MABTECH

ELISA Flex: Monkey IgA (ALP)

3860M-1A-6 I

ELISA Flex kit for quantitative determination of native monkey IgA in solution, e.g. serum/plasma samples or cell supernatants.

The kit includes		3860M-1A-6 for 6 plates
Capture mAb:	MT57 (0.5 mg/ml)	300 µl
Detection Ab:	anti-IgA, biotin (0.5 mg/ml)	50 µl
Streptavidin-ALP		80 µl
Purified human IgA ELISA standard for Monkey IgA ELISA		1 vial
Standard reconstitution buffer A5		1 ml

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

Shipping and storage

Shipped at ambient temperature. All reagents should be stored at 4-8 °C upon receipt, except the standard which should be stored at -20 °C. Antibodies are supplied in sterile-filtered PBS with sodium azide (0.02%). Streptavidin-ALP is supplied in 0.1 M Tris buffer with 0.002% Kathon CG. The expiry date indicates how long unopened products, stored according to instructions, are recommended for use.

General and Preparations

Specificity

The kit contains a matched pair of antibodies specific for human IgA. The antibodies cross-react with IgA from non-human primates (NHP). Please visit www.mabtech.com for reactivity on NHP species.

Standard range

2-200 ng/ml

Calibration

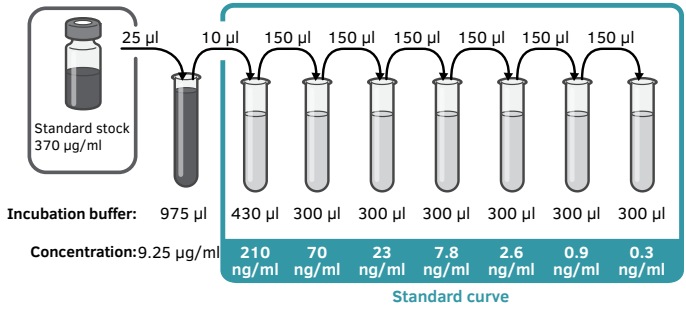
Purified lyophilised human IgA standard is included since international shipping of monkey derived material is prevented by CITES regulations. The standard has been calibrated to yield an ELISA curve corresponding to a standard curve obtained with purified IgA from cynomolgus and rhesus macaques.

Reconstitution of ELISA standard

Reconstitute the ELISA standard to a stock solution of 370 µg/ml by adding 0.5 ml of the standard reconstitution buffer. Allow the standard to dissolve for 5 minutes and mix thoroughly. The standard should be kept in aliquots at -20 °C. Avoid repeated freeze-thaw cycles.

Preparation of standard curve

Prepare within 30 minutes of use. Volumes are sufficient for duplicates.



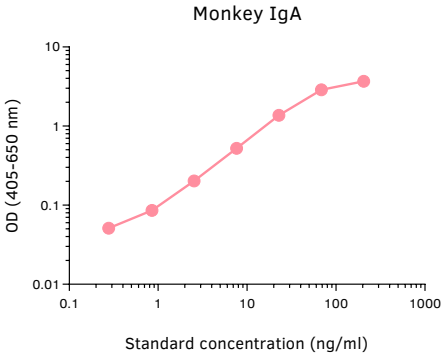
Protocol

Day 1

1. Dilute capture mAb MT57 to 2 µg/ml in PBS, pH 7.4, and filter the solution through a 0.2 µm filter. Add 100 µl/well of the solution. Incubate overnight at 4-8 °C.

Day 2

2. Empty the plate and add 200 µl/well of PBS with 0.05% Tween 20 and 0.1% BSA (incubation buffer) to block the plate. Incubate for 1 hour at room temperature.
3. Wash the plate 5 times with PBS containing 0.05% Tween 20 (300 µl/well).
4. Add 100 µl/well of samples or standards diluted in incubation buffer. Include assay background control, i.e. wells without standard. Incubate for 2 hours at room temperature.
5. Wash as above.
6. Add 100 µl/well of detection Ab IgA-biotin diluted to 0.25 µg/ml in incubation buffer. Incubate for 1 hour at room temperature.
7. Wash as above.
8. Add 100 µl/well of Streptavidin-ALP diluted 1:1000 in incubation buffer. Incubate for 1 hour at room temperature.
9. Wash as above.
10. Add 100 µl/well of pNPP substrate (product code: 3652-P10) and incubate the plate for approximately 60 minutes.
11. Measure the optical density in an ELISA reader at 405 nm. Preferably use a reader capable of subtracting a reference wavelength of between 570 and 650 nm. Representative standard curve shown below.



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