

Olerup SSP® HLA-A*30

Product number:	101.429-12u – without <i>Taq</i> polymerase
Lot number:	51M
Expiry date:	2014-February-01
Number of tests:	12
Number of wells per test:	24
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. 51M.

CHANGES COMPARED TO THE PREVIOUS *OLERUP SSP*® HLA-A*30 LOT

The HLA-A*30 specificity and interpretation tables have been updated for the HLA-A alleles described since the previous *Olerup SSP*® HLA-A*30 lot was made (Lot No. 02H).

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

Well	5'-primer	3'-primer	rationale
12	-	Added	3'-primer added for the A*30:35 allele.
22	-	Added	3'-primer added for the A*30:36 allele.
24	Added	Added	Primer pair added for the A*30:37 allele, exchanged positive control primer pair.

PRODUCT DESCRIPTION

HLA-A*30 SSP subtyping

CONTENT

The primer set contains 5'- and 3'-primers for identifying the A*30:01 to A*30:37 alleles.

PLATE LAYOUT

Each test consists of 24 PCR reactions in a 24 well cut PCR plate.

1	2	3	4	5	6	7	8
9	10	11	12	13	14	15	16
17	18	19	20	21	22	23	24

The 24 well cut PCR plate is marked with 'HLA-A*30' in silver/gray ink.

Well No. 1 is marked with '51M'.

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 covered with a PCR-compatible foil.

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

INTERPRETATION

The interpretation of HLA-A*30 SSP subtypings will be influenced by three A*01, two A*02, three A*03, the A*23:09, four A*24, two A*26, the A*29:14, most A*31, the A*32:07, two A*68 and the A*80 alleles when present on the other haplotype.

UNIQUELY IDENTIFIED ALLELES

All the HLA-A*30 alleles, i.e. **A*30:01 to A*30:37 alleles**, recognized by the HLA Nomenclature Committee in April 2010¹ will give rise to unique amplification patterns by the primers in the HLA-A*30 subtyping kit.

The HLA-A*30 kit cannot distinguish the A*30:01:01 to A*30:01:03 alleles, the A*30:02:01 to A*30:02:04 or the A*30:11:01 and A*30:11:02 alleles.

¹HLA-A 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

A total of 63 alleles generate 35 amplification patterns that can be combined in 630 homozygous and heterozygous combinations. 305 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.

++++-----	+++-----	+-----	*30:03, *30:18 = *30:11:01, *30:12
++++-----	+--+++--	+-----	*30:03, *30:16 = *30:11:01, *30:13
++++-----	+--+++--	+-----	*30:03, *30:15 = *30:11:01, *30:33
++++-----	+--+++--	+-----	*30:03, *30:20 = *30:11:01, *30:32
++++-----	+--+++--	+-----	*30:03, *30:23 = *30:11:01, *30:34
++++-----	+--+++--	+-----	*30:03, *30:24 = *30:03, *30:30
++++-----	+--+++--	+-----	*30:03, *30:36 = *30:11:01, *30:27N
++++-----	+--+++--	+-----	*30:03, *30:37 = *30:11:01, *30:25
++++-----	+--+++--	+-----	*30:01:01, *30:03 = *30:02:01, *30:11:01 = *30:02:05, *30:11:01
+++-+-----	+--+++--	+-----	*30:02:01, *30:19 = *30:02:05, *30:19
+++-+-----	+--+++--	+-----	*30:07, *30:24 = *30:07, *30:30
+++-+-----	+--+++--	+-----	*30:02:01, *30:08 = *30:02:05, *30:08
+++-+-----	+--+++--	+-----	*30:10, *30:24 = *30:10, *30:30
+++-+-----	++++++--	+-----	*30:12, *30:16 = *30:13, *30:18
+++-+-----	++++++--	+-----	*30:12, *30:15 = *30:18, *30:33
+++-+-----	++++++--	+-----	*30:12, *30:20 = *30:18, *30:32
+++-+-----	++++++--	+-----	*30:12, *30:23 = *30:18, *30:34
+++-+-----	++++++--	+-----	*30:12, *30:24 = *30:12, *30:30
+++-+-----	++++++--	+-----	*30:12, *30:36 = *30:18, *30:27N
+++-+-----	++++++--	+-----	*30:12, *30:37 = *30:18, *30:25
+++-+-----	++++++--	+-----	*30:01:01, *30:12 = *30:02:01, *30:18 = *30:02:05, *30:18
+++-+-----	+-----	+-----	*30:02:01, *30:35 = *30:02:05, *30:35
+++-+-----	+-----	+-----	*30:13, *30:15 = *30:16, *30:33
+++-+-----	+-----	+-----	*30:13, *30:20 = *30:16, *30:32
+++-+-----	+-----	+-----	*30:13, *30:31 = *30:16, *30:22
+++-+-----	+-----	+-----	*30:13, *30:23 = *30:16, *30:34
+++-+-----	+-----	+-----	*30:13, *30:24 = *30:13, *30:30
+++-+-----	+-----	+-----	*30:13, *30:36 = *30:16, *30:27N
+++-+-----	+-----	+-----	*30:13, *30:37 = *30:16, *30:25
+++-+-----	+-----	+-----	*30:01:01, *30:13 = *30:02:01, *30:16 = *30:02:05, *30:16 = *30:13, *30:16
+++-+-----	+-----	+-----	*30:02:01, *30:14L = *30:02:05, *30:14L
+++-+-----	+-----	+-----	*30:15, *30:32 = *30:20, *30:33
+++-+-----	+-----	+-----	*30:15, *30:22 = *30:31, *30:33
+++-+-----	+-----	+-----	*30:15, *30:34 = *30:23, *30:33
+++-+-----	+-----	+-----	*30:24, *30:33 = *30:30, *30:33
+++-+-----	+-----	+-----	*30:15, *30:27N = *30:33, *30:36
+++-+-----	+-----	+-----	*30:15, *30:25 = *30:33, *30:37
+++-+-----	+-----	+-----	*30:01:01, *30:33 = *30:02:01, *30:15 = *30:02:05, *30:15 = *30:15, *30:33
+++-+-----	+-----	+-----	*30:20, *30:22 = *30:31, *30:32

