Gel Documentation Form and Worksheet

HLA-A\*01 (101.411-24/06, -24u/06u) Lot No: 0E5 Expiry Date: 2018-12-01

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 mixes 8, 10, 12 and 31 have a tendency of giving rise to primer oligomer formation.

Primer mixes 5, 8, 9, 16, 19, 22, 23, 30, 45 and 47 may have tendencies of unspecific amplifications.

Primer mixes 2, 3 and 12 may give rise to a lower yield of HLA-specific PCR product than the other A\*01 primer mixes.

Primer mix 54 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\*01 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 3.21.1, August 2015.

**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 125 bp in the A\*01:60 and the A\*26:31 and in the C\*06:71 alleles. Specific PCR fragment of 210 bp in the A\*01:09 and in the C\*07:274 alleles.

Primer mix 7: Specific PCR fragment of 60 bp in the A\*01:06 and the A\*02:576 and 31:62 alleles. Specific PCR fragment of 115 bp in the A\*01:86 allele.

Primer mix 8: Specific PCR fragment of 110 bp in the A\*01:07, 01:23, 01:51 and 01:83:01-01:83:02 and the A\*24:243 and 31:35 alleles. Specific PCR fragment of 180 bp in the A\*01:01:01:02N allele.

Primer mix 10: Specific PCR fragment of 120 bp in the A\*01:50 allele. Specific PCR fragment of 155 bp in the A\*01:10 and the A\*11:14w, 11:50Q, 30:26, 80:01:01:01w-80:03w alleles. Specific PCR fragment of 240 bp in the A\*01:137 allele. Specific PCR fragment of 270 bp in the A\*01:29 allele.

Primer mix 11: Specific PCR fragment of 135 bp in the A\*01:13 and 01:28 and in the A\*31:35 alleles. Specific PCR fragment of 180 bp in the A\*01:106 allele. Specific PCR fragment of 275 bp in the A\*01:11N allele.

Primer mix 12: Specific PCR fragment of 90 bp in the A\*02:576, 03:187, 11:155, 31:62, 36:01-36:05 and 68:41 and in the B\*57:65, C\*04:31 and C\*06:137 alleles. Specific PCR fragment of 125 bp in the A\*01:86 and 01:115 allele.

Primer mix 14: Specific PCR fragment of 75 bp in the A\*01:59 allele. Specific PCR fragment of 120 bp in the A\*01:13 and 01:17 alleles.

Primer mix 16: Specific PCR fragment of 180 bp in the A\*01:01:01:02N allele. Specific PCR fragment of 235 bp in the A\*01:15N allele.

Primer mix 17: Specific PCR fragment of 180 bp in the A\*01:106 allele. Specific PCR fragment of 210 bp in the A\*01:16N allele. Specific PCR fragment of 285 bp in the A\*01:101 and the A\*03:87, 11:30 and 30:92 alleles.

Primer mix 21: Specific PCR fragment of 125 bp in the A\*01:44 allele. Specific PCR fragment of 220 bp in the A\*01:155 allele. Specific PCR fragment of 255 bp in the A\*01:20, 01:66 and 01:130 and the A\*02:19, 02:36-02:37, 02:54, 02:255, 02:417, 24:14 and 24:93 alleles.

Primer mix 25: Specific PCR fragment of 80 bp in the A\*01:31N, 01:51 and 01:59 alleles. Specific PCR fragment of 425 bp in the A\*01:19 and 01:173 alleles.

Primer mix 26: Specific PCR fragment of 90 bp in the A\*01:104 and 01:134 and the A\*02:346, 02:427, 11:06, 25:11, 26:03:01, 26:06, 26:21, 26:36, 26:78, 26:92, 26:111 and 80:01:01:01w alleles. Specific PCR fragment of 460 bp in the A\*01:32 allele. Specific PCR fragment of 545 bp in the A\*01:45 and the A\*02:453, 02:557, 03:78, 11:108, 24:271 and 66:17 alleles.

Primer mix 27: Specific PCR fragment of 110 bp in the A\*01:87N allele. Specific PCR fragment of 170 bp in the A\*01:33 and 01:141 and the 03:193 alleles. Specific PCR fragment of 195 bp in the A\*01:109 and the A\*03:182, 11:100 and 11:175 alleles.

Primer mix 28: Specific PCR fragment of 95 bp in the A\*01:01:38L and the A\*24:02:03Q alleles. Specific PCR fragment of 135 bp in the A\*01:132 allele. Specific PCR fragment of 195 bp in the A\*01:109 and the A\*03:182, 11:100 and 11:175 alleles.

Primer mix 29: Specific PCR fragment of 110 bp in the A\*01:69:01-01:69:02 and the A\*03:107, 11:17, 23:09, 24:129, 26:62, 26:72 and 32:24 alleles. Specific PCR fragment of 155 bp in the A\*01:35 and the A\*03:77 and 11:144 alleles. Specific PCR fragment of 230 bp in the A\*01:68 alleles. Specific PCR fragment of 285 bp in the A\*01:101 and the A\*03:87, 11:30 and 30:92 alleles.

Primer mix 30: Specific PCR fragment of 135 bp in the A\*01:95 allele. Specific PCR fragment of 175 bp in the A\*01:07 and 01:100 and the A\*31:35 alleles. Specific PCR fragment of 220 bp in the A\*01:155 alleles. Specific PCR fragment of 280 bp in the A\*01:142 and the A\*24:150 alleles. Specific PCR fragment of 135 bp and 175 bp in the A\*24:243 allele.

