Gel Documentation Form and Worksheet

HLA-B\*57 (101.567-12/12u) Lot No: 7E2 Expiry Date: 2019-07-01

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 Interpretation Table.

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

Primer mix 9 may have tendencies of unspecific amplifications.

Primer mix 15 has a tendency to giving rise to primer oligomer formation.

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.









**1**HLA-B\*57 alleles in bold lettering are listed as confirmed alleles on the on the IMGT/HLA web page [www.ebi.ac.uk/imgt/hla](http://www.ebi.ac.uk/imgt/hla), release 3.25.0, July 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 6: Specific PCR fragment of 95 bp in the B\*57:04:01-57:04:02 and 57:32 and the B\*44:153 alleles. Specific PCR fragment of 180 bp in the B\*57:06 allele. Specific PCR fragment of 205 bp in the B\*57:30 allele.

Primer mix 7: Specific PCR fragment of 105 bp in the B\*57:15 allele. Specific PCR fragment of 170 bp in the B\*57:37 allele. Specific PCR fragment of 295 bp in the B\*57:55 allele.

Primer mix 8: Specific PCR fragment of 90 bp in the B\*57:16 allele. Specific PCR fragment of 200 bp in the B\*57:49 allele. Specific PCR fragment of 250 bp in the B\*57:07 and the B\*44:153 alleles.

Primer mix 11: Specific PCR fragment of 215 bp in the B\*57:09 and 57:24 alleles. Specific PCR fragment of 250 bp in the B\*57:14:01-57:14:02 and 57:43 alleles.

Primer mix 14: Specific PCR fragment of 135 bp in the B\*57:17 and the B\*44:189 and in the C\*07:239 alleles. Specific PCR fragment of 195 bp in the B\*57:10 and the B\*07:219 alleles.

Primer mix 15: Specific PCR fragment of 110 bp in the B\*57:29 and 57:33 and the B\*15:214, 18:81, 35:250 and 51:165 alleles. Specific PCR fragment of 145 bp in the B\*57:11 and the B\*14:01:01-14:02:08, 14:02:10-14:04, 14:07N, 14:09, 14:11-14:12, 14:14-14:36, 14:38-14:52, 14:54, 18:44:01-18:44:02, 39:79, 58:02:01-58:02:02, 58:06-58:07, 58:25, 58:38, 58:43 and 58:60 and in the C\*01:32:01-01:32:02, C\*06:20 and C\*16:98 alleles.

Primer mix 16: Specific PCR fragment of 85 bp in the B\*57:12 and the B\*08:119, 14:24, 35:226, 39:92 and 58:64 alleles. Specific PCR fragment of 160 bp in the B\*57:18 allele.

Primer mix 17: Specific PCR fragment of 140 bp in the B\*57:31 and the B\*40:30 and 40:34 and in the C\*06:72 alleles. Specific PCR fragment of 210 bp in the B\*57:22, 57:27, 57:57 and 57:63 and the B\*55:14 and in the C\*06:72w alleles. Specific PCR fragment of 140 bp and 210 bp in the B\*57:13 and in the C\*07:227 alleles.

Primer mix 19: Specific PCR fragment of 90 bp in the B\*57:04:01-57:04:02 and 57:41 and the B\*44:153 and in the C\*06:72 alleles. Specific PCR fragment of 240 bp in the B\*57:13, 57:25 and 57:43 and the B\*07:227, 40:30 and 40:34 alleles.

Primer mix 20: Specific PCR fragment of 90 bp in the B\*57:20 allele. Specific PCR fragment of 240 bp in the B\*57:26 and the B\*55:14 and in the C\*06:72 alleles.

Primer mix 21: Specific PCR fragment of 110 bp in the B\*57:33 allele. Specific PCR fragment of 150 bp in the B\*57:21 and the B\*35:127 alleles.

Primer mix 22: Specific PCR fragment of 95 bp in the B\*57:34 and 58:54 alleles. Specific PCR fragment of 150 bp in the B\*57:40 and the B\*14:20 alleles.

Primer mix 23: Specific PCR fragment of 75 bp in the B\*57:35 allele. Specific PCR fragment of 100 bp in the B\*57:36 allele.

Specific PCR fragment of 465 bp in the B\*57:05 and 57:67:01 and the B\*13:02:06, 13:26:01, 13:72, 15:16:03, 15:20, 15:85, 15:194, 15:393, 35:46, 35:207, 46:01:07, 46:33 and 58:36 and in the C\*01:73 alleles.

Primer mix 24: Specific PCR fragment of 230 bp in the B\*57:77 and 44:36 and in the A\*03:174, A\*11:65, A\*24:247 and C\*04:83 alleles. Specific PCR fragment of 270 bp in the B\*57:40 and 40:284 and in the C\*07:511 and C\*15:121 alleles.

**4**The following HLA-B\*57 alleles can be distinguished by the different sizes of the HLA-specific PCR product:

|  |  |
| --- | --- |
| **Alleles** | **Primer mix** |
| B\*57:15, 57:37, 57:55 | 7 |
| B\*57:25, 57:41 | 19 |

The HLA-B\*57 subtyping kit cannot distinguish the silent mutations in the B\*57:01:01-57:01:04 and 57:01:06-57:01:22 alleles, the B\*57:02:01-57:02:02 alleles, the B\*57:03:01-57:03:02 alleles or the B\*57:14:01-57:14:02 alleles.

‘w’, might be weakly amplified.

Change in revision R01 compared to R00:

1. Primer mix 15 does not amplify the B\*07:120 and the B\*40:150 alleles. This has been corrected in the Specificity and Interpretation Tables.