

Research use only. Not for use in diagnostic procedures.

## LANCE<sup>®</sup> Ultra

# Eu-W1024 labeled Anti-phospho-CREB (Ser133) Antibody

Product number:	TRF0200-C	Lot	Number:	3270467	
Product Format:	TRF0200-C: 1.7 μg				$\overline{}$
Manufacturing date:	March 14, 2023	Document version:	1		
Product Information					
Antibody:	Europium-labeled rabbit monoclonal antibody recognizing phospho-Ser133 of the cAMP responsive element binding (CREB) protein.				
Storage Buffer:	50 mM Tris-HCl (pH 7.4), 0.9% NaCl, 0.1% BSA and 0.05% sodium azide as preservative.				
Molecular Weight:	160 000				
Stability:	This product is stable for at least 24 months from the manufacturing date when stored in its original packaging and at the recommended storage conditions.				
Storage Conditions:	Store at 4°C				

## **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:	5.99
Concentration:	100 μg/ml (0.625 μM)

## **Recommended Assay Conditions**

#### **PROTEIN KINASE A: ATP TITRATION**

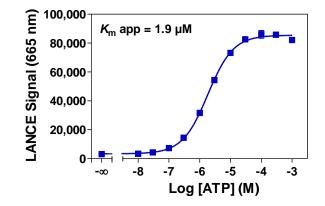
SUGGESTED METHOD: (Specific applications might require optimization)

#### **Reagent Preparation**

- Prepare 1X Kinase Assay Buffer: 50 mM Hepes pH 7.5, 1 mM EGTA, 10 mM MgCl<sub>2</sub>, 2 mM DTT and 0.01% Tween-20.
- Prepare a 2X PKA solution: dilute the enzyme to a concentration of 5 pM in Kinase Assay Buffer. Keep on ice.
- Prepare a 4X mix of ULight<sup>™</sup>-CREBtide: dilute ULight<sup>™</sup>-CREBtide to a concentration of 200 nM in Kinase Assay Buffer.
- Prepare a 4X mix of ATP: dilute ATP to concentrations ranging from 40 nM to 4 mM (serial half-log dilutions) in Kinase Assay 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 24 mM EDTA solution in 1X Detection Buffer.
- Prepare a 4X Detection Mix: dilute the Eu-anti-phospho-CREB antibody to a concentration of 8 nM in 1X Detection Buffer.

- Pipet 5 μL of 2X PKA solution into a 384-well white OptiPlate-384 (2.5 pM final concentration).
- Add 2.5 μL of 4X ULight- CREBtide (50 nM final concentration).
- Add 2.5 μL of 4X ATP solution (10 nM to 1 mM final concentrations).
- Cover plate with TopSeal-A and incubate 45 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-anti-phospho-CREB antibody final conc.) and mix.
- 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**



Supplier

**Revvity Inc** 

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Millipore

#### **Suggested Materials**

- Substrate: ULight<sup>™</sup>- CREBtide) Peptide
- Antibody: Eu- phospho-CREB (Ser133) Antibody
- Kinase: PKA, catalytic subunit, recombinant
- Detection Buffer: LANCE<sup>®</sup> Detection Buffer, 10X
- Plate: OptiPlate<sup>™</sup>-384, white
- TopSeal<sup>™</sup>-A
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TRF0200-C-R Rev01



**Revvity, Inc.** 940 Winter Street Waltham, MA 02451 USA (800) 762-4000 www.revvity.com

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