101.613-12 – including *Taq* polymerase, IFU-01
101.613-12u – without *Taq* polymerase, IFU-02

Visit www.olerup-ssp.com for **"Instructions for Use" (IFU)**

Lot No.: 02Y

Lot-specific information

Olerup SSP® HLA-C*05

Product number:	**101.613-12 – including *Taq* polymerase** **101.613-12u – without *Taq* polymerase**
Lot number:	**02Y**
Expiry date:	**2017-October-01**
Number of tests:	**12**
Number of wells per test:	**31+1**
Storage - pre-aliquoted primers:	**dark at -20ºC**
- PCR Master Mix:	**-20ºC**
- Adhesive PCR seals	**RT**
- Product Insert	**RT**

This Product Description is only valid for Lot No. 02Y.

Complete product documentation consists of generic Instructions for Use (IFU), lot specific Product Insert, Worksheet and Certificate.

CHANGES COMPARED TO THE PREVIOUS *OLERUP* SSP® HLA-C*05 LOT (24V)

The HLA-C*05 kit is updated for new alleles to enable separation of:

- Confirmed[1] 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

A well containing Negative Control primer pairs has been added.

The format of the Product Insert and Worksheet have been changed.

[1]As described in section Uniquely Identified Alleles.

The HLA-C*05 primer set, specificity and interpretation tables have been updated for the HLA-C alleles described since the previous *Olerup* SSP® HLA-C*05 lot was made **(Lot No. 24V)**. The kit design is based on IMGT/HLA database 3.19.0.

As of lot series V, the Specificity Table is included in the lot-specific Product Insert, and the Interpretation Table is included in the Worksheet.

101.613-12 – including *Taq* polymerase, IFU-01
101.613-12u – without *Taq* polymerase, IFU-02

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Lot No.: 02Y **Lot-specific information**

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	Added	-	5'-primer added for the C*05:01:28 allele.
4	-	Added	3'-primer added for the C*05:108 allele.
6	Added	-	5'-primer added for the C*05:99 allele.
9	Added	-	5'-primer added for the C*05:113N allele.
14	-	Added	3'-primer added for the C*05:80 allele.
15	-	Added	3'-primer added for the C*05:65 allele.
18	Added	-	5'-primer added for the C*05:99N allele.
25	-	Added	3'-primer added for the C*05:80 allele.
26	Added	-	5'-primer added for the C*05:113N allele.
28	-	Added	3'-primer added for the C*05:65 allele.
30	-	Added	3'-primer added for the C*05:108 allele.
32	-	-	Updated negative control.

Changes in revision R01 compared to R00:

1. Primer mixes 4 and 30 do not amplify the C*05:108 and the C*06:111, C*07:316 and C*07:359 alleles. This has been corrected in the Specificity and Interpretation Tables.

Well **32** contains Negative Control primer pairs, that will amplify more than 95% of the *Olerup* SSP® HLA Class I, DRB, DQB1, DPB1 and DQA1 amplicons as well as all the amplicons generated by the control primer pairs matching the human growth hormone gene.

HLA-specific PCR product sizes range from 75 to 200 base pairs.
The PCR product generated by the positive control primer pair is 430 base pairs.

Length of PCR product	**105**	**200**	**105**	**80**	**75**	**80**	**85**
5'-primer[1]	**164**	**340**	**440**	**45**	**45**	**43**	**36**
	5'-CAC3'	**5'-Agg3'**	**5'-TTA3'**	**5'-Tgg3'**	**5'-Tgg3'**	**5'-Tgg3'**	**5'-TAC3'**
							36
							5'-TAT3'
3'-primer[2]	**231**	**2nd I**	**507**	**59**	**58**	**57**	**47**
	5'-TgC3'	**5'-AAA3'**	**5'-TTg3'**	**5'-CTC3'**	**5'-ggC3'**	**5'-CTC3'**	**5'-ACA3'**
							48
							5'-gCA3'
							48
							5'-gCC3'
							52
							5'-TgT3'
A*	+	+	+				
B*	+	+	+				
C*	+	+	+				
DRB1				+	+		
DRB3				+	+		
DRB5				+			
DQB1					+		
DPB1						+	
DQA1							+

[1]The nucleotide position for HLA class I genes and the codon for HLA class II genes, in the 2nd or 3rd exon, matching the specificity-determining 3'-end of the primer is given. Nucleotide and codonnumbering as on the www.ebi.ac.uk/imgt/hla web site. The sequence of the 3 terminal nucleotides of the primer is given.

