

101.421-06 – including *Taq* polymerase, IFU-01
101.421-06u – without *Taq* polymerase, IFU-02

Visit <https://labproducts.caredx.com> for
“Instructions for Use” (IFU)

Lot No.: **7K4**

Lot-specific Information
Olerup SSP® HLA-A*23

Product number:	101.421-06 – including <i>Taq</i> polymerase 101.421-06u – without <i>Taq</i> polymerase
Lot number:	7K4
Expiry date:	2024-01-01
Number of tests:	6
Number of wells per test:	35+1
Storage - pre-aliquoted primers:	dark at -20°C
- PCR Master Mix:	-20°C
- Adhesive PCR seals	RT
- Product Insert	RT

This Product Description is only valid for Lot No. 7K4.

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*23 Lot (0G9)**

- The product documentation has been updated for new alleles of IMGT 3.38.0
- The kit resolution focuses on common and well documented (CWD) alleles¹.

¹As described in section Uniquely Identified Alleles.

The HLA-A*23 specificity and interpretation tables have been updated for the HLA alleles described since the previous *Olerup SSP®* HLA-A*23 lot was made (**Lot No. 0G9**).

The HLA-A*23 primer set is unchanged compared to the previous *Olerup SSP®* HLA-A*23 lot (**Lot No. 0G9**)

¹S. J. Mack, P. Cano, J. A. Hollenbach et al.
Common and well-documented HLA alleles: 2012 update to the CWD catalogue. Tissue Antigens, 2013, 81, 194–203

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Lot-specific Information

Well **36** contains Negative Control primer pairs, that will amplify a majority of the *Olerup SSP*® HLA Class I, DRB, DQB1, DPB1 and DQA1 amplicons as well as all the amplicons generated by the control primer pairs matching the human growth hormone gene.

HLA-specific PCR product sizes range from 75 to 200 base pairs.

The PCR product generated by the positive control primer pair is 200 base pairs.

Length of PCR product	105	200	105	80	75	80	85
5'-primer¹	164	340	440	45	45	43	36
	5'-CAC ^{3'}	5'-Agg ^{3'}	5'-TTA ^{3'}	5'-Tgg ^{3'}	5'-Tgg ^{3'}	5'-Tgg ^{3'}	5'-TAC ^{3'}
							36
							5'-TAT ^{3'}
3'-primer²	231	2nd I	507	59	58	57	47
	5'-TgC ^{3'}	5'-AAA ^{3'}	5'-TTg ^{3'}	5'-CTC ^{3'}	5'-ggC ^{3'}	5'-CTC ^{3'}	5'-ACA ^{3'}
							48
							5'-gCA ^{3'}
							48
							5'-gCC ^{3'}
							52
							5'-TgT ^{3'}
A*	+	+	+				
B*	+	+	+				
C*	+	+	+				
DRB1				+	+		
DRB3				+	+		
DRB5				+			
DQB1					+		
DPB1						+	
DQA1							+

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

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

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Lot-specific Information
PRODUCT DESCRIPTION

HLA-A*23 SSP subtyping

CONTENT

The primer set contains 5'- and 3'-primers for identifying the A*23:01 to A*23:102 alleles.

PLATE LAYOUT

Each test consists of 36 PCR reactions in a 48 well cut PCR plate. Wells 37 to 48 are empty.

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	32
33	34	35	NC	empty	empty	empty	empty
empty	empty	empty	empty	empty	empty	empty	empty

The 48 well cut PCR plate is marked with ‘HLA-A*23’ in silver/gray ink.

Well No. 1 is marked with the Lot Number ‘7K4’.

Wells 1 to 35 – HLA-A*23 high resolution primers.

Well 36 – 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 48 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*23 alleles will be amplified by some primer mixes. For further details see Specificity Table.

UNIQUELY IDENTIFIED ALLELES

All the HLA-A*23 alleles, i.e. **A*23:01 to A*23:102 alleles**, recognized by the HLA Nomenclature Committee in October 2019^{1,2} will be amplified by the primers in the HLA-A*23 subtyping kit.

