

101.623-12 – including *Taq* pol., IFU-01  
101.623-12u– without *Taq* pol., IFU-02

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“Instructions for Use” (IFU)

Lot No.: **72V**

Lot-specific information  
**Olerup SSP® HLA-C\*08**

<b>Product number:</b>	<b>101.623-12 – including <i>Taq</i> polymerase 101.623-12u – without <i>Taq</i> polymerase</b>
<b>Lot number:</b>	<b>72V</b>
<b>Expiry date:</b>	<b>2016-November-01</b>
<b>Number of tests:</b>	<b>12</b>
<b>Number of wells per test:</b>	<b>31+1</b>
<b>Storage - pre-aliquoted primers:</b>	<b>dark at -20°C</b>
- PCR Master Mix:	-20°C
- Adhesive PCR seals	RT
- Product Insert	RT

**This Product Description is only valid for Lot No. 72V.**

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\*08 LOT (13S)**

The HLA-C\*08 kit is updated for new alleles to enable separation of:

- Confirmed<sup>1</sup> 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.

<sup>1</sup>As described in section Uniquely Identified Alleles.

The HLA-C\*08 specificity and interpretation tables have been updated for the HLA-C alleles described since the previous *Olerup SSP®* HLA-C\*08 lot was made (**Lot No. 13S**).

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.

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

Well	5'-primer	3'-primer	rationale
6	Added	Added	Primer pair added for the C*08:102 allele.
7	Added	Added	Primer pair added for the C*08:87 allele.
9	Added	-	5'-primers added for the C*08:75 and C*08:88N alleles.
11	Added	Added	Primer pair added for the C*08:86 alleles.
15	-	Modified	3'-primer modified for improved HLA-specific amplification.
20	-	Added	3'-primer added from well 27.
23	Added	Added	Primer pairs added for the C*08:70Q and C*08:78 alleles.
24	Added	-	5'-primers added for the C*08:83 and C*08:89N alleles.
27	Moved, added	Moved, added	Primer pair moved to well 20, primer pairs added from well 32.
28	Added	Added	Primer pair added for the C*08:53 alleles.
30	Added	-	5'-primer added for the C*08:99 allele.
32	Moved	Moved	Primer pairs moved to well 27, Negative Control.

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## PRODUCT DESCRIPTION

### HLA-C\*08 SSP typing

#### CONTENT

The primer set contains 5'- and 3'-primers for identifying the C\*08:01 to C\*08:102 alleles.

#### PLATE LAYOUT

Each HLA-C\*08 test consists of 32 PCR reactions in a 32 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
25	26	27	28	29	30	31	NC

The 32 well PCR plate is marked with ‘HLA-C\*08’ in silver/gray ink.

Well No. 1 is marked with the Lot No. ‘72V’.

Wells 1 to 31– HLA-C\*08 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 24 well PCR plate, make sure that the remaining plates stay sealed. Use a scalpel or a similar instrument to carefully cut the foil between the plates.

#### INTERPRETATION

Due to the sharing of sequence motifs between HLA-C alleles non-HLA-C\*08 alleles will be amplified by primer mixes 1 to 9, 11 to 13, 15 to 17, 19 to 25, 27, 28, 30, 31 amplify non-HLA-C\*08 alleles. In addition, a few HLA-A and HLA-B alleles will be amplified by primer mixes 1, 3, 5, 8, 11 16, 20, 22, 25, 28 and 30.

For further details see Specificity Table.

#### UNIQUELY IDENTIFIED ALLELES

All the HLA-C\*08 alleles, i.e. **C\*08:01 to C\*08:102**, recognized by the HLA Nomenclature Committee in January 2014<sup>1</sup> will be amplified by the primers in the HLA-C\*08 SSP kit<sup>2,3</sup>.

The HLA-C\*08 kit enables separation of the confirmed HLA-C\*08 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\*08 alleles is listed below.

The HLA-C\*08 kit also enables identification of polymorphisms in exons outside of the region encoding the peptide binding domain and of null and alternatively expressed alleles.

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**Lot-specific information**

The HLA-C\*08 primer set cannot distinguish the following silent mutations: the C\*08:01:01-08:01:06 and C\*08:01:08-08:01:13 alleles, the C\*08:02:01-08:02:05 and C\*08:02:08-08:02:10 alleles, the 08:03:01-08:03:03 alleles, the C\*08:04:01-08:04:03 alleles, the C\*08:08:01-08:08:02 alleles, the 08:15:01-08:15:02 alleles, the C\*08:16:01-08:16:02 alleles or the C\*08:72:01-08:72:02 alleles.

