101.201-48/12 – including *Taq* pol., IFU-01
101.201-48u/12u – without *Taq* pol., IFU-02

Visit https://labproducts.caredx.com for **"Instructions for Use" (IFU)**

Lot No.: 5N9 **Lot-specific information**

Olerup SSP® DQ low

Product number:	**101.201-48/12 – including *Taq* pol.** **101.201-48u/12u – without *Taq* pol.**
Lot number:	**5N9**
Expiry date:	**2025-12-01**
Number of tests:	**48 tests – Product No. 101.201-48/48u** **12 tests – Product No. 101.201-12/12u**
Number of wells per test:	**15+1**
Storage - pre-aliquoted primers:	**dark at -20ºC**
- PCR Master Mix:	**-20ºC**
- Adhesive PCR seals	**RT**
- Product Insert	**RT**

This Product Description is only valid for Lot No. 5N9.

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

CHANGES COMPARED TO THE PREVIOUS *OLERUP* SSP® DQ LOW LOT (8L2)

The product documentation has been updated for new alleles of IMGT 3.45.0.

The DQ low primer set, specificity and interpretation tables have been updated for the HLA-DQB1 alleles described since the previous *Olerup* SSP® DQ low lot was made **(Lot No 8L2)**.

The primers of the wells detailed below have been exchanged, added or modified compared to the previous lot **(Lot No. 8L2)**.

Well	5'-primer	3'-primer	rationale
13	Added	-	5'-primer added for improved yield of the HLA-specific product.

101.201-48/12 – including *Taq* pol., IFU-01
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Lot No.: 5N9 **Lot-specific information**

Well **16** contains Negative Control primer pairs, that will amplify the 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'-primer[1]	**164**	**340**	**440**	**45**	**45**	**43**	**36**
	5'-CAC3'	**5'-Agg3'**	**5'-TTA3'**	**5'-Tgg3'**	**5'-Tgg3'**	**5'-Tgg3'**	**5'-TAC3'**
							36
							5'-TAT3'
3'-primer[2]	**231**	**2nd I**	**507**	**59**	**58**	**57**	**47**
	5'-TgC3'	**5'-AAA3'**	**5'-TTg3'**	**5'-CTC3'**	**5'-ggC3'**	**5'-CTC3'**	**5'-ACA3'**
							48
							5'-gCA3'
							48
							5'-gCC3'
							52
							5'-TgT3'
A*	**+**	**+**	**+**				
B*	**+**	**+**	**+**				
C*	**+**	**+**	**+**				
DRB1				**+**	**+**		
DRB3				**+**	**+**		
DRB5				**+**			
DQB1					**+**		
DPB1						**+**	
DQA1							**+**

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

101.201-48/12 – including *Taq* pol., IFU-01
101.201-48u/12u – without *Taq* pol., IFU-02

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Lot No.: 5N9 **Lot-specific information**

PRODUCT DESCRIPTION

DQ low SSP typing

CONTENT

The primer set contains 5'- and 3'-primers for grouping the DQB1 alleles into the serological groups DQ2 to DQ9.

Please note that DQB1 amplifications usually are somewhat less pronounced than e.g. DRB and DQA1 amplifications even when using the same DNA preparation and exactly the same experimental procedures.

PLATE LAYOUT

Each test consists of 16 PCR reactions in a 16 well PCR plate.

1	**2**	**3**	**4**	**5**	**6**	**7**	**8**
9	**10**	**11**	**12**	**13**	**14**	**15**	**NC**

The 16 well cut PCR plate is marked with 'DQ LOW' in silver/gray ink.
Well No. 1 is marked with the Lot No. '5N9'.
Wells 1 to 15 – DQ low primers.
Well 16 – 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 16 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 the DQB1 alleles will be amplified by the DQ low typing kit. Thus, the interpretation of DQ low typings is not influenced by the DQB2 and DQB3 genes.