The HLA-A*23 kit enables separation of the confirmed HLA-A*23 alleles as listed in the IMGT/HLA database 3.30.0. 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*23 alleles is listed below.

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Lot-specific Information

The HLA-A*23 kit also enables identification of many null and alternatively expressed alleles.

The following HLA-A*23 alleles can be distinguished by the different sizes of the HLA-specific PCR product:

Alleles	Primer mix	Alleles	Primer mix
A*23:08N, 23:22	7	A*23:16, 23:29	20
A*23:09, 23:26	8	A*23:38N, 23:56	30
A*23:10, 23:23	10	A*23:41, 23:42	25
A*23:13, 23:33	17		

¹HLA-A alleles listed on the IMGT/HLA web page 2019-October-17, release 3.38.0, www.ebi.ac.uk/imgt/hla.

²Alleles 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>.

ALLELE CONFIRMATION STATUS

Allele	Status ¹	Allele	Status ¹	Allele	Status ¹	Allele	Status ¹
A*23:01:01:01	Confirmed	A*23:08N	Confirmed	A*23:37:01	Unconfirmed	A*23:66	Unconfirmed
A*23:01:01:02	Confirmed	A*23:09	Unconfirmed	A*23:37:02	Unconfirmed	A*23:67	Unconfirmed
A*23:01:01:03	Unconfirmed	A*23:10	Unconfirmed	A*23:38N	Unconfirmed	A*23:68	Confirmed
A*23:01:01:04	Unconfirmed	A*23:11N	Confirmed	A*23:39	Unconfirmed	A*23:70	Unconfirmed
A*23:01:02	Confirmed	A*23:12	Confirmed	A*23:40	Unconfirmed	A*23:71	Unconfirmed
A*23:01:03	Confirmed	A*23:13	Unconfirmed	A*23:41	Unconfirmed	A*23:72	Unconfirmed
A*23:01:04	Confirmed	A*23:14:01	Unconfirmed	A*23:42	Unconfirmed	A*23:73	Unconfirmed
A*23:01:05	Unconfirmed	A*23:14:02	Unconfirmed	A*23:43	Confirmed	A*23:74	Unconfirmed
A*23:01:06	Confirmed	A*23:15	Confirmed	A*23:44	Unconfirmed	A*23:75	Unconfirmed
A*23:01:07	Unconfirmed	A*23:16	Unconfirmed	A*23:45	Unconfirmed	A*23:76	Unconfirmed
A*23:01:08	Unconfirmed	A*23:17	Confirmed	A*23:46	Unconfirmed	A*23:77	Unconfirmed
A*23:01:09	Confirmed	A*23:18	Confirmed	A*23:47	Confirmed	A*23:78	Unconfirmed
A*23:01:10	Confirmed	A*23:19N	Confirmed	A*23:48	Confirmed	A*23:79	Unconfirmed
A*23:01:11	Unconfirmed	A*23:20	Unconfirmed	A*23:49	Confirmed	A*23:80	Unconfirmed
A*23:01:12	Unconfirmed	A*23:21	Confirmed	A*23:50	Unconfirmed	A*23:81	Unconfirmed
A*23:01:13	Unconfirmed	A*23:22	Unconfirmed	A*23:51	Unconfirmed	A*23:82	Unconfirmed
A*23:01:14	Unconfirmed	A*23:23	Unconfirmed	A*23:52	Unconfirmed	A*23:83	Unconfirmed
A*23:01:15	Unconfirmed	A*23:24	Confirmed	A*23:53	Confirmed		
A*23:01:16	Unconfirmed	A*23:25	Unconfirmed	A*23:54	Unconfirmed		
A*23:01:17	Unconfirmed	A*23:26	Unconfirmed	A*23:55	Unconfirmed		
A*23:01:18	Unconfirmed	A*23:27	Confirmed	A*23:56	Confirmed		
A*23:01:19	Unconfirmed	A*23:28	Confirmed	A*23:57	Unconfirmed		
A*23:01:20	Unconfirmed	A*23:29	Unconfirmed	A*23:58	Unconfirmed		
A*23:02	Unconfirmed	A*23:30	Confirmed	A*23:59	Confirmed		
A*23:03:01	Unconfirmed	A*23:31	Confirmed	A*23:60	Unconfirmed		
A*23:03:02	Unconfirmed	A*23:32	Unconfirmed	A*23:61	Unconfirmed		
A*23:04	Confirmed	A*23:33	Unconfirmed	A*23:62	Unconfirmed		
A*23:05	Confirmed	A*23:34	Unconfirmed	A*23:63	Unconfirmed		
A*23:06	Confirmed	A*23:35	Unconfirmed	A*23:64	Unconfirmed		
A*23:07N	Unconfirmed	A*23:36	Unconfirmed	A*23:65	Confirmed		