The following HLA-C\*08 alleles can be distinguished by the different sizes of the HLA-specific PCR product:

Alleles	Primer mix	Alleles	Primer mix
C*08:01:07, 08:44	28	C*08:30, 08:32	19
C*08:20, 08:99	30	C*08:36N, 08:78	23
C*08:22, 08:80	16	C*08:55N, 05:64:02	27
C*08:24, 08:88N	9		

<sup>1</sup>HLA-C alleles listed on the IMGT/HLA web page 2014-January-17, release 3.15.0, [www.ebi.ac.uk/imgt/hla](http://www.ebi.ac.uk/imgt/hla).

<sup>2</sup>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>.

<sup>3</sup>The HLA-C\*08 primer set cannot separate the 08:15:01-08:15:02 and 08:51 alleles from the C\*07:148 and 07:161. These alleles can be distinguished by the HLA-C low resolution kit and/or the HLA-C\*07 high resolution kit.

The HLA-C\*08 primer set cannot separate the C\*08:55N from the C\*05:92N allele. These alleles can be distinguished by the HLA-C low resolution kit.

The HLA-C\*08 primer set cannot separate the C\*08:94 from the C\*05:29:01-05:29:02 alleles. These alleles can be distinguished by the HLA-C low resolution kit and/or the HLA-C\*05 high resolution kit.

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**ALLELE CONFIRMATION STATUS**

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

<sup>1</sup>Allele status “confirmed” or “unconfirmed” as listed on the IMGT/HLA web page 2014-January-17, release 3.15.0, [www.ebi.ac.uk/imgt/hla](http://www.ebi.ac.uk/imgt/hla).

**RESOLUTION IN HOMO- AND HETEROZYGOTES**

Results file with resolution in HLA-C\*08 homo- and heterozygotes is available upon request.



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**SPECIFICITY TABLE**

**HLA-C\*08 SSP subtyping**

Specificities and sizes of the PCR products of the 31+1 primer mixes used  
for HLA-C\*08 SSP subtyping

Primer Mix	Size of spec. PCR product <sup>1</sup>	Size of control band <sup>2</sup>	Amplified HLA-C*08 alleles <sup>3</sup>	Other amplified HLA Class I alleles <sup>4</sup>
<b>1</b>	250 bp	<b>800 bp</b>	*08:01:01-08:01:13, 08:03:01-08:04:03, 08:06, 08:08:01-08:11, 08:13- 08:14, 08:16:01-08:16:02, 08:20-08:22, 08:24, 08:26N, 08:36N, 08:38-08:42, 08:44, 08:46, 08:50, 08:54, 08:56- 08:61, 08:65-08:66, 08:72:01-08:72:02, 08:78- 08:89N, 08:91, 08:93, 08:95- 08:99, 08:101-08:102	*01:22, 01:35, 02:03, 02:16:01- 02:16:02, 02:18, 04:04:01-04:04:02, 04:06, 04:13, 04:34, 04:58, 04:122, 04:160, 05:11, 05:17, 05:27, 05:68, 05:79, 06:04, 06:118, 12:14:01- 12:14:02, 12:18:01-12:18:02, 12:20, 12:83, 14:06, 14:15, 14:53, 15:02:01- 15:07, 15:09-15:13, 15:15-15:19, 15:21-15:24, 15:26-15:50, 15:52- 15:73, 15:76-15:83, 16:35, 16:40, 16:48, 17:01:01:01-17:16, 17:18- 17:23, <b>B*58:02</b>
<b>2<sup>5</sup></b>	115 bp	1070 bp	*08:01:01-08:01:13, 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, 08:36N, 08:38, 08:40-08:42, 08:44, 08:46, 08:50, 08:56, 08:58-08:61, 08:78-08:89N, 08:91, 08:95-08:99, 08:101- 08:102	*05:79
<b>3<sup>5</sup></b>	115 bp	<b>800 bp</b>	*08:02:01:01-08:02:10, 08:04:01-08:05, 08:07, 08:12-08:13, 08:17-08:19, 08:23, 08:25, 08:27-08:35, 08:37, 08:43, 08:45, 08:48- 08:49, 08:52N-08:55N, 08:62, 08:66-08:71, 08:73- 08:76, 08:90, 08:92-08:94, 08:100	*04:120, 05:01:01:01-05:01:27, 05:03-05:08, 05:10-05:11, 05:13- 05:16, 05:18:01-05:51Q, 05:53- 05:61, 05:63-05:67, 05:69-05:75, 05:77-05:78, 05:80-05:87, 05:89- 05:99N, 07:41, <b>B*14:32</b>
<b>4<sup>5</sup></b>	110 bp	1070 bp	*08:03:01-08:03:03, 08:06, 08:14, 08:38, 08:40, 08:61, 08:101	*05:79
<b>5</b>	155 bp	1070 bp	*08:05, 08:15:01-08:15:02, 08:21, 08:51	*01:13, 02:51, 03:87, 04:129, 05:09:01-05:09:02, 05:17, 05:42, 05:46, 05:52, 06:67, 07:101, 07:130, 07:148, 07:161, 16:27, 17:05, <b>B*15:33, B*15:248</b>
<b>6<sup>5</sup></b>	65 bp 270 bp 310 bp	<b>800 bp</b>	*08:102 *08:06 *08:23	*07:06, 07:18-07:19 *16:33
<b>7<sup>5</sup></b>	100 bp 230 bp	1070 bp	*08:07, 08:47 *08:87	*05:23, 05:62, 07:02:35

