Gel Documentation Form and Worksheet

HLA-B\*49 (101.547-06/06u) Lot No: 4E8 Expiry Date: 2019-04-01

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 6, 13 and 15 may have tendencies of unspecific amplifications.

Primer mix 21 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\*49 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 2: Specific PCR fragment of 145 bp in the B\*49:06 and the B\*58:13 alleles. Specific PCR fragment of 190 bp in the B\*49:26 and the B\*39:31 alleles. Specific PCR fragment of 255 bp in the B\*49:02 and the B\*13:23, 13:55, 18:09, 27:01, 27:142, 37:10, 40:47, 40:96, 40:157, 40:201, 44:15, 44:55, 44:103, 44:188 and 44:212 alleles.

Primer mix 3: Specific PCR fragment of 115 bp in the B\*49:22 and the B\*15:33, 15:248, 51:126 and 57:54 and in the C\*03:87:01-03:87:02, C\*05:27, 05:39 and C\*08:115 alleles. Specific PCR fragment of 145 bp in the B\*49:02 and the B\*40:47, 40:96, 40:110, 40:157, 40:201, 44:02:01:01-44:02:12, 44:02:14-44:02:18, 44:02:20-44:05:02, 44:05:04, 44:07, 44:10, 44:12-44:17, 44:19N-44:24, 44:26-44:39, 44:41:01-44:43:02, 44:45, 44:47-44:49, 44:51-44:56N, 44:58N-44:59:02, 44:61N-44:74, 44:76-44:82, 44:84:01-44:89, 44:91-44:94, 44:96-44:128:02, 44:132-44:133, 44:135-44:151, 44:153-44:155, 44:157-44:169, 44:171N-44:195N, 44:197-44:200, 44:202-44:209, 44:211-44:245, 44:247-44:249 and 47:04 and in the C\*16:85 alleles.

Primer mix 4: Specific PCR fragment of 170 bp in the B\*49:03 and the B\*15:32:02, 18:04, 18:125, 40:01:11, 40:58, 44:02:10, 44:34:01, 51:07:01, 52:01:02, 52:01:04, 52:01:09, 52:02:01, 52:03, 52:09, 52:10:03, 52:21 and 52:31:02 alleles. Specific PCR fragment of 205 bp in the B\*49:09 and the B\*13:15, 14:37, 51:62, 51:106, 52:25:01-52:25:02 and 56:34 alleles.

Primer mix 7: Specific PCR fragment of 105 bp in the B\*49:04:01-49:05 and the B\*13:04, 13:46, 13:72, 15:04:01:01-15:04:04, 15:16:01:01-15:16:03, 15:67, 15:95, 15:137, 15:155, 15:222, 15:254, 15:293, 15:310, 15:362, 35:37, 35:235, 35:265, 35:276, 39:06:01-39:06:05, 39:33-39:34, 39:50, 39:57, 39:62, 39:64, 39:83, 39:90, 39:102, 39:109, 40:86, 40:159, 40:330, 41:30, 46:32, 50:09, 50:33, 51:13:01-51:13:02, 51:37, 51:63, 51:92:01-51:92:02, 51:97, 51:172, 52:14, 52:39, 54:14, 55:13, 55:23, 55:32, 55:58, 55:68, 56:22, 56:50, 59:03 and 73:01-73:02 and in the C\*07:463 and C\*12:183 alleles. Specific PCR fragment of 255 bp in the B\*49:20 and the B\*44:15, 44:18, 44:20, 44:47, 44:100, 44:197, 45:01:01-45:01:02, 45:03, 45:05-45:07, 45:11-45:19, 50:02, 51:23, 51:108 and 52:15 alleles. Specific PCR fragment of 105 bp and 255 bp in the B\*45:02 and 45:09 alleles.