¹Allele status “confirmed” or “unconfirmed” as listed on the IMGT/HLA web page 2017-October-27, release 3.30.0, www.ebi.ac.uk/imgt/hla.

RESOLUTION IN HOMO- AND HETEROZYGOTES

Results file with resolution in HLA-A*23 homo- and heterozygotes is available upon request.

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Lot-specific Information
SPECIFICITY TABLE
HLA-A*23 SSP subtyping

Specificities and sizes of the PCR products of the 35+1 primer mixes used for HLA-A*23 SSP subtyping

Primer Mix	Size of spec. PCR product ¹	Size of control band ²	Amplified HLA-A*23 alleles ³	Other amplified HLA Class I alleles
1	210 bp	800 bp	*23:01:01:01-23:01:28, 23:03:01-23:65, 23:67-23:68, 23:70-23:102	*02:17:02:01-02:17:04, 02:108, 02:110, 02:268, 02:300, 02:303, 02:617, 02:657, 03:347, 24:13:01-24:13:02, 24:18, 24:24, 24:94, 24:188, 24:207:01-24:207:02, 24:228, 24:315, 24:355, 24:392, 29:07, 29:49, 31:29, 33:187, B*18:27
2	160 bp	1070 bp	*23:01:01:01-23:02, 23:05-23:42, 23:44-23:68, 23:71-23:78, 23:80-23:82, 23:84N-23:102	*01:88:02-01:88:03, 01:236, 02:19, 02:36-02:37, 02:54, 02:255, 02:417, 02:723, 02:804, 02:848, 02:883, 11:209, 24:02:01:01-24:02:41, 24:02:43-24:02:69, 24:02:71-24:02:132, 24:04-24:09N, 24:11N, 24:13:01-24:15, 24:17, 24:19-24:20:01:02, 24:24-24:32, 24:34-24:64, 24:66-24:74:02, 24:76-24:93, 24:95-24:109, 24:111-24:124, 24:126-24:137, 24:139-24:157, 24:159-24:166, 24:168-24:203, 24:205-24:206, 24:209, 24:212-24:218, 24:220-24:282, 24:284-24:288, 24:290-24:298, 24:301-24:314, 24:316-24:338, 24:341-24:348, 24:350-24:356, 24:358-24:373, 24:375-24:386, 24:388N-24:389N, 24:391-24:440, 24:442-24:458, 24:460, 24:462-24:470, 24:472-24:477, 33:19, 68:26, 68:65, 68:115, 68:131
3⁴	125 bp	800 bp	*23:03:01-23:03:02, 23:70, 23:83	*01:301Q, 03:347, 11:139, 24:21:01, 24:21:03, 24:208:01-24:208:02, 29:07, 29:49, 31:29, 33:187, B*18:27
	270 bp		*23:25	*24:313:02
4	200 bp	1070 bp	*23:28	*24:77
5	230 bp	800 bp	*23:06	*31:36
6⁵	470 bp	1070 bp	*23:07N	*01:04:01:01N-01:04:01:02N, 03:21N, 11:21N, 24:11N, 30:132N
7⁴	95 bp 145 bp 205 bp	800 bp	*23:08N *23:39 *23:22	*02:82N, 02:773N *30:22, 30:104
8⁶	140 bp 170 bp 215 bp	1070 bp	*23:26, 23:39 *23:48 *23:09	*30:22, 30:104 *24:403 *01:02, 01:20, 01:188, 01:190, 01:307, 24:129
9	235 bp	1070 bp	*23:05	*24:25, 24:308
10	135 bp 240 bp	800 bp	*23:23 *23:10, 23:43	B*18:27 *01:301Q, 11:139 ^w , 24:10:01:01 ^w -24:10:02 ^w , 24:46, 24:210 ^w , 24:300 ^w , 24:340 ^w
11	200 bp 270 bp	800 bp	*23:11N *23:32	B*51:293
12	190 bp	1070 bp	*23:12	*24:30, 24:42, 24:309, 25:11, 32:08

