

ELISA PRODUCT INFORMATION & MANUAL

Bovine IL-5 ELISA Development Kit NBP3-18273

Enzyme-linked Immunosorbent Assay for quantitative detection. For research use only.

Not for diagnostic or therapeutic procedures.

Bovine IL-5 ELISA Development Kit

ELISA Flex kit for quantitative determination of native and recombinant bovine IL-5 in solution, e.g. cell supernatant and serum/plasma.

The kit includes	NBP3-18273 for 6 plates
Capture mAb: MT52B34 (0.5 mg/ml)	300 μΙ
Detection mAb: MT11B37, biotinylated (0.5 mg/ml)	150 μΙ
Streptavidin-HRP	اμ 08
Recombinant bovine IL-5 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 $^{\circ}$ C upon receipt, except the standard which should be stored at -20 $^{\circ}$ 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 monoclonal antibodies (mAbs) specific for native and recombinant bovine IL-5.

Standard range

4-1000 pg/ml

Calibration

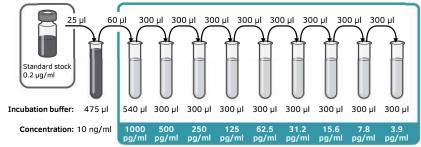
No international standard exists for calibration.

Reconstitution of ELISA standard

Reconstitute the ELISA standard to a stock solution of 0.2 μ g/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

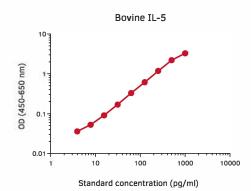
Prepare within 30 minutes of use. Volumes are sufficient for duplicates.



Standard curve

Protocol

- **1.** Add 100 μl/well of capture mAb MT52B34 diluted to 2 μg/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 μ l/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 μ l/well of detection mAb MT**11**B37-biotin diluted to 1 μ g/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.
 - 9. Wash as above.
 - 10. Add 100 µl/well of TMB substrate and incubate for 15 minutes.
 - 11. Add 100 µl/well of 0.2 M H₂SO₄ 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.



Quality management system complies with the standards ISO 9001:2015 & ISO 13485:2016.

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