

Mouse IgM ELISA development kit

Product Code: 3845-1AD-6

CONTENTS:

Vial 1 (red top)

Monoclonal anti-IgM antibody (150 μ l)
Concentration: 0.5 mg/ml

Vial 2 (yellow top)

ALP-conjugated anti-IgM antibody (150 μ l)

Vial 3

Lyophilised mouse IgM standard

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

STORAGE:

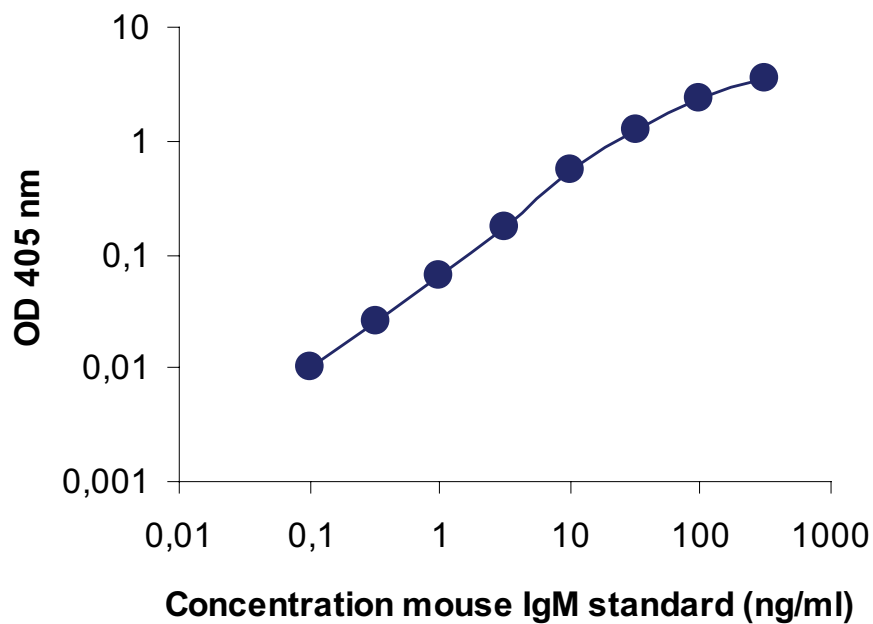
Shipped at ambient temperature. On arrival box 1 should be stored refrigerated at 4-8°C and box 2 should be stored frozen at -20°C.

General

Intended use: For quantitative determination of native mouse IgM in serum and plasma. May cross-react with immunoglobulins from other species.

Reagents: Monoclonal anti-IgM antibody is supplied in sterile-filtered (0.2 μm) PBS with sodium azide (0.02%). ALP-conjugated anti-IgM antibody is supplied in 0.1 M Tris-buffer with 0.002% Kathon CG.

Recommended standard dilution: 0.1-500 ng/ml



Guidelines for Mouse IgM ELISA

- Day 1**
1. Coat a high protein binding ELISA plate with monoclonal anti-IgM antibody, diluted to 1 $\mu\text{g}/\text{ml}$ in PBS, pH 7.4, by adding 100 $\mu\text{l}/\text{well}$. Incubate overnight at 4-8°C.
- Day 2**
2. Wash twice with PBS (200 $\mu\text{l}/\text{well}$).
 3. Block plate by adding 200 $\mu\text{l}/\text{well}$ 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 $\mu\text{g}/\text{ml}$. 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 $\mu\text{l}/\text{well}$ of samples or standards diluted in incubation buffer and incubate for 2 hours at room temperature.
 7. Wash as in step 4.
 8. Add 100 $\mu\text{l}/\text{well}$ of anti-IgM-ALP diluted 1:500 in incubation buffer. Incubate for 1 hour at room temperature.
 9. Wash as in step 4.
 10. Add 100 $\mu\text{l}/\text{well}$ 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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