CERTIFICATE OF ANALYSIS

Research use only. Not for use in diagnostic procedures.

AlphaScreen®

IgG (Protein A based) Detection Kit

Product number: 6760617HV **Lot Number:** 3355432

Product Format: 6760617C: 500 assay points

6760617M: 10 000 assay points

6760617R: 50 000 assay points

The number of assay points is based on an assay volume of 25 µL in 384-well assay plates using a final bead concentration of 20 µg/mL.

Manufacturing date: November 9, 2024 **Document version:** 1

Kit Components

Component	6760617C	6760617M	6760617R
Protein A Acceptor Beads at 5 mg/mL in 0.1M Tris, 0.05% Kathon, pH 8.0	1 x 50 µL (6760136)	1 x 1 mL (6760137)	1 x 5 mL (6760137B)
Streptavidin Donor Beads at 5 mg/mL in 0.1M Tris, 0.05% Kathon, pH 8.0	1 x 50 µL (6760007)	1 x 1 mL (6760008)	1 x 5 mL (6760008B)
Biotinylated-rIgG at 0.05 µM in 1X PBS, 0.1% BSA, 0.05% Kathon, pH 7.2	1 x 50 µL (6760264)	1 x 50 µL (6760264)	1 x 50 µL (6760264)
10x Buffer : 1M Tris, 0.1% Tween-20, 0.05% Kathon, pH 8.0	1 x 1.5 mL (6760028G)	1 x 1.5 mL (6760028G)	1 x 1.5 mL (6760028G)

Product Information

Antibody/Protein: The Protein A acceptor beads are coated with Protein A. Protein A binds the Fc region of many IgG.

Stability: This kit is stable for at least 9 months from the date of manufacture when stored in its original packaging and the recommended storage conditions.

Storage: Store undiluted at 4°C protected from light. Freeze-thaw is not recommended and cause the beads to form aggregates.

Recommended Use: AlphaScreen® Donor beads are light-sensitive. All Alpha assays using the Donor beads should be performed under subdued laboratory lighting (< 100 lux). Green filters (LEE 090 filters) can be applied to light fixtures.

Quality Control

Alpha maximum signal, minimum signal and EC50 are determined using a biotinylated rIgG titration assay performed on an EnVision® instrument. We certify that these results meet our requirements.

Maximum Signal:	782029 counts
Minimum Signal:	1081 counts
EC_{50}:	0.25 nM

Recommended Assay Conditions

Note: This protocol provides a method to verify kit performance and is not representative of an assay. Sufficient biotinylated-probe and 10x buffer is provided to perform 3 titration curves in triplicate as described.

1x Buffer:	Add 500 µL 10x buffer to 4.5 mL Milli-Q® H2O (or equivalent). Add 5 mg BSA (0.1% final concentration) and adjust pH to 7.4.
Acceptor Beads:	Add 5 µL Protein A Acceptor beads to 245 µL 1x buffer.
Donor beads:	Add 5 µL Streptavidin Donor beads to 245 µL 1x buffer.
Biotinylated-probe:	From the 0.5 µM biotin-rIgG, prepare a ½ log dilution series (15 nM to 1.5 pM) in 1x buffer. Include a buffer only control.

Titration Protocol

To a white opaque OptiPlate-384:

1) Add 5 µL biotin-rIgG dilutions (from lowest to highest concentration).

2) Add 10 µL of 1x buffer

3) Add 5 µL of Protein A Acceptor beads.

Incubate in the dark at room temperature for 30 minutes

4) Add 5 µL of Streptavidin Donor beads.

Incubate in the dark at room temperature for 60 minutes and analyze on your Alpha capable detection reader.

Typical Product Data

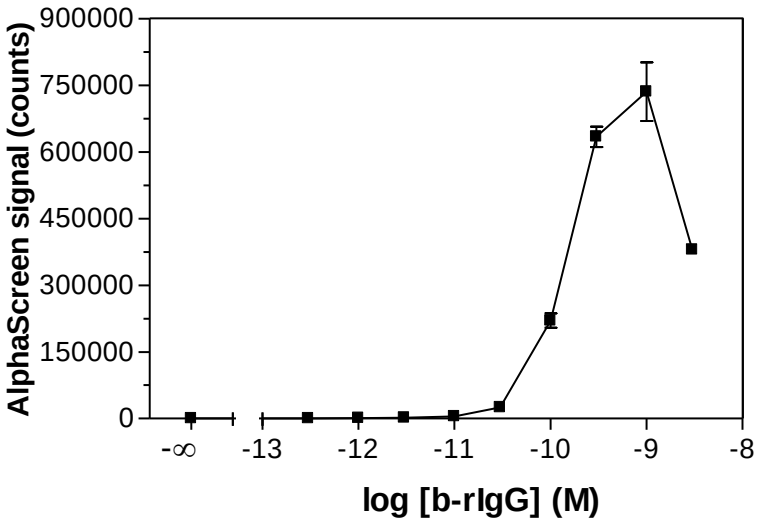

Figure 1: Biotinylated-probe titration assay 384-well biotinylated-rIgG titration curve (25 µL final volume; Reader: Envision). Note: Alpha signal will vary depending on instrument detection protocol, incubation temperature and incubation time.

Please visit our website for additional information on AlphaLISA technology at www.revvity.com

6760617-R Rev 01

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