

Olerup SSP[®] HLA-C*16

Product number:	101.627-12u – without <i>Taq</i> polymerase
Lot number:	18M
Expiry date:	2013-December-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. 18M.

CHANGES COMPARED TO THE PREVIOUS OLERUP SSP[®] HLA-C*16 LOT

The HLA-C*16 specificity and interpretation tables have been updated for the HLA-C alleles described since the previous *Olerup SSP[®]* HLA-C*16 lot was made (Lot No. 21K).

Eight wells has been added to the C*16 kit, wells **16 to 23**.
The amplification patterns for some rare HLA-C*16 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
1	Added	-	Primer added for the C*16:01:05 allele.
12	-	Added	Primer added for the C*16:22 allele.
16	New	New	New primer pair for the C*16:17:02 allele.
17	New	New	New primer pair for the C*16:26 allele.
18	New	New	New primer pair for the C*16:26 allele.
19	New	New	New primer pairs for the C*16:28, C*16:29 and C*16:31 alleles.
20	New	New	New primer pairs for the C*16:27 and C*16:32 alleles.
21	New	New	New primer pair for the C*16:23 allele.
22	New	New	New primer pair for the C*16:24 allele.
23	New	New	New primer pair for the C*16:30N allele.

PRODUCT DESCRIPTION

HLA-C*16 SSP typing

CONTENT

The primer set contains 5'- and 3'-primers for identifying the C*16:01 to C*16:32 alleles.

PLATE LAYOUT

Each HLA-C*16 test consists of 23 PCR reactions in a 24 well cut 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 16 well PCR plate is marked with ‘HLA-C*16’ in silver/gray ink.

Well No. 1 is marked with the Lot No. ‘18M’.

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-C*16 SSP subtypings will be influenced by many other HLA-C alleles, when present on the other haplotype. In addition, primer mix 4 will amplify the B*35:08:02 and B*67:02 alleles, primer mixes 5 and 7 will amplify the B*46:25 allele, primer mix 6 will amplify the A*24:106 allele and primer mix 11 will amplify the A*24:73, A*24:157, B*07:66, B*46:25 and B*51:55 alleles.

UNIQUELY IDENTIFIED ALLELES

All the HLA-C*16 alleles, i.e. **C*16:01 to C*16:32**, recognized by the HLA Nomenclature Committee in January 2011¹ will be amplified by the primers in the HLA-C*16 SSP kit.

The HLA-C*16 primer set cannot distinguish the C*16:01:01 and C*16:01:03-16:01:06 alleles, the C*16:02:01-16:02:06 alleles or the C*16:15:01-16:15:02 alleles.

The C*16:15:01-16:15:02 and C*16:20 alleles may be distinguished by the different sizes of the specific PCR products generated by primer mix 11.

The C*16:16Q and C*16:17 alleles may be distinguished by the different sizes of the specific PCR products generated by primer mix 12.

The C*16:27 and C*16:32 alleles may be distinguished by the different sizes of the specific PCR products generated by primer mix 20.

Lot No.: **18M**

Lot-specific information

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The C*16:28 and C*16:31 alleles may be distinguished by the different sizes of the specific PCR products generated by primer mix 19.

¹HLA-C alleles listed on the IMGT/HLA web page 2011-January-14, 3.3.0, www.ebi.ac.uk/imgt/hla.

RESOLUTION IN HOMO- AND HETEROZYGOTES

A total of 47 alleles generate 28 amplification patterns that can be combined in 406 homozygous and heterozygous combinations. 175 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.

++-++++-	-----	-----	*16:09, *16:10 = *16:09, *16:11
++-++++-	+-----	-----	*16:10, *16:12 = *16:11, *16:12
++-++++-	------	-----	*16:10, *16:19 = *16:11, *16:19
++-++++-	---+-----	-----	*16:10, *16:25 = *16:11, *16:25
++-++++-	-----	-----	*16:02:01, *16:10 = *16:02:01, *16:11
++-++++-	---+-----	-----	*16:09, *16:16Q = *16:09, *16:22
++-++++-	-----+	-----	*16:02:01, *16:07:01 = *16:07:01, *16:09 = *16:07:02, *16:09
++-++++-	+-----	-----	*16:12, *16:16Q = *16:12, *16:22
++-++++-	-++-----	-----	*16:13, *16:25 = *16:15:01, *16:19
++-++++-	-++-----	-----	*16:16Q, *16:19 = *16:19, *16:22
++-++++-	-+-----	-----	*16:01:01, *16:19 = *16:02:01, *16:13 = *16:13, *16:19
++-++++-	---+-----	-----	*16:16Q, *16:25 = *16:22, *16:25
++-++++-	---+-----	-----	*16:01:01, *16:25 = *16:02:01, *16:15:01 = *16:15:01, *16:25
++-++++-	---+-----	-----	*16:02:01, *16:16Q = *16:02:01, *16:22
++-++-+-	-----	-----	*16:01:02, *16:09 = *16:02:01, *16:09 = *16:09, *16:09
++-++-+-	+-----	-----	*16:01:02, *16:12 = *16:02:01, *16:12 = *16:12, *16:12
++-++-+-	-+-----	-----	*16:01:02, *16:19 = *16:02:01, *16:19 = *16:19, *16:19
++-++-+-	-++-----	-----	*16:01:02, *16:25 = *16:02:01, *16:25 = *16:25, *16:25
++-++-+-	-----	-----	*16:01:02, *16:02:01 = *16:02:01, *16:02:01
+-----	---+-----	---+-----	*16:16Q, *16:29 = *16:22, *16:29
+-----	---+-----	-----	*16:04:01, *16:16Q = *16:04:01, *16:22
+-----	-----	---+-----	*16:01:01, *16:29 = *16:01:02, *16:29 = *16:04:01, *16:28 = *16:28, *16:29
+-----	-----	-----	*16:01:01, *16:04:01 = *16:01:02, *16:04:01
+-----	-----	---+-----	*16:04:01, *16:29 = *16:29, *16:29
+-----	-----+	-----	*16:07:01, *16:10 = *16:07:01, *16:11
+-----	-----+	-----	*16:08, *16:10 = *16:08, *16:11
+-----	-+-----	-----	*16:10, *16:13 = *16:11, *16:13
+-----	-++-----	-----	*16:10, *16:15:01 = *16:11, *16:15:01
+-----	---+-----	-----	*16:10, *16:16Q = *16:10, *16:22 = *16:11, *16:16Q = *16:11, *16:22
+-----	---+-----	-----	*16:10, *16:14 = *16:11, *16:14
+-----	---+-----	-----	*16:10, *16:18 = *16:11, *16:18
+-----	---+-----	-----	*16:10, *16:21 = *16:11, *16:21
+-----	---+-----+	-----	*16:01:01, *16:06 = *16:01:02, *16:06 = *16:06, *16:10 = *16:06, *16:11 = *16:07:02, *16:10 = *16:07:02, *16:11
+-----	-----	++-----	*16:10, *16:26 = *16:11, *16:26
+-----	-----	---+-----	*16:10, *16:28 = *16:11, *16:28
+-----	-----	---+-----	*16:10, *16:27 = *16:11, *16:27
+-----	-----	-----+	*16:10, *16:23 = *16:11, *16:23
+-----	-----	-----+	*16:10, *16:24 = *16:11, *16:24
+-----	-----	-----+	*16:10, *16:30N = *16:11, *16:30N
+-----	-----	-----	*16:01:01, *16:10 = *16:01:01, *16:11 = *16:01:02, *16:10 = *16:01:02, *16:11 = *16:10, *16:11 = *16:11, *16:11
+-----	-----+	-----	*16:01:01, *16:07:01 = *16:01:02, *16:07:01
+-----	---+-----	-----	*16:08, *16:16Q = *16:08, *16:22

