Gel Documentation Form and Worksheet

HLA-C\*06 (101.614-12/12u) Lot No: 2E9 Expiry Date: 2019-03-01

Name:\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

Sample ID:\_\_\_\_\_\_\_\_\_\_\_\_\_\_

DNA Conc.(ng/ul):\_\_\_\_\_\_\_\_\_

Test Date:\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

Tested By:\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

Review Date:\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

Reviewed By:\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

*Interpretation:\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ Failed lanes*: \_\_\_\_\_\_\_\_\_\_\_\_ *Comments:\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_*

**Gel Picture**

|  |
| --- |
| PHOTO DOCUMENT |







‘ICB’ Internal Control Band,

‘AmpS’ Amplicon Size

**Notes:**

Product sizes are approximate. For detailed information, see the lot-specific Specificity Table and Interpretation Table.

This table is intended as a guide. For interpretation always use the Interpretation Table and/or Specificity Table.

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

Primer mix 16 has a tendency to giving rise to primer oligomer formation.

Primer mixes 19, 22, 39 and 42 may have tendencies of unspecific amplification.

Primer mix 14 may give rise to a lower yield of HLA-specific PCR product than the other HLA-C\*06 resolution primer mixes in the C\*03 alleles.

Primer mix 57 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.

Changes in R01 compared to R00:

1. Primer mixes 22 and 42 do not amplify the C\*06:86 allele. Thus, this lot cannot discriminate between the C\*06:86 and the C\*06:02:01:01-06:02:01:03, 06:02:03-06:02:45, 06:48, 06:51-06:52, 06:56, 06:62-06:64, 06:68, 06:75, 06:78, 06:80, 06:82, 06:84-06:85, 06:88-06:92, 06:94-06:95, 06:97-06:99, 06:102, 06:105, 06:108-06:110, 06:112-06:115, 06:117, 06:119, 06:121, 06:125, 06:130-06:131, 06:135, 06:137, 06:141, 06:145, 06:149-06:150, 06:154-06:157, 06:159-06:167, 06:169-06:170, 06:172-06:174 alleles. This has been corrected in the specificity and interpretation tables.







 













 **1**HLA-C\*06 alleles in bold lettering are listed as confirmed alleles on the IMGT/HLA web page [www.ebi.ac.uk/imgt/hla](http://www.ebi.ac.uk/imgt/hla), release 3.24.0, April 2016.

**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**Primer mix 5: Specific PCR fragment of 160 bp in the C\*06:05, 06:67 and 06:103 and the C\*02:94, 04:129, 05:01:01:01-05:01:27, 05:01:28w, 05:01:29-05:01:31, 05:03-05:28, 05:30-05:47, 05:49-05:91N, 05:93-05:128N, 08:10, 12:21, 12:33, 15:107, 17:05 alleles. Specific PCR fragment of 220 bp in the C\*06:18 and 06:23 and the C\*01:04, 01:09 and 02:21 alleles.

Primer mix 6: Specific PCR fragment of 250 bp in the C\*06:05-06:06 and the C\*01:02:01-01:03, 01:05-01:07:01, 01:08, 01:10-01:20, 01:23-01:34, 01:36-01:119, 04:110?, 05:01:01:01-05:01:31, 05:03-05:07N, 05:09:01-05:10, 05:12-05:16, 05:18:01-05:26, 05:28-05:50, 05:53-05:61, 05:63-05:67, 05:69, 05:71-05:78, 05:80-05:88, 05:90-05:105, 05:107-05:111, 05:113N-05:114, 05:116-05:129, 08:02:01:01-08:02:12, 08:05, 08:07, 08:12, 08:15:01-08:15:02, 08:17-08:19, 08:23, 08:25, 08:28, 08:30, 08:32-08:35, 08:37, 08:43, 08:45, 08:47-08:49, 08:51-08:53, 08:55N, 08:62-08:63, 08:67-08:71, 08:73-08:77, 08:90, 08:92, 08:94, 08:100, 08:103, 08:107-08:108, 08:110-08:112, 08:114-08:116, 08:118, 08:120, 08:123, 08:125-08:126, 12:09, 12:24, 12:85, 14:02:01-14:05, 14:07N-14:14, 14:17-14:52, 14:54-14:62, 14:64-14:73, 15:08, 15:102, 16:53, 16:68, 16:88, 17:17, 18:01-18:02 and 18:04-18:10 alleles. Specific PCR fragment of 290 bp in the C\*06:50 and the C\*12:53 alleles.

