

ELISA Flex: Mouse IgEa (allotype a)

3816-1A-6 |

ELISA Flex kit for quantitative determination of mouse IgEa (allotype a).

The kit includes	3816-1A-6 for 6 plates
Capture mAb: MT18E (0.5 mg/ml)	300 µl
Detection mAb: MT44E, biotin (0.5 mg/ml)	80 µl
Streptavidin-ALP	80 µl
Purified mouse IgE 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%). 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 monoclonal antibodies (mAbs) specific for mouse IgE^a (allotype a). The ELISA does not detect IgE^b (allotype b).

Standard range

30-3000 pg/ml

Calibration

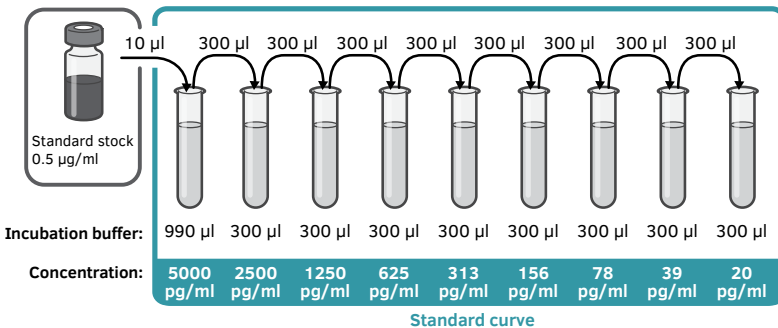
No international standard exists for calibration.

Reconstitution of ELISA standard

Reconstitute the ELISA standard to a stock solution of 0.5 µg/ml by adding 1 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.



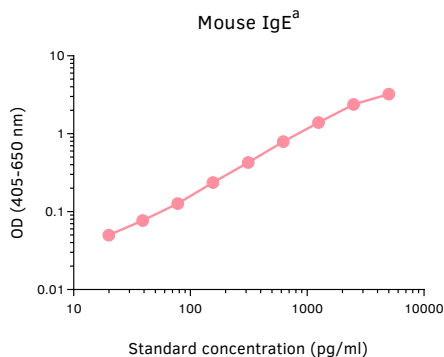
Protocol

Day 1

1. Add 100 μl /well of capture mAb MT18E diluted to 2 $\mu\text{g}/\text{ml}$ in PBS, pH 7.4. Use high protein binding ELISA plates. Incubate overnight at 4-8 $^{\circ}\text{C}$.

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 mAb MT44E-biotin diluted to 0.5 $\mu\text{g}/\text{ml}$ 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 at room temperature protected from direct light 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.



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



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