

101.403-48/12 – including *Taq* pol., IFU-01  
101.403-48u/12u – without *Taq* pol., IFU-02

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“Instructions for Use” (IFU)

Lot No.: **1F4**

Lot-specific information

## **Olerup SSP<sup>®</sup> HLA-A low resolution screening**

<b>Product number:</b>	101.403-48/12 – including <i>Taq</i> polymerase 101.403-48u/12u – without <i>Taq</i> polymerase
<b>Lot number:</b>	1F4
<b>Expiry date:</b>	2019-10-01
<b>Number of tests:</b>	48 tests – Product No. 101.403-48/48u 12 tests – Product No. 101.403-12/12u
<b>Number of wells per test:</b>	23 +1
<b>Storage - pre-aliquoted primers:</b>	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. 1F4.**

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

### **CHANGES COMPARED TO THE PREVIOUS OLERUP SSP<sup>®</sup> HLA-A LOW RESOLUTION SCREENING LOT (82Y)**

The Lot-specific Information for HLA-A low resolution screening including and without *Taq* polymerase is described in one common Product Insert.

The **HLA-A low resolution Screening** specificity and interpretation tables have been updated for the HLA-A alleles described since the previous *Olerup SSP<sup>®</sup>* HLA-A low resolution Screening lot was made (**Lot No. 82Y**). The kit design is based on IMGT/HLA database 3.26.0.

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.

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

Well	5'-primer	3'-primer	rationale
6	-	Modified	3'-primer modified for increased yield of the HLA-A*23 alleles.
8	Added	-	5'-primer added for the A*66:23 allele.
9	Added	-	5'-primer added for the A*25:39 allele.
12	-	Added	3'-primer added for the A*66:26Q allele.
13	Added	-	5'-primer added for the A*25:01:09 allele.
14	Added	-	5'-primer added for the A*11:130 allele.
17	Added	-	5'-primer added for the A*03:152 allele.
18	-	Added	3'-primer added for the A*31:08 allele.

Change in revision R01 compared to R00:

1. The section “Uniquely Identified Alleles”, the Interpretation Table and the footnotes to the Interpretation Table have been corrected with respect to alleles not possible to separate.

Change in revision R02 compared to R01:

1. Primer mix 21 may generate a false positive band of about 560 base pairs. This band should be disregarded when interpreting HLA-A low resolution typings. A footnote has been added in the Specificity Table.

Change in revision R03 compared to R02:

1. The reactivities of primer mix 17 have been corrected in the Specificity and Interpretation Tables.

Change in revision R04 compared to R03:

1. Primer mix 4 amplifies the A\*01:15N allele. This has been corrected in the Specificity and Interpretation Tables.

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Well **24** contains Negative Control primer pairs, that will amplify more than 95% of the *Olerup SSP*<sup>®</sup> 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
<b>5'-primer<sup>1</sup></b>	<b>164</b>	<b>340</b>	<b>440</b>	<b>45</b>	<b>45</b>	<b>43</b>	<b>36</b>
	5'-CAC <sup>3'</sup>	5'-Agg <sup>3'</sup>	5'-TTA <sup>3'</sup>	5'-Tgg <sup>3'</sup>	5'-Tgg <sup>3'</sup>	5'-Tgg <sup>3'</sup>	5'-TAC <sup>3'</sup>
							<b>36</b>
							5'-TAT <sup>3'</sup>
<b>3'-primer<sup>2</sup></b>	<b>231</b>	<b>2<sup>nd</sup> I</b>	<b>507</b>	<b>59</b>	<b>58</b>	<b>57</b>	<b>47</b>
	5'-TgC <sup>3'</sup>	5'-AAA <sup>3'</sup>	5'-TTg <sup>3'</sup>	5'-CTC <sup>3'</sup>	5'-ggC <sup>3'</sup>	5'-CTC <sup>3'</sup>	5'-ACA <sup>3'</sup>
							<b>48</b>
							5'-gCA <sup>3'</sup>
							<b>48</b>
							5'-gCC <sup>3'</sup>
							<b>52</b>
							5'-TgT <sup>3'</sup>
<b>A*</b>	<b>+</b>	<b>+</b>	<b>+</b>				
<b>B*</b>	<b>+</b>	<b>+</b>	<b>+</b>				
<b>C*</b>	<b>+</b>	<b>+</b>	<b>+</b>				
<b>DRB1</b>				<b>+</b>	<b>+</b>		
<b>DRB3</b>				<b>+</b>	<b>+</b>		
<b>DRB5</b>				<b>+</b>			
<b>DQB1</b>					<b>+</b>		
<b>DPB1</b>						<b>+</b>	
<b>DQA1</b>							<b>+</b>

