Gel Documentation Form and Worksheet

HLA-A\*23 Lot No: 7E3 Expiry Date: 2019-07-01

(101.421-06/06u)

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 mixes 6, 20, 22 and 27 may give rise to a lower yield of HLA-specific PCR product than the other A\*23 primer mixes.

Primer mix 8, 14 and 23 may have tendencies of unspecific amplifications.

Primer mix 36 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-A\*23 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 3: Specific PCR fragment of 125 bp in the A\*23:03:01-23:03:02 and 23:70 and the A\*11:139, 24:21:01, 24:21:03, 24:208, 29:07, 29:49 and 31:29 and in the B\*18:27 alleles. Specific PCR fragment of 270 bp in the A\*23:25 allele.

Primer mix 7: Specific PCR fragment of 95 bp in the A\*23:08N and the A\*02:82N alleles. Specific PCR fragment of 145 bp in the A\*23:39 and the A\*30:22 alleles. Specific PCR fragment of 205 bp in the A\*23:22 allele.

Primer mix 8: Specific PCR fragment of 140 bp in the A\*23:26 and 23:39 and the A\*30:22 allele. Specific PCR fragment of 170 bp in the A\*23:48 allele. Specific PCR fragment of 215 bp in the A\*23:09 and the A\*01:02, 01:20, 01:188, 01:190 and 24:129 alleles.

Primer mix 10: Specific PCR fragment of 135 bp in the A\*23:23 and in the B\*18:27 alleles. Specific PCR fragment of 240 bp in the A\*23:10 and 23:43 and the A\*11:139w, 24:10:01w-24:10:02w, 24:46, 24:210w, 24:300w and 24:340w alleles.

Primer mix 11: Specific PCR fragment of 200 bp in the A\*23:11N allele. Specific PCR fragment of 270 bp in the A\*23:32 allele.

Primer mix 12: Specific PCR fragment of 135 bp in the A\*23:69 allele. Specific PCR fragment of 190 bp in the A\*23:12 and the A\*24:30, 24:42, 24:309, 25:11 and 32:08 alleles.

Primer mix 13: Specific PCR fragment of 90 bp in the A\*23:30 allele. Specific PCR fragment of 210 bp in the A\*23:02, 23:24 and 23:66 and the A\*24:06, 24:87, 24:138, 24:167, 24:285, 24:289 and 24:339 alleles.

Primer mix 14: Specific PCR fragment of 85 bp in the A\*23:53 and 23:70 and the A\*24:17, 24:41 and 24:208 alleles. Specific PCR fragment of 190 bp in the A\*23:68 allele. Specific PCR fragment of 245 bp in the A\*23:04 and the A\*02:17:01-02:17:03, 02:108, 02:110, 02:268, 02:300, 02:303, 02:617, 24:03:01:01-24:03:03, 24:10:01-24:10:02, 24:18, 24:22, 24:33, 24:94, 24:125, 24:138, 24:167, 24:204, 24:207, 24:210, 24:289, 24:299-24:300, 24:315, 24:339-24:340 and 31:29 and in the B\*18:27 alleles. Specific PCR fragment of 85 bp and 245 bp in the A\*11:139, A\*29:07 and 29:49 alleles.

Primer mix 15: Specific PCR fragment of 210 bp in the A\*23:01:16 and 23:14:01 and the A\*24:13:02 and the 24:315 and in the B\*18:27 alleles. Specific PCR fragment of 290 bp in the A\*23:47 and the A\*24:234 and 24:339 alleles.

Primer mix 16: Specific PCR fragment of 175 bp in the A\*23:14:01-23:14:02 and the A\*02:17:01w-02:17:03w, 24:02:16, 24:06, 24:13:01-24:13:02, 24:18, 24:22, 24:54, 24:87, 24:91, 24:107, 24:133, 24:167, 24:207, 24:227, 24:285, 24:289, 24:315 24:327 and 24:339 alleles. Specific PCR fragment of 205 bp in the A\*24:05, 24:24, 24:130, 24:168, 24:194, 24:199, 24:293, 24:319, 24:344, 26:16, 68:45 and 68:117 alleles. Specific PCR fragment of 175 bp and 205 bp in the A\*11:139, 24:02:01:01-24:02:15, 24:02:17-24:04, 24:07:01-24:11N, 24:17, 24:19-24:21:03, 24:23, 24:25-24:50, 24:55-24:56, 24:58-24:63, 24:66-24:86N, 24:88-24:90:02N, 24:93, 24:95-24:106, 24:108-24:113, 24:115-24:129, 24:131-24:132N, 24:134-24:137, 24:139-24:166, 24:169-24:187, 24:189-24:193, 24:195-24:198, 24:200-24:206, 24:208-24:210, 24:212-24:221, 24:223-24:226:02, 24:229-24:284 24:286-24:288, 24:290, 24:292, 24:294Q-24:295, 24:297-24:298, 24:300-24:303N, 24:305-24:314, 24:317-24:318, 24:320-24:323N, 24:325-24:326, 24:328-24:338, 24:340-24:341, 24:343, 24:345-24:348 and 33:19 alleles. Both fragments may not always be visible.