UNIQUELY IDENTIFIED ALLELES

All the DQB1 alleles, i.e. **DQB1*05:01:01:01 to 05:282, DQB1*06:01:01:01 to 06:394N, DQB1*02:01:01 to 02:186, DQB1*03:01:01:01 to 03:466 and DQB1*04*01:01:01 to 04:86**, recognized by the HLA Nomenclature Committee in July 2020[1,2,4] will be amplified by the primers in the DQ low SSP kit. The DQB1 alleles will be grouped into their corresponding serological specificities, i.e.:

DQ5(1) = DQB1*05:01-05:05[3]
DQ6(1) = DQB1*06:01-06:33[3]
DQ2 = DQB1*02:01-02:05[3]
DQ3 = DQB1*03:01-03:20[3]
DQ4 = DQB1*04:01-04:02[3]

CE

Lot No.: 5N9 **Lot-specific information**

The DQ3 alleles may be further subdivided into the DQ3, DQ7, DQ8 and DQ9 based upon serology and expert assignment. Thus:

DQ3	=	DQB1*03:06, 03:10, 03:14
DQ7	=	DQB1*03:01:01-03:01:04, 03:04, 03:09, 03:13, 03:16, 03:19
DQ8	=	DQB1*03:02, 03:05, 03:07, 03:08, 03:11, 03:18
DQ9	=	DQB1*03:03, 03:12, 03:15, 03:17, 03:20

[1]HLA-DQB1 alleles listed on the IMGT/HLA web page 2021-July-12, release 3.45.0, www.ebi.ac.uk/imgt/hla.
[2]Alleles that have been deleted from or renamed in the official WHO HLA Nomenclature up to and including the last IMGT/HLA database release can be retrieved from web page http://hla.alleles.org/alleles/deleted.html.
[3]The serological split of the DQB1*05:05-05:282 alleles, the DQB1*06:06-06:07, DQB1*06:10, DQB1*06:13, DQB1*06:15-06:24 and DQB1*06:27-06:394N alleles, the DQB1*02:04-02:186 alleles, the DQB1*03:07-03:09 and DQB1*03:11 to 03:466, and the DQB1*04:03 to 04:86 alleles is not known. The grouping of not serologically defined alleles is taken from the expert-assigned serological grouping in Tissue Antigens (2009) 73:95-170.
[4]The DQ low alleles will be grouped into their corresponding serological specificities, except for the following alleles that will give rise to identical amplification patterns:

Alleles
DQB1*05:01:01:01-05:01:15, 05:01:17-05:03:23, 05:03:26-05:03:27, 05:05:01-05:43:02, 05:45-05:51, 05:53, 05:55-05:59, 05:61-05:71, 05:73-05:76, 05:78-05:81, 05:84-05:97, 05:99-05:104, 05:106-05:113, 05:115, 05:117-05:127, 05:129-05:131, 05:133-05:145, 05:147-05:174, 05:177-05:206N, 05:208N-05:217, 05:219-05:243, 05:245-05:257, 05:259-05:261, 05:263-05:282, DQB1*06:325

101.201-48/12 – including *Taq* pol., IFU-01
101.201-48u/12u – without *Taq* pol., IFU-02

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Lot No.: 5N9 **Lot-specific information**

SPECIFICITY TABLE

DQ low SSP typing

Specificities and sizes of the PCR products of the 15+1 primer mixes used for DQ low SSP typing