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+++-----	+--+-----	+--+-----	*30:20, *30:34 = *30:23, *30:32
+++-----	+--+-----	+--+-----	*30:24, *30:32 = *30:30, *30:32
+++-----	+--+-----	+--+-----	*30:20, *30:27N = *30:32, *30:36
+++-----	+--+-----	+--+-----	*30:20, *30:25 = *30:32, *30:37
+++-----	+--+-----	+--+-----	*30:01:01, *30:32 = *30:02:01, *30:20 = *30:02:05, *30:20 = *30:20, *30:32
+++-----	+--+-----	+--+-----	*30:22, *30:23 = *30:31, *30:34
+++-----	+--+-----	+--+-----	*30:22, *30:24 = *30:22, *30:30
+++-----	+--+-----	+--+-----	*30:22, *30:36 = *30:27N, *30:31
+++-----	+--+-----	+--+-----	*30:22, *30:37 = *30:25, *30:31
+++-----	+--+-----	+--+-----	*30:01:01, *30:22 = *30:02:01, *30:31 = *30:02:05, *30:31
+++-----	+--+-----	+--+-----	*30:24, *30:34 = *30:30, *30:34
+++-----	+--+-----	+--+-----	*30:23, *30:27N = *30:34, *30:36
+++-----	+--+-----	+--+-----	*30:23, *30:25 = *30:34, *30:37
+++-----	+--+-----	+--+-----	*30:01:01, *30:34 = *30:02:01, *30:23 = *30:02:05, *30:23 = *30:23, *30:34
+++-----	+--+-----	+--+-----	*30:24, *30:27N = *30:27N, *30:30
+++-----	+--+-----	+--+-----	*30:24, *30:25 = *30:25, *30:30
+++-----	+--+-----	+--+-----	*30:02:01, *30:24 = *30:02:01, *30:30 = *30:02:05, *30:24
+++-----	+--+-----	+--+-----	*30:25, *30:36 = *30:27N, *30:37
+++-----	+--+-----	+--+-----	*30:01:01, *30:27N = *30:02:01, *30:36 = *30:02:05, *30:36 = *30:27N, *30:36
+++-----	+--+-----	+--+-----	*30:01:01, *30:25 = *30:02:01, *30:37 = *30:02:05, *30:37 = *30:25, *30:37
+++-----	+--+-----	+--+-----	*30:01:01, *30:02:01 = *30:01:01, *30:02:05
+++-----	+--+-----	+--+-----	*30:11:01, *30:17 = *30:11:01, *30:19
+++-----	+--+-----	+--+-----	*30:09, *30:11:01 = *30:11:01, *30:35
+++-----	+--+-----	+--+-----	*30:11:01, *30:24 = *30:11:01, *30:30
+++-----	+--+-----	+--+-----	*30:01:01, *30:06 = *30:06, *30:19
+++-----	+--+-----	+--+-----	*30:08, *30:17 = *30:08, *30:19 = *30:19, *30:28
+++-----	+--+-----	+--+-----	*30:17, *30:18 = *30:18, *30:19
+++-----	+--+-----	+--+-----	*30:09, *30:19 = *30:17, *30:35 = *30:19, *30:35
+++-----	+--+-----	+--+-----	*30:16, *30:17 = *30:16, *30:19
+++-----	+--+-----	+--+-----	*30:01:01, *30:29 = *30:14L, *30:17 = *30:14L, *30:19 = *30:14L, *30:29 = *30:19, *30:29
+++-----	+--+-----	+--+-----	*30:15, *30:17 = *30:15, *30:19
+++-----	+--+-----	+--+-----	*30:17, *30:20 = *30:19, *30:20
+++-----	+--+-----	+--+-----	*30:17, *30:31 = *30:19, *30:31
+++-----	+--+-----	+--+-----	*30:17, *30:23 = *30:19, *30:23
+++-----	+--+-----	+--+-----	*30:17, *30:24 = *30:19, *30:24 = *30:19, *30:30
+++-----	+--+-----	+--+-----	*30:17, *30:36 = *30:19, *30:36
+++-----	+--+-----	+--+-----	*30:17, *30:37 = *30:19, *30:37
+++-----	+--+-----	+--+-----	*30:01:01, *30:17 = *30:01:01, *30:19 = *30:17, *30:19 = *30:19, *30:19
+++-----	+--+-----	+--+-----	*30:08, *30:18 = *30:18, *30:28
+++-----	+--+-----	+--+-----	*30:08, *30:35 = *30:28, *30:35
+++-----	+--+-----	+--+-----	*30:08, *30:16 = *30:16, *30:28
+++-----	+--+-----	+--+-----	*30:08, *30:14L = *30:14L, *30:28

