

Caution: For Laboratory Use. A research reagent for research purposes only

## Histidine (Nickel Chelate) Detection Kit

**Product No.:** 6760619C/M/R

**Lot No.:** 677-328-A

### Material Provided

<b>Format:</b>	6760619C	500 Assay Points
	6760619M	10000 Assay Points
	6760619R	50000 Assay Points

Note: The number of assay points is based on a final bead concentration of 20 µg/mL in a 25 µL/well reaction volume.

**Manufacturing Date:** July 06, 2011

### Kit Components:

Component	6760619C	6760619M	6760619R
<b>Nickel Chelate Acceptor Beads at 5 mg/mL in 25 mM Hepes, 100 mM NaCl, 0.05% Proclin-300, pH 7.4</b>	1 x 50 µL (6760140)	1 x 1 mL (6760141)	1 x 5 mL (6760141B)
<b>Streptavidin Donor Beads at 5 mg/mL in 25 mM Hepes, 100 mM NaCl, 0.05% Proclin-300, pH 7.4</b>	1 x 50 µL (6760001)	1 x 1 mL (6760002)	1 x 5 mL (6760002B)
<b>Biotinylated-(His)<sub>6</sub> at 0.5 µM in 25mM Hepes, 0.05% Proclin-300, 0.1% BSA, pH 7.4</b>	1 x 50 µL (6760302)	1 x 50 µL (6760302)	1 x 50 µL (6760302)
<b>10x Buffer : 250 mM Hepes, 1 M NaCl, 0.05% Proclin-300, pH 7.4</b>	1 x 1.5 mL (6760020G)	1 x 1.5 mL (6760020G)	1 x 1.5 mL (6760020G)

### Product Information

**Antibody/Protein:** The Nickel Chelate Acceptor beads are coated with chelated nickel which binds (His)<sub>6</sub> tagged proteins.

**Stability:** This product is stable for at least **12 months** from the manufacturing date if used and stored under recommended conditions.

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

**Recommended use:** AlphaScreen® donor beads are **light sensitive** and should be handled under subdued or green filtered light conditions (< 100 Lux). Vortex beads prior to use.

For additional information on running AlphaScreen® assays or on potential interfering compounds, please visit our website: [www.perkinelmer.com/AlphaTech](http://www.perkinelmer.com/AlphaTech)

## Quality Control

AlphaScreen<sup>®</sup> maximum signal, minimum signal and EC<sub>50</sub> are determined using a biotinylated-(His)<sub>6</sub> titration assay performed on an EnVision<sup>®</sup> HTS Alpha detection instrument. We certify that these results meet our requirements.

<u>TEST</u>	<u>RESULTS</u>
Maximum signal	736 330 cps
Minimum signal	338 cps
EC <sub>50</sub>	0.68 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<sup>®</sup> H<sub>2</sub>O (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 µM biotinylated-(His)<sub>6</sub>, prepare a ½ log dilution series (0.5 µM to 50 pM) in 1x buffer. Include a buffer only control.

### Titration Protocol:

To a white opaque 384-well Optiplate:

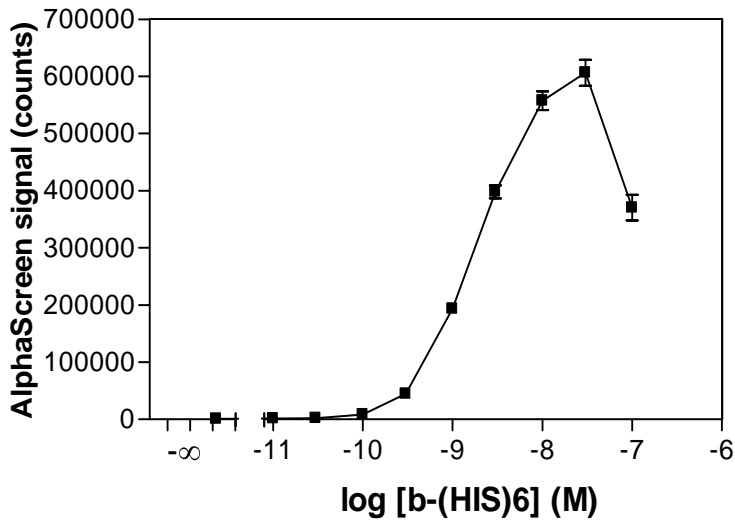
- 1- Add 5 µL biotinylated-(His)<sub>6</sub> dilutions (from lowest to highest concentration).
- 2- Add 10 µL of Nickel chelate Acceptor beads.

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 AlphaScreen<sup>®</sup> detection reader.

## Product Typical Data



**Figure 1: Biotinylated-probe titration assay**  
384-well biotinylated-(His)<sub>6</sub> titration curve (25  $\mu$ L final volume; Reader: Envision HTS Alpha).

**Note:** AlphaScreen<sup>®</sup> signal will vary depending on instrument detection protocol, incubation temperature and incubation time.

## Suggested Materials and Instrumentation

Please visit our website

[www.perkinelmer.com/AlphaTech](http://www.perkinelmer.com/AlphaTech)

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