Lot No.: **18M**

Lot-specific information

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+---+---+	-----	-----	*16:01:01, *16:08 = *16:01:02, *16:08 = *16:08, *16:08
+---+---+	-+-+---	-----	*16:13, *16:16Q = *16:13, *16:22
+---+---+	-+-----	-----	*16:01:01, *16:13 = *16:01:02, *16:13 = *16:13, *16:13
+---+---+	---+---	-----	*16:15:01, *16:16Q = *16:15:01, *16:22
+---+---+	---+---	-----	*16:01:01, *16:15:01 = *16:01:02, *16:15:01 = *16:15:01, *16:15:01
+---+---+	---+---	-----	*16:14, *16:16Q = *16:14, *16:22
+---+---+	---+---	-----	*16:16Q, *16:18 = *16:18, *16:22
+---+---+	---+---	-----	*16:16Q, *16:21 = *16:21, *16:22
+---+---+	---+---	++-----	*16:16Q, *16:26 = *16:22, *16:26
+---+---+	---+---	---+---	*16:16Q, *16:28 = *16:22, *16:28
+---+---+	---+---	---+---	*16:16Q, *16:27 = *16:22, *16:27
+---+---+	---+---	-----+--	*16:16Q, *16:23 = *16:22, *16:23
+---+---+	---+---	-----+--	*16:16Q, *16:24 = *16:22, *16:24
+---+---+	---+---	-----+--	*16:16Q, *16:30N = *16:22, *16:30N
+---+---+	---+---	-----+--	*16:01:01, *16:16Q = *16:01:01, *16:22 = *16:01:02, *16:16Q = *16:01:02, *16:22 = *16:16Q, *16:16Q = *16:16Q, *16:22
+---+---+	---+---	-----+--	*16:01:01, *16:14 = *16:01:02, *16:14 = *16:14, *16:14
+---+---+	---+---	-----+--	*16:01:01, *16:18 = *16:01:02, *16:18 = *16:18, *16:18
+---+---+	---+---	-----+--	*16:01:01, *16:21 = *16:01:02, *16:21 = *16:21, *16:21
+---+---+	---+---	-----+--	*16:01:01, *16:07:02 = *16:01:02, *16:07:02
+---+---+	---+---	++-----	*16:01:01, *16:26 = *16:01:02, *16:26 = *16:26, *16:26
+---+---+	---+---	---+---	*16:01:01, *16:28 = *16:01:02, *16:28 = *16:28, *16:28
+---+---+	---+---	---+---	*16:01:01, *16:27 = *16:01:02, *16:27 = *16:27, *16:27
+---+---+	---+---	-----+--	*16:01:01, *16:23 = *16:01:02, *16:23 = *16:23, *16:23
+---+---+	---+---	-----+--	*16:01:01, *16:24 = *16:01:02, *16:24 = *16:24, *16:24
+---+---+	---+---	-----+--	*16:01:01, *16:30N = *16:01:02, *16:30N = *16:30N, *16:30N
+---+---+	-----	-----	*16:01:01, *16:01:01 = *16:01:01, *16:01:02
---+---+	-----+	-----	*16:06, *16:06 = *16:06, *16:07:02
---+---+	-----+	-----	*16:07:01, *16:07:01 = *16:07:01, *16:07:02

*16:01:01 = *16:01:01, 16:01:03-16:01:06
*16:02:01 = *16:02:01-16:02:06
*16:15:01 = *16:15:01-16:15:02 and 16:20
*16:16Q = *16:16Q and 16:17
*16:27 = *16:27 and 16:32
*16:28 = *16:28 and 16:31

SPECIFICITY TABLE

HLA-C*16 SSP subtyping

Specificities and sizes of the PCR products of the 23 primer mixes used for HLA-C*16 SSP subtyping

Primer Mix	Size of spec. PCR product ¹	Size of control band ²	Amplified HLA-C*16 alleles ³	Other amplified HLA Class I alleles ⁴
1	205 bp	800 bp	*16:01:01- 16:02:06, 16:04:01, 16:08- 16:21, 16:23-16:32	*06:31
2	340 bp	1070 bp	*16:02:01- 16:02:06, 16:09, 16:12, 16:19, 16:25	*01:14, 02:02:01-02:02:03, 02:02:05-02:02:11, 02:02:13-02:11, 02:13- 02:26:02, 02:28-02:40, 02:42-02:47, 03:07, 03:15, 03:45, 04:01:01:01- 04:01:26, 04:03-04:10, 04:12-04:20, 04:23-04:28, 04:30-04:35, 04:37-04:54, 04:56-04:84, 05:01:01:01- 05:01:15, 05:03-05:53, 06:02:01:01-06:02:01:02, 06:02:03-06:10, 06:12- 06:51, 06:53-06:58, 07:07, 07:09, 07:49, 07:76, 08:10, 12:04:01-12:05, 12:09, 12:21, 12:33, 12:41, 14:04, 14:12, 15:02:01-15:06:03, 15:08-15:13, 15:15-15:20, 15:22-15:24, 15:26-15:42, 15:44-15:48, 17:01:01:01- 17:08, 18:01-18:04
3⁶	220 bp	800 bp	*16:04:01, 16:29	*01:04, 01:09, 02:05, 02:17, 06:02:01:01- 06:02:01:02, 06:02:03- 06:03, 06:07-06:13, 06:15- 06:34, 06:36-06:39, 06:41- 06:58, 12:03:01:01-12:07, 12:11-12:13, 12:15, 12:23, 12:25-12:26, 12:28-12:29, 12:31-12:35, 12:37- 12:39N, 12:42Q-12:43, 12:45-12:48, 12:50-12:51, 14:16

