

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

Protein A Acceptor Beads

Product No.: AL101C (250 µg)
AL101M (5 mg)
AL101R (25 mg)

Lot No.: 677-685-A

Material Provided

Formats:

| Catalog number | Size | Volume | Assay points |
|----------------|--------|--------|--------------|
| AL101C | 250 µg | 50 µL | 500 |
| AL101M | 5 mg | 1 mL | 10 000 |
| AL101R | 25 mg | 5 mL | 50 000 |

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:

July 14, 2011

Description:

AlphaLISA Protein A Acceptor Beads at 5 mg/mL in PBS, pH 7.2 supplemented with 0.05% Proclin-300 as a preservative.

Storage:

Store in the dark at 4°C.

Stability:

This product is stable for at least 12 months from the manufacturing date when stored in its original packaging under recommended storage conditions.

Product Information

Intended use:

This product is intended for use in homogeneous AlphaLISA assays for the capture of several IgG subtypes.

FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES.

Quality Control

Lot-to-lot consistency is confirmed by a Quality Control AlphaScreen[®] titration assay read on an EnVision[®] HTS Alpha instrument (see protocol below). We certify that the results meet our quality release criteria. *Note: maximum counts will vary depending on assay conditions as well as between lots. This variation has no impact on assay quality.*

Maximum signal: 418 256 counts
Minimum signal: 486 counts
EC₅₀: 0.70 nM

Titration Assay (Quality Control Protocol)

This protocol provides a means to verify product performance.

The following reagents and materials are recommended.

| Item | Suggested Source | Catalog # |
|---|-------------------------------|---|
| AlphaScreen® Streptavidin-coated Donor Beads | PerkinElmer LAS Inc. | 6760002S (1 mg) 6760002 (5 mg) 6760002B (50 mg) |
| Biotin-rabbit IgG | Thermo Fisher Scientific Inc. | 31826 |
| AlphaLISA Universal Assay Buffer, 5X | PerkinElmer LAS Inc. | AL001C (10 mL) AL001F (100 mL) |
| White OptiPlate™-384 | PerkinElmer LAS Inc. | 6007290 |
| TopSeal™-A Adhesive Sealing Film | PerkinElmer LAS Inc. | 6005185 |
| EnVision® Multilabel Reader with the Alpha Option | PerkinElmer LAS Inc. | - |

Recommendations:

- AlphaScreen® Donor beads are light-sensitive. All AlphaLISA assays using the Streptavidin-coated Donor Beads should be performed under subdued laboratory lighting of less than 100 lux. Alternatively, green filters (Roscolux filters #389 from Rosco, or the equivalent) can be applied to light fixtures. Incubation with AlphaScreen® Donor beads should always be performed in the dark. For example, assay reactions in a microplate can be covered with an opaque microplate.
- Sodium azide should not be added to stock solutions or assay components. Final concentrations of sodium azide higher than 0.001 % will decrease the AlphaLISA signal.
- Spin down tubes briefly before use to improve recovery of content (2,000 x g, 10-15 sec). Resuspend all reagents by vortexing before use.
- Use Milli-Q® grade water (18 MΩ•cm) to dilute the 5X AlphaLISA Universal Buffer.
- 1X AlphaLISA Universal Assay Buffer contains PBS, pH 7.5, 0.1% BSA, 0.01% Proclin-300. 1X AlphaLISA Universal Assay Buffer is used in the titration assay described below (Quality Control Protocol). Optimization of this assay buffer might be necessary in other assay types.
- Small volumes may be prone to evaporation. It is recommended to cover microplates with TopSeal-A Adhesive Sealing Film to reduce evaporation during incubation. Microplates are read with the TopSeal-A Film on the plate.
- Total signal varies with temperature and incubation time. For consistent results, identical incubation times and temperature should be used for all plates.
- The AlphaLISA signal is detected with an EnVision Multilabel Reader equipped with the ALPHA option using the AlphaScreen standard settings (e.g. Total Measurement Time: 550 ms, Excitation Time: 180 ms, Mirror: D640as, Emission Filter: M570w, Center Wavelength 570 nm, Bandwidth 100 nm, Transmittance 75%).

Protocol:

This protocol is recommended for generating one titration curve in a 25 μL final assay volume (12 concentrations, triplicate determinations with manual pipetting). If more assay points are needed, volumes should be adjusted accordingly.

1) Preparation of 1X AlphaLISA Universal Assay Buffer:

Add 1 mL of 5X AlphaLISA Universal Assay Buffer to 4 mL H_2O .

2) Preparation of 1.7X Biotin-rabbit IgG dilutions:

Dilute Biotin-rabbit IgG to a 50 nM stock solution.

Prepare 1.7X dilutions in 1X AlphaLISA Universal Assay Buffer as follows:

| Tube | Volume of Biotin-rabbit IgG | Volume of buffer (μL) | [Biotin-rabbit IgG] (M) in 15 μL (1.7X) | [Biotin-rabbit IgG] (M) in final assay volume (25 μL) |
|------|-----------------------------|------------------------------------|--|---|
| A | 51 μL of 50 nM | 99 | 1.7E-8 | 1.0E-8 |
| B | 60 μL of tube A | 140 | 5.1E-9 | 3.0E-9 |
| C | 60 μL of tube B | 120 | 1.7E-9 | 1.0E-9 |
| D | 60 μL of tube C | 140 | 5.1E-10 | 3.0E-10 |
| E | 60 μL of tube D | 120 | 1.7E-10 | 1.0E-10 |
| F | 60 μL of tube E | 140 | 5.1E-11 | 3.0E-11 |
| G | 60 μL of tube F | 120 | 1.7E-11 | 1.0E-11 |
| H | 60 μL of tube G | 140 | 5.1E-12 | 3.0E-12 |
| I | 60 μL of tube H | 120 | 1.7E-12 | 1.0E-12 |
| J | 60 μL of tube I | 140 | 5.1E-13 | 3.0E-13 |
| K | 60 μL of tube J | 120 | 1.7E-13 | 1.0E-13 |
| L | 0 | 140 | 0 | 0 |

