

Instrument Control Terbium TR-FRET

Catalog Number: A14138

Literature Part Number: A14138PIS (MAN0005193)

Literature Lot Number: V01.00

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Kit components	Part no.	Amount	Storage	Handling
HIGH Instrument Control	A14180	1 mL	4°C	Protect from light
LOW Instrument Control	A14181	1 mL	4°C	Protect from light

Additional materials required, but not provided	Recommended source	Part no.
White opaque 96-well assay plate, or	Corning	3917
White opaque 384-well assay plate	Corning	3570
Fluorescence plate reader with top-read and TR-FRET capability	See www.invitrogen.com/instrumentsetup for details	

Overview

This kit allows for the rapid assessment of proper instrument setup prior to the performance of a terbium (Tb)-based TR-FRET assay. Tb-based LanthaScreen® technology requires specific instrument settings that are critical to experimental success. We do not recommend using monochromator-based instruments as the sensitivity of these instruments is generally not sufficient to adequately detect the TR-FRET signal. For set-up information on instruments we have tested, refer to www.invitrogen.com/instrumentsetup.

Step 1: Aliquot the Controls onto Assay Plate

We recommend plating a minimum of 3 replicates of each control.

1. Add 60 µL/well of the HIGH control to empty assay plate wells for 96-well format (or 20 µL/well for 384-well format).
2. Add 60 µL/well of the LOW control to empty assay plate wells for 96-well format (or 20 µL/well for 384-well format).

Step 2: Read the Assay Plate

All measurements should be taken at room temperature from the top of the wells, with the plate lid removed.

1. Set the fluorescence plate reader to top-read and time-resolved fluorescence mode (allow the lamp in the plate reader to warm up for at least 10 minutes before taking measurements).
2. Remove the lid and read the plate using the LanthaScreen® Tb TR-FRET instrument-specific filter selection guidelines provided at www.invitrogen.com/instrumentsetup. Note that the filter bandwidths are critical and cannot be approximated.

Step 3: Analyze the Data

1. For each well, calculate the TR-FRET Emission Ratio (e.g., 520 nm/495 nm) by dividing the acceptor emission value (i.e., 520 nm) by the donor emission value (i.e., 495 nm).
2. Average the Emission Ratios for the HIGH control, and separately average the Emission Ratios for the LOW control.
3. Determine the HIGH to LOW fold-change by dividing the average Emission Ratio for the HIGH control by the average Emission Ratio for the LOW control.

Note: The HIGH/LOW fold-change should be 2–4, depending on the plate reader used. Values below 2 may indicate that the instrument is not setup properly and/or lacks enough sensitivity for Tb-based TR-FRET.

Technical Support

For assistance, contact our technical support team at drugdiscoverytech@lifetech.com or 760-603-7200, extension 40266.

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