

Research use only. Not for use in diagnostic procedures.

AlphaScreen®

Histidine (Nickel Chelate) Detection Kit

Product number: 6760619C Lot Number: 3330628

Product Format: 6760619C: 500 assay points

6760619M: 10 000 assay points

6760619R: 50 000 assay points

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

Manufacturing date: 10/03/2024 Document version: 1

Kit Components

Component	6760619C	6760619M	6760619R
Nickel Chelate Acceptor Beads at 5 mg/mL in	1 x 50 μL	1 x 1 mL	1 x 5 mL
25 mM Hepes, 100 mM NaCl, 0.05% Kathon, pH 7.4	(6760140)	(6760141)	(6760141B)
Streptavidin Donor Beads at 5 mg/mL in 25 mM Hepes,	1 x 50 μL	1 x 1 mL	1 x 5 mL
100 mM NaCl, 0.05% Kathon, pH 7.4	(6760001)	(6760002)	(6760002B)
Biotinylated-(His)6 at 0.5 μM in 25mM Hepes,	1 x 50 μL	1 x 50 μL	1 x 50 μL
0.05% Kathon, 0.1% BSA, pH 7.4	(6760302)	(6760302)	(6760302)
10x Buffer : 250 mM Hepes, 1 M NaCl, 0.05%	1 x 1.5 mL	1 x 1.5 mL	1 x 1.5 mL
Kathon, pH 7.4	(6760020G)	(6760020G)	(6760020G)

Product Information

Antibody/Protein: The Nickel Chelate Acceptor beads are coated with chelated nickel which binds (HIS)6 tagged

proteins.

Stability: This kit is stable for at least 12 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.

Alpha maximum signal, minimum signal and EC50 are determined using a biotinylated-(His)6 titration assay performed on an EnVision® instrument. We certify that these results meet our requirements.

Maximum Signal: 747,834 counts
Minimum Signal: 595 counts
EC₅₀: 1.74 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 Nickel chelate Acceptor beads to 495 µL 1x buffer.

Donor beads: Add 5 μL Streptavidin Donor beads to 495 μL 1x buffer.

Biotinylated-probe: From the 0.5 μ M biotinylated-(His)6, prepare a ½ log dilution series (0.5 μ M to 50

pM) in 1x buffer. Include a buffer only control.

Titration Protocol

To a white opaque OptiPlate-384:

- 1) Add 5 μL biotinylated-(His)₆ dilutions (from lowest to highest concentration).
- 2) Add 10 μ L of Nickel chelate Acceptor beads.

Incubate in the dark at room temperature for 30 minutes

3) Add 10 µL of Streptavidin Donor beads.

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

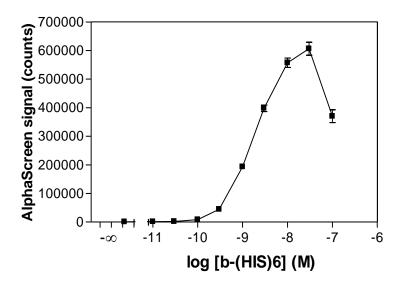


Figure 1: Biotinylated-probe titration assay 384-well biotinylated-(His)6 titration curve (25 μL final volume; Reader: Envision). Note: Alpha signal will vary depending on instrument detection protocol, incubation temperature and incubation time.

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

The information provided in this document is valid for the specified lot number and date of analysis. This information is for reference purposes only and does not constitute a warranty or guarantee of the product's suitability for any specific use. Revvity, Inc., its subsidiaries, and/or affiliates (collectively, "Revvity") do not assume any liability for any errors or damages arising from the use of this document or the product described herein. REVVITY EXPRESSLY DISCLAIMS ALL WARRANTIES, INCLUDING WARRANTIES OF MERCHANTABILITY OR FITNESS FOR A PARTICULAR PURPOSE, REGARDLESS OF WHETHER ORAL OR WRITTEN, EXPRESS OR IMPLIED, ALLEGEDLY ARISING FROM ANY USAGE OF ANY TRADE OR ANY COURSE OF DEALING, IN CONNECTION WITH THE USE OF INFORMATION CONTAINED HEREIN OR THE PRODUCT ITSELF.

6760619-R Rev 01