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				30:11:01-30:11:02, 30:14L-30:20, 30:23-30:26, 30:30-30:31, 30:35-30:43, 30:48-30:49, 30:52-30:54, 30:56, 30:58-30:60, 30:62-30:63, 30:65, 30:72-30:75, 30:78N, 30:81-30:83, 30:86-30:89, 30:91-30:98, 30:102, 30:104, 30:106, 30:109-30:116, 30:118, 30:120, 30:122-30:123N, 30:125-30:126, 30:128-30:132N, 30:134-30:138, 30:140-30:143, 30:145N, 30:147-30:148, 30:154, 30:159, 30:161, 68:45, 68:117
18⁴	120 bp	1070 bp	*23:01:01:01-23:01:28, 23:02 ^w , 23:04-23:23, 23:25-23:33, 23:35-23:56, 23:58-23:65, 23:67-23:68, 23:71-23:82, 23:84N-23:102	*02:01:22, 02:06:04, 02:40:01-02:40:02, 02:51, 02:99:02, 02:130, 02:171:01, 24:13:02, 24:24, 24:207:02, 24:315, 24:392, 29:101:01, 31:67-31:68, 32:28, 32:66, 33:32:01, 68:08:01:01-68:08:01:02, 68:51 ^w , 68:168, B*40:323:02, B*51:136, C*07:204:01, C*07:482
19^{4,7}	120 bp 195 bp	1070 bp	*23:15 *23:46	*24:66, 24:308, 30:75
20⁵	130 bp 195 bp 230 bp 270 bp	800 bp	*23:29 *23:46 *23:16 *23:32	*24:128 *24:66, 24:308, 30:75 *24:297 B*51:293
21⁴	90 bp	800 bp	*23:01:01:01-23:01:28, 23:02 [?] -23:03:01 [?] , 23:03:02-23:04, 23:06-23:07N, 23:08N [?] , 23:09, 23:10 [?] , 23:11N, 23:12 [?] -23:14:02 [?] , 23:15, 23:16 [?] , 23:18-23:19N, 23:20 [?] -23:25 [?] , 23:26, 23:27 [?] -23:29 [?] , 23:30, 23:31 [?] -23:37:02 [?] , 23:38N, 23:39 [?] -23:55 [?] , 23:56, 23:57 [?] -23:64 [?] , 23:65, 23:66 [?] -23:67 [?] , 23:68, 23:70 [?] -23:75 [?] , 23:76-23:80, 23:81 [?] -23:83 [?] , 23:85-23:95, 23:97-23:102	*24:463
22^{4,5}	90 bp	1070 bp	*23:31, 23:45, 23:53, 23:70	*01:301Q, 03:347, 11:139, 24:17, 24:41, 24:62, 24:106, 24:208:01-24:208:02, 24:330, 29:07, 29:49, 31:29, 33:187
23^{4,7}	290 bp 80 bp	1070 bp	*23:19N *23:45	*02:41, 02:80, 02:117, 02:289:01, 02:304, 02:454, 02:872, 24:62, 26:10, 31:67-31:68, 32:28, 32:66, 33:32:01
24⁴	170 bp 105 bp 180 bp 240 bp	800 bp	*23:20, 23:58 *23:49 *23:21 *23:40	*24:444, 30:06, C*04:361 C*04:344 *24:302
25⁴	95 bp 205 bp	800 bp	*23:41 *23:42	*02:221, 29:15, 31:78
26⁴	80 bp	1070 bp	*23:36	*24:32, 32:05, 32:79
27^{4,5}	85 bp	1070 bp	*23:53, 23:70	*01:301Q, 03:347, 11:139, 24:17, 24:41, 24:208:01-24:208:02, 29:07, 29:49

