

Titration of erythrocyte antibodies

(Art. Nr 098)

External quality assessment for quantification of irregular erythrocyte antibodies. The test material is plasma for titration against included and own test erythrocytes.

Frequency: 1/year

Examinations

Titration 1. Ref.erythrocyte + ref.method

Titration 2. Own testery. + ref. method

Titration 3. Ref.erythrocyte + own method

Titration 4. Own testery. + own method

	Dispatched	2022-10-25
	Last date for analysis	2022-11-08
	Closing date	2022-11-08
Scheme coordinator	Jenny Ullhagen	
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Test material

Label	Equalis Art. Nr: 98 2022:01/1 Equalis Art. Nr: 98 2022:01/Ref. eryt.
Description	The test material consists of one sample of plasma from a patient with anti-K (named /1) and one sample with reference erythrocyte (named /Ref. eryt.). Your own testing erythrocytes for titration of anti-K shall also be used.
Storage	On arrival, in refrigerator in +2 till +8 °C
Infectious diseases	HIV-antibodies: Negative HIV-antigen: Negative HBs-antigen: Negative HCV-antibodies: Negative Testing for infectious diseases is performed on individual patient samples. For safety reasons, the test material should always be handled using the same precautions as an unknown patient sample.

Included components

The table on the last page presents all components included in this scheme. The table also has a field for internal notes.

Instruction for investigation

The test shall be performed with two methods: the enclosed method (Reference method, page 2) and your own method.

Titration 1 = reference erythrocyte + reference method

Titration 2 = own testing erythrocyte + reference method

Titration 3 = reference erythrocyte + own method

Titration 4 = own testing erythrocyte + own method

Name your own testing erythrocyte's Rh-phenotype in the column *Rh-phenotype* at line (titration nr) 4 in the registration form.

Read and grade according to local routine for interpretation of results.

The test material should be handled in the same manner as a patient sample, when possible.

Registration of results

The results are registered on Equalis Online.

Specify which reaction strength determines which titer is indicated (for example, +/++).

Registered results may be changed until the closing date.

Reports

A summary of the results is sent to the participants within one month from the closing date.

**REFERENCE METHOD FOR QUANTITATION OF ANTIBODIES
THROUGH TITRATION IN IAT/GEL****Preparation**

- Make a 0.8 % suspension in ID-CellStab of enclosed reference erythrocyte (for titer 1) and of the own testing erythrocyte (for titer 2).
- Label 10 test tubes, or more if needed, with number, dilution (first sample not diluted) and testing erythrocyte code. Make the dilution in PBS with 2-step dilutions with start at dilution 1/2.
- Check the gel cards visually: The gel must be covered by buffer and should not contain any air bubbles.
- Label the gel cards with sample code, testing erythrocyte code and every position in the gel card should be labeled with the dilution.
- Pull off the foil of the gel card carefully.

Performance

1. Add 75 μ L PBS to all test tubes, except for the test tube labeled undiluted.
2. Add 75 μ L patient serum/plasma to the test tube labeled undiluted (sample 1) and the test tube labeled 1/2 (sample 2).
3. Perform the dilutions: Mix carefully and transfer 75 μ L from test tube 1/2 (sample 2) to test tube 1/4 (sample 3), etc.
4. Add 50 μ L testing erythrocyte suspension to each position of the gel card for IAT with wide spectrum AHG.
5. Starting from the test tube with the highest dilution; take 25 μ L of dilute to equal position at the gel card.
6. Incubate for 15 minutes at 37 °C.
7. Centrifuge for 10 minutes at 84 g.
8. Read and grade.
9. Report the titer value provided in the last step of the dilution series which gives a clear positive reaction. (For example, dilution 1:16 is titer 16)

Component*	Notes/ Results** /1	Report the reaction level for the given titer value
Titration 1. Ref.erythrocyt + Ref.metod (Titer)		
Titration 2. Egen testery. + Ref.metod (Titer)		
Titration 3. Ref.erythrocyt + egen metod (Titer)		
Titration 4. Egen testery. + egen metod (Titer)		

* All components included in the scheme.

** The results are registered on Equalis Online. Specify which reaction strength determines which titer is indicated (for example, +/+++).

Registered results may be changed until the closing date.

Lab code: _____

Instrument: _____

Date of analysis: _____

Round dispatched	2022-10-25
Closing date	2022-11-08
Scheme coordinator	Jenny Ullhagen
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Summary of results

40 out of 42 participants have reported results. Two participants reported results for two methods.

Expected result

The expected result is the median of the reference method and reference test erythrocyte results: 64. From this value, the titre should not deviate more than one titre step up or down, which is the expected interval.

Results outside the expected interval

Three participants have reported a total of six results outside of the expected interval, see table 1.

Table 1 Distribution of results outside of the expected interval.

Component	Number of results below the expected interval	Number of results above the expected interval
Titration 1. Reference erythrocyte + reference method	-	1
Titration 2. Own testing erythrocyte + reference method	-	2
Titration 3. Reference erythrocyte + own method	-	1
Titration 4. Own testing erythrocyte + own method	-	2

Comment by Equalis' advisory group for transfusion medicine

16 participants have reported identical answers regardless of the combination of erythrocyte and method, but two of these have not reported answers for all four components. Eight participants have obtained a higher titre in the two examinations containing their own testing erythrocyte, which is the most frequent recurring discrepancy, despite the fact that the phenotype in all cases can be expected to be K+k+. Very few participants specified whether the titrations were performed manually or whether an instrument was used, and if so, what kind of instrument.

Test material

The test material consists of one sample of plasma from a patient with anti-K and one sample with reference erythrocytes.