Lot No.: **18M**

Lot-specific information

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4	140 bp	800 bp	*16:01:01, 16:01:03-16:01:06, 16:04:01, 16:06- 16:08, 16:10- 16:11, 16:13- 16:18, 16:20- 16:24, 16:26-16:32	*01:21, 02:12 ^W , 02:27:01- 02:27:02, 04:11, 04:29, 04:36, 04:55, 07:02:09, 08:01:01-08:09, 08:11- 08:46, 12:02:01-12:03:03, 12:03:05-12:03:08, 12:03:10-12:03:15, 12:06- 12:08, 12:10:01-12:20, 12:22-12:26, 12:28-12:32, 12:34-12:40, 12:42Q- 12:51, 14:02:03, 14:03, 14:08, 14:10, 14:22, 15:07, 15:21 ^W , 15:25, B*35:08:02 , B*67:02
5	160 bp	800 bp	*16:01:01- 16:02:06, 16:06- 16:09, 16:11- 16:28, 16:30N- 16:32	*07:53, B*46:25
6^{5,7,8}	130 bp, 160 bp, 210 bp	1070 bp	*16:06, 16:10- 16:11	*02:21, A*24:106
7^{5,9}	100 bp, 210 bp	1070 bp	*16:07:01, 16:09	*02:34, B*46:25
8⁵	135 bp	1070 bp	*16:08	
9⁵	85 bp	1070 bp	*16:12	*04:69
10¹⁰	215 bp, 350 bp	800 bp	*16:13, 16:19	*07:24, 07:114, 12:45
11¹¹	170 bp, 540 bp	1070 bp	*16:15:01- 16:15:02, 16:20, 16:25	*04:14, 04:68, 07:53, A*24:73, A*24:157 , B*07:66, B*46:25 , B*51:55
12^{5,7,12}	105 bp, 210 bp, 245 bp	1070 bp	*16:16Q-16:17, 16:22	*01:27
13⁵	130 bp	1070 bp	*16:14	*06:32, 12:40
14⁷	210 bp	1070 bp	*16:18	
15	145 bp	1070 bp	*16:21	*02:14, 04:42, 05:43, 06:05, 07:02:09, 08:37, 12:16, 15:23
16	375 bp	1070 bp	*16:06-16:07:02	*01:05, 01:21, 01:36, 02:02:01-02:02:03, 02:02:05-02:02:08, 02:02:10-02:04, 02:06- 02:16:02, 02:18-02:36, 02:38N-02:40,

Lot No.: **18M**

Lot-specific information

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				02:42-02:47, 03:05, 03:13, 03:25, 03:27, 03:35, 04:01:01:01-04:01:23, 04:01:25-04:01:26, 04:03-04:20, 04:23-04:36, 04:38-04:39, 04:41-04:79, 04:81-04:84, 05:01:01:01-05:01:15, 05:03-05:06, 05:08-05:09, 05:11-05:15, 05:17-05:30, 05:32-05:53, 07:01:01-07:01:10, 07:01:12-07:03, 07:05-07:09, 07:13-07:30, 07:32N-07:33N, 07:35-07:42, 07:44, 07:46-07:62, 07:64-07:100, 07:102-07:138, 07:140-07:141, 07:143-07:171, 08:01:01-08:11, 08:13-08:33, 08:35-08:43, 08:45-08:46, 12:02:01-12:02:06, 12:08, 12:10:01-12:10:02, 12:14:01-12:14:02, 12:16-12:18, 12:21-12:22, 12:27, 12:30, 12:36, 12:40-12:41, 12:49, 14:09, 15:22, 18:01-18:04
17	180 bp	1070 bp	*16:26	*04:01:01:01-04:01:09, 04:01:11-04:01:22, 04:01:24-04:01:26, 04:03-04:10, 04:12-04:20, 04:23-04:26, 04:28-04:32, 04:34-04:51, 04:53-04:54, 04:56-04:84, 05:25, 05:42, 06:05, 07:02:09, 08:28, 12:28, 15:25
18	255 bp	1070 bp	*16:26	*01:23, 04:03, 04:06, 04:80, 05:25, 05:42, 06:02:01:01-06:02:01:02, 06:02:03-06:02:09, 06:02:11-06:25, 06:27-06:29, 06:31-06:52, 06:54-06:58, 07:01:01-07:02:10, 07:02:12-07:02:23, 07:04:01-07:06, 07:08-07:15, 07:17-07:19, 07:21-07:33N, 07:35,

				07:37-07:50, 07:52-07:55N, 07:57-07:58, 07:61N-07:63, 07:65-07:78, 07:80-07:87, 07:89-07:95, 07:97-07:108, 07:110-07:126, 07:128-07:171, 08:28, 12:28, 15:25, 18:01-18:04
19 ^{5,13}	110 bp, 240 bp	800 bp	*16:28-16:29, 16:31	*01:10, 02:05, 02:17, 06:08, 06:22, 14:25
20 ^{5,14}	95 bp, 145 bp	1070 bp	*16:27, 16:32	
21	210 bp	1070 bp	*16:23	
22	210 bp	800 bp	*16:24	*03:108, 07:25
23 ⁷	170 bp	1070 bp	*16:30N	

¹ Alleles 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-C*16 high resolution 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.

² The 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-C*16 SSP subtyping.

In addition, wells number 3 to 5, 10, 19 and 22 contain the primer pair giving rise to the shorter, 800 bp, internal positive control band.

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

³ The C*16:15:01-16:15:02 and C*16:20 alleles may be distinguished by the different sizes of the specific PCR products generated by primer mix 11.

The C*16:16Q and C*16:17 alleles may be distinguished by the different sizes of the specific PCR products generated by primer mix 12.

The C*16:27 and C*16:32 alleles may be distinguished by the different sizes of the specific PCR products generated by primer mix 20.

The C*16:28 and C*16:31 alleles may be distinguished by the different sizes of the specific PCR products generated by primer mix 19.

