# Mouse IgM ELISA development kit

Product Code: 3885-1AD-6

**CONTENTS:** development kit for 6 plates

### Vial 1 (green top)

Monoclonal antibody MT6A3 (300  $\mu l$ )

Concentration: 0.5 mg/ml

## Vial 2 (yellow top)

Monoclonal antibody MT9A2-ALP (80 μl)

#### Vial 3

Lyophilised mouse IgM standard

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

#### **STORAGE:**

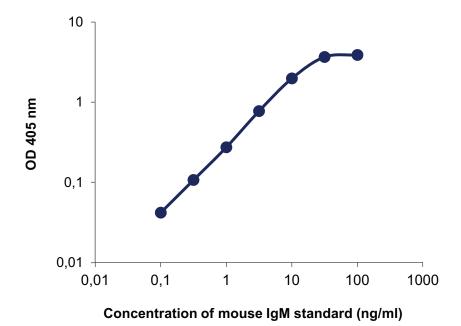
Shipped at ambient temperature. On arrival box 1 should be stored refrigerated at 4-8°C and box 2 should be stored frozen at -20°C.

## General

**Intended use:** For quantitative determination of native mouse IgM in serum, plasma or cell supernatant.

**Reagents:** mAb MT6A3 is supplied in sterile-filtered (0.2  $\mu$ m) PBS with sodium azide (0.02%). MT9A2-ALP is supplied in 0.1 M Tris-buffer with 1% BSA and 0.002% Kathon.

Standard range: 0.3-30 ng/ml



# Guidelines for Mouse IgM ELISA

- **Day 1** 1. Coat a high protein binding ELISA plate with mAbs MT6A3, diluted to 2  $\mu$ g/ml in PBS, pH 7.4, by adding 100  $\mu$ l/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 (PBS-Tween) containing 0.1% BSA (incubation buffer\*). Incubate for 1 hour at room temperature.
  - 4. Wash five times with PBS-Tween.
  - 5. Prepare mouse IgM standard by reconstituting contents of vial 3 in 500 μl PBS to make up a stock solution of 50 μg/ml. The stock solution should be used immediately or stored in aliquots at -20°C for future use. We recommend the aliquots not 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. It is recommended to make serial dilutions of samples to obtain results within the standard range. Incubate for 2 hours at room temperature.
  - 7. Wash as in step 4.
  - 8. Add 100 µl/well of MT9A2-ALP diluted 1:1000 in incubation buffer. Incubate for 1 hour at room temperature.
  - 9. Wash as in step 4.
  - 10. Add 100 μl/well of appropriate substrate solution e.g. p-nitrophenyl-phosphate (pNPP).
  - 11. Measure the optical density (405 nm for pNPP) in an ELISA reader after suitable developing time.
    - \* The same buffer is used for blocking and for dilution.

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