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HTRF® Europium cryptate donor / Red acceptor readout Setup recommendations for Infinite® M1000 pro

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.

Infinite[®] M1000 pro readers must be appropriately configured for HTRF[®] readout by setting up the measurement conditions in the "multilabeling" function of Tecan i-Control[™] software. In particular, these parameters should be entered as below. No special upgrade is required for HTRF[®] readout, as it is a monochromator-based instrument:

| | Label 1 | Label 2 |
|-----------------------|--|--|
| Measurement | Fluorescence intensity | Fluorescence intensity |
| Excitation wavelength | 317nm / 20nm | 317nm / 20nm |
| Emission wavelength | 665nm / 10nm | 620nm / 10nm |
| Mode | Тор | Тор |
| Flashes | Mode 2 [100Hz] : 100 | Mode 2 [100Hz] : 100 |
| Gain | Optimal | Optimal |
| Z position | Must be calculated from the well giving the highest signal | Must be calculated from the well giving the highest signal |
| Lag time | 60µs | 60µs |
| Integration time | 500µs | 500µs |

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 Infinite® M1000 pro

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 two fluorescence intensities 520/620 (acceptor/donor) enables the calculation of Delta F (%) which represents the relative energy transfer rate for each sample.

Infinite[®] M1000 pro readers must be appropriately configured for HTRF[®] readout by setting up the measurement conditions in the "multilabeling" function of Tecan i-Control[™] software. In particular, these parameters should be entered as below. No special upgrade is required for HTRF[®] readout, as it is a monochromator-based instrument:

| | Label 1 | Label 2 |
|-----------------------|--|--|
| Measurement | Fluorescence intensity | Fluorescence intensity |
| Excitation wavelength | 340nm / 20nm | 340nm / 20nm |
| Emission wavelength | 520nm / 10nm | 620nm / 10nm |
| Mode | Тор | Тор |
| Flashes | Mode 2 [100Hz] : 100 | Mode 2 [100Hz] : 100 |
| Gain | Optimal | Optimal |
| Z position | Must be calculated from the well giving the highest signal | Must be calculated from the well giving the highest signal |
| Lag time | 60µs | 60µs |
| Integration time | 500µs | 500µs |

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HTRF® Terbium cryptate donor / Red acceptor readout Setup recommendations for Infinite® M1000 pro

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.

Infinite® M1000 pro readers must be appropriately configured for HTRF® readout by setting up the measurement conditions in the "multilabeling" function of Tecan i-ControlTM software. In particular, these parameters should be entered as below. No special upgrade is required for HTRF® readout, as it is a monochromator-based instrument:

| | Label 1 | Label 2 |
|-----------------------|--|--|
| Measurement | Fluorescence intensity | Fluorescence intensity |
| Excitation wavelength | 340nm / 20nm | 340nm / 20nm |
| Emission wavelength | 665nm / 10nm | 620nm / 10nm |
| Mode | Тор | Тор |
| Flashes | Mode 2 [100Hz] : 100 | Mode 2 [100Hz] : 100 |
| Gain | Optimal | Optimal |
| Z position | Must be calculated from the well giving the highest signal | Must be calculated from the well giving the highest signal |
| Lag time | 60µs | 60µs |
| Integration time | 500µs | 500µs |

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