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

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## PRODUCT DESCRIPTION

### HLA-A low resolution screening

#### CONTENT

The primer set contains 5'- and 3'-primers for grouping the HLA-A\*01:01 to A\*80:03 alleles into the corresponding serological groups A1 to A80.

#### PLATE LAYOUT

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

<b>1</b>	<b>2</b>	<b>3</b>	<b>4</b>	<b>5</b>	<b>6</b>	<b>7</b>	<b>8</b>
<b>9</b>	<b>10</b>	<b>11</b>	<b>12</b>	<b>13</b>	<b>14</b>	<b>15</b>	<b>16</b>
<b>17</b>	<b>18</b>	<b>19</b>	<b>20</b>	<b>21</b>	<b>22</b>	<b>23</b>	<b>NC</b>

The 24 well PCR plate is marked with 'HLA-A low screening' in silver/gray ink.

Well No. 1 is marked with the Lot No. '1F4'.

Wells 1 to 23 – HLA-A low resolution screening primers.

Well 24 – Negative Control.

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

Only HLA-A alleles will be amplified by the 24 wells of the HLA-A low resolution screening primer set, except that a few HLA-B and HLA-C alleles will be amplified by primer mixes 4 to 6, 9, 11, 15, 22 and 23.

For further details see Specificity Table.

#### UNIQUELY IDENTIFIED ALLELES

All the HLA-A alleles, i.e. **A\*01:01 to A\*80:03**, recognized by the HLA Nomenclature Committee in October 2016<sup>1,2</sup> will be amplified by the primers in the HLA-A low resolution screening primer set<sup>3</sup>. The HLA-A alleles will be grouped into their corresponding serological specificities.

The following HLA-A low alleles can be distinguished by the different sizes of the HLA-specific PCR product:

Alleles	Primer mix
A*01:26, 01:136, 01:192, A*11:94, 11:112, 11:211	11
A*24:14:01:01-24:15, 24:51-24:53, 24:57, 24:64, 24:114, 24:222N, 24:291, 24:296, 24:304, 24:316, 24:324, C*04:01:03	5
A*68:01:32, 68:11N	22

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<sup>1</sup>HLA-A, HLA-B and HLA-C alleles listed on the IMGT/HLA web page 2016-October-14, release 3.26.0, [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 A\*23:14:01-23:14:02 and the A\*24:24, 24:71 and 24:315 alleles will give rise to identical amplification patterns. These alleles can be separated by the respective high resolution SSP primer sets.

The A\*23:57 and 23:66 and the A\*24:94, 24:138, 24:188 and 24:228 alleles will give rise to identical amplifications patterns. These alleles can be separated by the respective high resolution SSP primer sets.

The A\*66:01:01-66:01:02, 66:01:03, 66:04-66:05, 66:07-66:08, 66:11, 66:13-66:14, 66:17-66:20, 66:22, 66:24 and 66:27N and the A\*26:92 alleles will give rise to identical amplifications patterns. These alleles can be separated by the respective high resolution SSP primer sets.

The 30:01:01-30:02:11, 30:02:13-30:04:02, 30:06-30:07, 30:09-30:20, 30:22-30:54, 30:56-30:88, 30:90-30:100, 30:102-30:104 and 30:106-30:107 and the B\*07:260 alleles will give rise to identical amplifications patterns. These alleles can be separated by the respective high resolution SSP primer sets.

