

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

## *ULight*<sup>™</sup>-labeled MBP peptide

**Product No.:** TRF0109-M

**Lot No.:** 2927726

### Material Provided

**Format:** TRF0109-D 0.5 nmole (1 000 assay points\*)  
TRF0109-M 5 nmoles (10 000 assay points\*)  
\*Assuming 0.5 pmol/ assay point

**Volume:** 100 µL (TRF0109-D) or 1 mL (TFR0109-M)

**Manufacturing Date** November 15, 2021

### Product Information

**Phosphorylation Motif:** VTPRTPPPP  
Human Myelin Basic Protein phosphorylation site: Thr232 (Swiss-Prot: P02686); corresponds to Thr98 in other isoforms or species.

**Molecular Weight:** 2 808

**Storage Buffer:** 50 mM Tris-HCl (pH 7.4), 0.9% NaCl, 0.1% BSA and 0.05% sodium azide as preservative

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

**Storage Conditions:** Store at -20°C. Store protected from light. Repeated freezing and thawing should be avoided.

**Safety Note:** The storage buffer contains sodium azide (NaN<sub>3</sub>) as a preservative. Disposal of all waste should be in accordance with local regulations.

### Quality Control

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

**Labeling Ratio:** 1.0 (dye molecule/peptide)

**Concentration:** 14.05 µg/mL (5 µM)

## Recommended Assay Conditions

### ERK1 kinase: ATP titration

#### Reagent Preparation:

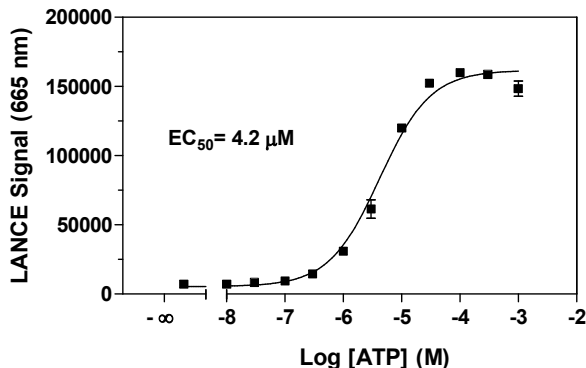
- Prepare 1X Kinase Assay Buffer: 50 mM Tris pH 7.5, 1 mM EGTA, 10 mM MgCl<sub>2</sub>, 2 mM DTT and 0.01% Tween-20.
- Prepare a 2X Erk1 solution: dilute the enzyme to a concentration of 2 nM in Kinase Assay Buffer. Keep on ice.
- Prepare a 4X mix of *ULight*-MBP: dilute *ULight*-MBP to a concentration 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 40 mM EDTA solution in 1X Detection Buffer.
- Prepare a 4X Detection Mix: dilute the Eu-anti-phospho-MBP antibody to a concentration of 8 nM in 1X Detection Buffer.

#### Protocol:

- Pipet 5  $\mu$ L of 2X Erk1 solution into a 384-well white OptiPlate™-384 (1 nM final concentration).
- Add 2.5  $\mu$ L of 4X *ULight*-MBP (50 nM final concentration).
- Add 2.5  $\mu$ L 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  $\mu$ L of 4X Stop Solution and incubate 5 min at 23°C.
- Add 5  $\mu$ L of Detection Mix (2 nM Eu-anti-phospho-MBP antibody final concentration) 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 ATP Titration Data

Erk1 kinase assay using *ULight*-MBP and Eu-anti-phospho-MBP antibody obtained using the EnVision® Multilabel Reader:



## Suggested Materials

	Supplier	Cat. No.
• Substrate: <i>ULight</i> <sup>™</sup> - MBP peptide	PerkinElmer	TRF0109
• Antibody: Eu-anti- phospho-Myelin Basic Protein (MBP)	PerkinElmer	TRF0201
• Kinase: MAP Kinase 1/Erk1, active	Upstate	14-439
• Detection Buffer: LANCE <sup>®</sup> Detection Buffer, 10X	PerkinElmer	CR97-100
• Plate: OptiPlate <sup>™</sup> -384, white	PerkinElmer	6007299
• TopSeal <sup>™</sup> : TopSeal-A	PerkinElmer	6050195

Please visit our website for additional resource:

[www.perkinelmer.com/LANCE](http://www.perkinelmer.com/LANCE)

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

*Ulight*<sup>™</sup> products may be covered by the following patents: US 7,250,517 and EP07102880.

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