

101.432-12 – including *Taq* polymerase, IFU-01  
101.432-12u – without *Taq* polymerase, IFU-02

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“Instructions for Use” (IFU)

Lot No.: **7F1**

Lot-specific information  
**Olerup SSP® HLA-A\*33**

<b>Product number:</b>	101.432-12 – including <i>Taq</i> polymerase 101.432-12u – without <i>Taq</i> polymerase
<b>Lot number:</b>	7F1
<b>Expiry date:</b>	2020-04-01
<b>Number of tests:</b>	12
<b>Number of wells per test:</b>	31+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. 7F1.**

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-A\*33 LOT (5D2)**

The HLA-A\*33 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

The format of the Worksheet has been changed.

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

The HLA-A\*33 specificity and interpretation tables have been updated for the HLA-A alleles described since the previous *Olerup SSP*® HLA-A\*33 lot was made (**Lot No. 5D2**). The kit design is based on IMGT/HLA database 3.29.0.

**101.432-12 – including Taq polymerase, IFU-01**  
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**Lot No.: 7F1**

**Lot-specific information**

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

<b>Well</b>	<b>5'-primer</b>	<b>3'-primer</b>	<b>rationale</b>
7	Added	-	5'-primer added for the A*33:119 allele.
21	Added	-	5'-primer added for the A*33:129N allele.
22	Added	Added	Primer pair added for the A*33:123N allele.
25	Added	-	5'-primers added for the A*33:109 and A*33:129N alleles.
26	-	Added	3'-primer added for the A*33:111 allele.
27	Added	-	5'-primer added for the A*33:109 allele.
28	Added	-	5'-primer added for the A*33:123N allele.
31	Added	Added	Primer pair added for the A*33:68 allele.

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Lot No.: **7F1**

Lot-specific information

Well **32** 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>
							36
							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>
							48
							5'-gCA <sup>3'</sup>
							48
							5'-gCC <sup>3'</sup>
							52
							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.

101.432-12 – including *Taq* polymerase, IFU-01  
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Lot No.: **7F1**

Lot-specific information

## PRODUCT DESCRIPTION

### HLA-A\*33 SSP subtyping

#### CONTENT

The primer set contains 5'- and 3'-primers for identifying the A\*33:01 to A\*33:130 alleles.

#### PLATE LAYOUT

Each 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 cut PCR plate is marked with 'HLA-A\*33' in silver/gray ink.

Well No. 1 is marked with the Lot No. '7F1'.

Wells 1 to 31 – HLA-A\*33 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 covered with a PCR-compatible foil.

**Please note:** When removing each 32 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

Due to the sharing of sequence motifs between HLA-A alleles non-HLA-A\*33 alleles will be amplified by some primer mixes. For further details see Specificity Table.

#### UNIQUELY IDENTIFIED ALLELES

All the HLA-A\*33 alleles, i.e. **A\*33:01 to A\*33:130 alleles**, recognized by the HLA Nomenclature Committee in August 2017<sup>1,2</sup> will be amplified by the primers in the HLA-A\*33 SSP kit<sup>3</sup>.

The HLA-A\*33 kit enables separation of the confirmed HLA-A\*33 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-A\*33 alleles is listed below.

The HLA-A\*33 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-A\*33 alleles can be distinguished by the different sizes of the HLA-specific PCR product:

**101.432-12 – including *Taq* polymerase, IFU-01**  
**101.432-12u – without *Taq* polymerase, IFU-02**

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**Lot No.: 7F1**

Alleles	Lot-specific information	
	Primer mix	Alleles
A*33:03:03Q, 33:86	30	A*33:29, 33:39
A*33:08, 31:99	10	A*33:30, 33:73N
A*33:11, 33:80N	12	A*33:31, 33:44
A*33:16, 33:64	15	

<sup>1</sup>HLA-A alleles listed on the IMGT/HLA web page 2017-August-10, release 3.29.0, [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\*33:09 and the A\*02:309, 26:22 and 66:09 alleles, the A\*33:51, 33:119 and A\*66:15 alleles give rise to identical amplification patterns with the HLA-A\*33 subtyping kit. These alleles can be distinguished by e.g. the HLA-A low resolution kit and/or the HLA-A\*02, HLA-A\*26 and HLA-A\*66 subtyping kits.