Primer Mix	Size of spec. PCR product[1]	Size of control band[2]	DQ serology[3]	Amplified DQB1 alleles[4]
1	135 bp, 230 bp	**515 bp**	5	*05:01:01:01-05:01:15, 05:01:17-05:03:24, 05:03:26-05:59, 05:61-05:81, 05:84-05:97, 05:99-05:115, 05:117-05:174, 05:176-05:217, 05:219-05:257, 05:259-05:282, 06:325, 06:389
2[8]	135 bp, 185 bp, 220 bp, 270 bp	**515 bp**	1, 5, 6	*02:03:02, 03:23:01-03:23:02, 03:217, 03:259, 03:355, 04:10, 05:176, 06:01:01:01-06:155, 06:157-06:161, 06:163-06:168, 06:170-06:219, 06:221-06:324, 06:326-06:382, 06:384-06:394N
3	210 bp	430 bp	2	*02:01:01:01-02:186
4	130 bp, 220 bp	**515 bp**	3, 7	*03:01:01:01-03:01:01:12, 03:01:01:14-03:01:50, 03:04:01:01-03:04:04, 03:09-03:10:03, 03:13-03:14:02, 03:16, 03:19:01:01-03:19:05, 03:21-03:22:02, 03:24, 03:27-03:29, 03:35-03:36, 03:42, 03:44, 03:46-03:60, 03:69, 03:71, 03:73, 03:75-03:77, 03:80, 03:82-03:84N, 03:92-03:94, 03:101-03:103, 03:108-03:109, 03:114-03:116, 03:118N-03:122, 03:127-03:131, 03:133-03:135, 03:138-03:140, 03:142-03:144, 03:147-03:148, 03:150, 03:152, 03:154, 03:157-03:160, 03:162-03:167, 03:169-03:173, 03:180, 03:182-03:183, 03:186-03:188, 03:191-03:198:02, 03:201-03:202, 03:206-03:208, 03:216, 03:218-03:219, 03:231-03:232, 03:235-03:236, 03:241-03:243, 03:246, 03:252-03:257, 03:260, 03:264, 03:266-03:268, 03:271, 03:275-03:276N, 03:281, 03:284-03:286, 03:288, 03:290-03:294, 03:297, 03:302-03:303N, 03:305-03:307, 03:309, 03:311-03:312, 03:317:01-03:318, 03:326-03:331, 03:335, 03:338N, 03:340N-03:342, 03:347, 03:350, 03:353-03:355, 03:358N, 03:360-03:361, 03:366, 03:370, 03:372-03:373, 03:376N-03:378, 03:380-03:381, 03:385N, 03:387, 03:389-03:391, 03:394, 03:396, 03:399N-03:401, 03:404, 03:407N-03:408, 03:417-03:421, 03:423-03:428, 03:430-03:432, 03:434-03:436, 03:438-03:439,

Lot No.: 5N9 **Lot-specific information**

				03:443, 03:448-03:449, 03:451, 03:454-03:455, 03:458, 03:460, 03:465, 06:209
5[6]	130 bp, 220 bp	**515 bp**	6, 8	*03:02:01:01-03:02:35, 03:05:01-03:05:05, 03:07-03:08, 03:11, 03:18, 03:32, 03:37, 03:45:01-03:45:02, 03:61, 03:63-03:64, 03:66N-03:68, 03:70, 03:85, 03:104, 03:106-03:107, 03:125, 03:132, 03:146, 03:153, 03:161, 03:174-03:175, 03:178-03:179, 03:181, 03:184-03:185, 03:189-03:190, 03:199, 03:203-03:205, 03:210-03:211, 03:213N-03:215, 03:220-03:221, 03:223-03:224, 03:226, 03:228-03:229, 03:233, 03:237N, 03:240, 03:245, 03:247, 03:250-03:251, 03:261-03:263:01:02, 03:265, 03:269N, 03:273-03:274, 03:277-03:279, 03:287, 03:289, 03:295-03:296, 03:298-03:301, 03:310N, 03:315, 03:320-03:324, 03:333-03:334N, 03:339N, 03:343-03:346, 03:348-03:349, 03:352, 03:362, 03:364, 03:367-03:369, 03:371, 03:379, 03:383, 03:386, 03:388, 03:392, 03:403N, 03:409-03:410, 03:412, 03:415-03:416, 03:422N, 03:429, 03:433, 03:437, 03:440N-03:442, 03:444, 03:446-03:447, 03:450, 03:452, 03:456-03:457, 03:459, 03:462-03:464, 03:466, 04:75, 06:29, 06:123, 06:139, 06:337
6[6]	135 bp	**515 bp**	2, 3, 9	*02:77, 02:180, 03:03:02:01-03:03:25, 03:03:27, 03:06, 03:12, 03:15, 03:20, 03:23:03, 03:25:01-03:26, 03:30-03:31, 03:33-03:34, 03:38:01-03:41, 03:43, 03:65, 03:74, 03:79, 03:86-03:91Q, 03:95N-03:99Q, 03:104-03:105, 03:111-03:113, 03:117, 03:123-03:124, 03:126, 03:136-03:137, 03:141, 03:145, 03:149, 03:155-03:156, 03:168, 03:176-03:177, 03:200, 03:209, 03:212, 03:222, 03:227, 03:230, 03:234, 03:238-03:239, 03:248-03:249, 03:258, 03:270, 03:280, 03:282N-03:283, 03:304, 03:313, 03:316, 03:319, 03:332, 03:336-03:337, 03:351, 03:356N-03:357N, 03:359, 03:363, 03:365, 03:374-03:375N, 03:382, 03:384, 03:393, 03:395, 03:397-03:398, 03:402, 03:405-03:406, 03:411N, 03:414, 03:445, 03:453, 03:461, 04:03:01-04:03:03, 06:02:43, 06:03:10, 06:03:33, 06:51:01, 06:66, 06:96:01, 06:168, 06:172, 06:322:01-06:322:02, 06:377
7[5,7]	80 bp	**515 bp**	3, 7, 8, 9	*03:01:01:01-03:01:01:12, 03:01:01:14-03:01:06, 03:01:07^{w}, 03:01:08-03:02:05, 03:02:07-03:02:17, 03:02:19-03:02:29, 03:02:31-03:05:05, 03:07, 03:08^{w}, 03:09-03:12, 03:13^{w}, 03:14:01-03:24, 03:26-03:57, 03:58^{w}, 03:59-03:64, 03:65^{w}, 03:66N-03:106, 03:107^{w}, 03:108-03:124, 03:126-03:136, 03:137^{w}, 03:138-03:146, 03:148-03:193,