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	200 bp		*23:34, 23:57	*02:17:02:01-02:17:04, 02:108, 02:110, 02:268, 02:300, 02:303, 02:617, 02:657, 24:13:01, 24:94, 24:188, 24:207:01, 24:228
	285 bp		*23:35	
28	385 bp	800 bp	*23:02 [?] -23:03:01 [?] , 23:05, 23:08N [?] , 23:10 [?] , 23:12 [?] -23:14:02 [?] , 23:16 [?] , 23:17:01:01-23:17:02, 23:20 [?] -23:25 [?] , 23:27 [?] -23:29 [?] , 23:31 [?] -23:37:02 [?] , 23:39 [?] -23:55 [?] , 23:57 [?] -23:64 [?] , 23:66 [?] -23:67 [?] , 23:70 [?] -23:75 [?] , 23:81 [?] -23:83 [?] , 23:84N, 23:96	*02:19, 02:804, 24:02:01:01-24:02:09, 24:02:11-24:02:96, 24:02:97 [?] , 24:02:98-24:02:122, 24:02:124-24:05:02, 24:06 [?] , 24:07:01:01-24:10:01:02, 24:10:02 [?] , 24:11N, 24:13:01 [?] , 24:13:02-24:14:01:04, 24:15 [?] , 24:17-24:18, 24:19 [?] , 24:20:01:01-24:20:01:02, 24:21:01 [?] -24:21:02 [?] , 24:21:03-24:23, 24:24 [?] , 24:25, 24:26 [?] -24:27 [?] , 24:28-24:32, 24:34 [?] , 24:35-24:37, 24:38 [?] -24:43 [?] , 24:44, 24:45N [?] -24:47 [?] , 24:48N, 24:49 [?] , 24:50, 24:51 [?] -24:52 [?] , 24:53-24:54, 24:55 [?] , 24:56-24:58, 24:59 [?] -24:60N [?] , 24:61, 24:62 [?] -24:64 [?] , 24:66 [?] -24:67 [?] , 24:68, 24:69 [?] -24:71 [?] , 24:72, 24:73 [?] -24:78 [?] , 24:79, 24:80 [?] , 24:81, 24:82 [?] -24:85 [?] , 24:86N, 24:87 [?] -24:89 [?] , 24:90:01N, 24:90:02N [?] -24:94 [?] , 24:95, 24:96 [?] -24:115 [?] , 24:116, 24:117 [?] -24:121 [?] , 24:122, 24:123 [?] -24:127 [?] , 24:128-24:129, 24:130 [?] -24:131 [?] , 24:132N, 24:133 [?] -24:134 [?] , 24:135:01, 24:135:02 [?] , 24:136, 24:137 [?] -24:141 [?] , 24:142:01, 24:142:02 [?] , 24:143, 24:144 [?] -24:147 [?] , 24:148, 24:149 [?] -24:151 [?] , 24:152, 24:153 [?] -24:162 [?] , 24:163N, 24:164 [?] -24:172:01 [?] , 24:172:02, 24:173 [?] -24:182 [?] , 24:183N, 24:184 [?] -24:193 [?] , 24:194, 24:195 [?] -24:207:01 [?] , 24:207:02, 24:208:01 [?] , 24:208:02, 24:209 [?] -24:210 [?] , 24:212 [?] -24:214 [?] , 24:215, 24:216 [?] -24:225:01 [?] , 24:225:02, 24:226:01 [?] , 24:226:02, 24:227 [?] -24:230 [?] , 24:231, 24:232N [?] , 24:233-24:235, 24:236 [?] -24:237 [?] , 24:238, 24:239 [?] -24:247 [?] , 24:248-24:252N, 24:253 [?] , 24:254-24:256, 24:257 [?] -24:258 [?] , 24:259-24:273, 24:274 [?] -24:277 [?] , 24:278N, 24:279 [?] , 24:280, 24:281 [?] -24:285 [?] , 24:286, 24:287 [?] -24:292 [?] , 24:293-24:294Q, 24:295 [?] -24:301 [?] , 24:302, 24:303N [?] -24:308 [?] , 24:309, 24:310:01 [?] -24:312N [?] , 24:313:01, 24:313:02 [?] , 24:314, 24:315 [?] -24:320 [?] , 24:321, 24:322 [?] -24:330 [?] , 24:331, 24:332 [?] -24:340 [?] , 24:341, 24:342 [?] -24:351 [?] , 24:352-24:353, 24:354 [?] , 24:355-24:359N, 24:360 [?] , 24:361-24:364, 24:365 [?] -24:366 [?] , 24:367-24:373, 24:374 [?] -24:386 [?] , 24:388N [?] , 24:389N, 24:390 [?] -24:393 [?] , 24:394, 24:395 [?] , 24:396N-24:412, 24:413 [?] , 24:414, 24:415 [?] -24:416 [?] , 24:417-24:420, 24:421 [?] , 24:422-24:431, 24:432 [?] , 24:433N-24:444, 24:445N [?] -24:446 [?] ,