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<b>8</b>	225 bp	1070 bp	*08:09, 08:11, 08:59	*05:04:01-05:04:02, 07:68, 07:260, 07:302, <b>B*18:83</b>
	290 bp		*08:34	
<b>9</b>	140 bp	1070 bp	*08:88N	
	375 bp		*08:24, 08:75	
	505 bp		*08:10	*05:79
<b>10<sup>5</sup></b>	110 bp	<b>800 bp</b>	*08:09	
	140 bp		*08:17	
<b>11<sup>6</sup></b>	255 bp	<b>800 bp</b>	*08:86	
	280 bp		*08:12	*05:16, 05:85, <b>B*14:32</b>
<b>12</b>	165 bp	<b>800 bp</b>	*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, 08:95-08:102	*01:43, 07:101, 07:148, 07:161
<b>13<sup>5</sup></b>	105 bp	<b>800 bp</b>	*08:02:01:01-08:02:10, 08:05, 08:07, 08:12, 08:17- 08:19, 08:23, 08:25, 08:28, 08:30, 08:32-08:35, 08:37, 08:43, 08:45, 08:47-08:48, 08:52N-08:53, 08:55N, 08:62-08:63, 08:67-08:71, 08:73-08:77, 08:90, 08:92, 08:94, 08:100	*04:120, 05:01:01:01-05:01:27, 05:03-05:07N, 05:10, 05:12-05:16, 05:18:01-05:26, 05:28-05:51Q, 05:53-05:61, 05:63-05:67, 05:69, 05:71-05:78, 05:80-05:88, 05:90- 05:99N, 07:41
<b>14</b>	170 bp	1070 bp	*08:18	
	280 bp		*08:08:01-08:08:02	
<b>15<sup>6</sup></b>	265 bp	1070 bp	*08:13, 08:16:01-08:16:02, 08:25, 08:94	*05:29:01-05:29:02
<b>16<sup>5,8</sup></b>	100 bp	1070 bp	*08:14, 08:80	*01:21, 02:42, 04:140, 05:98, 06:05, 07:02:09, 12:16, 15:63, <b>B*67:02</b>
	545 bp		*08:22, 08:56	*01:85, 15:29
<b>17</b>	375 bp	1070 bp	*08:05, 08:21, 08:25	*05:42 <sup>w</sup> , 05:46
	430 bp		*08:28	*05:25, 05:42
<b>18<sup>5</sup></b>	80 bp	1070 bp	*08:31	
	200 bp		*08:26N	
<b>19<sup>5,7</sup></b>	115 bp	<b>800 bp</b>	*08:27, 08:29, 08:30, 08:31	*05:08, 05:89
	250 bp		*08:32	
<b>20<sup>5</sup></b>	105 bp	1070 bp	*08:35, 08:43, 08:54	*05:44:01, <b>B*15:298</b>
<b>21<sup>6</sup></b>	195 bp	1070 bp	*08:01:01-08:01:13, 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, 08:72:01-08:72:02, 08:78- 08:89N, 08:91, 08:95-08:99, 08:101-08:102	*01:02:06, 01:04, 01:21, 02:02:05, 02:02:13, 02:05:01, 02:06, 02:10, 02:12, 02:14, 02:16:01, 02:17, 02:27:02, 02:33, 02:58, 02:73, 04:01:01:01-04:01:06, 04:01:09- 04:01:57, 04:03:01-04:20, 04:23- 04:159, 04:161-04:162, 04:164- 04:165, 05:12, 05:18:01, 06:02:01:01-06:02:01:03, 06:02:03- 06:02:26, 06:02:28-06:15, 06:17- 06:40, 06:42-06:61, 06:63-06:91, 06:93-06:111, 06:113-06:123, 12:02:01-12:03:15, 12:03:18- 12:03:26, 12:03:28-12:13, 12:14:02- 12:31, 12:33-12:115, 14:02:01- 14:02:04, 14:02:06-14:16,