Based on the previous tests and the results of this round, the samples are homogeneous, stable, and suitable for the external quality assessment scheme.

Expl of colours: Black = As exp. result/No exp. result., Light blue = Accepted, Red = Not as exp.

Sample	Component	Output group	Results	No. of res.	Prop. (%)
1	Titration 1. Ref.erythrocyte + Ref.method (Titer)	All results	32	11	28
			64	22	56
			128	5	13
			256	1	3
		Bio-Rad	32	10	31
			64	17	53
			128	5	16
		Grifols	64	1	100
		Not available	32	1	17
			64	4	67
			256	1	17
		Titration 2. Own testery. + Ref.method (Titer)	All results	32	8
	64			16	42
	128			12	32
	256			1	3
	512			1	3
	Bio-Rad			32	7
			64	13	42
			128	10	32
			256	1	3
	Grifols		128	1	100
	Not available		32	1	17
			64	3	50
		128	1	17	
512		1	17		

Overview

Colour your result: Black = As exp. result/Exp. result not available/Numerical/Text result. Red = Not as expected result

Sample	Component	n	Prop. with expected result (%)	Expected result	Your result
/1	Titration 1. Ref.erythrocyte + Ref.method (Titer)	39	97	32 el. 64 el. 128	64
	Titration 2. Own testery. + Ref.method (Titer)	38	95	32 el. 64 el. 128	64
	Titration 3. Ref.erythrocyte + own method (Titer)	41	98	32 el. 64 el. 128	64
	Titration 4. Own testery. + Own method (Titer)	44	95	32 el. 64 el. 128	64

Historic

	2022:01
	/1
Titration 1. Ref.erythrocyte + Ref.method (Titer)	
Titration 2. Own testery. + Ref.method (Titer)	
Titration 3. Ref.erythrocyte + own method (Titer)	
Titration 4. Own testery. + Own method (Titer)	

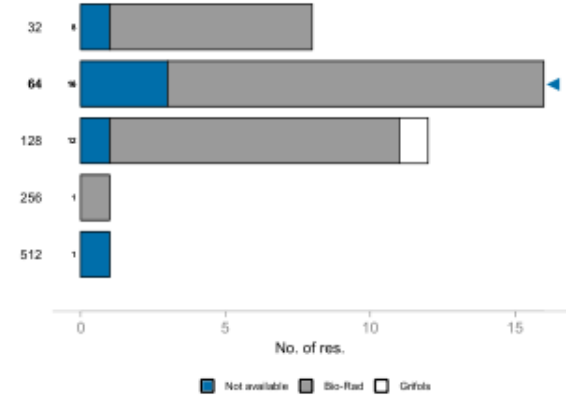
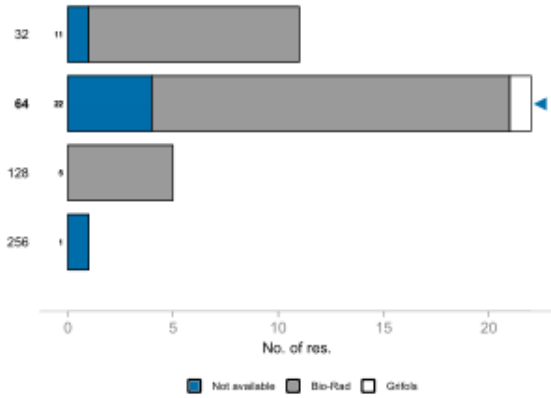
As expected result Not as expected result No result

Titration 1. Ref.erythrocyte + Ref.method (Titer)

Titration 2. Own testery. + Ref.method (Titer)

Expected result: 32 el. 64 el. 128 (Prop.: 97%)
Your result: 64

Expected result: 32 el. 64 el. 128 (Prop.: 95%)
Your result: 64

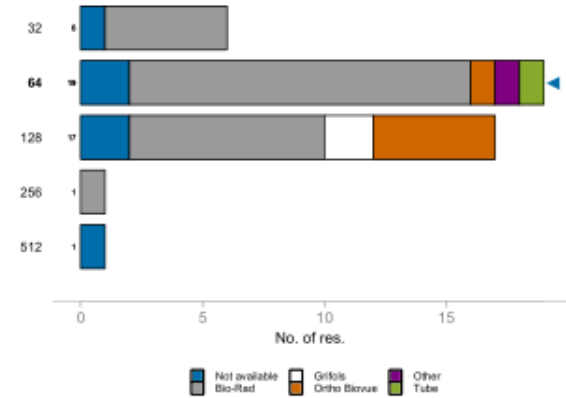
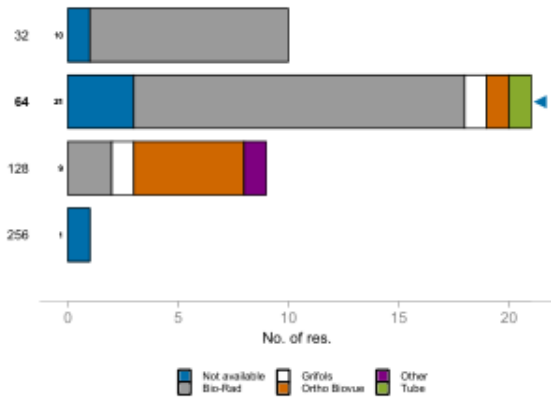


Titration 3. Ref.erythrocyte + own method (Titer)

Titration 4. Own testery. + Own method (Titer)

Expected result: 32 el. 64 el. 128 (Prop.: 98%)
Your result: 64

Expected result: 32 el. 64 el. 128 (Prop.: 95%)
Your result: 64



← Arrow points att bar with own result