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Lot-specific information  
**SPECIFICITY TABLE**

**HLA-A low resolution screening primer set**

Specificities and sizes of the PCR products of the 23+1 primer mixes used for  
HLA-A low resolution screening SSP typing

Primer Mix	Size of spec. PCR product <sup>1</sup>	Size of control band <sup>2</sup>	HLA-A serology <sup>3</sup>	Amplified HLA-A alleles <sup>4,5</sup>
<b>1<sup>6,7</sup></b>	120 bp, 145 bp, 225 bp	<b>800 bp</b>	A1, A36	*01:01:01:01-01:04N, 01:06-01:33, 01:35-01:211, 01:213-01:217, 03:18, 03:135, 03:260, 11:94, 11:112, 11:211, 11:226, 26:120, 36:01-36:05
<b>2<sup>7</sup></b>	175 bp, 215 bp, 255 bp, 365 bp, 415 bp	<b>800 bp</b>	A2, A203, A210, A19, A28	*02:01:01:01-02:01:15, 02:01:17-02:01:19, 02:01:21-02:01:81, 02:01:83-02:22:02, 02:24:01-02:35:01, 02:35:03-02:47, 02:49-02:77, 02:78 <sup>W</sup> , 02:79:01-02:97:02, 02:99, 02:101:01-02:128, 02:130-02:570, 02:572-02:641, 02:643N, 02:645-02:654
<b>3<sup>8,13</sup></b>	210 bp, 240 bp	1070 bp	A1, A3, A11, A32, A34, A36	*01:12, 01:19, 01:21, 01:126, 01:200, 02:338, 03:01:01:01-03:17:02, 03:19-03:74, 03:76-03:94, 03:96-03:134, 03:136-03:176, 03:178N-03:186, 03:188-03:193, 03:195-03:214, 03:216-03:224, 03:226-03:248, 03:250-03:265, 11:03, 11:20, 11:25:01-11:25:02, 11:60, 11:130, 11:158, 11:175, 11:183, 11:209, 24:92, 32:04, 34:02:01-34:04, 34:07-34:10N, 34:13, 34:15, 36:02, 74:23
<b>4</b>	190 bp	<b>800 bp</b>	A1, A2, A3, A11, A26, A30, A36	*01:01:01:01-01:01:22, 01:01:24-01:01:47, 01:01:49-01:01:64, 01:01:67-01:04N, 01:06-01:07, 01:09:01-01:11N, 01:13, 01:15N-01:18N, 01:20-01:29, 01:31N-01:33, 01:35-01:78, 01:80-01:98, 01:100-01:144, 01:146, 01:148, 01:150-01:158, 01:160N-01:166, 01:168-01:177, 01:180-01:199, 01:201-01:204, 01:206-01:207, 01:209-01:211, 01:213, 01:215-01:217, 02:78, 02:169, 03:12, 03:18, 03:88, 03:135, 11:01:01:01-11:27, 11:29-11:52Q, 11:54-11:246, 11:249-11:251N, 26:19, 26:72, 29:67, 30:08, 32:64, 36:04, 66:23, 68:13:01, 68:66, 68:134, 74:19, <b>C*12:131</b>
<b>5<sup>7</sup></b>	160 bp, 335 bp, 505 bp	1070 bp	A3, A9, A23, A24, A2403, A29, A31, A32	*03:15, 03:19, 03:30, 03:152, 11:139, 23:01:01-23:68, 23:70-23:77, 24:02:01:01-24:11N, 24:13:01-24:15, 24:17-24:64, 24:66-24:210, 24:212-24:341, 24:343-24:351, 24:354-24:367, 29:07, 29:49, 31:08, 31:29, 32:05, 32:79, 33:19, 33:53, <b>C*04:01:03</b>
<b>6</b>	135 bp, 200 bp	<b>800 bp</b>	A9, A23, A24, A29, A80	*11:166, 23:01:01-23:56, 23:58-23:65, 23:67-23:68, 23:70-23:77, 24:24, 24:71, 24:315, 29:07, 29:49, 31:29, 31:85, 32:72, 80:01:01:01-80:03, <b>B*18:27</b>
<b>7<sup>7</sup></b>	175 bp, 205 bp	1070 bp	A2, A23, A24, A26, A33	*02:17:01 <sup>W</sup> -02:17:03 <sup>W</sup> , 11:139, 23:14:01-23:14:02, 24:02:01:01-24:11N, 24:13:01-24:13:02, 24:17-24:50, 24:54-24:56, 24:58-24:63, 24:66-24:91, 24:93, 24:95-24:113, 24:115-24:137, 24:139-24:187, 24:189-24:210, 24:212-24:221, 24:223-24:227, 24:229-