Lot No.: 5N9 **Lot-specific information**

				03:194[w], 03:195-03:227, 03:229-03:231, 03:232[w], 03:233-03:261, 03:262[w], 03:263:01:01-03:282N, 03:283[w], 03:284-03:324, 03:326-03:410, 03:412-03:417, 03:419-03:448, 03:450-03:466
8[6]	135 bp, 160 bp, 185 bp, 210 bp	430 bp	4	*03:132, 04:01:01:01-04:02:01:01, 04:02:01:04-04:03:02, 04:04-04:86
9	225 bp	430 bp	4	*04:01:01:01-04:02:01:01, 04:02:01:04-04:02:18, 04:02:20-04:03:03, 04:06-04:21, 04:22[w], 04:23, 04:24[w], 04:25N-04:32, 04:34-04:37, 04:38[w], 04:39-04:48, 04:50-04:86
10	185 bp, 215 bp	430 bp	5	*05:01:01:01-05:03:23, 05:03:25-05:03:27, 05:05:01-05:43:02, 05:45-05:51, 05:53, 05:55-05:71, 05:73-05:76, 05:78-05:104, 05:106-05:113, 05:115-05:127, 05:129-05:131, 05:133-05:145, 05:147-05:175, 05:177-05:206N, 05:208N-05:243, 05:245-05:261, 05:263-05:282, 06:156, 06:162, 06:169, 06:325
11[6]	185 bp	430 bp	8, 9	*03:03:11, 03:05:01, 03:05:03, 03:05:05, 03:17:01, 03:61, 03:72, 03:100, 03:181, 03:250, 03:346
12[6]	185 bp	430 bp	4, 7, 8, 9	*03:01:01:01-03:01:01:12, 03:01:01:14-03:01:01:30, 03:01:03-03:01:07, 03:01:09-03:02:02, 03:02:04-03:02:12, 03:02:14-03:03:02:05, 03:03:04-03:03:15, 03:03:17-03:04:04, 03:05:03-03:05:04, 03:06-03:17:01, 03:18-03:19:05, 03:21-03:22:02, 03:23:02-03:36, 03:38:01, 03:39-03:60, 03:62-03:71, 03:74, 03:76-03:98, 03:101-03:103, 03:106-03:108, 03:110-03:111, 03:113-03:117, 03:119-03:131, 03:133-03:153, 03:155, 03:157-03:161, 03:163-03:180, 03:182, 03:184-03:188, 03:190-03:203, 03:205-03:222, 03:224-03:225, 03:227-03:232, 03:234-03:236, 03:239-03:249, 03:251, 03:253-03:257, 03:259-03:261, 03:263:01:01-03:290, 03:292-03:324, 03:326-03:334N, 03:337-03:345, 03:347-03:350, 03:353, 03:355-03:370, 03:372-03:390, 03:392-03:442, 03:444-03:466, 04:01:03, 04:02:16, 04:03:03
13[7]	185 bp	**515 bp**	6	*04:10, 06:02:01:01-06:02:42, 06:02:44-06:02:52, 06:14:01-06:16, 06:19:01-06:20, 06:23-06:24, 06:33, 06:37, 06:46-06:50, 06:51:02, 06:68, 06:70-06:84:01:02, 06:95, 06:96:02-06:97, 06:107, 06:109, 06:111-06:117, 06:122, 06:124-06:127, 06:136-06:138, 06:146:01-06:147, 06:150-06:152, 06:156, 06:159, 06:161-06:163, 06:166, 06:173-06:175, 06:178-06:179N, 06:182-06:183, 06:188, 06:192, 06:197-06:198,