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++-----+	+--+---+	+-----	*30:08, *30:15 = *30:15, *30:28
++-----+	+--+---+	+-----	*30:08, *30:20 = *30:20, *30:28
++-----+	+--+---+	+-----	*30:08, *30:23 = *30:23, *30:28
++-----+	+--+---+	+-----	*30:08, *30:24 = *30:08, *30:30 = *30:24, *30:28 = *30:28, *30:30
++-----+	+--+---+	+-----	*30:08, *30:36 = *30:28, *30:36
++-----+	+--+---+	+-----	*30:08, *30:26 = *30:26, *30:28
++-----+	+--+---+	+-----	*30:08, *30:37 = *30:28, *30:37
++-----+	+--+---+	+-----	*30:01:01, *30:08 = *30:01:01, *30:28
++-----	+++++---	+-----	*30:09, *30:18 = *30:18, *30:35
++-----	+++++---	+-----	*30:18, *30:24 = *30:18, *30:30
++-----	+++++---	+-----	*30:09, *30:16 = *30:16, *30:35
++-----	+++++---	+-----	*30:09, *30:14L = *30:14L, *30:35
++-----	+++++---	+-----	*30:09, *30:15 = *30:15, *30:35
++-----	+++++---	+-----	*30:09, *30:20 = *30:20, *30:35
++-----	+++++---	+-----	*30:09, *30:31 = *30:31, *30:35
++-----	+++++---	+-----	*30:09, *30:23 = *30:23, *30:35
++-----	+++++---	+-----	*30:09, *30:24 = *30:09, *30:30 = *30:24, *30:35 = *30:30, *30:35
++-----	+++++---	+-----	*30:09, *30:36 = *30:35, *30:36
++-----	+++++---	+-----	*30:09, *30:26 = *30:26, *30:35
++-----	+++++---	+-----	*30:09, *30:37 = *30:35, *30:37
++-----	+++++---	+-----	*30:01:01, *30:09 = *30:01:01, *30:35 = *30:09, *30:35 = *30:35, *30:35
++-----	+++++---	+-----	*30:16, *30:24 = *30:16, *30:30
++-----	+++++---	+-----	*30:01:01, *30:16 = *30:16, *30:16
++-----	+++++---	+-----	*30:14L, *30:24 = *30:14L, *30:30
++-----	+++++---	+-----	*30:01:01, *30:14L = *30:14L, *30:14L
++-----	+++++---	+-----	*30:15, *30:24 = *30:15, *30:30
++-----	+++++---	+-----	*30:01:01, *30:15 = *30:15, *30:15
++-----	+++++---	+-----	*30:20, *30:24 = *30:20, *30:30
++-----	+++++---	+-----	*30:01:01, *30:20 = *30:20, *30:20
++-----	+++++---	+-----	*30:24, *30:31 = *30:30, *30:31
++-----	+++++---	+-----	*30:23, *30:24 = *30:23, *30:30
++-----	+++++---	+-----	*30:01:01, *30:23 = *30:23, *30:23
++-----	+++++---	+-----	*30:24, *30:36 = *30:30, *30:36
++-----	+++++---	+-----	*30:24, *30:37 = *30:30, *30:37
++-----	+++++---	+-----	*30:01:01, *30:24 = *30:01:01, *30:30 = *30:24, *30:24 = *30:24, *30:30
++-----	+++++---	+-----	*30:01:01, *30:36 = *30:36, *30:36
++-----	+++++---	+-----	*30:01:01, *30:37 = *30:37, *30:37
++-----	+++++---	+-----	*30:02:01, *30:03 = *30:02:05, *30:03
++-----	+++++---	+-----	*30:02:01, *30:06 = *30:06, *30:07
++-----	+++++---	+-----	*30:02:01, *30:07 = *30:02:05, *30:07
++-----	+++++---	+-----	*30:02:01, *30:28 = *30:02:05, *30:28
++-----	+++++---	+-----	*30:02:01, *30:10 = *30:02:05, *30:10 = *30:10, *30:10
++-----	+++++---	+-----	*30:02:01, *30:12 = *30:02:05, *30:12
++-----	+++++---	+-----	*30:02:01, *30:09 = *30:02:05, *30:09
++-----	+++++---	+-----	*30:02:01, *30:13 = *30:02:05, *30:13 = *30:13, *30:13
++-----	+++++---	+-----	*30:02:01, *30:33 = *30:02:05, *30:33 = *30:33, *30:33