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

101.613-12 – including *Taq* polymerase, IFU-01
101.613-12u – without *Taq* polymerase, IFU-02

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Lot No.: 02Y **Lot-specific information**

PRODUCT DESCRIPTION

HLA-C*05 SSP subtyping

CONTENT

The primer set contains 5'- and 3'-primers for identifying the C*05:01 to C*05:114 alleles.

PLATE LAYOUT

Each test consists of 32 PCR reactions in a 32 well 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**
25	**26**	**27**	**28**	**29**	**30**	**31**	**NC**

The 32 well cut PCR plate is marked with 'HLA-C*05' in silver/gray ink.
Well No. 1 is marked with the Lot No. '02Y'.
Wells 1 to 31 – HLA-C*05 high resolution primers.
Well 32 – Negative Control (NC).
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 32 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

Due to the sharing of sequence motifs between HLA-C alleles, non-HLA-C*05 alleles will be amplified by primer mixes 1 to 7, 10 to 13, 15 to 17, 19, 21 to 24 and 26 to 31. In addition, a few HLA-A and HLA-B alleles will be amplified by primer mixes 3, 5, 6, 10 to 12, 17, 21, 26 and 31.
For further details see Specificity Table.

UNIQUELY IDENTIFIED ALLELES

All the HLA-C*05 alleles, i.e. **C*05:01 to C*05:114**, recognized by the HLA Nomenclature Committee in January 2015[1] will be amplified by the primers in the HLA-C*05 subtyping kit[2,3].

The HLA-C*05 kit enables separation of the confirmed HLA-C*05 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-C*05 alleles is listed below.

Lot No.: 02Y **Lot-specific information**

The HLA-C*05 kit also enables identification of polymorphisms in exons outside of the region encoding the peptide binding domain and of null and alternatively expressed alleles.

The following HLA-C*05 alleles can be distinguished by the different sizes of the HLA-specific PCR product:

Alleles	Primer mix	Alleles	Primer mix
C*05:03, 05:07N	4	C*05:24, 05:36	23
C*05:06, 05:55	8	C*05:25, 05:33	11
C*05:08, 05:30	10	C*05:28, 05:39	26
C*05:14, 05:93	16	C*05:32, 05:53	30
C*05:15, 05:91N	14	C*05:35, 05:40	25
C*05:21, 05:26	20	C*05:37, 05:41	29

The HLA-C*05 subtyping kit cannot distinguish the following silent mutations: the C*05:01:01:01-05:01:31, the C*05:04:01-05:04:02, the C*05:09:01-05:09:03, the C*05:18:02-05:18:03, the C*05:22:01-05:22:02, the C*05:29:01-05:29:02 and the C*05:44:01-05:44:02 alleles.

[1]HLA-C alleles listed on the IMGT/HLA web page 2015-January-19, release 3.19.0, www.ebi.ac.uk/imgt/hla.
[2]Alleles that have been deleted from or renamed in the official WHO HLA Nomenclature up to and including the last IMGT/HLA database release can be retrieved from web page http://hla.alleles.org/alleles/deleted.html.
[3]The C*05:18:01 and C*08:10 alleles, the C*05:29:01-05:29:02 and C*08:25 and C*08:94 alleles and the C*05:92N and C*08:55N alleles will give rise to identical amplification patterns with the HLA-C*05 subtyping kit. These alleles can be distinguished by the HLA-C low resolution kit and/or the C*08 subtyping kit.