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				24:290, 24:292-24:295, 24:297-24:303N, 24:305-24:315, 24:317-24:323N, 24:325-24:341, 24:343-24:351, 24:354-24:367, 26:16, 33:19, 33:119, 68:45, 68:117
<b>8<sup>7</sup></b>	160 bp, 200 bp	<b>800 bp</b>	A2, A3, A10, A11, A25, A26, A28, A32, A34, A43, A66, A68, A69	*01:51, 02:55, 02:135, 02:527, 02:582, 02:644, 03:24, 03:50, 11:10, 11:183, 11:191, 25:01:01-25:40, 26:01:01:01-26:01:39, 26:01:41-26:43:02, 26:45-26:132, 29:28, 32:15, 33:51, 34:01:01-34:17, 43:01, 66:01:01:01-66:27N, 68:01:01:01-68:151, 68:153-68:157, 69:01:01-69:03
<b>9<sup>6,9</sup></b>	80 bp	<b>800 bp</b>	A3, A25, A32	*25:01:01-25:40, 32:01:01-32:02, 32:04, 32:06-32:37, 32:39-32:59, 32:61-32:70, 32:72-32:77, 32:81-32:92N, <b>B*07:81, B*08:52, B*18:67, B*38:41, B*51:185, B*53:05, B*53:16, B*53:33</b>
<b>10<sup>6,7,10</sup></b>	80 bp, 240 bp	<b>800 bp</b>	A10, A26, A43	*01:43, 01:51, 02:644, 11:17, 11:40, 11:223, 26:01:01:01-26:01:39, 26:01:41-26:02:02, 26:04, 26:07:01-26:20, 26:22-26:29, 26:31-26:43:02, 26:45-26:77, 26:79-26:91, 26:93-26:110, 26:112-26:132, 33:13, 33:48, 43:01, 68:84
<b>11<sup>6,7</sup></b>	80 bp, 175 bp, 500 bp	1070 bp	A1, A11, A24, A26, A31, A34, A66	*01:13, 01:26, 01:28, 01:136, 01:176, 01:192, 03:63, 03:88, 11:01:01:01-11:27, 11:29-11:52Q, 11:54-11:246, 11:249-11:251N, 24:19, 24:44, 26:03:01, 26:06, 26:21, 26:78, 26:92, 26:111, 34:01:01-34:08, 34:10N-34:17, 66:01:01:01-66:01:03, 66:04-66:11, 66:13-66:14, 66:17-66:20, 66:22-66:24, 66:27N, 69:02, 80:02, <b>C*07:404</b>
<b>12<sup>6</sup></b>	125 bp, 185 bp	<b>800 bp</b>	A3, A10, A25, A26, A31, A34, A43, A66, A74	*02:309, 02:454, 03:01:19, 11:11, 25:05-25:06, 26:09, 26:54, 26:91, 31:03-31:04, 34:01:01-34:17, 43:01, 66:02-66:03, 66:16, 66:21, 66:25-66:26Q, 68:130, 74:01:03
<b>13</b>	125 bp, 185 bp	1070 bp	A1, A2, A3, A10, A25, A26, A34, A43, A66	*01:01:56 <sup>W</sup> , 01:13, 01:145 <sup>W</sup> , 01:176, 01:194, 02:34-02:35:03, 02:56:01-02:56:02, 02:62, 02:103, 02:135, 02:580, 03:01:01:01-03:01:22, 03:01:24-03:07, 03:09-03:11N, 03:13-03:31, 03:33-03:35, 03:37-03:40, 03:42-03:56, 03:58, 03:60-03:71, 03:73-03:87, 03:90-03:106, 03:109-03:110, 03:112-03:141, 03:143-03:151, 03:153-03:171, 03:174-03:175, 03:177, 03:179-03:193, 03:195-03:197N, 03:200Q-03:202, 03:204, 03:206-03:210, 03:212-03:218, 03:220-03:251, 03:253-03:265, 11:116, 11:140, 11:199:01-11:199:02, 11:222, 25:01:01-25:05, 25:07-25:35, 25:37-25:40, 26:01:01:01-26:01:20, 26:01:22-26:01:39, 26:01:41-26:03:01, 26:05-26:08, 26:10-26:28, 26:29 <sup>W</sup> , 26:30-26:33, 26:35-26:43:02, 26:45-26:48, 26:49 <sup>W</sup> , 26:50-26:72, 26:74-26:77, 26:79-26:90, 26:92-26:132, 30:55, 31:24, 33:61, 34:08, 43:01, 66:01:01:01-66:01:01:02, 66:01:03, 66:04-66:09, 66:10 <sup>W</sup> , 66:11-66:15, 66:17-66:20, 66:22-66:24, 66:27N, 68:71, 74:13
<b>14<sup>6,7,10</sup></b>	80 bp, 115 bp, 200 bp, 240 bp,	1070 bp	A3, A26, A29, A31, A34, A36, A68	*02:237, 02:309, 02:454, 03:01:38, 03:95, 03:123:02, 03:171, 03:231:01, 11:130, 26:19, 26:22, 29:01:01:01-29:94, 31:03-31:04, 32:42, 34:04, 36:02, 66:09, 68:19