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+--+-----	+--+-----	-+-----	*30:02:01, *30:32 = *30:02:05, *30:32 = *30:32, *30:32
+--+-----	+--+-----	--+-----	*30:02:01, *30:22 = *30:02:05, *30:22
+--+-----	+--+-----	---+-----	*30:02:01, *30:34 = *30:02:05, *30:34 = *30:34, *30:34
+--+-----	+--+-----	-----+--	*30:02:01, *30:27N = *30:02:05, *30:27N = *30:27N, *30:27N
+--+-----	+--+-----	-----+--	*30:02:01, *30:25 = *30:02:05, *30:25 = *30:25, *30:25
+--+-----	+--+-----	-----+--	*30:02:01, *30:02:01 = *30:02:01, *30:02:05
*30:01:01 = *30:01:01-30:01:03			
*30:02:01 = *30:02:01-30:02:04			
*30:11:01 = *30:11:01-30:11:02			

SPECIFICITY TABLE

HLA-A*30 SSP subtyping

Specificities and sizes of the PCR products of the 24 primer mixes used for HLA-A*30 SSP subtyping

Primer Mix	Size of spec. PCR product ¹	Size of control band ²	Amplified HLA-A*30 alleles	Other amplified HLA-A alleles ³
1	165 bp	800 bp	*30:01:01-30:02:04, 30:03, 30:07-30:16, 30:18-30:20, 30:22-30:25, 30:27N-30:28, 30:31-30:37	*02:52, 03:43
2	205 bp	800 bp	*30:01:01-30:01:03, 30:08, 30:11:01-30:11:02, 30:14L-30:16, 30:18-30:20, 30:23-30:24, 30:26, 30:30-30:31, 30:35-30:37	
3	210 bp	800 bp	*30:02:01-30:02:04, 30:02:05 ^w , 30:03, 30:07, 30:10, 30:12-30:13, 30:22, 30:25, 30:27N, 30:32-30:34	*80:01 ^w -80:02 ^w
4^{5,6}	155 bp	1070 bp	*30:03, 30:11:01-30:11:02	*01:02, 01:20
5⁷	150 bp, 245 bp	1070 bp	*30:04, 30:06, 30:17, 30:19, 30:29	*02:52, 24:66, 68:06
6^{4,8}	80 bp, 185 bp	1070 bp	*30:06-30:07	
7	200 bp	1070 bp	*30:08, 30:28	*02:185 ^w
8⁴	85 bp	800 bp	*30:10	
9	180 bp	1070 bp	*30:01:01-30:02:05, 30:04, 30:06-30:07, 30:09-30:10, 30:12-30:20, 30:23-30:37	
10	150 bp	1070 bp	*30:12, 30:18	*01:07, 02:185, 26:19, 29:14, 31:01:02-31:01:06, 31:02-31:07, 31:09-31:31