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“Instructions for Use” (IFU)

Lot No.: 7F1

Lot-specific information

**ALLELE CONFIRMATION STATUS**

Allele	Status <sup>1</sup>	Allele	Status <sup>1</sup>	Allele	Status <sup>1</sup>	Allele	Status <sup>1</sup>
A*33:01:01:01	Confirmed	A*33:11	Confirmed	A*33:60	Unconfirmed	A*33:110	Unconfirmed
A*33:01:01:02	Unconfirmed	A*33:12	Unconfirmed	A*33:61	Unconfirmed	A*33:111	Unconfirmed
A*33:01:02	Confirmed	A*33:13	Unconfirmed	A*33:62	Unconfirmed	A*33:112	Unconfirmed
A*33:01:03	Confirmed	A*33:14	Unconfirmed	A*33:63	Unconfirmed	A*33:113	Unconfirmed
A*33:01:04	Unconfirmed	A*33:15	Unconfirmed	A*33:64	Confirmed	A*33:114	Unconfirmed
A*33:01:05	Unconfirmed	A*33:16	Confirmed	A*33:65	Confirmed	A*33:115	Unconfirmed
A*33:01:06	Unconfirmed	A*33:17	Unconfirmed	A*33:66	Unconfirmed	A*33:116	Unconfirmed
A*33:01:07	Unconfirmed	A*33:18:01	Unconfirmed	A*33:67	Unconfirmed	A*33:117	Unconfirmed
A*33:01:08	Unconfirmed	A*33:18:02	Unconfirmed	A*33:68	Confirmed	A*33:118	Unconfirmed
A*33:01:09	Unconfirmed	A*33:19	Unconfirmed	A*33:69	Unconfirmed	A*33:119	Unconfirmed
A*33:01:10	Unconfirmed	A*33:20	Unconfirmed	A*33:70	Confirmed	A*33:120	Unconfirmed
A*33:03:01	Confirmed	A*33:21	Unconfirmed	A*33:71	Unconfirmed	A*33:121	Unconfirmed
A*33:03:02	Unconfirmed	A*33:22	Unconfirmed	A*33:72	Unconfirmed	A*33:122	Unconfirmed
A*33:03:03Q	Unconfirmed	A*33:23	Confirmed	A*33:73N	Confirmed	A*33:123N	Unconfirmed
A*33:03:04	Unconfirmed	A*33:24	Unconfirmed	A*33:74N	Unconfirmed	A*33:124	Unconfirmed
A*33:03:05	Unconfirmed	A*33:25	Confirmed	A*33:75	Unconfirmed	A*33:125	Unconfirmed
A*33:03:06	Unconfirmed	A*33:26	Confirmed	A*33:76	Unconfirmed	A*33:126	Unconfirmed
A*33:03:07	Confirmed	A*33:27	Confirmed	A*33:77	Confirmed	A*33:127	Unconfirmed
A*33:03:08	Unconfirmed	A*33:28	Unconfirmed	A*33:78	Unconfirmed	A*33:128	Unconfirmed
A*33:03:09	Unconfirmed	A*33:29	Unconfirmed	A*33:79	Unconfirmed	A*33:129N	Unconfirmed