101.613-12 – including *Taq* polymerase, IFU-01
101.613-12u – without *Taq* polymerase, IFU-02

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Lot No.: 02Y **Lot-specific information**

ALLELE CONFIRMATION STATUS

Allele	Status[1]	Allele	Status[1]	Allele	Status[1]	Allele	Status[1]
C*05:01:01:01	**Confirmed**	C*05:09:02	Unconfirmed	C*05:44:01	Unconfirmed	C*05:81	Unconfirmed
C*05:01:01:02	**Confirmed**	C*05:09:03	Unconfirmed	C*05:44:02	Unconfirmed	C*05:82	Unconfirmed
C*05:01:02	Unconfirmed	**C*05:10**	**Confirmed**	C*05:45	Unconfirmed	C*05:83	Unconfirmed
C*05:01:03	Unconfirmed	C*05:11	Unconfirmed	**C*05:46**	**Confirmed**	C*05:84	Unconfirmed
C*05:01:04	Unconfirmed	C*05:12	Unconfirmed	C*05:47	Unconfirmed	C*05:85	Unconfirmed
C*05:01:05	**Confirmed**	**C*05:13**	**Confirmed**	C*05:48N	Unconfirmed	C*05:86	Unconfirmed
C*05:01:06	Unconfirmed	**C*05:14**	**Confirmed**	C*05:49	Unconfirmed	C*05:87	Unconfirmed
C*05:01:07	**Confirmed**	C*05:15	Unconfirmed	C*05:50	Unconfirmed	C*05:88	Unconfirmed
C*05:01:08	**Confirmed**	C*05:16	Unconfirmed	C*05:51Q	Unconfirmed	C*05:89	Unconfirmed
C*05:01:09	Unconfirmed	C*05:17	Unconfirmed	C*05:52	Unconfirmed	C*05:90	Unconfirmed
C*05:01:10	Unconfirmed	**C*05:18:01**	**Confirmed**	C*05:53	Unconfirmed	C*05:91N	Unconfirmed
C*05:01:11	**Confirmed**	C*05:18:02	Unconfirmed	C*05:54	Unconfirmed	C*05:92N	Unconfirmed
C*05:01:12	**Confirmed**	C*05:18:03	Unconfirmed	**C*05:55**	**Confirmed**	C*05:93	Unconfirmed
C*05:01:13	Unconfirmed	**C*05:19**	**Confirmed**	C*05:56	Unconfirmed	C*05:94	Unconfirmed
C*05:01:14	Unconfirmed	C*05:20	Unconfirmed	C*05:57	Unconfirmed	C*05:95	Unconfirmed
C*05:01:15	Unconfirmed	C*05:21	Unconfirmed	C*05:58:01	Unconfirmed	C*05:96	Unconfirmed
C*05:01:16	**Confirmed**	**C*05:22:01**	**Confirmed**	C*05:58:02	Unconfirmed	C*05:97	Unconfirmed
C*05:01:17	**Confirmed**	**C*05:22:02**	**Confirmed**	C*05:59	Unconfirmed	C*05:98	Unconfirmed
C*05:01:18	**Confirmed**	C*05:23	Unconfirmed	C*05:60	Unconfirmed	C*05:99N	Unconfirmed
C*05:01:19	**Confirmed**	**C*05:24**	**Confirmed**	C*05:61	Unconfirmed	C*05:100	Unconfirmed
C*05:01:20	**Confirmed**	C*05:25	Unconfirmed	C*05:62	Unconfirmed	C*05:101	Unconfirmed
C*05:01:21	Unconfirmed	**C*05:26**	**Confirmed**	C*05:63	Unconfirmed	C*05:102	Unconfirmed
C*05:01:22	Unconfirmed	**C*05:27**	**Confirmed**	C*05:64:01	Unconfirmed	C*05:103	Unconfirmed
C*05:01:23	Unconfirmed	**C*05:28**	**Confirmed**	C*05:64:02	Unconfirmed	C*05:104	Unconfirmed
C*05:01:24	**Confirmed**	**C*05:29:01**	**Confirmed**	**C*05:65**	**Confirmed**	C*05:105	Unconfirmed
C*05:01:25	Unconfirmed	C*05:29:02	Unconfirmed	C*05:66	Unconfirmed	C*05:106	Unconfirmed
C*05:01:26	Unconfirmed	C*05:30	Unconfirmed	C*05:67	Unconfirmed	C*05:107	Unconfirmed
C*05:01:27	Unconfirmed	C*05:31	Unconfirmed	C*05:68	Unconfirmed	C*05:108	Unconfirmed
C*05:01:28	Unconfirmed	**C*05:32**	**Confirmed**	C*05:69	Unconfirmed	C*05:109	Unconfirmed
C*05:01:29	**Confirmed**	**C*05:33**	**Confirmed**	C*05:70	Unconfirmed	C*05:110	Unconfirmed
C*05:01:30	Unconfirmed	**C*05:34**	**Confirmed**	C*05:71	Unconfirmed	C*05:111	Unconfirmed
C*05:01:31	Unconfirmed	**C*05:35**	**Confirmed**	C*05:72	Unconfirmed	C*05:112	Unconfirmed
C*05:03	Unconfirmed	**C*05:36**	**Confirmed**	C*05:73	Unconfirmed	C*05:113N	Unconfirmed
C*05:04:01	**Confirmed**	**C*05:37**	**Confirmed**	C*05:74	Unconfirmed	C*05:114	Unconfirmed
C*05:04:02	Unconfirmed	C*05:38	Unconfirmed	C*05:75	Unconfirmed		
C*05:05	Unconfirmed	**C*05:39**	**Confirmed**	C*05:76	Unconfirmed		
C*05:06	**Confirmed**	C*05:40	Unconfirmed	C*05:77	Unconfirmed		
C*05:07N	**Confirmed**	C*05:41	Unconfirmed	C*05:78	Unconfirmed		
C*05:08	**Confirmed**	**C*05:42**	**Confirmed**	C*05:79	Unconfirmed		
C*05:09:01	**Confirmed**	C*05:43	Unconfirmed	**C*05:80**	**Confirmed**		