Primer mix 8: Specific PCR fragment of 70 bp in the B\*49:04:02-49:05 and the B\*07:78, 07:84, 08:09, 08:84, 13:02:01-13:02:05, 13:02:07-13:04, 13:08-13:09, 13:14-13:16, 13:18-13:19, 13:27, 13:30-13:35, 13:37-13:38, 13:40-13:42, 13:44-13:49N, 13:53-13:56:02N, 13:58-13:59, 13:62, 13:65-13:70, 13:75, 13:81-13:82, 13:84-13:85, 13:88-13:89, 13:91, 13:93, 15:04:01:01-15:04:04, 15:16:01:01-15:16:02, 15:42, 15:67, 15:83, 15:95, 15:137, 15:155, 15:222, 15:254, 15:293, 15:303, 15:310, 15:362, 27:14, 27:81, 27:130, 27:153, 35:60, 35:265, 35:276, 39:06:01-39:06:05, 39:33-39:34, 39:50, 39:57, 39:62, 39:64, 39:83, 39:90, 39:102, 39:109, 40:06:01:01-40:06:15, 40:44, 40:53, 40:70:01-40:70:02, 40:75, 40:83, 40:86, 40:93, 40:95-40:96, 40:103, 40:109-40:110, 40:127, 40:131, 40:148, 40:159, 40:161-40:162, 40:165, 40:177, 40:190, 40:230, 40:244, 40:256N, 40:268-40:270, 40:275, 40:294-40:295, 40:298, 40:306-40:307, 40:311, 40:313, 40:317-40:318, 40:330, 41:01:01-41:01:04, 41:05-41:07, 41:09, 41:12, 41:14, 41:16-41:17, 41:20-41:22, 41:25-41:26, 41:28-41:30, 41:32-41:33, 41:35, 41:37, 41:44, 42:04, 44:20, 44:47, 44:100, 44:197, 45:14, 46:11, 46:18, 46:32, 50:20, 51:01:01:01-51:01:18, 51:01:20-51:03, 51:05, 51:07:01-51:16, 51:18-51:24:05, 51:26-51:41N, 51:43-51:44N, 51:48-51:55, 51:57-51:58, 51:60-51:61:02, 51:63, 51:65-51:80, 51:82-51:93, 51:95-51:134, 51:136-51:138, 51:140-51:147, 51:149N-51:161, 51:163-51:193, 51:195-51:208, 52:01:01:01-52:40, 52:42-52:61, 54:01:01-54:05N, 54:07-54:08N, 54:10-54:14, 54:16-54:35, 55:01:01-55:01:06, 55:01:08-55:03, 55:05, 55:07, 55:09-55:13, 55:15-55:26, 55:28-55:48, 55:50, 55:52-55:78, 56:01:01:01-56:01:08, 56:05:01-56:08, 56:13-56:17, 56:19N-56:30, 56:33-56:50, 58:08:01-58:08:02, 59:01:01:01-59:09, 73:01-73:02 and 78:01:01-78:09 alleles. Specific PCR fragment of 155 bp in the B\*49:19N allele.

Primer mix 9: Specific PCR fragment of 155 bp in the B\*49:21 allele. Specific PCR fragment of 240 bp in the B\*49:07 and the B\*45:10 and 50:31 alleles.

Primer mix 10: Specific PCR fragment of 80 bp in the B\*49:16 allele. Specific PCR fragment of 250 bp in the B\*49:08 and the B\*52:33 alleles.

Primer mix 14: Specific PCR fragment of 155 bp in the B\*49:19N allele. Specific PCR fragment of 190 bp in the B\*49:12 and 49:17 and the B\*51:130 and 51:177 alleles.

Primer mix 15: Specific PCR fragment of 160 bp in the B\*49:13 allele. Specific PCR fragment of 190 bp in the B\*49:17 and the B\*51:177 alleles. Specific PCR fragment of 290 bp in the B\*49:14 allele.

**4**The following HLA-B\*49 alleles can be distinguished by the different sizes of the HLA-specific PCR product:

|  |  |
| --- | --- |
| **Alleles** | **Primer mix** |
| B\*49:07, B\*49:21 | 9 |
| B\*49:08, B\*49:16 | 10 |
| B\*49:13, B\*49:14 | 15 |

The HLA-B\*49 subtyping kit cannot distinguish the silent mutations in the B\*49:01:01-49:01:08 or the B\*49:18:01-49:18:02 alleles.

 ‘w’, may be weakly amplified.