

## Technical Data Sheet

## Phospho ERK1/2 (T202/Y204) Standard

## Product Information

Material Number:	560027
Component:	51-9004815
Description:	Phospho ERK1/2 (T202/Y204) Standard
Size:	1 vial (1 ea)
Storage Buffer:	Lyophilized in an aqueous buffered solution containing BSA and ProClin™ 150.

## Description

This product contains lyophilized phosphorylated recombinant extracellular signal regulated kinases 1/2 (ERK1/2). When reconstituted, this product can be used as a standard control with the BD™ CBA Phospho ERK1/2 (T202/Y204) Flex Set (Cat. No. 560012). Please refer to the technical data sheet included with the BD™ CBA Phospho ERK1/2 (T202/Y204) Flex Set for further information.

## Preparation and Storage

Store the lyophilized standard at 4°C prior to reconstitution.

Lyosphere(s) from 1 vial of Phospho ERK1/2 (T202/Y204) Standard, when reconstituted in 0.1 mL Assay Diluent, approximates the CBA activity of Phospho ERK1/2 (T202/Y204) at a concentration of 50,000 Units/mL. Following reconstitution, the Phospho ERK1/2 (T202/Y204) Standard may be stored at 4°C for up to 3 months. Any unused, reconstituted standard must be discarded after this period. When using reconstituted standard, warm to 37°C and vortex to mix thoroughly before use.

## Application Notes

## Application

Flow cytometry	Routinely Tested
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## Recommended Assay Procedure:

The Phospho ERK1/2 (T202/Y204) Standard has been tested to assure to function as a standard when used with the BD™ Cytometric Bead Array (CBA), such as the BD™ CBA Phospho ERK1/2 (T202/Y204) Flex Set (Cat. No. 560012). Investigators are encouraged to refer to the technical data sheet for the BD™ CBA Phospho ERK1/2 (T202/Y204) Flex Set (Cat. No. 560012) as well as the product manual for the BD™ CBA Cell Signaling Master Buffer Kit (Cat. No. 560005 or 560006). An abbreviated instruction has been provided below (Preparation of Phospho ERK1/2 (T202/Y204) Standard Protocol). The Phospho ERK1/2 (T202/Y204) Standard should be reconstituted using Assay Diluent, which is provided with the BD™ CBA Cell Signaling Master Buffer Kit (Cat. No. 560005 or 560006) or it may also be purchased separately (Cat. No. 560104).

**Preparation of Phospho ERK1/2 (T202/Y204) Standard:** The Phospho ERK1/2 (T202/Y204) Standard is lyophilized and should be reconstituted and serially diluted before mixing with CBA capture beads and PE detection reagents.

1. Open one vial of the lyophilized Phospho ERK1/2 (T202/Y204) Standard. Transfer the lyosphere(s) to a 1.5 mL microfuge tube.
2. Reconstitute the standard with 0.1 mL of Assay Diluent, warming the tube to 37°C, and vortexing. This creates a 50x stock solution.
3. Label 12 x 75 mm tubes (BD Falcon™, Cat. No. 352008) and arrange them in the following order: Top Standard, 1:2, 1:4, 1:8, 1:16, 1:32, 1:64, 1:128, and 1:256.
4. Add 20 µl of the 50x Standard to one tube. Label the tube "Top Standard".
5. Add Assay Diluent to Top Standard to achieve a final volume of 1 mL and a 1x concentration.
6. Add 500 µl of Assay Diluent to each of the remaining dilution tubes.
7. Perform a serial dilution by transferring 500 µl from the Top Standard to the 1:2 dilution tube and mix thoroughly. Continue making serial dilutions by transferring 500 µl from the 1:2 tube to the 1:4 tube and so on to the 1:256 tube and mix thoroughly. Prepare one tube containing only Assay Diluent to serve as the 0 Units/mL negative control.

## Suggested Companion Products

Catalog Number	Name	Size	Clone
560012	Phospho ERK1/2 (T202/Y204) Flex Set	100 tests	(none)
560005	Cell Signaling Master Buffer Kit	100 tests	(none)
560006	Cell Signaling Master Buffer Kit	500 tests	(none)
560104	Assay Diluent	30 ml	(none)
352008	BD Falcon™ Round Bottom Tubes, 6 ML		(none)

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## Product Notices

1. ProClin is a trademark of Rohm and Haas Company.
2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
3. Please refer to [www.bdbiosciences.com/pharmingen/protocols](http://www.bdbiosciences.com/pharmingen/protocols) for technical protocols.

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