



anti-C1q Ab ELISA

Procedure

EK-AC1QA

A Commitment to Diagnostics

Pre-Analytics

- Samples required: Serum or plasma
- Shipping samples: Samples may be transported at RT (within 24 hrs). Avoid temperatures above 30°C.
- Sample storage: Samples may be stored at 2 -8°C for up to 1 month, at -20°C for up to 6 months. Store samples in aliquots.
- Freeze/thaw-cycles: Not recommended

Special Equipment

- There is no need for any special equipment.

Critical Steps

Samples and sample preparation

- Samples: Samples must not be hemolytic, lipemic or icteric
- Sample preparation: Dilute - vortex - equilibrate for 15 mins prior to using samples in the assay.

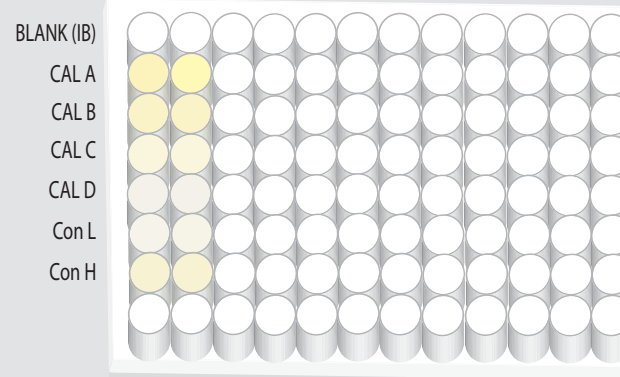
ELISA

- Washing steps: Wash wells 3 x with wash buffer prior to using it - empty wells completely.
Set program to „plate mode“ (serial dispensing followed by serial „aspiration“) when using automated washer.
- Incubations: All incubation steps have to be performed on a plate rotator.

Sample Preparation

Dilute samples 1:50 and put for 15 min on ice for equilibration.

Microtiter Plate Set-up



30'

Time to Result: ~3.0 hrs

ELISA

All reagents must be used between 18 and 28°C. (best assay performance is obtained between 20 and 25°C).

Precoated Microtiter Plate

↓ ↻ wash 2 x

add 100 µl of Standard, Controls and diluted Samples (1:50)

1 hr ↓ ↻ incubate 1 hr ± 5 min at 18-28 °C on a plate rotator
wash 3 x

add 100 µl of Enzyme Label

30' ↓ ↻ incubate 30 ± 5 min at 18-28°C on a plate rotator.
wash 3 x

add 100 µl of TMB Substrate

30' ↓ ↻ incubate 30 ± 2 min at 18-28°C on a plate rotator

add 100 µl of Stop Solution

→ Read at 450 nm (within 30 min)

2.5 hrs



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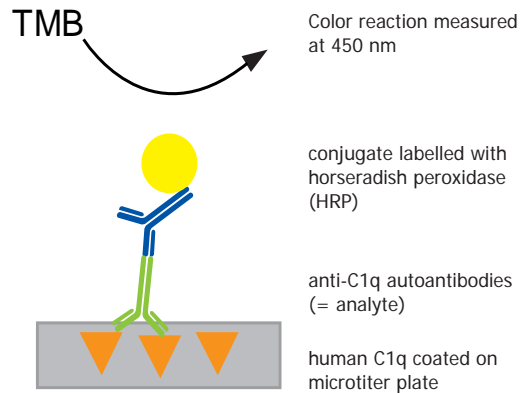
Characteristics

EK-AC1QA

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Intended Use

The anti-C1q autoantibody ELISA from BÜHLMANN is intended for the quantitative *in vitro* diagnostic determination of anti-C1q autoantibodies in human serum or plasma.



Kit Format

Code	Enzyme label	Wells
EK-AC1QA	IgG	1 x 96

Number of Tests in different Runs

Number of Runs	Number of Tests
1	41
2	34
3	27

Assay Performance Data

Intra-assay precision **5.0%**
 Calculated from the results of at least 20 pairs of values from each sample in a single run.

Inter-assay precision **10.8%**
 Calculated from the results of 5 pairs of values in 20 different runs.

Sensitivity **1.0 Units/mL**
 20 duplicates of incubation buffer (= blank) were assayed in a single run. Mean and standard deviation were calculated for the absorbance values. The minimal detectable dose of anti-C1q autoantibodies was calculated to be 1 Unit/mL by adding two standard deviations to the mean absorbance and intersecting this value with the standard curve obtained in the same run.

Dilution / Linearity parallelism **105.9%**
 3 human serum samples with high titers of anti-C1q autoantibodies were diluted with incubation buffer and subsequently assayed according to the assay procedure.

Expected Values and Cut-off
 The frequency of anti-C1q autoantibodies in normal human sera was determined using blood samples from apparently healthy asymptomatic volunteer blood donors (adult men and women at the age of 18-70 years). N=220 samples were assayed according to the assay procedure (values below: U/mL and after elimination).

Range: **2.0 - 22.1**
Mean: **5.6**
Median: **3.8**
SD: **4.2**
Mean + 3SD: **18.2**
Suggested Cut-off Value: **15.0**

Example of Results

EK-AC1Q	Mean OD	% CV
Blank	0.068	
Calibrator A	2.581	0.4
Calibrator B	1.097	0.8
Calibrator C	0.301	0.9
Calibrator D	0.040	10.6
Control low	0.033	8.6
Control high	1.633	2.5

Explanation: the above results were generated with a particular lot number (#1011) according to the Instructions for Use and are for demonstration purposes only. A standard curve must be generated for each set of samples to be assayed.

Results

The absorbance measured is proportional to the titer of anti-C1q autoantibodies bound in the initial step. The titer of the quantitative anti-C1q antibody ELISA is expressed as Units/mL.

Standardisation

The Calibrators of the anti-C1q autoantibody ELISA are calibrated against an internal reference pool, which consists of a plasmapheresis of clinically defined HUVS patients.

Ordering codes:
 EK-AC1QA (IgG)
 EK-CIC 96 wells
 96 wells



BÜHLMANN Laboratories AG
 Baselstrasse 55
 CH-4124 Schönenbuch/Basel
 Switzerland

Phone +41 61 487 12 12
 Fax orders +41 61 487 12 99
 info@buhlmannlabs.ch
 www.buhlmannlabs.ch