A*33:03:10	Unconfirmed	A*33:30	Unconfirmed	A*33:80N	Unconfirmed	A*33:130	Unconfirmed
A*33:03:11	Unconfirmed	A*33:31	Unconfirmed	A*33:81	Unconfirmed		
A*33:03:12	Unconfirmed	A*33:32:01	Unconfirmed	A*33:82	Unconfirmed		
A*33:03:13	Unconfirmed	A*33:32:02	Unconfirmed	A*33:83	Unconfirmed		
A*33:03:14	Unconfirmed	A*33:33	Confirmed	A*33:84	Unconfirmed		
A*33:03:15	Unconfirmed	A*33:34	Confirmed	A*33:85	Unconfirmed		
A*33:03:16	Confirmed	A*33:35	Unconfirmed	A*33:86	Confirmed		
A*33:03:17	Unconfirmed	A*33:36	Confirmed	A*33:87	Unconfirmed		
A*33:03:18	Unconfirmed	A*33:37	Unconfirmed	A*33:88	Unconfirmed		
A*33:03:19	Unconfirmed	A*33:39	Unconfirmed	A*33:89	Unconfirmed		
A*33:03:20	Unconfirmed	A*33:40	Unconfirmed	A*33:90	Confirmed		
A*33:03:21	Unconfirmed	A*33:41	Unconfirmed	A*33:91	Unconfirmed		
A*33:03:22	Unconfirmed	A*33:42	Confirmed	A*33:92	Unconfirmed		
A*33:03:23	Confirmed	A*33:43	Unconfirmed	A*33:93	Unconfirmed		
A*33:03:24	Unconfirmed	A*33:44	Confirmed	A*33:94	Unconfirmed		
A*33:03:25	Confirmed	A*33:45	Unconfirmed	A*33:95	Unconfirmed		
A*33:03:26	Unconfirmed	A*33:46	Unconfirmed	A*33:96N	Unconfirmed		
A*33:03:27	Unconfirmed	A*33:47	Unconfirmed	A*33:97	Unconfirmed		
A*33:03:28	Unconfirmed	A*33:48	Unconfirmed	A*33:98	Unconfirmed		
A*33:03:29	Unconfirmed	A*33:49	Unconfirmed	A*33:99	Unconfirmed		
A*33:03:30	Unconfirmed	A*33:50	Unconfirmed	A*33:100	Unconfirmed		
A*33:03:31	Unconfirmed	A*33:51	Unconfirmed	A*33:101	Unconfirmed		
A*33:03:32	Unconfirmed	A*33:52	Confirmed	A*33:102	Unconfirmed		
A*33:04	Confirmed	A*33:53	Unconfirmed	A*33:103	Unconfirmed		
A*33:05	Confirmed	A*33:54	Confirmed	A*33:104	Unconfirmed		
A*33:06	Unconfirmed	A*33:55	Unconfirmed	A*33:105	Unconfirmed		
A*33:07	Confirmed	A*33:56	Unconfirmed	A*33:106	Unconfirmed		
A*33:08	Confirmed	A*33:57	Confirmed	A*33:107	Unconfirmed		
A*33:09	Confirmed	A*33:58	Confirmed	A*33:108	Unconfirmed		
A*33:10	Unconfirmed	A*33:59	Unconfirmed	A*33:109	Confirmed		