[1]Allele status "confirmed" or "unconfirmed" as listed on the IMGT/HLA web page 2015-January-19, release 3.19.0, www.ebi.ac.uk/imgt/hla.

RESOLUTION IN HOMO- AND HETEROZYGOTES

Results file with resolution in HLA-C*05 homo- and heterozygotes is available upon request.

101.613-12 – including *Taq* polymerase, IFU-01
101.613-12u – without *Taq* polymerase, IFU-02

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Lot No.: 02Y **Lot-specific information**

SPECIFICITY TABLE

HLA-C*05 SSP subtyping

Specificities and sizes of the PCR products of the 31+1 primer mixes used for HLA-C*05 SSP subtyping

Primer Mix	Size of spec. PCR product[1]	Size of control band[2]	Amplified HLA-C*05 alleles[3]	Other amplified HLA Class I alleles[4]
1	155 bp	**800 bp**	*05:01:01:01-05:01:31, 05:03-05:08, 05:10-05:11, 05:13-05:16, 05:19-05:51Q, 05:53-05:86, 05:88-05:96, 05:98-05:102, 05:104-05:105, 05:108-05:113N	*07:41, 08:02:01:01-08:02:12, 08:04:01-08:05, 08:07, 08:12-08:13, 08:17-08:19, 08:23, 08:25, 08:28-08:32, 08:34, 08:37, 08:43, 08:45, 08:47-08:49, 08:52N-08:53, 08:55N, 08:57, 08:62-08:63, 08:67-08:71, 08:73-08:77, 08:90, 08:92-08:94, 08:100, 08:103-08:104, 08:107-08:108, 08:110-08:115
2	165 bp	1070 bp	*05:01:01:01-05:01:31, 05:03-05:28, 05:30-05:47, 05:49-05:91N, 05:93-05:114	*02:94, 04:129, 06:05, 06:67, 08:10, 12:21, 12:33, 17:05
3	150 bp	1070 bp	*05:09:01-05:09:03, 05:17, 05:44:01-05:44:02, 05:52, 05:79	*01:13, 02:51, 03:87:01-03:87:02, 07:130, 08:15:01-08:15:02, 08:51, 12:144, 16:27, **B*15:33, B*15:248**
4[5]	120 bp 285 bp 310 bp	**800 bp**	*05:03 *05:07N	*07:52
5	225 bp 285 bp	1070 bp	*05:04:01-05:04:02, 05:103 *05:31	*06:129, 07:68, 07:260, 07:302, 08:09, 08:11, 08:59, 08:113, **B*15:337, B*18:83**
6	255 bp 280 bp	1070 bp	*05:05, 05:99N *05:16, 05:85, 05:107	*03:251, 08:62, 08:82, **A*02:425, A*02:519, A*29:10, A*68:69, B*14:32, B*15:337, B*18:83, B*44:148** ***06:129, 07:364, 08:12, **B*14:32**
7	265 bp	1070 bp	*05:01:01:01-05:01:31, 05:03-05:04:02, 05:06-05:08, 05:10-05:16, 05:18:01-05:51Q, 05:53-05:60, 05:62-05:81, 05:83-05:84, 05:86-05:106, 05:108-05:114	*04:120, 06:129, 07:04:01-07:04:10, 07:11-07:12, 07:41, 07:63, 07:68, 07:101, 07:139, 07:142, 07:181, 07:199:01-07:199:02, 07:260, 07:272, 07:302, 07:323-07:324, 07:328-07:329N, 07:336, 07:338, 07:354-07:355, 07:357-07:358, 07:361, 07:365, 07:378, 07:394-07:395, 07:403, 07:406, 08:01:01-08:01:13, 08:01:15-08:08:01, 08:09-08:14, 08:16:01-08:50, 08:52N-08:61,