Primer mix 7: Specific PCR fragment of 110 bp in the C\*06:07 allele. Specific PCR fragment of 185 bp in the C\*06:19 allele. Specific PCR fragment of 235 bp in the C\*06:33 and 06:104 alleles.

Primer mix 9: Specific PCR fragment of 165 bp in the C\*06:09 and 06:144 and the C\*02:22, 04:94:01-04:94:02, 05:08, 05:52, 05:89, 05:106, 08:27, 08:29, 08:31, 12:31 and 12:144 and in the B\*15:137 alleles. Specific PCR fragment of 210 bp in the C\*06:23 and the C\*01:04 and 01:09 alleles. Specific PCR fragment of 435 bp in the C\*06:17 and the C\*07:07, 07:09, 07:76:01-07:76:02, 07:315, 07:328, 07:406, 18:01-18:02, 18:04-18:10 alleles. Specific PCR fragments of 165 and 435 bp in the C\*18:03 allele.

Primer mix 11: Specific PCR fragment of 130 bp in the C\*06:11, 06:122w, 06:124w and 06:147 and the C\*07:01:13, 07:04:01-07:04:10, 07:11-07:12, 07:45, 07:63, 07:68, 07:101, 07:139, 07:142, 07:181, 07:199:01-07:199:02, 07:272, 07:302, 07:323-07:324, 07:329N, 07:338, 07:354-07:355, 07:358, 07:364-07:365, 07:378, 07:394-07:395, 07:420, 07:426, 07:428, 07:447, 07:459, 07:466-07:467, 07:480, 07:487, 12:02:11, 12:03:09 and 16:01:16 alleles. Specific PCR fragment of 185 bp in the C\*06:22 and the C\*07:107, 07:224 and C\*16:31 and in the B\*15:193 and B\*35:283 alleles.

Primer mix 13: Specific PCR fragment of 155 bp in the C\*06:13 allele. Specific PCR fragment of 210 bp in the C\*06:59 allele.

Primer mix 14: Specific PCR fragment of 225 bp in the C\*06:55 allele. Specific PCR fragment of 305 bp in the C\*06:14 and 06:143 and the C\*03:32, 03:45, 03:136, 03:297, 04:80, 04:100, 04:110w, 04:178, 07:10w, 07:43, 07:196, 15:25 and 15:62 and in the B\*15:357, B\*35:178 an 35:282 alleles.

Primer mix 15: Specific PCR fragment of 135 bp in the C\*06:116N allele. Specific PCR fragment of 360 bp in the C\*06:15 allele.

Primer mix 16: Specific PCR fragment of 235 bp in the C\*06:16N allele. Specific PCR fragment of 340 bp in the C\*06:21 and the C\*15:91 alleles.

Primer mix 19: Specific PCR fragment of 85 bp in the C\*06:28 and the C\*02:14:01-02:14:02, 03:67, 04:42:01-04:42:02, 05:43, 07:20, 07:73, 07:172:01-07:172:02, 07:390, 08:37, 15:23, 16:21, 17:20, 18:04 and the B\*08:56:01-08:56:02, B\*15:142, B\*35:218, 35:256, B\*51:68 and B\*57:49 alleles. Specific PCR fragment of 160 bp in the C\*06:32 allele. Specific PCR fragment of 245 bp in the C\*06:122 and 06:124 alleles.

Primer mix 20: Specific PCR fragment of 115 bp in the C\*06:29 and the C\*07:134 alleles. Specific PCR fragment of 220 bp in the C\*06:05w, 06:44w, 06:124, 06:134N and 06:138w and the C\*02:14:01w-02:14:02w, 03:39, 03:67, 05:43w, 07:39-07:40, 07:177, 07:210w, 07:238w, 07:328w, 07:335, 08:37, 15:23w, 15:63w, 16:21 and 16:80 and in the B\*08:15 alleles. Specific PCR fragment of 275 bp in the C\*06:27 and the C\*16:47 alleles.

Primer mix 21: Specific PCR fragment of 190 bp in the C\*06:36 and 06:87 and the C\*02:113, 12:45, 12:56, 12:166 and 16:61 and in the A\*68:76:01-68:76:02 alleles. Specific PCR fragment of 215 bp in the C\*06:72 allele. Specific PCR fragment of 380 bp in the C\*06:25 and the C\*04:81 alleles.