Lot No.: 5N9 **Lot-specific information**

				06:200-06:201, 06:208, 06:211, 06:213, 06:215-06:216N, 06:219, 06:224-06:228, 06:232, 06:235-06:237, 06:240, 06:242, 06:249, 06:255-06:256, 06:262, 06:264, 06:270:02-06:271, 06:273, 06:284, 06:286, 06:289-06:290, 06:293-06:298, 06:300, 06:304N, 06:306N, 06:308N, 06:311, 06:314-06:315, 06:317N, 06:324, 06:326, 06:333, 06:335, 06:338, 06:341N, 06:344, 06:347, 06:354-06:357, 06:363-06:364, 06:366, 06:370, 06:372, 06:374, 06:376, 06:379N-06:380, 06:383N-06:384, 06:386, 06:388, 06:390
14	185 bp	430 bp	1, 5, 6	*06:02:01:01-06:02:12, 06:02:14-06:03:10, 06:03:12-06:03:41, 06:05:02[?]-06:06[?], 06:08:01-06:08:03, 06:10-06:11:04, 06:13:01-06:14:03, 06:16, 06:18:01-06:20, 06:23-06:24, 06:26N-06:27:02, 06:29-06:33, 06:37, 06:40-06:41:01:03, 06:44, 06:47-06:51:02, 06:59-06:65, 06:67-06:68, 06:70-06:78, 06:80-06:84:01:02, 06:87, 06:90-06:91, 06:95-06:97, 06:99:01-06:99:03, 06:106-06:107, 06:109-06:117, 06:122-06:128, 06:130-06:131, 06:133-06:134, 06:136-06:139, 06:141, 06:143, 06:145, 06:147-06:148, 06:150-06:152, 06:154, 06:156, 06:159, 06:161-06:163, 06:165-06:166, 06:169-06:170, 06:173-06:176, 06:178-06:179N, 06:182-06:185, 06:187-06:188, 06:190:01-06:192, 06:195-06:198, 06:200-06:201, 06:206:01-06:206:02, 06:208, 06:211, 06:213, 06:215-06:216N, 06:218-06:219, 06:221-06:228, 06:230, 06:232-06:234, 06:236-06:238, 06:240, 06:242, 06:244, 06:248-06:250, 06:253, 06:255-06:256, 06:259, 06:262, 06:264, 06:269-06:273, 06:276, 06:278-06:279, 06:284, 06:286, 06:289-06:290, 06:293-06:298, 06:300, 06:304N, 06:306N, 06:308N, 06:311, 06:314-06:317N, 06:319, 06:322:01-06:322:02, 06:324, 06:326-06:329, 06:331, 06:333-06:338, 06:341N, 06:344-06:347, 06:350, 06:352, 06:354-06:357, 06:360, 06:362-06:368, 06:370-06:374, 06:376, 06:378-06:380, 06:383N-06:384, 06:388-06:394N
15	135 bp	430 bp	3, 5, 6, 9	*03:10:01-03:10:02:02, 03:12, 03:14:01-03:14:02, 03:70, 03:179, 03:183, 03:195, 06:01:01:01-06:01:06, 06:01:08-06:01:30, 06:06[?], 06:43, 06:54N-06:58, 06:98, 06:99:02-06:105, 06:108, 06:120, 06:132, 06:140, 06:153:01-06:153:02, 06:157, 06:167-06:168, 06:177, 06:181, 06:194, 06:205, 06:209, 06:214, 06:229, 06:239, 06:243, 06:245-06:247, 06:251,

