52:01:17, 52:04-52:08, 52:10:01- 52:13, 52:16-52:18, 52:20, 52:22- 52:24, 52:26-52:31, 54:04, 54:11, 55:17, 55:20, 55:28, 56:05:01-56:06, 56:15, 56:21, 58:08:01-58:08:02, 59:02, 78:01:01-78:07
		11						*08:03, 08:52, 08:78, 37:34, 38:30, 44:06, 53:22, 57:09, 57:24
9	10	11	12	13	14	15	16	Well No.

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Length of spec.	145	145	115	170	105	160	105	70
PCR product(s)		190	145	205			255	155
		255						
Well No.	1	2	3	4	5	6	7	8
*13:01:01-13:01:06, 13:06-13:07N, 13:11-13:12:02, 13:17, 13:22:01-13:22:02, 13:25, 13:28-13:29, 13:36, 13:39, 13:43, 13:50, 13:52, 13:57, 13:60-13:61, 13:63N-13:64, 15:86, 15:224, 40:48, 40:71, 56:02, 56:04, 56:10						6		
*13:02:01-13:02:05, 13:02:07-13:02:14, 13:03, 13:08Q-13:09, 13:14, 13:19, 13:27, 13:30, 13:32-13:34, 13:37-13:38, 13:40, 13:42, 13:44-13:45, 13:49N, 13:53-13:54, 13:56N, 13:58, 13:65-13:67, 35:60, 46:11, 46:18, 51:15, 54:01:01-54:03, 54:05N, 54:07-54:08N, 54:12-54:13, 54:17-54:19, 54:21-54:28, 55:02:01-55:02:06, 55:07, 55:10, 55:12, 55:16, 55:18-55:19, 55:22, 55:26, 55:30, 55:34-55:35, 55:41, 55:43, 55:47-55:48, 55:50, 56:01:01-56:01:04, 56:07-56:08, 56:14, 56:16-56:17, 56:19N-56:20:02, 56:23-56:24, 56:26-56:29, 56:33, 56:35-56:36, 56:38N-56:39, 59:01:01:01-59:01:01:02, 59:04-59:05					5	6		8
*13:02:06					5	6		
*13:02:15, 13:47, 55:39, 55:42						6		8
*13:04, 13:46, 15:04, 15:16:01-15:16:02, 15:67, 15:95, 15:137, 15:155, 15:222, 15:254, 39:06:01-39:06:02, 39:33-39:34, 39:50, 39:57, 39:62, 39:64, 40:86, 40:159, 46:32, 51:13:01-51:13:02, 51:23, 51:37, 51:63, 51:92, 51:108, 52:14-52:15, 54:14, 55:13, 55:23, 55:32, 55:58, 56:22, 59:03, 73:01-73:02							7	8
*13:15, 56:34				4	5	6		8
Well No.	1	2	3	4	5	6	7	8

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155	80	505	135	150	155	160	95	Length of spec.
240	250				190	190	135	PCR product(s)
						290		
9	10	11	12	13	14	15	16	Well No.
				13				*13:01:01-13:01:06, 13:06-13:07N, 13:11-13:12:02, 13:17, 13:22:01- 13:22:02, 13:25, 13:28-13:29, 13:36, 13:39, 13:43, 13:50, 13:52, 13:57, 13:60-13:61, 13:63N-13:64, 15:86, 15:224, 40:48, 40:71, 56:02, 56:04, 56:10
				13				*13:02:01-13:02:05, 13:02:07- 13:02:14, 13:03, 13:08Q-13:09, 13:14, 13:19, 13:27, 13:30, 13:32- 13:34, 13:37-13:38, 13:40, 13:42, 13:44-13:45, 13:49N, 13:53-13:54, 13:56N, 13:58, 13:65-13:67, 35:60, 46:11, 46:18, 51:15, 54:01:01-54:03, 54:05N, 54:07-54:08N, 54:12-54:13, 54:17-54:19, 54:21-54:28, 55:02:01- 55:02:06, 55:07, 55:10, 55:12, 55:16, 55:18-55:19, 55:22, 55:26, 55:30, 55:34-55:35, 55:41, 55:43, 55:47- 55:48, 55:50, 56:01:01-56:01:04, 56:07-56:08, 56:14, 56:16-56:17, 56:19N-56:20:02, 56:23-56:24, 56:26- 56:29, 56:33, 56:35-56:36, 56:38N- 56:39, 59:01:01:01-59:01:01:02, 59:04-59:05
				13				*13:02:06
				13				*13:02:15, 13:47, 55:39, 55:42
								*13:04, 13:46, 15:04, 15:16:01- 15:16:02, 15:67, 15:95, 15:137, 15:155, 15:222, 15:254, 39:06:01- 39:06:02, 39:33-39:34, 39:50, 39:57, 39:62, 39:64, 40:86, 40:159, 46:32, 51:13:01-51:13:02, 51:23, 51:37, 51:63, 51:92, 51:108, 52:14-52:15, 54:14, 55:13, 55:23, 55:32, 55:58, 56:22, 59:03, 73:01-73:02
				13				*13:15, 56:34
9	10	11	12	13	14	15	16	Well No.