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				24:447Q-24:451, 24:452 [?] , 24:453-24:459, 24:461-24:462, 24:464, 24:466-24:470, 24:472-24:477, 33:19 [?]
29	170 bp	1070 bp	*23:48	*24:403
	200 bp		*23:37:01-23:37:02	*24:273
	240 bp		*23:40	*24:302
	290 bp		*23:47	*24:234, 24:339, 24:347:02
30⁴	110 bp	1070 bp	*23:56	
	245 bp		*23:38N	*24:61
31	185 bp	1070 bp	*23:44	
32⁴	85 bp	1070 bp	*23:59	*01:203, 02:671, 11:166, 30:56, 31:85, 32:72, 80:01:01:01-80:06
33	165 bp	1070 bp	*23:65	*24:298
34⁴	90 bp	1070 bp	*23:18	*24:267
	170 bp		*23:58	
35⁴	105 bp	800 bp	*23:49	*23:49, 24:444, 30:06, C*04:361
	260 bp		*23:27	*24:255
36⁸	-	-	Negative Control	

¹Alleles are assigned by the presence of specific PCR product(s). However, the sizes of the specific PCR products may be helpful in the interpretation of HLA-A*23 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.

²The 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.

³For 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.

⁴HLA-specific PCR products shorter than 125 base pairs have a lower intensity and are less sharp than longer PCR products.

⁵Primer mixes 6, 20, 22 and 27 may give rise to a lower yield of HLA-specific PCR product than the other A*23 primer mixes.

⁶Primer mixes 8 and 14 may have tendencies of unspecific amplifications.

⁷Primer mixes 19 and 23 have tendencies to giving rise to primer oligomer formation.

⁸Primer mix 36 contains a negative control, which will amplify a majority 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 200 base pairs.

Abbreviations

‘w’, may be weakly amplified.

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

101.421-06 – including *Taq* polymerase, IFU-01
101.421-06u – without *Taq* polymerase, IFU-02