Primer mix 24: Specific PCR fragment of 50 bp in the C\*01:108, 03:141, 03:233, 04:210, 07:394 and in the B\*55:53 alleles. Specific PCR fragment of 160 bp in the C\*06:09, 06:34:01-06:34:02, 06:103, 06:107, 06:144 and 06:168 and the C\*01:09, 02:19, 02:22, 02:47, 02:111, 03:21, 03:80, 03:142, 03:287, 04:94:01-04:94:02, 05:08, 05:52, 05:89, 05:106, 07:447, 08:27, 08:29, 08:31, 12:31, 12:144 and 18:03 and in the B\*13:31, 13:41, B\*15:58, 15:73, 15:137, 15:303, B\*39:36, B\*46:61, B\*54:33, B\*55:21 and B\*56:43 alleles. Specific PCR fragment of 210 bp in the C\*06:35 allele.

Primer mix 25: Specific PCR fragment of 85 bp in the C\*06:38 allele. Specific PCR fragment of 190 bp in the C\*06:118 and the C\*01:60, 07:31:01-07:31:02, 07:177, 07:364, 14:17 and 17:21 alleles.

Primer mix 26: Specific PCR fragment of 120 bp in the C\*06:39 and 06:128N and C\*02:100, 04:205N and 15:115N alleles. Specific PCR fragment of 225 bp in the C\*06:146 allele.

Primer mix 27: Specific PCR fragment of 190 bp in the C\*06:42:01-06:42:02 and the C\*14:29 alleles. Specific PCR fragment of 225 bp in the C\*06:40 and the C\*14:63 and in the B\*39:79 alleles.

Primer mix 28: Specific PCR fragment of 90 bp in the C\*06:41 and the C\*12:32 alleles. Specific PCR fragment of 115 bp in the C\*07:394 allele. Specific PCR fragment of 220 bp in the C\*06:47 and 06:123 and the C\*02:57, 07:447 and 12:11 alleles.

Primer mix 29: Specific PCR fragment of 125 bp in the C\*06:152 allele. Specific PCR fragment of 170 bp in the C\*06:43:01-06:43:02, 06:107 and the C\*01:09, 02:19, 03:21, 03:80, 03:142 and 03:287 and in the A\*26:79, A\*68:114, B\*15:207 and B\*15:349 alleles. Specific PCR fragment of 210 bp in the C\*06:47 and the C\*12:11 alleles.

Primer mix 30: Specific PCR fragment of 100 bp in the C\*06:44 and the C\*07:01:13 alleles. Specific PCR fragment of 160 bp in the C\*06:126 and 06:171N alleles.

Primer mix 31: Specific PCR fragment of 110 bp in the C\*06:128N and the C\*04:205N and 15:115N alleles. Specific PCR fragment of 155 bp in the C\*06:45 and the C\*02:85 alleles. Specific PCR fragment of 260 bp in the C\*06:111 and the C\*05:108 alleles.

Primer mix 32: Specific PCR fragment of 85 bp in the C\*06:71 allele. Specific PCR fragment of 195 bp in the C\*06:19 and 06:66 alleles.

Primer mix 33: Specific PCR fragment of 105 bp in the C\*06:60 and 07:427 and in the B\*08:138 alleles. Specific PCR fragment of 240 bp in the C\*06:69, 06:93, 06:122 and the 06:124 alleles.

Primer mix 34: Specific PCR fragment of 175 bp in the C\*06:74Q and the C\*15:32Q alleles. Specific PCR fragment of 275 bp in the C\*06:20 and the C\*01:32, 02:56, 03:102, 03:263, 04:180:01, 07:81, 07:168, 07:450, 08:123 and 12:50 alleles.

Primer mix 35: Specific PCR fragment of 210 bp in the C\*06:134N allele. Specific PCR fragment of 235 bp in the C\*06:65 and 06:93 alleles. Specific PCR fragment of 380 bp in the C\*06:46N allele.

Primer mix 36: Specific PCR fragment of 120 bp in the C\*06:57 allele. Specific PCR fragment of 195 bp in the C\*06:87 and the C\*12:45, 12:166, 16:61 and in the A\*68:76:01-68:76:02 alleles. Specific PCR fragment of 220 bp in the C\*06:58 and 06:81 and the C\*02:37 alleles.