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Length of spec.	145	145	115	170	105	160	105	70
PCR product(s)		190	145	205			255	155
		255						
Well No.	1	2	3	4	5	6	7	8
*13:16, 13:48, 15:42, 55:01:01-55:01:03, 55:01:05-55:01:06, 55:01:08-55:01:10, 55:03, 55:05, 55:11, 55:15, 55:24-55:25, 55:29, 55:31, 55:33, 55:36, 55:38, 55:40, 55:45, 55:54-55:55N, 55:57, 56:25, 56:30, 56:37					5	6		8
*13:18, 54:10, 55:37					5			8
*13:20, 39:17, 50:12, 50:16-50:17, 55:01:07						6		
*13:23		2				6		
*13:55		2			5	6		8
*15:16:03, 35:37, 45:02, 45:09, 50:09							7	
*15:33, 15:248, 44:03:16, 44:07, 44:49, 44:81, 44:111, 44:145, 57:54, C*03:87, C*05:27, C*05:39			3					
*15:46, 15:53, 15:106, 35:47, 35:154, 40:10:01-40:10:02, 40:40, 40:49, 40:129, 40:164, 40:210, 40:219, 40:229, 44:02:13, 44:02:19, 44:05:03, 44:09, 44:11, 44:46, 44:75, 44:90, 44:129, 44:152, 44:170, 47:01:01:01-47:03, 47:05-47:08								
*18:04, 40:01:11				4				
*18:09, 27:01, 37:10, 39:31, 58:13		2						
*35:01:10, 35:04:02, 40:28, 50:14, 51:56:01-51:56:02, 53:28, C*02:02:23								
*40:13, 40:19, 40:117, 57:51	1							
*40:47, 40:157, 40:201, 44:55, 44:103		2	3					
*40:58				4				
*40:96		2	3					8
*40:109, 51:112	1							8
*40:110			3					8
Well No.	1	2	3	4	5	6	7	8

101.547-06 – including *Taq* pol., IFU-01
 101.547-06u – without *Taq* pol., IFU-02

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 “Instructions for Use” (IFU)

Lot No.: 56S				Lot-specific information				www.olerup-ssp.com
155	80	505	135	150	155	160	95	Length of spec.
240	250				190	190	135	PCR product(s)
						290		
9	10	11	12	13	14	15	16	Well No.
								*13:16, 13:48, 15:42, 55:01:01-55:01:03, 55:01:05-55:01:06, 55:01:08-55:01:10, 55:03, 55:05, 55:11, 55:15, 55:24-55:25, 55:29, 55:31, 55:33, 55:36, 55:38, 55:40, 55:45, 55:54-55:55N, 55:57, 56:25, 56:30, 56:37
				w				*13:18, 54:10, 55:37
								*13:20, 39:17, 50:12, 50:16-50:17, 55:01:07
				13				*13:23
				13				*13:55
								*15:16:03, 35:37, 45:02, 45:09, 50:09
								*15:33, 15:248, 44:03:16, 44:07, 44:49, 44:81, 44:111, 44:145, 57:54, C*03:87, C*05:27, C*05:39
			12					*15:46, 15:53, 15:106, 35:47, 35:154, 40:10:01-40:10:02, 40:40, 40:49, 40:129, 40:164, 40:210, 40:219, 40:229, 44:02:13, 44:02:19, 44:05:03, 44:09, 44:11, 44:46, 44:75, 44:90, 44:129, 44:152, 44:170, 47:01:01:01-47:03, 47:05-47:08
								*18:04, 40:01:11
								*18:09, 27:01, 37:10, 39:31, 58:13
							16	*35:01:10, 35:04:02, 40:28, 50:14, 51:56:01-51:56:02, 53:28, C*02:02:23
								*40:13, 40:19, 40:117, 57:51
								*40:47, 40:157, 40:201, 44:55, 44:103
			12					*40:58
								*40:96
								*40:109, 51:112
			12					*40:110
9	10	11	12	13	14	15	16	Well No.