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	470 bp			
<b>15</b> <sup>6,7,12,13</sup>	90 bp, 135 bp, 205 bp	1070 bp	A1, A30	*01:13, 01:28, 01:176, 01:194, 03:43, 03:82, 03:186, 03:260, 11:113, 11:162, 30:01:01-30:02:11, 30:02:13-30:04:02, 30:06-30:20, 30:22-30:100, 30:102-30:104, 30:106-30:107, 31:35, <b>B*07:260</b>
<b>16</b>	240 bp, 380 bp, 410 bp	1070 bp	A24, A31, A32	*02:237, 03:95, 29:14, 31:01:02:01-31:115, 32:05, 32:79, 33:53, 74:13
<b>17</b>	140 bp, 180 bp, 235 bp, 260 bp	1070 bp	A3, A25, A32, A74	*01:95, 03:32, 03:43, 03:82, 03:152, 03:186, 03:219, 23:64, 24:104, 24:243, 25:03, 25:30, 29:13, 29:39, 30:89, 31:21, 31:35, 32:01:01-32:93, 74:07
<b>18</b> <sup>7</sup>	200 bp, 240 bp, 390 bp, 430 bp	1070 bp	A24, A31, A32, A33, A68, A74	*02:243:01-02:243:02, 24:82, 29:48, 31:07-31:08, 31:10, 31:109, 32:03, 32:15, 32:93, 33:01:01- 33:01:08, 33:03:01-33:37, 33:39-33:52, 33:54-33:120, 68:29, 74:04, 74:21
<b>19</b>	340 bp, 375 bp	<b>800 bp</b>	A2, A19, A68, A74	*01:121, 02:65, 02:407, 02:449, 03:246, 32:62, 68:25, 74:01:01-74:28
<b>20</b> <sup>10</sup>	210 bp, 240 bp	<b>800 bp</b>	A2, A210, A25, A28, A68	*02:34-02:35:03, 02:46, 02:48, 02:56:01-02:56:02, 02:62, 02:70, 02:78, 02:103, 02:129, 02:571, 02:580, 11:199:02, 23:01:13, 24:340, 25:05, 26:54, 26:122, 34:02:04, 68:01:01:01-68:151, 68:153-68:157
<b>21</b> <sup>14</sup>	200 bp, 240 bp, 375 bp, 545 bp	<b>800 bp</b>	A2, A23, A24 A26, A28, A66, A68, A69	*02:17:01-02:17:03, 02:55, 02:108, 02:110, 02:243:01-02:243:02, 02:268, 02:300, 02:303, 02:617, 02:644, 23:02, 23:57, 23:66, 24:06, 24:13:01, 24:22, 24:82, 24:94, 24:138, 24:167, 24:188, 24:207:01, 24:228, 24:339, 25:30, 26:22, 33:22, 66:06, 66:09, 68:08:02, 68:29, 68:105, 68:157, 69:01:01-69:03
<b>22</b> <sup>6,11</sup>	85 bp, 240 bp, 400 bp	<b>800 bp</b>	A2, A23, A34, A36	*02:34-02:35:03, 02:46, 02:48, 02:56:01-02:56:02, 02:62, 02:70, 02:78, 02:103, 02:129, 02:571, 02:580, 03:187, 11:155, 11:199:02, 11:226, 11:231 <sup>?</sup> , 23:01:13, 24:340, 31:62, 34:02:04, 36:01-36:05, 68:01:32, 68:11N, <b>C*07:481<sup>?</sup></b> , <b>C*07:504<sup>?</sup></b>
<b>23</b> <sup>6</sup>	75 bp, 160 bp, 240 bp, 495 bp	<b>800 bp</b>	A2, A3, A24 A26, A36, A68, A80	*02:55, 02:237, 03:41, 03:63, 03:75, 03:88, 03:95, 03:177, 11:130, 24:18, 24:204, 24:213, 26:03:01, 26:05-26:06, 26:21, 26:30, 26:78, 26:111, 33:24, 36:02, 68:05, 68:15, 68:20, 68:109, 68:136, 80:01:01:01-80:01:01:02, 80:03, <b>C*06:187</b>
<b>24</b> <sup>15</sup>	-	-	-	<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-A low resolution screening 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 inherent feature of the primer