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11	215 bp	800 bp	*30:01:01-30:04, 30:06, 30:09-30:20, 30:23- 30:30, 30:32-30:37	*01:02 ^W , 01:20 ^W , 03:72, 23:09 ^W , 24:24, 24:67, 26:16, 32:07, 68:45
12	195 bp	1070 bp	*30:09, 30:35	*03:42, 31:03- 31:04
13	155 bp	1070 bp	*30:01:01-30:02:05, 30:04, 30:06-30:10, 30:13-30:17, 30:19- 30:20, 30:23-30:37	
14	210 bp	1070 bp	*30:13, 30:16	
15	245 bp	800 bp	*30:14L, 30:29	
16	265 bp	1070 bp	*30:15, 30:33	
17	225 bp	1070 bp	*30:01:01-30:01:03, 30:11:01-30:11:02, 30:14L-30:20, 30:23- 30:24, 30:26, 30:30- 30:31, 30:35-30:37	*03:72, 24:19, 68:45
18⁹	130 bp, 210 bp	800 bp	*30:20, 30:32	
19¹⁰	160 bp, 200 bp	1070 bp	*30:22, 30:31	
20¹¹	210 bp, 275 bp	800 bp	*30:23, 30:34	
21¹²	155 bp, 185 bp	1070 bp	*30:24, 30:30	
22¹³	170 bp, 215 bp	800 bp	*30:27N, 30:36	
23	155 bp	1070 bp	*30:26	
24^{4,14}	80 bp, 200 bp	800 bp	*30:25, 30:37	

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-A*30 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

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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-A*30 subtyping.

In addition, wells number 2, 3, 8, 11, 15, 18, 20, 22 and 24 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.

³Due to the sharing of sequence motifs between HLA-A alleles non-HLA-A*30 alleles will be amplified by primer mixes 1, 3 to 5, 7, 10 to 12 and 17.

⁴Short specific PCR fragments are less intense and not as sharp as longer specific bands.

⁵Primer mix 4 may give rise to nonspecific amplifications.

⁶In primer mix 4 the positive control band may be weaker than for other HLA-A*30 primer mixes.

⁷Primer mix 5: Specific PCR fragment of 150 bp in the A*30:04, 30:06, 30:17 and 30:29 and the A*02:52, 24:66 and 68:06 alleles. Specific PCR fragment of 245 bp in the A*30:19 alleles.

⁸Primer mix 6: Specific PCR fragment of 80 bp in the A*30:06 allele. Specific PCR fragment of 185 bp in the A*30:07 allele.

⁹Primer mix 18: Specific PCR fragment of 130 bp in the A*30:32 allele. Specific PCR fragment of 210 bp in the A*30:20 allele.

¹⁰Primer mix 19: Specific PCR fragment of 160 bp in the A*30:22 allele. Specific PCR fragment of 200 bp in the A*30:31 allele.

¹¹Primer mix 20: Specific PCR fragment of 210 bp in the A*30:23 allele. Specific PCR fragment of 275 bp in the A*30:34 allele.

¹²Primer mix 21: Specific PCR fragment of 185 bp in the A*30:24 allele. Specific PCR fragment of 155 and 185 bp in the A*30:30 allele.

¹³Primer mix 22: Specific PCR fragment of 170 bp in the A*30:36 allele. Specific PCR fragment of 215 bp in the A*30:27N allele.

¹⁴Primer mix 24: Specific PCR fragment of 80 bp in the A*30:37 allele. Specific PCR fragment of 200 bp in the A*30:25 allele.

'w', might be weakly amplified.