101.547-06 – including *Taq* pol., IFU-01101.547-06u – without *Taq* pol., IFU-02Visit www.olerup-ssp.com for
“Instructions for Use” (IFU)Lot No.: **56S**

Lot-specific information

www.olerup-ssp.com

Length of spec.	145	145	115	170	105	160	105	70
PCR product(s)		190	145	205			255	155
		255						
Well No.	1	2	3	4	5	6	7	8
*44:02:01:01-44:02:09, 44:02:11-44:02:12, 44:02:14-44:02:18, 44:02:20-44:03:15, 44:03:17-44:05:02, 44:05:04, 44:12-44:14, 44:16-44:17, 44:19N, 44:21-44:24, 44:26-44:33, 44:34:02-44:39, 44:41:01-44:43:02, 44:45, 44:48, 44:51-44:54, 44:56N, 44:58N-44:59:02, 44:61N-44:74, 44:76-44:80, 44:82, 44:84-44:89, 44:91-44:94, 44:96-44:99, 44:101-44:102, 44:104-44:110, 44:112-44:128, 44:132-44:133, 44:135-44:144, 44:146-44:151, 44:153-44:155, 44:157-44:169, 44:171N-44:175, 47:04			3					
*44:02:10, 44:34:01			3	4				
*44:10			3			6		
*44:15		2	3		5		7	
*44:18	1				5		7	
*44:20, 44:47, 44:100			3				7	8
*44:25, 44:95	1							
*44:50, 53:30, 57:45	1							
*45:01, 45:03, 45:05-45:07, 45:11-45:13					5		7	
*45:04, 45:08					5			
*45:10					5			
*45:14					5		7	8
*50:01:01-50:01:03, 50:04-50:08, 50:10-50:11, 50:15, 50:18-50:19, 50:32					5	6		
*50:02					5	6	7	
*50:13					5	6		
*50:31					5	6		
*51:07:01, 52:01:02, 52:01:04, 52:01:09, 52:02-52:03, 52:09, 52:21				4				8
*51:08, 51:20, 51:36, 51:44N, 51:141, 52:19								8
*51:62				4		6		
Well No.	1	2	3	4	5	6	7	8

101.547-06 – including *Taq* pol., IFU-01
 101.547-06u – without *Taq* pol., IFU-02

Visit www.olerup-ssp.com for
 “Instructions for Use” (IFU)

Lot No.: **56S**

Lot-specific information

www.olerup-ssp.com

155	80	505	135	150	155	160	95	Length of spec.
240	250				190	190	135	PCR product(s)
						290		
9	10	11	12	13	14	15	16	Well No.
			12					*44:02:01:01-44:02:09, 44:02:11-44:02:12, 44:02:14-44:02:18, 44:02:20-44:03:15, 44:03:17-44:05:02, 44:05:04, 44:12-44:14, 44:16-44:17, 44:19N, 44:21-44:24, 44:26-44:33, 44:34:02-44:39, 44:41:01-44:43:02, 44:45, 44:48, 44:51-44:54, 44:56N, 44:58N-44:59:02, 44:61N-44:74, 44:76-44:80, 44:82, 44:84-44:89, 44:91-44:94, 44:96-44:99, 44:101-44:102, 44:104-44:110, 44:112-44:128, 44:132-44:133, 44:135-44:144, 44:146-44:151, 44:153-44:155, 44:157-44:169, 44:171N-44:175, 47:04
			12					*44:02:10, 44:34:01
			12	13				*44:10
								*44:15
		11						*44:18
			12					*44:20, 44:47, 44:100
		11	12					*44:25, 44:95
			12					*44:50, 53:30, 57:45
								*45:01, 45:03, 45:05-45:07, 45:11-45:13
								*45:04, 45:08
9								*45:10
								*45:14
								*50:01:01-50:01:03, 50:04-50:08, 50:10-50:11, 50:15, 50:18-50:19, 50:32
								*50:02
			12					*50:13
9								*50:31
								*51:07:01, 52:01:02, 52:01:04, 52:01:09, 52:02-52:03, 52:09, 52:21
		11						*51:08, 51:20, 51:36, 51:44N, 51:141, 52:19
								*51:62
9	10	11	12	13	14	15	16	Well No.