				08:63, 08:65-08:69, 08:71, 08:73-08:81, 08:83-08:115
8[5]	85 bp 210 bp	**800 bp**	*05:06 *05:55	
9[5]	105 bp 175 bp 245 bp	1070 bp	*05:51Q *05:48N *05:113N	
10[5]	95 bp 250 bp 320 bp	1070 bp	*05:08, 05:52, 05:89 *05:30 *05:92N	*02:51, 08:29, 08:31, 12:144, **B*15:33, B*15:248** *03:247, 06:125 *08:55N
11[5]	115 bp 205 bp	1070 bp	*05:33 *05:25, 05:42	*04:129, 06:05, 06:67, 07:101, 07:148, 07:161, 08:28, **A*11:166, A*80:01:01:01[w]-80:03[w]**
12[6]	155 bp 225 bp 285 bp	**800 bp**	*05:97 *05:38 *05:10	 *08:97 *03:251, 08:44, 08:61, 08:82, **B*44:148**
13[5]	95 bp	1070 bp	*05:11, 05:17, 05:27, 05:68, 05:70, 05:79	*03:87:01-03:87:02, 07:130, 08:04:01-08:04:03, 08:13, 08:57, 08:93, 08:104, 08:113
14[5]	120 bp 200 bp	1070 bp	*05:12, 05:15 *05:80, 05:91N	
15[5]	115 bp 185 bp 240 bp	1070 bp	*05:65 *05:34 *05:13	*04:96 *02:93, 06:13
16	195 bp 470 bp	1070 bp	*05:14 *05:93	 *03:171, 03:211:01, 04:144, 06:73, 08:20, 08:40, 12:109
17	155 bp	1070 bp	*05:12, 05:18:01	*03:251, 04:120, 06:129, 08:01:01-08:01:18, 08:03:01-08:03:03, 08:06, 08:08:01-08:11, 08:14, 08:16:01-08:16:02, 08:20-08:22, 08:24, 08:26N-08:27, 08:33:02-08:33:03, 08:35-08:36N, 08:38-08:42, 08:44, 08:46, 08:50, 08:54, 08:56, 08:58-08:61, 08:65-08:66, 08:78-08:89N, 08:91, 08:95-08:99, 08:101-08:102, 08:105-08:106, 08:109, **B*15:337**
18	160 bp 245 bp	1070 bp	*05:87 *05:19, 05:99N	
19	175 bp	**800 bp**	*05:20	*01:43, 02:87, 07:101, 07:148, 07:161, 08:01:01-08:09, 08:11-08:12, 08:14-08:15:02, 08:17, 08:19-08:24, 08:26N-08:54, 08:56-08:63, 08:65-08:93, 08:95-08:110, 08:112-08:115,

101.613-12 – including *Taq* polymerase, IFU-01
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Lot No.: 02Y **Lot-specific information**