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Lot No.: 7K4

Lot-specific Information
PRIMER SPECIFICATION

Well No.	1	2	3	4	5	6	7	8	9	10	11	12
Length of spec. PCR product	210	160	125 270	200	230	470	95 145 205	140 170 215	235	135 240	200 270	190
Length of int. pos. control ¹	800	1070	800	1070	800	1070	800	1070	1070	800	800	1070
5'-primer(s) ²	368 5'-gTT 3'	453 5'-AAA 3'	368 5'-gTT 3'	98 5'-CTC 3'	144 5'-gCC 3'	3rd In 5'-ATA 3'	98 5'-CTC 3' 564 5'-TgA 3'	98 5'-CTC 3'	28 5'-TCg 3'	368 5'-gTT 3'	90 5'-AgT 3' 160 5'-ACg 3'	144 920 5'-CCA 3'
3'-primer(s) ³	539 5'-TCA 3'	570 5'-CCg 3'	453 5'-TCg 3' 595 5'-CCg 3'	256 5'-CTg 3'	331 5'-CTC 3'	621 5'-ggg 3'	200 5'-TCC 3' 262 5'-TgC 3' 616 5'-CgT 3'	193 5'-CgA 3' 200 5'-TCC 3' 227 5'-CTg 3' 271 5'-CAT 3'	92 5'-AAC 3'	463 5'-gCT 3' 559 5'-CCg 3' 571 5'-CCT 3'	317 5'-ggA 3'	292 5'-gTg 3' 1013 5'-CTT 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	90 210	85 190 245	210 290	175 205	125 225	120	120 195	130 195 230 270	90	90 290	80 170	105 180 240
Length of int. pos. control ¹	800	1070	1070	800	1070	1070	1070	800	800	1070	1070	800
5'-primer(s) ²	368 5'-gTT 3'	368 5'-gTT 3'	368 5'-gTT 3'	98 5'-CTC 3' 368 5'-gTT 3'	98 5'-CTC 3'	453 5'-AAA 3'	418 5'-AgC 3' 493 5'-CTg 3'	90 5'-AgT 3' 228 5'-ATg 3' 379 5'-ACA 3' 418 5'-AgC 3'	920 5'-CCA 3'	368 5'-gTT 3'	414 5'-CAg 3' 678 5'-AgA 3'	98 5'-CTC 3'
3'-primer(s) ³	419 5'-CgC 3'	413 5'-gCC 3'	538 5'-CAg 3'	259 5'-gTT 3'	181 5'-gTA 3'	524 5'-CAC 3'	570 5'-CCg 3'	317 5'-ggA 3'	968 5'-CAg 3'	418 5'-gTC 3'	453 5'-TCT 3'	163 5'-CgC 3'
	530 5'-CCA 3'	518 5'-CCT 3'	616 5'-CgC 3'	502 5'-CTT 3'	282 5'-gAC 3'	538 5'-CAg 3'		570 5'-CCg 3'		619 5'-gTT 3'	806 5'-CTA 3'	238 5'-CCT 3'
	539 5'-TCC 3'	570 5'-CAC 3'		539 5'-TCT 3'	282 5'-gAC 3'						809 5'-CAA 3'	299 5'-TCg 3'
Well No.	13	14	15	16	17	18	19	20	21	22	23	24

101.421-06 – including *Taq* polymerase, IFU-01
101.421-06u – without *Taq* polymerase, IFU-02

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Lot No.: **7K4**

Lot-specific Information

Well No.	25	26	27	28	29	30	31	32	33	34	35
Length of spec. PCR product	95	80	85	385	170	110	185	85	165	90	105
	205		200		200	245				170	260
			285		240						
					290						
Length of int. pos. control ¹	800	1070	1070	800	1070	1070	1070	1070	1070	1070	800
5'-primer(s) ²	376	265	368	678	98	98	395	176	453	678	98
	5'-gCT 3'	5'-CAg 3'	5'-gTT 3'	5'-AgA 3'	5'-CTC 3'	5'-CTC 3'	5'-gCA 3'	5'-gCA 3'	5'-AAA 3'	5'-AgA 3'	5'-CTC 3'
	484				368						
	5'-ACg 3'				5'-gTT 3'						
3'-primer(s) ³	538	302	413	920	227	167	538	218	577	728	163
	5'-CAA 3'	5'-ggC 3'	5'-gCC 3'	5'-Tgg 3'	5'-CTg 3'	5'-ACC 3'	5'-CAA 3'	5'-gCg 3'	5'-gCC 3'	5'-CCT 3'	5'-CgC 3'
			524		255	303				809	316
			5'-CAT 3'		5'-TCC 3'	5'-AgT 3'				5'-CAA 3'	5'-gAA 3'
			534		255						
			5'-CgT 3'		5'-TCT 3'						
			614		299						
			5'-Tgg 3'		5'-TCg 3'						
					616						
					5'-CgC 3'						
Well No.	25	26	27	28	29	30	31	32	33	34	35

¹The 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.