<sup>1</sup>Allele status “confirmed” or “unconfirmed” as listed on the IMGT/HLA web page 2017-August-10, release 3.29.0, [www.ebi.ac.uk/imgt/hla](http://www.ebi.ac.uk/imgt/hla).

**RESOLUTION IN HOMO- AND HETEROZYGOTES**

Results file with resolution in HLA-A\*33 homo- and heterozygotes is available upon request.

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Lot No.: **7F1**

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**SPECIFICITY TABLE**

**HLA-A\*33 SSP subtyping**

**Specificities and sizes of the PCR products of the 31+1 primer mixes used for HLA-A\*33 SSP subtyping**

Primer Mix	Size of spec. PCR product <sup>1</sup>	Size of control band <sup>2</sup>	Amplified HLA-A*33 alleles <sup>3</sup>	Other amplified HLA-A alleles
<b>1</b>	205 bp	<b>800 bp</b>	*33:01:01:01-33:01:10, 33:03:01-33:07, 33:10-33:20, 33:22-33:37, 33:39-33:50, 33:52, 33:54-33:68, 33:70-33:91, 33:93-33:108, 33:110-33:118, 33:120-33:130	*68:29
<b>2</b>	205 bp	<b>800 bp</b>	*33:01:01:01-33:01:10, 33:04-33:05, 33:07, 33:16, 33:27, 33:32:01-33:32:02, 33:34, 33:49-33:50, 33:64, 33:67-33:69, 33:89, 33:91-33:92, 33:109, 33:111, 33:121-33:122, 33:125, 33:127, 33:129N	*03:104, 31:88, 66:04
<b>3<sup>8</sup></b>	155 bp	1070 bp	*33:01:01:01-33:01:10, 33:05, 33:07, 33:16, 33:27, 33:32:01-33:32:02, 33:34, 33:49-33:50, 33:64, 33:67-33:69, 33:89, 33:91-33:92, 33:109, 33:111, 33:121-33:122, 33:125, 33:127, 33:129N	*02:332, 03:104, 24:220, 31:88, 66:04
<b>4</b>	210 bp	1070 bp	*33:03:01-33:03:32, 33:06, 33:08-33:15, 33:17, 33:20-33:26, 33:28-33:31, 33:33, 33:35-33:37, 33:39-33:48, 33:51-33:63, 33:65-33:66, 33:70-33:88, 33:90, 33:93-33:105, 33:107-33:108, 33:110, 33:112-33:120, 33:123N-33:124, 33:126, 33:128, 33:130	*01:145, 02:41, 02:65, 02:80, 02:117, 02:135, 02:152, 02:289:01, 02:304, 02:309, 02:454, 03:103, 03:282, 11:116, 11:140, 23:45, 24:62, 25:01:01:01-25:42N, 25:44-25:45, 26:01:01:01-26:39, 26:41-26:43:02, 26:45-26:145N, 29:32, 31:01:02:01-31:02, 31:05, 31:07-31:68, 31:70-31:87, 31:89-31:127, 32:01:01:01-32:03, 32:05-32:29, 32:31, 32:33:01-32:40, 32:42-32:47, 32:49-32:100, 32:102-32:103, 34:01:01-34:01:02, 34:05-34:06, 34:11-34:12, 34:14, 34:16-34:17, 43:01, 66:01:01:01-66:03, 66:05-66:28N, 74:01:01-74:28
<b>5<sup>4</sup></b>	90 bp	<b>800 bp</b>	*33:04, 33:33	*26:68
<b>6<sup>4,5</sup></b>	105 bp 170 bp	1070 bp	*33:20 *33:05, 33:58	*31:94 *29:12, 29:92
<b>7<sup>4</sup></b>	75 bp 105 bp	1070 bp	*33:21, 33:53 *33:06	

