ELISA PRODUCT INFORMATION & MANUAL

IL-4 ELISA Development Kit

NBP3-11748

Enzyme-linked Immunosorbent Assay for quantitative detection. For research use only.
Not for diagnostic or therapeutic procedures.

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IL-4 ELISA DEVELOPMENT KIT

Development kit for quantitative determination of native and recombinant bovine IL-4 in solution, e.g. cell supernatant.

The kit includes		NBP3-11748 for 6 plates
Capture mAb:	bIL4-I (0.5 mg/ml)	300 µl
Detection mAb:	bIL4-II, biotinylated (0.5 mg/ml)	80 µl
Streptavidin-HRP		80 µl
Recombinant bovine IL-4 ELISA standard		1 vial
Standard reconstitution buffer A8		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-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-4. The mAbs cross-react with native IL-4 from sheep.

Standard range

20-2000 pg/ml

Calibration

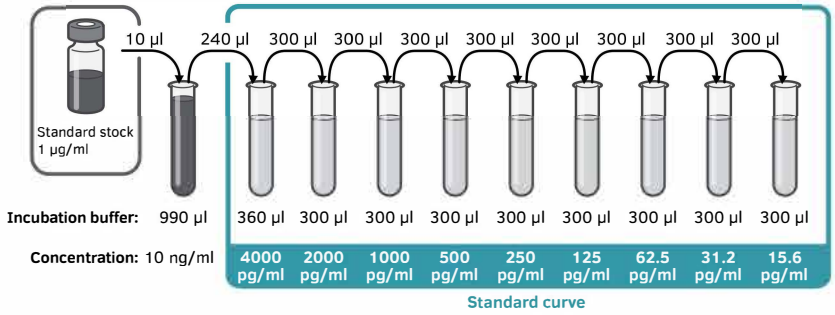
No international standard exists for calibration.

Reconstitution of ELISA standard

Reconstitute the ELISA standard to a stock solution of 1 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 bIL4-I 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 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 bIL4-II-biotin diluted to 0.5 µg/ml in incubation buffer. Incubate for 1 hour at room temperature.

7. Wash as above.

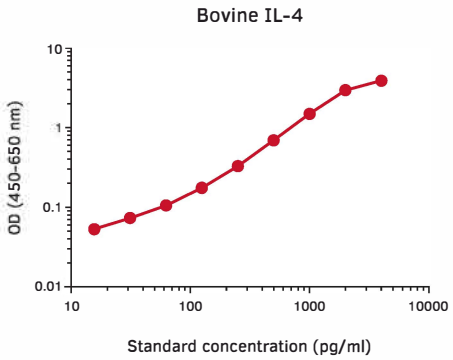
8. Add 100 µl/well of Streptavidin-HRP diluted 1:1000 in incubation buffer. Incubate for 1 hour at room temperature. Please note that sodium azide used in buffers will inhibit HRP activity.

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 H_2SO_4 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.



Updated on 2022/02/23

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

The products are for research use only.