⁴ Due to the sharing of sequence motifs between HLA-C alleles non-HLA-C*16 alleles will be amplified by primer mixes 1 to 7, 9 to 13, 15 to 19 and 22. In addition, primer mix 4 will amplify the B*35:08:02 and B*67:02 alleles, primer mixes 5 and 7 will amplify the B*46:25 allele, primer

Lot No.: **18M**

Lot-specific information

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mix 6 will amplify the A*24:106 allele and primer mix 11 will amplify the A*24:73, A*24:157, B*07:66, B*46:25 and B*51:55 alleles.

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

⁶Primer mix 3 may give a lower yield of specific PCR product than the other C*16 primer mixes.

⁷Primer mixes 6, 12, 14 and 23 may have tendencies of unspecific amplifications.

⁸Primer mix 6: Specific PCR fragment of 130 bp in the C*16:11 and in the C*02:21 allele. Specific PCR fragment of 160 bp in the C*16:10 and in the A*24:106 alleles. Specific PCR fragment of 210 bp in the C*16:06 allele.

⁹Primer mix 7: Specific PCR fragment of 100 bp in the C*16:09 and in the C*02:34 alleles. Specific PCR fragment of 210 bp in the C*16:07:01 and the B*46:25 alleles.

¹⁰Primer mix 10: Specific PCR fragment of 215 bp in the C*16:19 and the C*07:114 alleles. Specific PCR fragment of 350 bp in the C*16:13 and the C*07:24 and 12:45 alleles.

¹¹Primer mix 11: Specific PCR fragment of 170 bp in the C*16:20 and in the A*24:73, 24:157, B*07:66, B*46:25 and B*51:55 alleles. Specific PCR fragment of 540 bp in the C*16:15:01-16:15:02 and 16:25 and the C*04:14, 04:68 and 07:53 alleles.

¹²Primer mix 12: Specific PCR fragment of 105 bp in the C*16:17 and in the C*01:27 alleles. Specific PCR fragment of 210 bp in the C*16:22 allele. Specific PCR fragment of 245 bp in the C*16:16Q allele.

¹³Primer mix 19: Specific PCR fragment of 110 bp in the C*16:28 allele. Specific PCR fragment of 240 bp in the C*16:29 and 16:31 and the C*01:10, 02:05, 02:17, 06:08, 06:22 and 14:25 alleles.

¹⁴Primer mix 20: Specific PCR fragment of 95 bp in the C*16:27 allele. Specific PCR fragment of 145 bp in the C*16:32 allele.

‘w’ might be weakly amplified.

INTERPRETATION TABLE

HLA-C*16 SSP subtyping

Amplification patterns of the HLA-C*16:01 to 16:32 alleles

	Well ⁹											
	1	2	3	4	5	6	7	8	9	10	11	12
Length of spec. PCR product(s)	205	340	220	140	160	130	100	135	85	215	170	105
						160	210			350	540	210
						210						245
Length of int.	800	1070	800	800	800	1070	1070	1070	1070	800	1070	1070
pos. control ¹												
5'-primer(s) ²	360	1 st	361	201	419	113	244	126	256	385	289	361
	5'-CAG 3'	5'-CgA 3'	5'-AgT 3'	5'-CCA 3'	5'-gTC 3'	5'-CCA 3'	5'-CgC 3'	5'-ggA 3'	5'-ACg 3'	5'-ggT 3'	5'-Agg 3'	5'-AgT 3'
	361					368	369			523	409	
	5'-AgT 3'					5'-gTC 3'	5'-TAC 3'			5'-CCg 3'	5'-ggC 3'	
						418						
						5'-Agg 3'						
3'-primer(s) ³	527	302	538	302	539	201	302	220	302	3 rd	539	427
	5'-CCg 3'	5'-ggT 3'	5'-CCA 3'	5'-ggC 3'	5'-TCT 3'	5'-CTT 3'	5'-ggT 3'	5'-CgA 3'	5'-ggT 3'	5'-CTC 3'	5'-TCT 3'	5'-gTA 3'
	527					539	539					530
	5'-CCg 3'					5'-TCT 3'	5'-TCT 3'					5'-CCA 3'
												563
												5'-CgT 3'
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
HLA-C allele ⁴												
*16:01:01, 16:01:03-16:01:06	1			4	5							
*16:01:02	1				5							
*16:02:01-16:02:06	1	2			5							
*16:04:01	1		3	4								
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

INTERPRETATION TABLE											
HLA-C*16 SSP subtyping											
Amplification patterns of the HLA-C*16:01 to 16:32 alleles											
Well⁹											
13	14	15	16	17	18	19	20	21	22	23	
130	210	145	375	180	255	110	95	210	210	170	Length of spec. PCR product(s)
						240	145				
1070	1070	1070	1070	1070	1070	800	1070	1070	800	1070	Length of int.
											pos. control ¹
126	368	97	361	201	126	361	436	368	524	201	5'-primer(s) ²
5'-ggA 3'	5'-gTg 3'	5'-TCg 3'	5'-AgA 3'	5'-CCA 3'	5'-ggA 3'	5'-AgT 3'	5'-AgA 3'	5'-gTT 3'	5'-CCA 3'	5'-CCA 3'	
							485				
							5'-CAA 3'				
214	539	201	3 rd	341	341	430	539	539	3 rd	331	3'-primer(s) ³
5'-CCA 3'	5'-TCT 3'	5'-CTT 3'	5'-CTC 3'	5'-CgT 3'	5'-CgT 3'	5'-gCg 3'	5'-TCT 3'	5'-TCT 3'	5'-CTC 3'	5'-CTA 3'	
						559					
						5'-CTC 3'					
						565					
						5'-CAT 3'					
13	14	15	16	17	18	19	20	21	22	23	Well No.
											HLA-C allele ⁴
											*16:01:01, 16:01:03-16:01:06
											*16:01:02
											*16:02:01-16:02:06
											*16:04:01
13	14	15	16	17	18	19	20	21	22	23	Well No.