				12:127
20	260 bp 390 bp	1070 bp	*05:26 *05:21	
21	230 bp	1070 bp	*05:04:01-05:04:02, 05:22:01-05:22:02, 05:38, 05:103, 05:107	*03:251, 06:129, 07:04:01-07:04:10, 07:11-07:12, 07:63, 07:68, 07:101, 07:139, 07:142, 07:181, 07:199:01-07:199:02, 07:260, 07:272, 07:302, 07:323-07:324, 07:328-07:329N, 07:336, 07:338, 07:354-07:355, 07:357-07:358, 07:361, 07:364-07:365, 07:378, 07:394-07:395, 07:403, 07:406, 08:09, 08:11, 08:83, 08:97, 08:108, **B*44:148**
22[5]	100 bp	**800 bp**	*05:23, 05:62	*07:02:35, 08:07, 08:47, 08:104
23[5]	85 bp 135 bp	1070 bp	*05:24 *05:36	*07:148
24	185 bp 265 bp	1070 bp	*05:43 *05:29:01-05:29:02	*08:37 *08:13, 08:16:01, 08:25, 08:94
25[5]	105 bp 205 bp	1070 bp	*05:40 *05:35, 05:80	
26[5]	115 bp 185 bp 245 bp	1070 bp	*05:27, 05:39 *05:28 *05:113N	*03:87:01-03:87:02, 08:115, **B*15:33, B*15:248** *06:64
27	155 bp	1070 bp	*05:42, 05:46	*04:129, 06:67, 07:101, 07:148, 07:161, 08:05, 08:21, 12:127, 17:05
28[5,7]	115 bp 200 bp	**800 bp**	*05:65 *05:45	*04:96
29	140 bp 185 bp 260 bp	1070 bp	*05:41 *05:34 *05:37	*04:78 *02:93, 06:13
30[5]	70 bp 175 bp 315 bp	1070 bp	*05:53 *05:32	
31	155 bp	1070 bp	*05:18:02-05:18:03, 05:103, 05:107	*07:04:01-07:04:10, 07:11-07:12, 07:63, 07:68, 07:101, 07:139, 07:142, 07:181, 07:199:01-07:199:02, 07:260, 07:272, 07:302, 07:323-07:324, 07:328-07:329N, 07:336, 07:338, 07:354-07:355, 07:357-07:358, 07:361, 07:364-07:365, 07:378, 07:394-07:395, 07:403, 07:406, 08:33:01, **B*14:32, B*18:83, B*44:148**
32[8]	-	-	**Negative Control**	

[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 C*05 high resolution 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 alleles listed are specified according to amplicon length.

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 inherit 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 internal positive control bands are 1070 or 800 base pairs respectively, well distribution as outlined in the table. Well number 1 contains the shorter, 800 bp, internal positive control band. The well distribution of the internal controls can help in orientation of the kit on gel photo, as well as allow for kit identification. In the presence of a specific amplification the intensity of the control band often decreases.

[3]For several HLA Class I alleles 1st and/or 4th exon(s) and beyond, as well as intron nucleotide sequences, are not available. In these instances it is not known whether some of the primers of the SSP sets are completely matched with the target sequences or not. Assumption is made that unknown sequences in these regions are conserved within allelic groups.

[4]Due to the sharing of sequence motifs between HLA-C alleles, non-HLA-C*05 alleles will be amplified by primer mixes 1 to 7, 10 to 13, 15 to 17, 19, 21 to 24 and 26 to 31. In addition, a few HLA-A and HLA-B alleles will be amplified by primer mixes 3, 5, 6, 10 to 12, 17, 21, 26 and 31.

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

[6]Primer mix 12 may have tendencies of unspecific amplifications.

[7]Primer mix 28 may have a tendency to giving rise to primer oligomer formation.

[8]Primer mix 32 contains a negative control, which will amplify more than 95% of HLA amplicons as well as the amplicons generated by the control primer pairs matching the human growth hormone gene. HLA-specific PCR product sizes range from 75 to 200 base pairs and the PCR product generated by the HGH positive control primer pair is 430 base pairs.

'w', might be weakly amplified.