101.547-06 – including *Taq* pol., IFU-01
 101.547-06u – without *Taq* pol., IFU-02

Visit www.olerup-ssp.com for
 “Instructions for Use” (IFU)

Lot No.: **56S**

Lot-specific information

www.olerup-ssp.com

Length of spec.	145	145	115	170	105	160	105	70
PCR product(s)		190	145	205			255	155
		255						
Well No.	1	2	3	4	5	6	7	8
*51:97							7	8
*51:106				4		6		8
*51:126			3					8
*51:130								8
*52:25				4	5	6		8
*55:01:04					5	6		8
*55:44, 55:53						6		8
HLA-B allele								
Well No.	1	2	3	4	5	6	7	8

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*49 SSP subtyping.

In addition, wells number 3, 9 and 10 contains 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 the 2nd or 3rd exon, 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.

⁴HLA-B*49 alleles in bold lettering are listed as confirmed alleles on the on the IMGT/HLA web page www.ebi.ac.uk/imgt/hla, release 3.12.0, April 2013.

⁵The B*49:07 and 49:21 alleles can be distinguished by the different sizes of the specific PCR products generated by primer mix 9.

⁶The B*49:08 and 49:16 alleles can be distinguished by the different sizes of the specific PCR products generated by primer mix 10.

⁷The B*49:13 and 49:14 alleles can be distinguished by the different sizes of the specific PCR products generated by primer mix 15.

⁸Primer mix 2: Specific PCR fragment of 145 bp in the B*49:06 and the B*58:13 alleles. Specific PCR fragment of 190 bp in the B*49:26 and the B*39:31 alleles. Specific PCR fragment of 255 bp in the B*49:02 and the B*13:23, 13:55, 18:09, 27:01, 37:10, 40:47, 40:96, 40:157, 40:201, 44:15, 44:55 and 44:103 alleles.

Primer mix 3: Specific PCR fragment of 115 bp in the B*49:22 and the B*15:33, 15:248, 51:126 and 57:54 and in the C*03:87, C*05:27 and C*05:39 alleles. Specific PCR fragment of 145 bp in the B*49:02 and the B*40:47, 40:96, 40:110, 40:157, 40:201, 44:02:01:01-44:02:12, 44:02:14-44:02:18, 44:02:20-44:05:02, 44:05:04, 44:07, 44:10, 44:12-44:17, 44:19N-44:24, 44:26-44:39, 44:41:01-44:43:02, 44:45, 44:47-44:49, 44:51-44:56N, 44:58N-44:59:02, 44:61N-44:74, 44:76-44:82, 44:84-44:89, 44:91-44:94, 44:96-44:128, 44:132-44:133, 44:135-44:151, 44:153-44:155, 44:157-44:169, 44:171N-44:175 and 47:04 alleles.

Primer mix 4: Specific PCR fragment of 170 bp in the B*49:03 and the B*18:04, 40:01:11, 40:58, 44:02:10, 44:34:01, 51:07:01, 52:01:02, 52:01:04, 52:01:09, 52:02-52:03, 52:09, 52:21 alleles. Specific PCR fragment of 205 bp in the B*49:09 and the B*13:15, 51:62, 51:106, 52:25 and 56:34 alleles.

101.547-06 – including *Taq* pol., IFU-01
101.547-06u – without *Taq* pol., IFU-02

Visit www.olerup-ssp.com for
“Instructions for Use” (IFU)

Lot No.: 56S				Lot-specific information				www.olerup-ssp.com
155	80	505	135	150	155	160	95	Length of spec.
240	250				190	190	135	PCR product(s)
						290		
9	10	11	12	13	14	15	16	Well No.
		11						*51:97
								*51:106
								*51:126
					14			*51:130
								*52:25
							16	*55:01:04
								*55:44, 55:53
9	10	11	12	13	14	15	16	HLA-B allele
								Well No.