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**101.403-48u/12u – without Taq pol., IFU-02**

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**Lot-specific information**

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>The serological reactivity of all HLA-A alleles is not known. In this table we use the expert-assigned serological grouping in Tissue Antigens (2009) 73:95-170 and the serological grouping of the sequence-defined allele.

<sup>4</sup>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.

<sup>5</sup>Due to the sharing of sequence motifs between HLA-C alleles, non-HLA-A alleles will be amplified by primer mixes 4 to 6, 9, 11, 15, 22, 23.

<sup>6</sup>HLA-specific PCR products shorter than 125 base pairs have a lower intensity and are less sharp than longer PCR products.

<sup>7</sup>The primer pairs in wells 1, 2, 5, 7, 8, 10, 11, 14, 15 and 18 will in many samples give rise to two or three HLA-specific PCR fragments.

<sup>8</sup>Primer mix 3 may faintly amplify the A\*30:04:01-30:04:02, 30:06, 30:09, 30:17, 30:29, 30:46, 30:77, 30:90, 30:99 and 30:103 alleles.

<sup>9</sup>Primer mix 9 may weakly amplify the A\*34 alleles.

<sup>10</sup>Primer mixes 10, 14 and 20 have a tendency to giving rise to primer oligomer formation.

<sup>11</sup>Primer mix 22 might faintly amplify most A\*11 alleles.

<sup>12</sup>Primer mix 15 may give rise to a lower yield of HLA-specific PCR product than the other HLA-A low primer mixes.

<sup>13</sup>Primer mixes 3 and 15 may have tendency of unspecific amplification.

<sup>14</sup>Primer mix 21 may generate a false positive band of about 560 base pairs. This band should be disregarded when interpreting HLA-A low resolution typings.

<sup>15</sup>Primer mix 24 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.

‘?’, nucleotide sequence information not available for the primer matching sequence.

