

# KeyTec® TR-FRET

## mAb anti-FLAG-Solar Eu



CAT. & Size    A1020009S (1,000 tests)  
                    A1020009L (10,000 tests)  
Storage at     -60°C or below

VKEYBIO-01-2024  
For Research Use Only  
Not For Diagnostic Or Therapeutic Use

## KeyTec® TR-FRET

### mAb anti-FLAG-Solar Eu

#### Instruction Manual

#### 1. Introduction

KeyTec® TR-FRET mAb anti-FLAG-Solar Eu is designed for developing the TR-FRET Assay. The anti-FLAG antibody is a mouse monoclonal antibody. In the Protein-Protein Interaction assay, one FLAG-tagged protein binds to the donor (KeyTec® TR-FRET mAb anti-FLAG-Solar Eu<sup>\*1</sup>), and the other protein is labeled (directly or indirectly) with the acceptor (KeyTec® TR-FRET LA/HX<sup>\*2</sup>). When the two proteins interact, the donor molecule is brought into proximity with the acceptor molecule. Excitation of the donor will result in the generation of the TR-FRET signal at 665 nm, proportional to the extent of protein interaction.

\*1 KeyTec® TR-FRET Solar Eu: TR-FRET Donor Molecule

\*2 KeyTec® TR-FRET LA/HX: TR-FRET Acceptor Molecule

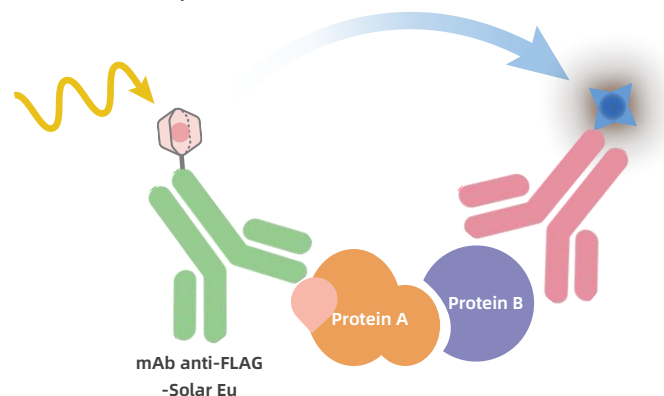


Figure 1. KeyTec® TR-FRET Protein-Protein Interaction assay model

## 2. Components

Components	A1020009S (1,000 tests)	A1020009L (10,000 tests)
KeyTec® TR-FRET	1 vial	1 vial
mAb anti-FLAG-Solar Eu (100X)	50 µL/vial	500 µL/vial

KeyTec® Materials Required But Not Supplied	CAT. & Size
KeyTec® TR-FRET Binding Assay Diluent Buffer	A1010001L (200 mL)
KeyTec® TR-FRET Solar Eu Detection Buffer	A1010002L (120 mL)
KeyTec® 384-Well White Flat Low-Volume Microplates, PS, Solid, Non-treated, No lid	M2000102N (40 Pcs/Box)
KeyTec® Fluorescent High-Transparency Microplate Top Seals	M1000102N (100 Pcs/Box)

## 3. Storage Conditions

- ◆ Upon receipt, store the reagent below -60 °C
- ◆ Up to 1 years from date of receipt, when stored and handled as recommended.
- ◆ When first thaw, aliquot the reagents as needed to avoid multiple freeze-thaw cycles.

## 4. Assay Procedure

### 1.1 Assay Format

Assay Format	Total Volume (20 $\mu\text{L}^3$ )
Other assay components	10 $\mu\text{L}$
KeyTec® TR-FRET Donor (Solar Eu/Tb) working solution (1X)	5 $\mu\text{L}$
KeyTec® TR-FRET Acceptor (LA/HX) working solution (1X)	5 $\mu\text{L}$

\*<sup>3</sup> The assay volume is optimized for 384-well microplates, and can be adjusted proportionally to perform in 96- or 1536-well microplates.

### 1.2 Reagents Handling

#### 1) Buffers

- ◆ KeyTec® TR-FRET Solar Eu Detection Buffer (A1010002L) has been optimized for maximum performance.
- ◆ Use the same buffer to prepare both the donor and the acceptor (LA/HX) conjugates.
- ◆ KeyTec® TR-FRET Binding Assay Diluent Buffer (A1010001L) is recommended for dilution and preparation of other components or samples.
- ◆ If using a homemade buffer solution, avoid SDS and ensure KF addition.

#### 2) Conjugates

- ◆ Thaw reagents on ice and equilibrate to room temperature before use.
- ◆ Prepare working solutions as per the purchased product instructions. The storage solution for KeyTec® TR-FRET mAb anti-FLAG-Solar Eu is 100X; dilute 100 times for a 1X working solution. For example, mix 50  $\mu\text{L}$  of the storage solution with 4950  $\mu\text{L}$  of KeyTec® TR-FRET Solar Eu Detection Buffer for a 1X working solution.
- ◆ Optimal amounts per well can be further optimized based on different assay format and conditions.

### 1.3 Data Calculating

- ◆ Calculate the ratio of 665 nm/615 nm (TR-FRET Ratio) and the CV for each individual well.

$$\text{TR-FRET Ratio} = \frac{\text{Signal 665 nm}}{\text{Signal 615 nm}} \times 10,000$$