Gel Documentation Form and Worksheet

DR low resolution screening Lot No: 4E9 Expiry Date: 2019-05-01

(101.103-48/12,-48u/12u)

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.

Individual alleles can give to rise to two differently sized specific PCR fragments in primer mix 1, 3, 5 to 11, 13, 14, 16 to 21.

Primer mixes 1, 3, 16, 19 and 22 have a tendency to giving rise to primer oligomer formation.

Primer mixes 3, 4 and 9 may have tendencies of unspecific amplification.

Primer mix 19 has a tendency of primer oligomer formation and also has an intense primer cloud due to the high number of primers present in the primer mix.

The DRB4\*01:03:01:02N allele is amplified by the primer pairs in wells 18 and 22, whereas the DRB4\*02:01N and DRB4\*03:01N null alleles are only amplified by the primer pairs in well 22.

Due to sharing of sequence motifs in codon 38 and 47, DRB3\*01:14 will also be amplified in primer mixes 5, 6, 17, and DRB3\*01:23 and \*02:32 will be amplified in primer mix 5, in addition to primer mix 21.

Due to sharing of sequence motifs, DRB3\*02:27 is amplified by the primer pairs in well 15 in addition to primer mix 21.

Primer mix 24 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**DRB alleles listed on the IMGT/HLA web page 2016-July-14, release 3.25.0, [www.ebi.ac.uk/imgt/hla](http://www.ebi.ac.uk/imgt/hla).

**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**The serological reactivity of all DRB alleles is not known. In this table we use the information in the HLA Dictionary 2004 on the www.ebi.ac.uk/imgt/hla web site, the information available at the [www.worldmarrow.org](http://www.worldmarrow.org) web site and the expert-assigned serological grouping in Tissue Antigens (2009) **73**:95-170.

**4**The following alleles give rise to identical amplification patterns with the DR low resolution screening primer set. These alleles can be separated by the respective high resolution primer sets.

|  |  |
| --- | --- |
| **Alleles** | **Alleles** |
| DRB1\*08:20, 13:18, 13:47, 13:55, 13:158, 13:164 | DRB1\*03:15:02, 13:02:02 |
| DRB1\*08:31, 08:41, 08:75, 11:67, 11:193 | DRB1\*13:13, 13:119, 13:154, 13:156, 14:84, 14:116, 14:144 |

 ‘w’, might be weakly amplified.

‘?’, nucleotide sequence of the primer matching region not known.

Change in revision R01 compared to R00:

1. Due to sharing of sequence motifs, DRB3\*01:14 will also be amplified in primer mixes 5, 6 and 17 in addition to primer mix 21. DRB3\*01:23 and DRB3\*02:32 will also be amplified in primer mix 5.
2. The DRB1\*13:02:02 allele is amplified in primer mix 6.Thus, the DRB1\*03:15:02 and DRB1\*13:02:02 alleles will give rise to identical amplification patterns with the DR low resolution screening primer set. These alleles can be separated by the respective high resolution primer sets.

The Specificity and Interpretation Tables have been changed.

Change in revision R02 compared to R01:

1. Primer mix 6 does not amplify the DRB1\*14:137N and 14:152N allele. Primer mix 15 does not amplify the DRB1\*14:137N allele. This has been corrected in the Specificity and Interpretation Tables.