101.613-12 – including *Taq* polymerase, IFU-01
101.613-12u – without *Taq* polymerase, IFU-02

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Lot No.: 02Y

Lot-specific information

PRIMER SPECIFICATION

Well No.	1	2	3	4	5	6	7	8	9	10	11	12
Length of spec.	155	165	150	120	225	255	265	85	105	95	115	155
PCR product				285	285	280		210	175	250	205	225
				310					245	320		285
Length of int.	800	1070	1070	800	1070	1070	1070	800	1070	1070	1070	800
pos. control[1]												
5'-primer(s)[2]	485	176	485	355	355	361	379	176	96	28	176	355
	5' -CAA 3'	5' -gCA 3'	5' -CAA 3'	5' -.CC 3'	5' -TCC 3'	5' -AgT 3'	5' -ACC 3'	5' -gCA 3'	5' -.TC 3'	5' -TCA 3'	5' -gCA 3'	5' -TCA 3'
		176		3rd I	419	379			166	485		416
		5' -gCA 3'		5' -Cgg 3'	5' -gTC 3'	5' -ACg 3'			5' -CgT 3'	5' -CAA 3'		5' -CCg 3'
						385			485			485
						5' -g.C 3'			5' -CAA 3'			5' -CAg 3'
3'-primer(s)[3]	601	302	595	601	601	601	601	221	302	106	248	601
	5' -CTT 3'	5' -ggT 3'	5' -CCT 3'	5' -CTT 3'	5' -CTT 3'	5' -CTT 3'	5' -CTT 3'	5' -ACC 3'	5' -ggT 3'	5' -CAT 3'	5' -AAC 3'	5' -CTT 3'
			595	668				343	550	175	341	
			5' -CCg 3'	5' -TgA 3'				5' -T 3'	5' -CAg 3'	5' -CTA 3'	5' -CgT 3'	
			601	862						538		
			5' -CTC 3'	5' -CTT 3'						5' -CCA 3'		
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

Well No.	13	14	15	16	17	18	19	20	21	22	23	24
Length of spec.	95	120	115	195	155	160	175	260	230	100	85	185
PCR product		200	185	470		245		390			135	265
			240									
Length of int.	1070	1070	1070	1070	1070	1070	800	1070	1070	800	1070	1070
pos. control[1]												
5'-primer(s)[2]	485	176	115	446	486	385	176	28	412	453	176	1st I
	5' -CAA 3'	5' -gCA 3'	5' -ggA 3'	5' -CgT 3'	5' -ACg 3'	5' -g.C 3'	5' -gCA 3'	5' -TCA 3'	5' -ATg 3'	5' -AAT 3'	5' -gCA 3'	5' -CgA 3'
		453	2nd I	652		406		322	416			
		5' -AAT 3'	5' -CCA 3'	5' -CCA 3'		5' -gCC 3'		5' -gCC 3'	5' -CCg 3'			
						483						
						5' -gAg 3'						
3'-primer(s)[3]	538	256	312	601	601	601	311	118	601	512	218	97
	5' -CAg 3'	5' -CCA 3'	5' -AgT 3'	5' -CTT 3'	5' -CTT 3'	5' -CTT 3'	5' -ggT 3'	5' -gCT 3'	5' -CTT 3'	5' -CCA 3'	5' -gCC 3'	5' -gTC 3'
		337	403	956				419			270	175
		5' -CTA 3'	5' -gCA 3'	5' -CAg 3'				5' -CgA 3'			5' -TAg 3'	5' -CCg 3'
		337	475									
		5' -CTT 3'	5' -ggT 3'									
		527										
		5' -CCA 3'										
Well No.	13	14	15	16	17	18	19	20	21	22	23	24

101.613-12 – including *Taq* polymerase, IFU-01
101.613-12u – without *Taq* polymerase, IFU-02

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Lot No.: 02Y **Lot-specific information**