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Lot No.: **7F1**

Lot-specific information

<b>8</b>	230 bp		*33:51, 33:119	*11:98, 11:250, 66:15, 68:04:01
	160 bp	1070 bp	*33:07	*02:444, 31:54
	235 bp		*33:24	*02:243:01-02:243:02, 29:19, 29:48
<b>9<sup>7</sup></b>	150 bp	<b>800 bp</b>	*33:14, 33:22, 33:58	*01:20, 01:66, 01:130, 02:24:01-02:24:02, 02:137, 02:309, 02:503, 03:95, 26:22, 29:12, 29:22, 29:92, 30:47, 31:99, 66:09, <b>C*02:74</b>
	185 bp		*33:08-33:09	*02:243:01-02:243:02, 29:48
<b>10<sup>6,7</sup></b>	135 bp	<b>800 bp</b>	*33:22	*02:24:02, 29:22, 31:99
	215 bp		*33:08, 33:53	*02:243:01-02:243:02, 24:82, 29:48, 31:02, 31:07-31:08, 31:91, 31:109
	285 bp		*33:13	*01:143, 11:43, 29:66, 31:03, <b>C*07:449</b>
<b>11<sup>5</sup></b>	165 bp	1070 bp	*33:10, 33:25	*23:03:01, 24:21:03, 24:208, 29:03, 29:33, 31:05, 32:13
<b>12<sup>4</sup></b>	105 bp	1070 bp	*33:36, 33:80N	*11:137:01N, 26:60N
	235 bp		*33:11, 33:125	*03:205, 11:43, 31:66, 31:89, 31:115, 68:29
<b>13<sup>4,5</sup></b>	95 bp	1070 bp	*33:12	
	165 bp		*33:25	
<b>14<sup>4</sup></b>	120 bp	1070 bp	*33:15, 33:84	*02:140, 26:99, 31:48
	335 bp		*33:19	*02:10, 02:17:01-02:17:04, 02:39, 02:108, 02:110, 02:148, 02:242, 02:244, 02:268, 02:300, 02:303, 02:398, 02:453, 02:604, 02:617, 02:628, 02:630, 02:657, 02:680, 03:15, 03:19, 11:139, 24:04, 24:19, 24:28, 24:44, 24:89, 24:109, 24:129, 24:290, 29:07, 29:49, 31:29
<b>15<sup>6</sup></b>	140 bp	1070 bp	*33:16, 33:23	
	215 bp		*33:65	
	255 bp		*33:64	
<b>16<sup>4</sup></b>	95 bp	1070 bp	*33:36	
	210 bp		*33:21	
	245 bp		*33:17	
<b>17</b>	145 bp	1070 bp	*33:18:01-33:18:02, 33:26	*03:42, 03:133, 23:53, 23:70, 29:01:01:01-29:04, 29:06-29:31, 29:34, 29:36-29:50, 29:52-29:70, 29:72-29:76, 29:78N, 29:81, 29:84-29:86, 29:88-29:103, 31:03-31:04, 31:06, 32:30:01-32:30:02, 32:32
	215 bp		*33:65	
<b>18<sup>4</sup></b>	100 bp	1070 bp	*33:27	
	145 bp		*33:57	
	240 bp		*33:82	
<b>19<sup>4</sup></b>	120 bp	1070 bp	*33:28	*03:01:18, 03:22:02, 29:83, 31:110, 32:10, 32:63, 74:28, <b>B*15:02:07, B*15:17:03, C*02:02:15, C*03:03:20</b>
<b>20<sup>4</sup></b>	90 bp	1070 bp	*33:29	
	225 bp		*33:39	*02:480, 32:53
<b>21<sup>4</sup></b>	115 bp	1070 bp	*33:30	*02:342
	245 bp		*33:73N, 33:82, 33:129N	
<b>22<sup>4</sup></b>	120 bp	1070 bp	*33:31, 33:84	*02:241, 02:684, 26:24



101.432-12 – including *Taq* polymerase, IFU-01  
101.432-12u – without *Taq* polymerase, IFU-02

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“Instructions for Use” (IFU)

Lot No.: **7F1**

Lot-specific information

	255 bp		*33:44	
	545 bp		*33:123N	*01:166
<b>23</b>	180 bp	1070 bp	*33:32:01-33:32:02, 33:52	*02:332, 24:220, <b>B*51:136</b>
<b>24<sup>4</sup></b>	95 bp	1070 bp	*33:34	*03:01:18, 11:01:28, 11:01:77, 24:21:03, 24:208, 29:09, 29:33, 31:24, 32:33:01
	205 bp		*33:54	*29:59
<b>25<sup>5</sup></b>	215 bp	1070 bp	*33:69, 33:83, 33:109	
	245 bp		*33:129N	
<b>26</b>	150 bp	1070 bp	*33:77	
	190 bp		*33:74N, 33:111	
<b>27<sup>5</sup></b>	200 bp	1070 bp	*33:74N, 33:85, 33:92, 33:109	
<b>28</b>	230 bp	1070 bp	*33:70, 33:90	*01:84, 02:214, 03:145:02, 11:54
	545 bp		*33:123N	*01:166
<b>29<sup>4</sup></b>	105 bp	1070 bp	*33:42	*02:292
<b>30<sup>4</sup></b>	120 bp	1070 bp	*33:03:03Q	*01:01:38L, 24:02:03Q
	260 bp		*33:86	*03:265, 32:51
<b>31</b>	155 bp	1070 bp	*33:96N	*01:52:01N, 26:107N
	220 bp		*33:90	*01:84, 02:214
	260 bp		*33:68	*24:114, 29:61, 32:98
<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-A\*33 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 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>HLA-specific PCR products shorter than 125 base pairs have a lower intensity and are less sharp than longer PCR products.