Lot No.: **18M**

Lot-specific information

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Length of spec.	205	340	220	140	160	130	100	135	85	215	170	105
PCR product(s)						160	210			350	540	210
						210						245
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
*16:06				4	5	6						
*16:07:01				4	5		7					
*16:07:02				4	5							
*16:08	1			4	5			8				
*16:09	1	2			5		7					
*16:10	1			4		6						
*16:11	1			4	5	6						
*16:12	1	2			5				9			
*16:13	1			4	5					10		
*16:14	1			4	5							
*16:15:01-16:15:02, 16:20 ⁵	1			4	5						11	
*16:16Q, 16:17 ⁶	1			4	5							12
*16:18	1			4	5							
*16:19	1	2			5					10		
*16:21	1			4	5							
*16:22				4	5							12
*16:23	1			4	5							
*16:24	1			4	5							
*16:25	1	2			5						11	
*16:26	1			4	5							
*16:27, 16:32 ⁷	1			4	5							
*16:28, 16:31 ⁸	1			4	5							
*16:29	1		3	4								
*16:30N	1			4	5							
*01:04, 01:09, 12:03:04, 12:03:09, 14:16			3									
*01:05, 01:36, 02:02:12, 03:05, 03:13, 03:25, 03:27, 03:35, 07:02:11, 07:03, 07:16, 07:20, 07:36, 07:51, 07:56:01- 07:56:02, 07:59-07:60, 07:64, 07:79, 07:88, 07:96, 07:109, 07:127, 12:27, 14:09												
*01:10, 14:25												
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

Lot No.: **18M**

Lot-specific information

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130	210	145	375	180	255	110	95	210	210	170	Length of spec. PCR product(s)
						240	145				
13	14	15	16	17	18	19	20	21	22	23	Well No.
			16								*16:06
			16								*16:07:01
			16								*16:07:02
											*16:08
											*16:09
											*16:10
											*16:11
											*16:12
13											*16:13
											*16:14
											*16:15:01-16:15:02, 16:20 ⁵
											*16:16Q, 16:17 ⁶
	14										*16:18
											*16:19
		15									*16:21
											*16:22
								21			*16:23
									22		*16:24
											*16:25
				17	18						*16:26
							20				*16:27, 16:32 ⁷
						19					*16:28, 16:31 ⁸
						19					*16:29
									23		*16:30N
			16								*01:04, 01:09, 12:03:04, 12:03:09, 14:16
											*01:05, 01:36, 02:02:12, 03:05, 03:13, 03:25, 03:27, 03:35, 07:02:11, 07:03, 07:16, 07:20, 07:36, 07:51, 07:56:01- 07:56:02, 07:59-07:60, 07:64, 07:79, 07:88, 07:96, 07:109, 07:127, 12:27, 14:09
						19					*01:10, 14:25
13	14	15	16	17	18	19	20	21	22	23	Well No.

Lot No.: **18M**

Lot-specific information

www.olerup-ssp.com

Length of spec.	205	340	220	140	160	130	100	135	85	215	170	105
PCR product(s)						160	210			350	540	210
						210						245
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
*01:14, 02:02:09, 02:37, 03:07, 03:15, 03:45, 05:07N, 05:10, 05:16, 05:31, 12:09, 14:04, 14:12, 15:02:01-15:06:03, 15:08-15:13, 15:15-15:20, 15:24, 15:26-15:42, 15:44-15:48, 17:01:01:01-17:08		2										
*01:21, 02:27:01-02:27:02, 04:11, 04:55, 08:01:01-08:09, 08:11, 08:13-08:27, 08:29-08:33, 08:35-08:36N, 08:38-08:43, 08:45-08:46, 12:02:01-12:02:06, 12:08, 12:10:01-12:10:02, 12:14:01-12:14:02, 12:17-12:18, 12:22, 12:30, 12:36, 12:49				4								
*01:23, 07:01:11, 07:04:01-07:04:04, 07:10-07:12, 07:31, 07:43, 07:45, 07:63, 07:101, 07:139, 07:142												
*01:27												12
*02:02:01-02:02:03, 02:02:05-02:02:08, 02:02:10-02:02:11, 02:02:13-02:04, 02:06-02:11, 02:13, 02:15-02:16:02, 02:18-02:20, 02:22-02:26:02, 02:28-02:33, 02:35-02:36, 02:38N-02:40, 02:42-02:47, 04:01:10, 04:01:23, 04:27, 04:33, 04:52, 05:01:01:01-05:01:15, 05:03-05:06, 05:08-05:09, 05:11-05:15, 05:17-05:24, 05:26-05:30, 05:32-05:41, 05:44-05:53, 07:07, 08:10, 12:21, 12:41, 15:22		2										
*02:05, 02:17		2	3									
*02:12				w								
*02:14, 05:43		2										
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

Lot No.: **18M**

Lot-specific information

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130	210	145	375	180	255	110	95	210	210	170	Length of spec. PCR product(s)
						240	145				
13	14	15	16	17	18	19	20	21	22	23	Well No.
											*01:14, 02:02:09, 02:37, 03:07, 03:15, 03:45, 05:07N, 05:10, 05:16, 05:31, 12:09, 14:04, 14:12, 15:02:01-15:06:03, 15:08-15:13, 15:15-15:20, 15:24, 15:26-15:42, 15:44- 15:48, 17:01:01-01-17:08
			16								*01:21, 02:27:01-02:27:02, 04:11, 04:55, 08:01:01-08:09, 08:11, 08:13-08:27, 08:29- 08:33, 08:35-08:36N, 08:38- 08:43, 08:45-08:46, 12:02:01- 12:02:06, 12:08, 12:10:01- 12:10:02, 12:14:01-12:14:02, 12:17-12:18, 12:22, 12:30, 12:36, 12:49
					18						*01:23, 07:01:11, 07:04:01- 07:04:04, 07:10-07:12, 07:31, 07:43, 07:45, 07:63, 07:101, 07:139, 07:142
											*01:27
			16								*02:02:01-02:02:03, 02:02:05- 02:02:08, 02:02:10-02:02:11, 02:02:13-02:04, 02:06-02:11, 02:13, 02:15-02:16:02, 02:18- 02:20, 02:22-02:26:02, 02:28- 02:33, 02:35-02:36, 02:38N- 02:40, 02:42-02:47, 04:01:10, 04:01:23, 04:27, 04:33, 04:52, 05:01:01-05:01:15, 05:03- 05:06, 05:08-05:09, 05:11- 05:15, 05:17-05:24, 05:26- 05:30, 05:32-05:41, 05:44- 05:53, 07:07, 08:10, 12:21, 12:41, 15:22
						19					*02:05, 02:17
			16								*02:12
		15	16								*02:14, 05:43
13	14	15	16	17	18	19	20	21	22	23	Well No.

