## Datasheet & Protocol

## MABTECH

# ELISA Flex: Monkey IgA (HRP)

3860M-1H-6 |

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

The kit includes		<b>3860M-1H-6</b> for 6 plates	
Capture mAb:	MT57 (0.5 mg/ml)	300 µl	
Detection Ab:	anti-IgA, biotin (0.5 mg/ml)	50 µl	
Streptavidin-HRP		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-HRP is supplied in PBS 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

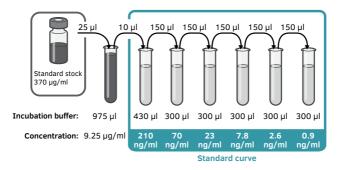
Purified lyophilised human IgA standard is included since international shipping of monkey derived material is prevented by CITES regulations. The standard has been calibrated to yield an ELISA curve corresponding to a standard curve obtained with purified IgA from cynomolgus and rhesus macaques.

#### **Reconstitution of ELISA standard**

Reconstitute the ELISA standard to a stock solution of 370  $\mu$ g/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.



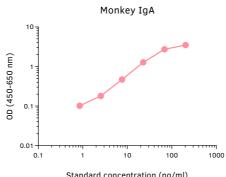
### Protocol

Day 1

**1.** Dilute capture mAb MT57 to 2  $\mu$ g/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 anti-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-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.
- 9. Wash as above.
- **10.** Add 100 μl/well of TMB substrate (product code: 3652-F10) and incubate for 15 minutes.
- **11.** Add 100  $\mu$ l/well of 0.2 M H<sub>2</sub>SO<sub>4</sub> to stop the reaction.
- **12.** Measure the optical density in an ELISA reader at 450 nm within 15 min. Preferably use a reader capable of subtracting a reference wavelength of between 570 and 650 nm. Representative standard curve shown below.



Standard concentration (ng/ml)



Developed and manufactured by MABTECH AB, Sweden, whose quality management system complies with the standards ISO 9001:2015 & ISO 13485:2016.



#### The products are for research use only.

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