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				14:18-14:61, 15:02:01-15:02:04, 15:02:06-15:02:12, 15:02:14- 15:05:05, 15:05:07-15:06:02, 15:07- 15:13, 15:15-15:19, 15:21-15:64, 15:66-15:84Q, 16:01:01-16:02:02, 16:02:04-16:02:11, 16:04:01, 16:06- 16:26, 16:28-16:63, 17:01:01:01- 17:05, 17:07-17:23, 18:01-18:07N
<b>22<sup>5</sup></b>	80 bp 155 bp	1070 bp	*08:41 *08:33:01	*05:27, 05:39, <b>B*44:148</b> *05:18:02-05:18:03, 07:04:01- 07:04:09, 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, <b>B*14:32, B*18:83, B*44:148</b>
<b>23<sup>5,7</sup></b>	95 bp 205 bp 240 bp	1070 bp	*08:36N *08:70Q *08:78	*03:236, 04:106
<b>24<sup>5</sup></b>	105 bp 150 bp	1070 bp	*08:56, 08:89N *08:37, 08:53	*02:14, 04:42, 05:43, 06:05, 07:02:09, 12:16, 15:23, 15:63, 16:21
<b>25<sup>5</sup></b>	115 bp 265 bp	1070 bp	*08:39 *08:62, 08:82	*05:12, <b>A*29:10, B*18:83, B*44:148</b> *05:05, <b>A*02:425, A*29:10, A*68:69,</b> <b>B*14:32, B*18:83, B*44:148</b>
<b>26<sup>5</sup></b>	125 bp	1070 bp	*08:42	
<b>27<sup>5</sup></b>	125 bp 265 bp	1070 bp	*08:19, 08:101 *08:55N	*05:64:01-05:64:02 *05:92N
<b>28<sup>7</sup></b>	155 bp 185 bp  285 bp	1070 bp	*08:53 *08:01:07, 08:02:07, 08:33:02 *08:44, 08:61, 08:82	*04:120  <b>*05:10, B*44:148</b>
<b>29<sup>7</sup></b>	205 bp	1070 bp	*08:38, 08:52N	
<b>30<sup>5</sup></b>	65 bp 405 bp  470 bp	1070 bp	*08:99 *08:28  *08:20, 08:40	*05:25, 05:42, 07:02:35, 07:41, <b>A*01:01:06</b> *03:171, 03:211:01, 04:144, 05:93, 06:73, 12:109
<b>31</b>	500 bp	1070 bp	*08:02:06, 08:19, 08:62	*04:129, 05:01:20, 05:64:01
<b>32<sup>9</sup></b>	-	-	<b>Negative Control</b>	

<sup>1</sup> 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\*08 SSP subtypings.

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.



101.623-12 – including *Taq* pol., IFU-01  
101.623-12u– without *Taq* pol., IFU-02

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**Lot No.: 72V**

**Lot-specific information**

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.

<sup>2</sup>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.

<sup>3</sup>For several HLA Class I alleles 1<sup>st</sup> and/or 4<sup>th</sup> 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.

<sup>4</sup>Due to the sharing of sequence motifs between HLA-C alleles non-HLA-C\*08 alleles will be amplified by primer mixes 1 to 9, 11 to 13, 15 to 17, 19 to 25, 27, 28, 30, 31 amplify non-HLA-C\*08 alleles. In addition, a few HLA-A and HLA-B alleles will be amplified by primer mixes 1, 3, 5, 8, 11, 16, 20, 22, 25, 28 and 30.