Lot No.: **18M**

Lot-specific information

www.olerup-ssp.com

Length of spec.	205	340	220	140	160	130	100	135	85	215	170	105
PCR product(s)						160	210			350	540	210
						210						245
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
*02:21		2				6						
*02:34		2					7					
*03:108												
*04:01:01-04:01:09, 04:01:11-04:01:22, 04:01:25- 04:01:26, 04:04:01-04:05, 04:07-04:10, 04:12-04:13, 04:15:01-04:20, 04:23-04:26, 04:28, 04:30-04:32, 04:34- 04:35, 04:38-04:39, 04:41, 04:43-04:51, 04:53-04:54, 04:56- 04:67, 04:70-04:79, 04:81-04:84		2										
*04:01:24, 04:37, 04:40		2										
*04:03, 04:06, 05:25, 05:42		2										
*04:14, 04:68		2									11	
*04:29, 04:36				4								
*04:42		2										
*04:69		2							9			
*04:80		2										
*06:02:01:01-06:02:01:02, 06:02:03-06:02:09, 06:02:11- 06:03, 06:07, 06:09-06:10, 06:12-06:13, 06:15-06:21, 06:23- 06:25, 06:27-06:29, 06:33- 06:34, 06:36-06:39, 06:41- 06:51, 06:54-06:58		2	3									
*06:02:10, 06:26, 06:30, 06:53, 12:04:01-12:05, 12:33		2	3									
*06:04, 06:06, 06:14, 06:35, 06:40		2										
*06:05		2										
*06:08, 06:22		2	3									
*06:11, 06:52			3									
*06:31	1	2	3									
*06:32		2	3									
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

Lot No.: **18M**

Lot-specific information

www.olerup-ssp.com

130	210	145	375	180	255	110	95	210	210	170	Length of spec. PCR product(s)
						240	145				
13	14	15	16	17	18	19	20	21	22	23	Well No.
			16								*02:21
			16								*02:34
									22		*03:108
			16	17							*04:01:01:01-04:01:09, 04:01:11-04:01:22, 04:01:25- 04:01:26, 04:04:01-04:05, 04:07-04:10, 04:12-04:13, 04:15:01-04:20, 04:23-04:26, 04:28, 04:30-04:32, 04:34- 04:35, 04:38-04:39, 04:41, 04:43-04:51, 04:53-04:54, 04:56- 04:67, 04:70-04:79, 04:81-04:84
				17							*04:01:24, 04:37, 04:40
			16	17	18						*04:03, 04:06, 05:25, 05:42
			16	17							*04:14, 04:68
			16	17							*04:29, 04:36
		15	16	17							*04:42
			16	17							*04:69
				17	18						*04:80
					18						*06:02:01:01-06:02:01:02, 06:02:03-06:02:09, 06:02:11- 06:03, 06:07, 06:09-06:10, 06:12-06:13, 06:15-06:21, 06:23- 06:25, 06:27-06:29, 06:33- 06:34, 06:36-06:39, 06:41- 06:51, 06:54-06:58
											*06:02:10, 06:26, 06:30, 06:53, 12:04:01-12:05, 12:33
					18						*06:04, 06:06, 06:14, 06:35, 06:40
		15		17	18						*06:05
					18	19					*06:08, 06:22
					18						*06:11, 06:52
					18						*06:31
13					18						*06:32
13	14	15	16	17	18	19	20	21	22	23	Well No.

Lot No.: **18M**

Lot-specific information

www.olerup-ssp.com

Length of spec.	205	340	220	140	160	130	100	135	85	215	170	105
PCR product(s)						160	210			350	540	210
						210						245
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
*07:01:01-07:01:10, 07:01:12-07:02:08, 07:02:10, 07:02:12-07:02:23, 07:05-07:06, 07:08, 07:13-07:15, 07:17-07:19, 07:21-07:23, 07:26-07:30, 07:32N-07:33N, 07:35, 07:37-07:42, 07:44, 07:46-07:48, 07:50, 07:52, 07:54-07:55N, 07:57-07:58, 07:61N-07:62, 07:65-07:75, 07:77-07:78, 07:80-07:87, 07:89-07:95, 07:97-07:100, 07:102-07:108, 07:110-07:113, 07:115-07:126, 07:128-07:138, 07:140-07:141, 07:143-07:171												
*07:02:09				4								
*07:09, 07:49, 07:76, 18:01-18:04		2										
*07:24, 07:114										10		
*07:25												
*07:53					5						11	
*08:12, 08:34, 08:44, 12:19-12:20, 12:24, 12:44, 14:02:03, 14:03, 14:08, 14:10, 14:22, 15:07, B*35:08:02, B*67:02				4								
*08:28				4								
*08:37, 12:16				4								
*12:03:01:01-12:03:03, 12:03:05-12:03:08, 12:03:10-12:03:15, 12:06-12:07, 12:11-12:13, 12:15, 12:23, 12:25-12:26, 12:29, 12:31-12:32, 12:34-12:35, 12:37-12:39N, 12:42Q-12:43, 12:46N-12:48, 12:50-12:51												
*12:28			3	4								
*12:40				4								
*12:45			3	4						10		
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

Lot No.: **18M**

Lot-specific information

www.olerup-ssp.com

130	210	145	375	180	255	110	95	210	210	170	Length of spec. PCR product(s)
						240	145				
13	14	15	16	17	18	19	20	21	22	23	Well No.
			16		18						*07:01:01-07:01:10, 07:01:12-07:02:08, 07:02:10, 07:02:12-07:02:23, 07:05-07:06, 07:08, 07:13-07:15, 07:17-07:19, 07:21-07:23, 07:26-07:30, 07:32N-07:33N, 07:35, 07:37-07:42, 07:44, 07:46-07:48, 07:50, 07:52, 07:54-07:55N, 07:57-07:58, 07:61N-07:62, 07:65-07:75, 07:77-07:78, 07:80-07:87, 07:89-07:95, 07:97-07:100, 07:102-07:108, 07:110-07:113, 07:115-07:126, 07:128-07:138, 07:140-07:141, 07:143-07:171
		15	16	17	18						*07:02:09
			16		18						*07:09, 07:49, 07:76, 18:01-18:04
			16		18						*07:24, 07:114
			16		18			22			*07:25
			16		18						*07:53
											*08:12, 08:34, 08:44, 12:19-12:20, 12:24, 12:44, 14:02:03, 14:03, 14:08, 14:10, 14:22, 15:07, B*35:08:02, B*67:02
			16	17	18						*08:28
	15	16									*08:37, 12:16
											*12:03:01:01-12:03:03, 12:03:05-12:03:08, 12:03:10-12:03:15, 12:06-12:07, 12:11-12:13, 12:15, 12:23, 12:25-12:26, 12:29, 12:31-12:32, 12:34-12:35, 12:37-12:39N, 12:42Q-12:43, 12:46N-12:48, 12:50-12:51
				17	18						*12:28
13			16								*12:40
											*12:45
13	14	15	16	17	18	19	20	21	22	23	Well No.

