Human Apolipoprotein A1 ELISA development kit

Product Code: 3710-1H-6

CONTENTS, development kit for 6 plates:

Vial 1 (green top)
Monoclonal antibody HDL 110 (150 µl)
Concentration: 1 mg/ml

Vial 2 (yellow top)
Biotinylated monoclonal antibody HDL 44 (80 µl)
Concentration: 1 mg/ml

Vial 3 (white top)
Streptavidin-Horseradish Peroxidase (80 µl)

Vial 4
Lyophilised purified apoA1 standard batch 9 (4 µg)

To ensure total recovery of stated quantity, vials have been overfilled.

STORAGE:
Shipped at ambient temperature. On arrival box 1 should be stored refrigerated at 4-8°C and box 2 should be stored frozen at -20°C.

General

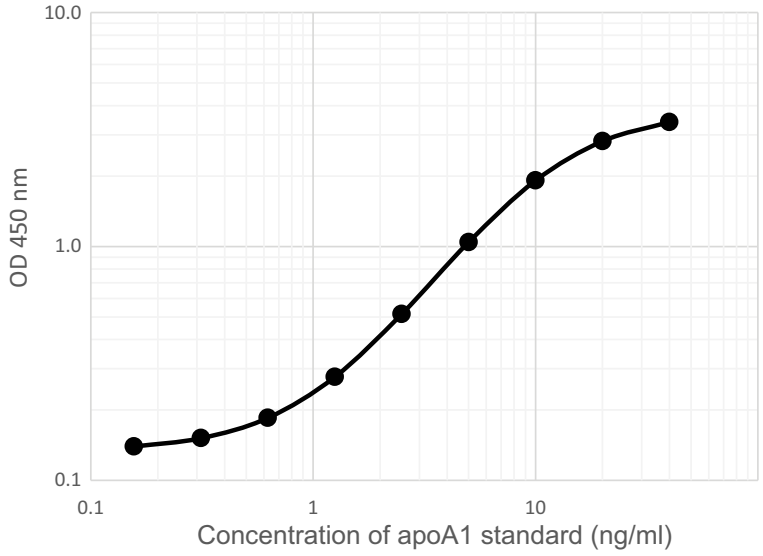
Intended use: For quantitative determination of human Apolipoprotein A1 (apoA1) in serum/plasma samples and cell culture supernatants.
Please note that wash-, block- and incubation buffers should contain detergent. Tween 20, Triton X-100 or NP40 can be used at a concentration of 0.05-0.5%. In block and incubation buffers it is recommended to use 0.1% BSA, but not bovine serum, as HDL 44 also binds bovine apoA1.

Serum/plasma samples: When analyzing human serum/plasma samples it is recommended to use Apo ELISA buffer (product code: 3652-M2) for dilution of samples, standard and detection antibody. The buffer prevents false positive read-outs which may be caused by interference of heterophilic antibodies commonly found in human plasma and serum. Triton X-treatment of samples, necessary for apoB analysis, will not interfere with apoA1 analysis. It is recommended to dilute serum/plasma samples 150,000x to 200,000x, see dilution guidelines at https://www.mabtech.com/knowledge-center/apodilution. Avoid repeated freezing-thawing cycles and do not store samples in -20°C. Samples stored in -20°C will give false high apoA1 values.

Reagents: Antibodies are supplied in sterile-filtered (0.2 µm) PBS with sodium azide (0.02%). Streptavidin-HRP is supplied in PBS with 1% BSA and 0.002% Kathon CG.

Standard range: 0.6-40 ng/ml

Standard calibration: The standard has been calibrated against an international standard from WHO. One ng of supplied standard equals one ng of WHO-IFCC:SP1-01 standard. Please note that the calibration is batch specific.



Guidelines for Human Apolipoprotein A1 ELISA

Day 1 1. Coat a high protein binding ELISA plate with mAb HDL 110, diluted to 2 µg/ml in PBS, pH 7.4, by adding 100 µl/well. Incubate overnight at 4-8°C.

Day 2 2. Wash twice with PBS (200 µl/well).

3. Block plate by adding 200 µl/well of PBS with 0.05% Tween 20 containing 0.1% BSA (incubation buffer). Incubate for 1 hour at room temperature.

4. Wash five times with PBS containing 0.05% Tween.

5. Prepare apoA1 standard by reconstituting contents of vial 4 in 1 ml PBS with 1% BSA, do not stir and leave at room temperature for 15 minutes followed by vortex for 3 sek. This gives a stock solution of 4 µg/ml which should be used immediately or stored in aliquots at -20°C for future use. We recommend the aliquots not to be refrozen after initial use. For the test, prepare dilutions of the stock using the standard range as a guideline.

6. Add 100 µl/well of samples or standards diluted in incubation buffer or Apo ELISA buffer for serum/plasma samples and incubate for 1 to 2 hours at room temperature. Dilution recommendations for serum/plasma samples can be found at https://www.mabtech.com/knowledge-center/apodilution.

7. Wash as in step 4.

8. Add 100 µl/well of mAb HDL 44-biotin at 0.5 µg/ml in incubation buffer or Assay buffer for serum/plasma samples. Incubate for 1 hour at room temperature.

9. Wash as in step 4.

10. Add 100 µl/well of Streptavidin-HRP diluted 1:1000 in incubation buffer. Incubate for 1 hour at room temperature. **Please note that sodium azide used in buffers will inhibit HRP activity.**

11. Wash as in step 4.

12. Add 100 µl/well of appropriate substrate solution e.g. TMB, available from Mabtech product code 3652-F10.

13. Measure the optical density in an ELISA reader after suitable developing time. If required stop the reaction first.

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Updated on 2020/03/31

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CERTIFIED ISO 9001 Quality management systems

CERTIFIED ISO 13485 Medical devices Quality management systems

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