<sup>5</sup>HLA-specific PCR products shorter than 125 base pairs have a lower intensity and are less sharp than longer PCR products.

<sup>6</sup>Primer mixes 11, 15 and 21 may have tendencies of unspecific amplifications.

<sup>7</sup>Primer mixes 19, 23, 28 and 29 have a tendency to giving rise to primer oligomer formation.

<sup>8</sup>Primer mix 16 may give rise to a lower yield of HLA-specific PCR product than the other HLA-C\*08 primer mixes.

<sup>9</sup>Primer mix 32 contains a negative control, which will amplify more than 95% of HLA amplicons as well as the amplicons generated by control primer pairs. PCR product sizes range from 75 to 200 base pairs. The PCR product generated by the control primer pair is 430 base pairs.

‘w’, might be weakly amplified.

101.623-12 – including *Taq* pol., IFU-01  
101.623-12u– without *Taq* pol., IFU-02

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Lot No.: **72V**

Lot-specific information  
**PRIMER SPECIFICATION**

Well No.	1	2	3	4	5	6	7	8	9	10	11	12
Length of spec. PCR product	250	115	115	110	155	65	100	225	140	110	255	165
						270	230	290	375	140	280	
						310			505			
Length of int. pos. control <sup>1</sup>	<b>800</b>	1070	<b>800</b>	1070	1070	<b>800</b>	1070	1070	1070	<b>800</b>	<b>800</b>	<b>800</b>
5'-primer(s) <sup>2</sup>	2 <sup>nd</sup> I	527	527	527	176	2 <sup>nd</sup> I	453	351	312	176	361	176
	5'-CCA 3'	5'-TAC 3'	5'-TgA 3'	5'-TAC 3'	5'-gCA 3'	5'-CCA 3'	5'-AAT 3'	5'-CAA 3'	5'-AAA 3'	5'-gCA 3'	5'-AgT 3'	5'-gCA 3'
					485	1018	658	419	429	527	634	
					5'-CAA 3'	5'-gTg 3'	5'-CTA 3'	5'-gTC 3'	5'-Ag 3'	5'-TAC 3'	5'-CAA 3'	
									736			
									5'-gCA 3'			
									757			
									5'-CCA 3'			
3'-primer(s) <sup>3</sup>	539	601	601	595	289	559	512	601	526	277	601	302
	5'-TCA 3'	5'-CTT 3'	5'-CTT 3'	5'-CCT 3'	5'-AgC 3'	5'-CgC 3'	5'-CCA 3'	5'-CTT 3'	5'-CgT 3'	5'-gCA 3'	5'-CTT 3'	5'-ggC 3'
					289	599	846		956	598	846	
					5'-AgC 3'	5'-TCC 3'	5'-CAC 3'		5'-CAg 3'	5'-CTC 3'	5'-CAC 3'	
					601	1043						
					5'-CTC 3'	5'-CAA 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. PCR product	105	170	265	100	375	80	115	105	195	80	95	105
		280		545	430	200	250			155	205	150
											240	
Length of int. pos. control <sup>1</sup>	<b>800</b>	1070	1070	1070	1070	1070	<b>800</b>	1070	1070	1070	1070	1070
5'-primer(s) <sup>2</sup>	539	173	1 <sup>st</sup> I	142	1 <sup>st</sup> I	441	391	527	2 <sup>nd</sup> I	486	246	88
	5'-gCg 3'	5'-CgC 3'	5'-CgA 3'	5'-TCT 3'	5'-CgA 3'	5'-TAg 3'	5'-ACT 3'	5'-TgA 3'	5'-CCA 3'	5'-ACC 3'	5'-CAg 3'	5'-Tgg 3'
		363		972		560	520			560	374	97
		5'-AgC 3'		5'-CTA 3'		5'-CgA 3'	5'-CgC 3'			5'-CCT 3'	5'-CTA 3'	5'-TCg 3'
							539				3 <sup>rd</sup> I	134
							5'-gTg 3'				5'-Cgg 3'	5'-CCg 3'
												527
												5'-TAC 3'
3'-primer(s) <sup>3</sup>	601	302	175	201	289	601	601	584	485	601	302	201
	5'-CTT 3'	5'-ggC 3'	5'-CCg 3'	5'-CTT 3'	5'-AgC 3'	5'-CTT 3'	5'-CTT 3'	5'-ggC 3'	5'-CCg 3'	5'-CTT 3'	5'-ggC 3'	5'-CTT 3'
		601	175	1034	341			595			538	587
		5'-CTT 3'	5'-CCT 3'	5'-AgT 3'	5'-CgT 3'			5'-CCT 3'			5'-CCg 3'	5'-CCg 3'
											787	
											5'-TCT 3'	
Well No.	13	14	15	16	17	18	19	20	21	22	23	24

