

AlphaLISA®

Anti-c-Myc Acceptor Beads**Product number:** AL111C **Lot Number:** 3219145**Material provided:** AlphaLISA Anti-c-Myc Acceptor Beads at 5 mg/mL in PBS, pH 7.2 supplemented with 0.05% Kathon as a preservative.**Product Format:** AL111C: 250 µg, 50 µL, 500 assay points

AL111M: 5 mg, 1 mL, 10 000 assay points

AL111R: 25 mg, 5 mL, 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: 10/25/2023 **Document version:** 1**Product Information****Application:** This product is intended for use in homogeneous AlphaLISA assays for the capture of c-Myc-tagged targets.**Storage:** Store product in the dark at 4 °C.**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.**Quality Control**

Lot to lot consistency is confirmed in an Alpha assay. Maximum and minimum signals and EC50 were measured on the EnVision Multilabel Plate Reader with Alpha option. We certify that these results meet our quality release criteria. Maximum counts may vary between bead lots and the instrument used, with no impact on assay quality

EC₅₀: 30.1 nM
Min counts: 531 counts
Max counts: 59900 counts

Titration Assay (Quality Control Procedure)

This protocol provides a means to verify product performance. The following reagents and materials are recommended.

| Item | Suggested source |
|---|------------------|
| White OptiPlate™-384 | Revvity Inc. |
| TopSeal™-A Plus Adhesive Sealing Film | Revvity Inc. |
| EnVision®-Alpha Reader | Revvity Inc. |
| AlphaScreen® Streptavidin-coated Donor Beads | Revvity Inc. |
| Biotin-ChromaLink-AEQKLISEEDL amide (Biotin-c-Myc) | Custom |
| AlphaLISA Universal Assay Buffer 5X | Revvity Inc. |

Recommendations

- 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.
- 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% Kathon. 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%).

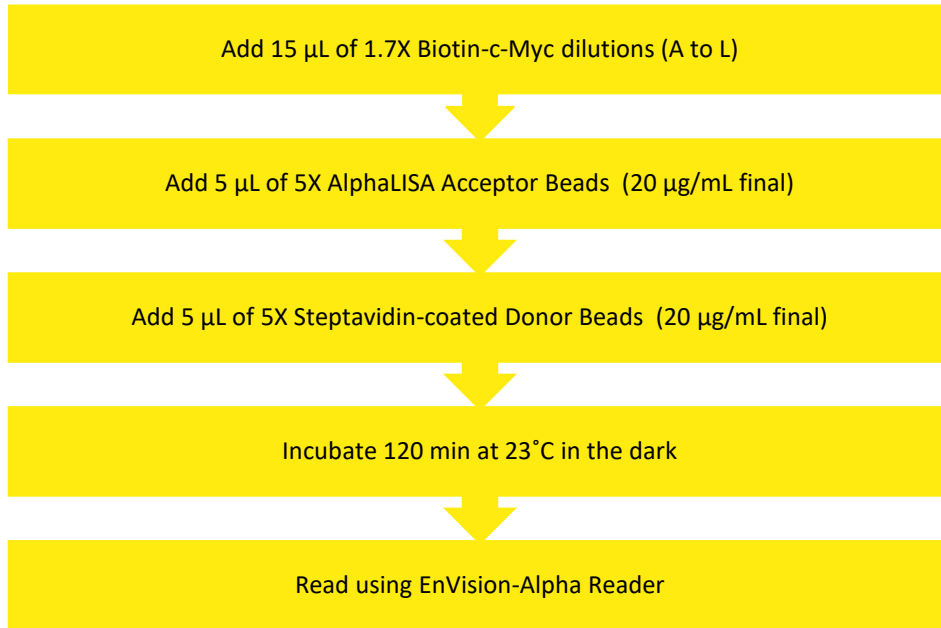
Protocol

- 1) Preparation of 1X AlphaLISA Universal Buffer:
Add 1 mL of 5X AlphaLISA Universal Buffer to 4 mL H₂O.
- 2) Preparation 1.7X Biotin-c-Myc dilutions:
Prepare 1.7X dilutions in 1X AlphaLISA Universal Assay Buffer as follows:

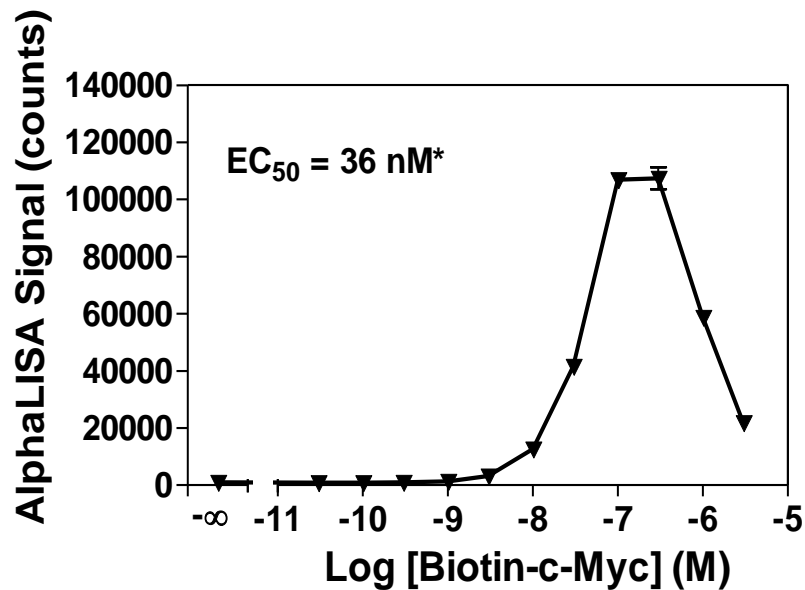
| Tube | Volume of Biotin-c-Myc | Volume of buffer (μL) | [Biotin-c-Myc] (M) in 15 μL (1.7X) | [Biotin-c0Myc] (M) in final assay volume (25 μL) |
|------|------------------------|-----------------------|------------------------------------|--|
| A | 8 μL of 100 μM | 149 | 5.1E-6 | 3.0E-6 |
| B | 60 μL of tube A | 120 | 1.7E-6 | 1.0E-6 |
| C | 60 μL of tube B | 140 | 5.1E-7 | 3.0E-7 |
| D | 60 μL of tube C | 120 | 1.7E-7 | 1.0E-7 |
| E | 60 μL of tube D | 140 | 5.1E-8 | 3.0E-8 |
| F | 60 μL of tube E | 120 | 1.7E-8 | 1.0E-8 |
| G | 60 μL of tube F | 140 | 5.1E-9 | 3.0E-9 |
| H | 60 μL of tube G | 120 | 1.7E-9 | 1.0E-9 |
| I | 60 μL of tube H | 140 | 5.1E-10 | 3.0E-10 |
| J | 60 μL of tube I | 120 | 1.7E-10 | 1.0E-10 |
| K | 60 μL of tube J | 140 | 5.1E-11 | 3.0E-11 |
| L | 0 | 120 | 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 μL of 5 mg/mL Streptavidin-coated Donor Beads to 245 μL of 1X AlphaLISA Universal Assay Buffer.

5) In a OptiPlate-384 microplate:



Typical Product Data

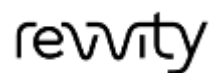


* The EC_{50} value was determined following a non-linear regression analysis using the sigmoidal dose-response curve model with variable slope. Only assay points up to the maximum signal were used for EC_{50} determination (in this case, up to 100 nM).

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

AL111-R Rev01



Revvity, Inc.
940 Winter Street
Waltham, MA 02451 USA

(800) 762-4000 www.revivity.com

For a complete listing of our global offices, visit www.revivity.com
Copyright ©2023, Revvity, Inc. All rights reserved.