Primer mix 7: Specific PCR fragment of 105 bp in the B*49:04 and 49:05 and the B*13:04, 13:46, 15:04, 15:16:01-15:16:03, 15:67, 15:95, 15:137, 15:155, 15:222, 35:37, 39:06:01-39:06:02, 39:33-39:34, 39:50, 39:57, 39:62, 39:64, 40:86, 40:159, 45:02, 45:09, 50:09, 51:13:01-51:13:02, 51:37, 51:63, 51:92, 51:97, 52:14, 54:14, 55:13, 55:23, 55:32, 55:58, 56:22, 59:03 and 73:01-73:02 alleles. Specific PCR fragment of 255 bp in the B*49:20 and the B*44:15, 44:18, 44:20, 44:47, 44:100, 45:01-45:03, 45:05-45:07, 45:09, 45:11-45:14, 50:02, 51:23, 51:108 and 52:15 alleles.

Primer mix 8: Specific PCR fragment of 70 bp in the B*49:05 and the B*07:78, 07:84, 08:09, 08:84, 13:02:01-13:02:05, 13:02:07-13:04, 13:08Q-13:09, 13:14-13:16, 13:18-13:19, 13:27, 13:30-13:35, 13:37-13:38, 13:40-13:42, 13:44-13:49N, 13:53-13:56N, 13:58-13:59, 13:62, 13:65-13:67, 15:04, 15:16:01-15:16:02, 15:42, 15:67, 15:83, 15:95, 15:137, 15:155, 15:222, 15:254, 27:14, 27:81, 35:60, 39:06:01-39:06:02, 39:33-39:34, 39:50, 39:57, 39:62, 39:64, 40:06:01:01-40:06:07, 40:44, 40:53, 40:70, 40:75, 40:83, 40:86, 40:93, 40:95-40:96, 40:103, 40:109-40:110, 40:127, 40:131, 40:148, 40:159, 40:161-40:162, 40:165, 40:177, 40:190, 40:230, 41:01, 41:05-41:07, 41:09, 41:12, 41:14, 41:16-41:17, 41:20-41:22, 41:25-41:26, 42:04, 44:20, 44:47, 44:100, 45:14, 46:11, 46:18, 46:32, 50:20, 51:01:01-51:01:18, 51:01:20-51:03, 51:05, 51:07:01-51:16, 51:18-51:24:04, 51:26-51:41N, 51:43-51:44N, 51:48-51:55, 51:57-51:58, 51:60-51:61:02, 51:63, 51:65-51:80, 51:82-51:93, 51:95-51:134, 51:136-51:138, 51:140-51:147, 51:149N-51:152, 52:01:01:01-52:31, 54:01:01-54:05N, 54:07-54:08N, 54:10-54:14, 54:16-54:28, 55:01:01-55:01:06, 55:01:08-55:03, 55:05, 55:07, 55:09-55:13, 55:15-55:26, 55:28-55:48, 55:50, 55:52-55:58, 56:01:01-56:01:05, 56:05:01-56:08, 56:13-56:17, 56:19N-56:30, 56:33-56:39, 58:08:01-58:08:02, 59:01:01:01-59:05, 73:01-73:02 and 78:01:01-78:07 alleles. Specific PCR fragment of 155 bp in the B*49:19N allele.

Primer mix 9: Specific PCR fragment of 155 bp in the B*49:21 allele. Specific PCR fragment of 240 bp in the B*49:07 and the B*45:10 and 50:31 alleles.

Primer mix 10: Specific PCR fragment of 80 bp in the B*49:16 allele. Specific PCR fragment of 250 bp in the B*49:08 allele.

Primer mix 14: Specific PCR fragment of 155 bp in the B*49:19N allele. Specific PCR fragment of 190 bp in the B*49:12 and 49:17 and the B*51:130 alleles.

Primer mix 15: Specific PCR fragment of 160 bp in the B*49:13 allele. Specific PCR fragment of 190 bp in the B*49:17 allele. Specific PCR fragment of 290 bp in the B*49:14 allele.

Primer mix 16: Specific PCR fragment of 95 bp in the B*49:18 and the B*35:01:10, 35:04:02, 40:28, 50:14, 51:56:01-51:56:02, 53:28 and 55:01:04 and in the C*02:02:23 alleles. Specific PCR fragment of 135 bp in the B*49:15 allele.