INTERPRETATION TABLE												
HLA-A*30 SSP subtyping												
Amplification patterns of the A*30:01 to 30:37 alleles												
	Well ⁵											
	1	2	3	4	5	6	7	8	9	10	11	12
Length of spec.	165	205	210	155	150	80	200	85	180	150	215	195
PCR product(s)					245	185						
Length of int.	800	800	800	1070	1070	1070	1070	800	1070	1070	800	1070
pos. control ¹												
5'-primer(s) ²	414	363	363	123	123	123	78	367	98	127	98	363
	5'-gAA 3'	5'-ATA 3'	5'-ATA 3'	5'-AgT 3'	5'-AgT 3'	5'-AgT 3'	5'-TCT 3'	5'-TgC 3'	5'-CTC 3'	5'-ggg 3'	5'-CTC 3'	5'-ATA 3'
					414		362					
					5'-gAA 3'		5'-ggT 3'					
3'-primer(s) ³	538	526	526	238	325	163	238	411	238	238	270	505
	5'-CAA 3'	5'-CCA 3'	5'-CCg 3'	5'-CCC 3'	5'-gTg 3'	5'-CgC 3'	5'-CCT 3'	5'-TCA 3'	5'-CCT 3'	5'-CCT 3'	5'-ACA 3'	5'-gCT 3'
			535		524	265	526					524
			5'-CTA 3'		5'-CAT 3'	5'-CCC 3'	5'-CCg 3'					5'-CAC 3'
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
HLA-A allele ⁴												
*30:01:01-30:01:03	1	2							9		11	
*30:02:01-30:02:04	1		3						9		11	
*30:02:05			W						9		11	
*30:03	1		3	4							11	
*30:04					5				9		11	
*30:06					5	6			9		11	
*30:07	1		3			6			9			
*30:08	1	2					7					
*30:09	1								9		11	12
*30:10	1		3					8	9		11	
*30:11:01-30:11:02	1	2		4							11	
*30:12	1		3						9	10	11	
*30:13	1		3						9		11	
*30:14L	1	2							9		11	
*30:15	1	2							9		11	
*30:16	1	2							9		11	
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

INTERPRETATION TABLE											
HLA-A*30 SSP subtyping											
Amplification patterns of the A*30:01 to 30:37 alleles											
Well ⁵											
13	14	15	16	17	18	19	20	21	22	23	24
155	210	245	265	225	130	160	210	155	170	155	80
					210	200	275	185	215		200
1070	1070	800	1070	1070	800	1070	800	1070	800	1070	800
123	123	363	363	98	123	123	363	410	363	413	123
5'-AgT 3'	5'-AgT 3'	5'-ATA 3'	5'-ATA 3'	5'-CTC 3'	5'-AgT 3'	5'-AgT 3'	5'-ATA 3'	5'-gTg 3'	5'-ATA 3'	5'-CCg 3'	5'-AgT 3'
			784					634			485
			5'-ggA 3'					5'-Cag 3'			5'-CAC 3'
238	292	563	586	282	212	240	530	526	494	526	282
5'-CCT 3'	5'-gTg 3'	5'-Cgg 3'	5'-CAC 3'	5'-gAC 3'	5'-gCC 3'	5'-ggA 3'	5'-CCT 3'	5'-CCA 3'	5'-TCC 3'	5'-CCA 3'	5'-gAC 3'
		572	899		294	281	595	777	535		526
		5'-gCg 3'	5'-ACA 3'		5'-CgT 3'	5'-ACC 3'	5'-CCT 3'	5'-gCA 3'	5'-CTA 3'		5'-CCA 3'
13	14	15	16	17	18	19	20	21	22	23	24
13				17							
13											
13											
13											
13											
13											
13											
13											
				17							
13	14										
13		15		17							
13			16	17							
13	14			17							
13	14	15	16	17	18	19	20	21	22	23	24

Lot No.: **51M**

Lot-specific information

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Length of spec.	165	205	210	155	150	80	200	85	180	150	215	195
PCR product(s)					245	185						
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
*30:17					5				9		11	
*30:18	1	2							9	10	11	
*30:19	1	2			5				9		11	
*30:20	1	2							9		11	
*30:22	1		3									
*30:23	1	2							9		11	
*30:24	1	2							9		11	
*30:25	1		3						9		11	
*30:26		2							9		11	
*30:27N	1		3						9		11	
*30:28	1						7		9		11	
*30:29					5				9		11	
*30:30		2							9		11	
*30:31	1	2							9			
*30:32	1		3						9		11	
*30:33	1		3						9		11	
*30:34	1		3						9		11	
*30:35	1	2							9		11	12
*30:36	1	2							9		11	
*30:37	1	2							9		11	
*01:02, 01:20				4							w	
*01:07, 26:19, 29:14, 31:01:02-31:01:06, 31:02, 31:05-31:07, 31:09-31:31										10		
*02:52	1				5							
*02:185							w			10		
*03:42												12
*03:43	1											
*03:72, 68:45											11	
*23:09											w	
*24:19												
*24:24, 24:67, 26:16, 32:07											11	
*24:66, 68:06					5							
*31:03-31:04										10		12
*80:01-80:02			w									
HLA-A allele ⁴												
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