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Lot No.: **1F4**

Lot-specific information  
**PRIMER SPECIFICATION**

Well No.	1	2	3	4	5	6	7	8	9	10	11	12
Length of spec.	120	175	210	190	160	135	175	160	80	80	80	125
PCR product	145	215	240		335	200	205	200		240	175	185
		225	255		505						500	
		365										
		415										
Length of int.	800	800	1070	800	1070	800	1070	800	800	800	1070	800
pos. control <sup>1</sup>												
5'-primer(s) <sup>2</sup>	98	48	357	98	144	176	98	98	259	98	301	103
	5'-CTT 3'	5'-gCT 3'	5'-ATg 3'	5'-CTA 3'	5'-gCC 3'	5'-gCA 3'	5'-CTC 3'	5'-CTA 3'	5'-AgA 3'	5'-CTA 3'	5'-Cgg 3'	5'-CCT 3'
	103	78	363	413	317	368	368	102	261	261	302	415
	5'-CCT 3'	5'-TCT 3'	5'-ATA 3'	5'-CCg 3'	5'-gCT 3'	5'-gTT 3'	5'-gTT 3'	5'-ACA 3'	5'-AAC 3'	5'-AAC 3'	5'-ggA 3'	5'-ggT 3'
	123	106	363					413	266		385	423
	5'-AgT 3'	5'-CCA 3'	5'-ATA 3'					5'-CCg 3'	5'-ACg 3'		5'-ggC 3'	5'-gCT 3'
	363							423				
	5'-ATA 3'							5'-gCT 3'				
3'-primer(s) <sup>3</sup>	203	240	527	256	265	270	259	259	302	299	341	257
	5'-TCT 3'	5'-ggA 3'	5'-CCA 3'	5'-CTg 3'	5'-CCC 3'	5'-ACA 3'	5'-gTT 3'	5'-gTT 3'	5'-ggC 3'	5'-TCg 3'	5'-CgT 3'	5'-gCA 3'
	545	292	527	559	368	521	502	259	303		521	506
	5'-AgA 3'	5'-gTg 3'	5'-CCT 3'	5'-CCg 3'	5'-CAA 3'	5'-ggg 3'	5'-CTT 3'	5'-gTT 3'	5'-AgA 3'		5'-ggg 3'	5'-TgT 3'
			527			534	539	538				559
			5'-CCT 3'			5'-CgT 3'	5'-TCT 3'	5'-CCA 3'				5'-CTC 3'
			555									559
			5'-CCA 3'									5'-CgT 3'
			555									559
			5'-gCA 3'									5'-CgC 3'
												563
												5'-CgA 3'
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