				06:257-06:258, 06:260, 06:263, 06:268, 06:274, 06:277, 06:285, 06:302, 06:305, 06:307, 06:309-06:310, 06:312, 06:321, 06:323, 06:330N, 06:342, 06:359, 06:377, 06:382
16[9]	-	-	-	**Negative Control**

[1]Alleles are assigned by the presence of specific PCR product(s). However, the sizes of the specific PCR products may be helpful in the interpretation of DQ low SSP typings.
When the primers in a primer mix can give rise to HLA-specific PCR products of more than one length this is indicated if the size difference is more than 20 base pairs. Size differences of 20 base pairs or less are not given. For high resolution SSP kits, the alleles listed are specified according to amplicon length.
Nonspecific amplifications, i.e. a ladder or a smear of bands, may sometimes be seen. GC-rich primers have a higher tendency of giving rise to nonspecific amplifications than other primers.
PCR fragments longer than the control bands may sometimes be observed. Such bands should be disregarded and do not influence the interpretation of the SSP typings.
PCR fragments migrating faster than the control bands, but slower than a 400 bp fragment may be seen in some gel read-outs. Such bands can be disregarded and do not influence the interpretation of the SSP typings.
Some primers may give rise to primer oligomer artifacts. Sometimes this phenomenon is an inherit feature of the primer pair(s) of a primer mix. More often it is due to other factors such as too low amount of DNA in the PCR reactions, taking too long time in setting up the PCR reactions, working at elevated room temperature or using thermal cyclers that are not pre-heated.

[2]The internal positive control primer pairs amplify segments of the human growth hormone gene. The internal positive control bands are 430 or 515 base pairs respectively, well distribution as outlined in the table. Well number 1 contains the longer, 515 bp, internal positive control band. The well distribution of the internal controls can help in orientation of the kit on gel photo, as well as allow for kit identification. In the presence of a specific amplification the intensity of the control band often decreases.

[3]The serological split of the DQB1*05:05-05:282 alleles, the DQB1*06:06-06:07, DQB1*06:10, DQB1*06:13, DQB1*06:15-06:24 and DQB1*06:27-06:394N alleles, the DQB1*02:04-02:186 alleles, the DQB1*03:07-03:09 and DQB1*03:11 to 03:466, and the DQB1*04:03 to 04:86 alleles is not known. The grouping of not serologically defined alleles is taken from the expert-assigned serological grouping in Tissue Antigens (2009) 73:95-170.

[4]For several DQB1 alleles 1st and/or 3rd 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.

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

[6]Primer mixes 5, 6, 8, 11 and 12 may give a lower yield of HLA-specific PCR products than the other DQ low primer mixes.

[7]Primer mixes 7 and 13 may have tendencies of unspecific amplification.

[8]Primer mix 2 may have tendencies for primer oligomer formation.

[9]Primer mix 16 contains a negative control, which will amplify the majority of the 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
ser: serological HLA specificity
w: might be weakly amplified.
?: nucleotide sequence information not available for the primer matching sequence.