Lot No.: **51M**

Lot-specific information

www.olerup-ssp.com

155	210	245	265	225	130	160	210	155	170	155	80	Length of spec. PCR product(s)
					210	200	275	185	215		200	
13	14	15	16	17	18	19	20	21	22	23	24	Well No.
13				17								*30:17
				17								*30:18
13				17								*30:19
13				17	18							*30:20
						19						*30:22
13				17			20					*30:23
13				17				21				*30:24
13											24	*30:25
13				17						23		*30:26
13									22			*30:27N
13												*30:28
13		15										*30:29
13				17				21				*30:30
13				17		19						*30:31
13					18							*30:32
13			16									*30:33
13							20					*30:34
13				17								*30:35
13				17					22			*30:36
13				17							24	*30:37
												*01:02, 01:20
												*01:07, 26:19, 29:14, 31:01:02-31:01:06, 31:02, 31:05-31:07, 31:09-31:31
												*02:52
												*02:185
												*03:42
												*03:43
				17								*03:72, 68:45
												*23:09
				17								*24:19
												*24:24, 24:67, 26:16, 32:07
												*24:66, 68:06
												*31:03-31:04
												*80:01-80:02
												HLA-A allele ⁴
13	14	15	16	17	18	19	20	21	22	23	24	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-A*30 subtyping.

In addition, wells number 2, 3, 8, 11, 15, 18, 20, 22 and 24 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, 3rd or 4th exons, 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, 3rd or 4th exons, 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 A*3005 allele has been shown to be identical to A*30:04.

A*3021 has been renamed A*30:11:02.

⁵Primer mix 5: Specific PCR fragment of 150 bp in the A*30:04, 30:06, 30:17 and 30:29 and the A*02:52, 24:66 and 68:06 alleles. Specific PCR fragment of 245 bp in the A*30:19 alleles.

Primer mix 6: Specific PCR fragment of 80 bp in the A*30:06 allele. Specific PCR fragment of 185 bp in the A*30:07 allele.

Primer mix 18: Specific PCR fragment of 130 bp in the A*30:32 allele. Specific PCR fragment of 210 bp in the A*30:20 allele.

Primer mix 19: Specific PCR fragment of 160 bp in the A*30:22 allele. Specific PCR fragment of 200 bp in the A*30:31 allele.

Primer mix 20: Specific PCR fragment of 210 bp in the A*30:23 allele. Specific PCR fragment of 275 bp in the A*30:34 allele.

Primer mix 21: Specific PCR fragment of 185 bp in the A*30:24 allele. Specific PCR fragment of 155 and 185 bp in the A*30:30 allele.

Primer mix 22: Specific PCR fragment of 170 bp in the A*30:36 allele. Specific PCR fragment of 215 bp in the A*30:27N allele.

Primer mix 24: Specific PCR fragment of 80 bp in the A*30:37 allele. Specific PCR fragment of 200 bp in the A*30:25 allele.

'w', might be weakly amplified.

[illegible]