Well No.	25	26	27	28	29	30	31
Length of spec.	105	115	155	115	140	70	155
PCR product	205	185		200	185	175	
		245			260	315	
Length of int.	1070	1070	1070	800	1070	1070	1070
pos. control[1]							
5'-primer(s)[2]	176	96	176	2nd I	2nd I	176	486
	5' -gCA 3'	5' -.TC 3'	5' -gCA 3'	5' -CCA 3'	5' -CCA 3'	5' -gCA 3'	5' -ACC 3'
		158			629	3rd I	
		5' -ggg 3'			5' -AAg 3'	5' -Cgg 3'	
		485					
		5' -CAA 3'					
3'-primer(s)[3]	241	302	289	403	430	311	601
	5' -CgA 3'	5' -ggT 3'	5' -AgC 3'	5' -gCA 3'	5' -gCA 3'	5' -gTC 3'	5' -CTT 3'
	337	559	289	488	475	621	
	5' -CTT 3'	5' -CAg 3'	5' -AgC 3'	5' -CCT 3'	5' -ggT 3'	5' -Tgg 3'	
	341				846	862	
	5' -CgA 3'				5' -CAC 3'	5' -CTT 3'	
Well No.	25	26	27	28	29	30	31

[1]The internal positive control primer pairs amplify segments of the human growth hormone gene. The internal positive control bands are 1070 or 800 base pairs respectively, well distribution as outlined in the table. Well number 1 contains the shorter, 800 bp, internal positive control band. The well distribution of the internal controls can help in orientation of the kit on gel photo, as well as allow for kit identification. In the presence of a specific amplification the intensity of the control band often decreases.

[2]The nucleotide position 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.

[3]The nucleotide position 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.

101.613-12 – including *Taq* polymerase, IFU-01
101.613-12u – without *Taq* polymerase, IFU-02

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Lot No.: 02Y **Lot-specific information**

CELL LINE VALIDATION SHEET

HLA-C*05 SSP subtyping kit

					Well[2]															
					1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
				Prod. No.:	201448501	201448502	201448503	201448504	201448505	201448506	201448507	201448508	201448509	201448510	201448511	201448512	201448513	201448514	201448515	201448516
	IHWC cell line[1]		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	9025	DEU	*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	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

101.613-12 – including *Taq* polymerase, IFU-01
101.613-12u – without *Taq* polymerase, IFU-02

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Lot No.: 02Y **Lot-specific information**

CELL LINE VALIDATION SHEET

HLA-C*05 SSP subtyping kit

					Well[2]													
					17	18	19	20	21	22	23	24	25	26	27	28	29	30	31
				Prod. No.:	201448517	201448518	201448519	201448520	201448521	201448522	201448523	201448524	201448525	201448526	201448527	201448528	201448529	201448530	201448531
	IHWC cell line[1]		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	9025	DEU	*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	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

[1]The provided cell line HLA specificities are retrieved from the http://www.ihwg.org/hla web site. The specificity of an individual cell line may thus be subject to change.

101.613-12 – including *Taq* polymerase, IFU-01
101.613-12u – without *Taq* polymerase, IFU-02

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Lot No.: 02Y **Lot-specific information**

[2]The specificity of each primer solution in the kit has been tested against 48 well characterized cell line DNAs and where applicable, additional cell line DNAs.
No DNAs carrying the alleles to be amplified by primer solutions 4 to 6, 8 to 12, 14 to 16, 18, 20 and 22 to 30 were available. The specificities of the primers in primer solutions 4 to 6, 8 to 12, 14, 15, 18, 20, 22 to 24, 26, 27 and 29 were tested by separately adding additional 5'-primers respectively 3'-primers.
In primer solutions 25, 28 and 30 it was only possible to test the 5'-primers, the 3'-primers were not possible to test. In primer solution 16 it was only possible to test the 3'-primer, the 5'-primer was not possible to test. In primer solutions 3, 4, 8 to 11, 14, 15, 20, 23 and 29 one or three 3'-primers were not possible to test. In primer solutions 2, 4, 6, 9, 12, 15, 18, 20, 21, 26 and 29 one or two 5'-primers were not possible to test. One additional 3'-primer in primer solution 3 was tested by separately adding one 5'-primer.

101.613-12 – including *Taq* polymerase, IFU-01
101.613-12u – without *Taq* polymerase, IFU-02

Visit www.olerup-ssp.com for **"Instructions for Use" (IFU)**

Lot No.: 02Y **Lot-specific information**

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