<sup>5</sup>Primer mixes 6, 11, 13, 25 and 27 may have tendencies of unspecific amplifications.

<sup>6</sup>Primer mixes 10 and 15 have a tendency to giving rise to primer oligomer formation.

<sup>7</sup>Primer mixes 9 and 10 may give rise to a long unspecific amplification product of approximately 640 bp. This should be disregarded when interpreting the HLA-A\*33 typings.

<sup>8</sup>Primer mix 3 may give rise to a lower yield of HLA-specific PCR product than the other A\*33 primer mixes.

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**Lot-specific information**

<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 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.

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Lot-specific information  
**PRIMER SPECIFICATION**

Well No.	1	2	3	4	5	6	7	8	9	10	11	12
Length of spec. PCR product	205	205	155	210	90	105	75	160	150	135	165	105
						170	105	235	185	215		235
							230			285		
Length of int. pos. control <sup>1</sup>	800	800	1070	1070	800	1070	1070	1070	800	800	1070	1070
5'-primer(s) <sup>2</sup>	97	418	468	414	414	97	98	97	97	97	448	97
	5'-TCA 3'	5'-Agg 3'	5'-TCT 3'	5'-CAg 3'	5'-CAg 3'	5'-TCA 3'	5'-CTC 3'	5'-TCA 3'	5'-TCA 3'	5'-TCA 3'	5'-CCT 3'	5'-TCA 3'
				414		413	103	413	355	355	652	355
				5'-CAg 3'		5'-CCA 3'	5'-CCT 3'	5'-CCA 3'	5'-CCg 3'	5'-CCg 3'	5'-CTg 3'	5'-CCA 3'
							228					
							5'-ATg 3'					
							257					
							5'-CgA 3'					
3'-primer(s) <sup>3</sup>	259	583	583	583	463	221	290	292	218	270	570	290
	5'-gTT 3'	5'-gTg 3'	5'-gTg 3'	5'-gTA 3'	5'-gCT 3'	5'-ACA 3'	5'-CAA 3'	5'-gTg 3'	5'-gCC 3'	5'-ACT 3'	5'-CCg 3'	5'-CAg 3'
						233		530	221	341	778	407
						5'-CCC 3'		5'-CCT 3'	5'-ACA 3'	5'-CgT 3'	5'-TgT 3'	5'-ACT 3'
						475			240	448		426
						5'-Cgg 3'			5'-ggA 3'	5'-CAA 3'		5'-TCC 3'
									453			
									5'-TCg 3'			
Well No.												

Well No.	13	14	15	16	17	18	19	20	21	22	23	24
Length of spec. PCR product	95	120	140	95	145	100	120	90	115	120	180	95
	165	335	215	210	215	145		225	245	255		205
			255	245		240				545		
Length of int. pos. control <sup>1</sup>	1070	1070	1070	1070	1070	1070	1070	1070	1070	1070	1070	1070
5'-primer(s) <sup>2</sup>	395	317	85	97	85	345	448	397	463	321	112	448
	5'-gCC 3'	5'-gCg 3'	5'-CCg 3'	5'-TCA 3'	5'-CCg 3'	5'-TT 3'	5'-CCT 3'	5'-gCg 3'	5'-TgT 3'	5'-g.C 3'	5'-CCT 3'	5'-CCT 3'
	652	652	158	355	161	390		649	629	652	453	
	5'-CTg 3'	5'-CTg 3'	5'-ggg 3'	5'-CCA 3'	5'-CgC 3'	5'-gAg 3'		5'-ACA 3'	5'-CAA 3'	5'-CTg 3'	5'-AAA 3'	
			370		413	632			629		453	
			5'-ATC 3'		5'-CCg 3'	5'-gAT 3'			5'-CAg 3'		5'-AAg 3'	
			482						632			
			5'-ggC 3'						5'-gAT 3'			
3'-primer(s) <sup>3</sup>	448	368	259	265	259	448	527	448	538	583	259	502
	5'-CAA 3'	5'-CAA 3'	5'-gTT 3'	5'-CCC 3'	5'-gTT 3'	5'-CAA 3'	5'-CCT 3'	5'-CAA 3'	5'-CAA 3'	5'-gTA 3'	5'-gTT 3'	5'-CTT 3'
	778	727	583	299	524	831		831	831	728	583	614
	5'-TgT 3'	5'-CCA 3'	5'-gTg 3'	5'-CCg 3'	5'-CAC 3'	5'-TCC 3'		5'-TCC 3'	5'-TCC 3'	5'-CCT 3'	5'-gTg 3'	5'-TgA 3'
		742		407						742		
		5'-CTC 3'		5'-ACT 3'						5'-CTC 3'		
										866		
										5'-gAT 3'		
Well No.												

