

LANCE® Ultra

**ULight™ -labeled Streptavidin****Product number:** TRF0102-M **Lot Number:** 3242476**Product Format:**  
TRF0102-D: 1 nmole (100µL)  
TRF0102-M: 10 nmoles (1mL)  
TRF0102-R: 100 nmoles (10mL)**Manufacturing date:** 1/4/2024 **Document version:** 1**Product Information****Storage Buffer:** 50 mM Tris-HCl (pH 7.4), 0.9% NaCl, 0.1% BSA and 0.05% sodium azide as preservative  
**Molecular Weight:** 60 000  
**Stability:** This product is stable for at least **24 months** from the manufacturing date when stored in its original packaging and the recommended storage conditions.  
**Storage Conditions:** Store at 4°C. Store protected from light.**Quality Control**

The QC release specifications are based on spectrophotometric analysis of the labeled antibody. We certify that results meet our quality release criteria.

**Labeling Ratio:** 2.8 (dye molecule/peptide)  
**Concentration:** 600 µg/mL (10 µM)**Recommended Assay Conditions**

Src kinase assay: ATP titration

## SUGGESTED METHOD:

(Specific applications might require optimization)

### Reagent Preparation

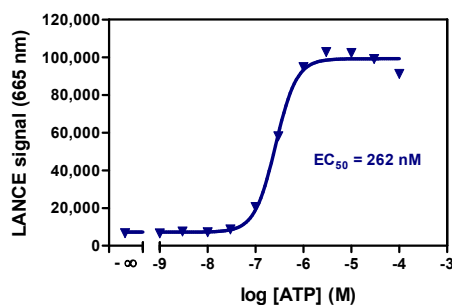
- Prepare 1X Kinase Buffer: 50 mM Tris-HCl pH 7.5, 1 mM EGTA, 10 mM MgCl<sub>2</sub>, 2 mM DTT and 0.01% Tween-20.
- Prepare a 2X c-Src enzyme solution: dilute the enzyme to a concentration of 2 nM in Kinase Buffer. Keep on ice.
- Prepare a 4X mix of biotin-poly GT: dilute biotin-poly GT to a concentration of 400 nM in Kinase Buffer. Keep on ice.
- Prepare a 4X mix of ATP: dilute ATP to concentrations ranging from 40 nM to 4 mM (serial half-log dilutions) in Kinase Buffer. Keep on ice.
- Prepare 1X Detection Buffer: dilute 0.5 mL of 10X detection buffer with 4.5 mL of H<sub>2</sub>O.
- Prepare a 4X Stop Solution: prepare a 40 mM EDTA solution in 1X Detection Buffer.
- Prepare a 4X Detection Mix: dilute the Eu-W1024-labeled PY20 antibody to 8 nM and *ULight*-Streptavidin to 200 nM in 1X Detection Buffer.

### Protocol

- Pipet 5 µL of 2X c-Src enzyme into a 384-well white OptiPlate™-384 (1 nM final concentration).
- Add 2.5 µL of 4X biotin-poly-GT (100 nM final concentration).
- Add 2.5 µL of 4X ATP mix (10 nM to 1 mM final concentrations).
- Cover plate with TopSeal-A and incubate 90 min at 23°C.
- Add 5 µL of 4X Stop Solution and incubate 5 min at 23°C.
- Add 5 µL of 4X Detection Mix (2 nM Eu-W1024-labeled PY20 antibody and 50 nM *ULight*-Streptavidin final concentrations).
- Cover plate with TopSeal-A and incubate 60 min at 23°C.
- Remove TopSeal-A and read in TR-FRET mode at 665 nm (excitation at 320 or 340 nm).

## Typical Product Data

Src kinase assay using *ULight*-Streptavidin, biotin poly GT and Eu-PY20 anti phosphotyrosine obtained using the EnVision® Multilabel Reader:



### Suggested Materials

- Detection: *ULight*<sup>™</sup> - Streptavidin
- Substrate: biotin-poly GT (4:1)
- Antibody: Eu-W1024 anti-phosphotyrosine (PY20)
- Kinase: c-Src

### Supplier

Revvity Inc  
Revvity Inc  
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UpState

- Detection Buffer: LANCE<sup>®</sup> Detection Buffer, 10X
- Plate: OptiPlate™-384, white
- TopSeal™-A

Revvity Inc  
Revvity Inc  
Revvity Inc

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TRF0102-R Rev01

The logo for Revvity, featuring the word "revvity" in a lowercase, sans-serif font.

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