CELL LINE VALIDATION SHEET													
HLA-A*30 SSP subtyping kit													
					Lot No.:	Well							
						17	18	19	20	21	22	23	24
						201070617	201070618	201070619	201070620	201070621	201189422	201189423	201189424
	IHCW cell line		A*	A*									
1	9001	SA	*24:02			-	-	-	-	-	-	-	-
2	9280	LK707	*02:01			-	-	-	-	-	-	-	-
3	9011	E4181324	*01:01			-	-	-	-	-	-	-	-
4	9275	GU373	*30:01			+	-	-	-	-	-	-	-
5	9009	KAS011	*01:01			-	-	-	-	-	-	-	-
6	9353	SM	*02:01	*26:03		-	-	-	-	-	-	-	-
7	9020	QBL	*26:01			-	-	-	-	-	-	-	-
8	9025	DEU	*31:01			-	-	-	-	-	-	-	-
9	9026	YAR	*26:01			-	-	-	-	-	-	-	-
10	9107	LKT3	*24:02			-	-	-	-	-	-	-	-
11	9051	PITOUT	*29:02			-	-	-	-	-	-	-	-
12	9052	DBB	*02:01			-	-	-	-	-	-	-	-
13	9004	JESTHOM	*02:01			-	-	-	-	-	-	-	-
14	9071	OLGA	*31:01			-	-	-	-	-	-	-	-
15	9075	DKB	*24:02			-	-	-	-	-	-	-	-
16	9037	SWEIG007	*29:02			-	-	-	-	-	-	-	-
17	9282	CTM3953540	*03:01	*80:01		-	-	-	-	-	-	-	-
18	9257	32367	*33:03	*74:01		-	-	-	-	-	-	-	-
19	9038	BM16	*02:01			-	-	-	-	-	-	-	-
20	9059	SLE005	*02:01			-	-	-	-	-	-	-	-
21	9064	AMALA	*02:17			-	-	-	-	-	-	-	-
22	9056	KOSE	*02:01			-	-	-	-	-	-	-	-
23	9124	IHL	*02:01	*34:01		-	-	-	-	-	-	-	-
24	9035	JBUSH	*32:01			-	-	-	-	-	-	-	-
25	9049	IBW9	*33:01			-	-	-	-	-	-	-	-
26	9285	WT49	*02:05			-	-	-	-	-	-	-	-
27	9191	CH1007	*24:10	*29:01		-	-	-	-	-	-	-	-
28	9320	BEL5GB	*02:01	*29:02		-	-	-	-	-	-	-	-
29	9050	MOU	*29:02			-	-	-	-	-	-	-	-
30	9021	RSH	*30:01	*68:02		+	-	-	-	-	-	-	-
31	9019	DUCAF	*30:02			-	-	-	-	-	-	-	-
32	9297	HAG	*02:01			-	-	-	-	-	-	-	-
33	9098	MT14B	*31:01			-	-	-	-	-	-	-	-
34	9104	DHIF	*31:01			-	-	-	-	-	-	-	-
35	9302	SSTO	*32:01			-	-	-	-	-	-	-	-
36	9024	KT17	*02:06	*11:01		-	-	-	-	-	-	-	-
37	9065	HHKB	*03:01			-	-	-	-	-	-	-	-
38	9099	LZL	*02:17			-	-	-	-	-	-	-	-
39	9315	CML	*01:01	*03:01		-	-	-	-	-	-	-	-
40	9134	WHONP199	*02:07	*30:01		+	-	-	-	-	-	-	-
41	9055	H0301	*03:01			-	-	-	-	-	-	-	-
42	9066	TAB089	*02:07			-	-	-	-	-	-	-	-
43	9076	T7526	*02:06	*02:07		-	-	-	-	-	-	-	-
44	9057	TEM	*66:01			-	-	-	-	-	-	-	-
45	9239	SHJO	*23:01	*24:02		-	-	-	-	-	-	-	-
46	9013	SCHU	*03:01			-	-	-	-	-	-	-	-
47	9045	TUBO	*02:16	*03:01		-	-	-	-	-	-	-	-
48	9303	TER-ND	*02:01	*11:01		-	-	-	-	-	-	-	-

CERTIFICATE OF ANALYSIS

Olerup SSP® HLA-A*30

Product number: 101.429-12u – without *Taq* polymerase
Lot number: 51M
Expiry date: 2014-February-01
Number of tests: 12
Number of wells per test: 24

Well specifications:

Well No.	Production No.	Well No.	Production No.
1	2010-706-01	13	2010-706-13
2	2010-706-02	14	2010-706-14
3	2010-706-03	15	2010-706-15
4	2010-706-04	16	2011-894-16
5	2010-706-05	17	2010-706-17
6	2010-706-06	18	2010-706-18
7	2011-894-07	19	2010-706-19
8	2010-706-08	20	2010-706-20
9	2010-706-09	21	2010-706-21
10	2010-706-10	22	2011-894-22
11	2010-706-11	23	2011-894-23
12	2011-894-12	24	2011-894-24

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 6 to 8, 14 to 16 and 18 to 24 were available. The specificities of the primers in primer 6, 7, 14 to 16, 19, 23 and 24 were tested by separately adding one additional 5'-primer, respectively one additional 3'-primer. In primer solutions 18, 20 and 22 it was only possible to test the 5'-primer, the 3'-primers were not possible to test. In primer solution 8 and 21 it was only possible to test the 3'-primers, the 5'-primers were not possible to test.

In primer solutions 7 and 16 one 5'-primer was not possible to test, and in primer solutions 2, 3, 5, 6, 12, 15, 16 and 19 one 3'-primer was not possible to test.

In addition, one 5'-primer in primer solution 5 was tested by adding an additional 3'-primer.

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

Date of approval: 2011-September-08

Approved by:

Quality Control, Supervisor

Lot No.: **51M**

Lot-specific information

www.olerup-ssp.com

Declaration of Conformity

Product name: Olerup SSP® HLA-A*30
Product number: 101.429-12u
Lot number: 51M

Intended use: HLA-A*30 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 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.

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.)

Stockholm, Sweden
2011-September-08

Ann-Cathrin Jareman
Head of QA and Regulatory Affairs

Lot No.: **51M**

Lot-specific information

www.olerup-ssp.com

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**.