²The nucleotide position matching the specificity-determining 3'-end of the primer is given. Nucleotide numbering as on the www.ebi.ac.uk/imgt/hla web site. The sequence of the 3 terminal nucleotides of the primer is given.

³The nucleotide position matching the specificity-determining 3'-end of the primer is given in the anti-sense direction. Nucleotide numbering as on the www.ebi.ac.uk/imgt/hla web site. The sequence of the 3 terminal nucleotides of the primer is given.

101.421-06 – including *Taq* polymerase, IFU-01
101.421-06u – without *Taq* polymerase, IFU-02

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“Instructions for Use” (IFU)

Lot No.: **7K4**

Lot-specific Information

CELL LINE VALIDATION SHEET																				
HLA-A*23 SSP subtyping kit ²																				
				Well																
				1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	
				Prod. No.:	201550401	201550402	201550403	201675604	201550405	201550406	201550407	201550408	201550409	201550410	201550411	201550412	201550413	201675614	201550415	201550416
	IHWC cell line ¹	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	-	-	-	-	-	-	-	-	-	W	-	-	-	+	-	-	+
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	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

101.421-06 – including *Taq* polymerase, IFU-01
101.421-06u – without *Taq* polymerase, IFU-02

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Lot No.: **7K4**

Lot-specific Information

CELL LINE VALIDATION SHEET																				
HLA-A*23 SSP subtyping kit ²																				
				Well																
				17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	
				Prod. No.:	201550417	201550418	201675619	201550420	201550421	201550422	201889123	201550424	201550425	201550426	201550427	201550428	201550429	201550430	201550431	201550432
	IHWC cell line ¹	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	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	

101.421-06 – including *Taq* polymerase, IFU-01
101.421-06u – without *Taq* polymerase, IFU-02

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Lot No.: **7K4**

Lot-specific Information

CELL LINE VALIDATION SHEET						
HLA-A*23 SSP subtyping kit ²						
						Well
						33 34 35
						Prod. No.:
						201550433
						201675634
						201550435
	IHWC cell line ¹	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	-	-	-

101.421-06 – including *Taq* polymerase, IFU-01
101.421-06u – without *Taq* polymerase, IFU-02

Visit <https://labproducts.caredx.com> for
“Instructions for Use” (IFU)

Lot No.: **7K4**

Lot-specific Information

¹The 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.

²The 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 3 to 5, 9, 11, 13, 15, 19, 20, 23 to 26, 29 to 31 and 33 to 35 were available.

The specificities of the primers in primer solutions 3, 13, 15, 19, 20, 23 and 26 were tested by separately adding one or two additional 5'-primers, respectively one or two additional 3'-primers.

In primer solutions 4, 5, 9, 24, 29, 30 and 33 to 35 it was only possible to test the 5'-primers, the 3'-primers were not possible to test.

In primer solutions 11, 25 and 31 it was only possible to test the 3'-primers, the 5'-primers was not possible to test.

In primer solutions 3, 7, 8, 10, 12 to 15, 17, 22, 23 and 27 one or more 3'-primers were not possible to test, and in primer solutions 19 and 20 one or more 5'-primer were not possible to test. In addition, one or more 3'-and/or 5' primers in primer solutions 7, 8, 10, 12, 14, 16, 18 and 27 were tested by separately adding one additional 5'-primer and/or one additional 3'-primer.

101.421-06 – including *Taq* polymerase, IFU-01
101.421-06u – without *Taq* polymerase, IFU-02

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Lot No.: **7K4**

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