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PRIMER SPECIFICATION

Well No.	1	2	3	4	5	6	7	8	9	10	11	12
Length of spec.	135	135	210	130	130	135	80	135	225	185	185	185
PCR product	230	185		220	220			160		215		
		220						185				
		270						210				
Length of int.	515	515	430	515	515	515	515	430	430	430	430	430
pos. control[1]												
5'-primer(s)[2]	26(173)	9(122)	28(178)	26(173)	28(179)	26(173)	72(312)	21(159)	9(122)	30(184)	21(159)	38(210)
	5' -ggg 3'	5' -gCT 3'	5' -TgC 3'	5' -TTA 3'	5' -gAC 3'	5' -TCT 3'	5' -Cgg 3'	5' -ACC 3'	5' -gTT 3'	5' -gAC 3'	5' -ACC 3'	5' -gCA 3'
	26(173)	25(169)	30(184)		28(179)			23(164)				
	5' -ggA 3'	5' -TgT 3'	5' -gAg 3'		5' -gAC 3'			5' -gCT 3'				
	26(173)	26(172)	30(185)		28(179)			38(210)				
	5' -ggg 3'	5' -ATC 3'	5' -AAg 3'		5' -gAC 3'			5' -gCg 3'				
	26(173)	26(173)	30(185)									
	5' -gTg 3'	5' -TTA 3'	5' -AAA 3'									
		26(173)										
		5' -TCT 3'										
		30(184)										
		5' -gAT 3'										
3'-primer(s)[3]	57(266)	57(266)	86(353)	55(260)	57(266)	57(266)	86(353)	70(304)	70(304)	77(327)	70(304)	86(353)
	5' -CAA 3'	5' -CAA 3'	5' -gCT 3'	5' -gCg 3'	5' -Cgg 3'	5' -CgT 3'	5' -gCT 3'	5' -CTC 3'	5' -CTC 3'	5' -ACT 3'	5' -CCT 3'	5' -gCT 3'
	87(356)	57(266)		86(353)	57(266)	57(266)	89(361)	77(327)		87(356)		87(357)
	5' -ggT 3'	5' -CAT 3'		5' -gCT 3'	5' -CAg 3'	5' -CgT 3'	5' -CgT 3'	5' -ACg 3'		5' -ggT 3'		5' -CgT 3'
	87(356)	86(353)		86(354)	57(266)					87(356)		
	5' -ggT 3'	5' -ACg 3'		5' -AgT 3'	5' -Cgg 3'					5' -ggA 3'		
	89(361)	86(353)			87(356)							
	5' -CCT 3'	5' -ACC 3'			5' -ggg 3'							
		86(354)										
		5' -AAT 3'										
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

Well No.	13	14	15
Length of spec.	185	185	135
PCR product			
Length of int.	515	430	430
pos. control[1]			
5'-primer(s)[2]	9(122)	38(209)	13(134)
	5' -gTT 3'	5' -CgC 3'	5' -ggC 3'
3'-primer(s)[3]	57(266)	86(353)	45(230)
	5' -CAT 3'	5' -ACg 3'	5' -CCC 3'
	58(270)		
	5' -TCC 3'		
Well No.	13	14	15

[1]The internal positive control primer pairs amplify segments of the human growth hormone gene. The internal positive control bands are 430 or 515 base pairs respectively, well distribution as outlined in the table. Well number 1 contains the longer, 515 bp, internal positive control band. The well distribution of the internal controls can help in orientation of the kit on gel photo, as well as allow for kit identification. In the presence of a specific amplification the intensity of the control band often decreases.

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

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

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CELL LINE VALIDATION SHEET

DQ low resolution primer set[2]

