TECHNICAL DATA SHEET

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

# Anti-Goat IgG Acceptor Beads

Product No.: AL107C (250 μg) AL107M (5 mg) AL107R (25 mg)

Lot No.: 663-964-A

### **Material Provided**

Formats:

| Catalog<br>number | Size   | Volume | Assay<br>points |
|-------------------|--------|--------|-----------------|
| AL107C            | 250 µg | 50 µL  | 500             |
| AL107M            | 5 mg   | 1 mL   | 10 000          |
| AL107R            | 25 mg  | 5 mL   | 50 000          |

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

Manufacturing Date: April 15, 2011

**Description:** AlphaLISA Anti-Goat IgG 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 goat IgG. The anti-goat antibody coupled to the beads targets the Fc region of goat IgG.

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

## **Quality Control**

Lot-to-lot consistency is confirmed by a Quality Control AlphaScreen<sup>®</sup> titration assay read on an EnVision<sup>®</sup> 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 Minimum signal: EC<sub>50</sub>: 25497 counts 194 counts 0.43 nM



TDS-AL107-02 Page 1 of 4

# **Titration Assay (Quality Control Protocol)**

This protocol provides a means to verify product performance.

The following reagents and materials are recommended.

| ltem  | Suggested Source                             | Catalog #   |  |
|---|--|---|--|
| AlphaScreen <sup>®</sup> Streptavidin-<br>coated Donor Beads  | PerkinElmer LAS, Inc.                        | 6760002S (1 mg)<br>6760002 (5 mg)<br>6760002B (50 mg) |  |
| Biotin-goat IgG   | Jackson ImmunoResearch<br>Laboratories, Inc. | 005-060-003   |  |
| AlphaLISA Universal Assay<br>Buffer, 5X                       | PerkinElmer LAS, Inc.                        | AL001C (10 mL)<br>AL001F (100 mL)                     |  |
| White OptiPlate™-384  | PerkinElmer LAS, Inc.                        | 6007290   |  |
| TopSeal™-A Adhesive<br>Sealing Film                           | PerkinElmer LAS, Inc.                        | 6005185   |  |
| EnVision <sup>®</sup> Multilabel Reader with the Alpha Option | PerkinElmer LAS, Inc.                        | -   |  |

#### **Recommendations:**

- AlphaScreen<sup>®</sup> 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<sup>®</sup> 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<sup>®</sup> 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<sub>2</sub>O.

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

Dilute Biotin-mouse IgG to a 50 nM stock solution. Prepare 1.7X dilutions in 1X AlphaLISA Universal Assay Buffer as follows:

| Tube | Volume of<br>Biotin-goat IgG | Volume of<br>buffer (µL) | [Biotin-goat IgG] (M)<br>in 15 μL (1.7X) | [Biotin-goat IgG] (M)<br>in final assay volume (25 μL) |
|------|------------------------------|--------------------------|--|--|
| Α    | 51 µL of 50 nM               | 99                       | 1.7E-8                                   | 1.0E-8   |
| В    | 60 µl of tube A              | 140                      | 5.1E-9                                   | 3.0E-9   |
| С    | 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  |
| Н    | 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 l              | 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  |

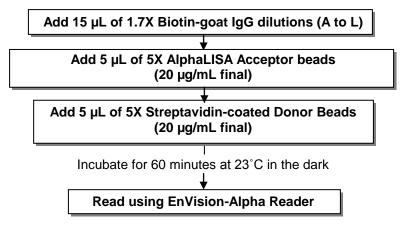
3) Preparation of 5X AlphaLISA Acceptor beads (100 µg/mL):

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

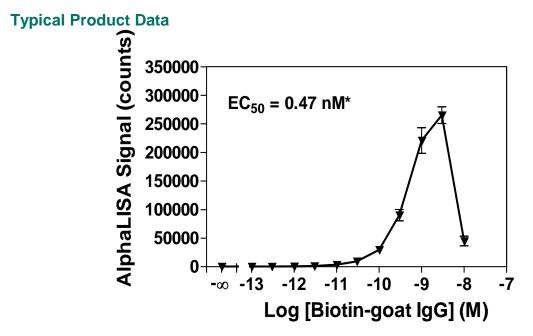
4) Preparation of 5X Streptavidin-coated Donor Beads (100 µg/mL):

Keep the beads under subdued laboratory lighting. Add 5  $\mu$ L of 5 mg/mL Streptavidin-coated Donor Beads to 245  $\mu$ L of 1X AlphaLISA Universal Assay Buffer.

5) In an OptiPlate-384 microplate:







\* The EC<sub>50</sub> 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<sub>50</sub> determination (in this case, up to 3 nM).

### **Suggested Materials and Instrumentation**

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www.perkinelmer.com/AlphaTech

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