Primer mix 37: Specific PCR fragment of 115 bp in the C\*06:148 and 06:151 alleles. Specific PCR fragment of 210 bp in the C\*06:49N and 06:101 and the C\*04:94:01-04:94:02, 12:10:01-12:10:02 and 18:03 and in the B\*15:27:01-15:27:03, 15:109, 15:327 and 15:344 alleles.

Primer mix 38: Specific PCR fragment of 130 bp in the C\*06:70:01-06:70:02, 06:120 and 06:132:01 and the C\*02:02:13 and 02:02:29 alleles. Specific PCR fragment of 470 bp in the C\*06:73 and the C\*03:171, 03:211:01, 04:144, 05:93, 08:20, 08:40 and 12:109 alleles.

Primer mix 40: Specific PCR fragment of 140 bp in the C\*06:133 and the C\*16:52 alleles. Specific PCR fragment of 275 bp in the C\*06:54 allele.

Primer mix 44: Specific PCR fragment of 120 bp in the C\*06:77 and the C\*03:278 and 16:86 and in the A\*11:47, 11:221, A\*26:89, B\*18:01:20, 18:72:02, B\*37:01:04, B\*40:94 and B\*54:02 alleles. Specific PCR fragment of 215 bp in the C\*06:100 allele.

Primer mix 49: Specific PCR fragment of 195 bp in the C\*06:175N allele. Specific PCR fragment of 280 bp in the C\*06:129 and the C\*05:16, 05:85, 05:107, 07:364 and 08:12 and in the B\*14:32 alleles.

Primer mix 50: Specific PCR fragment of 160 bp in the C\*06:158 allele. Specific PCR fragment of 195 bp in the C\*06:175N allele.

Primer mix 52: Specific PCR fragment of 115 bp in the C\*06:151 allele. Specific PCR fragment of 210 bp in the C\*06:47, 06:136 and 06:142 and the C\*02:17, 02:77, 07:138, 12:11, 12:118, 12:156 and 14:16 and in the B\*15:32:01-15:32:02 and 15:299 alleles.

**4**The HLA-C\*06 primer set cannot separate the C\*06:127:01-06:127:02 and C\*04:220 alleles. These alleles can be distinguished by the HLA-C low resolution kit and/or HLA-C\*04 high resolution kit.

**5**The following HLA-C\*06 alleles can be distinguished by the different sizes of the HLA-specific PCR product:

|  |  |  |  |
| --- | --- | --- | --- |
| **Alleles** | **Primer mix** | **Alleles** | **Primer mix** |
| C\*06:07, 06:33 | 7 | C\*06:45, 06:111 | 31 |
| C\*06:15, 06:116N | 15 | C\*06:49N, 06:148 | 37 |
| C\*06:16N, 06:21 | 16 | C\*06:54, 06:133 | 40 |
| C\*06:20, 06:74Q | 34 | C\*06:57, 06:58 | 36 |
| C\*06:25, 06:36 | 21 | C\*06:66, 06:71 | 32 |
| C\*06:27, 06:29 | 20 | C\*06:70:01-06:70:02, 06:73 | 38 |

**6**This lot of the C\*06 kit cannot discriminate between the C\*06:86 and the **C\***06:02:01:01-06:02:01:03, 06:02:03-06:02:45, 06:48, 06:51-06:52, 06:56, 06:62-06:64, 06:68, 06:75, 06:78, 06:80, 06:82, 06:84-06:85, 06:88-06:92, 06:94-06:95, 06:97-06:99, 06:102, 06:105, 06:108-06:110, 06:112-06:115, 06:117, 06:119, 06:121, 06:125, 06:130-06:131, 06:135, 06:137, 06:141, 06:145, 06:149-06:150, 06:154-06:157, 06:159-06:167, 06:169-06:170, 06:172-06:174 alleles.

The HLA-C\*06 primer set cannot distinguish the silent mutations in the C\*06:02:01:01-06:02:01:03, C\*06:02:03-06:02:45, the C\*06:04:01-06:04:02, the C\*06:34:01-06:34:02, the C\*06:42:01-06:42:02, the C\*06:43:01-06:43:02, the C\*06:53:01-06:53:02, the C\*06:70:01-06:70:02 alleles, the C\*06:106:01-06:106:02 or the C\*06:127:01-06:127:02 alleles.

‘w’, may be weakly amplified.

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