

anti-MAG Autoantibodies ELISA

Procedure

FK-MAG-II

Pre-Analytics

Specimen: Serum

Specimen storage: Serum samples are stable for

≥ 1 year if stored at ≤ -20°C.

Freeze/thaw-cycles: Not recommended

Sample Preparation

Dilute samples 1:1000 with incubation buffer.

Allow samples to set for 1 hour at 18-28 °C, vortex from

time to time.

Put samples for 10 minutes on ice prior to pipetting.

Critical Steps

Samples and sample preparation

Samples: Samples must not be hemolytic,

lipemic or icteric

Sample Dilute samples - let set for 1 hour preparation: at 18-28 °C - put for 10 minutes on

ice prior to pipetting.

ELISA

All reagents but TMB (substrate) Reagents

must be used refrigerated.

Washing Wash plate 4 x with refrigerated steps:

wash buffer prior to using it -

empty wells completely.

Use "plate mode" (serial dispensing followed by serial "aspiration") for

automated washing.

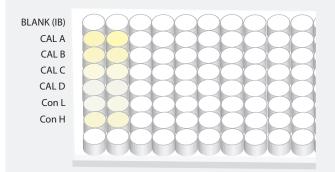
Incubations: Both, first and second incubation

have to be performed at 2-8°C.

Incubation with TMB substrate has to be performed on a plate rotator

at RT (18-28°C).

Microtiter Plate Set-up*



*always perform ELISA measurements in duplicate

ELISA

All reagents but TMB (substrate) must be used refrigerated

Precoated Microtiter Plate



add 100 µl of Controls and diluted Samples (1:1000)

2 h



incubate 2 hrs (±5 min) at 2-8°C

wash 4 x

add 100 µl of Enzyme Label

2 h



incubate 2 hrs (±5 min) at 2-8°C



add 100 µl of TMB Substrate

30'



incubate 30 (±5 min) at 18-28°C on a plate rotator

add 100 µl of Stop Solution

Time to Result: ~5 h





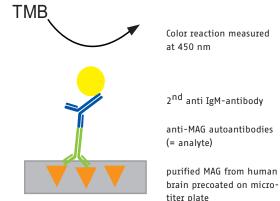
anti-MAG Autoantibodies ELISA

Characteristics EK-MAG-U

Intended Use

The anti-MAG Autoantibodies ELISA is intended for the quantitative determination of human IgM autoantibodies directed against Myelin Associated Glycoprotein (=MAG).

Assay Principle



Kit Formats

| Code | Enzyme label | Wells |
|--------|--------------|--------|
| EK-MAG | IgM | 1 x 96 |

Number of Tests in different Runs

| Number of Runs | Number of Tests | |
|----------------|-----------------|--|
| 1 | 41 | |
| 2 | 34 | |
| 3 | 27 | |
| 4 | 20 | |

Assay Performance Data

Intra-assay precision

The intra-assay precision was calculated from the results of n = 20 pairs of values obtained in a single run (CV range: 2.1 - 10.3%).

Inter-assay precision

15.4 %

6.5 %

The inter-assay precision was calculated from the results of n = 20 results of pairs of values obtained in 20 different runs.

Dilution Linearity / Parallelism

procedures.

147%

Human serum samples with high titers of anti-MAG autoantibodies were diluted from 1:1000 to 1:64000. It is suggested that the relatively high deviation in about 50% of samples is due to antibody aggregation.

BÜHLMANN anti-MAG Autoantibodies ELISA is available for Research Use Only in the US. Not for use in diagnostic

Example of Results

| EK-MAG (IgM) | absorbance (mean 0D450) | BTU | % CV |
|-----------------|----------------------------|-------|------|
| Blank | 0.048 | | |
| CAL A | 2.191 | 70000 | 0.2 |
| CAL B | 1.258 | 15000 | 1.5 |
| CAL C | 0.408 | 3000 | 2.9 |
| CAL D | 0.134 | 1000 | 1.5 |
| CRTL LOW | 0.368 | 2666 | 3.1 |
| CRTL HIGH | 1.389 | 18261 | 0.6 |

Results

The absorbance measured is proportional to the titer of anti-MAG autoantibodies present in a given sample. The titers of the quantitative anti-MAG Autoantibodies ELISA are expressed as BÜHLMANN Titer Units (BTU).

Standardization

The calibrators of the BÜHLMANN anti-MAG Autoantibodies ELISA kit were calibrated against an internal reference consisting of more than 10 human sera with low to high titers of anti-MAG antibodies.

Ordering code:

EK-MAG-U 96 tests (Ig



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