Datasheet & Protocol



ELISA Flex: Equine IgG (ALP)

3153-1A-6 |

ELISA Flex kit for quantitative determination of native equine IgG in solution, e.g. serum/plasma samples or cell supernatants.

The kit includes		3153-1A-6 for 6 plates	
Capture mAb:	MT329A4 (0.5 mg/ml)	300 μΙ	
Detection mAb:	MT310A61, biotin (0.5 mg/ml)	80 μΙ	
Streptavidin-ALP		80 μΙ	
Equine IgG 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 equine (horse) IqG.

Standard range

0.316-31.6 ng/ml

Calibration

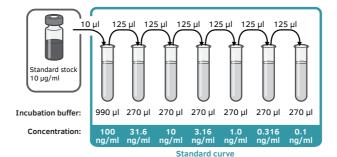
No international standard exists for calibration.

Reconstitution of ELISA standard

Reconstitute the ELISA standard to a stock solution of 10 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.



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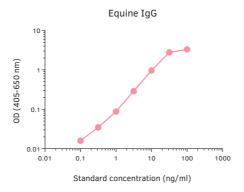
Protocol

Day 1

1. Add 100 μ l/well of capture mAb MT329A4 diluted to 2 μ g/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 μ l/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 MT310A61-biotin diluted to 0.5 μ g/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.



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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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