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HTRF® Europium cryptate donor / Red acceptor readout Setup recommendations for Synergy 2

Two sequential measurements should be carried out: at 620 nm for the cryptate mission, and at 665 nm for the specific signal emitted by the acceptor (XL665 or d2). The ratio of the two fluorescence intensities 665/620 (acceptor/donor) enables the calculation of Delta F (%) which represents the relative energy transfer rate for each sample.

The Synergy 2 must be equipped with a TRF module, i.e. SLFPTA, or SLFPTAD or custom equipped with a T module (TRF).

Synergy 2 readers must be appropriately configured for HTRF[®] readout by setting up the measurement conditions in the Gen5[™] Reader Control and Data Analysis Software. In particular, these parameters should be entered as defined in the table below.

HTRF® assays must be read using the filter-based detection mode only. The monochromator mode is **not** HTRF® compatible.

Measurement 1

Excitation filter	330 (80) nm	Ref.: 7082263
Emission filter	620 (10) nm	Ref.: 7082266
Optics position	top 365	Ref.: 7138365
Number of flashes	10	
Lag time	100 μs	
Integration time	300 μs	
Sensitivity	Read the plate, select the well with the highest signal, and set its value at 50,000 counts	
Z	Select the default value given in the software	
Measurement 2		
Excitation filter	330 (80) nm	Ref.: 7082263
Emission filter	665 (8) nm	Ref.: 7082265
Emission filter Optics position	665 (8) nm top 365	Ref.: 7082265 Ref.: 7138365
	` '	
Optics position	top 365	
Optics position Number of flashes	top 365 10	
Optics position Number of flashes Lag time	top 365 10 100 μs 300 μs	

This reader only allows high performance HTRF measurement when assays are run in WHITE plates.





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HTRF® Terbium cryptate donor / Green acceptor readout Setup recommendations for Synergy 2

Two sequential measurements should be carried out: at 620 nm for the cryptate emission, and at 520 nm for the specific signal emitted by the acceptor. The ratio of the fluorescence intensities 520/620 (acceptor/donor) enables the calculation of Delta F (%) which represents the relative energy transfer rate for each sample.

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HTRF[®] assays must be read using the filter-based detection mode only. The monochromator mode is **not** HTRF[®] compatible.

Measurement 1

Excitation filter	340 (30) nm	Ref.: 7082230
Emission filter	620 (10) nm	Ref.: 7082265
Optics position	top 400	Ref.: 7138400
Number of flashes	10	
Lag time	100 µs	
Integration time	300 μs	
Sensitivity	Read the plate, select the well with the highest signal, and set its value at 50,000 counts	
Z	Select the default value given in the software	
M		
Measurement 2		
Excitation filter	340 (30) nm	Ref.: 7082230
	340 (30) nm 520(10) nm	Ref.: 7082230 Ref.: 7092163
Excitation filter	,	
Excitation filter Emission filter	520(10) nm	Ref.: 7092163
Excitation filter Emission filter Optics position	520(10) nm top 400	Ref.: 7092163
Excitation filter Emission filter Optics position Number of flashes	520(10) nm top 400 10	Ref.: 7092163
Excitation filter Emission filter Optics position Number of flashes Lag time	520(10) nm top 400 10 100 μs 300 μs	Ref.: 7092163 Ref.: 7138400 ell with the highest signal, and

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HTRF® Terbium cryptate donor / Red acceptor readout Setup recommendations for Synergy 2

Two sequential measurements should be carried out: at 620 nm for the cryptate emission, and at 665 nm for the specific signal emitted by the acceptor (XL665 or d2). The ratio of the two fluorescence intensities 665/620 (acceptor/donor) enables the calculation of Delta F (%) which represents the relative energy transfer rate for each sample.

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Measurement 1

Excitation filter	340 (30) nm	Ref.: 7082230
Emission filter	620 (10) nm	Ref.: 7082265
Optics position	top 400	Ref.: 7138400
Number of flashes	10	
Lag time	100 µs	
Integration time	300 µs	
Sensitivity	Read the plate, select the well with the highest signal, and set its value at 50,000 counts	
Z	Select the default value given in the software	
Measurement 2		
	340 (30) nm	Ref.: 7082230
Excitation filter	3 4 0 (30) 11111	
Excitation filter Emission filter	665 (10) nm	Ref.: 7082266
	` ,	Ref.: 7082266 Ref.: 7138400
Emission filter	665 (10) nm	
Emission filter Optics position	665 (10) nm top 400	
Emission filter Optics position Number of flashes	665 (10) nm top 400 10 100 μs 300 μs	Ref.: 7138400
Emission filter Optics position Number of flashes Lag time	665 (10) nm top 400 10 100 μs 300 μs	Ref.: 7138400

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