Gel Documentation Form and Worksheet

HLA-B\*58 (101.568-06/06u) Lot No: 7E4 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**

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| --- |
| 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 mixes 9, 14, 16 and 17 may have tendencies of primer oligomer formation.

Primer mixes 14 and 17 may have tendencies of unspecific amplifications.

Primer mix 11 may give a lower yield of HLA-specific PCR product than the other HLA-B\*58 primer mixes.

Primer mix 28 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 mix 30 does not amplify the B\*58:62 allele. Thus, this lot cannot discriminate between the B\*58:62 and the 58:01:01:01-58:01:19, 58:32-58:35, 58:37, 58:41-58:42, 58:45:01-58:45:02, 58:47-58:50, 58:53-58:56, 58:63-58:64, 58:67-58:71, 58:73-58:75, 58:77, 58:79-58:81 alleles. This has been corrected in the specificity and interpretation tables.











**1**HLA-B\*58 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 4: Specific PCR fragment of 190 bp in the B\*58:04 allele. Specific PCR fragment of 230 bp in the B\*58:15 allele.

Primer mix 9: Specific PCR fragment of 90 bp in the B\*58:44 and the B\*07:219 alleles. Specific PCR fragment of 160 bp in the B\*58:08:01 and 58:23 and the B\*57:01:20 and in the A\*02:01:33 alleles. Specific PCR fragment of 190 bp in the B\*58:72N allele.

Primer mix 10: Specific PCR fragment of 115 bp in the B\*58:24 and in the C\*03:141, 03:233 and C\*15:18 alleles. Specific PCR fragment of 270 bp in the B\*58:09 and 58:76 and the B\*14:10, 18:22, 18:69, 18:105, 35:21, 35:24:01-35:24:02, 35:81, 35:96, 35:109, 35:157, 35:188, 35:190, 35:233, 35:287, 37:04:01-37:04:02, 40:28, 44:190, 44:203, 51:04, 51:46, 51:56:01-51:56:03, 51:139, 53:02, 53:06, 53:28 and 57:14:01-57:14:02 and in the C\*15:39 alleles.

Primer mix 12: Specific PCR fragment of 145 bp in the B\*58:13 and the B\*49:06 alleles. Specific PCR fragment of 235 bp in the B\*58:10N allele.

Primer mix 15: Specific PCR fragment of 90 bp in the B\*58:36 and the B\*57:01:01-57:04:01, 57:05-57:15, 57:17-57:19, 57:21-57:35, 57:37-57:44, 57:46-57:50, 57:52-57:57, 57:59-57:61, 57:63-57:68, 57:70-57:79N and 57:81-57:83 alleles. Specific PCR fragment of 155 bp in the B\*58:12 and the B\*07:219 and in the A\*02:42 and A\*02:310 alleles. Specific PCR fragment of 90 bp and 155 bp in the B\*57:58 and 57:80 alleles.

Primer mix 18: Specific PCR fragment of 70 bp in the B\*58:25 allele. Specific PCR fragment of 140 bp in the B\*58:17N allele.

Primer mix 20: Specific PCR fragment of 145 bp in the B\*58:39N allele. Specific PCR fragment of 175 bp in the B\*18:51 allele. Specific PCR fragment of 85 bp and 175 bp in the B\*58:29 allele. Specific PCR fragment of 175 bp and 275 bp in the B\*58:21 and the B\*53:34 alleles.

Primer mix 23: Specific PCR fragment of 150 bp in the B\*58:26 and the B\*35:75, 39:106 and 44:138Q alleles. Specific PCR fragment of 260 bp in the B\*58:31N and 58:52 and the B\*51:32 alleles.

Primer mix 24: Specific PCR fragment of 145 bp in the B\*58:39 allele. Specific PCR fragment of 190 bp in the B\*58:72N allele. Specific PCR fragment of 280 bp in the B\*58:40 allele.

Primer mix 25: Specific PCR fragment of 130 bp in the B\*58:57 allele. Specific PCR fragment of 260 bp in the B\*58:52 and the B\*51:32 alleles.

Primer mix 26: Specific PCR fragment of 75 bp in the B\*58:66 allele. Specific PCR fragment of 170 bp in the B\*58:58 allele.

**4**The following HLA-B\*58 alleles can be distinguished by the different sizes of the HLA-specific PCR product:

|  |  |
| --- | --- |
| **Alleles** | **Primer mix** |
| B\*58:10N, 58:13 | 12 |
| B\*58:21, 58:29 | 20 |
| B\*58:58, 58:66 | 26 |

5This lot of the B\*58 kit cannot discriminate between the B\*58:62 and the **B\***58:01:01:01-58:01:19, 58:32-58:35, 58:37, 58:41-58:42, 58:45:01-58:45:02, 58:47-58:50, 58:53-58:56, 58:63-58:64, 58:67-58:71, 58:73-58:75, 58:77 **and** 58:79-58:81 alleles.

The HLA-B\*58 primer set cannot distinguish the following silent mutations: the B\*58:01:01:01-58:01:19 alleles, the B\*58:02:01-58:02:02, the B\*58:16:01-58:16:02 alleles, the B\*58:28:01-58:28:02 or the B\*58:59:01-58:59:02 alleles.

‘w’, might be weakly amplified.

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