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101.623-12u– without *Taq* pol., IFU-02

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Lot No.: **72V**

Lot-specific information

Well No.	25	26	27	28	29	30	31
Length of spec.	115	125	125	155	205	65	500
PCR product	265		265	185		405	
				285		470	
Length of int.	1070	1070	1070	1070	1070	1070	1070
pos. control <sup>1</sup>							
5'-primer(s) <sup>2</sup>	379	2 <sup>nd</sup> I	1 <sup>st</sup> I	88	368	341	176
	5' -ACg 3' 5' -CCA 3'	5' -CgA 3'	5' -Tgg 3' 5' -gTC 3' 5' -ggA 3' 5' -gCA 3'				
	527		368	355	679	652	
	5' -TgT 3'		5' -gTT 3' 5' -TCA 3' 5' -g.T 3' 5' -CCA 3'				
				459		934	
				5' -gAT 3'		5' -TCA 3'	
3'-primer(s) <sup>3</sup>	601	412	175	201	526	453	387
	5' -CTT 3' 5' -CTT 3'	5' -CTA 3'	5' -CTT 3' 5' -CgT 3' 5' -TCA 3' 5' -TCC 3'				
			453	601	846	956	
			5' -TCA 3' 5' -CTT 3' 5' -CAC 3' 5' -CAg 3'				
Well No.	25	26	27	28	29	30	31

<sup>1</sup>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.

<sup>2</sup>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](http://www.ebi.ac.uk/imgt/hla) web site. The sequence of the 3 terminal nucleotides of the primer is given.

<sup>3</sup>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](http://www.ebi.ac.uk/imgt/hla) web site. The sequence of the 3 terminal nucleotides of the primer is given.

101.623-12 – including *Taq* pol., IFU-01  
101.623-12u– without *Taq* pol., IFU-02

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Lot No.: **72V**

Lot-specific information

CELL LINE VALIDATION SHEET																				
HLA-C*08 SSP primer set <sup>3</sup>																				
				Well <sup>2</sup>																
				1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	
				Prod. No.:	201075801	201075802	201075803	201075804	201075805	201434806	201434807	201075808	201434809	201075810	201434811	201075812	201075813	201075814	201434815	201316516
IHC cell line <sup>1</sup>		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	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

101.623-12 – including **Taq** pol., IFU-01  
101.623-12u– without **Taq** pol., IFU-02

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Lot No.: **72V**

Lot-specific information

<b>CELL LINE VALIDATION SHEET</b>																		
<b>HLA-C*08 SSP primer set<sup>3</sup></b>																		
			<b>Well<sup>2</sup></b>															
			17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	
			Prod. No.:	201075817	201075818	201075819	201434820	201075821	201189822	201434823	201434824	201316525	201189826	201202532	201434828	201202529	201434830	201202531
	<b>IHWC cell line<sup>1</sup></b>	<b>C*</b>																
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	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-

<sup>1</sup>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.



101.623-12 – including *Taq* pol., IFU-01  
101.623-12u– without *Taq* pol., IFU-02

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**Lot No.: 72V**

**Lot-specific information**

<sup>2</sup>The C\*07:18 allele is amplified by primer mix 6 in the 9285 (WT49) cell line.

<sup>3</sup>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 5, 7 to 11, 14, 15, 17 to 19, 23 to 31 were available.

The specificity of the primers in primer solutions 5, 7 to 9, 11, 14, 15, 17 to 19, 23, 24, 25, 27 to 31 were tested by adding additional 5'-primers respectively 3'-primers. In primer solutions 10 and 26 it was only possible to test the 5'-primer, the 3'-primer was not possible to test. In primer solution 7, 8, 9, 11, 14, 18, 19, 23, 24 and 28 to 30, one, two or three 5'-primers were not possible to test. In primer solutions 6, 24 and 27, one or two 3'-primers were not possible to test.

Additional primers in primer solutions 6, 16, 20 and 22 were tested by separately adding either 5'-primers and/or one or two 3'-primers.

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Lot-specific information

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