Length of spec.	205	340	220	140	160	130	100	135	85	215	170	105
PCR product(s)						160	210			350	540	210
						210						245
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
*15:21				w								
*15:23		2										
*15:25				4								
A*24:73, A*24:157, B*07:66, B*51:55											11	
A*24:106						6						
B*46:25					5		7				11	
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

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-C*16 SSP subtyping.

In addition, wells number 3 to 5, 10, 19 and 22 contain the primer pair giving rise to the shorter, 800 bp, internal positive control band.

²The nucleotide position, in the 2nd or 3rd exon or the 1st intron, 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 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 HLA-C*1603 nucleotide sequence has been shown to be identical to C*14:03.

The HLA-C*16042 nucleotide sequence has been shown to be identical to C*16:04:01.

The HLA-C*1605 nucleotide sequence has been shown to be identical to C*16:04:01.

⁵The C*16:15:01-16:15:02 and C*16:20 alleles may be distinguished by the different sizes of the specific PCR products generated by primer mix 11.

⁶The C*16:16Q and C*16:17 alleles may be distinguished by the different sizes of the specific PCR products generated by primer mix 12.

⁷The C*16:27 and C*16:32 alleles may be distinguished by the different sizes of the specific PCR products generated by primer mix 20.

⁸The C*16:28 and C*16:31 alleles may be distinguished by the different sizes of the specific PCR products generated by primer mix 19.

Lot No.: **18M**

Lot-specific information

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130	210	145	375	180	255	110	95	210	210	170	Length of spec. PCR product(s)
						240	145				
13	14	15	16	17	18	19	20	21	22	23	Well No.
											*15:21
		15									*15:23
				17	18						*15:25
											A*24:73, A*24:157, B*07:66, B*51:55
											A*24:106
											B*46:25
13	14	15	16	17	18	19	20	21	22	23	Well No.

⁹Primer mix 6: Specific PCR fragment of 130 bp in the C*16:11 and in the C*02:21 allele. Specific PCR fragment of 160 bp in the C*16:10 and in the A*24:106 alleles. Specific PCR fragment of 210 bp in the C*16:06 allele.

Primer mix 7: Specific PCR fragment of 100 bp in the C*16:09 and in the C*02:34 alleles. Specific PCR fragment of 210 bp in the C*16:07:01 and the B*46:25 alleles.

Primer mix 10: Specific PCR fragment of 215 bp in the C*16:19 and the C*07:114 alleles. Specific PCR fragment of 350 bp in the C*16:13 and the C*07:24 and 12:45 alleles.

Primer mix 11: Specific PCR fragment of 170 bp in the C*16:20 and in the A*24:73, 24:157, B*07:66, B*46:25 and B*51:55 alleles. Specific PCR fragment of 540 bp in the C*16:15:01-16:15:02 and 16:25 and the C*04:14, 04:68 and 07:53 alleles.

Primer mix 12: Specific PCR fragment of 105 bp in the C*16:17 and in the C*01:27 alleles. Specific PCR fragment of 210 bp in the C*16:22 allele. Specific PCR fragment of 245 bp in the C*16:16Q allele.

Primer mix 19: Specific PCR fragment of 110 bp in the C*16:28 allele. Specific PCR fragment of 240 bp in the C*16:29 and 16:31 and the C*01:10, 02:05, 02:17, 06:08, 06:22 and 14:25 alleles.

Primer mix 20: Specific PCR fragment of 95 bp in the C*16:27 allele. Specific PCR fragment of 145 bp in the C*16:32 allele.

'w' might be weakly amplified.

CELL LINE VALIDATION SHEET																				
HLA-C*16 SSP primer set																				
				Well																
				1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	
				Prod. No.:	201186001	201073702	201073703	201073704	201073705	201073706	201073707	201073708	201073709	201073710	201073711	201186012	201073713	201073714	201073715	201186016
IHCW cell line			C*																	
1	9001	SA	*07:02	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+
2	9280	LK707	*07:01	*15:05	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	+
3	9011	E4181324	*12:02		-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	+
4	9275	GU373	*03:04	*04:01	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	+
5	9009	KAS011	*06:02		-	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
6	9353	SM	*03:04	*07:02	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+
7	9020	QBL	*05:01		-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	+
8	9007	DEM	*04:01		-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	+
9	9026	YAR	*12:03		-	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-
10	9107	LKT3	*01:02		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
11	9051	PITOUT	*16:01		+	-	-	+	+	-	-	-	-	-	-	-	-	-	-	-
12	9052	DBB	*06:02		-	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
13	9004	JESTHOM	*01:02		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
14	9071	OLGA	*01:02	*03:04	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
15	9075	DKB	*03:04		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
16	9037	SWEIG007	*02:02		-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	+
17	9282	CTM3953540	*03:03	*07:01	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+
18	9257	32367	*01:02	*07:05	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+
19	9038	BM16	*07:01		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+
20	9059	SLE005	*03:04		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
21	9064	AMALA	*03:03		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
22	9056	KOSE	*12:03		-	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-
23	9124	IHL	*01:02	*15:02	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
24	9035	JBUSH	*12:03		-	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-
25	9049	IBW9	*08:02		-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	+
26	9285	WT49	*07:01		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+
27	9191	CH1007	*07:04	*15:05	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
28	9320	BEL5GB	*05:01	*16:01	+	+	-	+	+	-	-	-	-	-	-	-	-	-	-	+
29	9050	MOU	*16:01		+	-	-	+	+	-	-	-	-	-	-	-	-	-	-	-
30	9021	RSH	*17:01		-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
31	9019	DUCAF	*05:01		-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	+
32	9297	HAG	*17:01	*17:03	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
33	9098	MT14B	*03:04		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
34	9104	DHIF	*12:03		-	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-
35	9302	SSTO	*05:01		-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	+
36	9024	KT17	*03:03	*04:01	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	+
37	9065	HHKB	*07:02		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+
38	9099	LZL	*03:03		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
39	9315	CML	*02:02	*07:01	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	+
40	9134	WHONP199	*01:02	*06:02	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
41	9055	H0301	*08:02		-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	+
42	9066	TAB089	*01:02		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
43	9076	T7526	*01:02	*08:01	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	+
44	9057	TEM	*12:03		-	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-
45	9239	SHJO	*06:02	*17:01	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
46	9013	SCHU	*07:02		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+
47	9045	TUBO	*07:04	*15:02	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
48	9303	TER-ND	*04:01	*16:01	+	+	-	+	+	-	-	-	-	-	-	-	-	-	-	+

