## Datasheet & Protocol

## MABTECH

# ELISA Flex: Human IgA (ALP)

3860-1AD-6 |

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

The kit includes	<b>3860-1AD-6</b> for 6 plates	
Capture mAb: MT57 (0.5 mg/ml)	300 µl	
Detection mAb: MT20, ALP	80 µl	
Human IgA ELISA standard	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%). The detection antibody is supplied in 0.1 M Tris-buffer with 1% BSA and 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 monoclonal antibodies (mAbs) specific for human IgA.

Standard range 0.2-100 ng/ml

#### Calibration

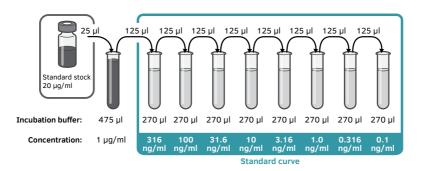
The ELISA standard has been calibrated against an international standard from the National Institute of Biological Standards and Control (NIBSC), Potters Bar, Hertfordshire EN6 3QG, UK. One  $\mu$ g of supplied standard equals 83 mU NIBSC-standard. Please note that the calibration is batch specific.

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

Reconstitute the ELISA standard to a stock solution of  $20 \ \mu$ 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

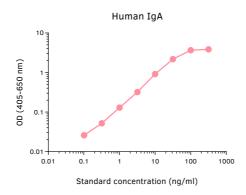
**1.** Add 100  $\mu$ /well of capture mAb MT57 diluted to 2  $\mu$ g/ml in PBS, pH 7.4. Use high protein binding ELISA plates. Incubate overnight at 4-8 °C.

Day 2

- **2.** Empty the plate and add 200  $\mu$ /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  $\mu$ l/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.
- Add 100 μl/well of detection mAb MT20-ALP diluted 1:1000 in incubation buffer. Incubate for
  hour at ream temperature

1 hour at room temperature.

- 7. Wash as above.
- **8.** Add 100 μl/well of pNPP substrate (product code: 3652-P10) and incubate the plate for approximately 60 minutes.
- **9.** 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.





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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