3) Preparation of 5X AlphaLISA Acceptor beads (100 $\mu\text{g}/\text{mL}$):

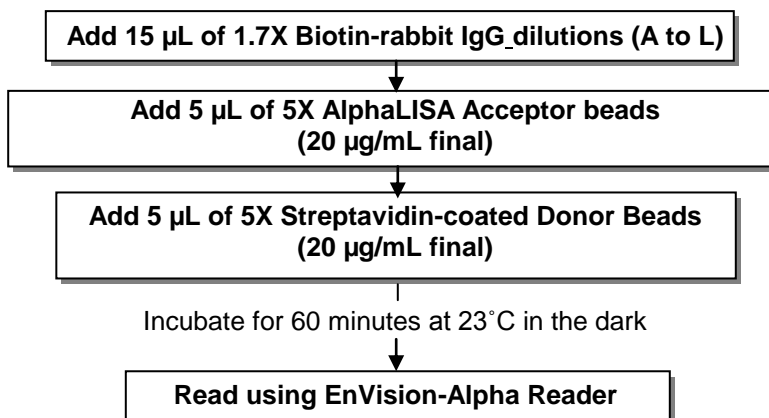
Add 5 μL of 5 mg/mL AlphaLISA beads to 245 μL of 1X AlphaLISA Universal Assay Buffer.

4) Preparation of 5X Streptavidin-coated Donor Beads (100 $\mu\text{g}/\text{mL}$):

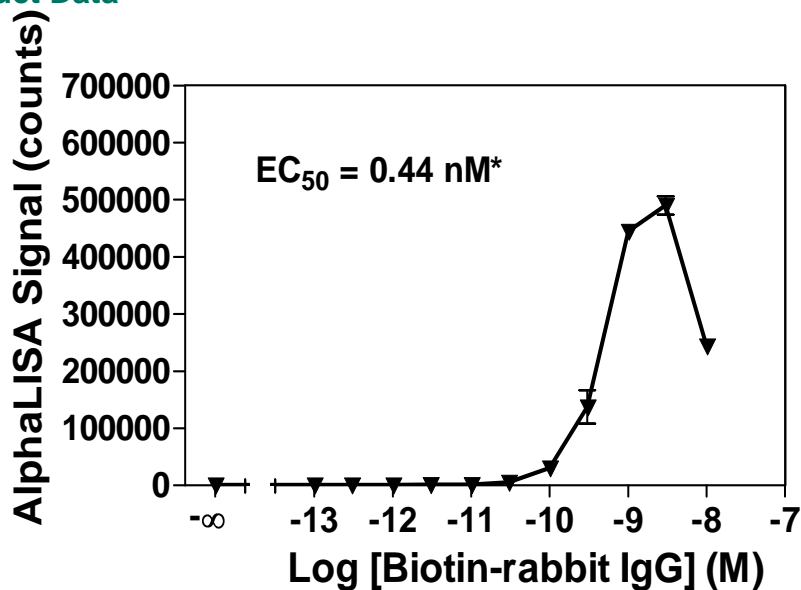
Keep the beads under subdued laboratory lighting.

Add 5 μL of 5 mg/mL Streptavidin-coated Donor Beads to 245 μL of 1X AlphaLISA Universal Assay Buffer.

5) In an OptiPlate-384 microplate:



Typical Product Data



* The EC_{50} value was determined following a non-linear regression analysis using the sigmoidal dose-response curve model with variable slope. Only assay points up to the maximum signal were used for EC_{50} determination (in this case, up to 3 nM).

Suggested Materials and Instrumentation

Please visit our website

www.perkinelmer.com/AlphaTech

This product is not for resale or distribution except by authorized distributors.

LIMITED WARRANTY: PerkinElmer BioSignal Inc. warrants that, at the time of shipment, the products sold by it are free from defects in material and workmanship and conform to specifications which accompany the product. PerkinElmer BioSignal Inc. makes no other warranty, express or implied with respect to the products, including any warranty of merchantability or fitness for any particular purpose. Notification of any breach of warranty must be made within 60 days of receipt unless otherwise provided in writing by PerkinElmer BioSignal Inc. No claim shall be honored if the customer fails to notify PerkinElmer BioSignal Inc. within the period specified. The sole and exclusive remedy of the customer for any liability of PerkinElmer BioSignal Inc. of any kind including liability based upon warranty (express or implied whether contained herein or elsewhere), strict liability contract or otherwise is limited to the replacement of the goods or the refunds of the invoice price of goods. PerkinElmer BioSignal Inc. shall not in any case be liable for special, incidental or consequential damages of any kind.

PerkinElmer, Inc.
940 Winter Street
Waltham, MA 02451 USA
P: (800) 762-4000 or
(+1) 203-925-4602
www.perkinelmer.com



For a complete listing of our global offices, visit www.perkinelmer.com/ContactUs

Copyright ©2009, PerkinElmer, Inc. All rights reserved. PerkinElmer® is a registered trademark of PerkinElmer, Inc. All other trademarks are the property of their respective owners.