CELL LINE VALIDATION SHEET					HLA-C*16 SSP primer set							
					Well							
					17	18	19	20	21	22	23	
					Prod. No.:	201186017	201186018	201186019	201186020	201186021	201186022	201186023
IHWC cell line			C*									
1	9001	SA	*07:02		-	+	-	-	-	-	-	
2	9280	LK707	*07:01	*15:05	-	+	-	-	-	-	-	
3	9011	E4181324	*12:02		-	-	-	-	-	-	-	
4	9275	GU373	*03:04	*04:01	+	-	-	-	-	-	-	
5	9009	KAS011	*06:02		-	+	-	-	-	-	-	
6	9353	SM	*03:04	*07:02	-	+	-	-	-	-	-	
7	9020	QBL	*05:01		-	-	-	-	-	-	-	
8	9007	DEM	*04:01		+	-	-	-	-	-	-	
9	9026	YAR	*12:03		-	-	-	-	-	-	-	
10	9107	LKT3	*01:02		-	-	-	-	-	-	-	
11	9051	PITOUT	*16:01		-	-	-	-	-	-	-	
12	9052	DBB	*06:02		-	+	-	-	-	-	-	
13	9004	JESTHOM	*01:02		-	-	-	-	-	-	-	
14	9071	OLGA	*01:02	*03:04	-	-	-	-	-	-	-	
15	9075	DKB	*03:04		-	-	-	-	-	-	-	
16	9037	SWEIG007	*02:02		-	-	-	-	-	-	-	
17	9282	CTM3953540	*03:03	*07:01	-	+	-	-	-	-	-	
18	9257	32367	*01:02	*07:05	-	+	-	-	-	-	-	
19	9038	BM16	*07:01		-	+	-	-	-	-	-	
20	9059	SLE005	*03:04		-	-	-	-	-	-	-	
21	9064	AMALA	*03:03		-	-	-	-	-	-	-	
22	9056	KOSE	*12:03		-	-	-	-	-	-	-	
23	9124	IHL	*01:02	*15:02	-	-	-	-	-	-	-	
24	9035	JBUSH	*12:03		-	-	-	-	-	-	-	
25	9049	IBW9	*08:02		-	-	-	-	-	-	-	
26	9285	WT49	*07:01		-	+	-	-	-	-	-	
27	9191	CH1007	*07:04	*15:05	-	+	-	-	-	-	-	
28	9320	BEL5GB	*05:01	*16:01	-	-	-	-	-	-	-	
29	9050	MOU	*16:01		-	-	-	-	-	-	-	
30	9021	RSH	*17:01		-	-	-	-	-	-	-	
31	9019	DUCAF	*05:01		-	-	-	-	-	-	-	
32	9297	HAG	*17:01	*17:03	-	-	-	-	-	-	-	
33	9098	MT14B	*03:04		-	-	-	-	-	-	-	
34	9104	DHIF	*12:03		-	-	-	-	-	-	-	
35	9302	SSTO	*05:01		-	-	-	-	-	-	-	
36	9024	KT17	*03:03	*04:01	+	-	-	-	-	-	-	
37	9065	HHKB	*07:02		-	+	-	-	-	-	-	
38	9099	LZL	*03:03		-	-	-	-	-	-	-	
39	9315	CML	*02:02	*07:01	-	+	-	-	-	-	-	
40	9134	WHONP199	*01:02	*06:02	-	+	-	-	-	-	-	
41	9055	H0301	*08:02		-	-	-	-	-	-	-	
42	9066	TAB089	*01:02		-	-	-	-	-	-	-	
43	9076	T7526	*01:02	*08:01	-	-	-	-	-	-	-	
44	9057	TEM	*12:03		-	-	-	-	-	-	-	
45	9239	SHJO	*06:02	*17:01	-	+	-	-	-	-	-	
46	9013	SCHU	*07:02		-	+	-	-	-	-	-	
47	9045	TUBO	*07:04	*15:02	-	+	-	-	-	-	-	
48	9303	TER-ND	*04:01	*16:01	+	-	-	-	-	-	-	

CERTIFICATE OF ANALYSIS

Olerup SSP® HLA-C*16 SSP

Product number: 101.627-12u – without *Taq* polymerase
Lot number: 18M
Expiry date: 2013-December-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	2011-860-01	9	2010-737-09	17	2011-860-17
2	2010-737-02	10	2010-737-10	18	2011-860-18
3	2010-737-03	11	2010-737-11	19	2011-860-19
4	2010-737-04	12	2011-860-12	20	2011-860-20
5	2010-737-05	13	2010-737-13	21	2011-860-21
6	2010-737-06	14	2010-737-14	22	2011-860-22
7	2010-737-07	15	2010-737-15	23	2011-860-23
8	2010-737-08	16	2011-860-16		

The specificity of each primer solution of the HLA-C*16 primer set has been tested against 48 well characterized cell line DNAs.

No DNAs carrying the alleles to be amplified by primer solutions 6 to 15 and 19 to 23 were available. The specificity of the primers in primer solutions 6, 7, 9, 11, 14, 15 and 19 to 22 were tested by separately adding one additional 5'-primer, respectively one additional 3'-primer.

In primer solutions 10 it was only possible to test the 3'-primer, the 5'-primer was not possible to test. In primer solutions 8, 12, 13 and 23 it was only possible to test the 5'-primer, the 3'-primer was not possible to test. In primer solutions 1, 6, 7 and 20 one or two of the 5'-primers were not possible to test, and in primer solution 19 two of the 3'-primers were not possible to test.

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

Date of approval: 2011-July-13

Approved by:

Quality Control, Supervisor

Lot No.: **18M**

Lot-specific information

www.olerup-ssp.com

Declaration of Conformity

Product name: *Olerup* SSP® HLA-C*16
Product number: 101.627-12u
Lot number: 18M

Intended use: HLA-C*16 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 13485:2003, following the provisions of the 98/79/EC Directive on *in vitro* diagnostic medical devices, Annex III, 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.

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

Stockholm, Sweden
2011-July-13

Olle Olerup
Managing Director

Lot No.: **18M**

Lot-specific information

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