‘w’, may be weakly amplified.

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CELL LINE VALIDATION SHEET																					
HLA-B*49 SSP primer set																					
					Well																
						1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
				Prod. No.:		201191101	201321002	201207703	201191104	201191105	201191106	201191107	201191108	201207709	201191110	201191111	201191112	201191113	201191114	201191115	201191116
	IHCW 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	9025	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	-	-	+	-	-	-	-	-	-	-	-	+	-	-	-	-

101.547-06 – including *Taq* pol., IFU-01
101.547-06u – without *Taq* pol., IFU-02

Visit www.olerup-ssp.com for
“Instructions for Use” (IFU)

Lot No.: **56S**

Lot-specific information

www.olerup-ssp.com

CERTIFICATE OF ANALYSIS

Olerup SSP® HLA-B*49 SSP

Product number: 101.547-06 – including *Taq* polymerase
101.547-06u – without *Taq* polymerase
Lot number: 56S
Expiry date: 2015-January-01
Number of tests: 6
Number of wells per test: 16

Well specifications:

Well No.	Production No.	Well No.	Production No.
1	2011-911-01	9	2012-077-09
2	2013-210-02	10	2011-911-10
3	2012-077-03	11	2011-911-11
4	2011-911-04	12	2011-911-12
5	2011-911-05	13	2011-911-13
6	2011-911-06	14	2011-911-14
7	2011-911-07	15	2011-911-15
8	2011-911-08	16	2011-911-16

The specificity of each primer solution of the HLA-B*49 primer set has been tested against 48 well characterized IHWC cell line DNAs.

No DNAs carrying the alleles to be amplified by primer solutions 2, 4, 9 to 11 and 14 to 16 were available. The specificities of the primers in primer solutions 2, 4, 9, 11 and 16 were tested by separately adding additional 3'-primers, respectively additional 5'-primers. In primer solution 10, 14 and 15, it was only possible to test the 5'-primer, the 3'-primer was not possible to test. In primer solutions 2 and 16, one 5'-primer was not possible to test, and in primer solutions 8 and 9, one 3'-primer was not possible to test. One additional 5'-primer and one additional 3'-primer in primer solution 3 were tested by separately adding one 3'-primer respectively one 3'-primer.

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

Date of approval: 2013-August-19

Approved by:

Head of R & D

101.547-06 – including *Taq* pol., IFU-01
101.547-06u – without *Taq* pol., IFU-02

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Lot No.: **56S**

Lot-specific information

www.olerup-ssp.com

Declaration of Conformity

Product name: Olerup SSP® HLA-B*49
Product number: 101.547-06/06u
Lot number: 56S

Intended use: HLA-B*49 high resolution histocompatibility testing

Manufacturer: Olerup SSP AB
Franzengatan 5
SE-112 51 Stockholm, 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 13485:2012, 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, Franzengatan 5, SE-112 51 Stockholm, Sweden.

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

Stockholm, Sweden
2013-August-19

Ann-Cathrin Jareman
Head of QA and Regulatory Affairs

101.547-06 – including *Taq* pol., IFU-01
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Lot No.: **56S**

Lot-specific information

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101.547-06 – including *Taq* pol., IFU-01
101.547-06u – without *Taq* pol., IFU-02

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Lot No.: **56S**

Lot-specific information

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ADDRESSES:

Manufacturer:

Olerup SSP AB, Franzengatan 5, SE-112 51 Stockholm, Sweden.

Tel: +46-8-717 88 27

Fax: +46-8-717 88 18

E-mail: info-ssp@olerup.com

Web page: <http://www.olerup-ssp.com>

Distributed by:

Olerup GmbH, Löwengasse 47 / 6, AT-1030 Vienna, Austria.

Tel: +43-1-710 15 00

Fax: +43-1-710 15 00 10

E-mail: support-at@olerup.com

Web page: <http://www.olerup.com>

Olerup Inc., 901 S. Bolmar St., Suite R, West Chester, PA 19382

Tel: 1-877-OLERUP1

Fax: 610-344-7989

E-mail: info.us@olerup.com

Web page: <http://www.olerup.com>

For information on *Olerup* SSP distributors worldwide, contact **Olerup GmbH**.