					Well														
					1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
				Production No.	202020301	202135902	202020303	202020304	202133505	202020306	202020307	202020308	202020309	202020310	202020311	202020312	202135913	202020314	202020315
	IHWC cell line[1]		DQB1																
1	9001	SA	*05:01		+	-	-	-	-	-	-	-	-	+	-	-	-	-	-
2	9280	LK707	*06:01	*02:02	-	+	+	-	-	-	-	-	-	-	-	-	-	-	+
3	9011	E4181324	*06:01		-	+	-	-	-	-	-	-	-	-	-	-	-	-	+
4	9275	GU373	*02:01		-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
5	9009	KAS011	*05:02		+	-	-	-	-	-	-	-	-	+	-	-	-	-	-
6	9353	SM	*03:02	*06:01	-	+	-	-	+	-	+	-	-	-	-	+	-	-	+
7	9020	QBL	*02:01		-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
8	9025	DEU	*03:01		-	-	-	+	-	-	+	-	-	-	-	+	-	-	-
9	9026	YAR	*03:02		-	-	-	-	+	-	+	-	-	-	-	+	-	-	-
10	9107	LKT3	*04:01		-	-	-	-	-	-	-	+	+	-	-	-	-	-	-
11	9051	PITOUT	*02:02		-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
12	9052	DBB	*03:03		-	-	-	-	-	+	+	-	-	-	-	+	-	-	-
13	9004	JESTHOM	*05:01		+	-	-	-	-	-	-	-	-	+	-	-	-	-	-
14	9071	OLGA	*04:02		-	-	-	-	-	-	-	+	+	-	-	-	-	-	-
15	9075	DKB	*03:03		-	-	-	-	-	+	+	-	-	-	-	+	-	-	-
16	9037	SWEIG007	*03:01		-	-	-	+	-	-	+	-	-	-	-	+	-	-	-
17	9282	CTM 3953540	*02:01	*06:03	-	+	+	-	-	-	-	-	-	-	-	-	-	+	-
18	9257	32367	*06:02	*02:02	-	+	+	-	-	-	-	-	-	-	-	-	+	+	-
19	9038	BM16	*03:01		-	-	-	+	-	-	+	-	-	-	-	+	-	-	-
20	9059	SLE005	*06:04		-	+	-	-	-	-	-	-	-	-	-	-	-	-	-
21	9064	AMALA	*03:01		-	-	-	+	-	-	+	-	-	-	-	+	-	-	-
22	9056	KOSE	*05:03	*06:04	+	+	-	-	-	-	-	-	-	+	-	-	-	-	-
23	9124	IHL	*05:03	*06:01	+	+	-	-	-	-	-	-	-	+	-	-	-	-	+
24	9035	JBUSH	*03:01		-	-	-	+	-	-	+	-	-	-	-	+	-	-	-
25	9049	IBW9	*02:02		-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
26	9285	WT49	*02:01		-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
27	9191	CH1007	*04:01	*05:01	+	-	-	-	-	-	-	+	+	+	-	-	-	-	-
28	9320	BEL5GB	*02:02	*03:01	-	-	+	+	-	-	+	-	-	-	-	+	-	-	-
29	9050	MOU	*02:02		-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
30	9021	RSH	*04:02		-	-	-	-	-	-	-	+	+	-	-	-	-	-	-
31	9019	DUCAF	*02:01		-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
32	9297	HAG	*03:01		-	-	-	+	-	-	+	-	-	-	-	+	-	-	-
33	9098	MT14B	*03:02		-	-	-	-	+	-	+	-	-	-	-	+	-	-	-
34	9104	DHIF	*03:01		-	-	-	+	-	-	+	-	-	-	-	+	-	-	-
35	9302	SSTO	*03:05		-	-	-	-	+	-	+	-	-	-	+	-	-	-	-
36	9024	KT17	*03:02		-	-	-	-	+	-	+	-	-	-	-	+	-	-	-
37	9065	HHKB	*06:03		-	+	-	-	-	-	-	-	-	-	-	-	-	+	-
38	9099	LZL	*03:01		-	-	-	+	-	-	+	-	-	-	-	+	-	-	-
39	9315	CML	*02:01	*03:01	-	-	+	+	-	-	+	-	-	-	-	+	-	-	-
40	9134	WHONP199	*02:02	*03:03	-	-	+	-	-	+	+	-	-	-	-	+	-	-	-
41	9055	H0301	*06:09		-	+	-	-	-	-	-	-	-	-	-	-	-	-	-
42	9066	TAB089	*06:01		-	+	-	-	-	-	-	-	-	-	-	-	-	-	+
43	9076	T7526	*03:03		-	-	-	-	-	+	+	-	-	-	-	+	-	-	-
44	9057	TEM	*05:03		+	-	-	-	-	-	-	-	-	+	-	-	-	-	-
45	9239	SHJO	*02:02		-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
46	9013	SCHU	*06:02		-	+	-	-	-	-	-	-	-	-	-	-	+	+	-
47	9045	TUBO	*03:01		-	-	-	+	-	-	+	-	-	-	-	+	-	-	-
48	9303	TER-ND	*05:01		+	-	-	-	-	-	-	-	-	+	-	-	-	-	-

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

[2]The specificity of each primer solution in the kit has been tested against 48 well characterized cell line DNAs and where applicable, additional cell line DNAs.

In primer solutions 1 to 3 and 5 one, two or three 5'-primers were not possible to be tested, and in primer solutions 1, 2, 4, 5, 12 and 13 one or more of the 3'-primers were not possible to be tested.

The specificities of the primers in primer solution 2 was tested by separately adding one additional 5'-primer, and one additional 3'-primer accordingly. In addition, one 3'-primer in primer solutions 6 and 10 was tested by separately adding one 5'-primer.

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