Primer mix 17: Specific PCR fragment of 125 bp in the A\*23:33 allele. Specific PCR fragment of 225 bp in the A\*23:13 and the A\*03:72, 11:88, 24:07:01-24:07:02, 24:19, 24:24, 24:131, 24:288, 24:290, 24:294Q, 24:339, 24:347, 29:37, 29:56, 30:01:01-30:01:11, 30:11:01-30:11:02, 30:14L-30:20, 30:23-30:26, 30:30-30:31, 30:35-30:43, 30:48-30:49, 30:52-30:54, 30:56, 30:58-30:60, 30:62-30:63, 30:65, 30:72-30:75, 30:78N, 30:81-30:83, 30:86-30:89, 30:91-30:98, 68:45 and 68:117 alleles. Specific PCR fragment of 125 bp and 225 bp in the A\*30:102 allele.

Primer mix 19: Specific PCR fragment of 120 bp in the A\*23:15 allele. Specific PCR fragment of 195 bp in the A\*23:46 and the A\*24:66, 24:308 and 30:75 alleles.

Primer mix 20: Specific PCR fragment of 130 bp in the A\*23:29 and the A\*24:128 alleles. Specific PCR fragment of 195 bp in the A\*23:46 and the A\*24:66, 24:308 and 30:75 alleles. Specific PCR fragment of 230 bp in the A\*23:16 and the A\*24:297 alleles. Specific PCR fragment of 270 bp in the A\*23:32 allele.

Primer mix 22: Specific PCR fragment of 90 bp in the A\*23:31, 23:45, 23:53 and 23:70 and the A\*11:139, 24:17, 24:41, 24:62, 24:106, 24:208, 24:330, 29:07, 29:49 and 31:29 alleles. Specific PCR fragment of 290 bp in the A\*23:19N allele.

Primer mix 23: Specific PCR fragment of 80 bp in the A\*23:45 and the A\*02:41, 02:80, 02:117, 02:289:01, 02:304, 02:454, 24:62, 26:10, 31:67-31:68, 32:28, 32:66 and 33:32:01 alleles. Specific PCR fragment of 170 bp in the A\*23:20 and 23:58 alleles.

Primer mix 24: Specific PCR fragment of 105 bp in the A\*23:49 and the A\*30:06 alleles. Specific PCR fragment of 180 bp in the A\*23:21 allele. Specific PCR fragment of 180 bp in the A\*23:21 allele. Specific PCR fragment of 240 bp in the A\*23:40 and the A\*24:302 alleles.

Primer mix 25: Specific PCR fragment of 95 bp in the A\*23:41 and the A\*02:221, 29:15 and 31:78 alleles. Specific PCR fragment of 205 bp in the A\*23:42 allele.

Primer mix 27: Specific PCR fragment of 85 bp in the A\*23:53 and 23:70 and the A\*11:139, 24:17, 24:41, 24:208, 29:07 and 29:49 alleles. Specific PCR fragment of 200 bp in the A\*23:34 and 23:57 and the A\*02:17:01-02:17:03, 02:108, 02:110, 02:268, 02:300, 02:303, 02:617, 24:13:01, 24:94, 24:188, 24:207 and 24:228 alleles. Specific PCR fragment of 285 bp in the A\*23:35 allele.

Primer mix 29: Specific PCR fragment of 170 bp in the A\*23:48 allele. Specific PCR fragment of 200 bp in the A\*23:37:01-23:37:02 and the A\*24:273 alleles. Specific PCR fragment of 240 bp in the A\*23:40 and the A\*24:302 alleles. Specific PCR fragment of 290 bp in the A\*23:47 and the A\*24:234 and 24:339 alleles.

Primer mix 30: Specific PCR fragment of 110 bp in the A\*23:56 allele. Specific PCR fragment of 245 bp in the A\*23:38N and the A\*24:61 alleles.

Primer mix 34: Specific PCR fragment of 90 bp in the A\*23:18 and the A\*24:267 alleles. Specific PCR fragment of 170 bp in the A\*23:58 allele.

Primer mix 35: Specific PCR fragment of 105 bp in the A\*23:49 and the A\*30:06 alleles. Specific PCR fragment of 260 bp in the A\*23:27 and the A\*24:255 alleles.

**4**The following HLA-A\*23 alleles can be distinguished by the different sizes of the HLA-specific PCR product:

|  |  |  |  |
| --- | --- | --- | --- |
| **Alleles** | **Primer mix** | **Alleles** | **Primer mix** |
| A\*23:08N, 23:22 | 7 | A\*23:13, 23:33 | 17 |
| A\*23:09, 23:26 | 8 | A\*23:16, 23:29 | 20 |
| A\*23:10, 23:23 | 10 | A\*23:41, 23:42 | 25 |

The HLA-A\*23 subtyping kit cannot distinguish the silent mutations in the A\*23:01:01-23:01:15 and 23:01:17-23:01:19 alleles, the A\*23:03:01-23:03:02 alleles and the A\*23:37:01-23:37:02 alleles.

‘w’, may be weakly amplified.

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

Change in revision R01 compared to R00:

1. Primer mix 14 may have tendencies of unspecific amplifications. A footnote has been added in the Specificity Table.