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Lot-specific information

Well No.	25	26	27	28	29	30	31
Length of spec. PCR product	215	150	200	230	105	120	155
	245	190		545		260	220
							260
Length of int. pos. control <sup>1</sup>	1070	1070	1070	1070	1070	1070	1070
5'-primer(s) <sup>2</sup>	94	652	95	321	277	28	365
	5' -ATC 3'	5' -CTg 3'	5' -TTC 3'	5' -g.C 3'	5' -AgT 3'	5' -TCC 3'	5' -gAC 3'
	95		97	391		627	404
	5' -TTC 3'		5' -TCg 3'	5' -ACA 3'		5' -CCC 3'	5' -CCC 3'
	629		652	404			470
	5' -CAg 3'		5' -CTg 3'	5' -CCC 3'			5' -TTA 3'
	646						
	5' -ACA 3'						
3'-primer(s) <sup>3</sup>	259	761	259	583	341	118	583
	5' -gTT 3'	5' -Cgg 3'	5' -gTT 3'	5' -gTA 3'	5' -Cgg 3'	5' -gCT 3'	5' -gTA 3'
	831	803	803			705	583
	5' -TCC 3'	5' -CCT 3'	5' -CCT 3'			5' -TCT 3'	5' -gTg 3'
		806	814				
		5' -ACA 3'	5' -CAA 3'				
Well No.							

<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.

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Lot-specific information

CELL LINE VALIDATION SHEET																				
HLA-A*33 SSP subtyping kit <sup>2</sup>																				
				Well																
				1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	
				Prod No.:	201442301	201442302	201442303	201442304	201442305	201442306	201785707	201442308	201442309	201442310	201442311	201442312	201442313	201442314	201442315	201442316
	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	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

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Lot-specific information

CELL LINE VALIDATION SHEET																			
HLA-A*33 SSP subtyping kit <sup>2</sup>																			
				Well															
				17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	
				Prod No.:	201442317	201785718	201442319	201785720	201785721	201785722	201442323	201442324	201785725	201785726	201785727	201785728	201442329	201662830	201785731
	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	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

**101.432-12 – including *Taq* polymerase, IFU-01**  
**101.432-12u – without *Taq* polymerase, IFU-02**

Visit [www.olerup.com](http://www.olerup.com) for  
**“Instructions for Use” (IFU)**

**Lot No.: 7F1**

**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.

No DNAs carrying the alleles to be amplified by primer solutions 5, 7 to 13, 15, 16, 18 to 21 and 23 to 31 were available. The specificities of the primers in primer solutions 5, 7 to 13, 19, 23, 24 and 27, were tested by separately adding additional 5'-primers respectively 3'-primers. In primer solutions 16, 26 and 30 it was only possible to test the 5'-primers, the 3'-primers were not possible to test. In primer solution 15, 18, 20, 21, 25, 28, 29 and 31 it was only possible to test the 3'-primers, the 5'-primers were not possible to test. In primer solutions 7, 13, 17, 22, 23 and 27 one or two 5'-primers were not possible to test, and in primer solutions 6, 8, 9, 11 to 14, 22, 24 and 27 one or two 3'-primers were not possible to test. Additional primers in primer solutions 6, 14, 17 and 22 were tested by separately adding one additional 5'-primer or one additional 3'-primer.

101.432-12 – including *Taq* polymerase, IFU-01  
101.432-12u – without *Taq* polymerase, IFU-02

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Lot No.: **7F1**

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