Primer mix 32: Specific PCR fragment of 110 bp in the A\*01:57N and in the B\*40:291N, C\*06:152N and C\*07:191N alleles. Specific PCR fragment of 140 bp in the A\*01:43 allele.

Primer mix 34: Specific PCR fragment of 130 bp in the A\*01:60, 01:71 and 01:115 and the A\*26:31 and in the C\*06:71 alleles. Specific PCR fragment of 215 bp in the A\*01:58 allele.

Primer mix 35: Specific PCR fragment of 110 bp in the A\*01:47 and 01:150 alleles. Specific PCR fragment of 180 bp in the A\*01:07, 01:49 and 01:100 and the A\*24:243 and 31:35 alleles. Specific PCR fragment of 240 bp in the A\*01:137 allele.

Primer mix 36: Specific PCR fragment of 90 bp in the A\*01:54 allele. Specific PCR fragment of 140 bp in the A\*01:48 allele. Specific PCR fragment of 175 bp in the A\*01:53N allele. Specific PCR fragment of 235 bp in the A\*01:123N allele.

Primer mix 37: Specific PCR fragment of 65 bp in the A\*01:65 allele. Specific PCR fragment of 120 bp in the A\*01:71 allele. Specific PCR fragment of 155 bp in the A\*01:77 and 01:92 alleles.

Primer mix 38: Specific PCR fragment of 100 bp in the A\*01:50 allele. Specific PCR fragment of 180 bp in the A\*01:62 and the A\*02:315 and 03:85 alleles. Specific PCR fragment of 240 bp in the A\*01:68 and 01:72 and the A\*30:45 alleles.

Primer mix 39: Specific PCR fragment of 160 bp in the A\*01:77 and 01:113 alleles. Specific PCR fragment of 205 bp in the A\*01:39 and the A\*24:26 and 24:314 alleles.

Primer mix 42: Specific PCR fragment of 110 bp in the A\*01:87N allele. Specific PCR fragment of 185 bp in the A\*01:64 and 01:141 alleles. Specific PCR fragment of 255 bp in the A\*01:142 and the A\*24:150 alleles.

Primer mix 43: Specific PCR fragment of 550 bp in the A\*01:45 and 01:56N and the A\*02:453, 02:557, 03:78, 11:69N, 11:108, 24:271 and 66:17 alleles. Specific PCR fragment of 590 bp in the A\*01:107 allele.

Primer mix 44: Specific PCR fragment of 65 bp in the A\*01:98 and the A\*24:87 and 24:285 and in the B\*07:64 alleles. Specific PCR fragment of 245 bp in the A\*01:46 allele.

Primer mix 45: Specific PCR fragment of 65 bp in the A\*01:98 and the A\*24:87 and 24:285 and in the B\*07:64 alleles. Specific PCR fragment of 140 bp in the A\*01:52:01N allele.

Primer mix 46: Specific PCR fragment of 85 bp in the A\*01:81 and the A\*03:26, 11:77 and 11:126 alleles. Specific PCR fragment of 155 bp in the A\*01:40 and 11:160 allele. Specific PCR fragment of 245 bp in the A\*01:72 and 01:147Q and the A\*30:45 alleles.

Primer mix 47: Specific PCR fragment of 295 bp in the A\*01:41 and A\*11:48 alleles. Specific PCR fragment of 325 bp in the A\*01:42 allele. Specific PCR fragment of 450 bp in the A\*01:48 allele. Specific PCR fragment of 550 bp in the A\*01:123N allele.

Primer mix 54: Specific PCR fragment of 70 bp in the A\*01:103 allele. Specific PCR fragment of 155 bp in the A\*01:132 allele.

**4**The following HLA-A\*01 alleles can be distinguished by the different sizes of the HLA-specific PCR product:

|  |  |  |  |
| --- | --- | --- | --- |
| **Alleles** | **Primer mix** | **Alleles** | **Primer mix** |
| A\*01:31N, 01:173 | 25 | A\*01:53N-01:54 | 36 |
| A\*01:32, 01:104 | 26 | A\*01:65, 01:92 | 37 |
| A\*01:47, 01:49 | 35 | A\*01:87N, 01:141 | 27, 42 |
| A\*01:48, 01:123N | 36 | A\*01:137, 01:150 | 10, 35 |
| A\*01:50, 01:62 | 38 |
|  |  |

5This lot of the HLA-A\*01 subtyping kit cannot distinguish the A\*01:01:01:01, 01:01:02-01:01:22, 01:01:24-01:01:37, 01:01:39-01:01:47 and 01:01:49-01:01:72 alleles and the A\*01:37 allele.

The HLA-A\*01 subtyping kit cannot distinguish the silent mutations in the A\*01:01:01:01, 01:01:02-01:01:22, 01:01:24-01:01:37, 01:01:39-01:01:47, 01:01:49-01:01:72 and 01:67:01-01:67:02 alleles, the 01:83:01-01:83:02 alleles or the 01:69:01-01:69:01 alleles.

‘w’, might be weakly amplified.

Change in revision R01 compared to R00:

1. Primer mix 30 does not amplify the A\*01:37 and the A\*11:172 alleles. This has been corrected in the Specificity and Interpretation Tables. Thus, this lot of the HLA-A\*01 subtyping kit cannot distinguish the A\*01:01:01:01, 01:01:02-01:01:22, 01:01:24-01:01:37, 01:01:39-01:01:47 and 01:01:49-01:01:72 alleles and the A\*01:37 allele.