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Lot-specific information

Well No.	13	14	15	16	17	18	19	20	21	22	23
Length of spec.	180	80	90	240	140	200	340	210	200	85	75
PCR product	225	115	135	380	180	240	375	240	240	240	160
		200	205	410	235	390			375	400	240
		240			260	430			545		495
		470									
Length of int. pos. control <sup>1</sup>	1070	1070	1070	1070	1070	1070	800	800	800	800	800
5'-primer(s) <sup>2</sup>	98	98	203	41	98	41	302	78	28	47	176
	5'-CTT 3'	5'-CAC 3'	5'-gAA 3'	5'-CTT 3'	5'-CTT 3'	5'-CTT 3'	5'-ggA 3'	5'-TCT 3'	5'-TCg 3'	5'-g.T 3'	5'-gCA 3'
	414	219	362	355	180	98	302	106	261	78	261
	5'-CAg 3'	5'-gCA 3'	5'-ggT 3'	5'-CCg 3'	5'-TTT 3'	5'-CAC 3'	5'-ggA 3'	5'-CCA 3'	5'-AAC 3'	5'-TCT 3'	5'-AAC 3'
	423	238	363		203		341	2 <sup>nd</sup> I	368	527	341
	5'-gCT 3'	5'-AgA 3'	5'-ATA 3'		5'-gAA 3'		5'-ggA 3'	5'-CCT 3'	5'-gTT 3'	5'-TgC 3'	5'-ggA 3'
		355	363		418						355
		5'-CCg 3'	5'-ATA 3'		5'-AgC 3'						5'-CCC 3'
		489	363								362
		5'-gCA 3'	5'-ATA 3'								5'-gAg 3'
			369								362
			5'-TAC 3'								5'-gAg 3'
3'-primer(s) <sup>3</sup>	282	180	299	238	290	256	397	265	97	265	292
	5'-gAC 3'	5'-TCA 3'	5'-CCA 3'	5'-CCT 3'	5'-CAA 3'	5'-CTC 3'	5'-gAg 3'	5'-CCC 3'	5'-ggT 3'	5'-CCC 3'	5'-gTg 3'
	282	257	411	238	317	256		282	355	282	292
	5'-gAC 3'	5'-gCA 3'	5'-TCA 3'	5'-CCT 3'	5'-ggA 3'	5'-CCC 3'		5'-gAC 3'	5'-gAC 3'	5'-gAC 3'	5'-gTT 3'
	559	418	526	243	555	259		282	524	282	299
	5'-CCC 3'	5'-gTC 3'	5'-CCA 3'	5'-TCA 3'	5'-CCA 3'	5'-gTT 3'		5'-gAC 3'	5'-CAT 3'	5'-gAC 3'	5'-TCT 3'
	559	555		265	299			502	538	570	555
	5'-CCg 3'	5'-CCA 3'		5'-CCC 3'	5'-TCT 3'			5'-CTT 3'	5'-CCA 3'	5'-CAC 3'	5'-CCA 3'
				282				506			
				5'-gAC 3'				5'-TgT 3'			
				555							
				5'-CCA 3'							
Well No.	13	14	15	16	17	18	19	20	21	22	23

<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 19 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.

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Lot-specific information

CELL LINE VALIDATION SHEET																				
HLA-A low resolution screening primer set <sup>2</sup>																				
				Well																
				1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	
				Prod. No.:	201668801	201779202	2016669103	201779204	201674905	201779206	201674907	201674908	201779209	201674910	201779211	201779212	201779213	201674914	201674915	201779216
	IHWC cell line <sup>1</sup>	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		-	+	-	-	-	-	W	-	-	-	-	-	-	-	-	-	-
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		-	+	-	-	-	-	W	-	-	-	-	-	-	-	-	-	-
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	-	+	-	+	-	-	-	-	-	-	+	-	-	-	-	-	-

101.403-48/12 – including *Taq* pol., IFU-01  
101.403-48u/12u – without *Taq* pol., IFU-02

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Lot No.: **1F4**

Lot-specific information

CELL LINE VALIDATION SHEET											
HLA-A low resolution screening primer set <sup>2</sup>											
				Well							
				17	18	19	20	21	22	23	
				Prod. No.:	201674917	201674918	201674919	201674920	201674921	201674922	201779223
	IHWC cell line <sup>1</sup>	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	-	-	-	-	-	-	-	-

101.403-48/12 – including *Taq* pol., IFU-01  
101.403-48u/12u – without *Taq* pol., IFU-02

Visit [www.olerup.com](http://www.olerup.com) for  
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**Lot No.: 1F4**

**Lot-specific information**

<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.

<sup>2</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.

Additional 5'- and 3'-primers in primer solutions 4, 5, 7, 8, 12, 14 to 17 and 20 to 23 were tested by separately adding 3'-primers, respectively 5'-primers. Additional 5'-primers in primer solution 1, 13 and 19 were tested by separately adding one or two 3'-primers. Additional 3'-primers in primer solutions 3 and 18 were tested by separately adding one or two 5'-primers.

In primer solutions 2, 3, 9, 11, 12, 15 and 22 one or two 5'-primers were not possible to test, and in primer solutions 3, 6, 8, 9, 12, 13, 16 and 18 one or two 3'-primers were not possible to test.

101.403-48/12 – including *Taq* pol., IFU-01  
101.403-48u/12u – without *Taq* pol., IFU-02

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Lot No.: **1F4**

Lot-specific information

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